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PROCEEDINGS

13th
ANNUAL

**BIOASSAY
&
ANALYTICAL
CHEMISTRY
MEETING**

OCTOBER 12-13 1967

Held at LAWRENCE
RADIATION
LABORATORY

University
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Proceedings of the 13th Annual
BIOASSAY AND ANALYTICAL CHEMISTRY MEETING

held at
Lawrence Radiation Laboratory
October 12 and 13, 1967

April 1968

Compiled by
Anne deG. Low-Beer
Lawrence Radiation Laboratory

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Berkeley, California

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Foreword

The Thirteenth Annual Meeting on Bioassay and Analytical Chemistry was held at the Lawrence Radiation Laboratory, Berkeley, California 94720.

These meetings are held to give AEC and AEC contractor personnel an opportunity to discuss procedures and problems associated with the collection, interpretation, and application of bioassay and other pertinent health data.

Proceedings of previous meetings have been issued as the following AEC reports:

1955	NLCO-595
1956	WASH-736
1957	none published
1958	WASH-1023
1959	TID-7591
1960	TID-7616
1961	ANL-6637
1962	DP-831
1963	TID-7696
1964	CONF-727
1965	CONF-651008
1966	CONF-661018

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ANALYSIS OF AMERICIUM AND PLUTONIUM
FOLLOWING SINGLE INHALATION EXPOSURES

S. E. Hammond and D. L. Bokowski

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The analytical method used for determination of americium and plutonium in feces is described. Fecal excretion by 25 persons incurring greater than maximum permissible lung burdens during a plutonium fire has been studied. These results are compared with lung retention studies by in vivo counting of ²⁴¹Am.

STABLE LEAD INVESTIGATIONS AT HASL

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An analytical procedure has been developed for the determination of stable lead concentrations in some biological and environmental samples. A freeze-drying technique is used prior to wet-ashing biological samples in order to speed up the digestion process. The lead is concentrated by solvent extraction of the dihydrogentetraiodo complex with 3-methyl, 2-butanone. Subsequently the lead is determined by atomic absorption spectrophotometry of the organic extract. An alternative method would be the spectrophotometric determination of the lead dithizone complex at 515 millimicrons. The chemical recovery is determined by isotope dilution by use of ^{212}Pb tracer (10.6 h), which is purified from thorium nitrate by a modified solvent-extraction procedure.

Data will be presented for stable lead concentrations in some foods, air, and water samples.

STABLE LEAD INVESTIGATIONS AT HASL

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INTRODUCTION

Recently, Patterson⁽¹⁾ pointed out that man's present blood lead concentration is approximately 40% of the threshold for lead poisoning. This has stimulated a controversy over the present lead concentration in man. To validate some of the existing data for stable lead, a program was initiated at HASL. The preliminary work on diet and air is reported in this paper.

New York City food samples from the laboratory's tri-city fallout sampling program were utilized to establish man's daily lead intake and lead concentration in food products. Air samples from the laboratory roof, routinely collected in the HASL surface air program, were used to determine particulate lead air concentration and calculate man's daily lead inhalation.

SAMPLE ANALYSIS

The initial consideration prior to chemical analysis was preparation of the sample so that losses and contamination could be minimized. Although reagent contamination can be reduced in some matrices by dry ashing, Gorsuch⁽²⁾ found losses of lead in food dry ashed at 500°C, but obtained average recoveries of 100% by wet ashing in a nitric acid system exclusive of sulfuric acid. However, the use of wet ashing is somewhat restrictive because large quantities of commercially obtained nitric acid contribute significant amounts of contaminating lead to the samples. Previously, it had been determined at the laboratory that wet ashing with small volumes of nitric acid was successful with sample

dehydration. This technique is satisfactory for food samples except for those with high fat content. New borosilicate glassware was used to reduce contamination caused by acid leaching of the beakers.

The daily air samples on 2-inch microsorban (polystyrene) filters were composited, wet ashed, and the resultant solution analyzed for stable lead. The losses of lead during solubilization of the polystyrene filters is considered negligible. This has been confirmed by studies of radioactive material collected on this type of filter⁽³⁾.

The preliminary chemical procedure was based upon the classical solvent extraction of lead with dithiozone and measurement of the color complex at 510 m μ ⁽⁴⁾. Steps were added to this procedure to separate and to further purify the lead prior to the measurement⁽⁵⁾.

An alternate and shorter procedure was developed using atomic absorption spectrophotometry. The separation step was a solvent extraction of the dihydrogentetraiodo lead (II) complex from 5% HCl into 3-methyl, 2-butanone⁽⁶⁾. The organic extract was diluted to a known volume and measured directly on a Perkin-Elmer Model 303 atomic absorption spectrophotometer. The 284-m μ secondary absorption line was used, since there is solvent interference with the primary 212-m μ line. This procedure was used for both the food and air samples.

The chemical recovery was obtained using the radioactive tracer Pb-212. The isolation and purification of the Pb-212 tracer from natural thorium nitrate was performed by a solvent extraction method described by Petrow⁽⁷⁾ and modified at HASL⁽⁵⁾.

RESULTS AND DISCUSSION

Chemistry

Table I shows an average recovery, after correction for blank contamination, of 94% of the lead which had been added to spike samples.

TABLE I. ANALYSIS OF SPIKED SAMPLES

Milk Sample No. (1 kg)	Pb Added (μg)	Pb Recovered (μg)
1	0	0
2	100	88
3	200	184
4	300	292
5	400	372
6	500	<u>504</u>

Average Lead Recovery - 94%

Table II indicates that the method is reproducible to within 10% for split samples.

TABLE II. MEASUREMENT OF SPLIT SAMPLES

Sample	1st Analysis ($\mu\text{g}/\text{kg}$)	2nd Analysis ($\mu\text{g}/\text{kg}$)	Average \pm % Mean Dev.
Milk A	42	36	39 \pm 8
Fresh vegetables	130	110	120 \pm 10
Canned vegetables	426	444	435 \pm 2
Milk B	36	38	37 \pm 4

It was necessary to measure the lead in the nitric acid used in wet ashing, since this reagent is a major source of contamination. We performed four measurements on the acid and found a lead concentration of 2-5 $\mu\text{g}/\text{liter}$. Therefore, the procedure was designed to never exceed two liters of nitric acid for the one kilogram sample size. The reagent contamination level of the entire technique, including sample preparation, was evaluated by analysis of several blanks and

found to be $20 \pm 5 \mu\text{g}$. During the investigation of overall contamination, we established that reagent-grade ammonium hydroxide contributed the most significant amount of lead contamination. This is probably due to the leaching of the lead from the walls of the reagent bottle.

The procedure for preparation of the tracer provides radiochemically pure Pb-212. This was verified by gamma counting prepared solutions of Pb-212 over a period of several days. The half-life of Pb-212 was determined to be 10.5 hours (Fig. 1). This is in good agreement with the established value of 10.6 hours⁽⁸⁾. Since the half-life of Pb-212 is 10.6 hours and the sample preparation time is 3-6 days, the tracer had to be added after solubilization.

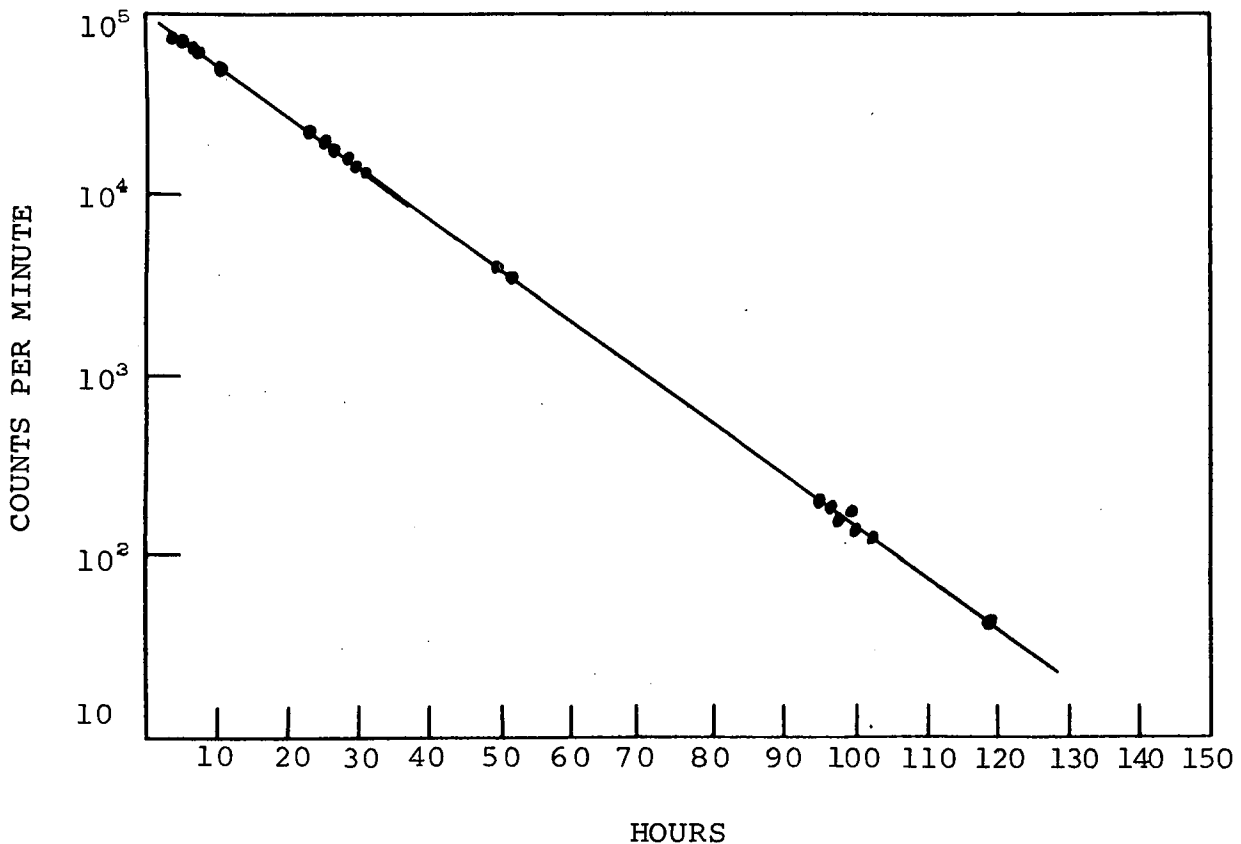


Figure 1. Decay of lead-212 tracer separated from thorium nitrate. Observed half-life of the tracer was 10.5 hours.

Analysis

The results for the food samples of this preliminary investigation are given in Table III.

TABLE III. LEAD IN FOOD

<u>Category</u>	<u>μg Pb/g Raw Food</u>
Fresh fish	0.157
Shell fish	0.311
Poultry	0.301
Meat	0.420
Eggs	0.222
Fresh fruit	0.072
Canned fruit	0.251
Fresh vegetables	0.120
Canned vegetables	0.435
Root vegetables	0.068
Potatoes	0.174
Macaroni	0.077
Rice	0.044
Juices	0.091
Dry beans	0.024
Flour	0.041
Bakery products	0.393
Whole grain products	0.129
Milk	0.038

Using established yearly average intake values⁽⁹⁾, the daily intake of stable lead was found to be 275 μg. Table IV gives the yearly intake of lead for the different food categories. Kehoe⁽¹⁰⁾ has reported intake values from 300-500 μg/day by analyses of fecal samples, so that our value of 275 μg is in general agreement with his data.

TABLE IV. YEARLY INTAKE OF LEAD IN FOOD

<u>Category</u>	<u>Yearly Average Intake (kg)</u>	<u>Total Yearly Lead Intake (μg)</u>
Fresh fish	8.09	1270
Shell fish	1.36	420
Poultry	16.85	5070
Meat	72.28	30360
Eggs	16.0	3550
Fresh fruit	67.89	4890
Canned fruit	14.95	3750
Fresh vegetables	43.20	5180
Canned vegetables	21.92	9540
Root vegetables	16.90	1150
Potatoes	44.62	7760
Macaroni	3.01	230
Rice	2.58	110
Juices	10.10	920
Dry beans	2.72	65
Flour	43.13	1770
Bakery products	36.27	14250
Whole grain products	10.90	1410
Milk	<u>214.1</u>	<u>8140</u>
	Total 647	99835

An interesting observation is the comparative results obtained for fresh and canned vegetables. The canned vegetables are a factor of 3.6 greater in lead concentration than fresh vegetables, and the analyses were performed in duplicate with good agreement ($\pm 2\%$). Similarly, the results for fresh and canned fruit show a factor of 3.5 greater lead concentration in the canned product. It appears that lead from the soldered seams of the can was solubilized.

The results for monthly air samples are reported in Table V. The values for April and June are lower than those

reported by Ludwig⁽¹¹⁾ in the U. S. Public Health Service tri-city air sampling program. His values were in the range of 1-3 $\mu\text{g}/\text{m}^3$, and the average value was 1.5 $\mu\text{g}/\text{m}^3$, except for Los Angeles. However, the lower value obtained for the April sample and the higher value for the November sample in our investigation are consistent with the seasonal variations observed in the U. S. Public Health Service program.

TABLE V. LEAD IN AIR*

Date	Total Air Volume (m^3)	Total Lead (μg)	μg Lead/ m^3
April 1966	2641	1700	0.64
June 1966	2953	2550	0.86
November 1966	2805	6650	2.37

*All air samples were collected at a height of about 120 feet and not at ground level.

The November sample filter collection was split into three groups of 10-day intervals because there was a temperature inversion during the period of November 23-28. The November values are listed in Table VI. It can be seen that during the period of the inversion concentrations of lead were higher by a factor of ≈ 2 .

TABLE VI. VARIATION IN LEAD AIR CONCENTRATION NOVEMBER 1966

Date	Total Air Volume (m^3)	Total Lead (μg)	μg Lead/ m^3
November 1-10	922	1880	2.04
November 11-20	976	1630	1.67
November 21-30**	906	3140	3.47

**Period of Inversion.

Based upon the measured minimum and maximum lead air concentrations and assuming a total daily air inhalation of 2×10^7 cc⁽¹²⁾, the calculated total lead inhaled daily ranges from 12 to 74 μ g. These calculated intake values are consistent with existing data⁽¹⁰⁾.

SUMMARY

Stable lead in food and air was measured to determine man's daily intake. Samples were prepared by freeze-drying and wet ashing to minimize contamination. Of the two chemical procedures developed, one based on colorimetry, and the other on atomic absorption spectrophotometry, the latter was used for sample analysis. The chemical recovery was determined by using a Pb-212 tracer. The overall contamination level was found to be 20 ± 5 μ g.

Measurement of the lead content in 19 foods of a typical diet gave a calculated daily lead intake of 275 μ g. Measured composited monthly New York City air samples had from 0.6 to 3.5 μ g/m³ stable lead. Based on these values the calculated daily inhalation of stable lead is 12 to 74 μ g. The observed food and air values are consistent with existing data.

REFERENCES

1. Patterson, C. C., Arch. Environ. Health 11, 344 (1965).
2. Gorsuch, T. T., Analyst 84, 137 (1959).
3. Collins, W. R., Unpublished data (1964).
4. Morrison, G. H., and Freiser, H., Solvent Extraction in Analytical Chemistry, John Wiley and Sons, New York (1957), p. 214.
5. Bogen, D. C., and Kleinman, M. T., Analyst 92, 611 (1967).
6. Talvitie, N. A., and Garcia, W. I., Anal. Chem. 37, 851 (1965).

7. Petrow, H. G., and Cover, A., Anal. Chem. 37, 1659 (1965).
8. Strominger, D., Hollander, J. M. and Seaborg, G. T., Rev. Mod. Phys. 30 (2), Part 2, 188 (1958).
9. Rivera, J., and Harley, J. H., USAEC Health and Safety Laboratory Report, HASL-147 (1964).
10. Kehoe, R. A., Arch. Environ. Health 2, 418 (1961).
11. Ludwig, J. H., Diggs, D. R., Hesselberg, H. E., and Maga, J. A., Am. Ind. Hyg. Assoc. J. 26, 270 (1965).
12. Blatz, H., Radiation Hygiene Handbook, McGraw-Hill Book Company, Inc., New York (1959).

LABORATORY BLANK EXPERIENCE IN LONG-LIVED FISSION PRODUCTS

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Tracerlab performs routine quantitative radiochemical analysis on thousands of samples annually. These samples derive from many sources. In particular, Tracerlab participates in various environmental and bioassay analysis programs. The High Altitude Balloon Sampling Program for AEC - HASL and the determination of the activity in single and aggregate fallout particles for the DOD and AEC DBM are representative. As part of a broader program on standards, calibration, spikes, and controls, a radioactivity blank program is carried out. This blank program is especially important because at high altitudes the small volume of air sampled precludes obtaining high activity levels. Thus, the counting rate of such long-lived fission products as ^{90}Sr , ^{137}Cs , ^{144}Ce , and ^{147}Pm is extremely low. Although the laboratory blank program encompasses induced activities, heavy elements, and natural radioisotopes as well, this paper reviews only blank experience with the above isotopes.

The analytical program under way at Tracerlab is periodically tested for radioactive blanks. The potential blank due to the chemistry and counting is discussed. Each contributor such as reagents, carrier, and planchets is examined. The potential for contribution by the laboratory environment, equipment, and glassware is determined. Blank procedures are carried through complete sample dissolution, separation, and purification. Since the blanks follow closely the route of actual samples through the chemistry and counting laboratories, they are specific for the isotope of interest. Wherever possible, the source and the activity found are identified so that steps to remove the undesired activity are effective. The effect of the counting instruments background relative to the limits of detection is assessed.

Laboratory blank data for these elements taken during the past ten years are presented and discussed. The methods and techniques developed to reduce the individual blanks are described. The total laboratory blank has been reduced to 0.2 to 0.3 cpm, of which most is due to the planchet assembly.

In conclusion, this experience has resulted in a mode of laboratory operation which leads to a minimum and consistent blank. These precautions are discussed and recommendations for further potential reduction proposed.

LABORATORY BLANK EXPERIENCE IN LONG-LIVED FISSION PRODUCTS

by

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Over the years, the measurement of radioactivity at low levels has become an order of magnitude more sensitive through the use of sophisticated low-background equipment. As a result, the establishment of a laboratory radioactivity blank has become an important part of the assessment of background or near background samples. This blank differs for every isotope and must be checked on a regular basis.

In setting up any quality control program, there must be a balance between theory and practicality. The extent of the blank program is set by the objectives of the analytical program. Past blank experience must be relied on at the outset to forecast the blank. The analytical schedule is then set up which includes blanks and spikes, and sample processing is commenced. In this respect, laboratory blanks, like counter backgrounds, are intrinsic. If, through some accident, contamination is present in the reagents, glassware, or elsewhere, the run of irreplaceable samples might be ruined. The blank should pick this up but it might be an after-the-fact finding. In a practical assessment, such a happening is always a possibility. In a well-kept, well-run laboratory, it is not probable but always must be guarded against. From another point of view, the blank values obtained with any given set of samples are used to give further credence to the average laboratory blank value.

Because Tracerlab performs routine quantitative radiochemical analysis on thousands of samples annually, blank studies must be performed. These samples derive from many sources. In particular, Tracerlab participates in various environmental and bioassay analysis programs. The High Altitude Balloon Sampling Program for AEC, HASL, and the determination of the activity in single and aggregate fallout particles for the DOD and

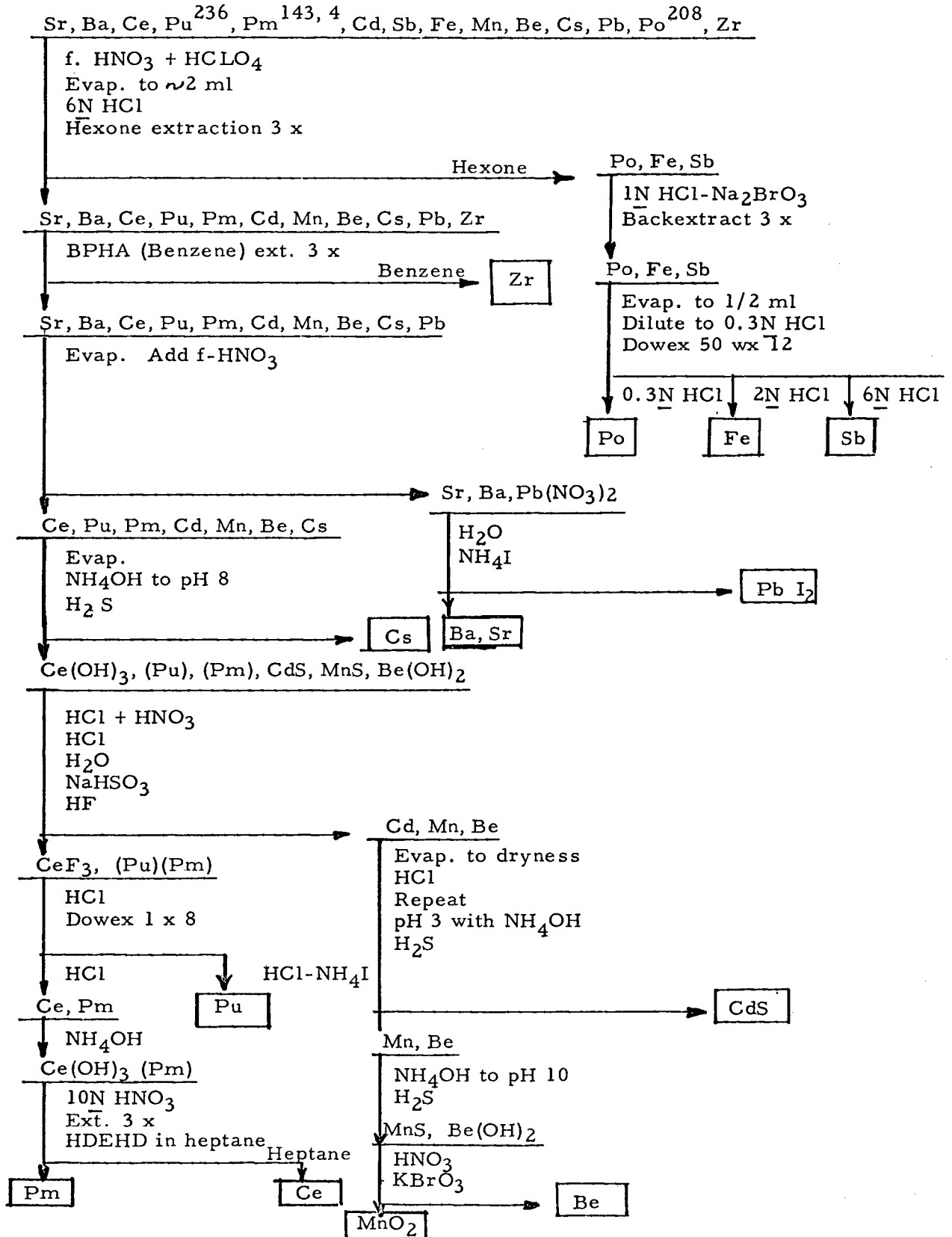
AEC, DBM, are representative. The radioactivity blank program is carried out as part of a broader standards, calibration, spikes and controls program. This blank program becomes especially important because at high altitudes the small volume of air sampled precludes obtaining high activity levels. Thus, the counting rate of such long-lived fission products as Sr^{90} , Zr^{95} , Cs^{137} , Ba^{140} , Ce^{144} , and Pm^{147} is extremely low. Although the laboratory blank program encompasses many of the induced activities, heavy elements and natural radioisotopes as well, this paper will review blank experience only with the above isotopes and Tl^{204} and Pb^{210} .

A typical fission product radioanalytical scheme at Tracerlab contains the following operations:

1. Sample dissolution - varies with sample.
2. Sequential separation of carrier elements - varies.
(Typical scheme used on fallout samples is illustrated in Fig. 1.)
3. Purification steps - usually standard.
4. Brass-backed sample mount assembly - standard for isotope.
5. Counting equipment - standard for isotope.

The definition of "blank" is generally taken to be the residual radioactivity measured in a sample which has been processed by the same procedure and materials as the real sample it represents. T. T. Sugihara, in his comprehensive monograph "Low-level Radiochemical Separations"⁽¹⁾ has a similar definition: "the contribution of the added reagents and other constituents to the quantity measured." The actual components included will vary with the case, depending upon the ability to reassign some components of the blank as a constant correction. The sources of potential blank may be the environment or due to chemical processing or counting. Each possible contributor such as reagents, carrier, and planchets is individually or collectively analyzed. The potential for contribution of the laboratory environment, equipment, and glassware is determined. The blank procedures are carried through complete sample dissolution, separation, and purification simulating actual individual laboratory procedures. Since the blanks follow closely the route of actual samples through the chemistry and counting laboratories, they are specific but necessarily applicable only to the isotope of interest. Wherever possible, the source

FIGURE 1. SEQUENTIAL SEPARATION SCHEME FOR FALLOUT SAMPLES.



and the activity found is identified so that steps to remove the undesired activity are effective. The isotope blank relative to the limits of detection of the counting instruments is assessed.

There seems to be disagreement among analysts on blanks, possibly because of the wide variety of specialized laboratories in low-level radiochemistry. From the previous definition, a blank need not be isotopic with that isotope being analyzed. In some discussions, workers consider a blank to be only isotopic. Bowen and Sugihara⁽²⁾ report they found no systematic radiochemical blank ascribable to either Sr⁹⁰ or Cs¹³⁷, whether in their reagents, in simulated samples, or in real samples. They also recommend considerable caution in the interpretation of reported values to which these blanks have made a significant contribution. Rocco and Broecker^{(3), (4)} on the other hand found considerable blanks in their analyses of large sea water samples for Cs¹³⁷ and Sr⁹⁰. However, the latter groups define their blanks in a rather unique way which is applicable only to a water or sea water sample. For instance they perform a Cs¹³⁷ analysis on sea water which has already been put through their normal Cs¹³⁷ separation. They correct any recovered activity for residual Cs¹³⁷ not removed in the initial Cs¹³⁷ analysis. Their (Ref. 3) corrected blank results for Cs¹³⁷ range in dpm from (-) 2.1 to + 3.1 (average 1.1 ± 0.4 dpm) and for Sr⁹⁰ from 1.6 to 7.2 dpm (average 4.5 ± 2.0 dpm). Sugihara⁽⁵⁾ points out how the use of such a technique on Ce¹⁴⁴ in sea water could result in ambiguous blank determinations.

Prior to 1956, counting measurements on purified fission product samples at Tracerlab were made on a general all-purpose proportional counter. The Tracerlab Methane-End-Window (MEW) counter has a background of approximately 15 cpm. The limit of detection was taken as 1 cpm. Sample measurements with low levels of activity in the region of 1 to 20 cpm were determined but used with large error limits.

It soon became apparent that an increase in analytical sensitivity was needed to obtain results below that limit of detection. A low-background beta counter with approximately 0.5 cpm background and efficiency equivalent to the MEW counter was developed and produced here as Model CE-14. A typical efficiency is 0.60 for a Y⁹⁰ sample. Many of the original CE-14 detectors are still in daily use.

The use of CE-14 counters expanded the ability of the radiochemists to provide usable data by an order of magnitude. They filled a badly needed gap, and thus were in constant use in sample analysis. Previously suspect measurements in the region of 1 to 20 cpm on long-lived samples from the pre-CE-14 era could be reassayed to obtain positive results. This new sensitivity made the radioactive blank an important contributor to the uncertainty assigned to the analytical results. Systemization and control of extraneous activities was necessary if good data was to be obtained.

The control and blank program at Tracerlab prior to the use of low-background counters had consisted mainly of assuring that no grossly contaminated material entered the processing sequence. The largest potential source of contamination being cross-contamination from laboratory isotope stocks or hot samples. A token, but adequate, blank program consisted of measuring activity in standardized carrier solutions and purified samples from any procedure tests and training runs.

As the new blank program evolved, emphasis was given to determining

1. Activity in the sample mount, individual parts and combined parts.
2. Activity in standardized carrier equivalent to a standard mount.
3. Activity remaining after correcting for (1) and (2).

Let us first discuss the possible sources of the "blank activity" as pertains to a mounting sandwich depicted in Fig. 2. The elemental carrier is filtered onto Whatman #42 paper, sandwiched between two Mylar films, and subsequently mounted on a brass backing planchet by use of an aluminum friction ring. This assembly has proven to provide extremely good sample integrity and reproducible geometry, and greatly increases counting efficiency when compared to synthetic materials due to its excellent backscatter characteristics.

Unfortunately, even carefully selected starting materials usually result in small planchet assembly count rates. Table 1 gives the results of counting typical bare planchets, the aluminum rings and the planchet ring assemblies as received from a manufacturer in August 1965. The Al rings measured 0.18 cpm, the brass disc 0.20 cpm, but the total activity when assembled was 0.26 cpm and less than the expected sum of 0.38 cpm. These results indicate some self-shielding of the planchet by the aluminum

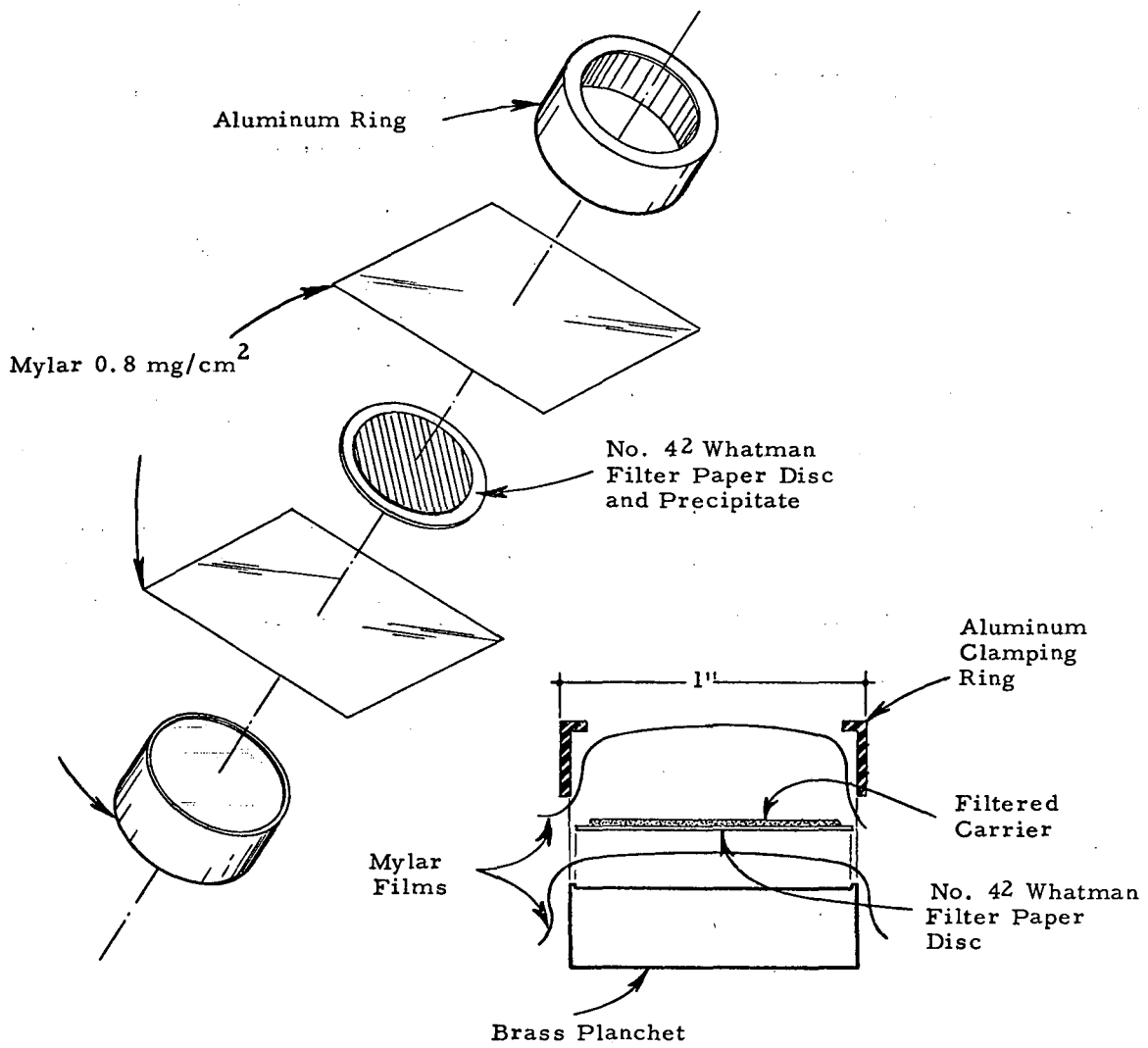


FIGURE 2. STANDARD TRACERLAB MOUNTING SANDWICH

TABLE 1. RESULTS OF COUNTING COMPONENTS OF PLANCHET ASSEMBLY.

Designation	Al Ring Only cpm	Planchet Only cpm	Ring and Planchet (a) cpm	Ring and Planchet (b) cpm
A	0.17	0.23	0.40	0.25
B	0.22	0.19	0.41	0.35
C	0.19	0.17	0.43	0.22
D	0.20	0.19	0.39	0.19
E	0.13	0.18	0.31	0.27
F	0.16	0.17	0.33	0.24
G	0.19	0.25	0.44	0.28
	<hr/>	<hr/>	<hr/>	<hr/>
Avg.	0.18	0.20	0.38	0.26

- (a) Value obtained by adding the results of counting the aluminum rings and brass planchets individually.
- (b) Value obtained by counting the aluminum rings and brass planchet as an assembly.

ring. In actuality, these results constitute only a receiving inspection, since none of this type of data are directly usable in actual blank corrections. This is because a good portion of the planchet assembly's activity is further shielded by the mounted sample.

Table 2 shows activity of several batches of planchets. The variation of activity from batch to batch is not large. A bad batch was received in October 1965 with an activity of 0.4 cpm per planchet assembly. Subsequently, a pre-production activity check was made in 1965 to insure that contamination-free material was used.

The data in Table 2 compares activity of ring and disc assemblies with or without filter paper and Mylar film. The filter and Mylar do not add any activity to the mounts. This leaves the last and most significant, and also the most difficult to determine source, the mounted carrier compound from the chemistry procedure. It would indeed be interesting to isolate this source of blank activity from all other sources and further to identify the relative abundances of isotopic and non-isotopic, elemental and non-elemental components. The blank activity is much too low to be subjected to normal qualitative analysis such as absorption curves, or gamma-ray spectrometry. A general idea of the energy of the radiation may be gained by absorptions, and these usually indicate the energies associated with the long-lived components of fission products. Identification of an isotopic or elemental species is not possible by our present analytical instrumental techniques. If the major component of the blank from this source is not due to the specific carrier element, chemical techniques can be applied to decontaminate the carrier from this activity.

CERIUM BLANK AS AN EXAMPLE

The cerium blank at Tracerlab will be discussed in detail. The cerium blank is not as perfect as some of the other elements and therefore presents some of the problems which might be encountered.

Carrier solution stocks are routinely checked for radioactivity every time a solution is standardized. Some carrier salts, however, have been found to contain radioactive impurities and are always purified before use. Cerium, lanthanum, zirconium, and beryllium are always purified at TLW. Table 3 shows measurements made on cerium carrier before and after

TABLE 2. BETA ACTIVITY IN SAMPLE MOUNT ASSEMBLIES.

Date	Manufacturer (Coded)	Metal Components Only cpm	Complete with Filter Paper Mylar Cover cpm
1960-61 ^(a)	Unknown	0.14 ± 0.02 (86)	---
Oct. '63	Unknown	---	0.27 ± 0.06 (8)
June '64	Unknown	---	0.40 ± 0.05 (6)
Oct. '65	VDV	0.26 ± 0.04 (4)	0.23 ± 0.05 (6)
July '66	KM	0.16 ± 0.05 (5)	0.17 ± 0.05 (5)

(a) Detailed data given in HASL-115, p. 387, Oct. 1961.

() Number of planchets counted.

TABLE 3. PURIFICATION OF CERIUM CARRIER.

Carrier-Material $Ce(NO_3)_3 \cdot 6 H_2O$ (99.9%, Lindsay).

Solution ^(a) Stock	Procedure	Th ²³² (b) dpm/10 mg Ce	Beta Activity cpm/10 mg Ce	Beta thru Cu Absorber ^(d) cpm/10 mg Ce
D-6	Unpurified Starting Material	3.46 ± 0.07 ^(c) (2) (0.094 wt. % as equiv. Th (NO ₃) ₄)	1.7 ± 0.3 (6)	0.3 ± 0.1 (6)
D-4	Purification by BaSO ₄ and Zr(10 ₄) ₄ scavenges	0.25 ± 0.03 (4)	0.1 ± 0.1 (4)	0.0 (4)
D-5	Purified by Anion Exchange from HNO ₃	0.09 ± 0.03 (6)	0.2 ± 0.1 (8)	0.0 (4)

(a) All carrier material from same stock for each analysis.

(b) Th²³² analysis using Th²³⁰ tracer, radiochemical purification and alpha spectroscopy.(c) About 1.5 dpm Th²²⁸ also observed.

(d) 0.3 cpm planchet activity subtracted.

() Number planchets counted.

purification. Radiochemical analysis and alpha spectrometry for thorium showed 3.5 dpm Th²³² and 1.5 dpm Th²²⁸ in the equivalent to a mounted sample of 10 mg Ce. Also, a corresponding 1.7 cpm of beta activity from the thorium daughters were found. This amount of Th²³² corresponds to 0.094 weight % in the original compound, a value within the range of values tabulated in DeVoe's monograph "Radioactive Contamination of Materials Used in Scientific Research"⁽⁶⁾. However, he makes no mention of the Th²²⁸ content of cerium.

Chemical purification of the carrier reduces the contamination considerably. The ion exchange method is preferred, and the activity is reduced to less than 0.1 dpm alpha and 0.2 cpm beta. Table 4 shows the beta activity in eight cerium carrier stocks purified since 1964.

In the case of cerium, activity from the thorium decay chain probably would not perturb a radio-cerium analysis, since the impurities would be removed in the purification. However, addition of this extraneous activity to a sample undergoing analysis for many radio-elements is undesirable. Often thorium alpha emitters are to be determined. Cerium carrier is often used in purification procedures for scavenging or hold-back of other activities. If added in one of the final steps in a procedure, the impurities might appear in the mounted sample.

No components of the chemical process other than carrier are individually checked for radioactivity. It is not economically feasible to check all. A reasonable compromise is made by checking activity in the processed sample.

The blank program in action is thus reduced to having technicians perform chemical and counting procedures precisely in the manner the samples are processed but without the sample itself. Where possible, a sample placebo, unexposed IPC or Microsorban, other collection media, or distilled water is used to more exactly simulate the conditions encountered during sample processing.

The blank value for an element is obtained at the cpm level initially. Table 5 shows the activity measured in cerium blanks over the past few years. Some of these samples were processed with relatively high levels of activity in the real samples processed alongside of them. In the case of one blank in 1965-66, the observed blank activities of 4.8 and 3.3, in reality, came from companion samples and pointed out the need for correction in technique of the technician.

TABLE 4. BETA ACTIVITY IN PURIFIED CERIUM CARRIER.

Solution Stock	Date Prepared	Beta Activity (a) cpm/2 ml	
E-7	April 1967	0.1	
E-6-A	Dec. 1966	0.7	Recheck of Stock E-6
E-6	Sept. 1966	0.2	
E-5-A	May 1966	0.5	Recheck of Stock E-5
E-5	March 1966	0.0	
E-4	Jan. 1966	0.1	
E-3	Sept. 1965	0.1	
E-2	July 1965	1.0	
E-1	March 1965	0.6	
D-5-B	Dec. 1964	0.3	

(a) 0.2 cpm or 0.3 cpm planchet activity subtracted as applicable

Stock D and E - American Potash and Chemical Corp., Rare Earth (Lindsay) Division, $\text{Ce}(\text{NO}_3)_3 \cdot 6 \text{H}_2\text{O}$, 99.9%. Different bottles of salts designated by stock D and E.

TABLE 5. BETA ACTIVITY IN PROCESSED CERIUM BLANKS.

Beta cpm Observed on Sample (a)							
No Absorber				Cu (b) Absorber Cover			
63-64	64-65	65-66	66-67	63-64	64-65	65-66	66-67
0.4	0.6	1.7	0.2	0.0	-0.1	-0.2	-0.1
0.5	0.4	1.8	0.1	0.1	-0.2	-0.2	-0.2
0.6	0.4	0.2	0.8		-0.1	-0.2	0.3
0.6		0.2	0.3			-0.2	0.0
1.5		0.7				0.0	
2.0		0.8				-0.1	
		0.4				-0.1	
		0.1				-0.1	
		-0.1				-0.4	
		(4.8)(c)				(3.3)(c)	
		-0.2				-0.3	
		0.0				-0.2	

(a) Counter background ≈ 0.5 cpm; efficiencies: Ce^{141} -0.38, $\text{Ce}^{144}(\text{Pr}^{144})$ -0.39. Planchet activity subtracted, 0.2 or 0.3 cpm as applicable.

(b) Cu absorber - 280 mg/cm^2 .

(c) High blank traced to relatively high levels of activity in companion samples processed simultaneously.

In other cases, unusually high blank results from a particular technician result in the discarding and restocking of his reagents and a rerun of the blank trial. If this is not successful, then correlative evidence with results of other technicians is sought to track the source of the blank activity to a master reagent stock supply. Of course, if the results of several or all technicians blank runs are high, then steps are necessary to improve the decontamination procedure or locate and remove the source of contamination.

When discussing blanks on an interlaboratory level, it is usually desirable to discuss the blank data in terms of equivalent dpm. Table 6 lists results on a few blank ceriums from a low-level radiochemistry program. They were reported to HASL and are applicable to the High Altitude Balloon Sampling Program (HABS) and the Surface Air Sampling Program (SASP). These are the uncorrected blanks and do not represent dpm that would be reported on an actual sample. Also, they do not mean that this is really Ce^{141} and Ce^{144} dpm but equivalent dpm which would be reported if the blank correction was to be ignored.

The blank control program for a given element is considered successful only when the activity level is both low and reproducible. At this point, then, an "average blank" and its error is calculated in terms of observed cpm. This blank, in cpm, is then directly subtracted from the observed sample count rate and contains the partially absorbed planchet blank. The correction is done on the cpm basis rather than on the dpm basis because it is universally applicable.

One test of the success of the blank program for cerium would be a result equal to zero on samples which contain no measurable cerium activity. HASL routinely submits coded blank samples to Tracerlab along with High Altitude Balloon Filters (HABS). They later publish the results in their quarterly reports. Table 7 shows Ce^{144} results on 14 samples for Ce published by HASL (7), (8), (9). All but two samples are listed as not detected.

BLANKS FOR OTHER ELEMENTS

The results for various phases of the blank program for the isotopes $\text{Sr}^{89, 90}$, Ba^{140} , Cs^{137} , Tl^{204} , and Pb^{210} will be given in less detail.

TABLE 6. TOTAL EQUIVALENT DPM^(a) IN CERIUM BLANKS
HABS AND SASP PROGRAM 1966-67.

equiv. Ce ¹⁴¹ dpm and error ^(b)	equiv. Ce ¹⁴⁴ dpm and error ^(b)	Radiochemical Yield %
0.2 B	0.5 A	79
0.0 C	0.6 A	83
0.6 A	0.2 C	79
0.5 B	0.5 A	83
1.0 A	0.6 A	73
0.5 C	0.3 C	84
0.5 C	0.3 C	84
---	0.6 A	83
---	0.3 A	80
---	0.1 B	64
---	0.1 B	79
---	0.1 C	77
---	0.4 B	88

(a) Equivalent dpm includes all observed cpm over background.

(b) Error: A 20-50%; B 51-100%; C 101-200%.

TABLE 7. CODED BLANK SAMPLES ANALYZED IN THE
HASL, HABS, PROGRAM^(a)

HASL Sample Number	Report Date	Ce ¹⁴⁴ dpm and error ^(b)
2323	5/66	ND
2376	7/66	ND
2384	8/66	ND
2100	1/66	ND
2206	2/66	ND
2207	3/66	ND
2281	4/66	0.4 B
2323	5/66	ND
2376	7/66	ND
2384	7/66	ND
2466	9/66	ND
2463	10/66	ND
2495	11/66	2.4 B ^(c)
2549	12/66	ND

(a) Results have been published in quarterly publications, HASL-174, 181, and 182.

(b) Error: A 20-50%; B 51-100%, C >200%; ND = not detected.

(c) #2495 - 2.4 dpm includes 25% decay correction.

Table 8 shows the beta activity in the carrier solutions. Each pair of values represents duplicate measurements at the time the carrier was standardized or restandardized. Cesium carrier from three bottles of salts up through late 1966 has the highest activity. In late 1966 a new bottle of CsNO_3 was started and the last two sets of duplicate checks show no measurable activity in the carrier.

Table 9 shows the beta activity in processed Ba and Cs blanks. These blanks have been obtained from different separation schemes, with samples involving count rates above the low-level range. They should not be averaged. Often, evaluation of high blanks in light of actual sample activities shows that the integrity of actual samples are not in question since they are so much hotter than the blanks. Rather, the potential of future lower-level samples must be determined. Four of the Cs^{137} blanks above 0.6 cpm are most likely due to slight contamination from relatively high-level samples they were simultaneously processed with. As in the case of the high cerium blanks, such outliers are investigated. A follow-on blank is processed if contamination of a chemist's bench or equipment is suspected.

Table 10 shows the beta activity in processed Tl and Pb blanks. Several high values for Tl show the effect of an unsuspected bottle of contaminated reagent. Used in small amounts at a time, portions of the solution reached working stock bottles at chemists' benches in an erratic pattern. The Tl^{204} blank values in Table 10 (column '63-64) show an erratic pattern as different chemists became involved. The contamination source was found to be a bottle of La scavenge carrier, with about 3 beta cpm/ml contamination. Lanthanum is added two times (total added approximately 1/2 ml) during the purification procedure.

The contamination apparently carried through the last steps of the purification procedure because steps which would have removed it had already been completed. Lanthanum compounds are known to be contaminated with heavy element activity, reportedly, ⁽¹⁰⁾ 22-year Ac^{227} and Th^{232} . Apparently, the clean-up of this particular La solution at Tracerlab was not completely effective.

TABLE 8. BETA ACTIVITY IN CARRIER SOLUTIONS.
 Average Beta cpm^(a) of Duplicate Checks on Each Carrier
 Stock Standardized.

Year	Sr Carrier cpm/20 mg	Ba Carrier cpm/20 mg	Cs Carrier cpm/20 mg	Tl Carrier cpm/20 mg	Pb Carrier cpm/20 mg
1963		0.0			
1964	0.0			0.1 0.0	0.0
1965	0.0 0.0	0.0 0.0	0.4 0.4 0.2 0.2 0.2 0.2	0.0 0.0 0.0	0.3
1966	0.0 0.0 0.0	0.0 0.0	0.2 0.2 0.2 0.2 0.0	0.0 0.0 0.1	0.0
1967		0.0 0.0	0.0 0.0	0.1 0.1	0.1 0.1 0.0

(a) Corrected for appropriate planchet blank of either 0.2 or 0.3 cpm.

TABLE 9. BETA ACTIVITY IN PROCESSED Ba and Cs BLANKS.

Beta cpm Observed on Sample ^(a)						
Ba ¹⁴⁰				Cs ¹³⁷		
63-64	64-65	65-66	66-67	64-65	65-66	66-67
0.1	-0.1	-0.1	-0.1	(6.6) ^(b)	0.0	0.0
0.0	-0.1	-0.3	0.0	0.2	0.1	0.1
0.0	0.0	0.0	0.3	0.1	0.0	0.7
0.0	-0.1	-0.1	0.1	(2.4) ^(b)	0.1	(34.) ^(b)
0.1	-0.1			(0.9) ^(b)	0.6	0.2
0.1	-0.1			0.2	0.0	0.1
				0.0	-0.1	
				0.3	0.0	

- (a) Counter background \approx 0.5 cpm; efficiencies: Ba¹⁴⁰-0.44, Cs¹³⁷-0.33. Planchet activity subtracted, 0.2 or 0.3 cpm as applicable.
- (b) Blanks greater than 0.6 cpm probably due to slight contamination from relatively high level sample they were simultaneously processed with.

TABLE 10. BETA ACTIVITY IN PROCESSED Tl AND Pb BLANKS.

Beta cpm Observed on Sample ^(a)					
Tl ²⁰⁴				Pb ²¹⁰ (b)	
63-64	64-65	65-66	66-67	65-66	66-67
0.8	(1.7) ^(c)	0.0	0.0	0.0	0.2
0.7	(1.7) ^(c)	-0.1	0.2	0.0	0.1
0.9	0.3	0.4	0.0	-0.1	0.0
1.5	-0.1	0.6	-0.1	0.0	-0.1
1.6	0.3	0.0	0.2	0.0	0.1
0.7	0.3	0.2	0.4	0.0	0.0
0.6	-0.1	0.7	0.3	0.0	(1.4) ^(d)
0.4	0.6	0.0	0.2	0.0	(1.0) ^(d)
0.2	0.1	-0.1	0.3	-0.1	0.2
	0.7	-0.1	0.3	0.2	0.2
	1.0	-0.1		-0.1	
		0.1		-0.1	

- (a) Counter background \approx 0.5 cpm; efficiencies: Tl²⁰⁴-0.45, Pb²¹⁰(Bi²¹⁰)-0.36. Planchet activity subtracted, 0.2 or 0.3 cpm as applicable.
- (b) Counted through 6.5 mg/cm² Al cap absorber.
- (c) Beta activity found in La scavenge carrier (\approx 3 cpm/ml) used in Tl processing.
- (d) Not Pb²¹⁰, no growth of Bi²¹⁰ observed.

Table 11 lists total equivalent dpm in Sr, Cs, and Pb. They are a portion of the internal blanks processed under the low-level HASL programs. This data includes all total activity observed over counter background converted to an equivalent dpm for each isotope, and it represents a "worst case" type of listing. Correction of actual samples is made only at the cpm level.

Results of blank samples submitted as real samples by HASL are listed in Table 12. The highest blanks listed are one Sr⁹⁰ at 3.0 dpm and the cerium sample previously discussed for Table 7. Two Pm¹⁴⁷ blanks are 1.8 dpm.

Pm analysis system used at TLW involves use of Pm^{143, 4} tracer for yielding. The tracer adds to the intrinsic beta background and the detection limit for Pm¹⁴⁷ is estimated at 1.3 ± 1.3 dpm.

Another method of assessing the validity of low-level analyses is by means of a dilution experiment. Decreasing amounts of activity are analyzed and compared with the expected value. The previously published⁽¹¹⁾ results of a dilution experiment performed on Sr⁹⁰ at TLW in 1960 are shown in Fig. 3. Eighteen samples ranging in count rate from 1200 to approximately 0.3 cpm were measured. The results show that the accuracy of the analysis is better than $\pm 10\%$ down to the 0.5 cpm level, after which the accuracy rapidly decreases. The measured counting precision below 1 cpm does not reflect the measured accuracy.

DISCUSSION

Analysis of many elements from a single sample increases the number of potential contamination sources. The blank program not only points out unexpected sources of contamination but provides a base line for the purpose of correcting low counting samples. Each analytical scheme is separately evaluated for blanks and the analytical scheme is usually specific to a given project.

The control can vary. Outside of the low-level lab, on higher-level samples an occasional blank of 1 or 2 cpm is acceptable. However, overall blank level in that laboratory must still be held at only a few tenths cpm above background.

TABLE 11. TOTAL EQUIVALENT DPM^(a) IN Sr, Cs, AND Pb.
HABS AND SASP PROGRAM 1966-67

equiv. Sr ⁸⁹ dpm and error ^(b)	equiv. Sr ⁹⁰ dpm and error ^(b)	equiv. Cs ¹³⁷ dpm and error ^(b)	equiv. Pb ²¹⁰ dpm and error ^(b)
0.6 A	1.0 A	1.4 A	0.7 A
0.0 C	0.6 A	1.4 A	0.4 A
0.7 A	0.6 A	1.5 A	0.6 A
0.4 A	0.3 B	1.4 A	0.8 A
	0.6 A	0.9 A	0.1 B
	0.4 A		0.0 B
	0.4 A		0.8 B
	0.3 A		
	0.4 A		
	0.9 A		
	1.2 A		
	0.5 A		

(a) Equivalent dpm includes all observed cpm over counter background.

(b) Error: A 20-50%; B 51-100%; C >200%.

TABLE 12. CODED BLANK SAMPLES ANALYZED IN THE HASL, HABS, PROGRAM^(a),
May-Dec '66.

HASL Sample #	Sr ⁸⁹ dpm (b)	Sr ⁹⁰ dpm (b)	Zr ⁹⁵ dpm (b)	Cs ¹³⁷ dpm (b)	Ce ¹⁴¹ dpm (b)	Ce ¹⁴⁴ dpm (b)	Pm ¹⁴⁷ dpm (b)	Pb ²¹⁰ dpm (b)
2323		ND		0.6 A		ND	1.8 B	ND
2376	ND	0.3 B				ND	ND	ND
2384	ND	1.6 B				ND		
2100		0.6 B				ND		
2206		0.8		ND		ND	ND	
2207		3.0				ND		0.5 A
2281		0.2 B		1.2		0.4 B		0.2 B
2323	ND	ND		0.6 A		ND	1.8 B	ND
2376	ND	0.3 B			ND	ND	ND	ND
2384	ND	1.6			ND	ND		
2466		0.9	ND		ND	ND		0.3 B
2463	ND	ND	ND		ND	ND	ND	
2495	ND	0.9 B	ND		ND	2.4 B		ND
2549	ND	1.0 B	ND		ND	ND		
2207								0.5 A
2281								0.2 B
2323								0.3 C
2376								0.2 C
2466								0.3 B

(a) Results have been published in quarterly publications, HASL-174, 181, and 182.

(b) Error: A 20-50%; B 51-100%; C >200%. C error only given; counting error >100%.

ND = not detected.

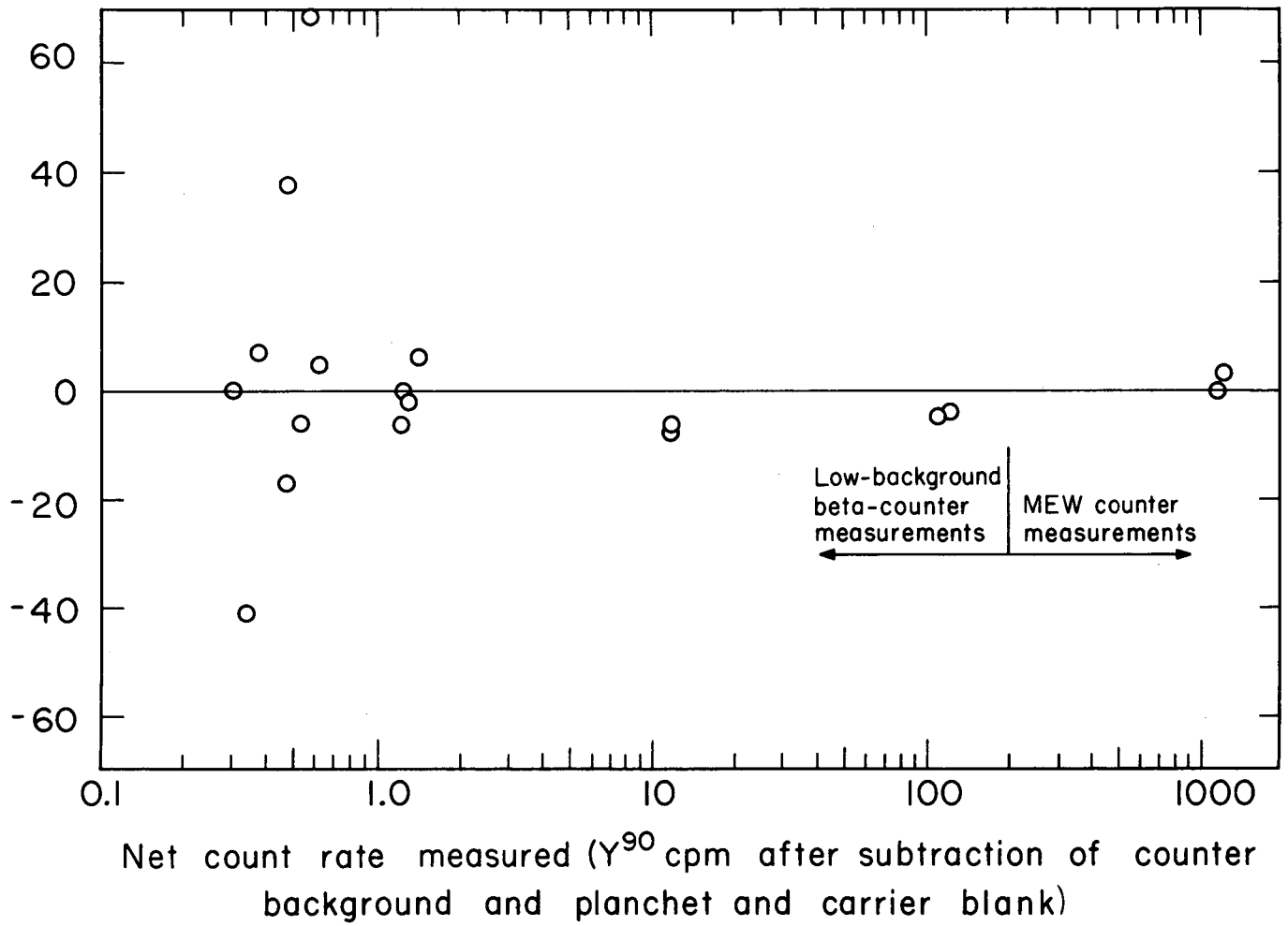


Fig. 3. Analysis of Sr^{90} at decreasing activity levels (Ref. 14). Samples obtained by successive dilutions of NBS standard No. 4919-B.

The activity level expected in each project determines if it should be carried out in Tracerlab's main chemistry lab, the intermediate lab, or the low-level lab.

Sources of blank activity have been found to be the sample-mounting material, metal carrier solutions, reagent solutions, and occasionally, accidental contamination. In one case, an unexpected potential contaminator was introduced into the low-level lab in the form of a hidden spike. A customer included a 50,000-dpm Sr^{90} spike in a shipment of samples for bioassay. Constant surveillance reduces the total laboratory blank to 0.2 - 0.3 cpm, of which most is due to the planchet assembly.

Extension of the sensitivity can be made using Nylon planchets which have 0.0 to 0.02 cpm background. Recalibration and preparation of precipitate curves for a large number of isotopes would be necessary at Tracerlab. The gain in figure of merit would not be large as the counting efficiency is less on Nylon. Thus, recalibration for Nylon is only being carried out for selected isotopes. Small volume, low background (0.03 to 0.08 cpm) beta counters are being used at Scripps by Somayajulu and co-workers⁽¹²⁾. Use of such counters to handle the lowest level of samples is one of the projects underway at Tracerlab.

REFERENCES

1. T. T. Sugihara, Low-level Radiochemical Separations, Publication 3103, National Academy of Sciences - National Research Council, Washington, D. C. 1961, p. 3.
2. V. T. Bowen and T. T. Sugihara, Oceanographic Implications of Radioactive Fall-out Distributions in the Atlantic Ocean: From 20°N to 25°S, from 1957 to 1961, Journal of Marine Research 23, 123 (1965).
3. G. G. Rocco and W. S. Broecker, The Vertical Distribution of Cesium-137 and Strontium-90 in the Oceans, J. Geophys. Res. 68, 4501-4512 (1963).
4. W. S. Broecker, E. R. Bonebakker, and G. G. Rocco, The Vertical Distribution of Cesium 137 and Strontium 90 in the Oceans, 2 J. Geophys. Res. 71, 1999 (1966).
5. T. T. Sugihara, op. cit., p 4.
6. J. R. De Voe, Radioactive Contamination of Materials Used in Scientific Research, Publication 895, National Academy of Sciences - National Research Council, Washington, D. C. 1961, p. 70.
7. HASL Quarterly
HASL-174, Jan. 1967.
8. HASL Quarterly Summary Report
HASL-181, April 1967.
9. HASL Quarterly Summary Report
HASL-182, July 1967.
10. J. R. De Voe, op. cit., p. 13.
11. R. Wessman and L. Leventhal, Quality Control at Tracerlab,
HASL-115, Oct. 1961.
12. B. L. K. Somayajulu and co-workers at Scripps Institute of Oceanography,
La Jolla, Calif.

BIOASSAY CORRELATION WITH BREATHING-ZONE SAMPLING

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Most bioassay-air-sampling correlation studies have been unsuccessful. Steep work-area concentration gradients make fixed-station air sampling a poor indicator of personnel exposure. In addition, urinalysis gives an uncertain demonstration of insoluble aerosol exposure.

NUMEC nuclear fuel production operators have been wearing battery-powered lapel air samplers for 2 years. When an exposure occurs, the operator is removed from radiation work, and fecal, urine, and nasal contamination measurements are performed. We have found a correspondence between breathing-zone sampling and early-clearance fecal data. Neither urinalysis or fixed-station air sampling is a reliable measure of personnel exposure.

Other current work includes: air-sampling badge development, multifilter aerosol analyzer, and aerosol solubility studies.

Bioassay Correlation with Breathing Zone Sampling

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The Purpose of Air Sampling

Because airborne radioactivity is the chief hazard in a nuclear fuel plant, the health physicist expends much effort in air sampling. He expects his air sampling program to accomplish several ends, ^(1,2,3) such as

- (1) warning when high air levels exist,
- (2) measuring the effectiveness of control measures,
- (3) determining general room air levels, or
- (4) following contamination trends.

But the primary purpose of air sampling is

- (5) the determination of personal exposure.

Indeed, since federal regulations⁽⁴⁾ are written in terms of individual exposure, the law compels the health physicist to estimate the radioactivity which each radiation worker inhales.

The problem is interpreting air samples. It is very easy to take an air sample, but it is often very difficult to know what the results mean. A few years ago, Harry Schulte⁽⁵⁾ of Los Alamos made the point that "air sampling is not a science but an empiric art."

There are two basic kinds of air sampling: fixed-station, commonly called general area (GA) sampling, and breathing-zone (BZ) sampling. Industrial hygienists have been taking BZ samples for decades by holding sample heads close to the worker's nose.⁽⁶⁾ Recently, battery-powered lapel samplers have been developed which are worn by the worker.⁽⁷⁾

Most nuclear fuel facilities we know about either assign fixed-station concentrations to workers or depend on bioassay to estimate exposure. This paper will show that personal breathing-zone air samplers not only yield exposure data which correlates with bioassay results, but most often are the only accurate means of measuring individual exposure.

The Nature of Industrial Radioaerosol Exposure

Some vague ideas about the nature of industrial airborne activity have led to elaborate fixed-station air monitoring systems.⁽⁸⁾ One common notion is that air activity takes the form of a rather large cloud which disperses throughout a room until it settles out on horizontal surfaces. Another concept is that, except in accident conditions, air activity consists of isolated particles randomly distributed in the room air.

Our experience presents a completely different picture. We believe almost all industrial radioaerosol exposures are extremely localized in space. An example of what we mean is shown in figure 1. Here an analytical chemist, moving a contaminated beaker from one hood to another, is exposed to a small local cloud. He is wearing a lapel sampler which should detect the release. Nearby a fixed-station air sampler is operating. This sampler, if it detects the release at all, will surely underestimate the exposure. We have also found the mere withdrawal of contaminated gloved hands from a hood can expose workers.

Plutonium glove box releases especially follow steep concentration gradients. Figure 2 shows a typical example. The operator is coming out of the gloves to check his hands on the alpha meter. He will find them contaminated because a hole developed in the left box glove.

As soon as he is aware of the contamination, he will put on a respirator, cover the glove port, survey the area, and change the glove. But he will have

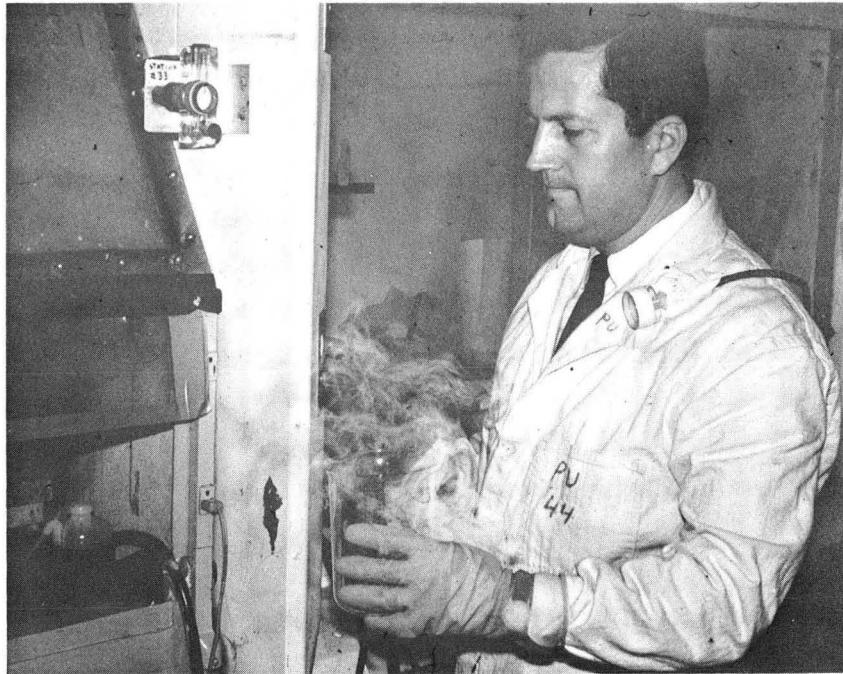


Fig. 1. Moving a "contaminated" beaker from one hood to another (aerosol cloud simulated by a smoke tube).

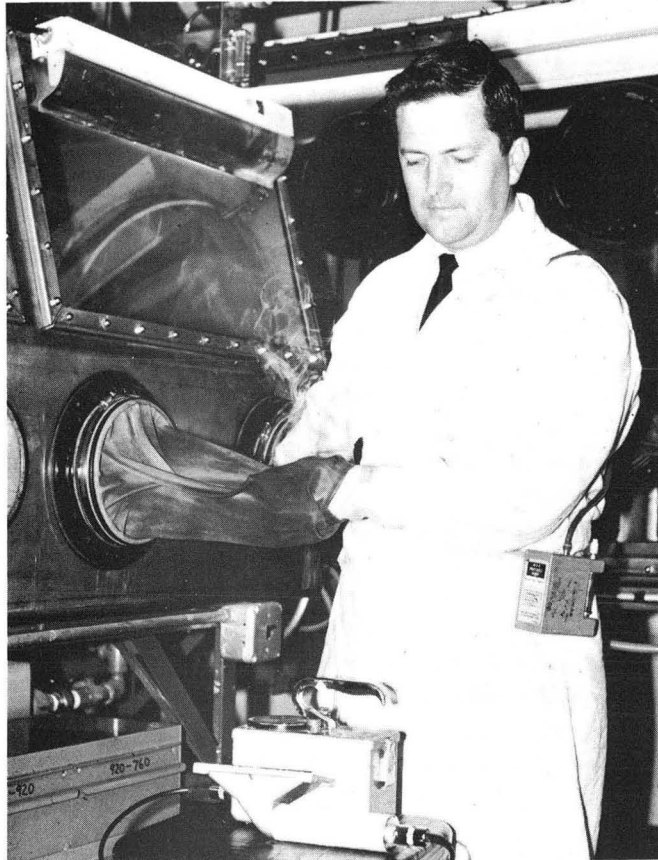


Fig. 2. Example of release of "contamination" (simulated by use of a smoke tube) when a glove is perforated.



Fig. 3. The typical radiation worker as viewed by the health physicist.

already been exposed by the small cloud generated when the glove was inverted. We have found that fixed-station samplers, like the one in the background, rarely detect these local releases. Also many times the releases are not accompanied by any appreciable floor fall-out contamination. This type of release is common in plutonium facilities. One major plant⁽⁹⁾ reports 900 glove failures per month.

Another common source of exposure is contaminated protective clothing. A worker wearing contaminated clothing generates a cloud or radioaerosol around himself. Figure 3 shows how the health physicist views the typical radiation worker. NUMEC experiments⁽¹⁰⁾ suggest that the function and design of protective clothing needs reevaluation.

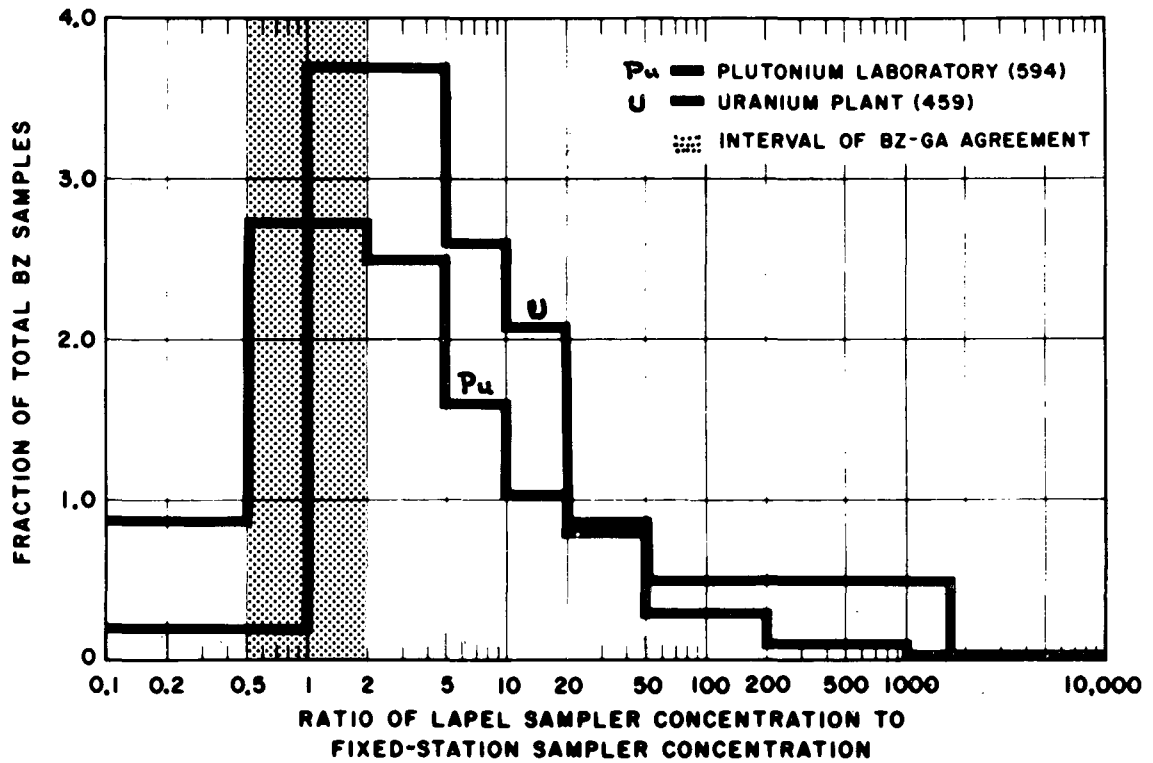
Although these photographs were staged with MSA smoke tubes, NUMEC experience with lapel samplers strongly suggests that uranium and plutonium aerosol clouds, although invisible, take exactly the same shape. The typical airborne release is a small cloud which quickly disperses to unmeasurable concentrations with relatively little surface contamination. In our experience floor contamination does not necessarily mean you have an air inhalation problem, but it surely means you had one earlier.

NUMEC Experience with Breathing-Zone Sampling

NUMEC uranium and plutonium workers have worn lapel samplers for two years. We find these samplers usually indicate higher concentrations than stationary samplers. Often the difference is orders of magnitude.

Figure 4 gives a two-year comparison of lapel samplers with fixed-station air samplers. It shows the lapel-to-fixed-station ratio distribution for 594 BZ samples at our plutonium laboratory and 459 at our uranium plant. The sample durations were for single shifts, an eight-hour workday. The fixed-station concentration is either the average of those in the worker's

COMPARISON OF LAPEL (BZ) TO FIXED-STATION (GA) AIR SAMPLING
NUMEC NUCLEAR FUEL FACILITIES
1966-1967



XBL 684-427

Fig. 4

area or the one closest to his work station. Actually we found little difference between fixed-station "breathing zone" samplers and those intended to cover general areas. The interval of general BZ - GA agreement (+ 100%, - 50%) covered 27% of the plutonium BZ samples and about 19% of uranium plant BZ samples. Notice that almost 9% of Pu BZs are less than 50% lower than the GAs. Sixty-four per cent of Pu BZ's exceeded the GA concentration by a factor of 2 or more, 23% by more than a factor of ten. The highest ratio we've ever detected was 9,870. Thirty-five per cent of uranium plant BZ concentrations exceeded 10 times the fixed station concentrations. While the median of these ratios is less than 10 for both plants, the very skewed distribution makes high-level exposures very important in computing the average exposures.

Figure 5 presents a clearer idea of how important personal samplers are when high-level exposures occur. The BZ/GA ratio data for all plutonium exposures exceeding 10 MPC for an eight-hour shift is plotted against the breathing-zone concentration.

The first impression from this graph is the extreme variability of the BZ/GA ratio for a given BZ concentration. As an example, for those BZs between 40 - 50 d/m/m³ the fixed station concentration varied from one half to one eight hundredth of the BZ concentration. It is difficult from this data to pick out a suitable factor (such as the UKAEA^(3,11,12) has done) by which to multiply the GA concentrations to obtain individual exposure.

Another thing to notice is the upward trend in the BZ/GA ratio as the BZ concentration increases. Basically this means the worse the problem is the wronger the fixed-station data.

We have drawn in the line where the fixed station air sample would indicate the soluble MPC_a for Plutonium. For all data above the line the GA was less than MPC. Only those GA's below the line even indicated that a

COMPARISON OF LAPEL (BZ) TO FIXED
STATION (GA) AIR SAMPLING
NUMEC PLUTONIUM LABORATORY
1966-1967

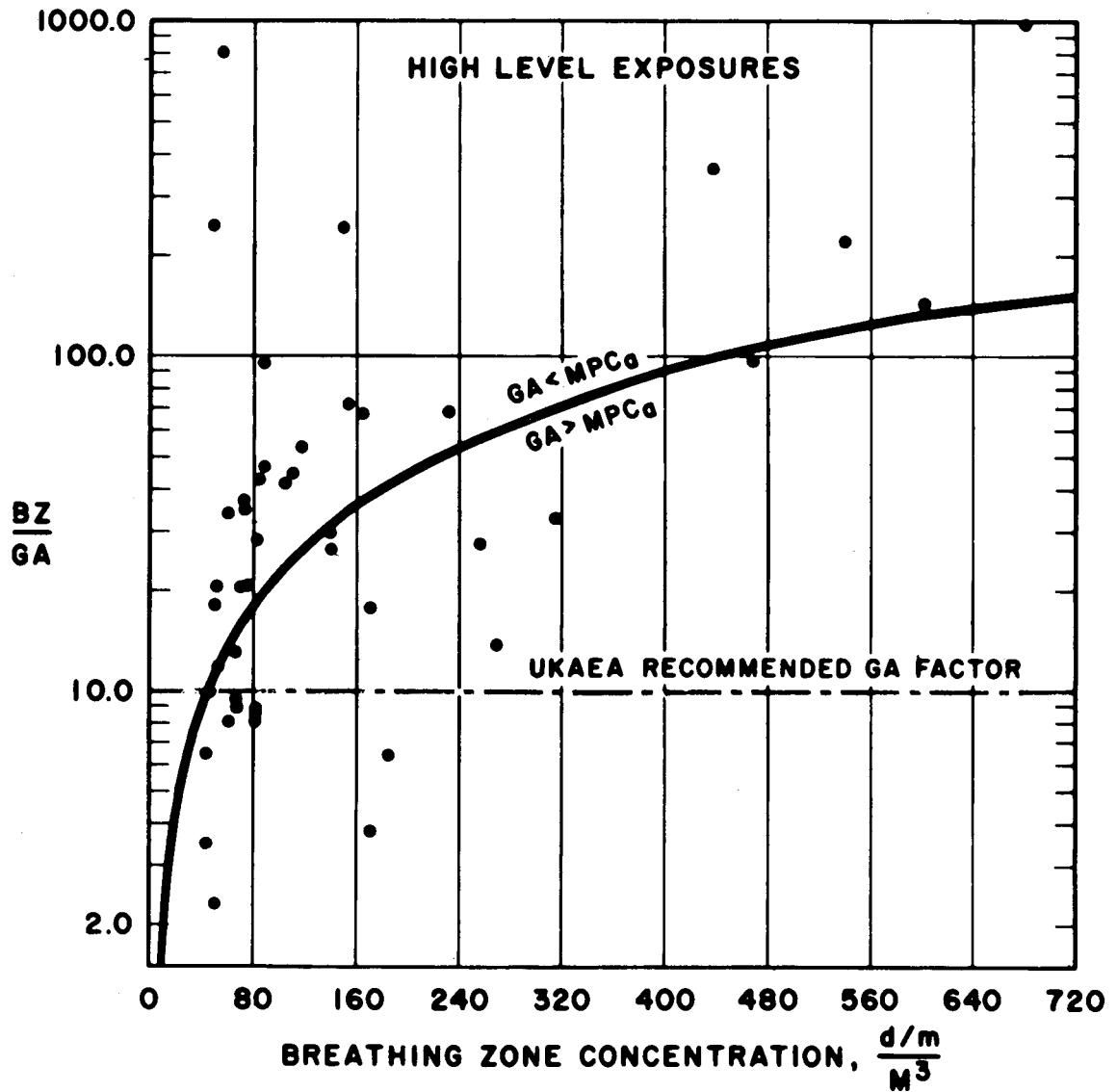


Fig. 5

hazard existed. This is an important point. Many industrial radioaerosol exposures are going unnoticed because the nuclear industry is depending on fixed-station air sampling.

The inability of stationary air samplers to indicate hazardous conditions in a uranium plant is shown on Figure 6. Nearly 73% of the time the GA sampling network failed to warn when greater than permissible exposure was occurring. Lest NUMEC be accused of not knowing how to place fixed air sample heads, please remember the location of the fixed air samplers in Figures 1 and 2. It has been our practice to place these sampling heads as close to the breathing zone in high-risk areas as is possible. There are 55 such samplers in our 20,000-ft.² plutonium laboratory and 34 in our 40,000-ft.² uranium plant.

It might be thought from reviewing this data that radiation control practices at NUMEC plants are not up to industry standards. We honestly don't think this is the case. In the first place we didn't have enough lapel samplers to continuously sample the breathing zone of all our workers. Consequently, we have chosen to use our available lapel samplers as diagnostic tools in areas where we feel that local "micro-climates" of radioaerosol may exist. Thus the high percentage of BZ samples above MPC_a is misleading. Our feeling is that it is the industry air sampling standards that are not adequate.

The Correspondence of Breathing-Zone Sampling with Early Fecal Clearance

The lapel sampler data would not be relevant, if it did not represent true exposure. For this reason, whenever an exposure occurred, the operator was removed from radiation work and both fecal and urine samples were collected. Figure 7 gives the correlation between BZ sampling and early fecal clearance for plutonium exposures. The eight cases shown were selected from almost a

HAZARD INDICABILITY
Lapel Samples (BZ) vs. Fixed Station Samples (GA)
 NUMEC URANIUM PLANT
 1966 - 67

CONDITION INDICATED	NUMBER RECORDED	FREQUENCY
BZ > MPC GA < MPC	300	.654
BZ > 10 MPC GA < MPC	33	.072
BZ < MPC GA < MPC	54	.118
BZ < MPC GA > MPC	2	.004
BZ > MPC GA > MPC	70	.152

Total BZ Samples 459
 Total GA > MPC 72
 Total BZ > MPC 403

Fig. 6

CORRELATION OF FECAL BIOASSAY WITH AIR SAMPLING

NUMEC PLUTONIUM LABORATORY
1966-1967

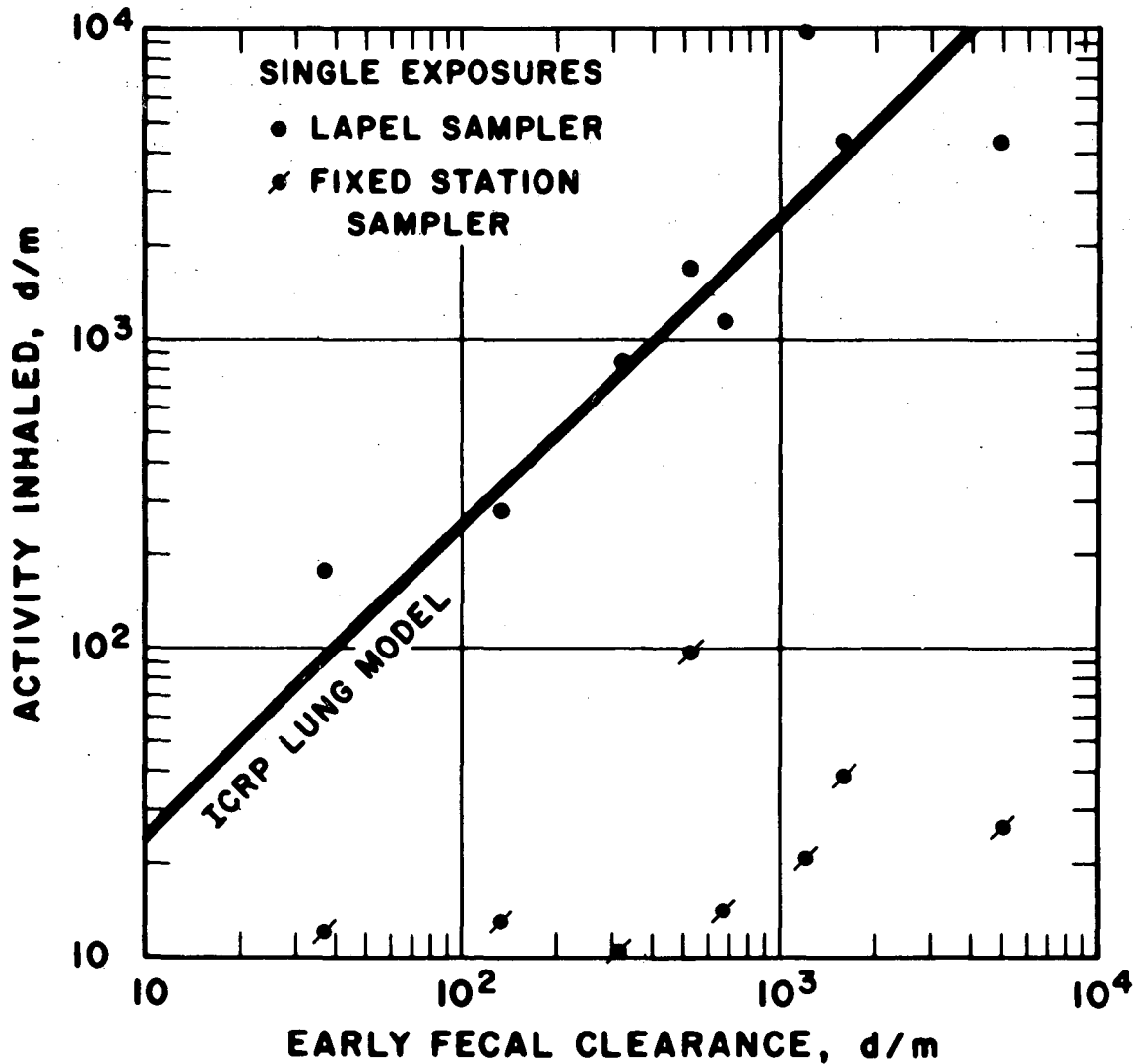


Fig. 7

UO₂ EXPOSURE AND BIOASSAY DATA
(W.S.) ACUTE EXPOSURE - May 22-23, 1967
NUMEC URANIUM PLANT

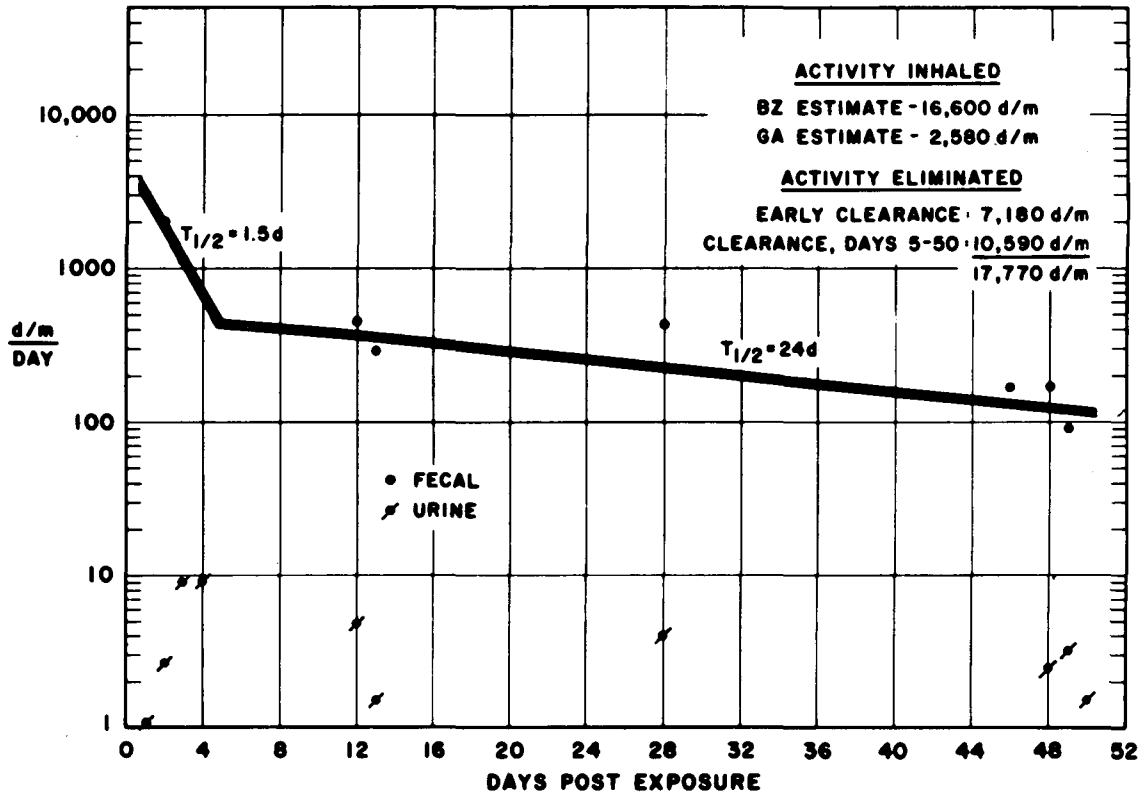


Fig. 8

hundred exposures because total fecal and urine data was available for the first seven days post exposure and because there was no recent prior exposure to complicate interpretation. Early fecal clearance was chosen as the exposure criteria because of earlier experience with urine and fecal sampling at our Plutonium laboratory.⁽¹³⁾

There is remarkable agreement between the proposed ICRP lung model⁽¹⁴⁾ and the lapel sampler data. The line represents expected 72-hour lung clearance for insoluble one-micron MAD PuO₂ particles.

The failure of fixed-station samplers could not be more graphic. We might also add that urine sampling did not demonstrate these exposures. Except for the highest plutonium exposures, no perturbation in urine excretion could be detected.

An example of inhalation inventory balancing is shown in Figure 8. A single UO₂ exposure was detected. We were fortunate enough to be able to follow this exposure unperturbed by subsequent exposures. Several items are interesting.

First, the activity inhaled as estimated by GA samples is less than the first day's fecal elimination. Secondly, the balance between the BZ estimate and that excreted over 50 days is noteworthy. Even the overage on excretion is consistent, since the individual had a history of recent exposure.

The early clearance half time of 1.5 days is interesting in that it agrees well with the lung model. It means that individuals must be removed from any possible exposure for at least seven days before fecal data can be used to estimate long-time lung burdens.

Fecal Sampling In a Uranium Plant

Figure 9 shows the importance of routine fecal sampling in a uranium plant. The excretion rates via feces and urine are plotted against each

URINE vs FECAL EXCRETION RATES
CHRONIC ^{234}U OXIDE EXPOSURES
NUMEC URANIUM PLANT

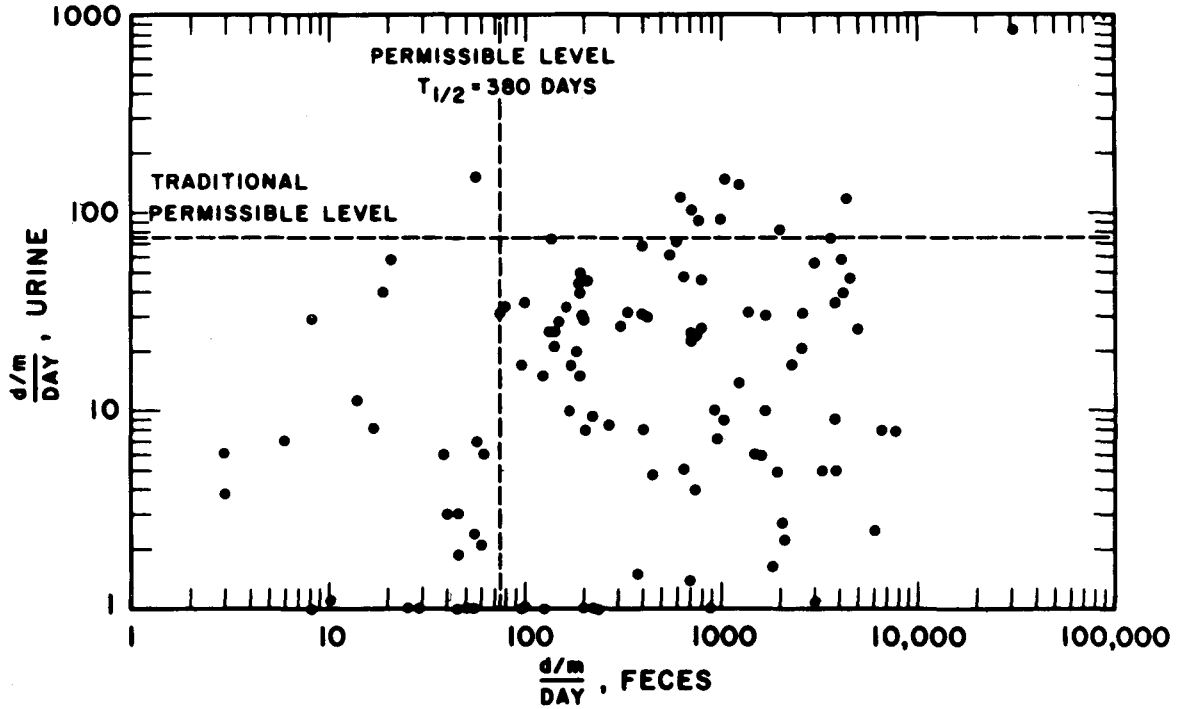


Fig. 9

other for all cases where the samples were collected the same day. The traditional permissible urine level is 75 d/m/day. The permissible fecal excretion rate was calculated to be 50 d/m/day, assuming the ICRP recommended 380-day half-time for chronic UO_2 exposures.

Many of the fecal data represent early clearance and are not necessarily unpermissible. However, it is obvious that urine data by itself gives a false impression of actual exposure. We were able to use this data to estimate the effectiveness of half-face respirators. We found that they are not very effective unless their use is closely supervised.

The Necessity for Breathing-Zone Sampling

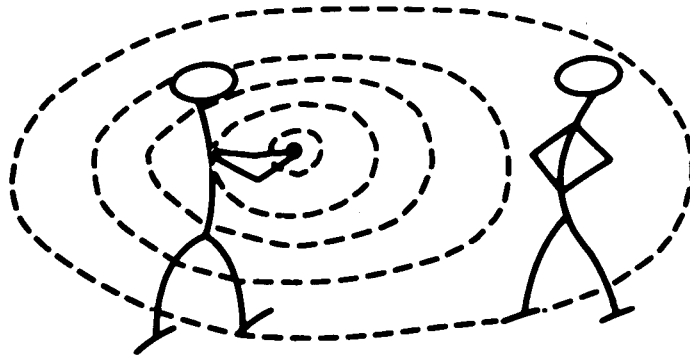
Many radiation protection workers feel that breathing-zone sampling is too fussy or is impossible. We believe it is absolutely necessary and can be easily done successfully with personal air samplers. Figure 10 demonstrates our concept. Nearly all radioaerosol sources are small; generally the worker's hands are the major aerosol generator. In static room air conditions concentrations will fall off with the inverse cube of the distance. If the distance from the hands is doubled, the concentration will be lower by a factor of eight. We have verified this by experiment.

The usual turbulent condition is more complicated. Still, the concentration gradient will be steep, and any fixed-station air sampler a few feet away will underestimate the man's exposure.

Breathing-zone sampling is usually recommended on the vague intuition that the closer the sampler is to the nose the better. NUMEC and UKAEA^(1,3,11,12,15) experience provides a more convincing basis for personal air sampling. The worker handling radioactive materials lives in a "micro-climate" which must be sampled if the health physicist is to detect industrial radioaerosol exposure.

NECESSITY FOR BZ SAMPLING

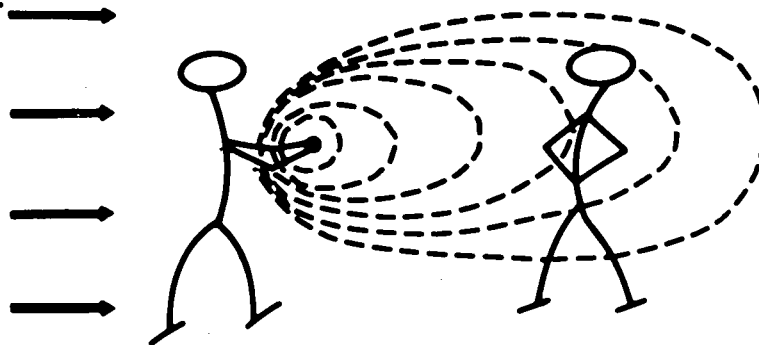
STATIC



$$C \propto \frac{1}{R^3}$$

TURBULENT

DRAFT →



$$C \propto \frac{1}{x^2} e^{-\frac{k}{x^2}}$$

Fig. 10

BIBLIOGRAPHY

1. B.A.J. Lister, Health Physics Aspects of Plutonium Handling, AERE-L151, 1964.
2. J. Pomarola et al., Assessment of Individual Risk During Airborne Contamination by Plutonium, IRPA Congress, Rome, 1966.
3. W.A. Langmead and D.T. O'Connor, The Significance of Radioactive Aerosol Measurements made in the Working Environment, IRPA Congress, Rome, 1966.
4. Code of Federal Regulations, Title 10, Part 20.103.
5. H.F. Schulte, The Contribution of Industrial Hygiene to the Protection of the Radiation Worker, 1966.
6. E.C. Hyatt and H.F. Schulte, Air Sampling Procedures in Evaluating Exposures to Uranium, HASL-58, 1958.
7. R.J. Sherwood and D.M.S. Greenbough, A Personal Air Sampler, Annual Occupational Hygiene 2:127, 1960.
8. M. Sanders, Innovations in Air Monitoring Techniques for Large Scale Programs, Y-KB-78, 1966.
9. E.A. Putzier, Plutonium Hazards and Accident Experiences, RFP-621, 1965.
10. E. Schnell and R. Crosby, Breathing Zone Air Levels as Caused by Contaminated Clothing, unpublished NUMEC report, 1966.
11. R.J. Sherwood, On the Interpretation of Air Sampling for Radioactive Particles, American Industrial Hygiene 27:2, pp 98-109, 1966.
12. R.T. Brunskill and S.T. Hermiston, The Detection and Measurement of Plutonium Airborne Contamination in Major Plutonium Facilities, IRPA Congress, Rome, 1966.
13. R. Caldwell, The Detection of Insoluble Alpha Emitters in the Lung, AEC Conference on Bioassay and Analytical Chemistry, Gatlingsburg, Tenn., 1966.
14. Task Group on Lung Dynamics, Deposition and Retention Models for Internal Dosimetry of the Human Respiratory Tract, Health Physics 12:173, 1966.
15. D.C. Fraser, Health Physics Problems Associated with the Production of Experimental Reactor Fuels Containing PuO₂, Health Physics 13:1133, 1967.

RADIOCHEMICAL DETERMINATION OF SULFUR-35

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ABSTRACT

A rapid, accurate method was developed to determine ^{35}S in Savannah River Plant waste streams. ^{35}S is partially purified by extracting other radionuclides into tri-isooctylamine from 2N HCl . ^{35}S is further purified by precipitating BaSO_4 from the aqueous phase, dissolving this precipitate in hot alkaline EDTA, and reprecipitating BaSO_4 .

Liquid scintillation counting is used for radioassay. A gelling agent, "Cab-O-Sil,"* suspends the BaSO_4 precipitate in the scintillation solution. Counting efficiency of ^{35}S as BaSO_4 is 58%.

The chemical recovery of ^{35}S from aqueous samples is about 90%; therefore, the overall efficiency of the procedure is about 52%. Decontamination factors for radionuclides likely to occur in plant waste streams are at least 10^3 .

* Registered trademark of Cabot Corporation, Boston, Mass.

INTRODUCTION

Population exposure is controlled by restricting the entry of radioactive materials into the environment. Effective restrictions require that the amount of individual radionuclides is known before release. A method was required to analyze for ^{35}S in effluent water from reactor operations. An important requirement for the method was that the analytical time be short.

^{35}S is one of the more difficult isotopes to determine. It decays by emission of a 0.168-MeV beta particle with no associated gamma ray. Consequently, it must be determined by beta counting a solid-free mount, or by some technique in which solids do not interfere.

A simple, rapid, and accurate method was developed to determine ^{35}S in reactor effluent water. The method consists of preliminary purification of the sample by liquid ion exchange using tri-isooctylamine, followed by two BaSO_4 precipitations. Liquid scintillation counting is used for radioassay.

EXPERIMENTAL

Procedure. 1. Adjust the sample to 2N in HCl .

2. Transfer 50 ml of sample to a 125-ml separating funnel, and add 50 ml of 10% v./v. tri-isooctylamine (TIOA) in xylene. Mix for 1 minute and drain the aqueous phase into a 100-ml centrifuge tube.

DP-MS-67-40

3. Add 25 mg of sulfate carrier, as NH_4HSO_4 , to the centrifuge tube and heat the sample in a boiling-water bath. Add 10 ml of 5% BaCl_2 , swirl, and let the sample stand in the hot water for 10 minutes. Centrifuge at 2,000 rpm for 10 minutes and discard the supernate.

4. Add 20 ml alkaline EDTA (0.25M EDTA in 1.0M NaOH) to precipitate and heat in a boiling-water bath until the precipitate is completely dissolved. Add glacial acetic acid dropwise until BaSO_4 reprecipitates (1); add 1 ml excess. Centrifuge and discard supernate. Wash the precipitate with distilled water, centrifuge, and discard the wash.

5. Add about 250 mg of "Cab-O-Sil" to the BaSO_4 precipitate, slurry with 5 ml of scintillation solution (2), and transfer to a 25-ml polyethylene counting vial. Wash the centrifuge tube with \approx 5-ml aliquots of scintillation solution until the vial is full. Shake the sample vigorously for 1 minute and refrigerate until counted in liquid scintillation spectrometer (Packard "Tri-Carb" Liquid Scintillation Spectrometer Model 4312).

DISCUSSION OF THE PROCEDURE

Solvent Extraction. A study was made to determine if ^{35}S could be analyzed by liquid ion exchange using TIOA. ^{35}S was extracted from various normalities of hydrochloric acid with 10% v./v. TIOA in xylene (Fig. 1). The per cent of the ^{35}S exchanged to TIOA is plotted vs. normality of the HCl.

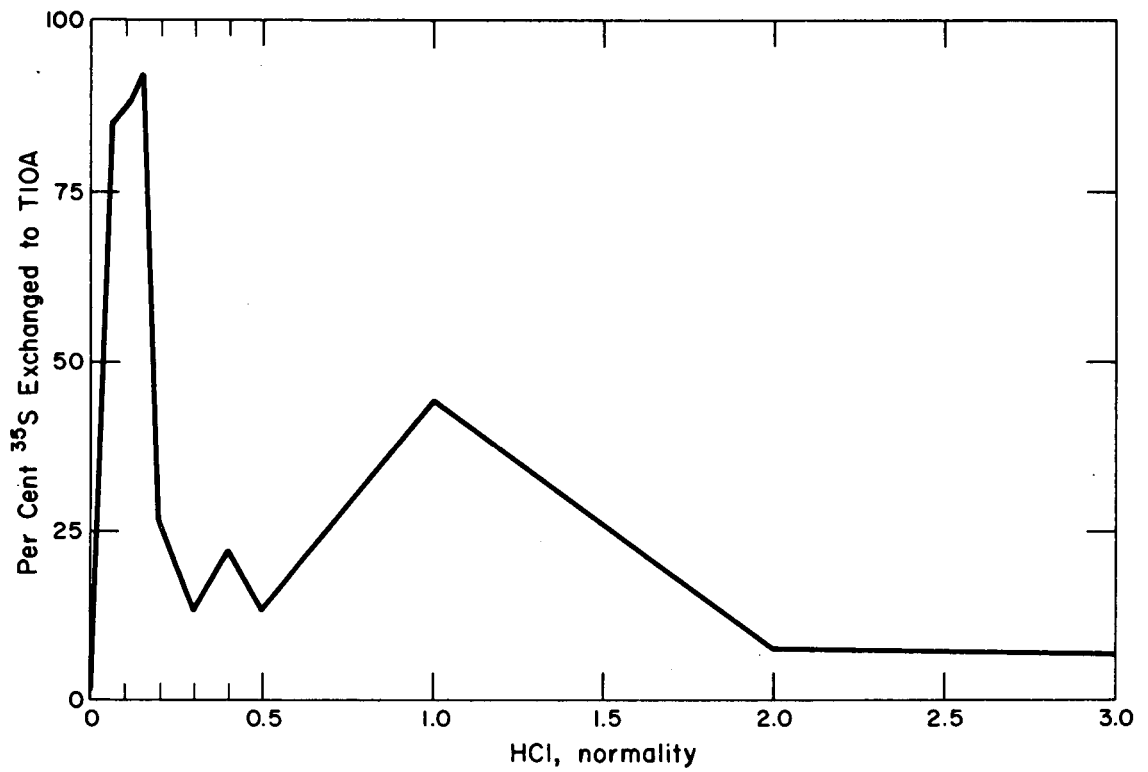


FIG. 1. EXTRACTION OF ^{35}S BY TIOA FROM VARIOUS NORMALITIES OF HCl.

DP-MS-67-40

Greater than 90% of the ^{35}S is extracted into TIOA from 0.15N HCl. The complex is readily destroyed with 4N HNO_3 , and the ^{35}S is transferred to the aqueous phase. The sample is solid-free when the HNO_3 is evaporated and mounted. ^{35}S added to 0.15N HCl was recovered very well, although decontamination from other radioisotopes was poor.

Recovery of ^{35}S from plant effluent stream samples was erratic and low. Nitric acid that is added to aqueous samples to prevent loss of radioactivity on the container walls during storage hinders the formation of the sulfur-TIOA complex. Nitric acid can be removed by evaporating the sample to dryness, but the residue is difficult to dissolve in 0.15N HCl.

Emulsions, sometimes formed between the dilute acid and TIOA/xylene, were reduced by adding toluene (3) to the TIOA/xylene.

Butler (4) has shown that many radioisotopes exchange to TIOA from 2N HCl. Figure 1 shows that less than 10% of the ^{35}S exchanges to TIOA from 2N HCl. Therefore, ^{35}S was partially purified by extracting other isotopes from 2N HCl with TIOA. Figure 2 shows the decontamination from one extraction with TIOA. Other isotopes removed by TIOA from 2N HCl are iron, technetium, silver, polonium, and most of the uranium.

Barium Sulfate Precipitation. ^{35}S is further purified by two BaSO_4 precipitations. The first precipitation is in the aqueous phase from the TIOA extraction step.

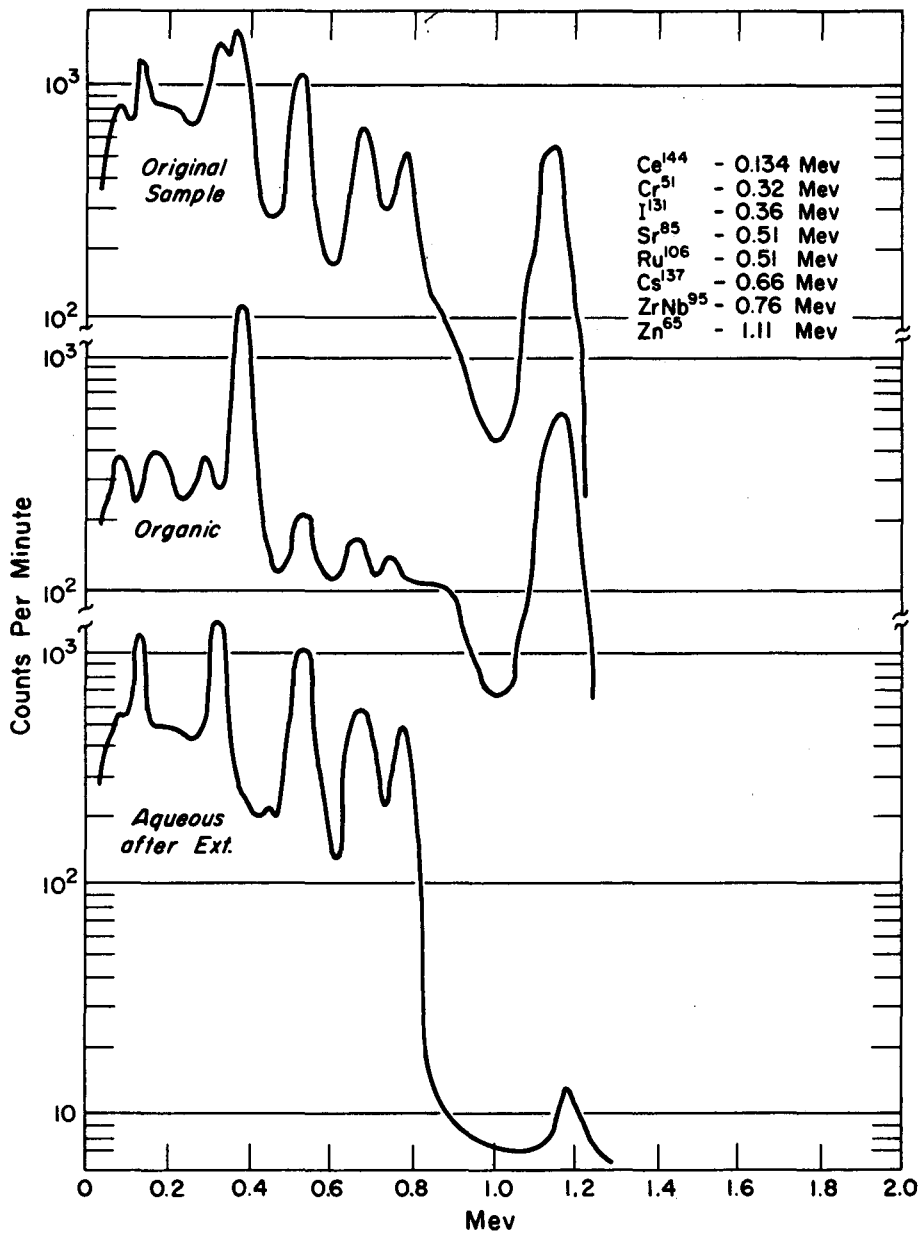


FIG. 2. DISTRIBUTION OF RADIONUCLIDES AFTER EXTRACTION WITH T10A.

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The first BaSO_4 precipitate is dissolved in 20 ml of alkaline EDTA (1) in a hot water bath. Acetic acid is added dropwise to reprecipitate BaSO_4 . The volume of the alkaline EDTA must be kept as small as possible, so that the precipitate will be dense enough to centrifuge. Figure 3 shows the purification obtained by BaSO_4 precipitations.

Liquid Scintillation Counting. The BaSO_4 precipitate is transferred to a 25-ml polyethylene counting vial containing scintillation solution and "Cab-O-Sil", a gelling agent, is used to suspend the precipitate in the scintillation solution. Liquid scintillation counting was chosen because it is more efficient than beta-proportional counting. Table I gives the counting efficiency of ^{35}S and shows that 60 mg of BaSO_4 reduces the counting efficiency of ^{35}S less in liquid scintillation counting than in beta proportional counting. The ^{35}S counting efficiency is determined with each batch of samples by counting a ^{35}S standard.

Table I. Counting Efficiency of ^{35}S

Method of Counting	Efficiency, Per cent	
	Solid-free	With 60 mg BaSO_4
Beta proportional	20	7
Liquid scintillation	72	58

The same scintillation solution and spectrometer settings used for counting tritium (2) at SRP are conveniently used for counting ^{35}S .

Effect of Cab-O-Sil. A shake-count test with $\text{Ba}^{35}\text{SO}_4$ showed that when "Cab-O-Sil" was added the counting efficiency

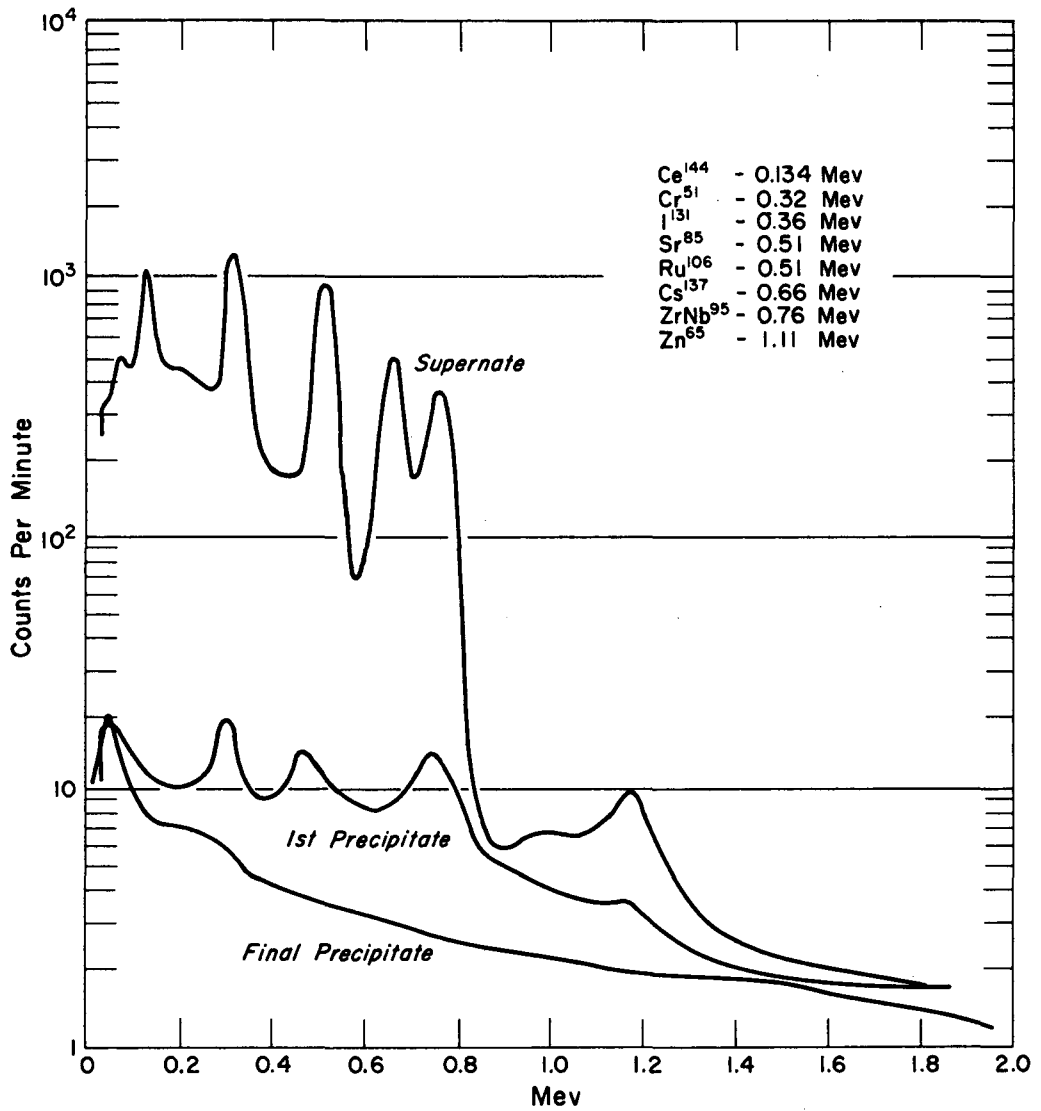


FIG. 3. PURIFICATION ACHIEVED BY BaSO₄ PRECIPITATIONS.

DP-MS-67-40

of $\text{Ba}^{35}\text{SO}_4$ remained constant, whereas without "Cab-O-Sil" the count increased by a factor of 2 but then dropped rapidly until the count rate was almost the initial count (Figure 4). Both samples were initially shaken, cooled for 1 hour, and counted. After the initial count, they were shaken and counted repeatedly without further shaking.

Counting Statistics. Duplicate samples and spikes were analyzed each time. The background was determined with a blank scintillation solution. The concentration of ^{35}S is calculated by

$$A = \frac{C \left(\frac{S^2}{(D)(P)} \right)}{V},$$

where

- A = concentration of ^{35}S in dpm/ml,
- S = dpm of the ^{35}S standard,
- C = corrected counts per minute of the sample,
- D = corrected counts per minute of the ^{35}S standard added directly to the scintillation solution,
- P = corrected counts per minute of the ^{35}S standard analyzed by the procedure,
- V = volume of sample analyzed.

Recovery and Decontamination Factors. A series of samples spiked with ^{35}S was analyzed by the procedure. The overall recovery was $52.5 \pm 7.2\%$ at the 90% confidence level. The counting efficiency of $\text{Ba}^{35}\text{SO}_4$ is 58%; thus the chemical recovery is 90%.

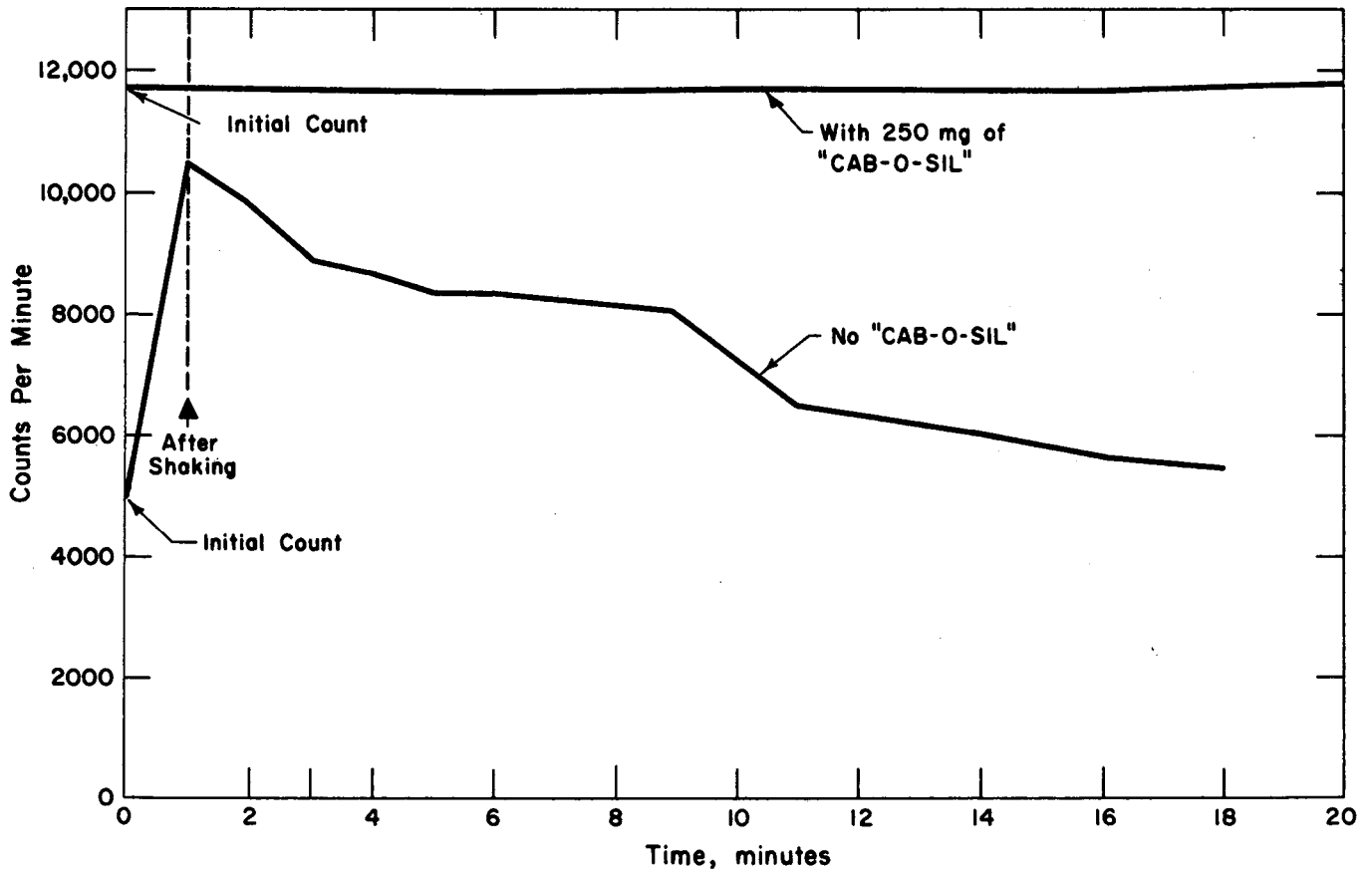


FIG. 4. EFFECT OF "CAB-O-SIL" ON COUNTING EFFICIENCY.

Three waste-water samples containing ^{35}S were analyzed in replicate to determine precision. Results are shown in Table II.

Table II. Replicate Analysis of Waste Water, counts/minute

Sample 1	Sample 2	Sample 3
19550	11419	11515
20091	11926	10468
19062	11502	11508
20045	11382	10292
Avg. = 19687 \pm 2.5% standard deviation	11552	11410
	12076	11746
	11701	Avg. = 11157 \pm 5.5% standard deviation
	10804	
	11172	
	10455	
	11277	
	11600	
	Avg. = 11406 \pm 3.9% standard deviation	

Decontamination factors for radionuclides likely to occur in plant waste water (Table III) are 10^3 or greater.

Table III. Decontamination Factors
for Radionuclides

<u>Nuclide</u>	<u>D. F.</u>
^{144}Ce	10^4
^{137}Cs	10^4
^{65}Zn	10^4
^{85}Sr	10^4
^{106}Ru	10^4
^{60}Co	10^4
$^{95}\text{ZrNb}$	10^4
^{32}P	10^3
^{239}Pu	10^3
^{51}Cr	10^4
^{131}I	10^4

REFERENCES

1. A. S. Goldin, Anal. Chem. 33, 406 (1961).
2. F. E. Butler, Anal. Chem. 33, 409 (1961).
3. G. H. Morrison and H. Freiser, "Solvent Extraction in Analytical Chemistry," John Wiley and Sons, New York, 1957 p. 109.
4. F. E. Butler, A. R. Boulogne, and E. A. Whitley, Health Phys. 12, 927 (1966).

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INCORPORATION OF RADIOACTIVE TRACERS
DURING CONTROLLED GROWTH
OF MONODISPERSE SILICA PARTICLES

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A new technique is described facilitating the controlled growth of monodisperse suspensions of silica spheres from solutions containing radioactive tracers. By using certain tetra acyl silicates as basic reactants, the uniform growth of silica spheres of diameters up to 2 microns can be effected by hydrolysis of the silicate and subsequent condensation of the silicic acid formed in the presence of a morphological catalyst. Radioactive elements which form oxides of low solubility are easily incorporated when present during the growth of the silica particles.

All particles are in the respirable size range. The suspensions can easily be diluted and nebulized in aerosol generators. The uniformity of the air-borne particles makes such aerosols an excellent tool for studies on inhalation, deposition, and clearance in the lung.

INCORPORATION OF RADIOACTIVE TRACERS DURING CONTROLLED
GROWTH OF MONODISPERSE SILICA PARTICLES*

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University of Rochester, N. Y.

Introduction

In a recent study (1) a chemical system for the generation of silica particles of uniform spherical size has been reported. Tetraalkyl esters of silicic acid in alcoholic solution react with water in the presence of ammonia in such a way that the hydrolysis of the ester is subsequently followed by a condensation of the silicic acid causing a growth of silica particles. This growth is very uniform for a number of different tetraalkyl esters and the final size of the spheres can be controlled up to 2 μ diameters by varying the concentrations of water and ammonia in the initial alcoholic solution. Figure 1 shows typical results for the final size in the tetraethyl ester/ethanol system. Figure 2 is an electron micrograph of silica particles grown in such a system and Figure 3 shows the cumulative particle size distribution of this sample plotted on probability paper versus the logarithm of the particle diameter. A very narrow logarithmic normal distribution is revealed by the steep straight-line relationship.

*This paper is based on work performed under contract with the U. S. Atomic Energy Commission at the University of Rochester Atomic Energy Project and has been assigned Report No. UR-49- 867.

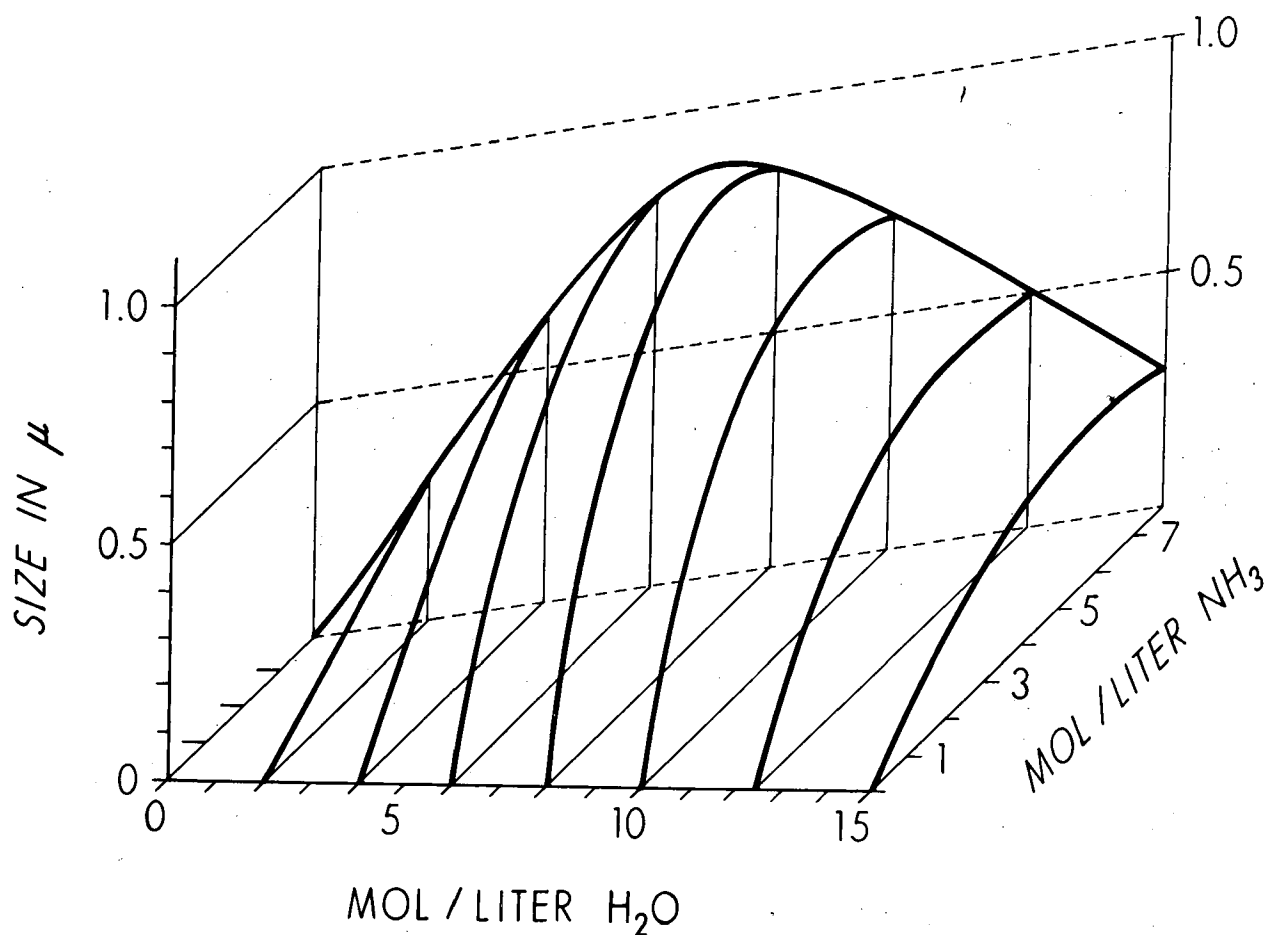


Fig. 1. Final particle size of silica spheres as obtained by reacting 0.28 mole/liter of tetraethyl silicate with various concentrations of water and ammonia in ethanol.

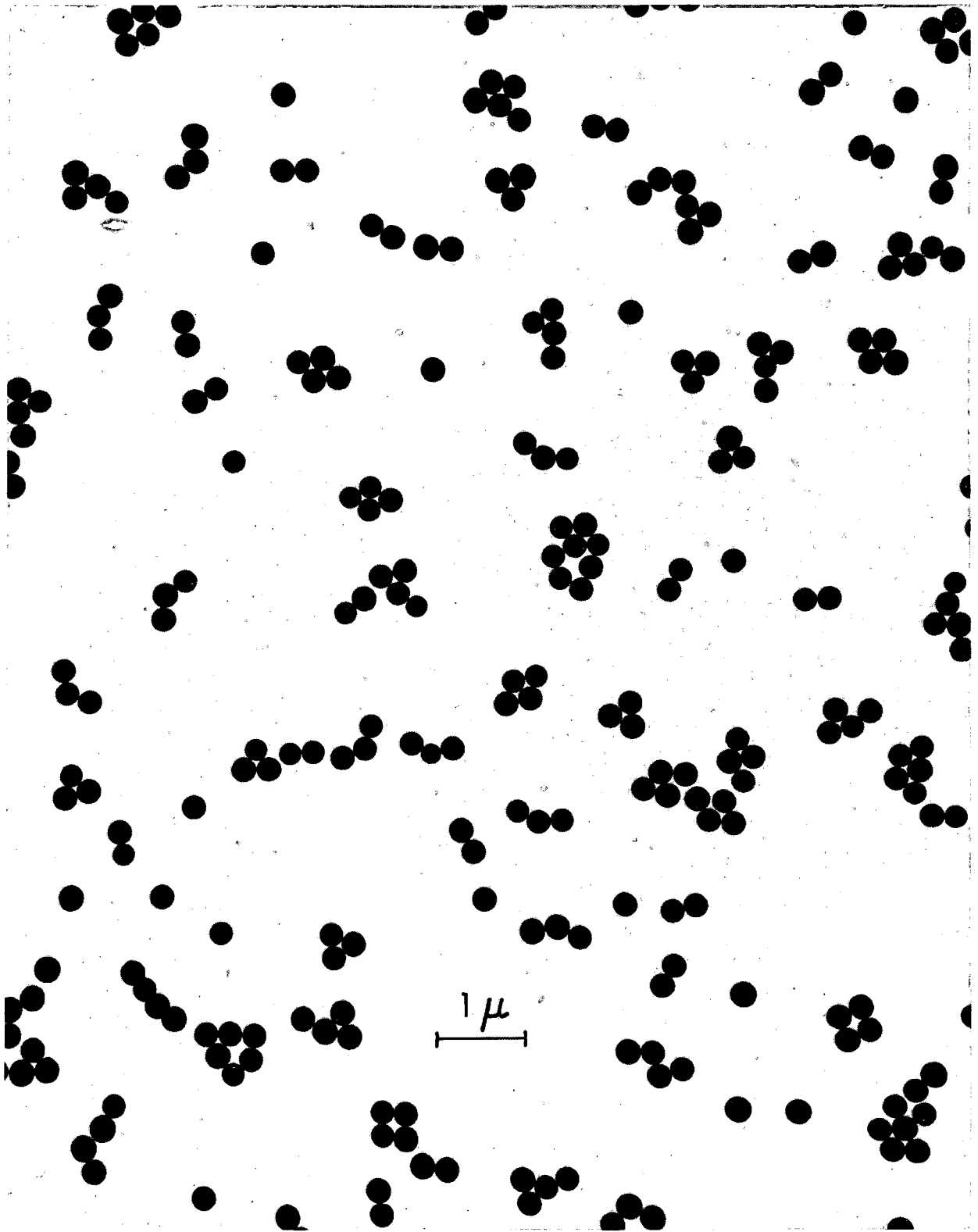


Fig. 2. Electron micrograph of a sample of silica spheres obtained in the ethanol-ethyl ester system.

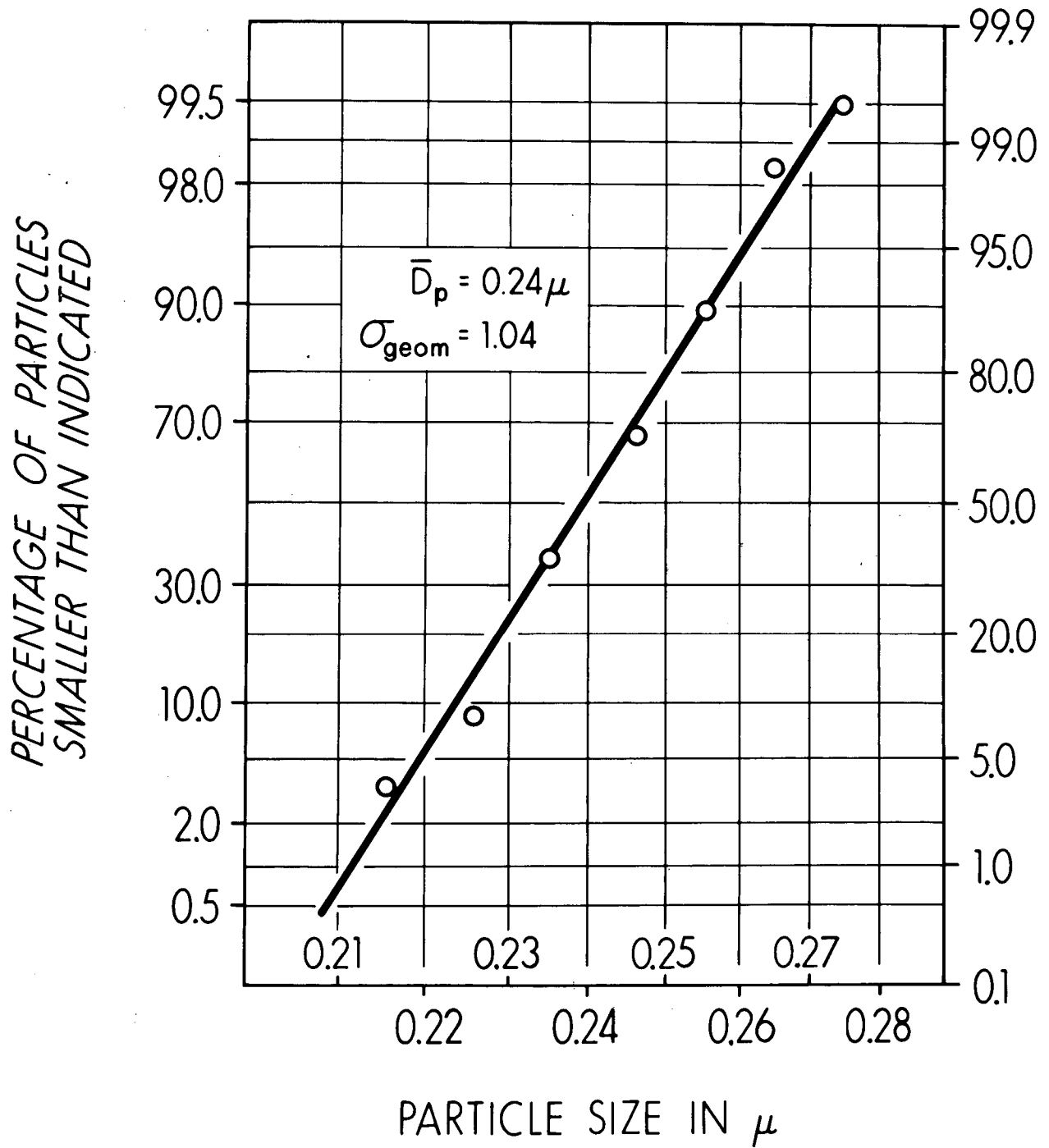


Fig. 3. Log-probability plot of the cumulative size distribution curve of the sample shown in Fig. 2.

This new technique of producing quasi-monodisperse silica particles for hydrosol and aerosol studies facilitates attempts to incorporate radioactive tracers into the bulk of the spheres by inclusion or substitution in the framework of the SiO_4 tetrahedrons during the growth of the silica particles. In order to explore this possibility, which would make the silica particles a versatile tool for many investigations, a systematic study with a number of radioactive isotopes was conducted.

Experimental Methods

The experimental procedure of attempting the incorporation of radioactive isotopes by growing silica particles was simple. Approximately $1 \mu\text{c}$ of dissolved radioactive material amounting in most cases to not more than a few μg of foreign matter was added to a 50 ml solution that would finally yield 500 mg of silica.

Initial tests were conducted to investigate the influence of the radioactive tracers on the nucleation and condensation pattern. No significant difference in particle size was detected between this series and the controls.

A standard procedure was then used to grow the silica particles. After mixing 40 ml of pure ethanol and 10 ml of saturated ammonium hydroxide, 2 ml of tetrapropyl ester of silicic acid and the radioactive tracer were added to the solution. After a few minutes the solution turned turbid and the final size of the growing particles was obtained after 2 hours. Electron micrographs indicated sizes close to 0.8μ . The suspension was then membrane filtered (Millipore filter GS 0.22μ) and the activity of the filter cake and in the filtrate were measured separately in a well-type crystal counter. Throughout the experiments

the activity of the filtrate was essentially background level and all the activity was associated with the particles on the filter.

The filter cakes were then resuspended for 24 hours under various conditions in 100 ml of alcoholic or aqueous suspensions over a wide p_H range and with different dissolved agents selected to remove the exchangeable radioactive material from the particles.

Results

1. $^{134}C_s$

This isotope was added in the form of cesium chloride. The amount of 1 ml of a solution containing less than 1 μg of C_s in 0.5N HCl. All radioactivity could be removed from the resuspended silica particles by adding 1 g of C_sCl to the suspension. Two hours later, after the material was filtered, very little activity could be detected on the filter cake.

2. ^{54}Mn

One ml of manganese chloride in acidic solution containing 2.5 $\mu g/ml$ and an activity of 1 $\mu c/ml$ was used in these tests. The removal of the activity from the resuspended particulates could not be accomplished in alcoholic suspensions and in aqueous suspensions at $p_H > 7$. However, at low p_H values, particularly after adding 1 g of $MnCl_2$ and/or tartaric acid (20 ml of 20% acid) a gradual removal of activity from the particles amounting to a loss of 50% within 36 hours was observed in a series of runs.

3. ^{65}Zn

In these tests 1 ml of a solution containing zinc in a concentration of $3\ \mu\text{g/ml}$ and an activity of $0.3\ \mu\text{c/ml}$ was added to the test solution. The resuspended particles did not release the activity in alcoholic suspension, but a gradual release was observed in aqueous suspensions. It was rather slow for high p_{H} values but increased rapidly with decreasing p_{H} values. At $p_{\text{H}} 2$ the activity of the particles was nearly instantaneously released. Adding $\text{Zn}(\text{NO}_3)_2$ generally increased the rate of removal of activity from the particles at high p_{H} values and adding 20 ml of 0.1 molar EDTA gave the highest rates of removal of the activity at any p_{H} value.

4. ^{124}Sb

A tracer concentration of $4.5\ \mu\text{g/ml}$ was used in these tests and 1 ml containing $0.25\ \mu\text{c}$ of activity was added to each test solution. Resuspending the silica particles in various solution showed little effect on the activity associated with the particles. No changes were observed in alcoholic suspension and adding SbCl_3 or tartaric acid to aqueous suspension did not increase a small release of activity. The highest releases were obtained at high p_{H} values ($p_{\text{H}} > 9$) and amounted to about 7 per cent.

5. ^{59}Fe

One ml of a tracer solution of $2.6\ \mu\text{g Fe/ml}$ containing $1\ \mu\text{c/ml}$ of activity was added to the test solutions of this series. The results after resuspension of the particles indicated that the various solutions

applied had little influence on the activity associated with the particles. Most prominent releases occurred at high p_H values and did not exceed 5 per cent.

Discussion

As expected, the chemical nature of the isotope used as the tracer is of great influence on the mode of attachment to the growing silica particles. As a representative of elements which remain in the ionic dissociated state over the entire p_H range, cesium showed in the experiments that no incorporation takes place. Apparently, the cesium ions always remain on the surface of the silica particles from where they can be removed by simple exchange with material dissolved in the liquid phase. The same seems to be true for zinc and manganese although significant removal occurred primarily at low p_H values and was incomplete in the case of manganese. The results, however, strongly suggest that surface phenomena are encountered.

A different mechanism seems to occur in case of antimony and iron. Here the results indicate that the release of tracer activity is not dependent upon the constituents of the liquid phase but that a small release is related to the solubility of silica. The saturation concentration is about 100 $\mu\text{g/ml}$ for dissolved silica at $p_H \leq 8$ and increases to about 400 $\mu\text{g/ml}$ at $p_H 10$ (2). Under the experimental conditions of the tests, this should cause an activity loss of about 8 per cent at $p_H 10$ if the activity is evenly distributed over the volume of the silica particles. The experimental results are in keeping with this assumption.

Investigations on other isotopes are anticipated.

References

1. W. Stöber, E. Bohn, and A. Fink, J. Colloid and Interface Sci.,
in press
2. G. B. Alexander, W. M. Heston, and R. K. Iler, J. Phys. Chem.
58, 453 (1954)

NLCO-1007

THE ESTIMATION OF THORIUM IN THE BODY

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ABSTRACT

Three methods for the estimation of exposure to airborne thorium are discussed: in vivo counting, the neutron-activation determination of thorium in urine, and breath thoron measurements. The limited data obtained in tests to date indicate that the present limits for inhalation exposure to thorium are adequate.

BACKGROUND AND PROCEDURES

Thorium operations at the National Lead Company of Ohio have increased as a result of the AEC's renewed interest in thorium as a reactor fuel. An evaluation of possible health problems in processing thorium feed materials revealed apparent inconsistencies in recommended limits for inhalation exposure to natural thorium, Th-232, and the isotope Th-230. While investigating these inconsistencies, we learned that lower limits were being considered by the International Commission on Radiological Protection. Our opposition to lower limits and our reservations about existing limits were discussed in a paper presented at the 1965 meeting of this group.¹

A session of the 1965 meeting was devoted to the thorium MPC's. During one discussion, Dr. Walter Snyder of ORNL pointed out that the basic limits are in terms of annual or quarterly doses to organs and, hence, the organ burden is the basic criterion for control. He said that while recommended air limits are definitely needed, they are not the primary standard for control. Hence, if the MPC is temporarily exceeded, even for a quarter, this only constitutes presumptive evidence that the basic standard may have been exceeded. If it can be shown that the organ burdens are, in fact, within the accepted guides or standards, this establishes compliance with the basic standard.

We wholeheartedly concurred with this opinion and set out to develop the capabilities to estimate thorium in the body. Our attempts to gather data on thorium body burdens were directed at in vivo counting, breath thoron measurements, and a determination of thorium in urine by neutron activation. In vivo counting was to be our basic method, against which we could assess or calibrate the other two techniques. These efforts are briefly described in this report.

In Vivo Counting

Five employees with varying degrees of thorium exposure were counted in the Oak Ridge Y-12 In Vivo Radiation Monitoring Facility. No thorium was detected in any employee; the results were not statistically different from counts obtained with nonexposed persons. The lower limit of detection for thorium was about 20% of a maximum permissible lung burden.

One of the five employees was a chemical operator who had worked for about 1-1/2 years in a thorium remelt area eleven years ago. Insoluble thorium dusts are associated with this operation. About 11 months before the in vivo count he returned to thorium operations when the plant began small-scale production of purified thorium compounds. He was an excellent subject for the in vivo count because his total thorium exposure was probably more than that of any other of our employees.

The results of the in vivo counts indicated that exposure controls used in the past were adequate. They also provided a basis for the comparison of urine results and breath thoron measurements.

Four of the five employees counted at Y-12 were also counted at the in vivo facility operated by the U. S. Air Force Radiological Health Laboratory at Wright-Patterson Air Force Base. This was their first attempt at in vivo counting for thorium. No thorium was detected in three employees. A lung burden of 25% of the maximum permissible was estimated for the chemical operator with the most exposure. However, the counting was done in the middle of the work week, which would not have allowed out-of-equilibrium, short half-life daughters to decay. Rn-220 is easily driven off in thorium operations, and can be present in above-equilibrium amounts. Both the Y-12 and Wright-Patterson counting techniques utilize the gamma activity from several of these daughters.

In air samples from our recent thorium operations, the activity from Rn-220 and daughters has been about ten times higher than the activity would be if equilibrium conditions existed. The decay of these daughters is controlled by the 10.6-hour half-life of Pb-212, and several days would be required for essentially complete decay. While the

Wright-Patterson count was made in the middle of the work week, the Y-12 count was made after the employee with the most exposure had been on vacation and away from thorium work for 12 days. Therefore, there is good justification for placing greater reliance on the Y-12 measurement.

Additional counts are planned for the Oak Ridge portable in-vivo counter when it becomes operational.

Breath Thoron Measurement

Measurement of thoron in the breath of thorocontrast patients with known Ra-224 burdens indicates that about 10% of the Rn-220 formed in the body reaches the expired breath.^{2,3} Despite the uncertainties surrounding the state of thorium radioactive equilibrium in exposed workers, it appeared that breath thoron measurements, coupled with occasional in vivo counts, would allow a reasonable estimation of thorium lung burdens. Accordingly, a system for breath thoron measurements was assembled with the assistance of Dr. John B. Hursh, University of Rochester, who had developed a successful system.²

In the measurement system, the subject's breath flows through a narrow absorption-detection chamber. The bottom of the chamber is a 5-inch-diameter aluminum disk with an adhering

thin layer of finely divided activated charcoal. The top of the chamber is a clear plastic disk coated with ZnS scintillator. The disk is coupled to a 5-inch phototube. As breath flows through the chamber, the thoron is absorbed on the charcoal and the alpha decay is detected by the ZnS-phototube arrangement. A detailed description of the system is given in Reference 2.

The system we have assembled requires final adjustments and some refinement. However, sufficient data have been obtained to permit an estimation of the thorium burden in the chemical worker with the most thorium exposure. His breath-thoron measurements indicate a lung burden certainly less than 50% of the suggested limit and probably less than 10%.

Neutron Activation Analysis

Spectrophotometric or fluorometric techniques are not sensitive enough for the determination of thorium in the urine from nonexposed persons or workers exposed to the small amounts of airborne dusts resulting from most industrial thorium operations. The usual thorium compounds, including oxides, hydroxides, and oxalates, are normally considered insoluble when deposited in the lung. There are, no doubt, fluctuations in the thorium-in-urine level which correspond to the amount

deposited. Since routine methods of thorium determination do not permit observation of these fluctuations, nuclear activation analysis was considered as the best method to obtain the necessary analytical sensitivity.

A neutron activation method was developed using the reactor facilities at Wright-Patterson Air Force Base, Dayton, Ohio.⁴ When spiked whole-urine samples were irradiated and counted, the thorium peak was masked by the Compton scattering from Na-23 and Cl-38. In the final procedure, pre-irradiation and post-irradiation separations were used to concentrate the thorium and reduce the interferences.

For each analysis, duplicate 30-ml aliquots of acidified urine were transferred to centrifuge tubes, and one of the aliquots was spiked with a known amount of thorium. Ammonium hydroxide was added to obtain a precipitate which was collected and then dissolved with dilute acid. The precipitation was repeated. Using 7 ml of dilute nitric acid, the second precipitate was dissolved and transferred to a small plastic vial. After heat-sealing, each sample and its spike were placed in the same rabbit and activated for 15 minutes at a thermal flux of 10^{14} n/(cm² sec). Immediately after irradiation the vials were opened, and the contents were transferred to centrifuge tubes. Lanthanum carrier was added and a precipitation was made, twice, with NH₄OH. Sodium chloride

was added before each precipitation to act as a hold-back carrier for the sodium and chloride which might have been carried through the double pre-irradiation separations.

Radiation levels of the irradiated samples imposed no limitations. After irradiation, the unopened rabbits had radiation levels up to 1300 mr/hr. The vials, several minutes after removal from the rabbits, had levels of 15-50 mr/hr.

So far, data have been obtained from four urine samples from nonexposed persons and one sample from the chemical operator with the most thorium exposure (see Table I). The nonexposed samples were composites of urine from laboratory personnel with no industrial exposure to thorium.

TABLE I

Thorium in Urine

Nonexposed "A"	0.13×10^{-8} g/l
Nonexposed "B"	0.25×10^{-8} g/l
Nonexposed "C"	0.43×10^{-8} g/l
Nonexposed "D"	0.16×10^{-8} g/l
Exposed "E"	6.02×10^{-8} g/l

Standard deviation (σ) for "A" = 3.7%

"E" = 7.1%

Although we hesitate in drawing definite conclusions from the results of only five samples, we can make two observations based on these data and on the other work described here.

(1) The concentration of thorium in the urine of persons with no industrial exposure is in the area of $0.1 - 0.4 \times 10^{-8}$ g/l.

(2) The concentration of 6×10^{-8} g/l for the thorium worker is above the background level and is associated with less than a maximum permissible lung burden, probably less than 20% of a MPLB and possibly less than 10%.

CONCLUSIONS

In vivo counts and breath-thoron measurements have shown that our thorium workers have not accumulated significant amounts of thorium. The data obtained show no need for a lower MPC for thorium in air. Furthermore, to achieve the recommended lower air concentration levels would be a costly undertaking.

The Y-12 in vivo counts are supported by breath-thoron measurements. A difference in thorium-in-urine concentrations was found between one exposed worker and several nonexposed persons. Efforts will continue in an attempt to relate urine levels to body burdens as determined by in vivo counting and breath-thoron measurements.

REFERENCES

1. R. C. Heatherton and M. W. Boback, Thorium and Its MPC's, Proceedings of the 11th Annual Bio-Assay and Analytical Chemistry Meeting, October 7-8, 1965, USAEC Report NLCO-962, September 1, 1965 (CONF-651008, p.187).
2. J. B. Hursh and A. Lovass, A Device for Measurement of Thoron in the Breath, Health Physics ,9: 621, 1963.
3. J. B. Hursh, Body Content of Thorium-232 Daughters After Thorotrast Injection, Brit. J. Radiol. 38: 776. 1965.
4. B. L. Twitty and M. W. Boback, A Determination of Thorium In Urine by Thermal Neutron Activation Analysis. (To be published.)

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RAPID DETERMINATION OF THORIUM AIR FILTERS
BY NEUTRON-ACTIVATION ANALYSIS

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ABSTRACT

A rapid neutron-activation analysis method has been developed to determine concentrations of ^{232}Th in air filters down to 5 micrograms, a level well below that necessary for monitoring occupational exposure to thorium.

INTRODUCTION

Urine analysis is commonly used to estimate the body burden of many radionuclides; however, exposure to or the body burden of thorium is not reliably estimated by this technique. Consequently, exposure to thorium is normally estimated by analyzing for particulate thorium in air. State and Federal codes require that the thorium content of air should not exceed 270 micrograms/cubic meter of air. The purpose of this investigation was to develop an accurate, fast, sensitive, and inexpensive technique for the analysis of Thorium-232 in air filters. In our laboratory all these criteria are filled by the neutron-activation method. (1)

Neutron-activation analysis is an elemental method of analysis based on the quantitative measurement of radioactivity produced in a sample by neutron irradiation. The technique is essentially divided into three parts: activation, isolation, and quanti-

tative measurement. First, the sample is irradiated in a neutron source. In this source, neutrons interact with nuclei of isotopes in the sample to produce radioactive nuclei. Every radionuclide formed is characterized by its decay rate and radiation. Second, physical, chemical, and instrumental means are used to isolate the emissions of a particular radioactive species from all the others in the irradiated sample. Third, the intensity of the radiation from each radioactive species is a quantitative measure of the isotope that produced it. This technique has been used to measure approximately 70 elements in the weight range of 10^{-6} to 10^{-12} gram.

When thorium is irradiated in a fast neutron flux of a nuclear reactor, the high energy neutrons interact with ^{232}Th producing, primarily, a large number of fission fragments. The concentration of ^{232}Th is then determined by counting the number of neutrons emitted in a decay of a number of short-lived fission products⁽²⁾ and by comparing this number with that obtained from a thorium standard which was irradiated and counted in the same manner as the sample.

EXPERIMENTAL

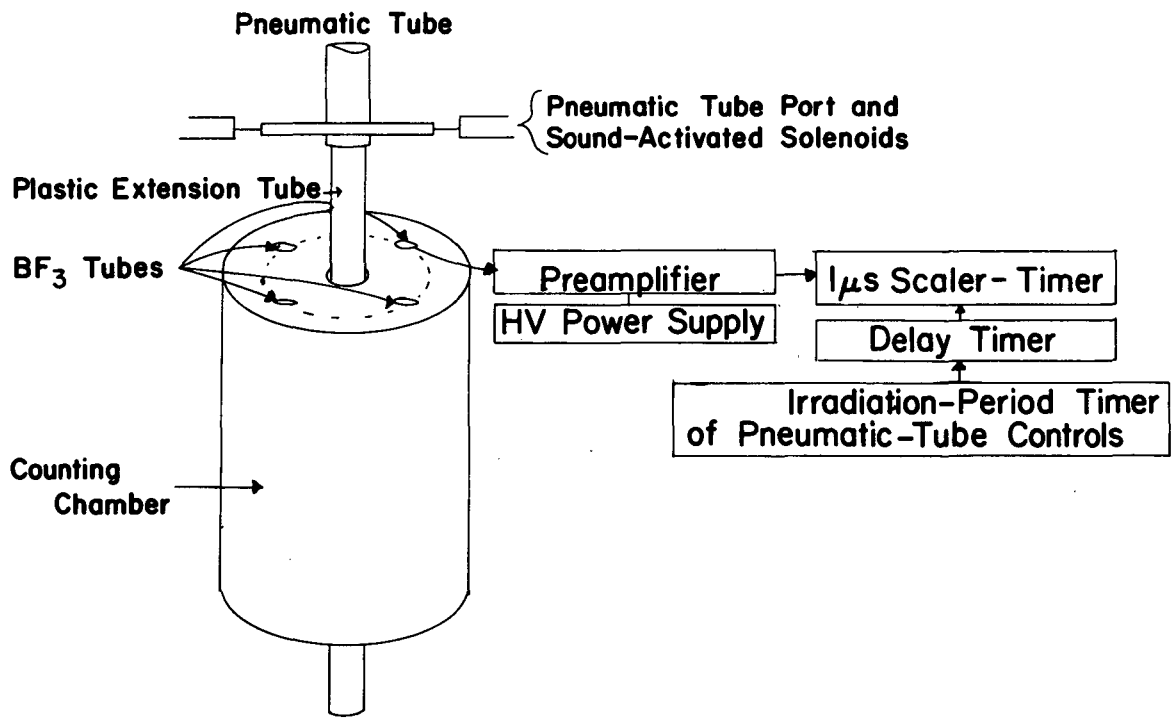
Air filters are carefully rolled into a small tight cylinder and placed into polyethylene medical tubing. The samples are not weighed, since the results are reported in micrograms of thorium per filter. This, in turn, represents the micrograms of thorium per cubic meter of air pulled through the filter. The neutron-activation technique for thorium is not limited to the analysis for thorium on air filter samples. Other samples of virtually any matrix may be analyzed for thorium and are packaged in a similar manner. Thorium standards

are prepared by weighing 1 to 4 milligrams of dried thorium dioxide in the same type of package as the sample.

Samples and standards are placed inside separate cadmium-lined pneumatic tube transfer containers (rabbits). The wall thickness of the cadmium liner is 40 mils. The samples and standards are then irradiated one at a time for a period of 30 seconds in a pneumatic transfer tube facility of the Union Carbide Research Reactor.⁽³⁾ The method of controlling and recording the irradiation period, as well as the method of sample coding, have been described previously.⁽⁴⁾

After the irradiation period, the sample is returned to an apparatus (Fig. 1) for ^{232}Th analysis. When the rabbit container strikes the pneumatic tube port, the impact noise activates a sound transducer that turns off the air flow in the pneumatic tube system and opens the port, permitting the irradiated container to drop by gravity into a counting chamber similar to that used at Oak Ridge.⁽²⁾ When the elapsed timer⁽⁴⁾ of the pneumatic tube system times out, in addition to changing the air directional control, it triggers a 20-second delay timer in the delayed neutron counting apparatus. Hence, 20 seconds after the sample is irradiated, a scaler is activated and the BF_3 tubes count the neutrons emitted from the fission products of ^{232}Th for the next 60 seconds. The total number of neutrons counted is converted directly into micrograms of thorium by calibrating the system with thorium standards.

At an equivalent fission flux of $1 \times 10^{13} \text{ n/cm}^2 \cdot \text{sec}$, the response of the delayed neutron detection system is $6.8 \pm 0.2 \times 10^6$ counts/gram of thorium over a weight range of 10^{-5} to 10^{-2} gram. The combined



DELAYED NEUTRON APPARATUS

Fig. 1

background count of empty, cadmium-lined irradiated rabbits and of the detection system is 80^{+10} .

When the thorium content exceeds 15 milligrams, the coincidence counting losses due to high count rates exceed 5%. This problem is circumvented by irradiating high-thorium-content samples in a much lower equivalent fission flux by varying the position of the pneumatic tube with respect to the reactor core, since the response of the system (as well as the counts due to the irradiated rabbit) is directly proportional to the neutron flux.

EXPERIMENTAL RESULTS

To demonstrate the utility of the delayed neutron technique for the analysis of thorium on air filters, a number of filter papers were exposed to varying concentrations of thorium. These filter papers, as well as blank filter papers and empty cadmium-lined rabbits, were irradiated and processed in the manner described above. The results of this experiment are shown in Table I. These data show that the response of the detection system for thorium is essentially linear over the concentration range of 10^{-5} to 10^{-2} gram. The data also indicate that thorium contents down to 10 micrograms are readily measured and that the lower limit of detection for our system is approximately 5 micrograms. When the concentrations approach this level (5-10 micrograms) the associated errors are very large (± 20 or $\pm 50\%$). However, when the concentration of thorium is greater than 50 micrograms, the relative standard deviation is better than $\pm 5\%$.

Our laboratory now routinely analyzes air filter samples obtained from a smelting operation of thorium-bearing ore. Table II

TABLE ITHORIUM DEPOSITED ON FILTER PAPER

<u>Sample</u>	<u>Counts</u>
Empty Cadmium-Lined Rabbit	80
Blank Filter Paper	10
1 μg Th deposited on Filter Paper	5
10 μg " " " " "	70
100 " " " " "	689
500 " " " " "	3375
1000 " " " " "	6588

TABLE IISAMPLES FROM A SMELTING OPERATION

<u>Sample</u>	<u>Thorium</u>
Original Ore	2.24%
Slag from first melt	1.45%
Flue particulate matter	8 μg
Air sample from smelting area	< 6 μg
Air sample from loading area	< 6 μg

shows typical results obtained from such samples. In addition to these types of samples, we routinely use this technique to analyze for thorium in urine, metal, and vegetation.

DISCUSSION

The process of neutron emission following an irradiation by neutrons can only be attributed to the presence of a fissionable nuclide, 0.17-second ${}^9\text{Li}$ and/or 4.14-second ${}^{17}\text{N}$. These latter two are produced in oxygen, lithium-oxygen, and beryllium-containing samples. The naturally abundant fissionable heavy nuclides are ${}^{235}\text{U}$, ${}^{238}\text{U}$, and ${}^{232}\text{Th}$. Consequently, the measurement of neutron emission after neutron activation is specific for thorium if the sample does not contain uranium, and if the sample is permitted to decay for 20 seconds before the start of the measurement. It was observed by us and others⁽²⁾ that a delay of 20 seconds before counting is sufficient to reduce the ${}^{17}\text{N}$ activity and the ${}^9\text{Li}$ activity to a negligible amount even in aqueous samples.

If, however, the sample should contain uranium, the cadmium-lined containers, which absorb thermal neutrons, reduce interferences from ${}^{235}\text{U}$ but not from ${}^{238}\text{U}$. It should be pointed out that the cadmium-covered irradiation method reduces the interference but does not eliminate the interferences due to uranium. To circumvent the problems associated with the analysis of samples containing both thorium and uranium, it is advisable to irradiate the samples and the standards in rabbits, with and without cadmium liners. The data obtained from these irradiations will consist of: the counts for a bare irradiated sample, the counts for a cadmium-covered irradiated sample, and the response of the detector system for bare and cadmium-covered irradiations of thorium

and uranium standards. These data permit, by simple simultaneous equations, the analysis of both thorium and uranium in the same sample. However, when the uranium content is greater than 10 times that of the thorium, it is, for practical purposes, not possible to obtain a thorium assay by the delayed neutron activation technique.

CONCLUSIONS

Air filter samples can be analyzed rapidly, simply, and directly for thorium by the neutron-activation analysis technique employing the delayed neutron counting method. In samples containing no uranium, the method is matrix-independent and interference-free, and has a lower limit of detection of 5 micrograms--a level that is well below the State and Federal regulations for the content in 1 cubic meter of air. It takes less than 10 minutes per sample for total processing time, which includes: logging, packaging, irradiating, and counting the sample; recording of the data; and calculating the results.

If the sample contains uranium, a double irradiation method (with and without cadmium liners) is employed. This double irradiation permits accurate analysis technique for thorium and uranium up to uranium-to-thorium ratio greater than ten. Other than the detection limit, this uranium-to-thorium ratio maximum is virtually the only limitation to the analysis of thorium by this delayed neutron-activation analysis method.

REFERENCES

- (1) W. H. Wahl and H. H. Kramer, Scientific American 216, 68 (1967).
- (2) F. F. Dyer et al., ORNL 3342.
- (3) D. B. Holzgraf, Res. React. J. 2, 3, 1 (1962).
- (4) J. W. Nostrand, Jr., and H. H. Kramer, Preprint 2.4-2-64
19th Annual ISA Conference, Instrument Society of America.

RECENT DEVELOPMENTS IN THE SEQUENTIAL DETERMINATION OF URANIUM
AND TRANSURANIUM ELEMENTS USING BARIUM SULFATE

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Precipitation of large ter- and quadrivalent elements by barium sulfate has been extended to the separation of uranium and the trans-uranium elements both from other elements and from each other. From 25 ml of solution containing sulfuric acid, potassium sulfate, and hydrogen peroxide, carrier-free quantities of all elements from lead through at least californium, except astatine, radon, francium, and uranium, are precipitated to approximately 99% by only 5 mg of barium ion. Uranium can also be included in the list of elements precipitated by reducing it to the quadrivalent state with titanium trichloride either before or after precipitation of barium sulfate. Like uranium, the trans-uranium elements through americium can be prevented from precipitating by oxidizing them to the hexavalent state in which the oxygenated cation is too large to fit into the barium sulfate lattice. Also, because of the marked differences in their oxidation potentials, uranium and the first four transuranium elements can be separated from other non-oxidizable ions and from each other by proper control of the oxidizing agent used. Ammonium peroxydisulfate in the presence of silver catalyst oxidizes americium but not curium; permanganate oxidizes plutonium but not americium; dichromate oxidizes neptunium but not plutonium; hydrogen peroxide oxidizes uranium but not neptunium; and uranium can finally be reduced with titanium trichloride. The precipitate can be mounted on a 2-inch stainless steel plate by centrifugation and counted directly in an alpha counter with less

than about 10% loss of counting efficiency due to the solid absorber present. Particularly important, the barium sulfate can be dissolved in water after evaporation with hydriodic and perchloric acids to reduce and volatilize the sulfate ion. The radionuclides present in the barium sulfate can then be electrodeposited from the resulting aqueous solution and each specific isotope present identified by alpha spectrometry. This procedure permits not only traditionally difficult separations such as americium from curium or the determination of extremely minute quantities of one transuranium element in the presence of large quantities of another to be made simply and quickly, but mixtures of uranium and the first four transuranium elements can be separated sequentially from each other and identified on the same sample in a single procedure. Furthermore, since virtually every alpha-emitting nuclide known can be precipitated on barium sulfate under the proper conditions, a chemical separation can be employed to eliminate the absorbing material from large samples without losing the concept of a true gross alpha procedure.

PLUTONIUM CONTENT OF SEVERAL INTERNAL
ORGANS FOLLOWING OCCUPATIONAL EXPOSURE

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ABSTRACT

Tissue samples from several internal organs were analyzed for plutonium following the natural death of an employee of a plutonium production plant. This employee had two contaminated puncture wounds and had several small inhalation exposures over a 9-year period. He had been excreting measurable quantities of plutonium in his urine since 1958.

A body burden calculation based on these urine results indicated the presence of 13.5 percent of the maximum permissible amount at the time of death. An extrapolation of tissue analysis to the whole body indicates the presence of 2.4 percent of the maximum permissible amount.

INTRODUCTION

Many articles (1-5) have been written describing the uncertainties in calculating plutonium body burdens from urine analysis. These articles all agree, however, that at the present time an alternative method does not exist. Human data relating known body burdens to urine analysis comes from Langham's work (6) so his equation ($D_r = 435 U t^{0.78}$) or some close variation of it (7-8) is used for estimating body burdens.

In the case reported here (Number 667) a comparison is made between the body burden calculation and the amount of plutonium found in several tissues at autopsy following death from natural causes. This employee had worked in a plutonium production plant for a period of 9 years and had an estimated 13.5 percent of a maximum permissible body burden at the time of death based on his urine analyses.

Histopathological and autoradiographic studies are also being conducted on some of these tissues but their results are not included in this report.

The need for obtaining tissue analyses has been long known in the plutonium industry. In fact, efforts are currently under way to create a "plutonium registry" which

is designed to help fill the gaps in our knowledge of determining plutonium body burdens.

RECORD OF INVOLVEMENT

Records indicate that on 20 different occasions this employee was involved in contamination incidents. In each case a hole developed in a box glove permitting the spread of material which was most likely PuO_2 . Contamination levels exceeded 100,000 d/m alpha per 100 cm^2 on the floor and nearby surfaces in most of the incidents. General room air contamination, however, was always measured to be lower than maximum permissible amounts. Contamination was usually found on his hands, arms, coveralls, and occasionally his face. He probably suffered a small inhalation exposure from one or more of these incidents because measurable quantities of plutonium were found in his urine before any contaminated puncture wounds were discovered. (See Table 1.)

On three different occasions, wounds were checked for possible contamination. Two were found to be positive. On December 31, 1959, 0.24 μg of plutonium was measured in a wound on his right thumb. An excision removed all but 0.022 μg . This wound was rechecked periodically. (See Table 2.) The second contaminated wound occurred on

Table 1
Plutonium and Americium Measured in 24-Hour
Urine Samples Since June 26, 1958

Date	Disintegrations per Minute		Date	Disintegrations per Minute	
	Pu	Am		Pu	Am
6-26-58	Bkgd.		8-12-63	0.27	
9-15-58	0.31		9-13-63	0.07	
12-15-58	0.58		10-21-63	0.28	
6-25-59	Bkgd.		12-13-63	0.10	
9-16-59	0.09		4-28-64	0.07	0.30
1-04-60	1.11		6-14-64	0.37	
2-22-60	0.27		7-17-64	0.23	Bkgd.
3-07-60	0.27		8-21-64	0.17	
1-05-61	0.09		12-10-64	0.03	
12-14-61	0.18		6-17-65	0.29	
6-13-62	0.57		7-30-65	Bkgd.	
12-21-62	0.32		9-30-65	0.13	0.08
2-21-63	0.21		12-10-65	0.11	0.09
3-14-63	0.17		6-21-66	Bkgd.	Bkgd.
4-04-63	0.21		7-19-66	0.13	
6-17-63	0.55		12-19-66	Bkgd.	
7-12-63	0.41		6-07-67	Bkgd.	Bkgd.

Table 2

Amount of Plutonium Measured
in Two Contaminated Wounds

<u>Wound Number 1</u>		<u>Wound Number 2</u>	
<u>Date</u>	<u>Amount (µg)</u>	<u>Date</u>	<u>Amount (µg)</u>
12-31-59	0.240	7-11-66	0.004
12-31-59	0.022	1- 9-67	0.004
2-15-61	0.015		
2- 2-62	0.013		
1-16-63	0.016		
1- 9-64	0.018		
1- 7-65	0.016		
1-24-66	0.005*		

* This reading is questioned.

July 11, 1966, when 0.004 ug was measured in his second finger, right hand. No medical action was taken on this wound.

Recounts of wounds show good consistency except for the recount of the first wound on January 24, 1966. This count does not seem to be correct in view of the five previous recounts that showed nearly 3 times the amount. We feel that this last reading was in error and that he had about 0.016 ug (the average of the other recounts) in his right thumb at the time of death.

ESTIMATION OF PLUTONIUM BODY BURDEN FROM URINE ANALYSIS

Results for plutonium on the analysis of 34 urine samples collected after January, 1958, are shown in Table 1 and are displayed on a linear plot in Figure 1. Prior to January, 1958, he worked exclusively with uranium and submitted 12 urine samples which were all determined to be background for uranium.

Body burden estimates were made using the results from the urine analyses. Langham's equation ($D_r = 435 U t^{0.78}$) was used with allowances made for multiple exposures. The calculations were done using a computer program similar to the programs reported by Lawrence (9) and Snyder (3)

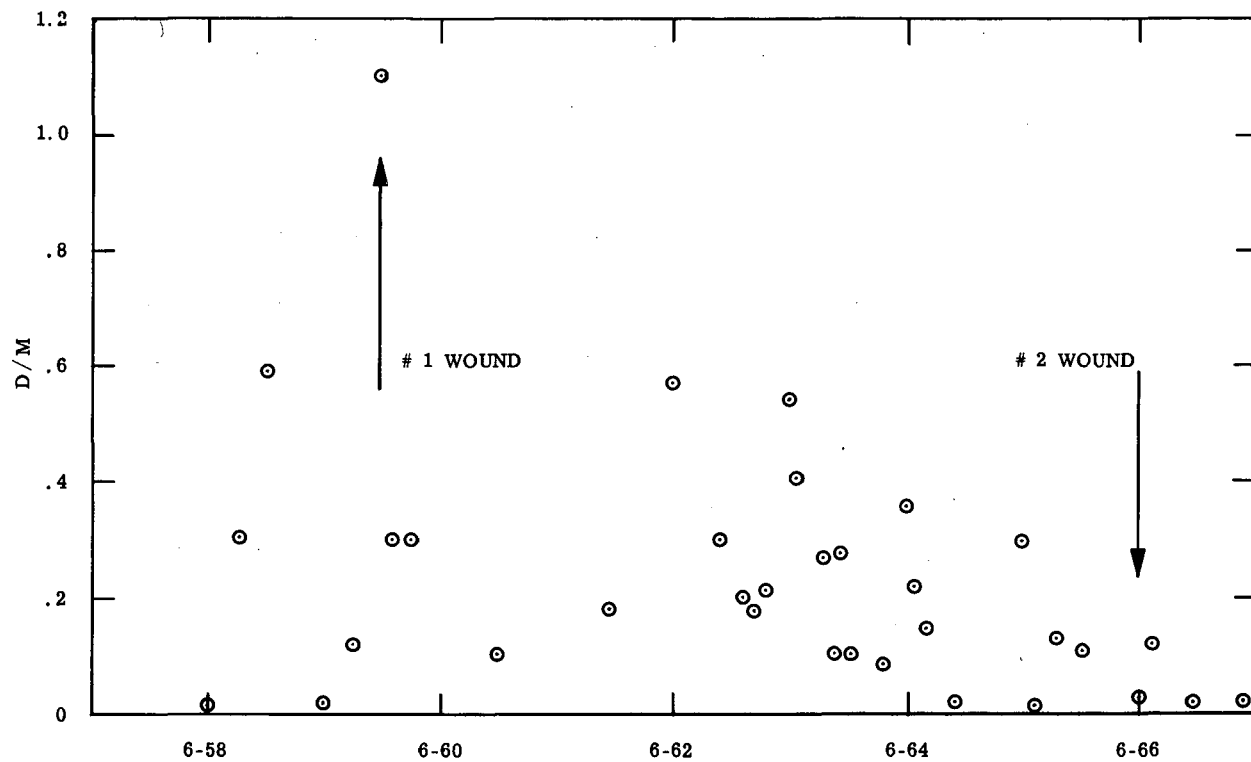


Fig. 1. Plutonium in 24-hour urine sample.

except our program does not cast out data nor does it allow for continuous excretion.

Calculations were done eight times since 1962 with the final result indicating that there was 13.5 percent of a maximum permissible body burden present at the time of death (Table 3).

METHOD OF TISSUE ANALYSIS

The samples were wet ashed in concentrated nitric acid and taken to dryness several times. The residual organic material was then destroyed with nitric acid containing small amounts of 30 percent hydrogen peroxide. The inorganic pyrophosphates were hydrolyzed by repeated evaporation and redissolution with dilute hydrochloric acid. The plutonium was isolated by anion exchange according to the method of Campbell (10) using Bio-Rad AG 1x2 resin.

Insoluble residues remaining after dissolution of the wet-ashed samples were fused with sodium carbonate and analyzed separately for the plutonium.

Table 3

Estimated Percent of Maximum
Permissible Body Burden (MPBB)

<u>Date</u>	<u>Percent of MPBB</u>
Sept., 1962	6.8
Jan., 1964	16.2
Jan., 1965	15.6
July, 1965	14.3
Jan., 1966	16.0
June, 1966	14.6
Jan., 1967	13.8
July, 1967	13.5

RESULTS OF TISSUE ANALYSES

The results of the individual tissue analyses for plutonium are shown in Table 4. Figure 2 shows the average plutonium concentrations in disintegrations per minute per gram which represent the relative dose rates to the different organs. The highest concentration was in the bronchial lymph nodes with 1.4 d/m per gram. This represents an absorbed dose of about 600 mrem per year. The next highest concentration was found in the liver with 0.32 d/m per gram followed by the lung at 0.21 d/m per gram and the skeleton at 0.13 d/m per gram. The concentration in the remaining tissue was taken as 0.003 d/m per gram which is the average result from the kidney, spleen, and a 32-gram tissue sample taken from the rib area.

Table 5 shows the total body content of plutonium extrapolated from the individual tissue samples. This information is also displayed in Figure 3 where the stippled areas represent the fraction of the organ that was analyzed. Only in the case of liver were we able to analyze the entire organ. More than half of the total body content was determined to be in the skeleton (58%). The liver had the second highest amount with 23 percent, followed by the lungs and remaining tissues at about 10 percent each.

Table 4

Results of Individual Tissue Analyses

<u>Tissue</u>	<u>Weight of Wet Sample, grams</u>	<u>Plutonium, d/m/gram</u>
Liver (1)	242.7	0.20
(2)	789.8	0.33
(3)	616.3	0.34
Lung (1)	261.0	0.11
(2)	381.8	Lost
(3)	372.5	0.28
Kidney(1)	144.5	≈ 0.0015
(2)	144.5	≈ 0.0030
Spleen	120.1	≈ 0.004
Bronchial lymph nodes	1.0	1.40
Other tissues	31.7	≈ 0.003
Rib (1)	71.3	0.09
(2)	9.0	0.39
Vertebra	56.2	0.22
Sternum (1)	35.5	0.18
(2)	69.5	0.04

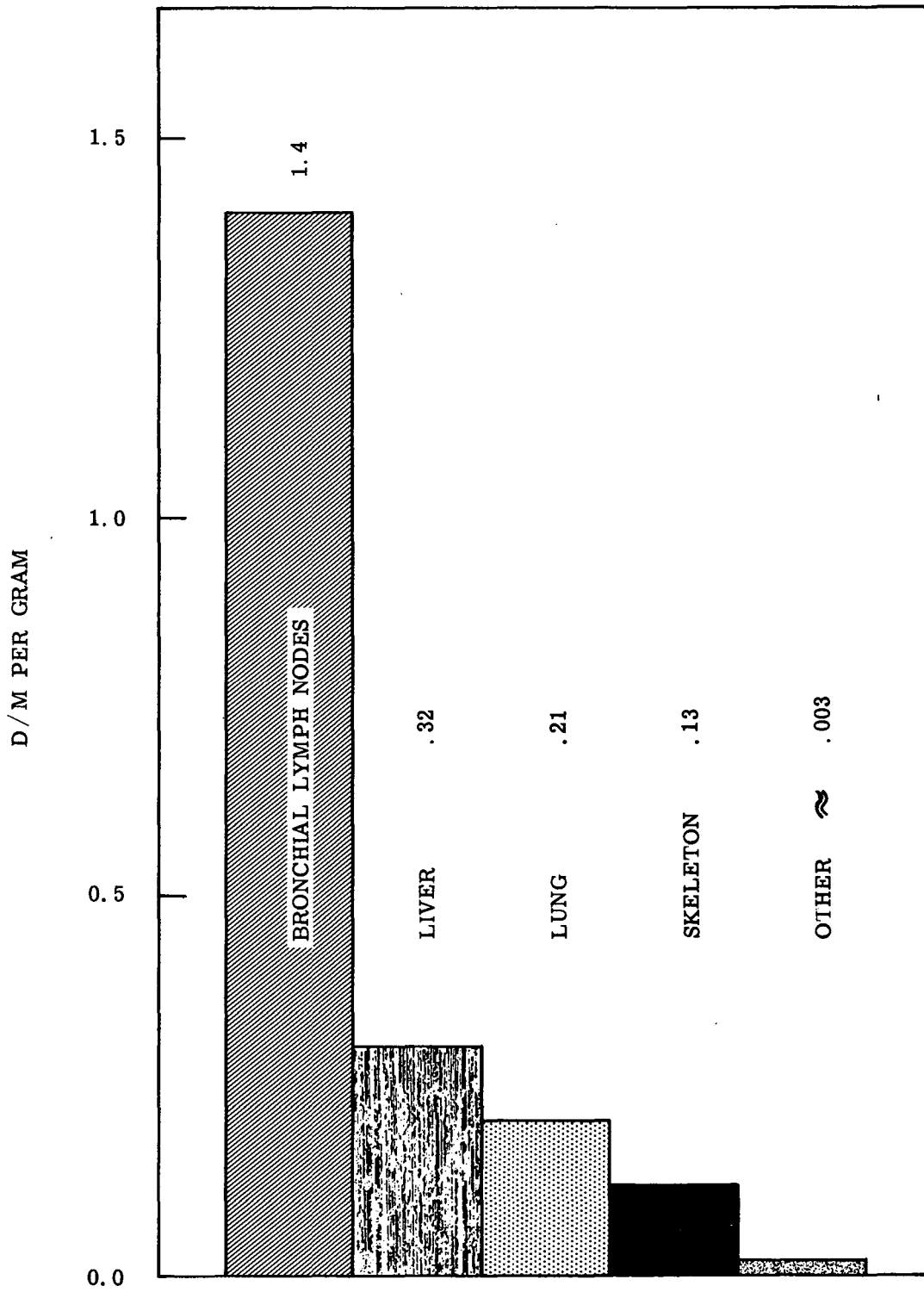


Fig. 2. Average plutonium concentrations representing relative dose rates to different organs.

Table 5

Extrapolated Body Burden of Plutonium

<u>Tissue</u>	<u>Weight (gram)</u>	<u>Average d/m/g</u>	<u>Total Content (d/m)</u>
Liver	1,649	0.32	526
Lung	1,015	0.21	214
Kidney	289	≈ 0.002	--
Spleen	120	≈ 0.004	--
Skeleton	10,000 [*]	0.13	1,300
Bronchial lymph nodes	3 ^{**}	1.4	≈ 4
Remaining tissue	69,500 ^{***}	≈ 0.003	≈ 208
			<u>2,252</u>
			(10 ⁻³ μCi) (2.4% MPBB)

* Taken from skeletal weight of the standard man.

** Estimation based on visual observation.

*** Assuming plutonium content of remaining tissue is the same as the average of the kidney, spleen, and 32-gram tissue sample from the rib area.

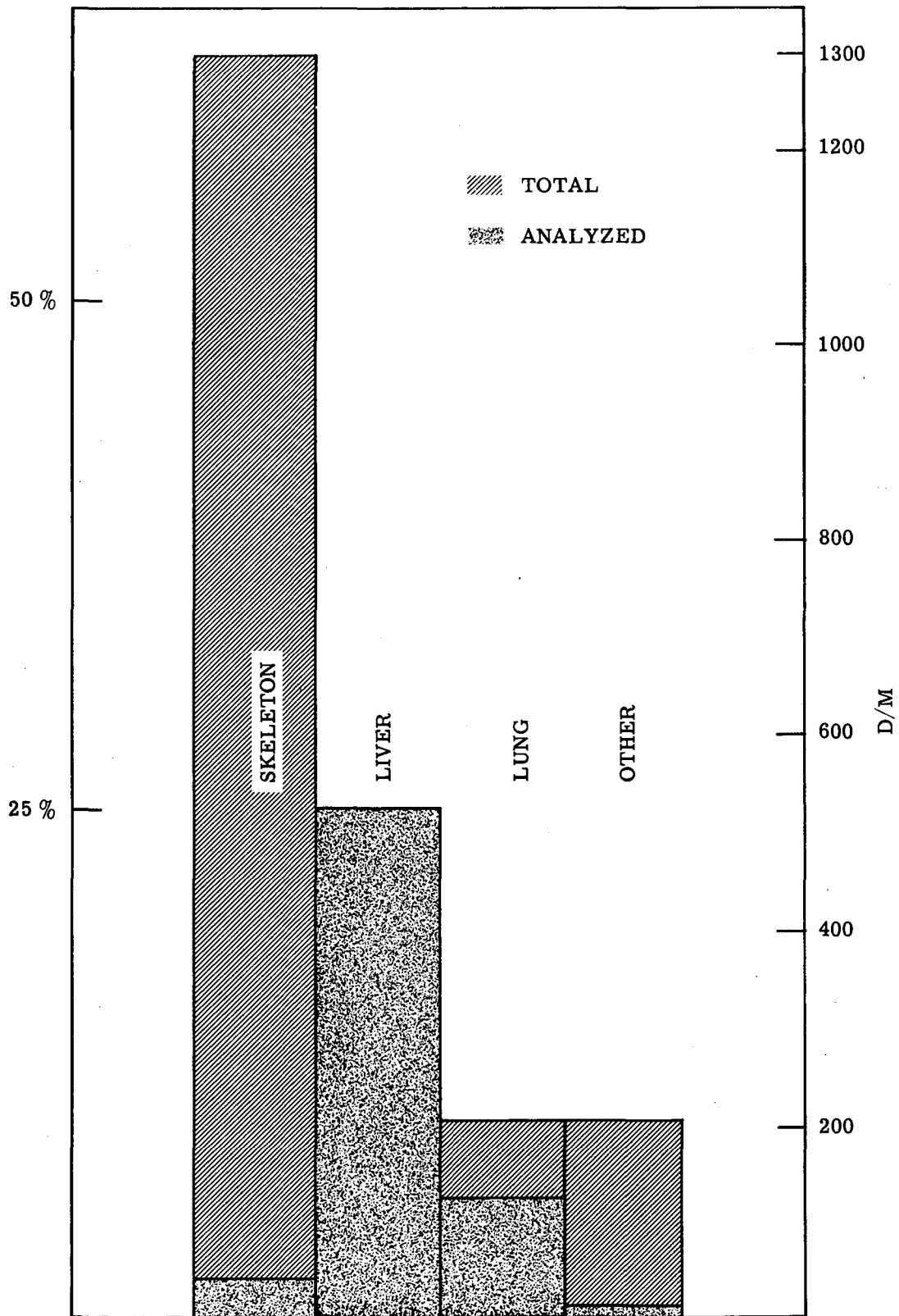


Fig. 3. Total body content of plutonium extrapolated from individual tissue samples.

Several assumptions were made in order that a total body content could be determined. First, the total content of the skeleton was determined by using the average concentration of the rib, sternum, and vertebra samples. Second, the 69,500 grams of tissue that were not analyzed were assumed to contain the same concentration as the kidney, spleen, and small (32-gram) tissue sample. Third, the total weight of the bronchial lymph nodes was taken as 3 grams. This was an estimate based on the visual observation that about one-third of the nodes was recovered and analyzed.

DISCUSSION

In the case reported here (667), more than half of the total body content was found in the skeleton compared to only about 23 percent in the liver. This is in contrast to the distribution of material in the case reported by Foreman (11) where nearly 50 percent was reported to be in the liver with about 35 percent in the skeleton. Other reported autopsy data (12,13) show that the fractional content of the skeleton, liver, and lungs will vary from case to case. This is probably caused by different physical and chemical states of the plutonium as well as different modes of exposure and lengths of time that the material has been in the body.

The extrapolation of the results from the tissue samples to the whole body indicates that there was about 2.4 percent of a maximum permissible body burden of plutonium present at the time of death. This is about a factor of 5 less than calculated using urine results.

Even though this discrepancy is large, it is not surprising. Some of the sources of error which would contribute to this discrepancy are as follows:

1. The extrapolation of the average concentration of several small samples of rib, sternum, and vertebra samples to the entire skeleton could be the source of a large error. This is particularly important since more than half of the total body content was involved.
2. The amount of plutonium in a 24-hour urine sample is so small that large percentage deviations are seen from sample to sample.
3. Snyder (3) has pointed out deviations as high as a factor of 2 in body burden calculations simply from choosing two different methods of calculation.

4. In wound cases, we suspect, there is a "short-circuiting" effect where plutonium goes directly from the wound to the urine which leads to artificially high body burden estimates.

When Foreman reported the Los Alamos case (10) he noted that the agreement between body burden found at autopsy and body burden calculated from urine results was so good as to be called "fortuitous." This good agreement may also have been somewhat unfortunate because it made the process of body burden calculation look better than it is. This case has, again, brought out the need for better knowledge of the relationship between plutonium in the urine and body burden for plutonium workers.

No attempt has yet been made, by us, to test any of the proposed excretion models that are designed to include the movement of plutonium from a reservoir in the body. Perhaps the use of one of these models with the proper selection of retention time would lead to better agreement between the calculated body burden and the extrapolated one.

REFERENCES

1. S.E. Hammond and E.A. Putzier, Observed Effects of Plutonium in Wounds over a Long Period of Time, Health Physics 10, 6 (1964).
2. G.W. Dolphin and S. Jackson, Interpretation of Bio-Assay Data, Assessment of Radioactivity in Man, Vol. I, 329, IAEA, Vienna (1964).
3. W.S. Snyder, Major Sources of Error in Interpreting Urinalysis Data to Estimate the Body Burden of Pu²³⁹: A Preliminary Study, Health Physics 8, 6 (1962).
4. S.A. Beach, G.W. Dolphin, K.P. Duncan, and H.J. Dunster, A Basis for Routine Urine Sampling of Workers Exposed to Plutonium-239, Health Physics 12, 12 (1966).
5. W.R. Wood, Jr., and W.E. Sheehan, Evaluation of the PUQFUA Method of Calculating Body Burdens, Proceedings of the 12th Annual Bio-Assay and Analytical Chemistry Meeting, CONF-661018 (1966).

6. W.H. Langham, Determination of Internally Deposited Radioactive Isotopes from Excretion Analyses, Am. Ind. Hyg. Q. 17, 3 (1956).
7. J.W. Healy, Estimation of Plutonium Lung Burden by Urine Analysis, Am. Ind. Hyg. Q. 18, 261 (1957).
8. S.A. Beach and G.W. Dolphin, Determination of Plutonium Body Burdens from Measurements of Daily Urine Excretion, Assessment of Radioactivity in Man, Vol. II, 603, IAEA, Vienna (1964).
9. J.N.P. Lawrence, PUQFUA, an IBM 704 Code for Computing Plutonium Body Burdens, Health Physics 8, 1 (1962).
10. E.E. Campbell and W.D. Moss, Determination of Plutonium in Urine by Anion Exchange, Health Physics 11, 737 (1965).
11. H. Foreman, W. Moss, and W. Langham, Plutonium Accumulation from Long-Term Occupational Exposure, Health Physics 2, 4 (1960).

12. C.E. Newton, Jr., K.R. Heid, H.V. Larson, and I.C. Nelson, Tissue Sampling for Plutonium Through an Autopsy Program, Proceedings of the 12th Annual Bio-Assay and Analytical Chemistry Meeting, CONF-661018 (1966).

13. W.H. Langham, J.N.P. Lawrence, J. McClelland, and L.H. Hempelmann, The Los Alamos Scientific Laboratory's Experience with Plutonium in Man, Health Physics 8, 753 (1962).

A STUDY OF INDIVIDUAL VARIATION OF EXCRETION OF PLUTONIUM BY MAN
AND OF ITS SIGNIFICANCE IN ESTIMATING THE SYSTEMIC BURDEN*

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Abstract

The excretion data of 15 terminal patients, reported by Langham,⁽¹⁾ who received ^{239}Pu by IV injection have been analyzed and studied in the context of the title. Power functions $bt^{-\beta}$ have been fitted to the urinary excretion data $U(t)$ by minimizing (1) the sum of deviations and (2) the sum of percent deviations. This corresponds to minimizing the sums

$$\sum_t | U(t) - bt^{-\beta} | \quad \text{and} \quad \sum_t \left| 1 - \frac{U(t)}{bt^{-\beta}} \right|,$$

respectively. This minimization produces parameter values a and α which provide a best fit to an individual's data; and by pooling all the data, one obtains values of a and α for the group. The distribution of the data $U(t)$ of an individual about his own curve of best fit and about the curve determined for the group is studied, and confidence limits P and p are determined so that the data $U(t)$ only exceed $P bt^{-\beta}$ or fall below $p bt^{-\beta}$ on a preassigned percentage of days. For example, it appears from analysis of these data that an individual's daily excretion lies between 0.3 and 3 times the power function $bt^{-\beta}$ for the group about 90% of the time. Thus, if a plutonium worker's excretion on a certain day is higher than his previous trend by more than a factor of 3, one may conclude with a

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rather high degree of confidence, perhaps 90%, that he had a new intake to blood.

If the employee's excretion is high on a certain day, but not by as much as a factor of 3, then one may conclude there is a chance of 10% or more that the excess represents only a temporary fluctuation and one need not postulate that a new intake has occurred.

The same type of analysis is used with the integral form of the power function, e.g.,

$$\int_{t-1}^t bx^{-\beta} dx = b [t^{1-\beta} - (t-1)^{1-\beta}] / (1-\beta)$$

in place of $bt^{-\beta}$. The use of these distributions in estimating body burdens is discussed and illustrated by examples.

A number of computer codes have been described which interpret the urinalysis data of an employee to obtain an estimate of his intake of plutonium or, alternatively, his body burden. ⁽²⁻⁶⁾ All of these codes are based essentially on a model for excretion following a single intake to blood which predicts excretion in terms of a power function, i. e., fraction of a single intake (to blood) excreted t days postexposure is given as $b t^{-\beta}$. An employee's exposure plausibly may be considered to consist of a series of single exposures, and the computer codes define a series of intakes at various times which, according to the model, would suffice to some degree of approximation to account for the excretion pattern as measured.

Langham et al. ⁽¹⁾ seem to have been the first to use this model for excretion of plutonium when they analyzed the data on some 15 terminal patients who were given ^{239}Pu complexed with citrate by IV injection. They also analyzed excretion data of employees and found that the excretion curves which described the trend of the patient data seemed to be in accord with the data on the employees. In this sense the model may be said to be supported by data out to five or more years postexposure.

Few attempts have been reported for taking statistical fluctuations of the excretion data into account in a systematic way. This paper is a preliminary analysis of the statistical day-to-day fluctuations of excretion data of the hospital patients reported by Langham et al., ⁽¹⁾ the different trends seen in the individual patients, and the different results found

when the data are analyzed in terms of various models or methods of curve fitting. For brevity these three sources of difference might be referred to as "statistical fluctuations," "individual differences," and "differences in models." Finally, a code is described which allows one to explore to some extent the influence of each of these sources of variability on the estimated systemic burden, and the results of some of the cases studied are shown.

The Basic Model

The basic model for excretion of plutonium which has entered blood and its use in interpreting the urinalysis data of an employee have been described in reference 6. Briefly summarized, the excretion following a single unit intake of plutonium to blood is described by a function $f(t)$ which represents the fraction of initial activity reaching blood at time 0 which is excreted on day t . Langham et al.⁽¹⁾ gave the relation

$$f(t) = b t^{-\beta} = 0.0023 t^{-0.77} \quad (1)$$

based on the study of the data on the terminal patients mentioned above. In this paper we retain the same essential idea, i. e., to represent excretion by a power function, but use different fitting procedures to obtain b and β .

If the function f is defined as above, t should be considered as a discrete variable, that is, t takes on values 1, 2, 3, ..., but $f(t)$ is not defined for fractions of a day and hence does not define an instantaneous rate of excretion. Alternatively, one may define

an instantaneous rate of excretion, and in that case the fraction of intake reaching blood at time 0 which is excreted on day t is given by the formula

$$f(t) = \int_{t-1}^t b \tau^{-\beta} d\tau = \frac{b}{1-\beta} [t^{1-\beta} - (t-1)^{1-\beta}]. \quad (2)$$

In what follows, the function defined as a power function, that is, as in Eq. (1), will be denoted by f_d to indicate that it is a function fitted to the discrete points where $t = 1, 2, 3, \dots$; whereas the excretion function defined by Eq. (2) will be denoted by f_c to indicate that it is defined in terms of a continuous rate function and represents the area under the graph of the power function representing the instantaneous rate of excretion.

In either case the parameters b and β may be determined by fitting the appropriate function to the data on the hospital patients. In this paper the curve fitting is done in two ways.

The first, represented by Eq. (3), minimizes the sum of absolute deviations of the formula about the sample urinary excretion values U_i ; and the second curve-fitting procedure, represented by Eq. (4), minimizes the sum of percent deviations of the sample values about the trend curve defined by the function f . Either the point function f_d or the area function f_c may be determined by Eqs. (3) and (4). Thus four different models for excretion following a single intake are studied in the present paper. That is, a function f may be of either type f_d or of type f_c , and the curve fitting may be done by minimizing either the sum of absolute deviations or by minimizing the sum of percent deviations as indicated by Eqs. (3) and (4).

$$\sum_{i=1}^n |U_i - f(t_i)| = \text{Min} \quad (3)$$

$$\sum_{i=1}^n \left| \frac{U_i}{f(t_i)} - 1 \right| = \text{Min.} \quad (4)$$

The curve-fitting procedures defined by Eqs. (3) and (4) have been applied to determine b and β for each of the 15 hospital patients, and the values of b and β determined in this way are shown in Tables I and II which are quoted from reference 6. It will be noted that there is a considerable spread of the values of b and β for different individuals, and this is to some extent a measure of the extent of individual variability among the patients of this group. Only the urinary data will be used in this paper, although the fecal data can be fitted by the same methods. All of the urinary excretion data of the patients may be pooled and the same minimization procedures applied to determine values of b and β which are in a sense representative for the group rather than for an individual patient. The excretion model determined for the group will be referred to in what follows as the "typical" excretion formula or excretion model. The values of b and β so determined are shown in Tables I and II.

Day-to-Day Fluctuations of Excretion Data

The excretion models determined in the preceding section only establish a trend of these data, and it is well known that there are wide fluctuations in the day-to-day excretion data. To establish some measure of the extent of this fluctuation, the distribution of the ratios $U_i/f(t_i)$ has been studied. When this ratio is less than 1, the sample value lies below the curve defining the trend of the data, and when it exceeds 1, the sample value is in excess of the trend curve. The cumulative curve for the distribution

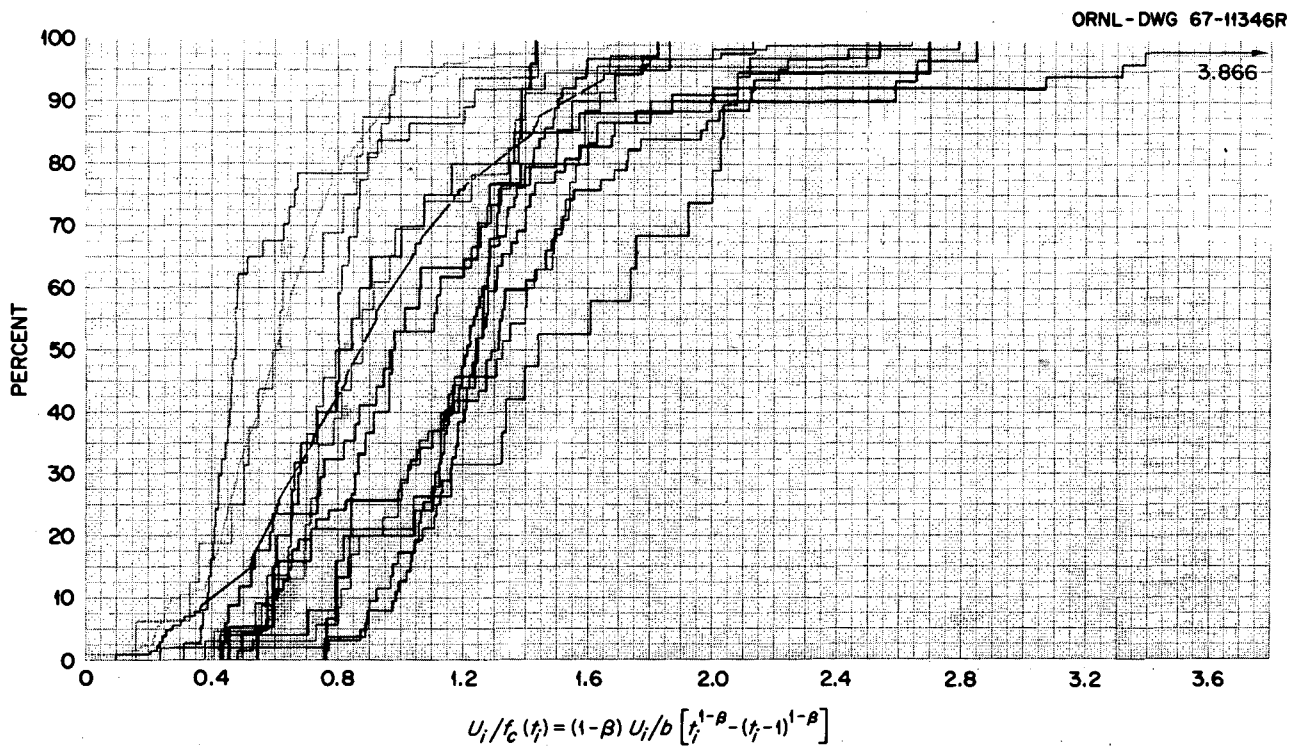
Table I. Urinary Excretion Formulas, $bt^{-\beta}$, for Fifteen Individuals
(Obtained by minimizing the sum of the absolute deviations)

Patient	"Point Fit"		"Area Fit"	
	b	β	b	β
Hp-1	0.24	0.74	0.10	0.42
Hp-2	0.47	1.04	0.17	0.63
Hp-3	0.38	0.99	0.17	0.71
Hp-4	0.44	0.89	0.20	0.54
Hp-5	0.30	1.10	0.11	0.62
Hp-6	0.50	1.21	0.31	0.99
Hp-7	0.24	1.02	0.078	0.64
Hp-8	0.38	0.88	0.16	0.58
Hp-9	0.12	0.48	0.096	0.40
Hp-10	0.41	0.98	0.17	0.60
Hp-12	0.15	0.58	0.087	0.40
Chi-I	0.86	1.85	0.18	0.79
Chi-II	0.25	0.73	0.20	0.66
Chi-III	0.15	0.86	0.071	0.53
Cal-I	0.48	1.19	0.12	0.74
All the patients	0.32	0.93	0.15	0.65

Table II. Urinary Excretion Formulas, $bt^{-\beta}$, for Fifteen Individuals
(Obtained by minimizing the sum of the percent deviations)

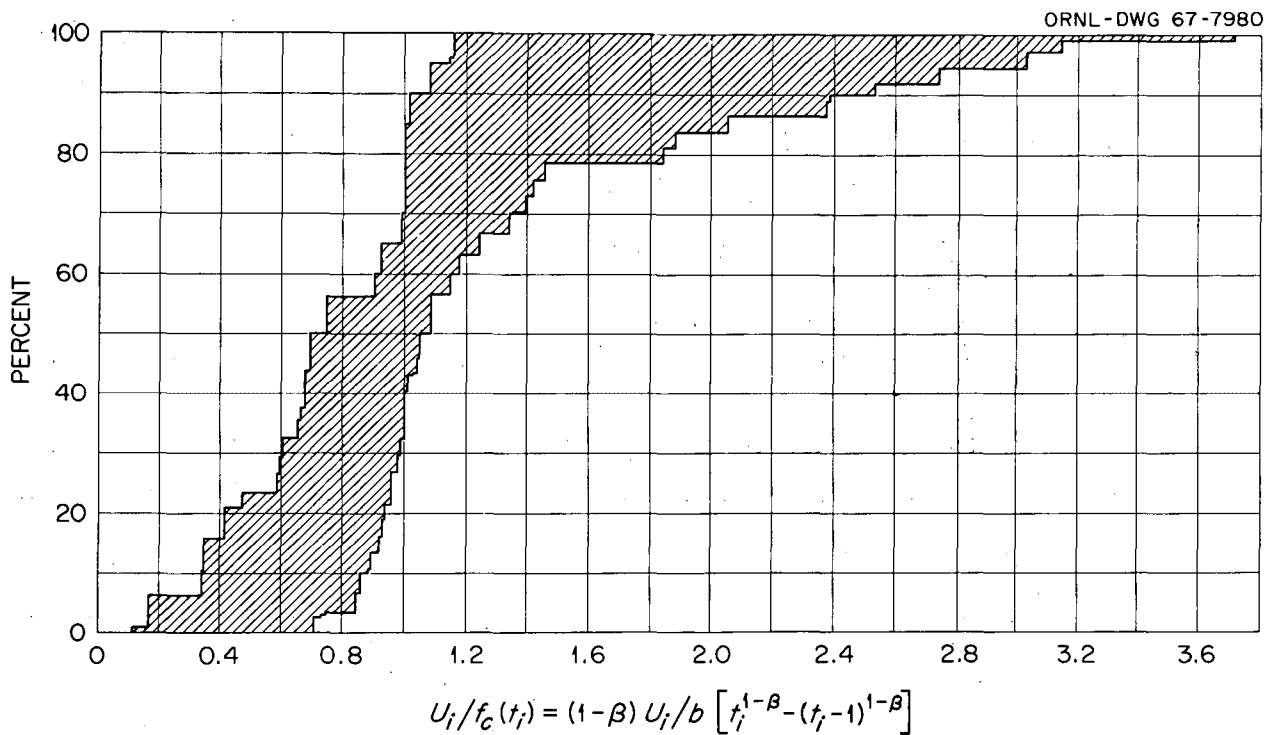
Patient	"Point Fit"		"Area Fit"	
	b	β	b	β
Hp-1	0.28	0.82	0.18	0.65
Hp-2	0.66	1.21	0.17	0.63
Hp-3	0.57	1.20	0.16	0.72
Hp-4	0.44	0.89	0.19	0.56
Hp-5	0.18	0.79	0.11	0.62
Hp-6	0.50	1.21	0.31	0.99
Hp-7	0.48	1.26	0.089	0.75
Hp-8	0.32	0.81	0.23	0.73
Hp-9	0.12	0.47	0.096	0.40
Hp-10	0.73	1.22	0.16	0.72
Hp-12	0.15	0.57	0.12	0.52
Chi-I	0.11	0.61	0.099	0.57
Chi-II	0.17	0.62	0.16	0.60
Chi-III	0.15	0.83	0.083	0.45
Cal-I	0.26	0.88	0.13	0.72
All the patients	0.21	0.73	0.14	0.61

of these ratios has been determined in each case. This cumulative curve rises from the value 0 at the smallest value of the ratio and attains the value 1 for the largest value of the ratio in each individual case. Figure 1 shows a typical example of these curves for the 15 hospital patients. The excretion function f_c used here is that determined for the fluctuations around the typical excretion formula defined by Eq. (3) for the group. Figures 2, 3, and 4 show the similar data for other versions of the excretion model, but the individual curves have been omitted so that only the broad band containing all the individual curves is shown. From this band one can read off the extreme values of the ratio which would contain a specified percentage of the daily excretion values for each of the individuals. For example, using Fig. 1 it is apparent that for none of the patients was there more than 10% of the daily excretion data for which the ratio fell below 0.30 and for no patient were as many as 10% of the daily excretion values in excess of the number 2.59. Thus an 80% range for the ratio of the daily sample to the trend curve for the group is given by the range of ratios from 0.30 to 2.59. These distributions provide a measure of the extent of day-to-day fluctuations of an individual about his own trend curve or of an arbitrary individual of the group about the group trend curve. One may well hesitate to apply either of these measures of day-to-day fluctuations to employees whose individual trend curves are unknown and where there is little information on the extent of fluctuations of the group. Nevertheless, this is almost the totality of data of this kind that are available. One may mention, however, the study of Beach and Dolphin⁽⁷⁾ who obtained distributions grossly similar to these based on employee data which were normalized in terms of their body burdens of plutonium.



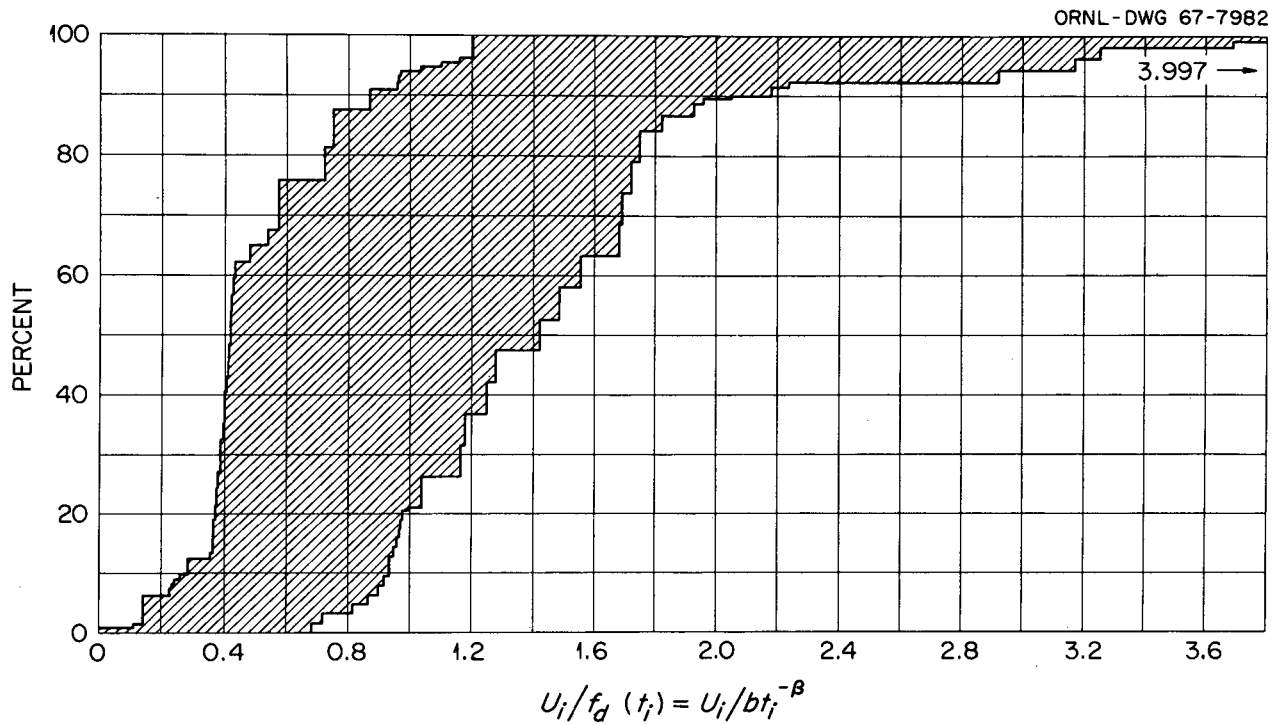
Fluctuations of Daily Urinary Excretion Data of all Patients (Langham-1950) about the "Typical" Formula of Best Fit - "Area Fit" Formula Obtained by Minimizing the Sum of Absolute Deviations.

Fig. 1



Fluctuations of Daily Urinary Excretion Data of all Patients (Langham-1950) about their Own Formula of Best Fit — "Area Fit" Formulas Obtained by Minimizing the Sum of Percent Deviations.

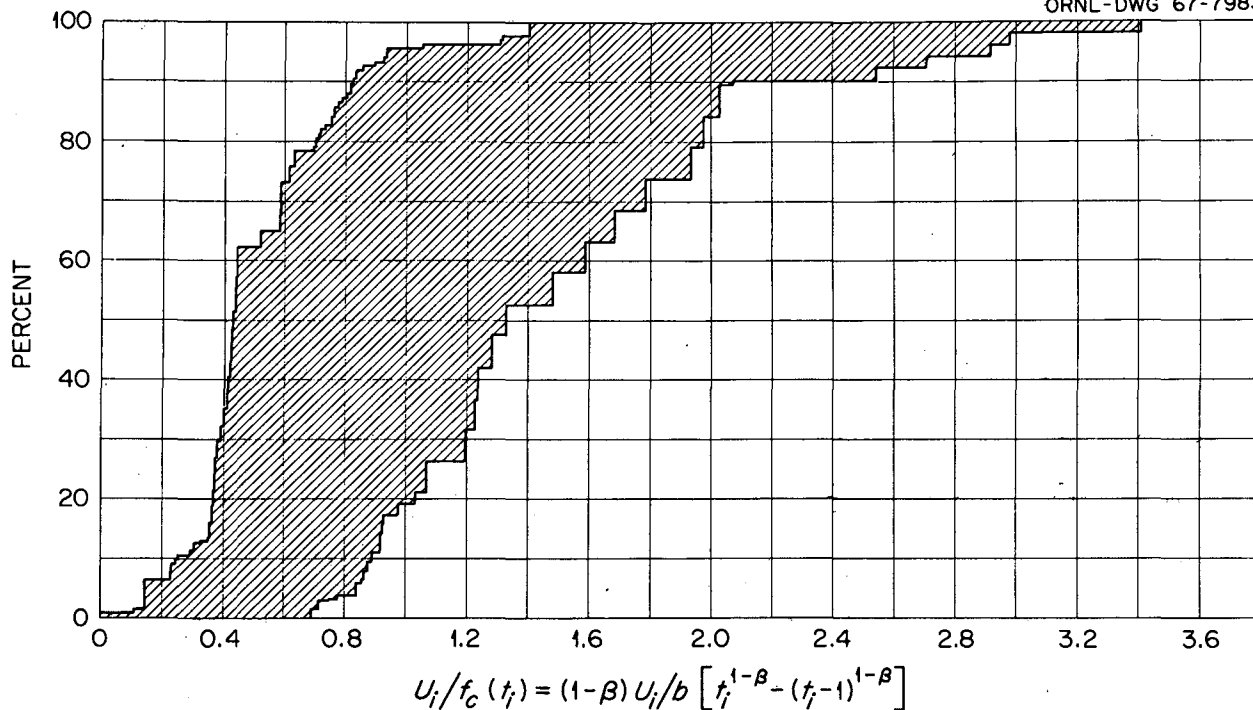
Fig. 2



Fluctuations of Daily Urinary Excretion Data of all Patients (Langham-1950) about the "Typical" Formula of Best Fit - "Point Fit" Formula Obtained by Minimizing the Sum of Percent Deviations.

Fig. 3

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Fluctuations of Daily Urinary Excretion Data of all Patients (Langham-1950) about the "Typical" Formula of Best Fit - "Area Fit" Formula Obtained by Minimizing the Sum of Percent Deviations:

Fig. 4

A Computer Code for Estimation of the Intake of Plutonium to Blood
on the Basis of Excretion Data

The input for the code consists of the sample values U_i and the days t_i on which the sample was taken. It is assumed that each sample represents a 24-hr period of excretion. Essentially the design of the code is based on the supposition that an intake is potentially possible between each successive pair of sample values, and the code is designed to produce intakes which will reproduce the excretion data as nearly as possible on the basis of the model. Basically this is achieved in the following way. Assuming that the data on excretion are given by

$$U_i, t_i \quad i = 1, 2, 3, \dots, n \quad (5)$$

one seeks to determine intake values

$$I_i \text{ on day } \tau_i \quad i = 1, 2, 3, \dots, n. \quad (6)$$

Since the intakes to blood must be spaced between the sample values, one requires that

$$t_{i-1} < \tau_i \leq t_i \quad i = 1, 2, 3, \dots, n. \quad (7)$$

The first intake is determined as in Eq. (8) where the quantity $t_1 - \tau_1 + 1$ represents the number of days from the first intake on day τ_1 to the first sample day t_1 . The convention has been made that intake is assumed to occur at the beginning of the day while the sample collection is only complete at the end of the day. There is a certain

degree of indeterminacy in Eq. (8) in that the function f may be any one of the functions as defined in Tables I and II and the variable τ_1 may be any day subject to the inequalities given by (7), that is, $0 < \tau_1 \leq t_1$. Whatever the choice of the function f or of the day τ_1 , the intake I_1 on day τ_1 will exactly reproduce the sample value U_1 according to the chosen model. In principle, the computer code then proceeds by induction. If intakes I_j have been defined for the first $i - 1$ cases, i.e., $j = 1, 2, \dots, i - 1$, these intakes already account for some of the activity found in the sample on day t_i . This amount is represented by the sum in formula (9) which is then subtracted from U_i , so that only the remainder, i.e., the bracketed quantity, needs to be accounted for by the new intake I_i .

$$I_1 = U_1 / f(t_1 - \tau_1 + 1) \quad (8)$$

$$I_i = \left[U_i - \sum_{j=1}^{i-1} I_j f(t_i - \tau_j + 1) \right] / f(t_i - \tau_i + 1) \quad (9)$$

One continues in this way to define successively the intakes I_i , and this would be a complete and rigorous solution to the problem if individuals exactly followed the trend formula defining f . Unfortunately, there are wide day-to-day fluctuations of the excretion data about the trend curve, and one cannot proceed in such a simple fashion as that indicated above. It may, for example, happen that the bracketed term in Eq. (9) may be negative, i.e., the previous intakes already more than account for the excretion as seen on day i . In this case no new intake is necessary, since an excess has already been accounted for according to the model. That this situation may occur is no cause for

surprise since it may happen that the sample collected on day i is one which fell below the trend curve, that is, it was an unusually low output for that particular day. However, in such case, one need not seek for a positive intake, and one merely sets $I_i = 0$.

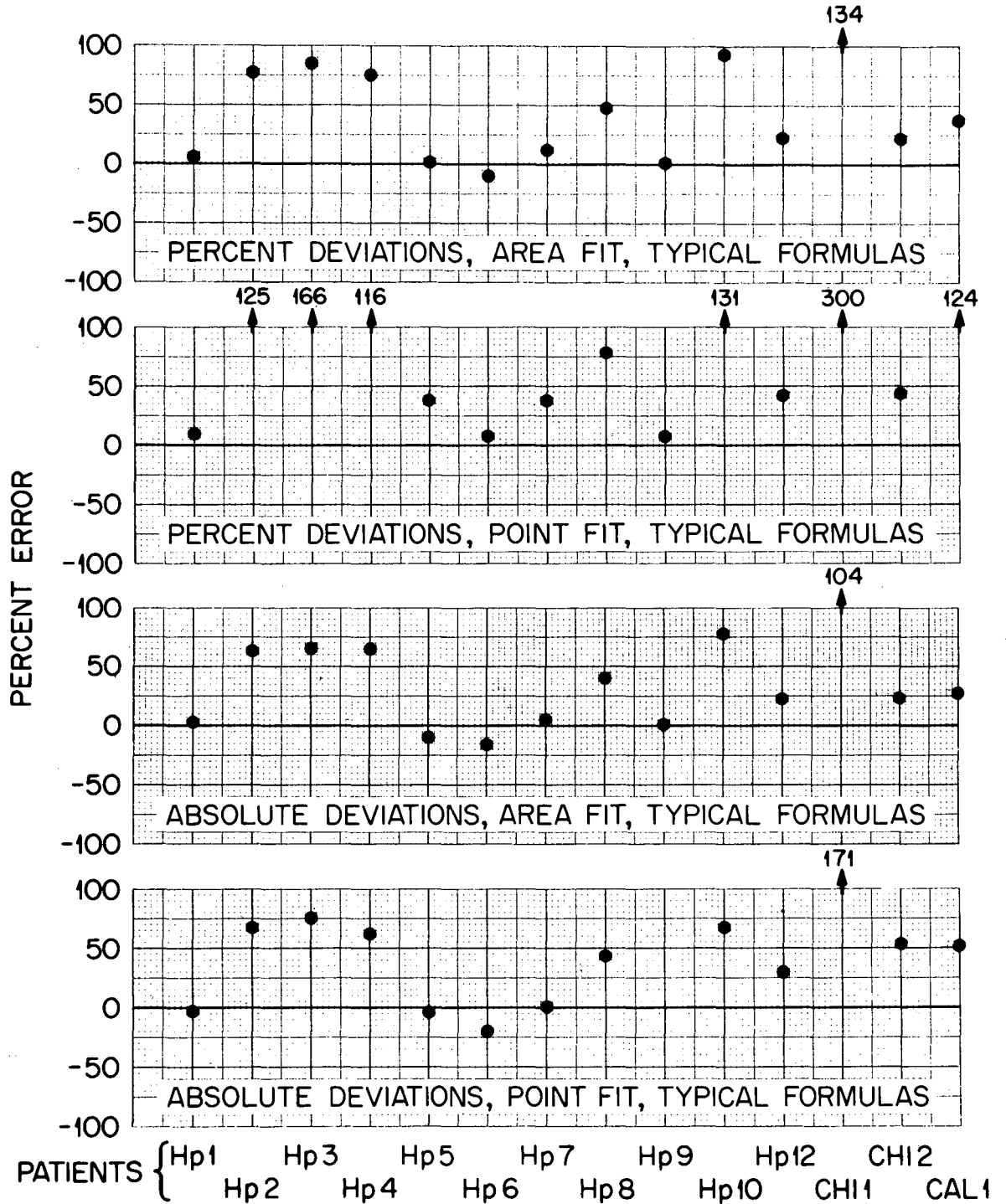
Likewise, it may happen that the numerator of formula (9) is not very large compared to U_i ; that is, the previous intakes already account for most of U_i , or perhaps account for U_i within a fairly small factor. In this case one may be tempted to regard the excess as merely due to chance, i. e., as one of the high daily fluctuations. One may make this precise by using the distributions given in Figs. 1-4; that is, one may assign confidence limits within which the day-to-day fluctuations appear to lie with a certain degree of probability. Thus the values 0.30 and 2.59 specified earlier and based on Fig. (1) would be values such that, for that excretion model, in no patient did the ratio of the daily excretion to the trend formula lie outside this range in more than 20% of the cases. Using only the high value of 2.59, one may say that if an excess by more than this amount occurs, one has 90% confidence that there has been additional intake. One may then introduce into the code a test so that if the ratio satisfies the inequality (10) where P is chosen to represent the upper limit of daily fluctuations at the prescribed confidence level, then and only then does one postulate a new intake. Otherwise, if the ratio does not exceed P , one may postulate no additional intake but regard the excess represented by the bracket in formula (9) as being merely due to chance fluctuations.

$$U_i / \sum_{j=1}^{i-1} I_j f(t_i - \tau_j + 1) > P \quad (10)$$

The procedures mentioned above represent only a few of the possibilities which can be made a part of the code for estimation of systemic body burden or, alternatively, of intake to blood of plutonium. One may take the code as so designed, test it by using the excretion data of any or all of the hospital patients, and see with what degree of accuracy the code will predict the actual intake. This has been done for a number of the different models discussed above and at various confidence levels for the fluctuations, and the results are shown graphically in Figs. 5 and 6. Actually, no one of the models appears to be greatly superior to any one of the others, nor do the confidence levels tested thus far for taking account of daily fluctuations seem to make any great difference in the final estimate. In a few cases excessively high values of intake seem to be obtained. These are notably absent when the model uses the area formula, especially that where the absolute deviations are minimized. It appears that when no allowance is made for fluctuations the estimates tend to be high, and this is understandable because each high fluctuation may require some new intake to account for the excess. When allowance is made for fluctuations, this tendency is less pronounced. The cases tried here represent only a few of the many possibilities which may be tested, and various other ways of taking daily fluctuations into account are under study. It is hoped that this approach will eventually lead to a procedure which will not require that a new intake be prescribed for each high value that occurs since some of these may indeed be merely chance fluctuations of the data.

There remains one additional case in which the body burden is known, and that is the case reported by Foreman et al.⁽⁸⁾ of a plutonium worker who was killed

ORNL-DWG 67-11018

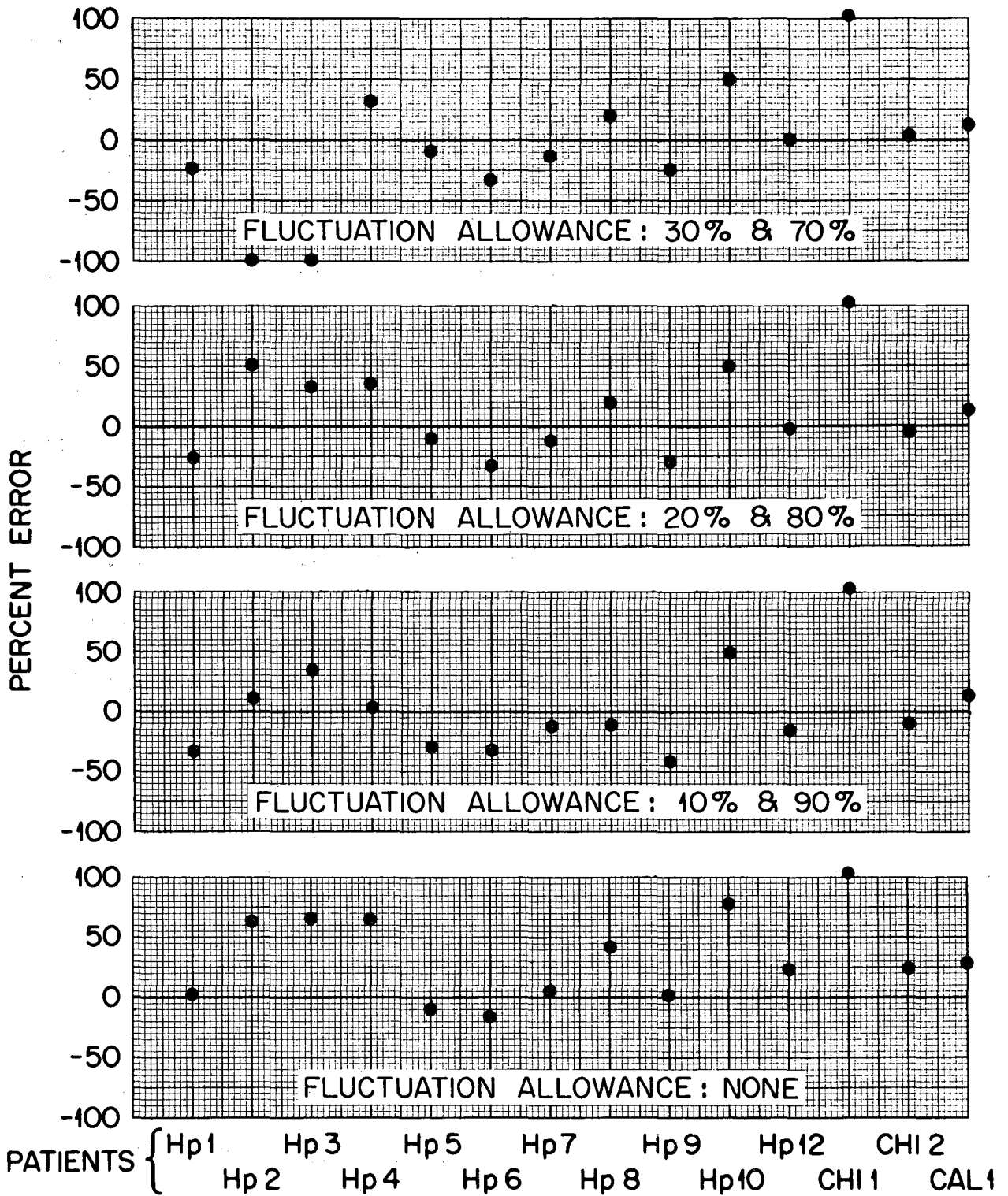


Percent Error of Estimated Intake.

(FLUCTUATION ALLOWANCE: NONE)

Fig. 5

ORNL-DWG 67-11200



Percent Error of Estimated Intake.

(ABSOLUTE DEVIATIONS, AREA FIT, TYPICAL FORMULA)

Fig. 6

in a criticality accident in 1958. The case has been studied previously by these methods (see refs. 2 and 3), but it may be of interest to compare the values obtained by the present code with some of those reported earlier. Our study of the case leads to estimates of about 0.0075 μCi if the area model is used and 0.013 μCi if the point model is used. As with the hospital patients, the confidence level used does not materially affect these estimates. Since excretion is small, one may regard these as an estimate of the systemic body burden which on the basis of autopsy data would be about 0.015 μCi . This estimate includes the amounts present in skeleton, liver, muscle, heart, spleen, kidneys, and the balance of the body but excludes that present in lung and pulmonary lymph nodes.

It would be of great value to test any or all of these models on further employee data that may be available, and the author invites any who may have such data to supply it to him if possible, and he will be glad to provide the estimates of systemic body burden that are obtained by these methods.

References

1. W. H. Langham, S. H. Bassett, P. S. Harris, and R. E. Carter, Los Alamos Scientific Laboratory, LA-1151 (1950).
2. James N. P. Lawrence, Health Phys. 8(1), 61 (1962).
3. W. S. Snyder, Argonne National Laboratory, ANL-6637, p. 13 (1961).
4. W. S. Snyder, Health Phys. 8(6), 767 (1962).
5. W. S. Snyder, p. 583 in Assessment of Radioactivity in Man, Vol. II (International Atomic Energy Agency, Vienna, 1964).

6. W. S. Snyder, Mary R. Ford, and G. G. Warner, "The Use of Excretion Data to Predict the Systemic Body Burden of Plutonium," Proceedings of Symposium on Diagnosis and Treatment of Deposited Radionuclides, Richland, Washington, May 15-17, 1967 (to be published).
7. S. A. Beach and G. W. Dolphin, p. 603 in Assessment of Radioactivity in Man, Vol. II (International Atomic Energy Agency, Vienna, 1964).
8. H. Foreman, W. Moss, and W. H. Langham, Health Phys. 2(4), 326 (1960).

DISTRIBUTION OF ^{210}Pb AND ^{226}Ra IN SOIL

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The ^{226}Ra and ^{210}Pb content of typical tobacco-growing soil was studied as a function of depth. The depth profile samples, including surface vegetation, were collected at an undisturbed area in Maryland. It was of interest to determine whether these nuclides concentrate in the upper horizons of this soil type.

^{210}Pb generally decreased with increasing depth, resulting in a decrease in the $^{210}\text{Pb}/^{226}\text{Ra}$ ratio. An attempt was made to correlate this trend with the known radon emanation rates from soil.

DISTRIBUTION OF LEAD-210 AND RADIUM-226 IN SOIL

Introduction

Soil is an essential link in the food chain. For artificial radionuclides introduced into the atmosphere, soil acts as a fallout collector. In this respect, soil is an integrating collector and has been used for many years as an indication of total strontium-90 fallout on the earth's surface. In the case of naturally occurring nuclides, soil can act both as a source of radioactivity and as a collector. An important naturally occurring nuclide in soil is radium-226. Radon-222, the gaseous daughter of radium, decays through a series of short-lived solid daughter products to 22-year lead-210 then to 5-day bismuth-210, 138-day polonium-210, and ultimately to stable lead-206. Since radon is a gas, it diffuses from the soil surface and is transported in a complex way by turbulence to great heights. Radon levels have been measured recently well into the stratosphere⁽¹⁾. While it is in the atmosphere radon decays, forming particulate lead-210, which in turn is carried back to the earth's surface by the usual fallout mechanisms.

In 1964, Radford and Hunt⁽²⁾ found polonium-210 in cigarettes. This finding was of some interest, since polonium-210 is volatile at the combustion temperature of manufactured tobacco. The same year, the U. S. Department of Agriculture requested that HASL verify these data, and we received a number of tobacco samples and tobacco soils from their laboratories in Beltsville, Maryland. The samples included typical cigarette and cigar tobaccos as well as corresponding fertilized soils from the southern growing region of the United States. These samples were processed not only for polonium-210, but

also radium-226. The average radium content of the tobacco-growing soil was found to be 0.8 pCi/g and the polonium-210 0.5 pCi/g⁽³⁾. The radium value is in good agreement with the abundance of radium in soil reported by others of about 1 pCi/g⁽⁴⁾. The average polonium-210/radium-226 ratio in these soils was about 0.5 and in the tobacco samples about 1.6. With the polonium-210 and the harvest time known, the lead-210/radium-226 ratio of 1.8 was calculated in these tobacco samples. Thus, an "observed ratio" for the lead-210/radium-226 in this tobacco-soil system may be computed as 3.6.

Because of the results of these initial measurements, the distribution of polonium-210, lead-210, and radium-226 in tobacco and soils has been under continuous study^(5,6). Most recently, an investigation was begun to determine the total lead-210 concentrations in soil as a function of depth to better understand its effect on uptake in the tobacco plant. For this, Dr. Lyle Alexander of the USDA collected a soil profile in an undisturbed area in Maryland from the surface to a depth of two feet for the lead-210 and radium-226 measurements. The core was divided into either one- or two-inch increment samples. Surface vegetation was also collected.

The over-all evaluation of these samples required not only a chemical analysis for lead-210 and radium-226 in the various increments of the core, but the construction of a theoretical model to account for the various complex physical parameters affecting the distribution of the lead-210.

Methods

Each soil sample was well mixed to assure uniform composition. The surface vegetation and soil samples were processed radiochemically for lead-210 using a solvent extraction technique developed by Petrow and Cover⁽⁷⁾. Briefly,

five grams of soil containing 10 mg of lead carrier were dissolved in concentrated nitric acid and 48% hydrofluoric acid. The clear solution was converted to a 3M hydrobromic acid medium and transferred to a separatory funnel. The solution was extracted with 50 ml of brominated Aliquat-336. Lead, bismuth, and polonium remained in the Aliquat-336, which was washed three times with 25-ml portions of 0.1M hydrobromic acid. The lead and a small amount of bismuth were extracted from the organic phase with two 25-ml portions of concentrated hydrochloric acid. The strip solutions were combined. Five milligrams of bismuth hold-back carrier and 4 ml of 9M sulfuric acid were added to the strip solution. The solvent, containing polonium-210 and impurities, was discarded. The solution was evaporated to dense sulfuric acid fumes and the organic matter volatilized with a few drops of nitric acid. The solution was cooled and 25 ml of distilled water were added to precipitate lead sulfate. The lead sulfate precipitate was filtered onto preweighted Whatman #42 filter paper and dried at 110°C. The chemical yield was then determined gravimetrically.

The precipitates were mounted on 1" Nylon disc, covered with a diffuse beta phosphor⁽⁸⁾ and Mylar. The samples were counted after 14 days to allow for the build-up of bismuth-210. The activity of the reagent blank averaged about 1 net count per minute. The beta counter background averaged about 2 counts per minute, and the efficiency for bismuth-210 was 33% for a sample containing 10 milligrams of lead carrier. The average chemical yield was 92%.

For radium-226 determination, one gram of soil was fused with sodium carbonate along with barium carrier and barium-133 tracer. The melt was water leached, filtered, and dissolved in hydrochloric acid. The radium and barium were precipitated with ammonium sulfate. The radium-barium sulfate was fumed

with sulfuric and hydrofluoric acids to remove traces of silica. The purified radium-barium sulfate was dissolved in EDTA and triethanolamine. The chemical recovery was determined by gamma counting the barium-133 tracer in the final solution.

The radium-226 content was determined by the radon emanation technique⁽⁹⁾. The reagent blank activity averaged 0.04 pCi per sample. The average radium recovery was 94%.

Discussion

The results of the radium and lead measurements along with the soil densities are reported in Table I. In this shallow soil profile, the lead-210 concentrations decrease rapidly from a surface value of 2.5 pCi/g to a minimum value of 0.59 pCi/g at about 11 inches. The profiles are shown in Figure 1.

The lead-210/radium-226 ratios, as a function of depth, are presented in Figure 2. A least-squares fit of the data shows that the ratio crosses unity (radioactive equilibrium) at about 11 inches. Although this ratio generally decreases with increasing depth over the measured range, the ratio should return to unity at some point beyond the depth of our measurements. Within the study depth an attempt has been made to explain the lead-210 results in the following manner. The total lead-210 concentration is the sum of three components: lead-210 produced within the soil crystal from radon which never diffused out of the crystal, lead-210 produced from radon diffusing and decaying in the soil gas and, lastly, lead-210 fallout, produced by the decay of atmospheric radon, which is washed back into the soil.

In order to separate the lead-210 into its three components, the assumption was made that the distribution of lead-210 and strontium-90 fallout in

Table I

Measured and Calculated Lead-210 and Radium-226 as a Function of Depth

Depth (Inches)	pCi Pb ²¹⁰ g	pCi Ra ²²⁶ g	Soil or Vegetation Density (g/cm ³)	pCi Pb ²¹⁰ cm ³	pCi Ra ²²⁶ cm ³	Pb ²¹⁰ Ra ²²⁶
Surface Vegetation	3.45 ± .08*	0.34 ± .03*	0.36	1.25 ± .03	0.123 ± .02	10.1
0-1	2.54 ± .20	1.52 ± .08	1.20	3.04 ± .23	1.82 ± .10	1.67
1-2	1.77 ± .17	1.34 ± 0	1.16	2.05 ± .20	1.55 ± 0	1.32
2-3	1.82 ± .16	1.18 ± .05	1.09	1.99 ± .17	1.29 ± .05	1.54
3-4	1.39 ± .18	1.02 ± .01	1.25	1.74 ± .23	1.28 ± .01	1.36
4-5	1.01 ± .16	1.14 ± .02	1.22	1.23 ± .20	1.39 ± .02	0.89
5-6	1.07 ± .15	1.33 ± .16	1.14	1.22 ± .17	1.51 ± .19	0.80
6-8	0.91 ± .14	0.80 ± 0	1.36	1.23 ± .19	1.08 ± 0	1.14
8-10	0.81 ± .14	0.65 ± .06	1.06	0.86 ± .15	0.69 ± .06	1.25
10-12	0.62 ± .14	0.59 ± .04	1.06	0.66 ± .15	0.62 ± .04	1.05
12-14	0.86 ± .14	0.70 ± .03	1.12	0.97 ± .16	0.79 ± .03	1.23
14-16	0.59 ± .14	0.76 ± .05	1.26	0.74 ± .18	0.96 ± .06	0.78
16-18	0.77 ± .14	0.78 ± .01	1.35	1.04 ± .19	1.05 ± .01	0.99
18-20	0.72 ± .14	0.94 ± 0	1.37	0.98 ± .19	1.28 ± 0	0.77
20-22	0.85 ± .14	1.20 ± .05	1.49	1.27 ± .21	1.79 ± .08	0.71
22-24	0.61 ± .14	1.10 ± .16	1.52	0.92 ± .21	1.67 ± .24	0.55

* One standard deviation due to counting error.

+ One standard deviation (Gaussian) of duplicate radon emanation.

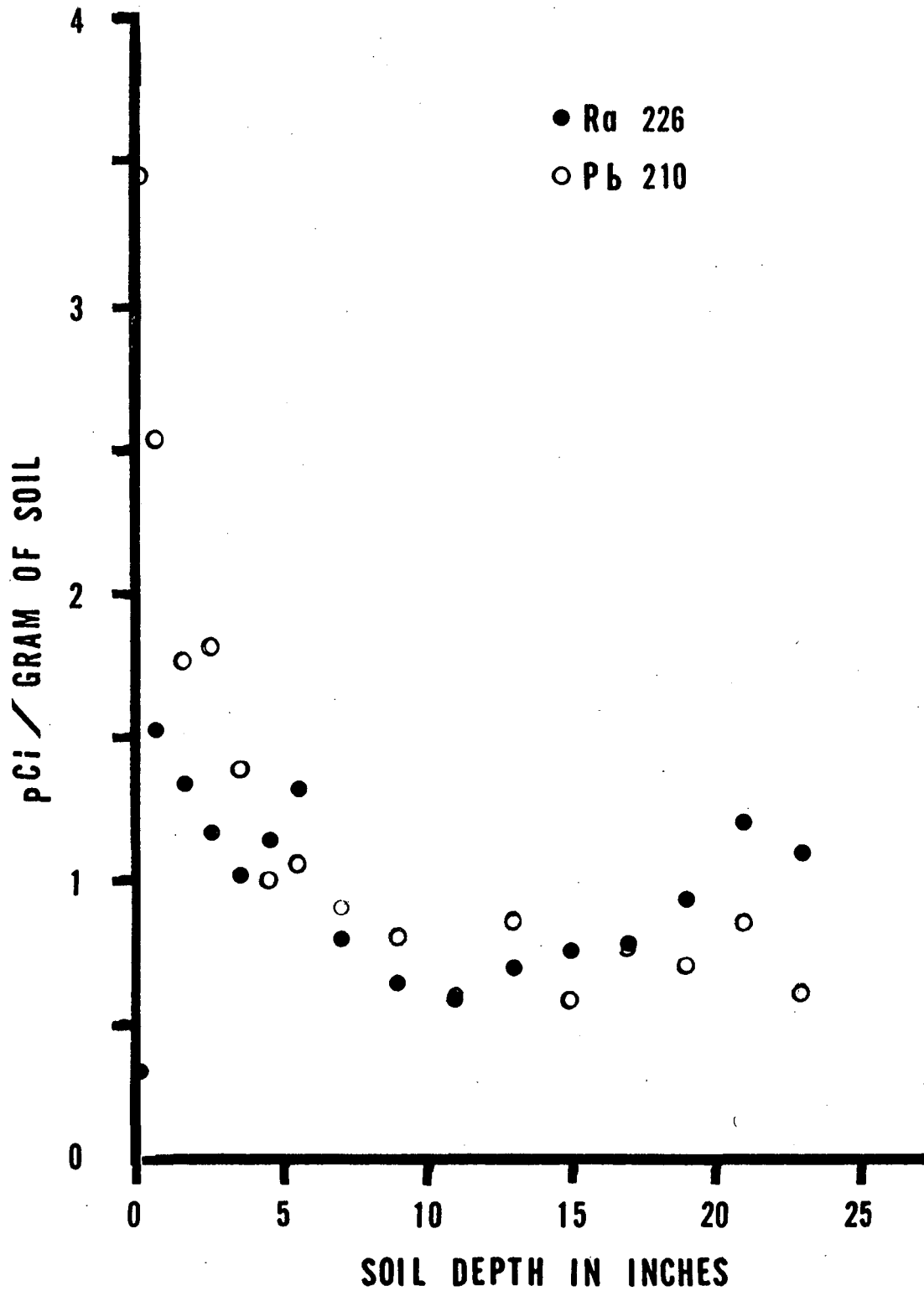


Fig. 1. Measured Lead-210 and Radium-226 as a Function of Depth.

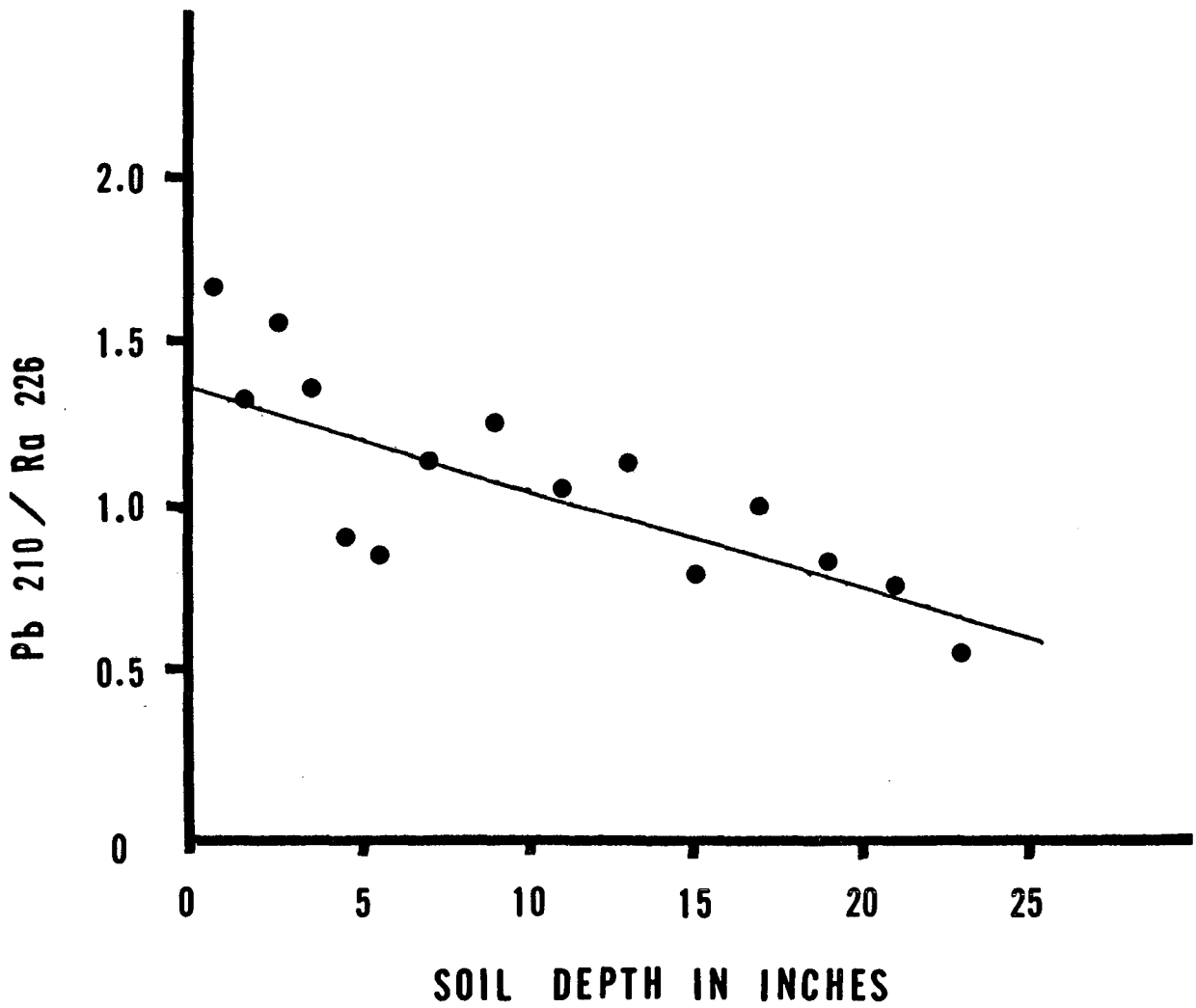


Fig. 2. Lead-210/Radium-226 Ratio as a Function of Depth.

soil are similar. These elements are chemically similar and the half-lives are comparable, so this assumption appears to be reasonable. Essentially one hundred per cent of the strontium-90 fallout which deposits on soil is found from 0 to 8 inches^(10,11). Therefore, it is assumed that the lead-210 concentrations below about 10 inches were unaffected by fallout. The lead-210 produced directly within the soil crystal from radon which never diffused into the soil gas may be expressed simply as a fraction of the radium-226 concentration of the soil at a given depth. The remaining fraction of the radium may be thought to support radon which diffuses through the soil gas in a random manner. Some radon is lost from the soil surface and it is this radon which gives rise to the radon and lead-210 levels observed in the atmosphere.

All of the radon diffusing within the soil gas is not lost to the atmosphere. We may think of a steady-state radon profile existing in soil supported by the radium which sustains the radon diffusion. Lead-210 will then be formed in the soil gas by this radon. Since we are considering the long-term formation of lead-210 from a steady-state radon concentration, the lead disintegration rate is equal to the radon disintegration rate. Obviously, at some great depth in soil the radium and lead activities should be equal.

Under these conditions, lead-210 concentrations below 10 inches may be represented by the expression

$$\text{Measured Pb-210 concentration} = [(\text{Rn concentration in soil gas})] + [f(\text{Measured Ra-226 concentration})], \quad (1)$$

The next step is to construct the steady-state soil gas radon profile as a

function of depth. The equilibrium concentration of radon in soil gas is expressed by the diffusion equation:

$$\frac{\partial C_s}{\partial t} = 0 = D \frac{\partial^2 C_s}{\partial z^2} + a - \lambda C_s. \quad (2)$$

In order to solve this equation we must consider the simple case of a constant source of radium with depth. The equation then simplifies to

$$C_s = \frac{a}{\lambda} \left[1 - \exp(-\sqrt{\lambda/d} \cdot z) \right], \quad (3)$$

where C_s = concentration of radon in soil gas (atoms/cm³),
 a = production rate of radon in soil gas (atoms/cm³·sec),
 λ = decay constant of radon (sec⁻¹),
 d = diffusion coefficient (cm²/sec),
 z = soil depth (cm).

An average radium-226 value of 1.25 pCi/cm³ was computed from the measured values in pCi/g and the known soil densities. Although the radium profile is not constant with depth, this assumption should not lead to gross errors in the calculated radon profile. Assuming a diffusion coefficient of 0.05 cm²/sec, a radon profile was calculated from 10 to 24 inches, or 25 to 59 centimeters. The radon increases exponentially with depth. To determine the two components of lead-210 distribution described by equation (1) we work first with the data below 10 inches, where we assume no fallout lead-210 exists. It was found that if in equation (1) we assume that 75% of the radon decays within the soil crystal to form lead-210, and 25% of the radon is available

for diffusion, then the calculated lead-210 below 10 inches is in good agreement with the measured lead values.

Subtracting the calculated lead-210 due to diffusible radon from the lead-210 measurements yields lead-210 values which are the sum of lead-210 from non-diffusible radon and fallout lead-210. Again, by subtracting 0.75 of the radium value, which is equal to the lead-210 from non-diffusing radon, a net lead-210 curve is computed which represents fallout lead-210. The fallout lead-210 appears to penetrate to a depth of about 11 inches, which is in fair agreement with known strontium-90 soil profiles. The separated components are shown in Figure 3.

In the theorized system a material balance of lead-210 and radium-226 activities should exist. However, the sampling depth is too shallow to justify further computations. It should be noted that lead-210 decay while it is in the atmosphere has essentially no effect on the total system. Lead-210 is thought to be uniformly mixed in the troposphere⁽¹²⁾. The integrated amount of lead-210 in 1 cm² from 0 to 60 centimeters is 73 pCi and therefore the amount of lead-210 supported in the atmosphere is negligible compared to that in the source. Patterson and Lockhart⁽¹³⁾ have reported ground-level lead-210 measurements of about 4×10^{-2} dpm/m³ with an associated radon concentration of about 270 dpm/m³. A column of air above a 1-cm² area of soil to a height of 40,000 feet contains a volume of 1.22 m³. Based on Patterson's measurements, the air column would contain 4.9×10^{-2} dpm of lead or 0.02 pCi of lead-210 from the surface to 40 KFT.

The influence of surface vegetation on the system is small. Although the lead-210/radium-226 ratio in vegetation is large, the activities expressed

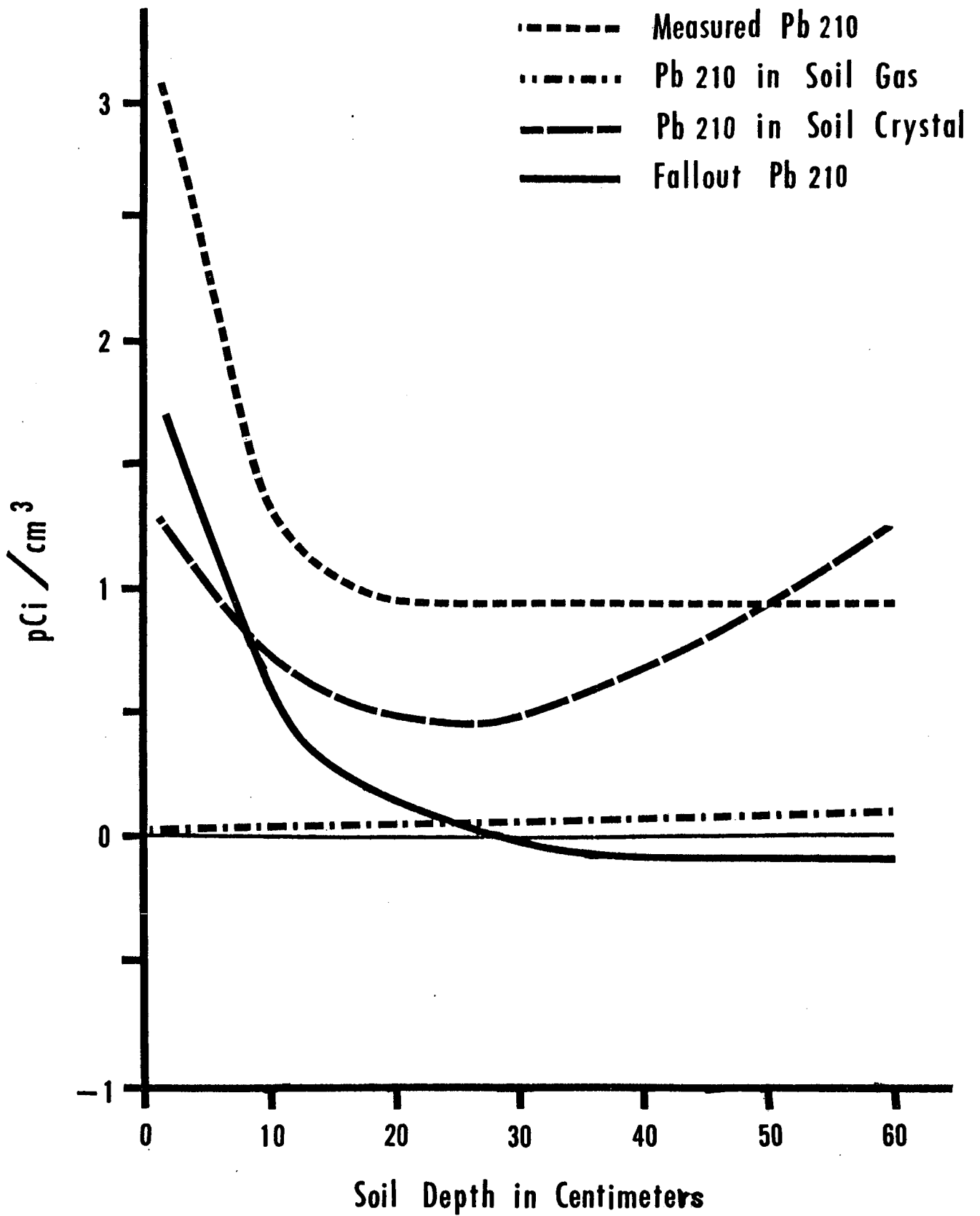


Fig. 3. Measured Lead-210 and Calculated Components as a Function of Depth.

per cm^3 are about a picocurie and less than a picocurie. From this we may conclude that in a lead-210, radium-226 activity balance only soil need be considered.

Conclusions

The lead-210 and radium-226 concentrations in a soil profile from 0 to 2 feet were measured. The concentrations measured vary as a function of depth. Lead-210 is concentrated in the upper soil horizons due mainly to natural lead fallout. The lead-210 profile was considered to be composed of three components: lead-210 formed directly from radium within the soil crystal, lead-210 formed from radon which diffused out of a soil crystal and migrated, and lead-210 fallout arising from radon which diffused from the soil and decayed in the atmosphere. By assuming a simple steady state radon model it was possible to calculate the first two lead-210 components from the radium data and therefore separate lead-210 fallout from the total lead-210 measured. This "natural fallout" penetrates to about 11 inches in this particular soil core. This penetration depth is in good agreement with published data on strontium-90 and other "weapons fallout" nuclide distribution in soil.

References

1. Memoranda - Fisenne to Welford (1967)
2. Radford, E. P., and Hunt, V. R. Science 146, 1943 (1964)
3. Tso, T. C., Hallden, N. A., and Alexander, L. T. Science 146, 1043 (1964)

4. Report of the United Nations Scientific Committee on the Effects of Atomic Radiation (New York, 1962).
5. Tso, T. C., Harley, N. A., and Alexander, L. T.
Science 153, 880 (1966)
6. Tso, T. C., Harley, N. A., and Alexander, L. T.
Tobacco Science 10, 105 (1966)
7. Petrow, H. G., and Cover, A.
Anal. Chem. 37, 1659 (1965)
8. Available from Pilot Chemical Company, Watertown, Massachusetts
9. HASL Manual of Standard Procedures
Report NYO-4700 (2d Issuance, 1967)
10. Alexander, L.T., Hardy, E.P., Myer, M.W., Allen, J.S., and Valassis, V.T.
Report HASL-171 (1966)
11. Walter, A.
J. Geophys. Res. 68, 1485 (1963)
12. Jacobi, W.
Biophysik 1, 175 (1963)
13. Patterson, R. L., Jr., and Lockhart, L. B., Jr.
Part I, Radioactivity in the Atmosphere, 383
The Natural Radiation Environment (Editors - Adams and Lowder)
University of Chicago Press (1964)

The Deposition of ^{226}Ra and ^{210}Pb in
the Human Aorta

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Abstract

The ^{226}Ra and ^{210}Pb contents of various soft tissues from two former radium dial painters have been measured to determine the distribution and concentration levels of these nuclides in the organs of exposed individuals. In both cases, the aorta contained significantly higher concentrations of radium and lead than were present in other tissues.

The significance of this finding is discussed with reference to normal values, concentration mechanism, radio-sensitivity of the aortic wall, and a preliminary estimation of α dose.

Introduction

For the past seven years, New York University has taken part in a study of the distribution and retention of ^{226}Ra in the tissues of former New Jersey radium dial painters. In two cases, a representative sample of the body soft tissue was available for measurement of ^{226}Ra and ^{210}Pb (1).

The relative specific activities of wet tissue have indicated that the aorta concentrates both of these nuclides, which has led to the beginning of an investigation to determine

the significance of the aortic dose in exposed individuals. Concentration levels of radium in the aortas of dial painters, as well as in normal individuals, are presented along with a discussion of the possible concentration mechanism, the radio-sensitivity of the aortic wall, and the α dosimetry in this tissue.

Methods

Radium-226 content was determined by radon emanation techniques (2) after preliminary wet ashing with nitric and perchloric acids. Lead-210 concentration was obtained by solvent extraction of a lead bromide complex ion into a quaternary ammonium bromide (Aliquat-336) and subsequent beta counting of the bismuth-210 daughter after a suitable ingrowth period (3).

Results

Specific activities for ^{226}Ra and ^{210}Pb in the aorta of Case #5281 with a 0.6 μCi terminal body burden were approximately 43 and 23 times greater than the average values obtained for 31 other soft tissues. In dial painter #5278 with 0.05 μCi body burden, the ^{226}Ra and ^{210}Pb contents were more than 17 and 112 times the respective average values obtained for various other tissues. A summary of the data for both cases is given in Table I. It has been estimated (1) for Case #5281, using standard man data, that approximately 0.05% of the whole body ^{226}Ra and 0.65% of the ^{210}Pb at death were present in soft tissue. To date, there are still too

Table I. Radium Dial Painter Soft Tissue Analysis -
Summary

<u>Case Number</u>	<u>^{226}Ra content pCi/g wet weight</u>	<u>^{210}Pb content pCi/g wet weight</u>
Case #5281 (Body burden \approx 0.6 uCi)		
aorta	0.43 \pm 0.02	1.01 \pm 0.05
Mean - Other tissues (31)	0.010 \pm 0.015	0.045 \pm 0.001
Case #5278 (Body burden \approx 0.05 uCi)		
aorta	0.089 \pm 0.008	1.01 \pm 0.05
Mean - Other tissues (6)	0.005 \pm 0.002	0.009 \pm 0.001

few tissues available from Case #5278 to make a similar comparison.

Discussion

a. Normal Values

Considering the relatively high concentrations of lead and the alkaline earth elements in the "normal" human aorta (4,5), it is reasonable to expect that levels of ^{226}Ra and ^{210}Pb in aortas of dial painters would be elevated above values in other soft tissues. Elkeles (6), for example, has measured higher α activities in calcified human aorta than in other soft tissues of the body. He attributes this difference to preferential deposition in aorta by members of the radium and thorium series. In addition, Lamberts and Van Andel (7) have demonstrated a greater in vivo affinity of the rat aorta for ^{137}Ba and ^{85}Sr than found for other soft tissues.

The deposition of calcium in the aorta is considered one of the fundamental processes in the biology of aging (8). Since radium and lead both act as chemical congeners of calcium, it follows that increased mineralization should result in higher levels of tissue radioactivity. Elkeles (6) has recently reported that the wet tissue gross α activities in calcified aortas are not only greater than found in non-sclerotic tissue but in some cases approach bone levels.

Radium-226 values for 4 "healthy" and 4 sclerotic aortas from unexposed persons have been measured and are

Table II

Radium-226 in Sclerotic and Non-Sclerotic Human Aorta Tissue

<u>Case</u>	<u>Sample Weight,g</u>	<u>Diagnosis</u>	<u>Ca in Dry Tissue g/g</u>	<u>Ra/Ca pCi/g</u>	<u>Ra in Dry Tissue,pCi/g</u>
1	4.52 (a)	Non-Sclerotic	0.040 ± 0.002 (b)	0.518	0.021 ± 0.004
2	9.50 (a)	Non-Sclerotic	0.040 ± 0.002	0.067	0.003 ± 0.001
3	27.24	Non-Sclerotic	0.037 ± 0.002	0.077	0.003 ± 0.001
4	6.99	Non-Sclerotic	0.043 ± 0.002	0.047	0.002 ± 0.002
5	31.68	Sclerotic	0.073 ± 0.004	0.276	0.020 ± 0.001
6	29.00	Sclerotic	0.058 ± 0.003	0.249	0.014 ± 0.001
7	21.81	Sclerotic	0.064 ± 0.003	0.234	0.015 ± 0.001
8	21.10	Sclerotic	0.066 ± 0.003	0.288	0.019 ± 0.001

(a) Cases 1 and 2 did not represent the entire aorta

(b) All errors reported = 1 S.D.

presented in Table II. From the data obtained, it appears that the specific activity of radium in aortic tissue may be related to the degree of mineralization that has occurred. Aside from Case #1, which did not represent more than 25% of the aorta, the range of values in nonsclerotic aortas was from 0.05-0.08 pCiRa/gCa. The corresponding range for this ratio in aortas diagnosed as sclerotic was from 0.23 to 0.29 pCiRa/gCa. Since only a small number of samples have been assayed thus far, this conclusion requires further statistical verification. Elkeles (9) has noted a similar relation in his measurements of the gross α activities in 107 aortas, assuming the mineral content is proportional to the percent ash.

"Normal" ^{226}Ra values in bone have been reported as ≈ 0.012 pCi/g ash (10), which gives a ratio of specific activity of aorta to that in bone of about 1.0 in unexposed individuals, whereas in dial painter #5281 it was found to be about 0.002.

This large difference in soft tissue to bone ratios may possibly be explained by the realization that for normal individuals the daily dietary intake of approximately 2 pCi of ^{226}Ra (10) is significant relative to a normal skeletal burden of 30-50 pCi. The soft tissue, therefore, is being continuously exposed to a level of ^{226}Ra which is a few percent of the skeletal burden. For the dial painter, however, where the biological half-life of skeletal-bound radium is

about 10-15 years (11), only 0.02 percent of the radium in the body is resorbed daily and much less radium is available to soft tissue relative to the amount in the skeleton (1,12).

b. Deposition Mechanism

The biological mechanism whereby alkaline earth elements are concentrated in the aorta is probably an ion-exchange uptake of divalent cations by acid mucopolysaccharides in the medial ground substance (13). Rothstein (14) was among the first to postulate that mucopolysaccharides behave as ion exchangers in vivo and Dunstone (15) has demonstrated that the bonding of the alkaline earth elements with compounds such as chondroitin sulfate is of an ion-exchange type with nonspecific electrostatic bonds rather than the more specific chelate type. Since chondroitin sulfates are present in the aorta in relatively high concentrations (8) they may well be the cation binding agents.

c. Radiobiological Implications

The in vitro sensitivity of the mucopolysaccharide matrix to X-rays has been demonstrated by Lamberts and DeBoer (16) and Brinkman and Lamberts (17) in experiments involving changes in membrane permeability and viscosity. In addition, Lindsay et al. (18) have irradiated the abdominal aorta in dogs and found atherosclerosis in the irradiated vessel which could not be distinguished from the "natural" process. All of these effects have been attributed to a depolymerization of the mucopolysaccharide matrix which has been shown to be

sensitive at exposure doses as low as 10-20 R (19).

Dosimetry

In order to estimate the cumulative α dose to the dial painters' aortas, the short-term radium kinetics and loci of deposition in this tissue must be studied. Since only the terminal concentrations are known and there exists little information about the kinetics of short-term radium metabolism in the aorta, levels that occurred during the exposure period are only speculative. Concentrations may have been of such magnitude during the ingestion period as to make subsequent levels negligible; or if there was a gradual translocation of calcium and radium from bone to aorta combined with exponential biological loss, concentration levels might have peaked at some time during the 40-year exposure period and terminal values may then represent an approximate average of cumulative exposure.

a. Short Term Radium Kinetics

To gain some insight into short-term soft tissue distribution of radium, an animal study was begun. The purpose of the initial pilot program was to produce a suitable experimental design and to gain familiarity with the methodology and techniques. Preliminary results using I.V.-injected ^{224}Ra and ^{223}Ra in rabbits produced the expected rapid transfer of radium from blood to bone with little permanent deposition in soft tissues within a 10-day period.

At this time, the aorta appeared to contain a greater specific activity than most other soft tissues with the exception of kidney. The possibility of subsequent translocation of radium from bone to aorta remains to be investigated in a longer-term study.

b. Deposition Site

Cross sections of the abdominal aorta from beagles given single injection doses of ^{226}Ra (20) were studied by α track autoradiography in order to determine the specific radium deposition site. The tissue was prepared in paraffin blocks, and sections approximately 5-6 μ thick were fixed on slides and dipped in emulsion. Three months later tissue sections from a dog given a dose level of 9.28 $\mu\text{Ci/kg}$ body weight were developed and stained with hematoxylin and eosin.

More than 98% of all the α tracks identified (about 40 per aortic cross section) were present in the medial layer of the aorta and in almost every case they appeared to originate extracellularly in the ground substance. Since the medial layer accounts for approximately 70% of the cross sectional area, this represents a concentration factor of about 40% above the expected value if uniform deposition is assumed. This is in good agreement with Bertelsen's data, which indicated that the medial layer is also the site of normal calcium deposition in humans (8).

In addition to deposition within a given portion of the aortic wall, other factors which may affect the concen-

tration of calcium and radium are the greater mineral involvement of the abdominal as compared to the thoracic aorta and the possibility of "hot spots" due to calcium plaque formation. Mayneord (21) noted, for example, that when the "calcarous nodules" present in the prostate of a radium chemist were dissected and measured separately from the prostate, the total α activity per gram of ash was approximately ten times higher.

If terminal concentration levels are taken to represent possible averages for the total burden period and the media is taken as a uniform deposition site, the integrated alpha dose from radium alone, assuming total energy deposition in this layer, is approximately 2.5 rads for Case #5281 and 0.5 rads for Case #5278. These dose estimates might conceivably be increased by significant factors if consideration is given to radium daughters decaying at their formation site, irradiation by activity in the blood during early exposure times, and any of the concentrating factors previously mentioned.

On the other hand the possibility exists that the majority of the radium activity measured was present in concentrated calcium plaque areas, thereby producing little energy deposition in soft aortic tissue. Since there is a substantial increase of calcium with age in the medial layer while definitive plaque formation usually occurs in old age, it is possible, nevertheless, that the final condition is unimportant in

relation to that at earlier exposure times.

Future Studies

If there is a translocation of calcium from bone to aorta in man, it appears to be an age-related phenomenon which may in some way be involved with lipid metabolism and ultimately with the morphological integrity of the aortic membranes. Cholesterol levels in blood and calcium levels in the aorta have both been shown to increase directly with age (8). It has been established that excessive circulating cholesterol in the rabbit results in plaque formation. In addition it has been shown that excessive doses of Vitamin D₂ produce a "generalized disorder characterized principally by resorption of bone and abnormal deposition of calcium salts in many extra-osseous tissues" (22). In view of this it would be informative to investigate the metabolism of radium in the soft tissues of animals exposed to these agents. In this way it may be possible to induce transfer of calcium and radium from bone to aorta, thereby approximating a test of the presumed mechanism of translocation in man.

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Literature Cited

1. Petrow, H.G., A Study of the Distribution of ^{226}Ra , ^{228}Ra , ^{210}Pb , ^{228}Th in Bone and Soft Tissue of Radium Dial Painters, Ph.D. Thesis, New York University, 1966.
2. Rushing, D.E., W.J. Garcia, and D.A. Clark, The Determination of Radium-226, AM/41-44 Public Health Service, 1965
3. Petrow, H.G., and A. Cover, Direct Radiochemical Determination of Lead-210 in Bone, Analytical Chemistry 37:13, 1659-60 (1965).
4. Tipton, J.H., and M.J. Cook, Trace Elements in Human Tissue, Part II, Adult Subjects from the U.S., Health Physics 9:2, 103-145 (1963).
5. Schroeder, H.A., and J.J. Balassa, Abnormal Trace Metals in Man, Lead, J. Chron. Dis. 14:4 (1961).
6. Elkeles, A., Radioactivity in Calcified Atherosclerosis, The British Journal of Radiology, Vol. XXXIV No. 405, 602-605 (1961).
7. Lamberts, H.B., and J.G. Van Andel, The Deposition of Ba and Sr in the Aortic Wall, Kininklijke Nederlandse Akademic Van Proceedings Series C Biological and Medical Sciences Vol. LXVIII #4, North Holland Publishing Co., Amsterdam (1965).
8. Bertelsen, S., The Role of Ground Substance, Collagen and Elastic Fibers in the Genesis of Atherosclerosis; in Atherosclerosis and its Origin, Sandler, S., and G.H. Bourne, Academic Press (1963).

9. Elkeles, A., Atherosclerosis and Radioactivity, Journal of the American Geriatrics Society 14:9, 895-901 (1966).
10. Hallden, N.A., I.M. Fisenne, J.H. Harley, Radium-226 in Human Diet and Bone, Science 140, 1327-9 (1963).
11. Cohen, N., A Summary of Body Burden Measurements and Elimination Rates in the New Jersey Radium Dial Painters, in N.Y.U. Annual Report USAEC Contract AT(30-1)3086 Eisenbud, M., and T.J. Kneip (1967).
12. Lucas, H.F., Jr., Correlation of the National Radioactivity of the Human Body to that of its Environment ANL 6297, 55-67 (1960).
13. Cohen, N., Discussion of Possible Mechanisms Involved in the Apparent Concentration of ^{226}Ra and ^{210}Pb in Aorta and Thyroid, in N.Y.U. Annual Report USAEC Contract AT(30-1)3086, Eisenbud, M., and H. Petrow (1966).
14. Rothstein, A.L., Ion Exchanges in Organic and Biochemistry, Interscience, New York, 213-232 (1957).
15. Dunstone, J.R., Ion Exchange Reactions Between Acid Mucopolysaccharides and Various Cations, J. Biochem 85, 336-351 (1962).
16. Lamberts, H.B., and W.G.R.M. DeBoer, X-ray Induced Atheromatous Lesions in the Arterial Wall of Hypercholesterolaemic Rabbits, Int. J. Rad. Biol. 6:4, 343-350 (1963).
17. Brinkman, R., and H.B. Lamberts, Radiopathology of Extracellular Substances, Current Topics in Radiation Research Vol. II, Ed. by Ebert and Howard, 281-302, North Holland Publ. Co. (1966).

18. Lindsay, S., H.J. Kohn, R.L. Darkin, and J. Jew, Circulation Research 10:51 (1962).
19. Brinkman, R., H.B. Lamberts, and J. Zuideveld, The Filtration of Water and of Red Cells Through Thin Connective Tissue Corium Membranes Under Low-Level X-Irradiation, Int. J. Rad. Biol. 3, 509 (1961).
20. Dougherty, T.F., Research in Radiobiology, COO-119-235 (Sept. 1966).
21. Mayneord, W.V., Some Problems in the Metabolism of Radioactive Materials in the Human Body, Clinical Radiology Vol. II, 2-13 (1960).
22. Hass, G.M., R.E. Trueheart, and A. Hemmens, Experimental Arteriosclerosis Due to Hypervitaminosis D, American Journal of Pathology 5:37, 521-539 (1960).

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CONTENT AND DISTRIBUTION OF STABLE STRONTIUM AND ^{226}Ra
 IN HUMAN SKELETONS FROM WISCONSIN DECEDENTS, 1957-1961

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ABSTRACT

A large collection of essentially complete human skeletons provided an opportunity for simultaneous measurement of stable strontium and ^{226}Ra in a group of 75 subjects. The skeletons were from Wisconsin residents who died during the period 1957 to 1961 and whose ages at death ranged from 24 to 95 years (avg \approx 70 yr). Strontium-90 was not measured because estimated levels on the order of 0.1 pCi/g calcium were too low to permit adequate comparisons, particularly for smaller bone categories.

Stable strontium and ^{226}Ra were determined in midsections of the combined tibiae and fibulae from each of the 75 skeletons. Their frequency distributions were found to be log-normal. Concentrations varied from 0.29 to 2.20 mg strontium/g calcium (log-mean = 0.51 mg strontium/g calcium) and from 13 to 112 fCi ^{226}Ra /g calcium (log-mean = 33 fCi ^{226}Ra /g calcium). There was no statistical correlation between strontium and ^{226}Ra levels.

To evaluate the relative distribution of strontium and ^{226}Ra within these skeletons, 19 component parts (excluding teeth) from three of the 75 were analyzed. Stable strontium and ^{226}Ra concentrations were found to be nearly uniform within any one skeleton. Although the levels of ^{226}Ra in long bone ends were only slightly higher and the concentrations of strontium slightly lower than in corresponding long bone shafts, the differences were statistically significant.

The relationship of these data to reported high levels of stable strontium and ^{226}Ra in certain Wisconsin municipal drinking water supplies is discussed and a comparison of these findings with those from similar studies is given. Descriptions of the analytical techniques employed in the study are included in the report.

INTRODUCTION

Numerous reports and summaries of human skeletal content of strontium, ^{90}Sr , and ^{226}Ra have appeared in the literature.¹⁻⁶ In this paper, data are presented for the content of stable strontium and ^{226}Ra in bones of 75 skeletons from Wisconsin.

The skeletons were obtained from Marquette University Medical School. As far as could be determined from available records, the subjects were Wisconsin residents who died during the period 1957-1961. Most of them had been mental patients who lived in various state institutions for a substantial fraction of their lives. The average age at death was ≈ 70 yr. (range: 24-90 yr). Long bones, either combined mid-thirds of left and right tibiae and fibulae or mid-humeri, were analyzed for strontium and ^{226}Ra . Three of the 75 skeletons were subdivided into 19 bone categories (see Tables 2 and 3) and each category analyzed separately.

An attempt to measure ^{90}Sr was abandoned because concentrations were found to be 0.1 pCi/g calcium or less, as Kulp's data⁷ would predict. Errors in measuring such low levels of activity, especially in smaller bone samples, were too large to permit meaningful comparisons to be made.

Municipal drinking water supplies of several eastern Wisconsin communities, obtained from deep (1000+ ft) wells, contain elevated levels of strontium and/or ^{226}Ra .⁸⁻¹⁰ A few of the 75 subjects did reside in institutions located in such municipalities. But, in each case, investigation of the water supplies in those institutions revealed that drinking water was derived from private, shallow wells and that, in addition, it was softened.^{11,12} Hence, unusually elevated concentrations of either strontium

or ^{226}Ra in these skeletons were not anticipated. Moreover, because it has been impossible to obtain complete histories of the subjects, it is difficult to relate their body burdens of strontium or ^{226}Ra to dietary intake.

SAMPLE PREPARATION

The skeletons, as supplied by Marquette University, had been well dissected. About half of them had been further cleaned by washing in petroleum ether. They were stored in canvas bags within steel drums for 2-4 years prior to time of analysis. For the three skeletons analyzed by parts, length of the mid-thirds of long bones was estimated visually. In the case of the remaining 72 skeletons from which only representative long bone shaft was taken, the mid-thirds were measured precisely.

Bone samples, sawed into convenient 3/4 to 1-1/2 chunks, were ashed for a minimum of 18 hr at 800°C in muffle furnaces lined with carborundum bricks. Afterwards, they were ground to a 50-mesh powder in a Wiley cutting mill* and homogenized by shaking in plastic containers. The bones from the three totally analyzed skeletons were ashed in Pyroceram* dishes, but all other samples were ashed in aluminum oxide trays. The procedure has been described earlier.¹³

Three control ash samples were prepared similarly in aluminum oxide trays. Two of these controls consisted of single totally ashed skeletons. The third was a homogenous composite of ash from three entire skeletons.

The samples were analyzed on a "blind" basis by being submitted, with controls, to an independent quality-evaluation section. Duplicate aliquots of each sample were dispensed in small plastic containers bearing different numbers. Control samples amounted in number to approximately

* Mention of commercial products does not constitute endorsement by the Public Health Service.

20% of the regular ash samples and were inserted randomly among them. These controls were useful both for checking analytical consistency and for evaluating the normal range of variation in sample results. Analytical determinations were reported directly to the quality-evaluation section, where duplicate values were matched. Repeat samples were assigned new numbers and analyzed "blindly" once more.

CHEMICAL ANALYSES

Calcium was determined by a standard oxalate-permanganate procedure.¹⁴

Stable strontium was measured by an ion-exchange flame-photometric method. In accordance with the technique described by Wade and Seim,¹⁵ 1-g ash samples were dissolved directly in a basic EDTA solution. Strontium was separated from calcium and minor impurities on a cation exchange column and measured flame-photometrically, by the method of Elfers et al.¹⁶ Chemical yields of strontium, monitored with ⁸⁵Sr for each sample, averaged between 90% and 95%.

A ²²²Rn emanation procedure¹⁷ was used to determine ²²⁶Ra. Ten-gram samples of ash were dissolved in 6 N HCl, filtered through acid-resistant membrane filters, evaporated to approximately 50 ml, and transferred quantitatively with 6 N HCl to 75-ml glass emanation ampules. Following a 30-day ingrowth period, the accumulated ²²²Rn daughter was transferred into an appropriate counting chamber, stored for 2 hr, and placed on a photomultiplier detector.

Interference from ²²⁸Ra was not considered to be serious and, for this reason, radon was not transferred through an intermediate charcoal trap. That is, ²²²Rn was not separated from any ²²⁰Rn daughters (from ²²⁸Ra) that might have been present. While there is evidence¹⁸ to suggest

that concentrations of ^{228}Ra in well waters from some Wisconsin communities may, in some cases, have been comparable to ^{226}Ra levels, several factors argue against significant ^{228}Ra interference in the analysis of the bone samples. In the first instance, there is indication¹⁹ that the normal ratio of ^{228}Ra : ^{226}Ra in human bone is approximately 1:4. Secondly, ^{228}Ra is counted by the emanation technique with only 1/3 the efficiency of ^{226}Ra . Thirdly, approximately one half-life of ^{228}Ra (6.7 yrs.) had elapsed between death of the subjects and the time analyses were performed. Thus, it is reasoned that the extent of ^{228}Ra interference would be no more than + 4%, which is insignificant compared to the normal uncertainties (20%-26%, at one S.D.) in the ^{226}Ra /calcium values.

ANALYTICAL RESULTS

Tabulations of the stable strontium and ^{226}Ra results, expressed with reference to calcium, are given in Tables 1-3. For the set of 75 skeletons, the strontium concentrations ranged from 0.29 to 2.20 mg strontium/g calcium (log-mean = 0.51 mg strontium/g calcium) and the ^{226}Ra concentrations, from 13 to 112 femto Curie (fCi) ^{226}Ra /g calcium (log-mean = 33 fCi ^{226}Ra /g calcium). It should be noted that concentrations of both substances varied over approximately one order of magnitude.

The concentrations of calcium in bones from the three skeletons analyzed by parts were constant to about 1-2%, that is, to within the precision of the analytical procedure. For this reason, the number of samples from the entire set of 75 skeletons analyzed (in duplicate) for calcium was limited to a total of 11. An average calcium value of 39.27%, thus determined, was used in calculations for the entire group of 75

TABLE 1
 Ratios of Stable Strontium and ^{226}Ra to Calcium
 in 75 Skeletons from Wisconsin

(Average of duplicate analyses of mid-thirds of combined left and right tibiae and fibulae, except where noted.)

<u>Skeleton Number</u>	<u>Sex</u>	<u>Age at Death</u>	<u>Year of Death</u>	<u>Sr/Ca (mg/g)</u>	<u>$^{226}\text{Ra/Ca}$ (fCi/g)</u>
1	F	34	1957	0.38	36
2	F	92	1958	0.37	23
3	M	67	1958	0.76	69
4	M	95	1959	0.48	33
5 ^{a,b}	F	68	1959	0.46	46
6 ^a	M	63	1959	0.55	28
7 ^c	M	76	1959	0.76	33 ^d
8	M	91	1959	0.35	15
9	M	70	1957	0.39	48
10	M	68	1957	0.36	13
11	M	79	1958	0.46	41
12	M	67	1959	0.38	46
13	M	61	1959	0.59	33
14 ^{a,e}	M	70	1959	0.56	28
15 ^c	M	89	1959	0.71	66
16 ^f	M	57	1959	0.45	51
17	M	71	1959	0.62	51
18	M	61	1959	1.90	20
19 ^f	F	84	1959	0.96 ^d	18 ^d
20	M	66	1959	0.41	31
21	g	g	g	0.64	31
22	g	g	g	0.35	13
23	M	66	1960	0.40	53
24 ^a	M	74	1960	0.32	89
25	M	68	1960	0.37	20
26	M	79	1960	0.44	23
27	M	85	1960	0.89	41

TABLE 1 (Cont'd.)

<u>Skeleton Number</u>	<u>Sex</u>	<u>Age at Death</u>	<u>Year of Death</u>	<u>Sr/Ca (mg/g)</u>	<u>²²⁶Ra/Ca (fCi/g)</u>
28	M	58	1960	0.42	23
29	F	66	1960	0.32	48
30	M	59	1960	0.48	15
31	M	49	1960	0.41	31
32	M	84	1960	0.52	20
33 ^a	F	83	1960	2.20	38
34	F	40	1960	0.71	20
35	F	24	1960	1.24	33
36	F	54	1960	0.46	76
37	M	79	1960	1.45	79
38	M	75	1960	0.55	33
39	M	48	1960	0.41	38
40 ^a	M	63	1960	1.16	102
41	M	75	1960	0.57	64
42	M	62	1961	0.68	46
43	M	94	1961	0.86	51
44	g	g	g	0.57	71
45 ^b	g	g	g	0.51	28
46	g	g	g	0.56	15
47	g	g	g	0.72	38
48 ^a	g	g	g	0.49	23
49	g	g	g	0.62	15
50	g	g	g	0.51	28
51 ^b	g	g	g	0.51	23
52 ^b	g	g	g	0.56	53 ^d
53	g	g	g	0.52	15
54	g	g	g	0.70	36
55	g	g	g	0.36	15

TABLE 1 (Cont'd.)

<u>Skeleton Number</u>	<u>Sex</u>	<u>Age at Death</u>	<u>Year of Death</u>	<u>Sr/Ca (mg/g)</u>	<u>²²⁶Ra/Ca (fCi/g)</u>
56	g	g	g	0.61	23
57 ^b	g	g	g	0.41	18
58	g	g	g	0.47	28
59	g	g	g	0.46	28
60	g	g	g	0.51	31
61 ^a	M	37	1957	0.29	61
62	M	87	1957	0.69	23
63	M	79	1957	0.41	76
64	M	62	1957	0.57	76
65	M	70	1958	0.37	13
66 ^c	F	75	1958	0.46	25
67 ^c	F	68(?)	1958	0.31	71
68	M	75	1958	0.30	18
69	M	85	1958	0.33	20
70	M	83	1959	0.34	31
71	M	83	1959	0.46	13
72 ^c	F	73	1959	0.61	51
73	M	58	1960	0.62	28
74 ^c	M	64	1960	0.41	28
75 ^b	g	g	g	0.48	112 ^d

FOOTNOTES:

^aCategory not complete, i.e., some portions of analyzed bone were missing.

^bVisual evidence of osteoporosis.

^cMid-third lengths of bone were estimated visually.

^dSingle analysis.

^eFlesh not dissected from bone.

^fFemurs not available, mid-thirds of both humeri substituted.

^gInformation unavailable.

Note: To calculate ratios of strontium and ²²⁶Ra to calcium, an average calcium content of 39.27% was used.

TABLE 2

Ratios of Stable Strontium to Calcium for Designated Bone Categories

of Three Wisconsin Skeletons

(Average of duplicate analyses, except where noted.)

Part No.	Sample Description	Sr/Ca (mg/g)		
		Skeleton No. 66	Skeleton No. 67	Skeleton No. 74
1	Hands and Feet	0.35	0.31	0.33
2	Patellae	-	-	-
3	Pelvis	0.26	0.32	0.27
4	Vertebrae	0.34	0.28	0.27
5	Ribs	0.39	0.29	0.27
6	Sternum	-	-	-
7	Clavicle	0.38	0.30	0.34*
8	Scapulae	0.31	0.28*	0.28
9	Calvarium & Rmdr. Skull & Mand.	0.37	0.31	0.35
10	Lt. Femur, Ends	0.33	0.29	0.34
11	Lt. Femur, Shafts	0.35	0.32	0.34
12	Rt. Femur, Ends	0.31	0.30	0.34
13	Rt. Femur, Shafts	0.38	0.29*	0.37
14	Lt. & Rt. Humeri, Ends	0.33	0.30*	0.32
15	Lt. & Rt. Humeri, Shafts	0.40	0.29*	0.34
16	Lt. & Rt. Ulnae & Radii, Ends	0.42*	0.31*	0.34*
17	Lt. & Rt. Ulnae & Radii, Shafts	0.41	0.30*	0.36
18	Lt. & Rt. Tibiae & Fibulae, Ends	0.37	0.30	0.36
19	Lt. & Rt. Tibiae & Fibulae, Shafts	0.47	0.31	0.42
20	Teeth	-	-	-

* Single Analysis

TABLE 3
 Ratios of ^{226}Ra to Calcium for Designated Bone Categories
 of Three Wisconsin Skeletons
 (Average of duplicate analyses, except where noted.)

Part No.	Sample Description	$^{226}\text{Ra}/\text{Ca}$ (fCi/g)		
		Skeleton No. 66	Skeleton No. 67	Skeleton No. 74
1	Hands and Feet	32	101*	36
2	Patellae	-	-	29*
3	Pelvis	29	74	28
4	Vertebrae	27	84	24
5	Ribs	24	-	26*
6	Sternum	-	-	36*
7	Clavicle	16*	66*	44*
8	Scapulae	23	-	31
9	Calvarium & Rmdr. Skull & Mand.	20	70	23
10	Lt. Femur, Ends	26	75	26
11	Lt. Femur, Shafts	23	59	28
12	Rt. Femur, Ends	23	65	34
13	Rt. Femur, Shafts	18	60*	28
14	Lt. & Rt. Humeri, Ends	28	85*	28
15	Lt. & Rt. Humeri, Shafts	26	58*	28
16	Lt. & Rt. Ulnae & Radii, Ends	29*	81*	-
17	Lt. & Rt. Ulnae & Radii, Shafts	26	55*	23
18	Lt. & Rt. Tibiae & Fibulae, Ends	33	80*	39
19	Lt. & Rt. Tibiae & Fibulae, Shafts	26	71*	29
20	Teeth	-	-	-

* Single Analysis

skeletons (Table 1). However, individual calcium values for each sample were applied for calculations of the three skeletons analyzed in detail (Tables 2 and 3).

Based on the amount of strontium or ^{226}Ra required to double the appropriate blank sample values (0.25 ppm equivalent intensity of strontium for distilled water and 20 fCi ^{226}Ra for 75 ml of 6 N HCl), the detection limits were, respectively, 0.02 mg strontium/g calcium and 5 fCi ^{226}Ra /g calcium.

The analytical determinations for the control ash samples are presented in Table 4. At the 95% confidence level (2σ), the uncertainties in the values of the controls are: for calcium, about 1.6%; for strontium/calcium, 12-14%; and for ^{226}Ra /calcium, 20-26%. Since the concentrations of strontium and ^{226}Ra in the control ash samples lie close to the averages for bones from the 75 skeletons, it is reasonable to assume that the ranges of uncertainty in the control measurements can be applied to most of the reported data, excepting the low values for which percentages of uncertainty would be greater.

DISCUSSION

The distribution of strontium and ^{226}Ra in the three skeletons (see Table 5 for personal histories) is generally uniform, that is, to within analytical uncertainties of $\pm 14\%$ and $\pm 20\%$ to $\pm 26\%$, respectively (at the 95% confidence level).

By analysis of variance, it was possible to demonstrate that for the three skeletons, the strontium/calcium ratios in long bone shafts are higher than in matching ends (F significant at the 5% level). The reverse is true for ^{226}Ra , viz, that long bone ends contain more ^{226}Ra

TABLE 4

Control Ash Analyses

(The number of single measurements is indicated in brackets.)

Control Designation	% Ca ($\pm 2 \sigma$)	Sr/Ca, mg/g ($\pm 2 \sigma$)	$^{226}\text{Ra}/\text{Ca}$, fCi/g ($\pm 2 \sigma$)
Skeleton "A"	38.83 \pm 0.62 [23]	0.643 \pm 0.072 [23]	39 \pm 10 [22]
Skeleton "B"	---	0.543 \pm 0.076 [6]	27 \pm 6 [6]
Composite of Three Skeletons	---	0.503 \pm 0.066 [17]	42 \pm 8 [17]

TABLE 5

Known Histories of the Three Subjects

Whose Skeletons were Analyzed by Parts

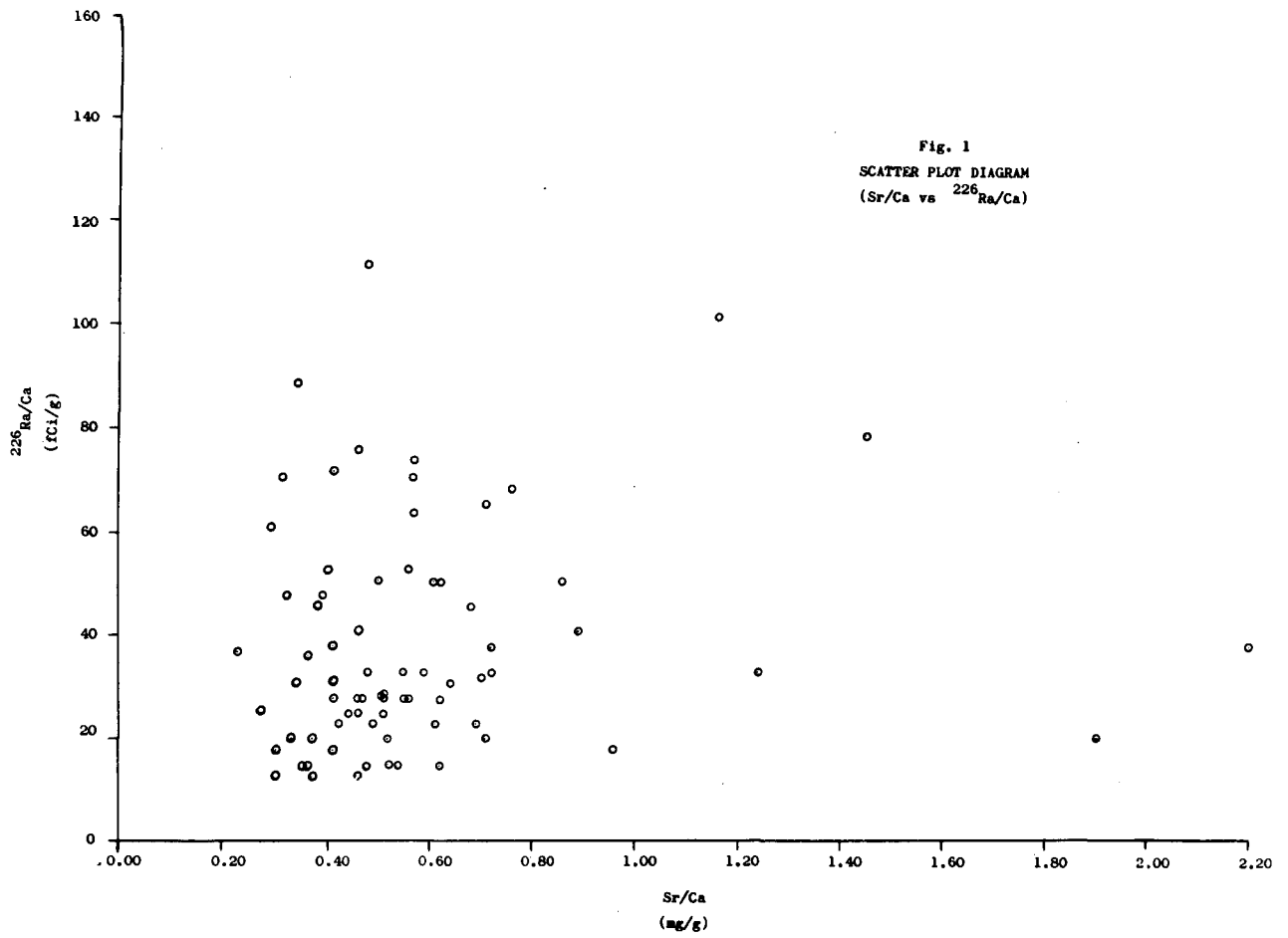
Subject No.	Sex	Year of Death	Age at Death	Remarks
66	F	1958	75	Inanition due to schizophrenia. Lived last 17-1/2 yrs. at Mendota State Hospital, Madison, Wisconsin.
67	F	1958	68(?)	Coronary occlusion. Lived last 31 yrs. at Douglas County Hospital and Sanitarium, Parkland, Wisconsin. Prior 9 yrs. lived at Mendota State Hospital, Madison, Wisconsin.
74	M	1960	64	Lobar pneumonia. Lived last 29 yrs. at Grant County Hospital, Lancaster, Wisconsin. Prior three yrs. at Northern Wisconsin Colony and Training School, Chippewa Falls, Wisconsin.

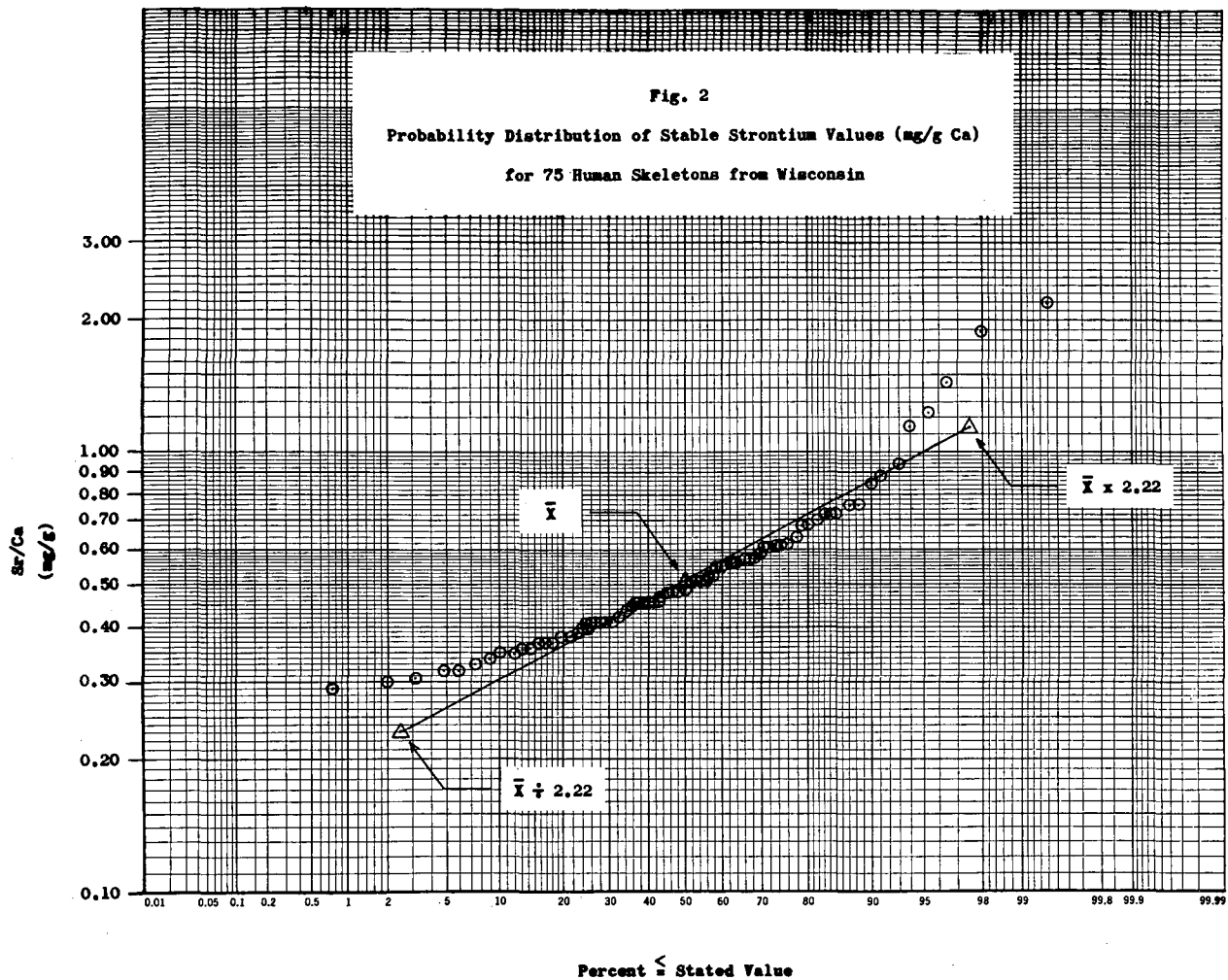
with respect to calcium than corresponding shafts (F significant at the 0.1% level). These differences are statistically consistent regardless of the bone type and, since turnover of bone mineral is more rapid in long bone ends than in shafts, are presumed to reflect dietary intake levels during the latter lifetime of those three subjects.

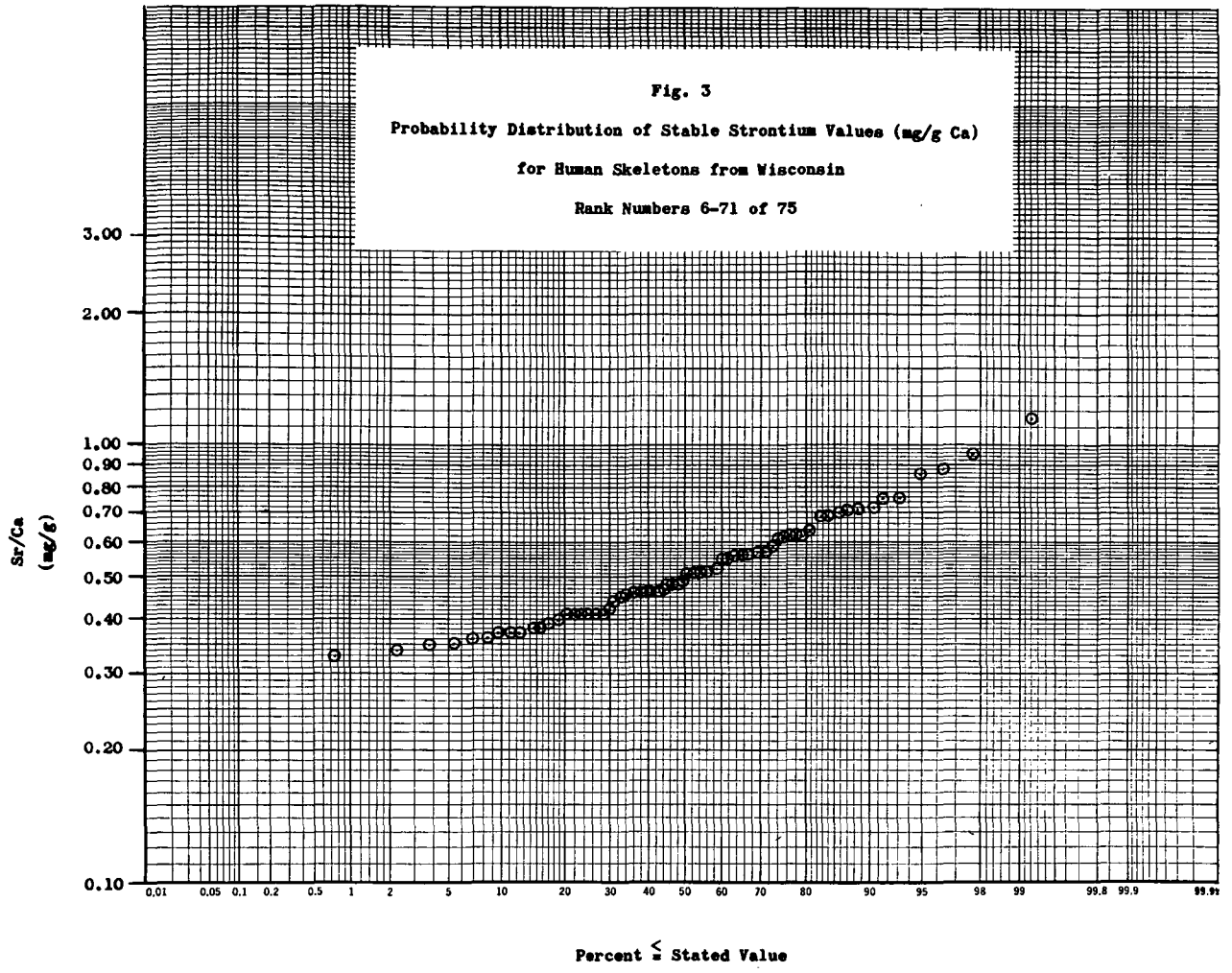
There is no correlation between strontium and ^{226}Ra concentrations in the skeletons, as the scatter-plot diagram in Figure 1 indicates. Minimum values lie sufficiently above the detection limits of 0.02 mg strontium/g calcium and 5 fCi ^{226}Ra /g calcium to be significant.

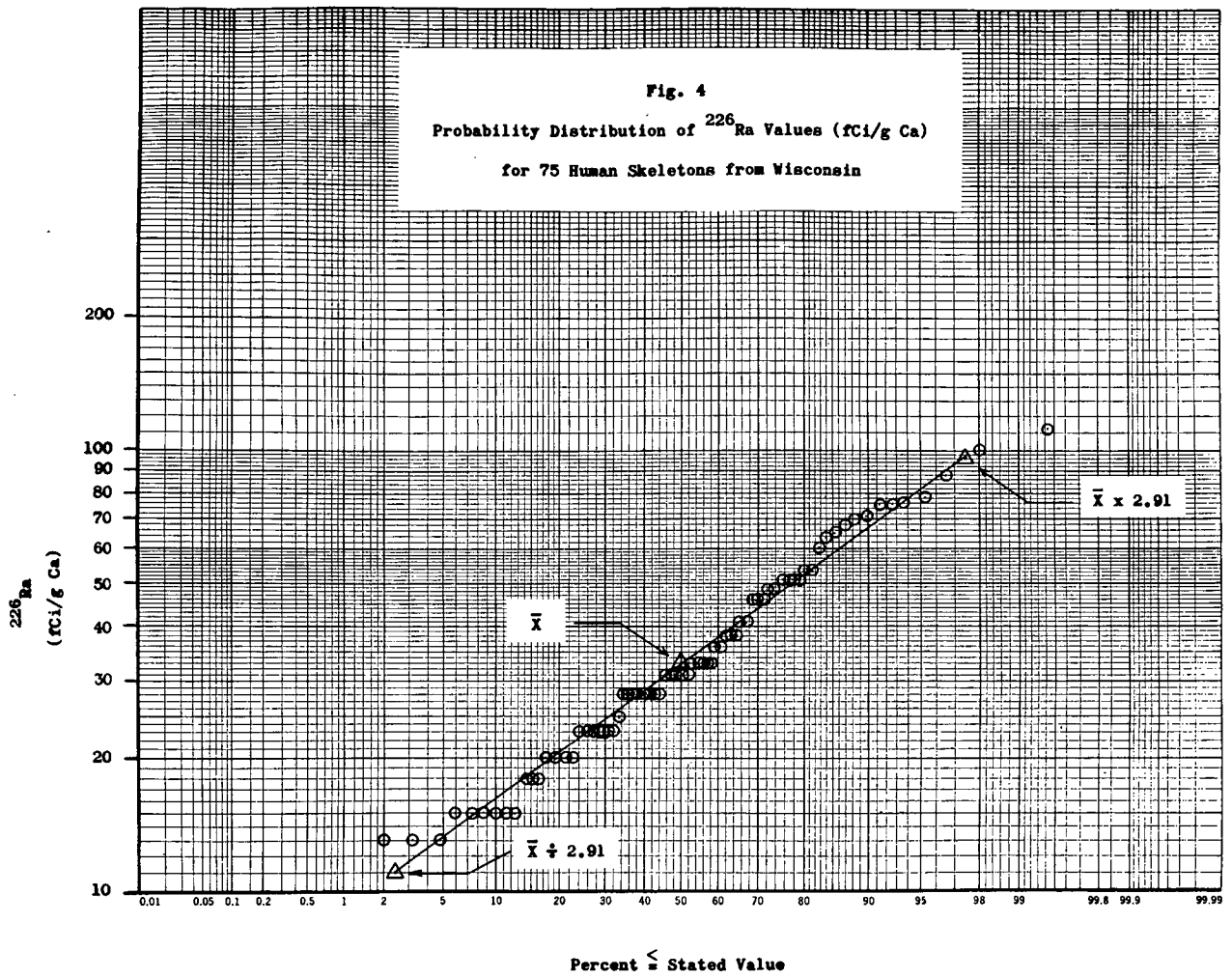
Distribution of both strontium and ^{226}Ra among the 75 skeletons approaches log-normal. (See Figs. 2-4). The highest four and possibly the lowest five strontium values do not conform with the distribution of the other points; hence, in Figure 3 only the mid-66 ranked data points are plotted. No explanation is offered to account for the highest strontium and radium values. Case histories for the subjects in question give no indication of how they might have ingested larger quantities of strontium or ^{226}Ra than any of the other subjects. In fact, as mentioned above, it is not feasible to account for body burdens of either substance in terms of case histories. Log-normal distributions of radionuclides in human populations are the rule, rather than the exception, as recent papers by Koch²⁰ and Schubert, et al.²¹ indicate.

Assuming the skeleton of a standard (70 kg) man contains 1.04 kg of calcium²² and that 95% of all the body Sr is located in the skeleton,²³ the body burden of strontium for the 75 skeletons ranged from 320 mg to 2400 mg (log-mean = 560 mg). Making the further assumption that 80% of the total body content of ^{226}Ra is in the skeleton,^{24,25} the ^{226}Ra body









burden for these subjects varied from 17 pCi to 146 pCi (log-mean = 43 pCi).

The concentrations of stable strontium and ^{226}Ra in the Wisconsin skeletons are of the same order as those reported by Kulp and Schulert² for human bones collected in the USA and from around the world. The ^{226}Ra content of the skeletons is also in reasonable agreement with values reported by Hursh^{26,27} (38-353 pCi and 47-130 pCi for 20 and 14 bodies, respectively, from Rochester, New York), Palmer and Queen²⁸ (13-139 pCi for 50 bodies from the Pacific Northwest), Walton et al.²⁹ (3-150 pCi for 140+ bodies from New York City), and the 130 pCi average body burden given more recently by Muth et al.³⁰ for skeletons from Germany. These data are summarized conveniently elsewhere.^{31,32}

Rivera³³ (HASL) determined strontium and ^{226}Ra in several bone categories of two adult human skeletons, also obtained at about the same time from Wisconsin. Using his data, it is possible to calculate that the strontium content of each of the two bodies he studied is 2460 mg and 620 mg, respectively. Assuming again that 80% of the body burden of ^{226}Ra is in the skeleton,^{24,25} these two bodies contained, respectively, 90 pCi and 60 pCi ^{226}Ra .

Thus, the data cited in this report are in substantial agreement with earlier findings.

CONCLUSIONS

Distribution of strontium and ^{226}Ra among long bones from the 75 Wisconsin skeletons approaches log-normal. Log-mean body burdens of these substances for strontium are 560 mg (range: 320 mg to 2400 mg) and for ^{226}Ra , 43 pCi (range: 17 pCi to 146 pCi). For the three skeletons analyzed by parts, strontium and ^{226}Ra were, within experimental errors, uniformly distributed among bone types. Statistical analysis of variance

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showed, however, that for these particular three skeletons, long bone ends contained more ^{226}Ra and less strontium than the shafts.

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Note:

Any number of the seventy-two remaining skeletons, minus sampled bones, are available to interested researchers.

REFERENCES

1. Kulp, J. L., and Schubert, A. R., NYO-9934, Vol. I (1962).
2. Ibid., Vol. II (1961).
3. Ibid., Vol. III (1961).
4. "Report of the United Nations Scientific Committee on the Effects of Atomic Radiation," General Assembly Official Records, 17th Session, Suppl. No. 16 (A/5216), 1962.
5. "Report of the United Nations Scientific Committee on the Effects of Atomic Radiation," General Assembly Official Records, 19th Session, Suppl. No. 14 (A/5814), 1964.
6. "Report of the United Nations Scientific Committee on the Effects of Atomic Radiation," General Assembly Official Records, 21st Session, Suppl. No. 14 (A/6314), 1966.
7. Kulp, J. L., J. Agr. Food Chem. 9, 122 (1961).
8. Nichols, M. S., and McNall, D. R., J. Am. Water Works Assoc. 49, 1493 (1957).
9. Scott, R. G., and Barber, F. B., Proc. Second United Nations Intern. Conf. on Peaceful Uses of Atomic Energy, Vol. 2, 153, Geneva (1958).
10. Lucas, H. F., Jr., private communication (through N. Petersen), 1965.
11. Lucas, H. F., Jr., ANL-6297, 55 (1961).
12. Adams, J. A. S., and Lowder, W. M., eds., The Natural Radiation Environment, 227-251, Univ. of Chicago Press, Chicago (1964).
13. Martin, A., Health Physics 13, 1348 (1967).
14. Hillebrand, W. F., et al., Applied Inorganic Analysis, 2nd ed., 627, Wiley, New York (1953).
15. Wade, M. A. and Seim, H. J., Anal. Chem. 33, 793 (1961).
16. Elfers, L. A., et al., Anal. Chem. 36, 540 (1964).

17. Krieger, H. L., et al., eds., Radionuclide Analysis of Environmental Samples. A Laboratory Manual of Methodology, Technical Report R-59-6, rev. Feb. 1966, p. 48 and p. 50, U. S. Public Health Service.
18. Krause, D. P., ANL-6199, 85 (1960).
19. Lucas, H. F., et al., Science 144, 1573 (1964).
20. Koch, A. L., J. Theoret. Biol. 12, 276 (1966).
21. Schubert, J., et al., Health Physics 13, 1187 (1967).
22. ICRP, Publ. 2, Report of Committee II on Permissible Dose for Internal Radiation, pp. 146-147 (1959).
23. Ibid., p. 176.
24. Hursh, J. B., and Lovaas, A., Nature 198, 265 (1963).
25. Lucas, H. F., Jr., ANL-6297, 55 (1961).
26. Hursh, J. B., and Gates, A. A., Nucleonics 7, 46 (1950).
27. Hursh, J. B., Brit. J. Radiol. Suppl. 7, 45 (1957).
28. Palmer, R. F., and Queen, F. B., Amer. J. Roentgenol. Ra Ther. and Nucl. Med. 79, 521 (1958).
29. Walton, A., et al., Health Physics 1, 409 (1959).
30. Muth, H., et al., Health Physics 2, 239 (1960).
31. "Report of the United Nations Scientific Committee on the Effects of Atomic Radiation," General Assembly Official Records, 17th Session, Suppl. No. 16 (A/5216), 1962, p. 222.
32. "Report of the United Nations Scientific Committee on the Effects of Atomic Radiation," General Assembly Official Records, 21st Session, Suppl. No. 14 (A/6314), 1966, p. 33.
33. Rivera, J., HASL-149, 134 (1964). See also HASL-144, 278 (1964).

THE ESTABLISHMENT AND EFFECTS OF A ^{226}Ra BODY BURDEN

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The subject, a 45-year-old married man with three living children and with a known exposure to ^{226}Ra and ^{210}Po , has been studied for the past 15 years to determine the extent of any resulting body burden and its possible effects.

The existence of a ^{226}Ra body burden was established by whole-body counting and expired-breath radon measurements at several different facilities. Possible effects have been studied by means of medical examinations, skeletal x rays, clinical blood tests, and analysis of short-term lymphocyte cultures in peripheral blood for assessment of chromosomal damage.

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Introduction

Use of radium, in the United States, started at the beginning of the Twentieth Century when it was first employed in cancer therapy. Since then usage has greatly diminished, although gynecological treatments still account for its greatest use in this country.⁽¹⁾ From about 1925, reports based on the experiences of American radium dial painters led to the recognition of the serious consequences that could follow accidental ingestion of such materials and consequently the effects of radium ingestion have been intensively studied with respect to both occupational⁽²⁻⁸⁾ and environmental⁽⁹⁻¹³⁾ exposure.

In this study, a 45-year-old married man with three living children and a known occupational exposure to ^{226}Ra and ^{210}Po has been under investigation for the past fifteen years to determine the extent of his body burden and any observable effects.

Radiation History

The subject received his ^{226}Ra body burden between June 1951 and June 1952 while working in a radium recovery plant, by inhalation and ingestion. He was also directly exposed to ^{210}Po during that time. He has since worked in analytical radiochemistry but has not been involved in any radium or polonium recovery work. The subject's occupational radiation exposure history is given in Table 1.

TABLE 1

<u>Occupational Radiation Exposure</u>	
<u>Year</u>	<u>Dose</u>
Before 1951	Not measureable
1951-52	Known exposure to ^{226}Ra and ^{210}Po Dose Unknown
1952-55	Not measurable
1955-60	1200 millirem
1960-Present	Not measureable

His x-ray exposure history is normal and diagnostic in nature.

Establishment of Body Burden

The presence of a ^{226}Ra body burden was established by a series of expired breath radon measurements (Table 2) and whole body counts (Table 3) between 1951 and 1967.

The maximum permissible ^{226}Ra skeletal body burden for occupational workers is $0.1 \mu\text{Ci}$ ⁽¹⁴⁾. The recommended RPG for skeletal concentration of ^{226}Ra

in the general population is 0.003 μCi for individuals and 0.001 μCi for an average population group⁽¹⁵⁾. A world-wide arithmetic average for the radium body burden of man has been calculated to be approximately 0.00003 μCi ⁽¹⁰⁾.

TABLE 2

Expired Breath Radon Measurements

<u>Date</u>	<u>Institution</u>	<u>Results (pCi/l)</u>
12/51	Fordham University	3.11
6/52	New York Operations Office	0.60
	Atomic Energy Commission	0.50
2/53	New York Operations Office	1.40
	Atomic Energy Commission	0.60
		0.40
		0.60
		0.50
		0.40
		0.40
5/56	New York Operations Office	0.23
	Atomic Energy Commission	0.27
7/59	Massachusetts Institute of Technology	0.14
8/63	University of Rochester	0.10
7/67	Massachusetts Institute of Technology	0.16
8/67	New York Operations Office	0.20
	Atomic Energy Commission	0.12

TABLE 3

Whole Body Counts

<u>Date</u>	<u>Facility</u>	<u>Burden ($\mu\text{Ci}^{226}\text{Ra}$)</u>
7/59	Massachusetts Institute of Technology	0.014
10/60	New York University Medical Center	0.009
1/63	New York University Medical Center	0.009
7/63	University of Rochester School of Medicine	0.010
7/67	Massachusetts Institute of Technology	0.016
9/67	New York University Medical Center	0.010

Thus, 0.1 μCi of ^{226}Ra and its daughter products in the body correspond to an average absorbed dose of 0.06 rad/week or an average dose to the bone of 0.56 rem/week equal to 29 rem/year⁽¹⁶⁾. Table 4 indicates the probable average dose to the bone received by the subject since his exposure.

TABLE 4

Average Dose to the Bone			
<u>Year</u>	<u>Body Burden ($\mu\text{Ci}^{226}\text{Ra}$)</u>	<u>Percent of MPC</u>	<u>Average Bone Dose (rem/year)</u>
1951-52	0.31	310	90
1952-56	0.04-0.14	40-140	12-41
1956-67	0.01-0.03	10-30	3-9

Thus the cumulative dose to the bone since 1951 ranges from 171-353 rem.

Determination of Possible Somatic Effects

Potential somatic effects due to the existing body burden have been studied by means of medical examinations, skeletal x-rays, clinical blood tests and chromosome analysis of cultured peripheral lymphocytes.

Medical Examinations

Complete physical examinations including urinalyses were made in July 1959 and again in July 1967 by the Massachusetts Institute of Technology Medical Department. The findings were negative with no effects due to radiation observed.

Skeletal X-ray

Skeletal X-ray examinations were made in July 1959 and again in July 1967. No bone damage or changes in bone structure were detected.

Clinical Blood Tests

Hematological tests were made monthly during the subject's exposure period, and later in 1959 and again in 1967. Table 5 lists the results of these tests. Laboratory findings were within normal limits and the variations observed in the total leukocyte counts are not considered to be related to radium body burden.

Blood chemistry and electrophoresis pattern tests were made in 1959 and again in 1967 and are presented in Table 6.

TABLE 5Hematological Results

<u>Period</u>	<u>Date</u>	<u>Total Leukocytes</u>	<u>Differential Neutrophils</u>	<u>Differential Lymphocytes</u>	<u>Differential Monocytes</u>
Normal Range		5-10,000/mm ³	50-70%	25-40%	1-4
Pre-exposure	6/7/51	11,400	57	39	4
	7/9/51	10,800	65	33	2
	8/9/51	8,100	61	35	2
	9/5/51	11,600	68	31	1
	10/31/51	11,700	62	33	3
During Exposure	12/7/51	6,250	58	39	3
	1/7/51	7,800	57	36	6
	2/6/52	9,700	68	32	-
	3/7/52	10,700	68	32	-
	4/8/52	6,500	57	41	2
	5/6/52	10,650	71	27	2
Post-Exposure	7/28/59	9,200	59	39	1
	7/24/67	7,600	63	32	1

TABLE 6Blood Chemistry and Electrophoresis Pattern Results

<u>Test</u>	<u>7/28/59</u>	<u>8/24/67</u>	<u>Normal Range</u>
<u>Blood Chemistry</u>			
Alkaline Phosphatase (units)	5.3	2.2	2-9
Blood Urine Nitrogen (mg/%)	17.5	22.8	10-18
Blood Sugar (mg%)	96	78	
Hinton	Negative	Negative	
<u>Electrophoresis Pattern</u>			
Total Protein (gram %)	7.6	7.75	6.0-8.0
Albumin (gram %)	4.88	3.07	4.0-5.0
Total Globulin(gram %)	2.72	4.68	2.0-3.0
Alpha 1 (gram%)	0.32	0.53	0.3-0.5
Alpha 2 (gram %)	0.60	0.84	0.5-0.7
Beta (gram %)	0.83	1.4	0.8-1.0
Gamma (gram %)	0.97	1.91	0.8-1.2

The only unusual finding is a reversal of the usual values of the albumin and globulin in the most recent tests. Electrophoresis tests will be made again to confirm these findings although they are not necessarily indicative of radiation damage.

Hematocrit values and sedimentation rates were normal.

Chromosome Analyses

Radiation produces breaks in chromosomes. The resulting aberrations may be restored with no apparent morphological change in the chromosomes, or the broken ends of adjacent chromosomes may join to produce chromosomes recognizably different, in size and morphology, from normal chromosomes.

The in vivo production of chromosome aberrations in peripheral leukocytes has been demonstrated in several population groups exposed to ionizing radiation. This includes diagnostic and therapeutic x-ray^(17,18) and therapeutic irradiation⁽¹⁹⁾, chronic occupational exposure^(20,6), radiation accidents^(21,22), thorotrast applications⁽²³⁾, fallout⁽²⁴⁾, and survivors of the bombing of Hiroshima and Nagasaki^(25,26).

Peripheral blood samples were processed for chromosomes from buffy coat cells⁽²⁷⁾ in 1962 and 1963. An improved micromethod⁽²⁸⁾ was used from 1964 to 1967. All chromosome preparations were stained with natural aceto-orcein and were photographed in phase 3 contrast (100/1.30 neofluar) on 35mm plus-X film using a Carl Zeiss photomicroscope. Projection prints were enlarged five times the primary film image of 400 diameters.

Control cultures from six healthy people were assessed and found to be normal.

The subject's chromosome counts showed an aneuploid spread which depressed the percentage of 46 chromosome cells below the normal controls by 11 percent (Table 7).

Typical chromosomal aberrations induced by ionizing radiation are shown in Fig. 1⁽²⁹⁾. Some of the subject's chromosomal aberrations are shown in Figure 2, 3, and 4.

Two abnormal chromosomes, one of which appears to be a reciprocal translocation involving one of the A and F homologs, and the other a mediocentric chromosome thought to be an E with a partial deletion of the long arms, are shown in Fig. 2. As in many of the subject's cells, two of the G chromosomes are in close association. The commonest cell with 46 chromosomes has a Y chromosome missing, and an extra chromosome which morphologically could be put into the C6-12 group is shown in Fig. 3. Figure 4a shows a 46-chromosome cell with a long chromosome and two secondary constrictions; this type is common after in vitro X-irradiation of leukocytes⁽³⁰⁾. Chromatid associations, Fig. 4b, 4c, and 4d, suggest stickiness. Fig. 4e shows the reciprocal translocation seen previously. A triradius type configuration (Fig. 4f) is the result of breakage and translocation.

The atypia of the subject's cell line is chiefly pseudodiploid with an extra chromosome often similar to the C group chromosomes. Some cells were also found with a missing Y chromosome.

TABLE 7
Chromosome Analysis

Patient		Subject							Control			
		Chromosome Frequency Distribution							Control		Cultures	
Culture No.	Date	Cells Counted	<45	45	46	47	47>	%(46)	No.	Sex	Age	%(46)
1	5/18/62	16	0	1	13	2	0	81.2	1	F	22	96.4
2	7/14/62	14	3	0	7	2	2	50.0	2	M	23	95.7
3	5/13/63	45	2	3	37	3	0	82.2	3	M	47	91.1
4	8/30/63	86(37)*	7	2	73	3	1	84.8	4	M	46	91.1
5	2/2/65	30(12)	4	2	17	6	1	56.6	5	M	46	91.7
6	4/7/65	84(29)	3	13	64	1	3	76.2	6	M	28	92.3
7	1/4/66	15(2)	2	1	12	0	0	80.0				
8	1/4/66	55(12)	3	2	44	6	0	80.0				$\bar{x} = 93.0$
9	4/1/66	94(55)	3	3	87	1	0	92.5				
10	10/7/66	56(19)	3	1	52	0	0	92.8				
Total:		495(186)	30	28	406	24	7	$\bar{x} = 82.0$				

* Number in parenthesis refers to number of cells karyotyped

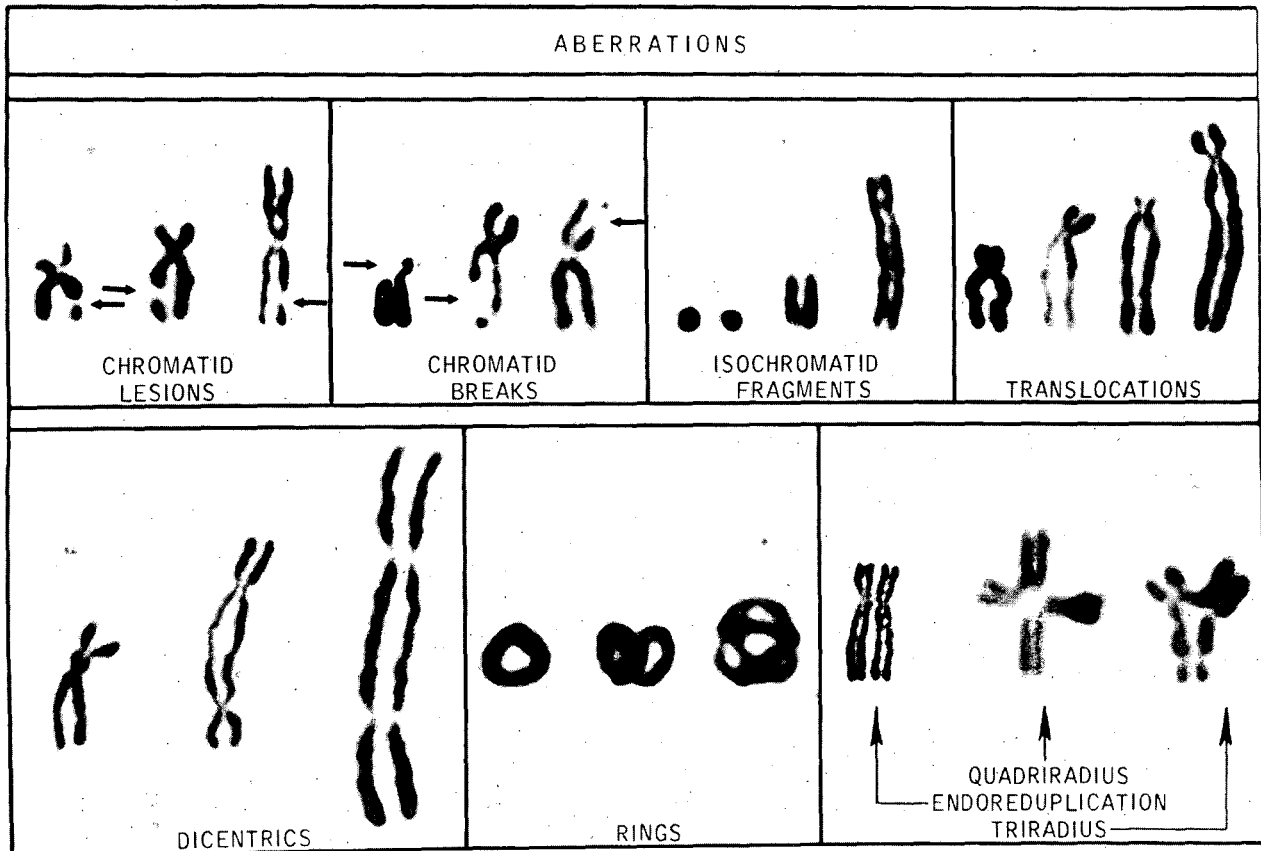


Fig. 1. Typical aberrations induced by ionizing radiation. [From Amarose et al., *Exptl. Molec. Pathol.* 7, 58-91 (1967).]

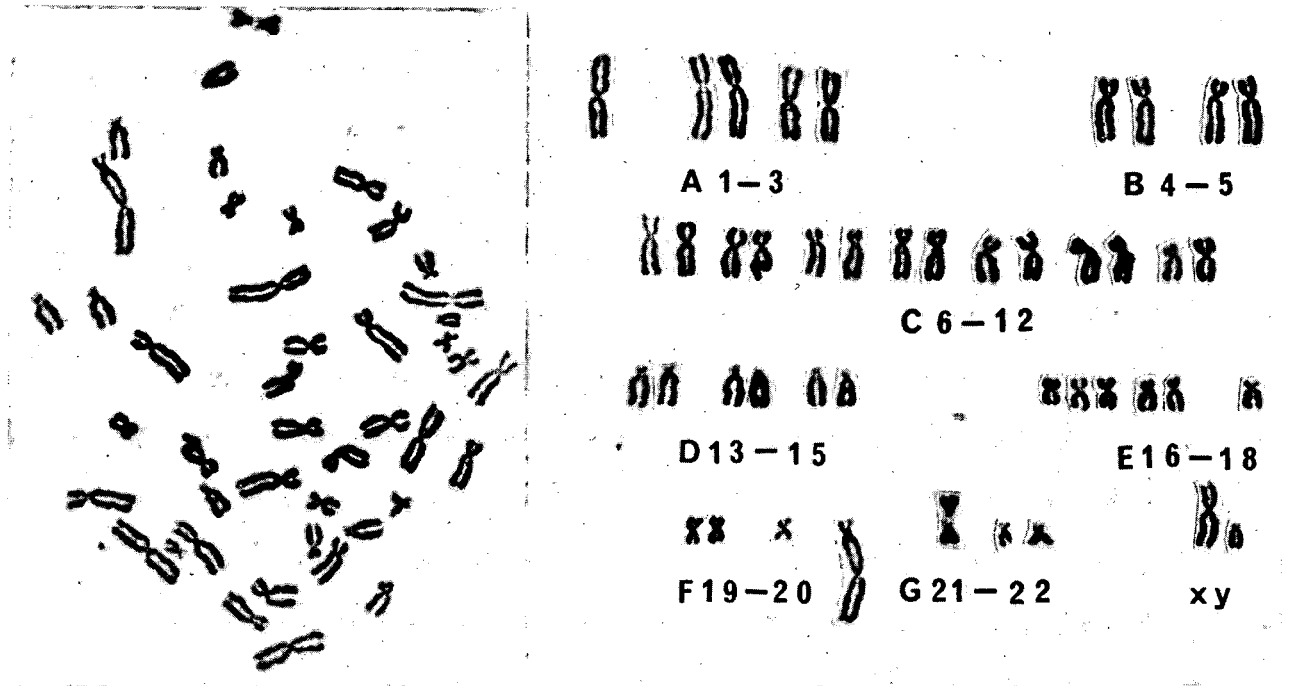


Fig. 2. Abnormal karyotype, showing reciprocal translocation.

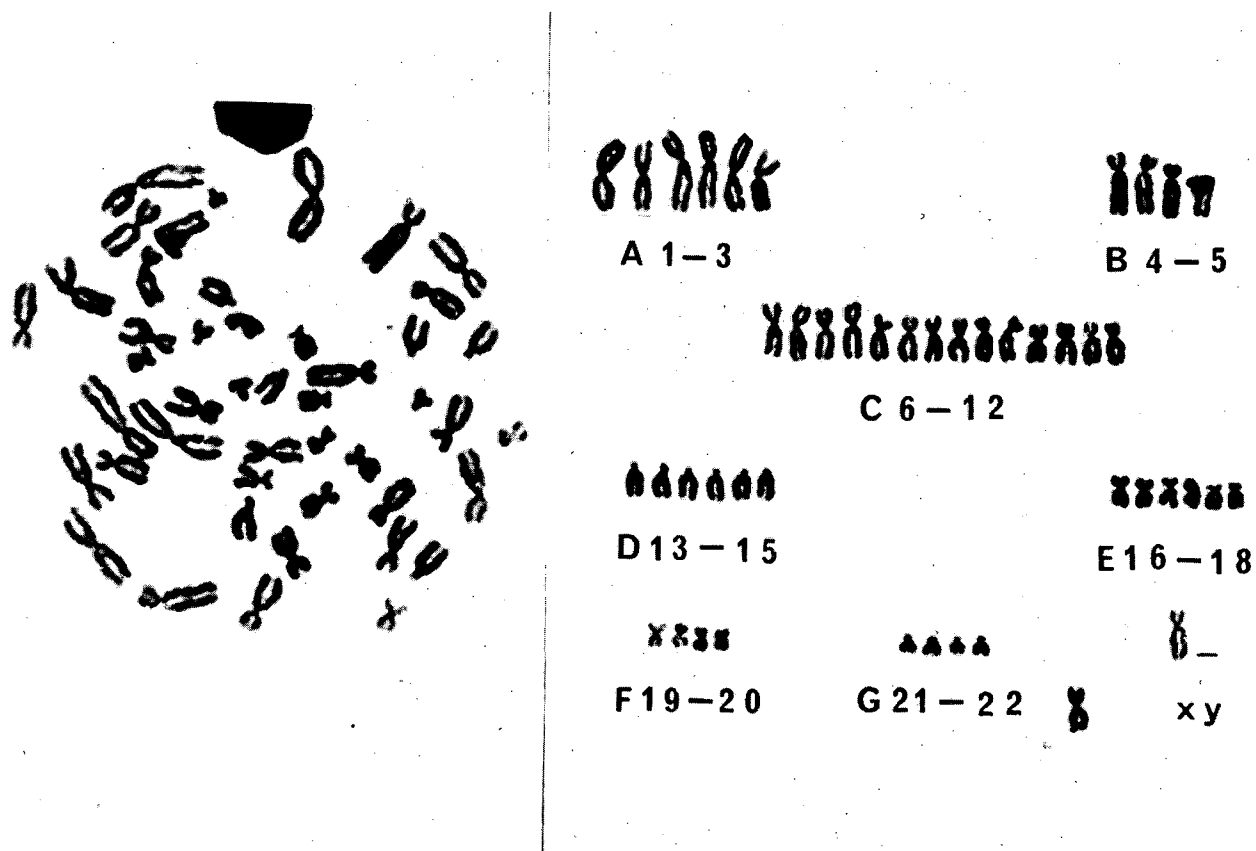


Fig. 3. Abnormal karyotype, showing an extra chromosome and a missing Y.

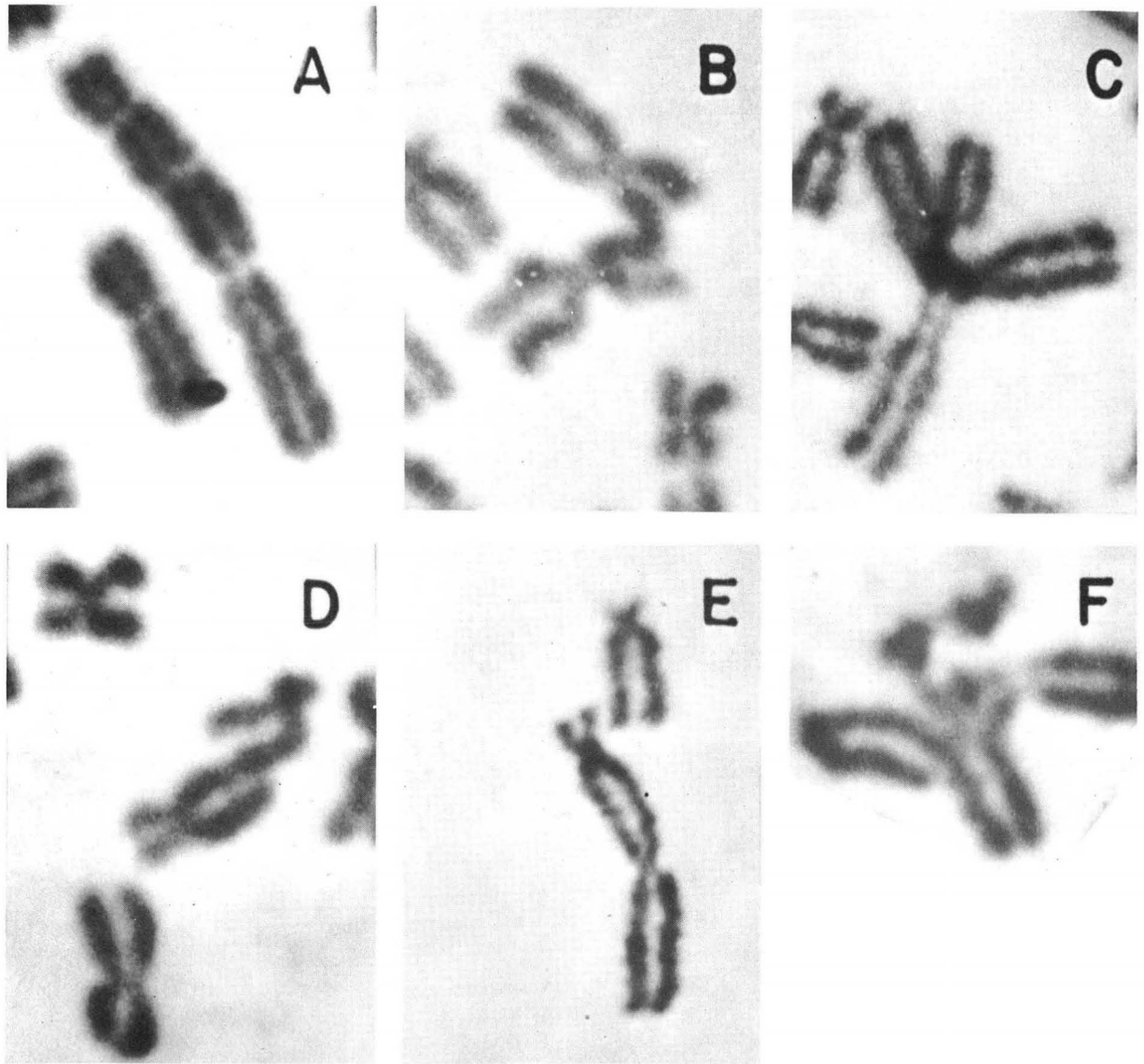


Fig. 4. A composite of abnormal chromosomes.

The abnormal chromosomes found were mainly acentrics and translocations. Such types are consistently seen after exposure to ionizing radiation. A few endoreduplicated cells were also observed. The frequency of such cells increases with dose⁽³¹⁾ and are rarely found in routine diagnostic cytogenetics. Dicentrics and rings were not found.

The percentage of abnormal chromosomes found in the karyotyped cells is shown in Table 8.

TABLE 8

Total Karyotypes with Abnormal Chromosomes

<u>Culture No.</u>	<u>Total Cells Karyotyped</u>	<u>★ Cells with Abnormal Chromosomes</u>	<u>Percent Abnormal</u>
4 (134)	37	7	18.9
5 (152)	12	2	16.7
6 (154)	29	3	10.3
9 (166)	55	3	5.5

() = months after exposure.

The percentage of abnormal chromosomes decreases with time (in months) after exposure. Analysis of culture No. 10 (Table 7) revealed no gross morphological abnormalities of the chromosomes. In January 1967, 200 months after exposure, an additional culture was done and found to be of normal pattern. Thus, the relationship of percent abnormality to elapsed time is shown in Figure 5, and appears to vary inversely with time.

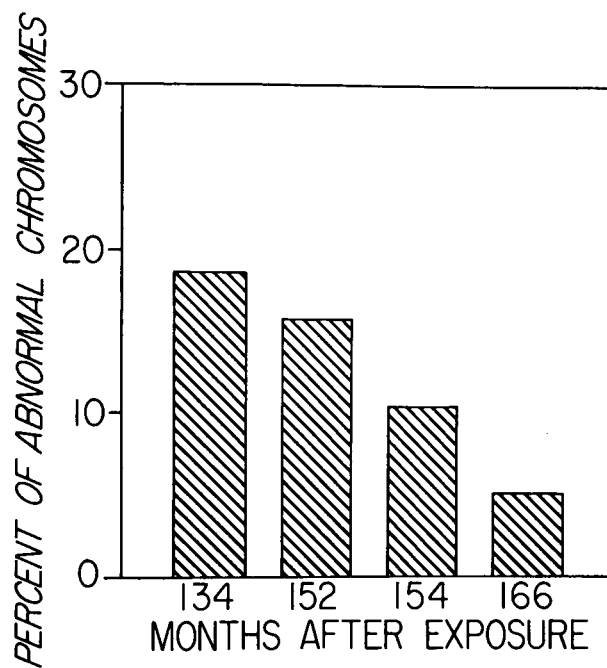
Abnormalities of the type seen here are caused by exposure to large doses of radiation. These progressively decrease with time after exposure, but are known to be detectable in significant number 10 or more years after exposure⁽⁶⁾.

Determination of Possible Genetic Effects

The subject's children were conceived after his exposure to radium. The first boy was born in 1954 and died at two months with congenital malformation of the heart (complete transposition of the great vessels with an interventricular septum defect). This anomaly occurs with sufficient frequency so that it cannot be considered a positive radiation-induced genetic effect.

The second boy was born in 1956 with an undescended testicle on the right side. The third boy was born in 1957 with an innocent heart murmur and a herniated hydrocele. A normal girl was born in 1959.

Examination of peripheral leukocyte cultures for the two male children indicated no numerical or structural abnormalities.



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Fig. 5. The relationship of percent abnormality with time.

Summary

The subject, presently with a 10-30 percent ^{226}Ra body burden, is in apparent good health. Medical examination and skeletal x-rays show no changes or conditions attributable to his exposure. Clinical laboratory findings were largely normal.

Chromosome changes in the form of numerical as well as structural aberrations were noted. There was an increase beyond control values in the frequency of cells not having the normal chromosome number of 46, while structural aberrations consisted of chromosome fragments, deletions, and translocations.

References

1. Quimby, E. H., Am. J. Roentgenol., Radium Therapy and Nucl. Med. 75, 443 (1956).
2. Silberstein, H. E., AECD-2122, "Radium Poisoning", May 26, 1945.
3. Baker, W. H., Bulkley, J. B., Dudley, R.A., Evans, R.D., McCluskey, H.B., Reeves, J. D., Jr., Ryder, R. H., Salter, L. P., and Shanahan, M. M., New England J. Med. 265, 1023 (1961).
4. Muth, H., and Oberhausen, E., "Assessment of Radioactivity in Man", Vol. II, 211 (1965), IAEA, Vienna.
5. Maletskos, C. J., Braun, A. G., Shanahan, M. M., and Evans, R. D., "Assessment of Radioactivity in Man", Vol. II, 225 (1965), IAEA, Vienna.
6. Boyd, J. T., Court-Brown, W. M., Vennart, J., and Woodcock, G. E., Brit. Med. J. 1, 377 (1966).
7. Muller, J., Klener, V., Tuscany, R., Thomas, J., Brezikova, D., and Houskova, M., Health Phys. 12, 993 (1966).
8. Evans, R. D., Health Phys. 13, 267 (1967).
9. Hursh, J. B., Brit. J. Radiol. Suppl. No. 7, 45 (1957).
10. Walton, A., Kologrivov, R., and Kulp, J. L., Health Phys. 1, 409 (1959).
11. Muth, H., Rajewski, B., Hanthe, H.-J., and Aurand, K., Health Phys. 2, 239 (1960).
12. Turner, R. C., Brit. J. Cancer 16, 200 (1962).
13. Hallden, N. A., Fisenne, I. M., and Harley, J. H., Science 140, 1327 (1963).

14. Handbook 69, Maximum Permissible Body Burdens and Maximum Permissible Concentrations of Radionuclides in Air and in Water for Occupational Exposure, National Bureau of Standards, August 1963.
15. Report No. 2, Background Material for the Development of Radiation Protection Standards. Federal Radiation Council, September 1961.
16. Report of ICRP Committee II on Permissible Dose for Internal Radiation, Health Physics 3, 205 (1960)
17. Tough, I. M., Buckton, K. E., Baikie, A. G., and Court-Brown, W. M., Lancet 2, 849 (1960).
18. Bloom, A. D., and Tjio, J. H., New Engl. J. Med. 270, 1341 (1964).
19. Millard, R. E., Cytogenetics 4, 277 (1965).
20. Norman, A., Sasaki, M., Ottoman, R. E., and Veomett, R. C., Radiation Res. 23, 282 (1964).
21. Bender, M. A., and Gooch, P. C., Radiation Res. 16, 44 (1962).
22. Bender, M.A., and Gooch, P. C., Radiation Res. 18, 389(1963).
23. Fischer, P., Golob, E., Kunze-Muhl, E., Ben Haim, A., Dudley, R. A., Mullner, T., Parr, R. M., and Vetter, H., Radiation Res. 29, 505 (1966).
24. Lisco, H., and Conard, R. A., Science 157, 445 (1967).
25. Doida, Y., Sugahara, T., and Horikawe, M., Radiation Res. 26, 69 (1965).
26. Bloom, A. D., Neriishi, S., Kamadu, N., Iseki, T., and Keeha, R. J., Lancet 2, 672 (1966).
27. Brown, C. D., Gabay, J. J., and Sax, N. I., Zeiss Mitteil. 3, 128 (1963).
28. Brown, C. D., Fleming, L. A., Mammalian Chromosomes No. 15, 110 (1965).
29. Amerose, A., Plotz, E., and Stein, A. A., Exp. Molec. Pathol. 7, 58 (1967).
30. Brown, C. D., unpublished data.
31. Kelly, S., and Brown, C. D., Amer. J. Public Health 55, 1419 (1965).

IN VIVO SCANNING FOR ^{131}I IN THE RAT THYROID

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In determining if low-level amounts of radionuclides could be detected and precisely located in a biological monitor as small as the rat without first sacrificing the animal and employing the usual analytical methods, a technique has recently been investigated which produced satisfactory results. The data show that concentrations on the order of 150 to 300 pCi of radioiodine in the thyroid of the intact rat can be detected and scanned in vivo.

One group of two rats was exposed to a controlled-field release of an aerosol consisting of submicron diatomaceous earth particles tagged with ^{131}I . Another group of two rats was injected intraperitoneally with an isotonic solution of Na^{131}I , approximately 10 times the expected field exposure.

The deposition of radioiodine in the thyroid of each rat in the two groups of animals was recorded 55 hours postexposure by use of the Picker-Nuclear Magnascanner V with color dot recorder. The thyroids were excised, then beta-counted and gamma-spectroanalyzed.

THE PHARMACOLOGIC ACTION
AND THE INDUSTRIAL HYGIENE ASPECTS
OF BERYLLIUM AND ITS COMPOUNDS

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The nature and history of berylliosis are reviewed, and the similarities and differences of experimental model systems correlated to emphasize the research areas in which our incomplete knowledge has served as a basis for doubt rather than confidence.

The manifestation of the disease in man is contrasted with that in animals. Correlation of the disease incidence with the level and kind of exposure before 1950 is compared with correlation of incidence and exposure after 1950. For instance, after the AEC limits for safe exposure were set forth in 1950, the total cases of disease in the industry declined from more than 600 during the pre-1950 era to only 8 during the 17 years since 1950, even though the industry markedly increased in size during the post-1950 period.

The chemical nature of beryllium in solution and beryllium in the biological system is discussed with particular reference to the chemical behavior of beryllium salts in the living solutions (cells). The data demonstrate the incompleteness of our knowledge regarding the relation between exposure and manifestation of disease.

The evidence collected over the past 20 years concerning the level that is toxic to man is discussed in the light of our present experience with AEC limits for beryllium.

ABSORPTION OF ^{212}Pb FROM THE
GASTROINTESTINAL TRACT OF MAN

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When ^{212}Pb was administered by mouth to three human subjects, gastrointestinal absorptions of 1.3, 8.1, and 16.0% were found. The method used was to compare 24-hour urinary excretion of these subjects with the average excretion in the same period by two subjects who received ^{212}Pb intravenously. The only obvious factor related to the wide range in absorption was age, increasing age being associated with decreased absorption. The average value of 8% is the same as that used for lead by ICRP - NCRP. It is pointed out that this choice may not be conservative for an appreciable fraction of the population. The ^{212}Pb concentration in the blood cells as function of time suggests that loss may occur only after red cell death. The implications of this to chemical toxicity of lead are considered.

ABSORPTION OF Pb-212 FROM THE GASTROINTESTINAL TRACT OF MAN

John B. Hursh¹⁾ and Jorma Suomela²⁾

For the calculation of radiation hazard from ingestion of lead isotopes it is necessary to specify the fractional amount which is absorbed from the gastrointestinal tract. The ICRP report (1) lists a value of 8 per cent based on experiments with stable lead. Because of the possibility that carrier-free lead of high specific radioactivity might behave differently, Pb-212, the 10.6 hour half-life daughter of thoron, was administered to 4 volunteer subjects. Subject A received one oral and one intravenous dose, subjects B and C each received a single oral dose, and subject D received a single intravenous dose. Based on the fractional amount excreted in the urine after intravenous injection, it was estimated that absorptions of 1.3, 8.1, and 16 per cent had occurred when Pb-212 was taken by mouth.

EXPERIMENTAL PROCEDURE

Preparation of dose

The Pb-212 dose for oral consumption was prepared by passing a stream of 50 per cent O₂ - 50 per cent CO₂ through a 0.7 mCi, Th-228 stearate, filter cartridge source (2), sweeping the released thoron gas through a fritted glass bubbler stick into 250 ml of beer at a gas flow rate of 50 ml per min. The beer was contained in a 2-liter glass bottle. Recovery of Pb-212 using beer as the vehicle was superior to recovery using coca-cola, weak ascorbic acid solution, or physiological saline. It was assumed that the foam generated by the bubbling process tended to hold the 55.6 sec. half-life thoron captive until decay occurred. Although the end-

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product was somewhat "flat", the subjects found it palatable. The period of thoron flow was adjusted so as to provide a Pb-212 activity equal to about 5 μCi at time of ingestion. A delay of > 10 hours was introduced between preparation and ingestion to ensure transient equilibrium between Pb-212 and its 60.5 min. half-life Bi-212 daughter.

A similar method was used to prepare the intravenous dose except that 20 ml of physiological saline in a 1-liter plastic bottle was used as the vehicle. Subsequently the saline solution containing Pb-212 was transferred to a silicone-coated penicillin bottle for sterilization prior to the injection of 10 ml (ca. 1 μCi Pb-212) into the antecubital vein.

In all cases gamma measurement of the preparation bottles, plastic syringes or drinking vessels, and transfer vessels showed that delivery of dose was better than 99 per cent complete. Aliquots of the dose were routinely set aside for later verification.

Doses were administered 1-2 hours after a light breakfast.

Sample collection and measurement

The subjects saved all urine and feces voided until the radioactive content decreased below the sensitivity of the counting method. Collection was made directly into 1-liter plastic cartons. The samples were measured for gamma activity by placing the collection carton on the upper surface of a 10 cm diameter, 10 cm thick NaI(Tl) crystal detector surrounded by a shielded compartment. The phototube output was fed into the first quarter of a 512-channel spectrum analyzer, calibrated to 16 keV per channel. The Pb-212 band was taken from 0-0.4 MeV and the Bi-212 band from 0.416 to 2.00 MeV. Calibration constants were established by measuring freshly prepared standardized Pb-212 solutions at intervals during the growth of Bi-212 and by including a solution volume range equivalent to that of the samples. These data enabled calculation of the correction factor to be applied to the counts in the Pb-212 band to take account of the Compton contribution from the Bi-212 gammas. For example, given a urine sample of 200 ml, the conversion constant for Pb-212 was 195 cpm per mCi and for Bi-212 was 130 cpm per nCi. The Compton correction was 0.87 times the cpm in the Bi-212 band. Backgrounds were about 350 cpm for the Pb-212 band and 215 cpm for the Bi-212 band. Counting times were 20 to 40 min. per sample. Sample volumes were estimated by weighing.

20 ml blood samples were taken from the antecubital vein, using heparin as anticoagulant, and sampling at times as noted in report of results. The blood was centrifuged within 3 min. after collection and the plasma was separated from the cells. Each fraction was weighed and counted for gamma radioactivity. A 5 cm diameter, 5 cm thick NaI(Tl) crystal was used as detector and 2 measurements were taken for each sample using single channel analyzer settings such that the Pb-212 and Bi-212 activity could be estimated separately. The blood fractions were next digested with conc. HNO_3 and hydrogen peroxide. The Pb-212 without addition of carrier was separated by use of the dithizone method (3). The 10 ml weak nitric acid solution containing the separated Pb-212 was evaporated on a 5 cm diameter stainless steel planchet yielding an essentially weight-free deposit. After a lapse of 6 hours or more to permit Pb-212-Bi-212 equilibrium to occur, the planchet was counted for alpha-activity in close proximity to a zinc-sulfide layer phototube detector. The overall counting efficiency was 34 per cent as determined by comparison with a RaD + E standard. Chemical recovery was estimated by performing 10 analyses in which known amounts of Pb-212 were added to red cell and plasma samples from non-radioactive donors. The results indicated an average recovery of 90 per cent with a standard deviation of ± 7 per cent. This correction was applied to the experimental data.

Whole body counting

At intervals, the subjects were counted in the low background laboratory (4) at this institute. The procedure included scanning measurements and a series of localization studies. For both purposes the subject lay in a supine position. The arrangement for scanning used three 12.7 x 10.2 cm NaI(Tl) crystals placed equidistant on a collar surrounding the subject with each crystal face 40 cm from the subject's longitudinal axis. During measurement the collar has a reciprocating motion as well as a movement parallel to the subject axis. Details are available in the IAEA Whole Body Counter Directory (5). For localization studies, a single crystal looked through a 5-cm-wide aperture formed by 2 1-cm-thick lead plates. The counting rate of a point source (Pb-212) moved along a line in the longitudinal axis of the subject position decreased by half when removed in either direction from a point directly under the crystal center to a point 10 cm distant. In the experimental procedure each position was counted

from the dorsal and the ventral aspect of the subject. The counts obtained were stored in a 400 channel spectrum analyzer, read out, and converted to activity according to techniques in general use.

Conversion to absolute activity units

The basis for estimating absolute activity depended on direct or indirect alpha counting rate comparison with a U.S. Bureau of Standards RaD + E standard. Secondary standards prepared by passing thoron through 0.5 N HNO₃ solutions were calibrated by evaporating aliquots on stainless steel planchets and by counting rate comparisons with the standard. The standardized solutions were useful for evaluating, by gamma counting rate comparison, the activity of substances such as beer and urine which leave residues when evaporated and were therefore not suitable for direct alpha counting.

RESULTS

Pb-212 in feces and urine

In the original planning of this research it was hoped that the personal habits of the subjects would conform to the ICRP gastrointestinal model and that the oral dose would be completely cleaned from the gut by 31 hours as specified. In sober fact, by 31 hours after intake subjects A, B, and C had cleared 23, 0, and 17 per cent of the oral dose; by 48 hours, 82, 69, and 77 per cent; by 72 hours, 82, 69, and 77 per cent; and by 96 hours, 99, 69, and 77 per cent. The above data are based on fecal sample measurements, with the measured activity decay-adjusted to time of oral intake. The intervals without increase are periods in which no fecal samples were voided. As this pattern of excretion became apparent in the first oral experiment, it became evident that gastrointestinal absorption of Pb-212 could not uniformly be determined by whole body counting techniques. For example, if 10 per cent of a 5 μ Ci oral dose were absorbed and if complete gut clearance required 96 hours, the absorbed Pb-212 would have decayed to 0.95 nCi, an activity below the threshold for measurement by this method. Accordingly other methods were sought. The use of fecal measurements alone for a direct assessment of the unabsorbed Pb-212 was unsatisfactorily imprecise because of the large decay correction factor for low activity samples excreted at late times. Determination of absorption on the basis of the amount excreted in the urine in the first 24 hours was

the method chosen. Accordingly as stated above, the oral experiments were supplement by 2 experiments in which Pb-212 was injected intravenously in order to define the per cent of the systemic burden which would appear in the urine.

The results of the measurements of urine samples from all 5 experiments appear in fig. 1. Each sample was decay-adjusted from the time of measurement to the mid-point of the collection period. The cumulative excretion from 0 to 24 hours is noted for each experiment. By reference to the injected doses listed in Table 1, it may be calculated that, when Pb-212 was administered intravenously, 4.9 per cent (subject A) and 3.4 per cent (subject B) of the dose appeared in the 24-hour urine collection. The average value, 4.15 per cent, was used to estimate absorption in the oral experiments yielding the results entered in Table 1.

After intravenous injection, subject A excreted 0.29 per cent dose in the fecal collection period 0 to 48 hours. Subject D excreted 0.25 per cent dose for the same fecal collection period. These estimates are maximized since all sample measurements were decay-adjusted to injection time.

Pb-212 uptake by blood cells

As an adjunct to the main experiments a preliminary in vitro experiment was performed in which 10 ml volumes of heparinized blood were incubated with added Pb-212 + Bi-212 at 37°C in a water bath. At pre-selected times the blood samples were removed and centrifuged. The plasma and cell fractions were then assayed for Pb-212 and Bi-212 activity by gamma measurement as described above. The results are shown in fig. 2. It is seen that at 16 min. the cells have taken up 99 per cent of the Pb-212 added to the whole blood. The uptake of Bi-212 by the cells occurs at a slower rate and gives no evidence of a tendency to concentrate with respect to the plasma.

In view of these tendencies, it is not surprising that the results after oral or intravenous administration as shown in Table 2 demonstrate only small amounts of Pb-212 in the plasma fraction of the drawn blood samples. The estimation of Pb-212 in the cells shows satisfactory agreement between the two measurement methods. The last column of the table lists results obtained by assuming that the total mass of circulating blood cells in grams is equal to 40.3 times the body weight in kg (6). Since the Pb-212 content per gram cell (as shown in the next to last column) has been decay-corrected to time of intake, it may be seen that little if any Pb-212 is

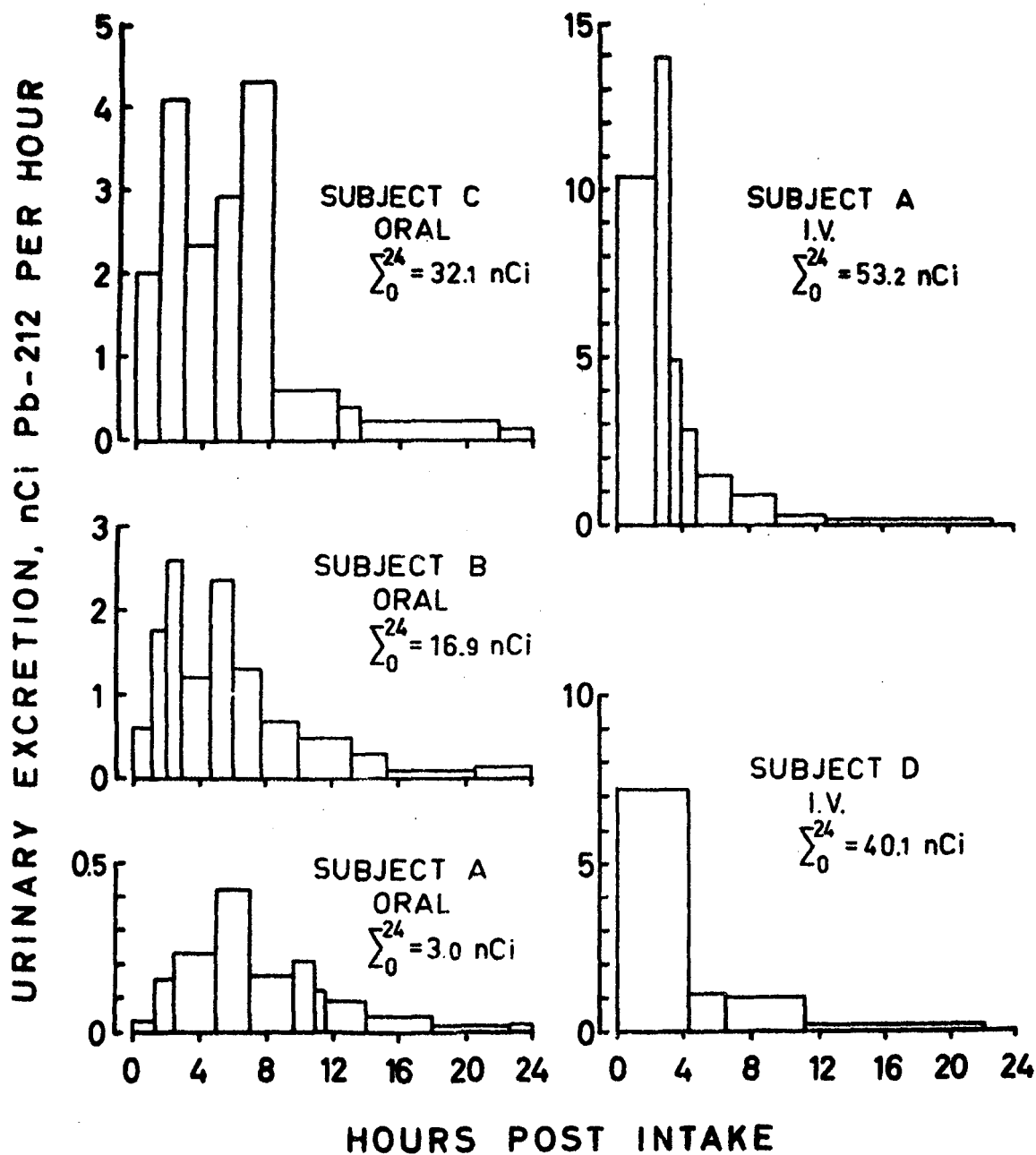


Fig. 1. The ²¹²Pb urinary excretion rate as a function of time for oral and intravenous intake.

Table 1

Absorption of orally administered Pb-212 calculated by comparing the urinary excretion with that after intravenous administration.

Subject	Age yr.	Wgt. kg	Dose μ Ci	Mode	Urinary Loss ^{x)} nCi/24 hours	Absorption	
						μ Ci	% dose
A	59	75	1.08	iv	53.2	-	-
D	39	76	1.17	iv	40.1	-	-
A	59	75	5.05	oral	2.98	0.067	1.3
B	40	84	5.01	oral	16.9	0.404	8.1
C	27	63	4.80	oral	32.1	0.768	16.0

x) Activity decay-corrected to midpoint of collection interval.

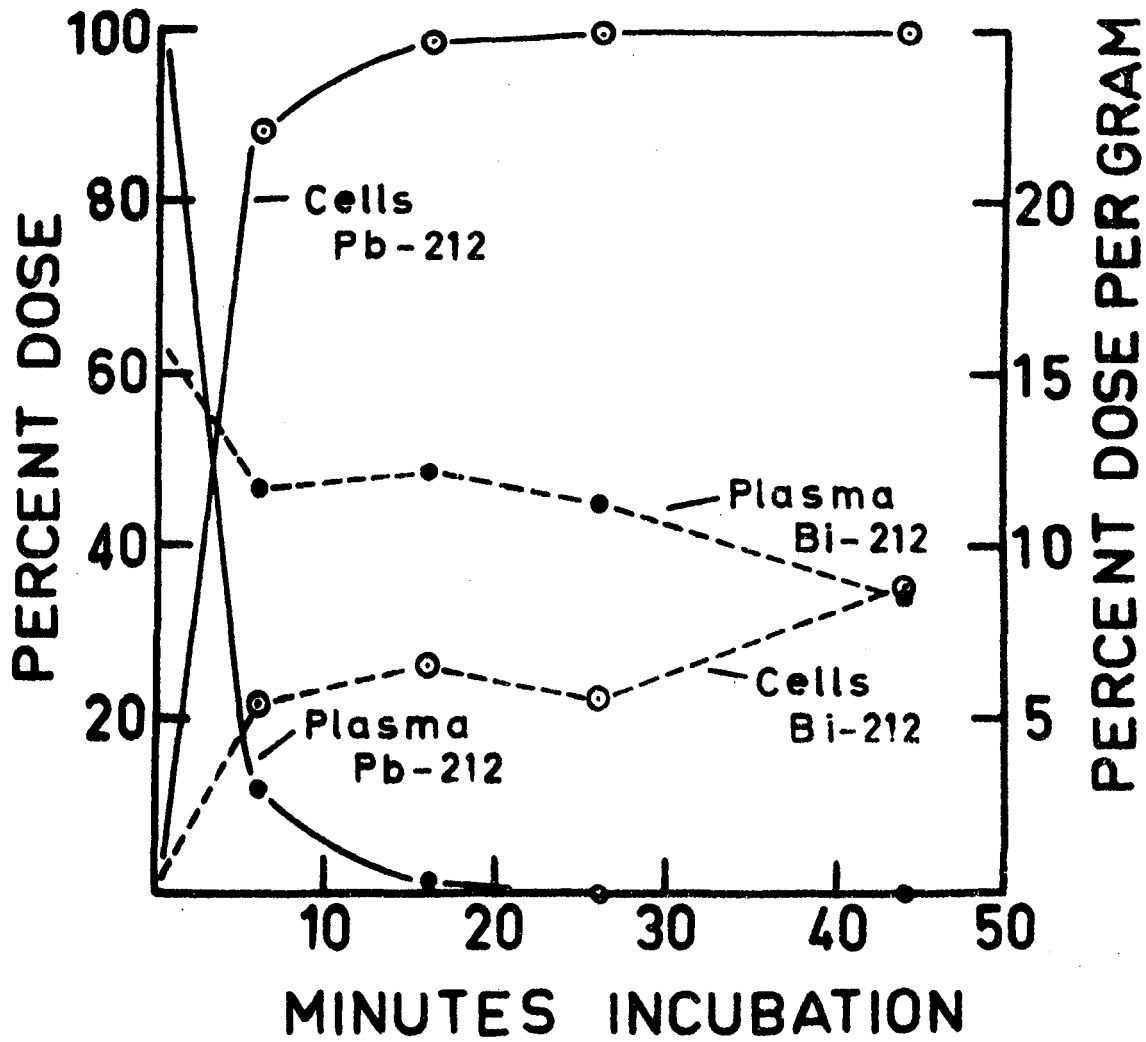


Fig. 2. The in vitro uptake of ^{212}Pb and ^{212}Bi by the blood cells. The solid lines pertain to the left ordinate scale and the dotted lines to the right ordinate scale.

Table 2

Pb-212 and Bi-212 content of plasma and blood cells in per cent absorbed or injected dose per gram referred to sampling time except as otherwise indicated.

Subject and Mode	Sample	Hours Post Dose	Plasma	Cells	Cells	Cells	Cells Pb-212 t = 0	Total Cells Pb-212 t = 0 % dose
			Pb-212 ^{x)}	Pb-212 ^{x)}	Pb-212 ^{xx)}	Bi-212 ^{xx)}		
A i.v.	1	0.33	0.00036	0.0106	0.0104	-	0.0107	32
	2	1.67	0.00009	0.0124	0.0140	-	0.0147	44
	3	4.05	-	-	0.0150	-	0.0195	59
	4	6.75	-	0.0124	0.0113	-	0.0184	56
	5	49.75	-	0.00054	-	-	0.0139	42
B oral	1	2.50	0.00009	0.0073	0.0057	0.0033	0.0077	26
	2	6.00	0.00009	0.0130	0.0116	0.0115	0.0182	62
	3	8.00	0.00006	0.0136	0.0142	0.0111	0.0234	79
	4	24.50	0.00006	0.0058	0.0031	0.0079	0.0221	75
C oral	1	2.60	0.00025	0.0085	0.0076	0.0052	0.0096	24
	2	6.10	0.00008	0.0124	0.0123	0.0096	0.0184	47
	3	8.00	0.00011	0.0132	0.0124	0.0117	0.0216	55
	4	24.50	-	0.0048	0.0055	0.0031	0.0255	65
	5	48.50	-	0.0010	0.0010	0.0012	0.0236	60
D i.v.	1	0.35	0.00033	0.0221	-	-	0.0226	69
	2	1.58	0.00011	0.0250	0.0260	0.0210	0.0285	89
	3	7.10	0.000052	0.0190	0.0200	0.0190	0.0310	93
	4	23.63	-	0.0059	0.0061	0.0060	0.0280	86
	5	47.57	-	0.0012	0.0016	-	0.0325	100

x) Measured by alpha counting

xx) Measured by gamma counting

lost except by decay. The data for all subjects show an increase in cell content (last 2 columns) for the early sampling times, believed to be due to the return of Pb-212 from the tissues to the blood as the plasma concentration is decreased, leading to further uptake by the cells. The generally higher estimates of Pb-212 uptake in the total cells for subject D are obviously in error and it seems likely that the formula used to calculate total cell mass resulted in an overestimate in this case. It is recognized (7) that such formulae may be in error when applied to specific normal individuals.

Comparison of Bi-212 and Pb-212 metabolism

Although the data on the fate of Bi-212 are less complete than are the data for Pb-212, it has been found that at the time of voiding, the Bi activity in the urine samples is about twice that of the Pb. The blood cells in general show (Table 2) slightly less Bi-212 than Pb-212, leading to the interpretation that the rate of Bi-212 loss from the cells must be low in respect to its rate of formation from Pb-212. Harbers (8), with ThX injected into rabbits, finds an excess of Bi-212 over Pb-212 in the urine and the reverse to be true for the blood cells. Stover (9), after intravenous injection of Pb-212 into dogs, finds an excess of Bi-212 over Pb-212 in the urine, and no disequilibrium for blood cells measured 20 min. after sampling. Finally the whole body counter localization studies in the present experiments showed no marked differences in the overall body distribution of Pb-212 and Bi-212.

Whole body counter findings

Scanning measurements made with the whole body counter facility in the two intravenous experiments showed the expected 10.6 hr half-life decrease when small corrections were made for urinary loss. For the three oral experiments the activity found at the latest scan (ca. 48 hr), decay corrected to $t = 0$, yielded values of 18, 39, and 23 per cent of the ingested dose for subjects A, B, and C. As explained above, these amounts include activity in the lower bowel which was voided subsequently. With the exception of subject A who voided 17.1 per cent at 73 hours, the delay in voiding resulted in activities, near the threshold of detection, subject to large correction factors for decay, and not of use in estimation of absorbed fraction.

The measurements with the lead-columnated crystal yielded results of which the data from examination of subject A presented in fig. 3 is a typical example. The oral experiment data, measured 28 hours after intake and normalized to time of intake and to 1 μ Ci dose, shows an activity peak in the lower bowel region. The survey 6 hours after intravenous injection (counts corrected to injection time) shows a concentration of activity in the blood-rich area of the heart, lungs, and liver. This result agrees well with what might be expected from the high Pb-212 content of the blood cells.

DISCUSSION

Absorption of Pb-212

The wide variation between the absorption of subject A and subject C (12 times) calls for comment. The validity of the indirect method used to calculate absorption depends on the assumptions: 1) that the fraction of the systemic Pb-212 excreted in the urine will be the same whether entry is via the gut or by intravenous injection, and 2) that this fraction does not vary greatly from one healthy individual to another. Both these assumptions are biologically plausible but proof that they apply to excretion of Pb by man is lacking.

Support for the calculated absorption values comes from the blood data in Table 2. When the absolute concentrations of Pb-212 in the blood of subjects B and C are converted into per cent absorbed dose, the resulting values are quite comparable to those obtained in the two intravenous experiments. This would not be expected if the calculated systemic burdens of subjects B and C were greatly in error.

Proceeding on the grounds that the wide range in fraction absorbed is real, it is pertinent to inquire as to possible causes of the variation. The 3 subjects for the oral experiment were healthy active individuals. Compared with the ICRP reference man (1) who has 2 grams potassium per kg body weight, subjects A, B, and C had 1.93, 1.89, and 2.63 grams per kg. These values suggest normal and above average ratios of lean body mass to total weight. The only suggestive relationship is the decrease in absorption of Pb-212 with increase in subject age. The present data are insufficient to prove age dependence and may represent fortuitous sampling of extremes on a curve of normal biological variability. Either interpretation makes it likely that there is a group of individuals for which the

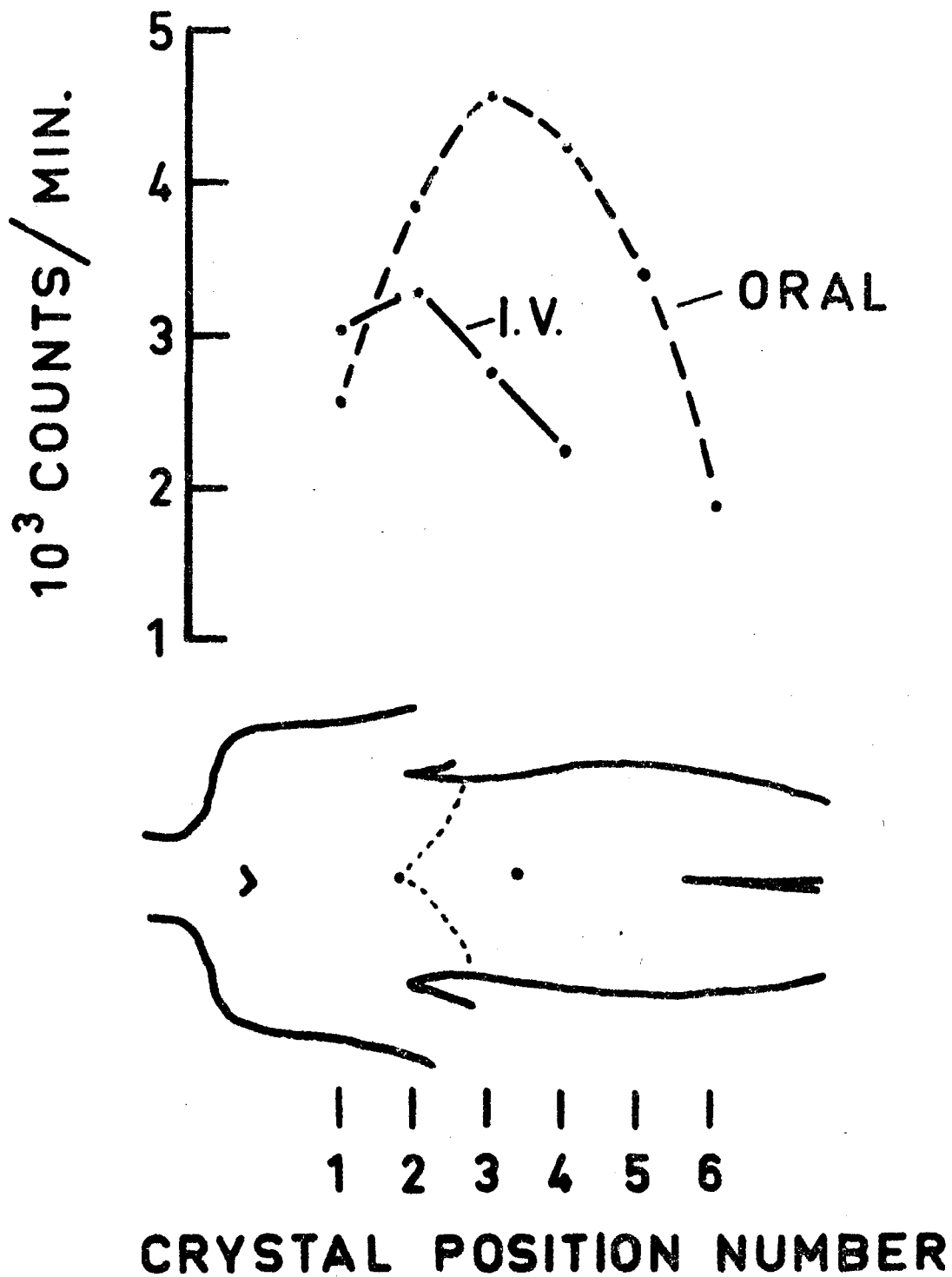


Fig. 3. Comparison of radioactivity profile after intravenous (+ 6 hours) and oral (+ 28 hours) intake for subject A. Actual counting rate was decay adjusted to time of intake and, for oral results, normalized to 1 μ Ci intake.

absorption value of 8 per cent is not conservative for health protection uses. Further experiments are needed to define the size of this group.

Concentration of Pb-212 in the blood cells

The concentration of lead in the blood cells was observed by early workers (10, 11) and confirmed many times since. Hevesy and Nylin (12) report that 6 hours after intravenous injection of Pb-212 into man, 45 per cent of the injected amount is in the blood cells. Stover (9) found, for dogs, that 65 per cent of an intravenous dose of Pb-212 was in the blood cells at 5 hours. Schubert and White (13) injected carrier-free Pb-210 into rats and detected 17 per cent of the dose in the blood cells at 30 minutes. The rate of loss from the red cells (not including radioactive decay) was 0.0187 per hour for dogs (9) and 0.0231 per hour for rats (13). Hevesy and Nylin (12) state that, in man, the loss rate is less than 0.04 per hour. The data in Table 2 show no consistent loss rate and render unlikely a loss rate greater than 0.007 per hour. If it be assumed for man that the death of the red cell releases the fixed lead and that the lead is removed from the body for example by secretion in the bile, a minimum release rate can be calculated. Since the average life span of the human red cell is 120 days, the equivalent lead loss rate would be 0.0083 per day or 0.00035 per hour. Neither the data of Hevesy and Nylin nor our findings can be regarded as excluding this interpretation. On the positive side a test of its feasibility can be applied by using data from the classical lead studies of Kehoe (14). He finds that the daily food intake of stable lead by occupationally unexposed individuals is about 0.35 mg and that the corresponding level in the blood is 0.030 mg per 100 ml. If the blood volume of an adult is taken as 5400 ml (1), the total lead burden carried by the blood becomes 1.62 mg. Assuming an average absorption of 8 per cent from the gastrointestinal tract and that 50 per cent of the absorbed lead is fixed by the blood cells, the daily uptake of lead becomes 0.014 mg. Since the lead concentration in the blood cells must be in a steady state the loss rate constant is 0.014 divided by 1.62 = 0.0086 per day, a value very near to the rate of 0.0083 per day based on the proposal that lead is lost only by death of the blood cell.

If this interpretation is valid it enables a circumstantial statement of the protection against lead poisoning given by the lead fixing property of the red blood cell. The maximum lead binding capacity of the cells can

be estimated from the report by Kehoe et al (15). They performed experiments in which 2 human subjects were given supplementary soluble lead by mouth at the rate of 1 and 2 mg per day over periods of years. From examination of their data it is found that the blood level rises to a mean value of about 0.06 mg lead per 100 ml and does not greatly exceed this. If this concentration represents saturation, the blood cell mechanism could turn over about 0.028 mg lead per day with only transient increases in the plasma concentration. Since by hypothesis the cells are able to pick up only half of the lead introduced from the gut, this would conform to a total systemic intake of about 0.056 mg per day. If the systemic intake exceeds this rate, the average plasma lead concentration must rise. This leads to proportionally increased skeletal deposition and urinary excretion. Toxicity symptoms might be anticipated at plasma lead concentrations up 4-5 times above unexposed mean levels on the grounds that equivalent increases in urinary excretion rates have been observed (14) to be associated with lead intoxication.

Comparison of behavior of "carrier-free" Pb-212 and stable lead

In terms of the discussion above, it is apparent that the Pb-212 in oral and intravenous administration was no longer "carrier-free" after it entered the gut or the circulatory system. Accordingly absorption from the gut should not differentiate between Pb-212 and stable lead ions as such. If Pb-212 is introduced with supplementary lead in amounts so large as to saturate the binding sites of the blood cells, marked differences in tissue distribution and urinary excretion might be expected.

SUMMARY

When Pb-212 was administered by mouth to 3 human subjects gastrointestinal absorptions of 1.3, 8.1, and 16.0 per cent were found. The method used to determine absorption was to compare the 24-hour urinary excretion of these subjects with the average excretion in the same period by 2 subjects who received Pb-212 intravenously. The only obvious factor related to the wide range of absorption was that of age, increasing age associated with decreasing absorption. The average value of 8 per cent is equal to that used by ICRP. It is pointed out that this choice may not be conservative for an appreciable fraction of the population. The Pb-212 concentrations in the blood cells as a function of time after oral or

intravenous administration suggest that lead may be released from the binding sites only when the red cell dies. The relationship of this proposal to permissible intakes of stable lead is considered.

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BIBLIOGRAPHY

1. Recommendations of the International Commission on Radiological Protection, ICRP Publication 2. Report of Committee II on Permissible Dose for Internal Radiation, Pergamon Press, New York (1959).
2. Hursh, J.B., and Lovaas, A., Preparation of a dry Th-228 source of thoron, *J. inorg. nucl. Chem.* (1966).
3. Gibson, W.M., The radiochemistry of lead, NAS-NS-3040.
4. Lindell, B., and Magi, A., A new laboratory for whole body counting, *Acta Radiol. Suppl.* 254, Stockholm (1966), p. 135-137.
5. Directory of whole-body monitors (new edition, in preparation), IAEA, Vienna.
6. Best, C.H., and Taylor, N.B., The Physiological Basis of Medical Practice, 5th edition p. 18, 19. Williams and Wilkins Co., Baltimore (1950).
7. Glasser, O., Medical Physics Vol. 1, p. 123. The Year Book Publishers Inc., Chicago (1944).
8. Harbers, E., Untersuchungen über Verteilung und Ausscheidung der in vivo entstehenden Folgeprodukte des Thorium X, *Zeit. f. Naturforschung*, Band 7 b, Heft 6, p. 363-364 (1952).
9. Stover, B.J., Pb²¹² (ThB) Tracer studies in adult beagle dogs, *Proc. Soc. Exp. Biol. Med.* 100, p. 269 (1959).
10. Dauwe, O., Contribution à l'étude expérimentale du Saturnisme Aigu, *Arch. intern. pharmacodynamie* 17, p. 387 (1907).
11. Behrens, B., and Pachur, R., Die Verteilung und der Zustand Kleinster Bleimengen im Blut, *Arch. Exp. Path. u. Pharmacol.* 122, p. 319 (1927).
12. Hevesy, G., and Nylin, G., Application of "Thorium B" labelled red corpuscles in blood volume studies, *Circulation Res.* 1, p. 102 (1953).
13. Schubert, J., and White, M.R., Effect of sodium and zirconium citrates on distribution and excretion of injected Radiolead, *J. Lab. and Clin. Med.* 39, p. 260 (1952).
14. Kehoe, R.A., Exposure to lead. *Occupational Med.* 3, p. 156 (1943).
15. Kehoe, R.A., Cholak, J., Hubbard, D.M., Bambach, K., and McNary, R.R., Experimental studies on lead absorption and excretion and their relation to the diagnosis and treatment of lead poisoning, *J. Ind. Hygiene and Toxicol.* 25, p. 71 (1943).

HIGHLIGHTS OF FLUORIDE TOXICOLOGY*

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The discussion of the toxicology of inorganic fluorides is based on six properties studied in sufficient quantitative detail that reliable evaluations can be made. The characteristics of fluoride are described briefly, e. g., its ubiquitous occurrence, its typical accumulation in the skeleton, its nature as a characteristic bone seeker, and the well-characterized acute and chronic poisonings. The hazards of fluoride poisoning from industrial uses, from pesticidal applications, and from use as a public health measure to improve tooth health and most recently as a possible therapeutic agent in certain bone disorders, make safety evaluation important. The six thoroughly studied effects of fluoride are (1) acute fluoride poisoning, (2) kidney injury, (3) thyroid effects, (4) growth effects, (5) crippling fluorosis, and (6) mottled enamel. The mechanisms of these effects are unknown.

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Introduction

The continued and increasing use of fluorides makes timely a brief resumé of some of the highlights of fluoride toxicology.

Fluoride, biologically, is a ubiquitous bone seeker with a variety of physiologic and toxic effects some of which constitute substantial hazards if neglected, but also with properties which permit a ready control of the hazards and an assurance of safety. Six properties have been so well studied that they may serve as landmarks of fluoride toxicology; these will be discussed in some detail.

Acute Poisoning

The certainly lethal dose for the standard 70-kg adult man is estimated to be 2500 to 5000 mg F. Because sodium fluoride is roughly half F, the lethal dose is 5 to 10 g of sodium fluoride or 1 to 2 teaspoonfuls.¹, p. 12

The course of acute poisoning is rapid. Taken orally, death often occurs in 2 to 4 hours; many patients who live longer than 4 hours recover. The reasons for the rapidity are not hard to find: in high concentrations fluoride is a powerful metabolic inhibitor. Rapid absorption and rapid distribution throughout the body of a lethal or near lethal dose quickly permit dangerously

high concentrations of fluoride ion to develop. Patients surviving for 4 or more hours have an improved prognosis because fluoride is rapidly deposited in the skeleton and rapidly excreted in the urine, thereby decreasing blood and tissue concentrations below fatal levels.² The remarkable ability of the body to dispose of fluoride accounts for the fact that large (but not lethal) doses of fluoride may be tolerated without severe toxic symptoms. For example, in unsuccessful chemotherapeutic trials some years ago, one patient suffering from leukemia received as much as 23 mg F/kg intravenously daily for nine days without apparent toxic effects. Each such dose must have exceeded a quarter of the acute lethal dose given by mouth. Toxic doses of fluorides taken orally have a salty or soapy taste; nausea, vomiting, diarrhea, cramping develop promptly; with large enough doses, collapse, coma, and death ensue. Death is usually attributed to a blockade of necessary enzyme or transport systems; which such system(s) are responsible for death cannot be said. Calcium-binding by overwhelming doses of fluoride probably accounts for the unclotted state of the blood reported in some autopsy examinations.

Kidney

When experimental animals are maintained for periods of months on diets or on drinking water containing over 100 ppm F, various changes in kidney structure and function can be demonstrated. Tubular cells die and regenerate, interstitial fibrosis develops, and in some animals a remarkable dilatation of certain tubules begins in the loop of Henle, later involving the distal convoluted tubule.³ Such dilated tubules have not been reported in man; in fact the only kidney effects ascribed to prolonged human exposures to fluorides are those in certain hospitalized patients in India or China with diagnoses of skeletal fluorosis who exhibited reduced kidney function (e.g., urea clearance).⁴ These patients were poverty-stricken laborers from small villages, chronically malnourished, and afflicted with various intercurrent diseases. The causative role of fluoride in their kidney impairment has by no means been established. In certain patients suffering from advanced kidney disease, or in animals with specifically injured kidneys (such as occurs in uranium poisoning), the ability to excrete fluoride in the urine is not seriously impaired until the kidneys fail. In terminal uremia, fluoride is excreted more slowly than usual, blood fluoride levels increase and, as a result,

the concentrations of fluoride in the skeleton increase markedly.⁵

Thyroid

Many studies have been made of the effects of fluoride on the thyroid, perhaps partly because fluoride's being a halogen raised the question whether fluoride, like iodine, might be taken up preferentially and stored by the thyroid. It is not. Perhaps interest in the thyroid has continued because textbooks have quoted an observation made years ago of a struma or goiter in a dog repeatedly given very large doses of fluoride. Large amounts of F do alter the thyroid. More than 50 ppm F in the ration of drinking water administered over periods of days to years have been responsible for structural or functional changes in the thyroid in a number of species of animals.¹, P. 98 When the diets contained less than 50 ppm of fluoride, few thyroid alterations were found. The question whether fluoride is associated with human goiter can be answered simply. If the drinking water contains practically none or up to 3 ppm of fluoride in areas where iodine is deficient, goiters develop regardless of the fluoride content of the water. On the other hand, when iodine is given in such areas, for example as iodized salt, whether the

drinking water contains only a trace or up to several parts per million of fluoride, the prophylactic effect of iodine is exerted. There is no direct relation of fluoride to goiter in such exposures.

Growth

The dairy cow, the most sensitive species, given a ration containing 40 parts per million of fluoride for four or five years, will lose weight and become unthrifty. Other species studied sustain growth impairment only with higher levels of dietary fluoride.¹, p. 337 If a single report by Japanese investigators is discounted, no adverse effects on human growth are known. In this study, the heights and weights of children living in two small settlements in a mountainous area of Japan were compared. For one group living up the mountainside, the water supply obtained from shallow wells presumably contained excess fluoride because teeth were mottled. The children in this settlement were somewhat shorter and weighed somewhat less at a given age than the children living below in the valley whose teeth were not mottled and where presumably the fluoride intake was lower.⁶ The lack of specific information on the fluoride intake, on fluoride excretion, on the nutritional status, on the hereditary background, and of other pertinent

information about these two groups prevents acceptance of the investigator's conclusion that fluoride was responsible for the growth difference. On the other hand, reliable evidence of the normal growth of children in a fluoridated community has been gathered. Possible growth effects of fluoride consumed at 1 ppm in the drinking water were sought during a period of 10 years in the two cities of Newburgh and Kingston, New York. The water supply of Newburgh was fluoridated, one of the earliest in the country; Kingston, with about 0.05 ppm in the community drinking water, was maintained as a control city. Thorough examinations of hundreds of children annually showed beyond doubt that growth, as revealed by height, weight, bone age estimated from wrist and knee radiographs, and by other indices, was exactly comparable in the Newburgh and in the Kingston children.⁷ A few more cortical defects were found in the radiographs of the bones of the Newburgh children; in the opinion of the radiologist, Dr. Caffey, the difference was within the limits of normal variation.

The question of adverse effects on reproduction has been examined. Large doses of fluoride interfere with fertility and normal reproductive performance.¹, p. 121 Pregnant animals of several species have been fed fluoride

without teratogenic effects. Relatively large doses of fluorides to female rats during gestation caused changes in the jaw bone and in the teeth of neonatal rats.⁸

Chronic Fluorosis

Crippling fluorosis was first described as an industrial disease. Certain workmen handling powdered cryolite in a Danish factory developed a "poker back" characterized by a fixation of the spine. X-ray examination showed (a) that the broad ligaments had calcified, (b) that the skeleton exhibited generalized hypermineralization with "moth-eaten" areas of hypo-mineralization, and (c) that exostoses projected both from flat and from long bones.⁹ Crippling fluorosis developed when men inhaled as dust 20 to 80 or perhaps more mg F per day for protracted periods, perhaps 10 to 20 years.¹⁰ This readily preventable illness should never recur. When fluoride exposures are unavoidable, urine analyses offer a reliable quantitative index of the exposures. The lengthy latent period permits the degree of exposure to be identified and controlled by appropriate safety measures. Radiographic examination will reveal asymptomatic osteosclerosis long before joint function is impaired. Skeletal fluoride is mobilized and slowly removed from the body when exposure is reduced.

Interest has recently heightened in the possibility of using fluorides to induce new bone formation, in treating osteoporosis, or more importantly, as a prophylactic agent against it.¹¹ Large doses of fluoride, 30 to 100 mg F per day, produced in some patients with osteoporosis or other bone lesions a positive calcium balance, a reduction in skeletal calcium-47 retention, and ultimately the formation of new bone which, although abnormal, may at least have a splinting effect. A major health benefit to the elderly would accrue if the fluoride in community drinking water (especially at elevated concentrations) lessened the incidence of spontaneously occurring osteoporosis. Some encouraging evidence to this effect was recently gathered in North Dakota¹² by comparing the incidence in the northwestern part of the state, where certain community drinking water supplies contain 3 to 6 ppm F, with that in the southeastern part, where drinking water supplies contain 0.1 to 0.3 ppm F. A radiographic survey of about a thousand individuals in these two areas showed a lessened incidence of osteoporosis with age in women living in the high-fluoride area. No such contrast was noted among the men. Unexpectedly, calcification of the aorta was markedly reduced both in men and women in the high-fluor-

ide areas.

Preliminary studies of the osteosclerotic changes have by no means clarified the mechanism of this effect. Recent attempts to understand the mechanism have produced several remarkable findings.

(a) The rapidity of calcium-47 deposition in the skeleton of osteoporotic subjects was reduced after prolonged fluoride treatment, i.e., less bone participated in the rapid exchange.¹³ Remineralization, or new bone formation, which reduced the amount of bone "available" to the circulation would plausibly account for such a difference.

(b) A narrowing of the x-ray diffraction peaks of the bones of fluoride-treated men or animals is interpreted as an increase in the average crystal size of the apatite mineral.¹⁴ Larger crystals possess smaller surface-to-weight ratios and have therefore less exchangeable calcium. Furthermore, the dissolution rates of larger crystals are less than of smaller ones, an effect which together with the considerably lower solubility attributed to the fluorapatite lattice would contribute greater "stability" to bone. The role of these factors in the reduced osteoporosis of elderly women (see above) has not been evaluated nor has it in

the new bone formation either in chronic fluorosis or in fluoride-treated osteoporotic patients.

(c) Paradoxically, prolonged treatment with large doses of fluoride reduces the incorporation of proline into bone collagen.¹⁵ Carbon-14-hydroxyproline tends to accumulate in the cell rather than in collagen. Such an interference with osteocytic metabolism would be expected to accompany a reduction in new bone formation. Perhaps this effect accounts for the fact that in patients suffering from crippling fluorosis certain local areas of bone may become hypomineralized; in these areas, described as "moth-eaten," bone formation is notably impaired.

(d) Several observations direct attention to an intriguing possibility that the parathormone is somehow involved in the osteosclerotic response to repeated large doses of fluoride. The parathyroid gland enlarges and becomes hyperplastic in sheep and in rabbits receiving 200 ppm F in their rations.¹⁶ The rat seems not to be responsive; at any rate, rat bone, whether it contains extra fluoride or not, serves successfully as a source of calcium when parathormone is put into the tissue culture.¹⁷ According to a complicated hypothesis, the parathyroid-mediated osteosclerosis involves two se-

quences.¹⁸ First, parathyroid stimulation. Fluoride deposited in bone renders the bone calcium somewhat less available, thereby lowering slightly the concentration of blood calcium, which stimulates parathormone secretion, increasing the activity of the bone cells and re-establishing the normal blood calcium. Second, increased collagen formation. Recent preliminary evidence indicates that a secondary effect of parathyroid stimulation occurring later than the well-established mobilization of bone mineral may under certain circumstances be a speeded-up collagen formation. In an animal chronically exposed to excessive amounts of fluoride, new bone formation might follow.

The brightest aspect of these sophisticated attempts to understand how toxic doses of fluoride alter the structure and function of bone may well be not these and other findings but the enthusiasm of the attack.

Mottled Enamel

The term "mottled enamel" or dental fluorosis embraces several degrees of severity. "Very mild mottling" and "mild mottling" (the first and second degrees) describe hypoplasias that are not esthetically damaging, i.e., are not brown-stained teeth. Brown stain, the image understandably misapplied by lay readers to any de-

gree of mottling, accompanies only the more severe hypoplasias: "moderate mottling" -- brown stain under a smooth enamel surface (third degree), and "severe mottling" -- brown stain in teeth with pitted or grooved surfaces (fourth degree). Dean's classic epidemiological studies of the incidence and severity of mottling in the permanent teeth of 12-14-year-old residents of American communities where the drinking waters contained from traces up to several ppm F established the increase in mottling with increasing F concentration above 1 ppm F.¹⁹ The community index of mottling, Dean's device for summarizing data, takes the average based on the two most severely mottled teeth for each child; the community index increases in a linear fashion with the logarithm of F concentration above 1 ppm. Brown stain from fluoride is not observed in communities in temperate climates if the drinking water contains two ppm F or less. Thus in water fluoridation, the safety margin against brown stain, while not large, is established with an exceptional reliability.

Mottled enamel develops only while teeth are forming prior to eruption. Excessive fluoride impairs the function of the ameloblasts, the cells that form the enamel, and hypoplasias result. Hypoplasias of the

enamel surface are by no means uniquely due to fluoride; these changes are non-specific at least to the extent that several other causative agents are known, e.g., trauma, vitamin deficiency, febrile illness. Even in communities with only traces of F in the drinking water, about one child in five has detectable non-specific hypoplasia. With sufficiently elevated water F concentrations, brown stains develop, but not uniformly in every child. For example, in Colorado Springs, when the water supply contained about 2.5 ppm F, while an occasional child had severely mottled teeth, about 10% of the children exhibited moderate mottling, most of the children showed only very mild or mild degrees of mottling, and many had "normal" tooth surfaces. The factors controlling this distribution of effect are unknown.

Dental Caries

Despite the evidence that slowly accumulated, during a score of years, of caries resistance conferred by fluoride, experts in dental health were slow to accept Dean's convincing demonstration that the DMF (decayed, missing, and filled teeth) rates decreased with increasing F concentrations in community water supplies in temperate areas of the United States.²⁰ The DMF rates in the permanent teeth of 12-14-year-old children

ranged from 6-10 "bad" teeth per child with traces of F in the water, to 4-5 with 0.5 ppm F, to 2-3 with 1 to 4 ppm F in the water. At 1 ppm F, the hazard of brown stain is minimal; this concentration in a temperate climate thus offers optimal tooth health with minimal hazard of injury. Extensive studies in the succeeding score of years have established the wisdom of this choice. No ill effects are established in any individual regardless of age or state of health in communities using fluoridated drinking water supplies with recommended concentrations (ranging from 1 ppm F down, depending on the annual mean temperature, to 0.7 ppm F). The fluoridation of public water supplies is widely acclaimed as one of the most important public health developments of our time.

1. HODGE, H. C., and SMITH, F. A. Fluorine chemistry
Vol. IV, ed. J. H. Simons, Academic Press, New
York, 786 pp., 1965.
2. HALL, L. L. Acute fluoride toxicity, Master's Thesis,
Univ. Rochester, 1967.
3. PINDBORG, J. J. The effect of 0.05 per cent dietary
sodium fluoride on the rat kidney, Acta Pharmacol
Toxicol 13:36-45 (Dec.), 1956.
4. SIDDIQUI, A. H. Fluorosis in Nalgonda district,
Hyderabad-Deccan, Brit Med J 2:1408-1413 (Dec.),
1955.
5. TAVES, D. R., TERRY, R., SMITH, F. A., and GARDNER,
D. E. Use of fluoridated water in long-term
hemodialysis, Arch Intern Med 115:167-172
(Feb.), 1965.
6. BABA, H., and KAWAHARA, S. Shikoku Acta Med 1:78-83,
1950.
7. SCHLESINGER, E. R., OVERTON, D. E., CHASE, H. C., and
CANTWELL, K. T. Newburgh-Kingston caries-fluorine
study, XIII. Pediatric findings after ten years,
J Amer Dent Assoc 52:296-306 (March), 1956.
8. FLEMING, H. S., and GREENFIELD, V. S. Changes in the
teeth and jaws of neonatal Webster mice after
administration of NaF and CaF₂ to the female

- parent during gestation, J Dent Res 33:780-788 (Dec.), 1954.
9. ROHOLM, K. Fluorine intoxication, H. K. Lewis, London, 364 pp., 1937.
 10. MØLLER, P. F., and GUDJONSSON, Dk. V. Massive Fluorosis of Bones and Ligaments, Acta Radiol 13:269-294, 1932.
 11. HODGE, H. C., and SMITH, F. A. Fluorides and Man, in press, Ann Rev Pharmacol, 1968.
 12. BERNSTEIN, D. S., SADOWSKY, N., HEGSTED, D. M., GURI, C. D., and STARE, F. J. Prevalence of osteoporosis in High- and Low-Fluoride Areas in North Dakota, J A M A 198:499-504 (Oct.), 1966.
 13. NEER, R. M., ZIPKIN, I., CARBONE, P. P., and ROSENBERG, L. E. Effect of sodium fluoride therapy on calcium metabolism in multiple myeloma, J Clin Endocrinol and Metab 26:2059-68 (Oct.), 1966.
 14. BERNSTEIN, D. S., and COHEN, P. Use of Sodium Fluoride in the treatment of Osteoporosis, J Clin Endocrinol and Metab 27:197-210 (Feb.), 1967.
 15. PROFFIT, W. R., and ACKERMAN, J. L. Fluoride: Its

- effect on two parameters of bone growth in organ culture, Science 145:932-34 (Aug.), 1964.
16. FACCINI, J. M. Inhibition of bone resorption in the rabbit by fluoride, Nature 214:1269-71 (June), 1967.
17. RAISZ, L. G., and TAVES, D. R. Calcified Tissue Res. in press.
18. NICHOLS, G., Jr., FLANAGAN, B., and WOODS, J. F. "Parathyroid influences on bone biosynthetic mechanisms," in The Parathyroid Glands: Ultra-structure, Secretion, and Function, ed. by P. J. Gaillard, R. V. Talmage and A. M. Budy, Univ. Chicago Press, Chicago, pp. 243-60, 1965.
19. DEAN, H. T. The Investigation of physiological effects by the epidemiological method, in Fluorine and Dental Health, Amer Assoc for the Advancement of Science, Washington, D. C., 1942.
20. DEAN, H. T. Endemic fluorosis and its relation to dental caries, U S Public Health Reports 53:1443-1452 (Aug.), 1938.

EFFECT OF THE VALENCE STATE OF ARSENIC ON THE ANALYSIS AND
INTERPRETATION OF BIOASSAY SAMPLES

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ABSTRACT

Among the many complicating factors in using biological samples for estimation of exposure is lack of knowledge of the metabolic pathways involved. In the case of arsenic a large amount of information is available on its metabolic behavior because of its historic role as a toxic material and its similiarity to metabolically important phosphorous. A complicating factor results from the fact that arsenic has two valence states which are markedly different in metabolism and presumably toxic effects. This poses a special problem in analysis since biological material is usually destroyed by oxidation which also serves to oxidize trivalent arsenic.

This report encompasses a discussion of some of the metabolic differences between trivalent and pentavalent arsenic as well as some experimental results on the micro-coulometric determination of arsenic. Attempts to determine the distribution of the two valence states in biological samples will also be discussed.

Introduction

The use of excreta samples for assessing exposure to toxic materials requires that some information on likely metabolic pathways be available. Even when this information is available, relating severity of exposure to excretion levels is difficult.

In the case of arsenic substantial information is available with regard to interaction with enzyme systems. There are, however, two factors which complicate the study of this element in man. One is its ubiquity in nature and the second is the difference in metabolic behavior of the penta- and trivalent forms.

An excellent review of this problem, which included extensive experimental data, was published by Schroedor and Balassa in 1966⁽¹⁾. Among the authors' conclusions were that pentavalent arsenic as arsenate is non-toxic in normal amounts, is rapidly excreted, chiefly renally, and is a normal constituent of food. They further concluded that trivalent arsenic as arsenite is toxic, accumulates in the mammalian body, is largely excreted from the intestine, and is slowly oxidized to the pentavalent form in vivo.

Our primary interest was to develop an ultra-micro-coulometric technique that would be useful in evaluation of exposures by analysis of both air and excreta samples. The above mentioned conclusions would clearly modify the type of analytical technique one could use as well as the interpretation of the data.

The purpose of this study was threefold.

- (1) Examine the metabolic schemes to determine whether one can establish the relative toxicity of each valence state.
- (2) Examine the excretory patterns to determine whether one can establish the route of excretion, if indeed they are different.
- (3) Establish analytical techniques for determining each state in the presence of the other in air and excreta samples.

Distribution in Nature

The ubiquity of arsenic is a well established fact. The concentration of the element has been measured in many materials among which are sea water, marine plants, and sea food. Values of about 2 ppb for sea water have been reported by Portmann and Riley⁽²⁾.

In drinking water, levels of about 10 ppb occur⁽¹⁾. This can be compared to the limit of 50 ppb required by Illinois⁽³⁾ and many other states.

Values in sea food have been reported by many authors and levels in the 1 - 3 ppm range are not uncommon. Other foods contain varying amounts of arsenic ranging from <0.02 µg/g wet weight for coffee to about 3 ppm for table salt⁽¹⁾.

All evidence indicates that arsenic taken in by this manner is rapidly excreted and it obviously is not toxic. On the basis of thermodynamic considerations one would expect arsenite to be oxidized to arsenate in the presence of oxygen. Indeed Goldschmidt⁽⁴⁾ states that arsenates are found, in general, in zones contacting the free oxygen of the atmosphere. Portmann and Riley⁽²⁾ also indicate that the form in sea water is likely pentavalent and Armstrong and Harvey⁽⁵⁾ stated that arsenite is oxidized in sea water to arsenate. In man the arsenic content is about 20 mg, which makes this element as abundant as iodine.

Both As(V) and As(III) form acids in aqueous solution but their acidities are widely different. Arsenious acid has a pKa value of (1st proton) 9.2. At the pH of blood (7.4), about 2% would be ionized and at the pH of tissues, even less. In contrast, arsenic acid has two dissociation contents in the range of interest. The pKa values are 2.3, 6.8, and 11.6. At the pH of blood, approximately 15% would be present as $\text{H}_2\text{AsO}_4^{-1}$ and 85% as HAsO_4^{-2} . Although these ions probably do not exist in these simple forms in solution, they will be so considered in this discussion.

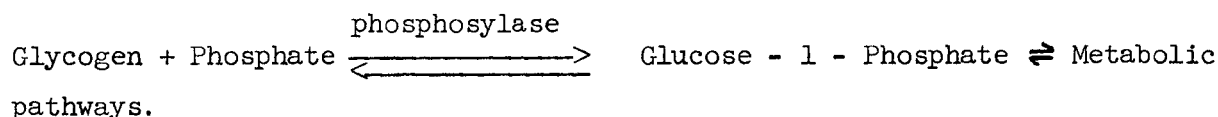
Biochemical Interaction

Some insight into the relative effects of the two valence states of arsenic may be gained from examining effects on enzyme systems.

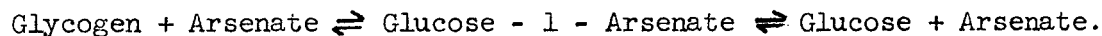
Because biochemical systems are intimately interrelated, and indeed require each other in vivo, it is not possible to extend in vitro data directly from isolated systems to the whole animal. However, one can gain general impressions.

Arsenate reacts in physiological media quite similarly to phosphate and interferes in reactions involving phosphate.

Glycogen is phosphorylated for use in the mammalian body in a reaction catalyzed by phosphoralyase:



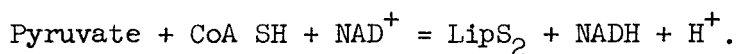
Similarly,



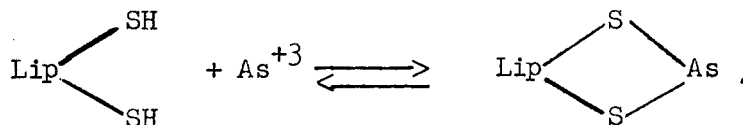
However, the arsenolysis product is quite unstable and glucose and arsenate are released.

It is characteristic of these enzyme systems that they are inhibited by their products. In the arsenolysis of sucrose in vitro inhibition occurs after a time lag presumably due to the build-up of glucose. There is no evidence that this occurs in living systems. Arsenate probably enters into many phosphorylation reactions producing easily hydrolyzed esters. There is also a possibility that cell permeability is a factor in excluding this ion from more and/or different interactions. There does not, however, seem to be any evidence of accumulation.

In contrast, the trivalent form has long been known to be highly inhibitory to enzyme systems requiring thiol groups⁽⁶⁾. With the discovery of the role of γ -lipoic acid in α -keto acid oxidations it appeared that this could well be the site of inhibition, since this would produce an accumulation of keto acids, as is observed. In the oxidation of pyruvate,



Arsenite can react with lipoic acid,



The interference of arsenite in glucose metabolism appears to be confined to this utilization of pyruvate rather than to any interference in glycolysis. A look at the citric acid oxidation cycle of aerobic glycolysis reveals other areas in which inhibition can occur.

The oxidation of α -keto glutaric acid is inhibited by arsenite, probably for the same reason that pyruvate oxidation is inhibited. In general, one could say that any system in which the thiols are needed will show some sensitivity to arsenite.

The problem in attempting to make general conclusions about arsenic metabolism centers around the fact that much of the data has been obtained using widely different concentrations of arsenite and the results in many cases are much different in vivo than in vitro.

For instance, the synthesis of cholesterol is inhibited by arsenite in vitro. However, in humans it has been observed that the blood cholesterol levels rise where one is exposed to As(III)⁽⁷⁾. This could be due to the blocking of acetate in the citric acid cycle and diversion of the acetate to cholesterol synthesis⁽⁶⁾.

The one general conclusion that can be drawn is that trivalent arsenic interferes more in vital enzyme systems than pentavalent. The next question that must be answered is whether this valence state is excreted via the renal system and, if so, can it be determined separately from the pentavalent form.

Schroeder in his summary states that As(V) is mainly excreted in the urine while As(III) is mainly excreted in the feces. There is considerable confusion on this point.

In vitro experiments have shown that pentavalent arsenic can be reduced to trivalent in varying degrees. Ginsberg and Latspeich⁽⁸⁾ have shown with dogs that there is a small but constant reduction of As(V) in both plasma and urine during infusion of As(V). Their data also indicated that there is a net tubular secretion of As(III). The site of this in vivo reduction cannot be definitely defined, but the proportion of As(III) in plasma is much lower than in urine, which would indicate that, in addition to the previously mentioned net secretion, more reduction occurs in the urinary system.

These latter authors did not analyze feces, since their studies were mainly urinary transport. The ratio of As(V) and As(III) in urine and the feces will, of course, depend upon the compound.

In 1964 S. Peoples reported on an experimental study of arsenic toxicity in cattle⁽⁹⁾. He concludes that in the cow arsenic acid is absorbed and is rapidly excreted in the urine. He indicated that there was no evidence of reduction to the trivalent state. One other important point was that the rat apparently metabolizes arsenic in a manner quite different from other mammals, and information obtained with this animal

should be evaluated in this light. On the basis of current evidence we believe that for urinalysis to be meaningful for assessment of arsenic intoxication one must determine both valence states.

Analytical Technique

The procedure employed for all of our As analysis has been a micro-coulometric titration with iodine. Briefly, iodine is generated in the solution and reacts with trivalent arsenic. The end point is determined using two platinum electrodes and amperometric detection. The titration is carried out in a bicarbonate or phosphate buffer which contains potassium iodide from which iodine is formed by oxidation. We have worked at generating currents ranging from 5 milliamperes down to 200 μ A, and currently have an instrument which will work at much lower levels. A current of 200 μ A for one minute is equivalent to about 5 μ g of As. The experimental set-up is similar to that reported previously⁽¹⁰⁾.

The current is recorded during generation and the end point determined by extrapolation to the base line. The length of the trace is measured and related to generation rate. Using this procedure, one is somewhat limited by the accuracy of the linear measurement.

In order for this titration to work the arsenic must be in the trivalent state and should be reasonably pure. A method for the separation and coulometric determination of arsenic in glass was reported by Williams and Wise in 1963⁽¹¹⁾. Briefly, the method consists of solution of the sample, reduction to As(III) with HI in 1-2N acid, and separation of As(III) on a Dowex III column. The eluant of the column can be titrated into a cooled beaker containing NaOH. Sodium bicarbonate and potassium iodide are added and iodine generated. The recovery obtained by us in each step is shown in Table 1.

As can be seen, all of the recoveries are slightly high when pure arsenic is titrated. When trivalent arsenic is carried through the HI reduction and subsequent separation, recoveries at 250 and 50 μ g are about the same and are higher at 500 μ g. When a sample is oxidized with HNO_3 or $\text{HNO}_3\text{-H}_2\text{SO}_4$ and carried through the entire procedure, iodine carries through and results are low, as shown. The losses in the reduction step are eliminated when hydrazine is used in place of HI. The hydrazine must

then be removed, and this can be accomplished by using a controlled pyrosulfate fusion. When this is used recoveries at the 50, 250, and 500 μg levels were 100.2, 100.5, and 99.1%.

This technique should be applicable to most forms of arsenic, and this was tested with sodium arsenate and p-arsanilic acid at the 50 μg (As) level. Recoveries were 100.9 and 100.0%, respectively.

The technique provides a general method for air analysis. If the samples are collected with electrostatic precipitation one would determine 10% of the TLV for As in 12 minutes. In addition, both tri- and pentavalent As could be determined in this sample by reduction of a portion and direct analysis of another portion.

For biological studies these techniques have to be modified. The amount of arsenic to be determined is considerably less. For these lower level studies a small current source of 200 μA was used initially. Under these conditions one can determine one microgram in a sample with about 1 μg deviation. Replicate analysis at 10.4 μg As(III) level yielded 95.0% recovery with a standard deviation of 2%. We currently have a Beckman Electroscan which has a generating current lower limit of 0.20 μA . This would be equivalent to 0.005 μg As, but it is not likely that we will be able to control the blank sufficiently to utilize this lower level. However, we should be able to extend our lower limit appreciably.

The determination of pentavalent arsenic in urine samples can be accomplished by many published techniques. The steps used in oxidizing the organic material will oxidize trivalent arsenic to the pentavalent state. In 1944 Crawford and Storey⁽¹²⁾ published an extraction procedure which extracts trivalent but not pentavalent arsenic, and they applied this to urine. It was necessary to dialyze the urine sample to prevent emulsion formation. This resulting extract is then oxidized and analyzed. Total arsenic can be obtained by oxidizing the urine and analyzing. In 1964 Portmann and Riley⁽²⁾ reported a procedure for cocrystallization of trivalent arsenic from sea water using thionalide. They reduced the arsenate present with ascorbic acid before treatment. It might be possible to utilize this technique for direct precipitation of trivalent arsenic from urine.

There appears to be sufficient evidence to indicate that for urinalysis results to be useful one must determine both the tri- and pentavalent forms in the sample. The bulk of our future work will be in this direction.

Summary

The distribution of arsenic in nature has been discussed. It appears that the predominant natural form of the element is the pentavalent state. An examination of selected enzymes systems indicates that the trivalent form is more metabolically active. The interconversion of inorganic arsenite and arsenate in vivo is somewhat obscure, but it would seem that an analysis to determine both valence states would be useful both for obtaining information on excretory patterns and for assessing exposures.

Finally an analytical technique has been described as well as two separation techniques which should be useful in studying this problem.

TABLE I
PERCENT RECOVERY OF ARSENIC IN
EACH STEP OF DETERMINATION

μg Arsenic	Titration	Reduction and Ion-exchange	Total Procedure
50	100.2 ± 1.5	94.1 ± 1.3	76.8 ± 2.6
250	100.2 ± 1.8	93.1 ± 2.0	93.1 ± 0.9
500	100.3 ± 0.7	96.8 ± 0.6	95.9 ± 0.6

References

1. Schroeder, H., and Balassa, J., J. Chron. Dis. 19, pps. 85-106, 1966.
2. Portmann, J., and Riley, J., Anal. Chim. Acta 31, pps. 509-19, 1964.
3. Illinois Sanitary Water Board, Rules and Regulations, SWB-11.
4. Goldschmidt, V.M., Geochemistry, Clarendon Press, Oxford, pps. 468-478, 1954.
5. Armstrong, F., and Harvey, J., Marine Biol. Assoc. U.K. 29, p. 147, 1950.
6. Webb, J., Enzymes and Metabolic Inhibitors, Academic Press, pps. 595-895, 1966.
7. Auken, G., Acta Pharmacol. Toxicol. 1, p. 369, 1945.
8. Ginsberg, J., and Latspeich, W., Am. J. of Physiol. 205, pps. 709-714, 1963.
9. Peoples, S., Ann. N. Y. Acad. Sci. 111, pps. 644-649, 1964.
10. Duffy, T., and Pelton, P., Amer. Ind. Hyg. Assoc. J. 26, pps. 544-548, 1965.
11. Williams, J., and Wise, W., Anal. Chem. 36, pps. 1863-1864, 1964.
12. Crawford, T., and Storey, I., Biochem. J. 38, p. 195, 1944.

BIOCHEMICAL ASPECTS OF MERCURY POISONING*

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The effects of mercury compounds on experimental animals and humans are well known. But the mechanisms leading to these effects are only partially understood and are the subject of continuing and intensive research in many laboratories.

A hazard from mercury poisoning still exists. It is compounded by the fact that the metal is used in a variety of chemical forms. These in turn differ markedly in the nature and extent of their toxic effect. Some organic compounds of mercury (i. e., compounds in which at least one valency of mercury is to a carbon atom of the organic moiety) produce specific kidney damage or damage to the nervous system, whereas others have very low toxicities. Inorganic compounds of mercury damage a variety of organs.

In order to discover the underlying biochemical lesions, it is important to identify the specific chemical form of mercury which is toxic. For example, the various inorganic forms of mercury appear to act via the release of or by conversion to the divalent ion, Hg^{++} . Recent studies have shown that the elemental vapor is rapidly converted to Hg^{++} in blood, and it has been known for a long time that mercurous mercury, Hg_2^+ , is unstable in the presence of proteins and releases Hg^{++} . The question arises whether the organic compounds of mercury also act in this fashion.

Organic mercurials differ greatly in their ability to cause death by kidney damage, and at least one compound, p-Mercuribenzoate, exhibits very different renal toxicity in various animal species. By labeling the mercurials with the ^{203}Hg isotope, it has been possible to demonstrate that these compounds break down in animal tissues to release inorganic mercury. The rate of release of inorganic mercury is related to the renal injury. Compounds not producing kidney damage release inorganic mercury slowly or do not accumulate in kidney tissues.

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BIOCHEMICAL ASPECTS OF MERCURY POISONING

The biochemistry of mercury poisoning has been studied from two points of view, the effects of mercury on tissue enzymes and the biotransformation of mercury, i.e., the changes in the chemical state of mercury which occur when it is exposed to living systems.

The importance of distribution and biotransformation studies

Webb¹ has recently reviewed the inhibitory actions of mercury on biochemical processes. Unfortunately no clear picture emerges. Mercury combines preferentially with thiol groups and is capable of inhibiting any enzyme containing such groups. At higher concentrations, mercury is a protein precipitant. Consequently a wide variety of enzymes are inhibited when mercury is added to in vitro preparations. It seems likely that, when mercury is administered in vivo, tissue enzymes will be affected wherever the metal is accumulated.

Berlin and Ullberg² have made effective use of the radioautographic techniques to study distribution of mercury within body tissues. Recent evidence indicates mercury entering the brain may become localized in special areas. Even at the cellular level marked differences in localization have been seen, with notable accumulation in the nuclei of some nerve cells³.

Norseth⁴ has applied modern methods of centrifugal separation of sub-cellular particulates in studies of the distribution of mercury compound in liver cells. He observed an accumulation in the lysosomes - the so-called "suicide bags" of the cell - disruption of which releases hydrolytic enzymes. He has suggested that damage to the lysosomes by mercury may be the primary lesion responsible for cell destruction. Information obtained by the techniques used by Berlin and Norseth may serve as useful guidelines in our efforts to

pinpoint the enzymes or structural macromolecules primarily affected in mercury poisoning.

The biotransformation of mercury in the body is an equally important factor in determining its effects on biochemical processes. Mercury exists in three oxidation states - as the elemental metal (vapor) and the mercurous and mercuric salt - as well as in a variety of organo-metallic compounds. Studies in the test-tube on the action of a certain organic mercury compound on enzymes may be completely misleading if the same compound is rapidly transformed in the body to inorganic mercury. This very point has led to considerable controversy over the interpretation of experimental work on the action of organo-mercurials on kidney function^{5, 6}. Until we possess more knowledge on the local distribution of mercury with tissues and cells, and on the chemical fate of mercury within the organism, it is likely that more basic biochemical studies will be irrelevant. The remainder of this article will be devoted to a review of our present knowledge on the biotransformation of mercury and its compounds.

The Biotransformation of Mercury and its Compounds

Studies carried out in the last fifteen or so years on the metabolism of foreign compounds have given an important insight into many problems associated with the use of drugs⁷. Marked species differences in the response to a drug are frequently due to different rates of detoxication. When two animals are given the same dose per kg body weight, different plasma levels result frequently because one animal metabolizes the compound to an inert product more rapidly than the other. Dangerous side effects of some drugs are due to the formation to toxic metabolites. Tolerance to a certain compound, e.g., a barbiturate, has been found to be due to the stimulation of its own metabolism by repeated administration. Marked sensitivity in the newborn, e.g., as with chloramphenicol, is in part due to the low activity of the drug-metabolizing

enzymes in the perinatal period. Genetically controlled differences in response, e.g., to succinylcholine, arises from a deficiency in plasma esterase enzymes in a small fraction of the population. Some synergistic and antagonistic effects between two or more compounds have been found to arise when one compound inhibits or stimulates the metabolism of the other. Some compounds are transformed to more active products, others to less active. This area of study, generally referred to as biochemical pharmacology, is now regarded as an essential part of the evaluation of any new drug.

Studies on the biotransformation of heavy metals have been few compared to studies on organic compounds. Perhaps investigators did not expect such potent enzyme inhibitors to undergo chemical changes catalyzed by enzymes.

Tetraethyl lead and tetraethyl tin were amongst the first metallic compounds to be studied⁸. In the last decade work on mercury and its compounds has been started and has already revealed some unexpected information on the behavior of the metal in the body and in ecological systems.

Biotransformation of Inorganic Mercury

Studies on the biotransformation of inorganic mercury, i.e., in the elemental form, mercurous or mercuric salts, has been reviewed⁹ and will be briefly recapitulated here. In vitro experiments revealed that samples of whole blood are capable of absorbing mercury vapor and rapidly oxidizing the metal to the toxic mercuric ion. It was estimated that the in vitro rate of oxidation in the body of all the absorbed vapor. This would explain the observation of Rothstein and Hayes¹⁰ that the general pattern of distribution of mercury in the body is the same after exposure to mercury as after the injection of HgCl_2 . In both cases the uptake in the brain is small, accounting for less than 2% of the injected dose. However, the brain levels from mercury vapor exposure may be ten times higher than after HgCl_2 . Recent work by Magos¹¹ indicates that

the increased brain uptake may be due to small amounts of unchanged vapor dissolved in blood. He points out that circulation time from lung to brain is short and the vapor need exist only transiently in the blood stream to account for the brain uptake. In vitro experiments demonstrated that dissolved elemental mercury is detectable in blood up to 15 minutes after exposure to the vapor.

When HgCl_2 is injected into rats, somewhat less than 20% of the excreted mercury is lost from the animal in the volatile form¹². The mechanism is unknown and the chemical form of the volatile mercury has not yet been identified. It could be elemental vapor or a volatile alkyl mercurial.

Micro-organisms are also capable of volatilizing mercury from solutions of mercuric chloride¹³. Some of these microorganisms are present in the usual laboratory environment and even in the water supply. The contamination of urine samples by such organisms caused a rapid loss of mercury from the samples. In a recent review¹⁴ Goldwater has noted that urinary mercury analyses are erratic and correlate poorly with exposure.

Studies associated with the famous Minamata Bay disaster¹⁵ indicated that inorganic mercury could be converted to an alkyl mercurial in non-mammalian systems. Effluent containing mercuric salts from a factory was discharged into the bay. People who had eaten fish from the affected area developed symptoms typical of poisoning by alkylmercurial compounds. Significant quantities of mercury were detected in the fish, and it was suggested by Kurland¹⁶ that inorganic mercury had been converted to the alkylmercurial compound. Subsequently, Uchida, Hirakawa, and Inoue showed that the mercury compound was methyl (methyl thio) mercury.¹⁷

The Biotransformation of Organo-Mercurial Compounds

The biotransformation of organo-mercurial compounds has been studied only in recent years. Investigations have been concentrated on two distinct

areas - first, the degradation of organo-mercurials in mammalian tissues, and second, the fate of organo-mercurial fungicides used as seed dressing and released into the soil and concentrated in various food chains.

Recent studies by Westöo¹⁸ using gas and thin-layer chromatography have indicated that the release of phenyl mercury into rivers and streams in Sweden results in the appearance of methyl mercury compounds in the fish. Jensen and Jernelov¹⁹ have presented evidence that inorganic mercury is converted to methyl mercury by micro-organisms present in mud and suggest that phenyl mercury compounds first release inorganic mercury which is converted into methyl mercury. Granted that inorganic mercury is transformed in nature to methyl mercury, the question arises as to how many organo-mercurials are capable of being split to release inorganic mercury. From an ecological standpoint, this is now an important question in view of the potential conversion to methyl mercury which is a cumulative and irreversible poison²⁰.

The release of inorganic mercury from organo-mercurial compounds is also of interest in relation to the toxic effects of these compounds on animals. However, only in recent years have the techniques been developed which make this type of study possible. Miller, Klavano, and Csonka²¹ and Gage and Swan²² were the first people to publish such studies. The analytical technique is based upon the extraction of the organic mercury into benzene from an acidified digest of the tissue. Inorganic mercury was not extracted by the benzene. These authors concluded that the non-extractable mercury represented inorganic mercury split off from the mercurial. As pointed out by Webb²³ strictly speaking this conclusion is not necessarily valid. It is possible that the organic moiety might undergo biotransformation making it insoluble or partly soluble in benzene. However, with this reservation, the results of the studies are of some interest. Phenyl mercury is rapidly broken down in the body, resulting in an accumulation of inorganic mercury in the kidneys. On the other hand, the alkylmercurials

(methyl mercury and ethyl mercury) are much more stable, and the concentrations of mercury in brain and blood are higher than those found after administration of phenyl mercury.

The differences in rates of metabolism very likely play an important role in the toxic effects. The alkyl mercurials give no symptoms until after a latency period of 1 to 2 weeks. Toxic symptoms indicate damage to the central nervous system²⁴. It is an intriguing possibility that the latency period may be related to the slow rate of cleavage of these compounds. The intact mercurial is known to penetrate the brain^{22, 23}. The inorganic mercury released from the compound may be responsible for the toxic symptoms. In view of the slow rate of breakdown it may well take 1 to 2 weeks for sufficient inorganic mercury to accumulate. However, there have been no published accounts to date of attempts to discover if the intact organo-mercurial or the released inorganic mercury is responsible for the effects.

The aryl and alkyl mercurial are used mainly as fungicides, and human exposure is accidental or unintentional. However, another class of organo-mercurials, the diuretics, have been administered to humans for many years. These mercurials and some of their derivatives are now used in radiology for diagnostic scintillation scanning of various organs²⁵.

The release of inorganic mercury from mercurial diuretics may be of toxicological or pharmacological significance for three reasons: first, it may be responsible for the diuretic effect, second, it may be the cause of the renal damage observed at high doses, and thirdly, since inorganic mercury remains in the kidney for long periods, the rate of cleavage of the compound should be important in determining cumulative effects resulting from repeated doses.

For these reasons many attempts have been made to develop analytical techniques capable of distinguishing between the intact diuretic molecule and

inorganic mercury in animal tissues and in urine. Since the organo-mercurial diuretics are water soluble, the benzene extraction method is not applicable. Attempts with the polarograph have been unsuccessful due to the low sensitivity of the method⁶.

Weiner, Levy, and Mudge²⁶ developed an in vitro method for testing for lability. This involved placing the mercurial into a solution of cysteine or other mercury-complexing agent and measuring the rate of breakdown. They were able to predict the diuretic potency of some 34 organo-mercurial compounds, namely that a compound would be diuretic if it released mercury in the in vitro test and if the compound were rapidly excreted in the urine. The effectiveness of these two criteria to predict the diuretic potency of so many organo-mercurial compounds is impressive, and suggests that the in vitro test correctly predicts the in vivo lability of the compound. Such a test would be very useful in assessing toxic hazard from new compounds of mercury. However, recent evidence indicates that the test is not completely reliable. For example, the in vitro test predicts that phenyl mercury should be stable, whereas it has been shown to breakdown rapidly in rat tissues²¹.

The introduction of the isotope-exchange technique²⁷ has made it possible to distinguish between inorganic mercury and the water-soluble organo-mercurials (e.g., the diuretics). This method is based on the finding that radioactive mercury attached covalently to a carbon atom in an organo-mercurial compound will exchange very slowly with mercury in the vapor phase. In contrast, labelled inorganic mercury exchanges rapidly. Thus after administration (to an animal) of organo-mercurial compound, labelled with the isotope ²⁰³Hg, the amount of inorganic mercury in any tissue is equal to the quantity of exchangeable radioactivity.

This technique is now in use to investigate two questions related to the biotransformation of mercurials: (1) is the inorganic mercury released from

the compounds responsible for the toxic effects and secondly, (2) does the inorganic mercury released from the compound remain in the tissues for long periods of time?

Several organo-mercurial compounds available commercially in the radioactive form were tested in rats and mice for acute toxicity. The approximate seven-day L.D.₅₀ values are given in Table I for two of these compounds.

TABLE I.

Approximate subcutaneous seven-day L.D.₅₀ values in male albino rats and mice.

<u>Mercury Compound</u>	<u>Rats (mg Hg/kg)</u>	<u>Mice</u>
HgCl ₂	3	6
p-chloromercuribenzoate (PMB)	3	50
Chlormerodrin	20	50

It will be seen that L.D.₅₀ of p-chloromercuribenzoate (PMB) was about 8 times lower than that of chlormerodrin and about 10 to 20 times lower than the L.D.₅₀ of PMB in mice. Our evidence to date suggests that both effects are due to differences in rates of release of Hg⁺⁺. This evidence may be summarized as follows: The animals died exhibiting typical symptoms of acute renal failure. The blood urea nitrogen rose to very high levels, urea and endogenous creatinine clearance values fell, and complete anuria developed on the day preceding death. Death usually occurred between 50 and 110 hr after injection. Autopsy examination indicates severe damage to the kidneys with the cortical tissue being completely ischemic. Damage to the kidneys correlated with the degree of conversion of the organo-mercurial. The data of Table II indicate that the levels of inorganic mercury in kidneys of rats, 24 hr after an L.D.₅₀ dose,

were approximately the same for the three compounds tested. PMB breaks down to inorganic mercury much more rapidly than chlormerodrin²⁸. Consequently the

TABLE II.

The concentrations of inorganic mercury in kidney tissue of rats 24 hours after an L.D.₅₀ dose of HgCl₂, p-chloromercuribenzoate and chlormerodrin.

Mercury Compound	Inorganic Mercury in Kidney µg. Hg/g wet wt.
HgCl ₂	30
p-chloromercuribenzoate	40
Chlormerodrin	25

much lower L.D.₅₀ dose (3 mg Hg/kg) gave the same kidney concentration as the higher L.D.₅₀ dose (20 mg/kg) of chlormerodrin. These results suggest that the kidney damage was produced by the inorganic mercury split off from these organo-mercurials.

TABLE III.

Species differences in metabolism of p-mercuribenzoate

	<u>Rat</u>	<u>Mouse</u>
L.D. ₅₀ mg Hg/kg	3.4	61
Total kidney Hg at 24 hr, % Inj. dose	10	11
% metabolized to Hg ⁺⁺ in 4 hr	100	<5

Differences in rates of metabolism may also account for the fact that the L.D.₅₀ for PMB in mice is larger than in rats. As indicated by the data of Table III, for a given dose of PMB (4 mg Hg/kg), the total mercury in the kidney is the same in both species, but the rate of metabolism differs greatly. In the rat after 24 hr all the mercury was present as Hg⁺⁺ whereas in the mouse less than 5% was in the inorganic form.

The importance of inorganic mercury concentrations in renal tissue in determining the toxic effects raises the second question as to how long mercury remains in the kidney. Previous studies with chlormerodrin²⁷ indicate that the organo-mercurial is rapidly accumulated and rapidly excreted. Forty-eight hours after injection, the intact organo-mercurial is not detectable in kidney tissue. In contrast, inorganic mercury split off from the mercurial slowly accumulated in the kidney. It reached a maximum level about 24 hr after injection of chlormerodrin, subsequently the level fell very slowly. After administration of PMB, levels of inorganic mercury rise quickly to a steady level. These results lend experimental support to the concern frequently expressed²⁹ that repeated injection of organo-mercurial diuretics may lead to cumulative toxicity.

It is a sobering thought that man has been aware of the toxicity of mercury for at least two thousand years and yet knows very little of its biochemical mechanisms of action. This review of our present knowledge suggests that studies should be directed towards following the biotransformation and microdistribution of mercury and its compounds in cells and tissues. Until our knowledge is more complete in these two areas, it will probably be very difficult to pinpoint the specific biochemical lesions underlying mercury poisoning.

1. Webb, J.L.: Enzyme and Metabolic Inhibitors, Vol. II, Aced. Press, New York (1966) p 729.
2. Berlin, M.;Ullberg, S.: Accumulation and retention of mercury in the mouse, I. An autoradiographic study after a single intravenous injection of mercuric chloride. Arch. Envir. Health 6, (1963).
3. Berlin, M.: Recent progress in mercury toxicology research and its consequences for current occupational mercury problems. 25th International Congress of Occupational Medicine, Vienna 1966 .
4. Norseth, T.: The intracellular distribution of mercury in rat liver after a single injection of mercuric chloride. Biochem. Pharmacol. 17 (1968).
5. Kessler, R.H.; Lozano, R.; and Pitts, R.F.: Studies on structure diuretic activity relationships of organic compounds of mercury. J. Clin. Invest. 36, 656 (1957).
6. Mudge, G.H.; and Weiner, I.M.: The mechanism of action of mercurial and xanthine diuretics. Annals N.Y. Acad. Sci. 71, 344 (1958).
7. Brodie, B.B.: Distribution and fate of drugs; therapeutic implications in "Absorption and distribution of drugs", Ed. by Binns., Williams and Wilkins, Baltimore (1964).
8. Cremer, J.E.: Tetraethyl lead toxicity in rats. Nature 195, 607 (1962).
9. Clarkson, T.: Mercury: Toxicological Aspects. Ann. Occup. Hyg. 8, 73 (1965).
10. Rothstein, A.; and Hayes, A.: The turnover of mercury in rats exposed repeatedly to inhalation of vapor. Health Physics 10, 1099 (1964).
11. Magos, L.: Transport of elemental mercury by blood. Arch. Pharmak. exp. Path. 259, 183 (1968).
12. Clarkson, T.W.; and Rothstein, A.: The excretion of volatile mercury by rats injected with mercuric salts. Health Physics 10, 1115 (1964).
13. Magos, L.; Tuffery, A.A.; and Clarkson, T.W.: Volatilization of mercury by bacteria. Brit. J. Ind. Med. 21, 294 (1964).

14. Goldwater, L.J.: Occupational Exposure to Mercury. 15th International Congress of Occupation Medicine 3, 117 (1966).
15. Bidstrup, P.L.: Toxicity of mercury and its compounds. Elsevier Pub. Co., New York (1964).
16. Kurland, L.T.; Fard, S.N.; and Siedler, H.: Minamata disease. Wld. Neurol. 1, 370 (1960).
17. Uchida, M.; Hirakawa, K.; and Inoue, T.: Kumamoto. Med. J. 14, 181 (1961).
18. Westoo, G.: Determination of methylmercury compounds in foodstuffs. Acta Chem. Scand. 21, 1790 (1967).
19. Jensen, S.; and Jernelov, A.: Biosynthesis of methyl mercury. Nordforsk, Biocid-Information No. 10, 5 (1967).
20. Hunter, D.; and Russell, D.S.: Focal cerebral and cerebellar atrophy in a human subject due to organic mercury compounds. J. Neurol. Neurosurg. Psychiat. 17, 235 (1954).
21. Miller, V.L.; Klavano, P.A.; and Csonka, E.: Absorption, distribution and excretion of phenyl mercury acetate. Toxic. Appl. Pharmacol. 2, 344 (1960).
22. Gage, J.C.; and Swan, A.A.B.: The toxicity of alkyl and aryl mercury salts. Biochem. Pharmacol. 8, 77 (1961).
23. Webb, J.L.: Enzyme and Metabolic Inhibitors. Vol. II. Aced. Press, New York (1966) p 961.
24. Swensson, A.: Toxicology of different organic mercurials used as fungicides. 15th International Congress of Occupational Medicine 3, 129(1966).
25. Blau, N.; and Bender, M.A.: Radiomercury (Hg^{203}) labelled neohydrin: A new agent for brain tumor localization. J. Nuclear Med. 3, 83 (1962).
26. Weiner, Levy, and Mudge: Studies on mercurial diuresis; renal excretion, acid stability and structure-activity relationships of organic mercurials. J. Pharmacol. exp. ther. 138, 96 (1962).

27. Clarkson, T.W.; Rothstein, A. and Sutherland, R.: The mechanism of action of mercurial diuretics in rats; the metabolism of ^{203}Hg -labelled chlormerodrin. *Brit. J. Pharmacol.* 24, 1 (1965).
28. Clarkson, T.W.; and Greenwood, M.: The mechanism of action of mercurial diuretics in rats; the renal metabolism of p-chloromercuribenzoate and its effects on urinary excretion. *Brit. J. Pharmacol.* 26, 50 (1966).
29. Freeman, R.B.; Maher, J.F.; Schreiner, G.E.; and Mostofi, F.K.: Renal tubular necrosis due to nephrotoxicity of organo-mercurial diuretics. *Ann. Intern. Med.* 57, 34-43 (1962).

TECHNO-LEGAL FACTORS TO BE CONSIDERED
IN DEVELOPING THE RADIATION DOSE RECORD*†

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"Recorda sunt vestigia vetustatis et veritatis."

Legal considerations appear to make it necessary to program for the long-term retention of certain parts of an individual's radiation exposure record, even though only a few legal precedents have been set in this particular field.

Some rules relating to record-keeping techniques as associated with personnel exposure have been established as a matter of law. Other rules, based on technical considerations as well as legal considerations, have been recommended by various agencies and advisory groups, but these recommendations usually are stated in broad terms so as to take into consideration the lack of uniformity in the laws of the various States.

Legal factors to be considered involve principles of law that are embodied generally in (i) the workmen's compensation acts, (ii) the wrongful-death statutes, (iii) a consideration for property rights in certain types of personnel records, and (iv) the law of evidence. Complications arise in that in each of the above enumerated categories the rules may vary from state to state, or even between the judicial systems within a particular state.

Pending legislative action or the establishment of precedent by the courts to the contrary, or both, it seems likely that the ideal record-keeping system will stress (a) retention of those data used in the evaluation of radiation dose, where key factors that affect the dose number derived are subject to individual interpretation, (b) identification and qualifications of those individuals responsible for dose evaluation and the recording of the data associated with it, and (c) a time of retention based on the applicable statutes of limitation, which, for practical purposes, appears to be commensurate with life expectancy rather than with tenure of employment or the duration of the exposure.

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† Opinions expressed are personal and not necessarily those of the Atomic Energy Commission or the Union Carbide Corporation.
Recorda sunt vestigia vetustatis et veritatis: Records are the vestiges of antiquity and truth.

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IN DEVELOPING THE RADIATION DOSE RECORD*/

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"Recorda sunt vestigia vetustatis et veritatis."

Records are the vestiges of antiquity and truth. This legal principle had become entrenched in the law of evidence by the time the early settlers were landing on Plymouth Rock. But, as time wore on the rules which governed the admissibility of testimony based on the written word developed to a complex point. The expert witness who is called upon to testify regarding causal relationships in a radiation injury case usually is faced with using records that have been generated by persons other than himself. Or, the radiation record itself may be placed before the court. In either case, the record becomes a subject for appraisal and the issues involve not only questions of admissibility but technical questions about which scientists themselves are in disagreement.

Techno-Legal Problems⁽¹⁾⁽²⁾

All persons, not just those who are occupationally involved, are subjected to radiation exposure. In fact, occupational exposure as such is but a small percentage of the radiation dose received by the population at large. Consequently, where an individual is suffering from what may appear to be a radiation-induced disease, establishing that occupational exposure was the cause may be difficult, if not impossible, to determine. Courts are not inclined to give a judgment against a defendant in tort unless there is a preponderance of evidence pointing

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to the fact that there is causal relation between the injury and some act of negligence on the part of the defendant. However, in workmen's compensation cases, the element of doubt often is resolved in favor of the injured party.

Medical knowledge today is such as to tell us something about radiation damage resulting from acute massive doses of radiation, but a great deal of uncertainty arises regarding the effect of small doses of radiation spread over a long period of time. An even more involved problem involves the unresolved conflict between the linear and threshold theories of radiation effects, the latent injury effect, and radiation-induced diseases (such as leukemia, skin cancer, and aplastic anemia) which may be produced or aggravated by other causes.

Some bodily malfunctions which radiation may induce, such as epilation and sterility, do not necessarily constitute a compensable disability under the typical workmen's compensation law. How valuable is a person's hair in terms of dollars? Can sterility be evaluated in terms of dollars? Or does sterility occur in fact, say, for example, at the maximum permissible dose limit? In workmen's compensation, if a radiation-induced injury is not covered by the act the injured party cannot recover under workmen's compensation, but he may be able to recover in tort.

Maximum permissible dose limits have been developed on the basis of an acceptable risk based on the premise that the assumed risk is deemed to be acceptable to the individual and to society in view of the benefits derived. It is said that the maximum permissible exposures for workers are on the conservative side so long as only a few employees average these maximum dose limits over an extended period of time. Thus, the risk taken by a particular individual when subjected to the maximum permissible dose limits is considered to be insignificant, with injury highly improbable. However, it is expected that some radiation-induced injuries will occur at the maximum permissible dose limits, with the result that the injured party may seek to recover compensation for the injury.

Radiation dose-measurement techniques have certain limitations. The accuracy of personnel monitoring devices depends upon considerations of geometry, the reliability of the radiation detector itself, and the overall efficiency of the total dosimetry system. Because of biological differences between individuals, the establishment of radiation dose resulting from ingestion, inhalation, or injection of radioactive materials is far from precise and the techniques for estimating such doses complicated.

The above technical problems are complex ones which the scientist and law-maker may in time resolve. However, unless there is a preemption by Federal law or until agreements between the states are reached, there will continue to be an appreciable lack of uniformity in procedural and substantive law as it exists between states and between judicial systems within a particular state. (The judicial system within a particular state may include the various state courts, the Federal courts, and certain quasi-judicial bodies such as an "industrial commission.")

Legal Considerations

It is a well-settled principle of law that the substantive law of the state in which a cause of action arises will be applied unless the action involves a field of law preempted by Federal law. Thus, even a Federal court will apply the substantive law of the state in which the cause of action arises. But, the substantive law of each state can reach different results.

In devising a radiation-exposure record system, it would appear that one must concern himself primarily with state law, where the principal areas of interest include (i) the interpretation placed on the various workmen's compensation acts adopted by the different states, (ii) the wrongful death statutes in certain states which give a cause of action to a third party, (iii) a consideration for property rights in certain types of records maintained by one party on behalf of

a second party, and (iv) the law of evidence as it is interpreted by the states.

Workmen's Compensation. Workmen's compensation may be considered as a form of insurance wherein the worker usually gives up his common law rights to a remedy in tort for assurances that he will receive some compensation in the event of an occupational injury. The courts uniformly hold that statutory compensation is the sole remedy of the employee against his employer if the injury to the employee is found to be covered by a workmen's compensation act with the result that recovery in tort is barred.⁽³⁾ However, where the injury is found not to come under a workmen's compensation act, the employee may seek recovery under tort procedures and a jury can award damages without reference to any statutory compensation which may be provided for in the applicable workmen's compensation act.

A 1964 survey⁽⁴⁾ showed the average maximum recovery for death under workmen's compensation in 36 states to be about \$15,000. Among these 36 states the maximum varied from \$10,000 to as high as \$25,000 in Hawaii and Montana. In nine states no maximum limit was specified as such, but the maximum wage subject to compensation was set at a value which controlled the periodic payment to levels comparable with the above figures. In the remaining five states and the District of Columbia there was no limit as to the period of time which periodic payments would be made to the widow of a worker so long as the widow remained unmarried.

Since compensation is limited, usually less than an estimate which a jury might place upon damages under tort proceedings, the compromise by which a workman accepts limited compensation in return for the extended liability of the employer has prompted provisions in most of the acts to the effect that the courts are instructed to interpret liberally in favor of the injured worker. The lack of technical certainty as to causation may be a factor that will favor the worker, and such is exemplified in a New York decision handed down by the Supreme Court of that state in 1965.⁽⁵⁾ Here, the widow of a myoblastic leukemia victim won a

workmen's compensation award when expert testimony was to the effect that "there is really no 'threshold' or 'safe' dosage of radiation because at the present stage of scientific knowledge it cannot be ascertained exactly what effects radiation has on the human body." The evidence in this case showed that the employee's potential for radiation exposure existed for a period of about one year, that he was not a radiation worker himself but had visited on occasion a building where radioactive materials were used, and that his exposure, if any, should have been well below maximum permissible levels. It was reasoned by the court that radiation could have caused the onset of leukemia in the worker's case and, in the absence of expert testimony to the contrary, doubt as to causation was resolved in favor of the worker. But, a somewhat opposite position seems to have been taken in the State of Texas.⁽⁶⁾ In a workmen's compensation case decided in 1967 that involved a metastatic carcinoma of the cervical lymph node, evidence was presented to the effect that the injured party may have received more than a casual exposure to radiation. There was medical testimony to the effect that "exposure to radiation can cause cancer and that a person working around radiation does have a higher than normal risk of developing malignant changes in his tissues." But the court noted that "no medical expert testified that radiation was the probable cause of the plaintiff's cancer" and held that "may have," "could have," or "possibly" was not sufficient standing alone to establish causation, and denied compensation. Although these two cases are of interest as to the degree of proof required in establishing causation, and even though as a general practice the worker is to get the benefit of the doubt, it is indicated that the radiation exposure record should be as complete as possible and that the record which shows that no exposure has in fact occurred is of extreme value where nonthreshold cases are involved.

The workmen's compensation acts include provisions which limit the time for filing a claim (usually to about one year after an injury). However, opinion varies as to the effect of the time of limitation where an occupational disease

occurs and the disease is associated with latent effects or there is evidence of low-level chronic exposures.

Wrongful Death Statutes. The wrongful death statutes provide a cause of action to next of kin under tort procedure, and usually limit the time of the commencement of the lawsuit to about one year after the death of the party in question.⁽³⁾ Here, a third party, not necessarily connected with the employer's operation, is provided with a cause of action against the employer if the employee himself would have had a cause of action and his injury was not covered by workmen's compensation. The problem of latent effects applies to these cases as it does to the workmen's compensation cases except that time begins to run following the death of the individual rather than after the injury is manifested.

Property Rights in Records. Does an injured party have the right to examine records maintained on his behalf by another? That a patient has a property right in medical records accrued for him by his physician seems to be established.⁽⁷⁾ Statutory provisions in the State of Wisconsin give a patient and his lawyers the right to inspect and copy medical records accrued for him by hospitals and physicians. Of course, administrative procedures formulated by the Atomic Energy Commission provide that an employee be given radiation dose records concerning himself upon request, and it would appear that responsibility for maintaining the exposure record rests with the employer, as set out in the Atomic Energy Commission Manual Chapters.⁽⁸⁾ Although one might conclude that an employee has a legal right to a copy of his radiation exposure record, there may be a question as to what matters the record should include.

The Law of Evidence. The law of evidence applies equally to workmen's compensation and tort actions.⁽⁹⁾ Perhaps a brief statement regarding the so-called "business records rule" will suffice. The business records rule admits records (i) if the records are made in the regular course of business, (ii) if, in the regular course of that business, the record is made by an employee or representative

who has personal knowledge of the matters recorded, and (iii) if the record was made at or near the time of the act or reasonably soon thereafter. It overcomes the "hearsay" rule which requires that only firsthand information or observations can be admitted into evidence. It is an exception to the hearsay rule and has been adopted by most of the states and federal courts in one form or another.⁽¹⁰⁾⁽¹¹⁾⁽¹²⁾

Actually, the law of evidence simply seeks to produce the best evidence, which has come to mean that testimony made by eye witnesses takes precedence over testimony based on second-hand information. In the case of a record it is the original record that is considered to be best evidence, and copies of originals may be rejected by the courts if the original record itself is available.

Suggested Guidelines

When viewed in a legal perspective the requirements for an adequate radiation exposure record system are not uniquely different from other types of record systems. Of course, the information recorded needs to tell the story, and the latent injury effect suggests long-term retention. The business records rule appears to constitute a reasonable formula for developing the radiation exposure record system. However, certain records must be kept, either as an administrative requirement or as a matter of law, pursuant to the provisions of the AEC Manual Chapters, state, and local regulations as they apply.

It has been a common practice in the case of personnel monitoring systems to retain in the record the piece of film that is worn for monitoring purposes and from which radiation dose is estimated. Whether the record of the estimated dose, as compared with the actual piece of film, should constitute an adequate record with the implication that films need not be produced in court continues to be debated, even though records generated from film data have been offered and admitted in evidence.⁽¹³⁾ It has been argued that the business records rule itself gives sufficient grounds for admitting in evidence the record generated from film data.⁽¹⁴⁾ In fact, it would appear on the surface that the admission of such evidence has gone unchallenged to a point where precedent has been set and the question of

saving films for legal reasons should have been laid to rest. Perhaps it is significant that it does not appear that there has ever been a serious objection on the part of counsel regarding the question of producing films, with the result that the courts have not been asked to rule upon the question. As a practical matter, it has been my own observation that under the usual conditions that films are stored, films deteriorate to a point where their reliability after a period of time is open to question. In fact, it is entirely reasonable to conclude that one should have a greater degree of confidence in the accuracy of the reading made at the time that such films were processed. Even so, until a firm precedent has been set, present administrative requirements have been changed, or aged stored films have been shown to have lesser technical significance it appears that films should be kept for legal reasons.

Internal dose determinations are complex, requiring in many cases long-term sampling, precise analytical techniques, and mathematical treatment. The question has been asked if raw data generated by the analytical chemist in a bioassay program should be retained to allow for a reconstruction of the dose estimated. Dose estimation in these cases usually involves an element of professional judgment, which may include consideration of certain particular physical characteristics of the exposed individual (e.g., body size, elimination rates, etc.), and the availability of raw data from which dose estimates can be reevaluated enhances the credibility of testimony involving such data. It is recommended that raw data be recorded.

The question of offering into evidence punch card data generated by automatic processing techniques appears to be resolved generally as the punched holes in the cards would be acceptable as regular entries when accompanied by testimony explaining their meaning under the code used in the particular system employed. (9)

A critical problem posed for the record-keeping system has to do with the length of time that the record should be maintained. A rational approach to the latent injury problem in workmen's compensation is a proposal that the time limitation on filing claims should not begin to run (1) until the date on which

the employee has knowledge of the disability and its relationship to his job and (ii) until after disablement.⁽¹⁵⁾ However, only a few of the fifty states have adopted the proposal, and many of the states are yet to include radiation-induced diseases as compensable. In any case, the problem posed by latent effects appears to make it necessary to retain the exposure record for a particular individual until some period of time after the death of the individual is confirmed. USA Standard N2.2 recommends that exposure records be maintained until the year when the individual would have reached the age of 75 years or until ten years after the known death of the individual.⁽¹⁶⁾ It appears that USA Standard N2.2 seeks to strike a balance between technical requirements, administrative requirements, and legal considerations representing a practical approach to the problem.

The emphasis placed upon systems that generate a permanent record has led to a somewhat erroneous conclusion that thermoluminescent dosimetry cannot replace film badge dosimetry because of the fact that the radiation dose indicated by a thermoluminescent dosimeter is erased when the dosimeter is processed. The best evidence rule will be applied to the record generated by the system under consideration by the court, and there would be no legal requirement that the system provide a permanent record within itself even though such might be desirable from a technical point of view. The evidentiary value of data generated from a particular scientific technique is usually based on whether the technique has gained scientific acceptance among experts in that field.⁽¹⁷⁾

More than likely the expert witness will be using data generated by persons other than himself. Although he may be recognized by the court as qualified to interpret such data, the ground rules are such that it may be advisable to produce in court the record from which his conclusions are drawn as a back-up to his testimony. Thus, it is advisable to identify those persons in the record who contributed toward the derivation of that record. In addition, the technical qualifications of persons who generate dose data should be documented along with procedural techniques,

and, when the dose-evaluation stage is reached, the particular contribution of these persons should be identified in the record by some appropriate system. It is the identification of the person who bears the responsibility of dose evaluation, along with the documentation of acceptable procedural techniques, that has a bearing on the credibility of the testimony offered in evidence and protects the expert whose job it is to discuss it.

Summary

It seems likely that the record-keeping system should stress (i) the retention of that data used in the evaluation of radiation dose where key factors that affect the dose number derived are subject to individual interpretation, (ii) the identification and qualifications of those individuals responsible for dose evaluation and the recording of the data associated with it, and (iii) a time of retention based on the applicable statutes of limitation which, for practical purposes, appears to be commensurate with life expectancy rather than with tenure of employment or the duration of the exposure.

REFERENCES

1. H. E. Plaine, Health Phys. 9, 7 (1963).
2. J. C. Hart, Health Phys. 13, 319 (1967).
3. William L. Prosser, The Law of Torts. St. Paul, Minnesota: West Publishing Co. (1955).
4. Analysis of Workmen's Compensation Laws. Chamber of Commerce of the United States, Washington, D. C., January 1964.
5. Besner v. Walter Kidde Nuclear Laboratory et al., New York Supreme Court, Appellate Division, Third Judicial District, December 20, 1965, 265 New York Sup. 2nd 312.
6. Employers Mutual Liability Insurance Company of Wisconsin v. Parker, Texas Court of Civil Appeals, Fourth District, July 31, 1967.
7. John R. Lindsey, Medical Economics 37, 172 (1960).
8. U. S. Atomic Energy Commission Manual. Chapter 0502: "Reporting and Investigating Accidents and Radiation Exposure." Chapter 0524: "Standards for Radiation Protection." Chapter 0525: "Occupational Radiation Exposure Information." Washington, D. C.: U. S. Government Printing Office.
9. Charles T. McCormick, Law of Evidence. St. Paul, Minnesota: West Publishing Co. (1954).
10. Uniform Laws Ann., Vol. 9, p. 387 (1951).
11. State of Texas, Texas Laws 1951, Chapter 321.
12. Roy R. Ray, Southwestern Law Journal, 5, 33 (1951).
13. McVey et al. v. Phillips Petroleum Co., U. S. Court of Appeals, Fifth Circuit, March 10, 1961, 288 F. 2nd 53.
14. Edward Forgotson, Health Phys. 9, 741 (1963).
15. U. S. Department of Labor and the U. S. Atomic Energy Commission, Studies in Workmen's Compensation and Radiation Injury, Vol. 1, Appendix B. Washington, D. C.: U. S. Government Printing Office.
16. American Standard Practice for Occupational Radiation Exposure Records Systems. (USA N2.2, March 1966.) United States of America Standards Institute, New York, N. Y.
17. Frye v. United States, 293 Fed. 1013, 1014 (C.A. D.C. 1923).

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