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In Vivo Analysis

STUDIES RELATING TO THE ACCURACY OF IN VIVO MEASUREMENTS OF LIVER AND KIDNEY CADMIUM

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A greater understanding of the inaccuracies involved in *in vivo* measurements of liver and kidney cadmium by neutron activation was necessary. Studies were therefore carried out, using phantoms, into the effect on *in vivo* measurement accuracy of organ depth and organ mass. In addition an independent *in vitro* technique was developed, and used to analyse autopsy samples from three people previously measured *in vivo*, thus making direct comparisons possible. No evidence of bias in the *in vivo* results was found, but the need to correct for organ mass and depth was highlighted. Current measurement procedures result in residual uncertainties of ±25%.

Introduction

In vivo measurements of liver and kidney cadmium burdens by prompt gamma neutron activation analysis, using the thermal neutron capture reaction $^{113}Cd(n,\gamma)^{114}Cd$, have been carried out by several laboratories for a number of years (see review in Ref. /1/). At Birmingham detection limits, (2 standard deviations of the computed net peak area as the area tends to zero), of 6.5 μg g^{-1} have been achieved for liver measurements for a skin dose of 0.5 mSv, and 6.4 mg for kidney measurements, for a skin dose of 0.9 mSv. Such detection limits are slightly higher than the cadmium values observed in normal individuals, but are adequate for studying industrially exposed populations. The main problem, however, with the in vivo measurements, is not the precision but the accuracy, which is largely dependent on the measurement geometry, which is itself dependent on the build and state of health of the subject. So, although precision could be improved by increasing the dose, or the size and number of detectors, this would be of little value without a greater understanding of the sources of inaccuracy and preferably, a method of correcting for them. In this paper we present a series of investigations into these inaccuracies.

METHOD; Phantom Studies

***In vivo* measurements of liver and kidney cadmium are calibrated using a torso sized polythene bucket filled with water, in which organ phantoms of various cadmium concentrations are placed. The arrangement for the liver measurement system is shown in Figure 1; that for the kidney measurement is very similar. As with the *in vivo* measurements themselves, irradiation and counting take 1000s (live time) and the gamma spectra obtained are typically**

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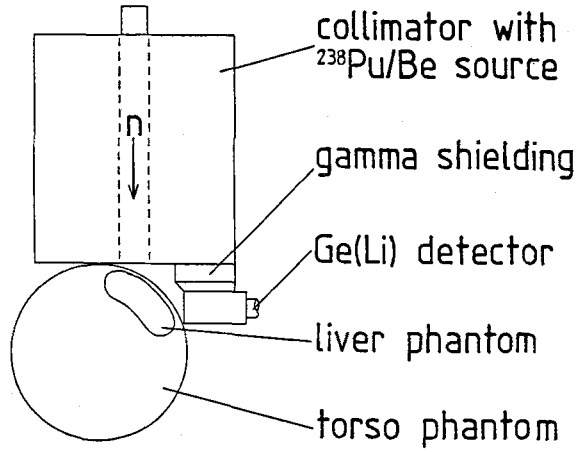

Fig. 1. *In vivo* liver cadmium portable assembly

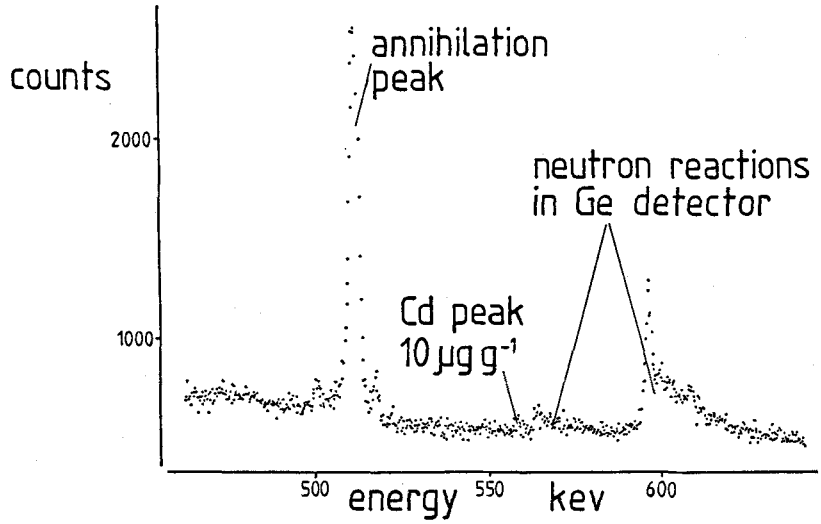

Fig. 2. *In vivo* gamma-ray spectrum

as shown in Figure 2. The spectra are analysed using a Marquardt algorithm /2/ to fit a parameterised function to the background simultaneously with an appropriate function for the 559 keV peak. Thus the area under the cadmium peak is obtained for each of the gamma spectra collected and a calibration line can be calculated.

The liver and kidney phantoms are made up to the masses given for Reference Man /3/ and are usually positioned in the bucket at an average depth, representing Reference Man (11 mm to the front surface of the liver,

55 mm to the centre of the kidney). Clearly however, few subjects measured have the build or organ sizes of Reference Man, and these differences lead to uncertainties in the results of the measurements. Two lines of investigation have therefore been followed. Firstly, the effect on measurement accuracy of organ depth into the body and secondly the effect of organ mass.

The effect of organ depth into the body is considerable, as it affects both the incident thermal neutron flux, and the attenutation of the emitted gamma rays. Before *in vivo* measurements are made, the liver and kidney depths are measured using ultrasound, so if the effect of depth on the measurement accuracy is known it can be corrected for in the final result. To investigate this the highest cadmium content phantom for each organ (150 $\mu g\ g^{-1}$ for liver, 60 mg for kidney) was used, and a series of spectra collected with each phantom progressively deeper into the torso phantom bucket. These spectra were analysed as described to obtain the area under the cadmium gamma peak as a function of organ depth.

The units of the results obtained from the two *in vivo* measurements are different, the kidney result being given as a total organ burden in mg and the liver result as an organ concentration in $\mu g\ g^{-1}$. The reason for this is that the kidney is a sufficiently small organ for the whole of it to be within the neutron beam, whereas the liver extends outside the beam and is therefore sampled. Thus when investigating the effect of organ mass on measurement sensitivity only the liver was studied.

In order to be able to vary the mass of the liver phantom, whilst keeping its relative proportions as near as possible constant, a set of sausage like phantoms /4/, containing different quantities of 150 $\mu g\ g^{-1}$ cadmium solution was used. A series of gamma spectra was then collected for liver phantoms of different masses, and analysed as before.

RESULTS; Phantom Studies

The results of the depth investigations are shown in Figure 3 for the liver and Figure 4 for the kidney, together with histograms showing the relevant range of organ depths observed at a recent survey. The results of the liver mass investigations are shown in Figure 5.

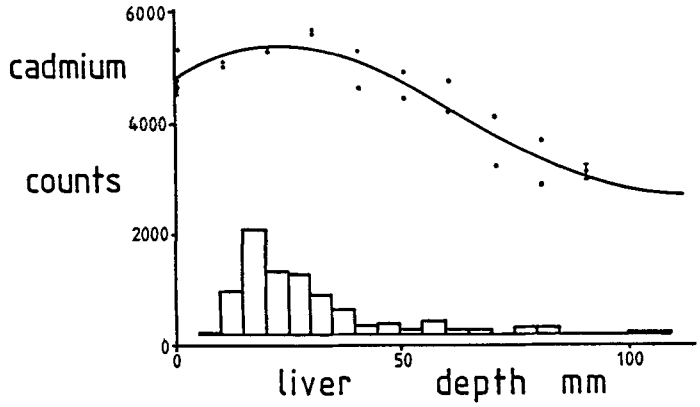

Fig. 3. Effect of liver depth on *in vivo* measurement

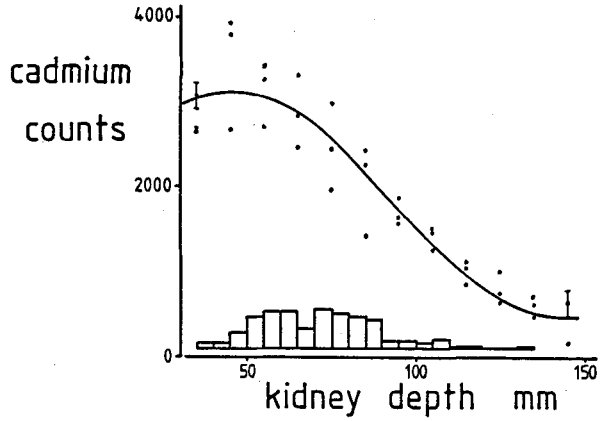

Fig. 4. Effect of kidney depth on *in vivo* measurement

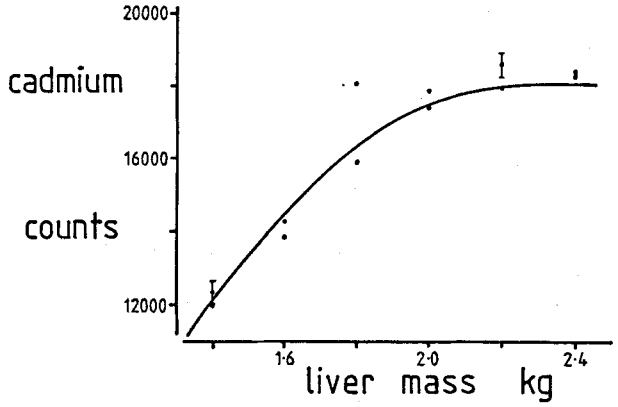

Fig. 5. Effect of liver mass on *in vivo* measurement

METHOD; In Vitro Analysis

In vitro analysis of tissue samples for cadmium is normally carried out by atomic absorption spectroscopy, but in this case it was decided to use neutron activation analysis since this permitted the analysis to be performed non-destructively. The 48.6 min half-life of ^{111m}Cd is ideal for such work because irradiations of 1 1/2 hours produce high activity and, on decaying, it emits two prominent gamma rays, at 245 keV (94%) and 151 keV (31%) /5/. It can be produced from natural cadmium by three different neutron reactions which are summarised in Table 1.

The main sources of interference were expected to be from activation of sodium and chlorine as a result of thermal neutron capture ($^{23}Na(n,\gamma)^{24}Na$, σ = 0.4b; $^{37}Cl(n,\gamma)^{38}Cl$, σ = 0.433b), both these elements being present in quite high levels in all tissues. It was decided to reduce the level of

Table 1
Neutron reactions forming the 48.6 min half-life ^{111m}Cd in natural cadmium (4)

Reaction	Isotopic Abundance	Threshold	Cross Sections
^{110}Cd(n,γ)	12.4%	0	100±30 mb at thermal 17 ± 3 mb at 25 keV
^{111}Cd(n,n')	12.8%	~0.4 MeV	average over 0.1 – 10 MeV
^{112}Cd(n,2n)	24.0%	~9.9 MeV	30 ± 2 mb

interfering activity produced by encasing the samples in cadmium sheet during irradation, thus reducing the incident thermal neutron flux. This also reduced the production of ^{111m}Cd by the ^{110}Cd(n,γ) reaction.

The Nuffield cyclotron was used for neutron production by bombarding a thick beryllium target with 20 MeV deuterons. The resulting neutron spectrum extends up to about 21 MeV, but peaks around 7–8 MeV /6/. As the total cross-section for the production of ^{111m}Cd from natural cadmium by epicadmium neutrons also peaks to 80 mb around 7–8 MeV, yields would be expected to be high.

The samples, which were irregular in shape, were frozen and heat sealed in two layers of polythene sheet. The calibration phantoms consisted of aqueous solutions of sodium and chlorine of approximately the correct physiological concentrations /3/, so that interference in their measurement would be of the same order as that for the samples, and the cadmium contents ranged from 0–25 mg. These were made up to 0.009 m^3 (this being close to the mean volume of the samples) heat sealed in polythene and frozen to have approximately the same dimensions as the samples.

During irradiations the neutron beam flux inevitably varies, so a method of normalising the results obtained was necessary. The most reliable method was found to be by simultaneous irradiation of high cadmium content (40 mg in 0.009 m^3) reference phantoms, which were made in an identical fashion to the calibration phantoms.

The sample, or calibration phantom, and a reference phantom were each encased in cadmium, but separated from it by polythene to avoid contamination, and positioned symmetrically in a polythene tub. To keep the sample frozen during the 1½ hour irradiation they were packed in dry ice. The tub was placed on a platform 100 mm back from the beryllium target, (Figure 6) to ensure a uniform incident beam. The platform was rotated at 4 r.p.m. throughout the irradiation in order to make the irradiation as uniform as possible for both sample and standard.

After irradiation the sample, or calibration phantom, and reference phantom were counted simultaneously on a pair of Ge(Li) detectors. To minimise background the samples were surrounded by lead and the detectors by bismuth annuli. Gamma spectra were collected for 5000s (live time) starting about 3 minutes after the end of irradiation. The spectrum from Subject 3's left kidney is shown in Figure 7. Although the cadmium wrapping prevented thermal neutron activation of sodium and chlorine, these elements still gave the main interferences, via epicadmium activation to produce ^{34m}Cl and ^{22}Na. Both isotopes were produced by (η,2η) reactions and were positron emitters;

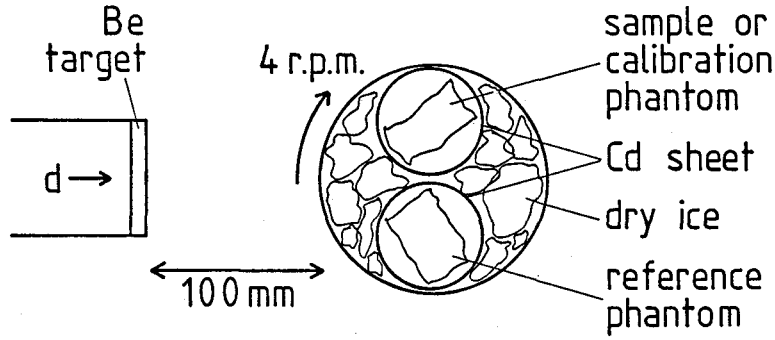

Fig. 6. Irradiation geometry for *in vitro* analysis.

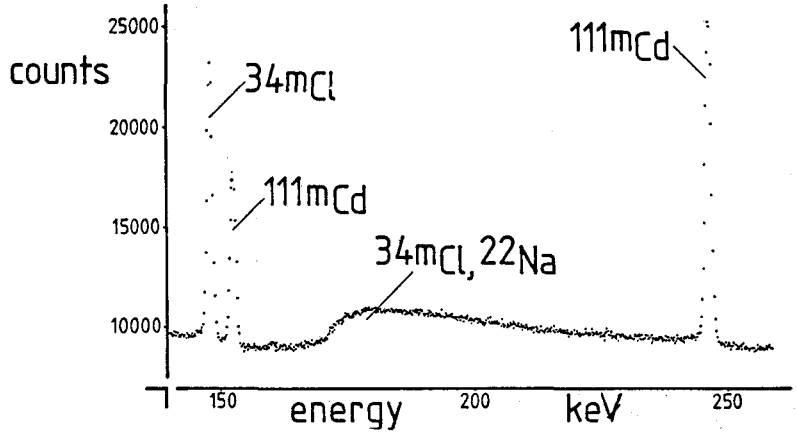

Fig. 7. *In vitro* gamma-ray spectrum

they were therefore responsible for the Compton backscatter edge seen at 170 keV in Figure 7.

The gamma spectra obtained were also analysed using a Marquardt algorithm. For each pair of samples the ratio of the 245 keV photopeaks was determined.

The accuracy of the technique was assessed by irradiating several pairs of reference phantoms, and a coefficent of variation in the peak ratio of 15% was found. Such a spread could arise from a number of factors, but the principal one is thought to be that of positional uncertainties in gamma ray counting, due to the irregular shapes involved.

RESULTS; *In Vitro* Analysis

The results of the *in vitro* analyses along with the corresponding *in vivo* results are shown in Table 2. The errors quoted are those from counting statistics added in quadrature with estimates of geometrical uncertainties, these being ±25% for *in vivo* results and ± 15% for *in vitro* results.

Table 2
Comparison of *in vivo* and *in vitro* results

	Subject 1		Subject 2		Subject 3	
	liver	kidney	liver	kidney	liver	kidney
total organ weight (g)	1120	ℓ 200 r 195	1280	ℓ 100 r 90	1415	ℓ 174 r 150
sample weight (g)	6.9	1.9	97.4	ℓ 104.5 r	1187.2	ℓ 174.0 r 142.8
in vivo result	45.2 ±11.9	ℓ 17.1 ±4.8	−3.1 ±10.9	ℓ 4.9 ±3.5	47.1 ±12.3	ℓ 26.5 ±7.4
in vitro result (scaled to whole)*	43.4 ±9.1	13.2 ±5.7	10.5 ±3.3	7.8 ±1.5	76.7 ±11.8	ℓ 14.2 ±3.6 r 26.4 ±4.1
Δ	0.120	0.523	−1.194	−0.762	−1.737	1.495
σ_Δ						
time lapse	2 months		4 years		1 year	

(liver results in μg g^{-1}, kidney results in mg.)
Δ = *in vivo* − *in vitro*
* whole liver (average concentration) or whole left kidney (total amount).

Table 3
Results of analysis of subject 3's liver

Sample	Sample mass	Cd concentration (μg g^{-1})
1	94.9	68.49 ± 8.96
2	139.4	49.93 ± 6.03
3	130.1	69.25 ± 6.15
4	122.3	78.94 ± 4.76
5	152.3	95.99 ± 4.99
6	112.9	40.57 ± 8.06
7	180.4	84.20 ± 4.21
8	94.8	70.46 ± 8.97
9	98.1	122.53 ± 7.75
10	62.0	95.16 ± 14.03
mean	118.7	77.55 ± 23.69
total	1187.2	76.74 ± 2.38

Subject 2's liver sample was a large proportion of the whole organ, and thus too large to analyse as one sample. It was therefore analysed in ten sections, allowing the gross distribution of cadmium within the organ to be studied. A sketch of the sectioning of the liver is shown in Figure 8, and the results of the analyses in Table 3. The errors quoted here are purely those from counting statistics.

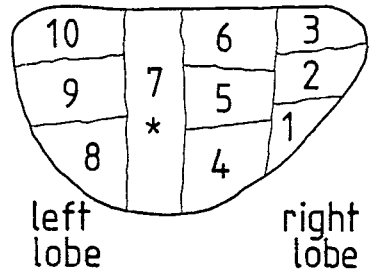

Fig. 8. Sectioning of Subject 3's liver; *includes gall bladder

Discussion

As can be seen from Figure 3 the sensitivity of the *in vivo* liver cadmium measurements to organ depth is not very great. This, combined with a relatively small range of organ depths, results in 90% of depth correction factors lying in the range 0.9–1.2. In contrast, because the kidney is a much smaller organ and tends to be deeper into the body, measurement sensitivity falls off much more (Figure 4). When this effect is combined with a wider distribution of organ depths the result is a 90% range of depth correction factors of 1–2.1 with outliers as high as 3.9. The use of these corrections is therefore very important to the accuracy of the final results. It should, however, be noted that the uncertainty in the depth measurements is of the order of ±2 mm, which is equivalent to an uncertainty in depth correction factor (depending on depth) of around ±0.1.

Reference Man has an 1800g liver, but the 80% range of liver masses for adult men of working age is roughly 1400g–2300g, and if older men are included the lower bound is reduced to 1000g. Reference Woman has a 1400g liver and an 80% range of masses of 1000g–1900g. It follows that the liver cadmium results for most men are of the correct order, but may be inaccurate by ±12% due to organ mass variations. The *in vivo* liver cadmium concentrations for most women, and for small or elderly men, could however, be underestimates, some by as much as 30%. Ideally, liver mass corrections would be made to the *in vivo* measurement results, but the small portable ultrasound machine currently used for organ depth measurements is not capable of measuring liver volume. In addition, if such corrections were to be made, a more detailed study, including organ shape as well as mass would have to be made. If detailed corrections are not possible it may be that there is a sufficient correlation between frame size, (height^3 or height^2 x shoulder width), and liver mass, to use that to remove some of the measurement uncertainty due to organ shape and mass. However, this has not yet been investigated.

Having shown how large uncertainties in *in vivo* results can be due to geometrical variations it is surprising to see, Table 2, how close the *in vivo* results are to the more accurate *in vitro* results. Indeed the agreement between the two sets of results for subjects 1 and 2 is very close, with those for subject 3 less so. There are two possible explanations for subject 3's *in vivo* liver result being low. Firstly it is possible that he slumped away from the collimator during the measurement, thus moving his liver further from both the neutron source and the detector. Secondly it could be that the *in vivo* result is an underestimate due to the small organ size. If the results in Figure 5 are used to correct the *in vivo* results for liver mass it is increased to 62.0 $\mu g\ g^{-1}$, thus improving the agreement. However, if similar corrections are applied to subjects 1 and 2 *in vitro* liver results the agreement is worsened. The difference in subject 3's two left kidney cadmium results is only just outside the quoted experimental errors.

When the *in vivo* results are compared in detail with the *in vitro* results they show no evidence of bias or systematic error greater than already assumed, the mean and standard deviation of Δ/σ_Δ being −0.26 and 1.2, which are not significantly different from 0.0 and 1.0 respectively. The comparison does, however, confirm the need for allowances of ±15% on the *in vitro* results and ±25% in the *in vivo* results.

Finally there is the distribution of cadmium within the liver. There was some concern that, because only a section of the liver was being measured *in vivo*, the results might not be representative of the whole organ. The sectioned *in vitro* analysis, (Table 3) shows no evidence for systematic variation with lobe or other position within the organ. It would seem likely that the cadmium accumulated in any small volume of liver depends on the blood supply to that volume, which will vary randomly through the organ. Thus provided the proportion of organ sampled is relatively large, which it is, the result should be representative.

*

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References

1. M. C. SCOTT, D. R. CHETTLE, Scand. J. Work Environ. Health, 12 (1986) in press.
2. P. R. BEVINGTON, Data Reduction and Error Analysis for the Physical Sciences, McGraw Hill, New York, 1968.
3. ICRP 1975 Publication 23, Report of the Task Group on Reference Man, Pergamon, Oxford.
4. C. B. OXBY, K. BROOKS, Phys. Med. Biol., 24 (1979) 440.
5. D. L. SMITH, J. W. MEADOWS, Nucl. Sci. Eng., 81 (1982) 525.
6. M. CATTERALL, D. K. BEWLEY, Fast Neutrons in the Treatment of Cancer, Academic Press, London, 1979.