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QUARTERLY TECHNICAL REPORT

October 1, 1949 thru December 31, 1949

Submitted by: Henry A. Blair
Director

Date of Report: 1/31/50

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INTRODUCTION

The scientific work presented herein has been coded at the program and problem levels according to the scheme given on Pages 7 and 8. In the report all contributions to a given problem have been assembled together without regard to the administrative organization except that the number of the section which did the work is prefixed in each case. By using this number, it can be found on Page 12 what administrative officer can be approached for information about particular work.

It should be noted that the Quarterly Technical Reports of The University of Rochester Atomic Energy Project do not attempt to describe progress in all of the research programs but only in those in which some significant results have been achieved but which are not sufficiently complete to be written up as a final report.

EXPLANATION OF PROGRAM AND PROBLEM CODES

The scientific work at The University of Rochester Atomic Energy Project has been coded at the program and problem levels. The programs, in general, indicate broad fields of investigative or service activities while the problems indicate divisions of these fields. Although no consistent method of division in problems was possible, an attempt was made to achieve a natural division in the sense that each problem would encompass a subject normally written up and generally considered as a unit. The program or chemical toxicity of uranium, for example, has been broken down into problems according to the divisions commonly employed by toxicologists.

The problem codes are not related directly to the administrative organization of the Project. Consequently, the smallest administrative unit, the section, may work on more than one of the coded problems. Conversely, more than one section may work on the same coded problem. The administrative organization will be ignored in making this quarterly report of our research and service activities, all material being assembled according to the program and problem codes. The contribution of each section to a Quarterly Technical Report will be prefixed by the section number, however, to permit reference to the administrative organization if necessary.

It has not been possible to code the problems sufficiently broadly to avoid all overlapping. In cases in which various parts of a given investigation might be coded differently, the whole work was coded according to its principal subject matter as long as the minor subjects were relatively unimportant. Otherwise, the work was divided under appropriate codes.

PROGRAM AND PROBLEM CODES

I. X.R. BIOLOGICAL EFFECTS OF EXTERNAL RADIATION (X-RAYS AND γ RAYS)

- X.R.1 Tolerance Studies (dose levels, surivial time, gross and histo-pathology)
- X.R.2 Mechanism of Effects (physiological and biochemical)
- X.R.3 Therapy (measures against radiation effects)
- X.R.4 Hematology
- X.R.5 Genetics (histogenetics)
- X.R.6 Embryology
- X.R.7 Bacteriology and Immunology

II. I.R. BIOLOGICAL EFFECTS OF EXTERNAL RADIATION (INFRA-RED & ULTRA-VIOLET)

- I.R.1 Flash Burns

III. R.M. BIOLOGICAL EFFECTS OF RADIOACTIVE MATERIALS (CONTACT, INGESTION, ETC.)

- R.M.1 Polonium
- R.M.2 Radon
- R.M.3 Thoron
- R.M.4 Miscellaneous Project Metals

IV. U. URANIUM

- U.1 Physical and Chemical Properties
- U.2 Toxic Effects (description of acute and chronic toxicity)
- U.3 Toxic Limits (respiratory; oral; skin; eye; parenteral)
- U.4 Fate (distribution and excretion)
- U.5 Mechanism of Toxic Effects
- U.6 Methods of Detection of Poisoning, Prophylaxis, Treatment and Protection

V. Be. BERYLLIUM

Be.1 Physical and Chemical Properties

Be.2 Toxic Effects (description of acute and chronic toxicity)

Be.3 Toxic Limits (respiratory; oral; skin; eye; parenteral)

Be.4 Fate (distribution and excretion)

Be.5 Mechanism of Toxic Effects

Be.6 Methods of Detection of Poisoning, Prophylaxis, Treatment and Protection

VI. Th. THORIUM

Th.1 Physical and Chemical Properties

Th.2 Toxic Effects (description of acute and chronic toxicity)

Th.3 Toxic Limits (respiratory; oral; skin; eye; parenteral)

Th.4 Fate (distribution and excretion)

Th.5 Mechanism of Toxic Effects

Th.6 Methods of Detection of Poisoning, Prophylaxis, Treatment and Protection

VII. F. FLUORIDE

F.1 Physical and Chemical Properties

F.2 Toxic Effects (description of acute and chronic toxicity)

F.3 Toxic Limits (respiratory; oral; skin; eye; parenteral)

F.4 Fate (distribution and excretion)

F.5 Mechanism of Toxic Effect

F.6 Methods of Detection of Poisoning, Prophylaxis, Treatment and Protection

VIII. Zr. ZIRCONIUM

Zr.1 Physical and Chemical Properties

Zr.2 Toxic Effects (description of acute and chronic toxicity)

Zr.3 Toxic Limits (respiratory; oral; skin; eye; parenteral)

Zr.4 Fate (distribution and excretion)

Zr.5 Mechanism of Toxic Effect

Zr.6 Methods of Detection of Poisoning, Prophylaxis, Treatment and Protection

IX. S.M. SPECIAL MATERIALS

S.M.1 Physical and Chemical Properties

S.M.2 Toxic Effects (description of acute and chronic toxicity)

S.M.3 Toxic Limits (respiratory; oral; skin; eye; parenteral)

S.M.4 Fate (distribution and excretion)

S.M.5 Mechanism of Toxic Effect

S.M.6 Methods of Detection of Poisoning, Prophylaxis, Treatment and Protection

X. I.S. ISOTOPES

I.S.1 Tracer Chemistry

I.S.2 Radioautography

I.S.3 Therapy

XI. O.S. OUTSIDE SERVICES

XII. P.H. PROJECT HEALTH

XIII. H.P. HEALTH PHYSICS

H.P.1 Research and Development

H.P.2 Service

XIV. C.S. SPECIAL CLINICAL SERVICE

XV. I.N. INSTRUMENTATION (SPECTROSCOPY, ELECTRON MICROSCOPY, X-RAY AND NUCLEAR RADIATION DETECTORS, X-RAY DIFFRACTION, ELECTRONICS)

I.N.1 Research and Development

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2604	Zirconium	Herbert E. Stokinger

PROGRAM X.R.

BIOLOGICAL EFFECTS OF EXTERNAL RADIATION (X-RAYS AND RAYS)

Problem Code: X.R.2 (Mechanism of Effects)

Section Code: 3140

Authors: K. Salomon, K. I. Altman, R. Della Rosa, S. J. Sanfilippo

Studies on the Biosynthesis of Chlorophyll.

Background. Since it has been shown in this laboratory that the alpha-carbon atom of glycine is incorporated in hemin (1), it seemed of interest to investigate the mechanism of the biosynthesis of the tetra-pyrrol structure characteristic of both hemin and chlorophyll. Granick has shown (2,3) that exposure of chlorella vulgaris to x-radiation can result in the formation of mutants which are unable to carry to completion the synthesis of chlorophyll. For this reason chlorella vulgaris was considered a suitable tool for the elucidation of the chain of biosynthetic events leading to the tetra-pyrrol structure. Thus, it might be possible to produce mutants which would synthesize various precursors of lower order. Since there is great chemical similarity between the porphyrin skeletons of hemin and chlorophyll, it might be expected that chemically very similar or even identical precursors would be encountered in the biosynthesis of these two tetra-pyrrol structures. This approach is the alternative to an investigation of the *in vivo* utilization of hypothetical precursors which could be synthesized by organic chemical means. Efforts to establish the sequence of events with respect to the biosynthesis of the tetra-pyrrol structure seem essential in view of the importance of an understanding of the biochemical mechanism of the effect of ionizing radiation on hematopoiesis.

In the experiments to be described, glycine and acetate labeled in their

respective alpha-carbon atoms with C^{14} were used, since both of them had been shown to function as precursors in the biosynthesis of the tetra-pyrrol structure of hemin (1,4).

Methods. A. Culturing of chlorella vulgaris.

Chlorella vulgaris was grown under sterile conditions in a medium of the following composition*

KNO_3 (molar)		3 ml.
$Ca(NO_3)_2 \cdot 4H_2O$ $(\frac{M}{2})$		2 ml.
KH_2PO_4 (M)		1 ml.
$CaCL_2$ $(\frac{M}{2})$		8 ml.
$MgSO_4$ $(\frac{M}{2})$		4 ml.
Fe Citrate (0.5%)		1 ml.
Trace Mixture		1 ml.
H_2O	to make	1 l.
Glucose	at	10 gm/l.
Agar	at	2%

Trace Mixture - One liter of trace mixture contains:

H_3BO_3	2.86 gm.
$MnCl_2 \cdot 4H_2O$	1.81 gm.
$ZnSO_4 \cdot 7H_2O$	0.22 gm.
$CuSO_4 \cdot 5H_2O$	0.08 gm.
Ammonium Molybdate	0.05 gm.

The organisms were grown under stationary conditions in 125 ml. Erlenmeyer flasks, each containing 30 ml. of liquid medium to which either labeled glycine or labeled acetate had been added. The glycine had a C^{14}

*We wish to acknowledge the advice given us by Dr. S. Caplin of the Department of Botany, University of Rochester, in the selection of this medium.

activity of 2.55×10^3 disintegrations per minute per micromole of carbon and the acetate had a C^{14}-activity of 2.57×10^3 disintegrations per minute per micromole of carbon. The total amount of glycine or acetate in each flask was 22 and 24 milligrams respectively, providing equimolar concentrations and equivalent activities. The cells were grown under daylight lamps at pH 5.8 for one week and then harvested by centrifugation.

B. Preparation and analysis of methylpheophorbide.

From approximately 40 flasks about 10 grams wet weight of dark green cells were obtained. The centrifuged cells were washed repeatedly with distilled water and finally with a dilute solution of inert glycine or acetate. The cells were then suspended in dioxane and ground in a glass homogenizer using 100 mesh pyrex glass powder. This procedure was repeated until the dioxane solution remained colorless. The chlorophyll thus extracted was precipitated several times with an aqueous salt solution, dissolved in acetone, and hydrolyzed in an hydrochloric acid solution of a final concentration of 34%. Pheophorbide thus formed was extracted with ether and esterified with methyl alcohol. Methylpheophorbide was crystallized several times from chloroform-ligroin boiling at 60-90°C. Absorption spectra of the two labeled methylpheophorbide preparations were compared with that of methylpheophorbide prepared from chlorella vulgaris cultures grown in a medium containing inert glycine. All three preparations showed the same absorption curve with a peak at approximately 665 mμ which is characteristic for methylpheophorbide a, and an increased absorption in the region of 550 to 500 mμ lacking, however, the well defined known absorption peak for methylpheophorbide b. The masking of the latter peak is probably due to the presence of an absorbing impurity.

Since spectroscopy proved inconclusive in the case at hand, carbon and

hydrogen analyses for the various methylpheophorbide preparations were carried out. The values obtained are presented in Table 1 below.

Table 1

Carbon and Hydrogen Content of Various Methylpheophorbide Preparations

	No labeled material added - but inert acetate present.	$C^{14}H_2NH_2COOH$ Added	$C^{14}H_3COONa$ Added	Calculated Values for Methylpheophorbide a.	b.
%C	67.55	73.80	66.61	71.30	69.0
%H	7.32	9.92	8.86	6.27	5.80

For the purpose of comparison, theoretical values for methylpheophorbide a and b are shown in the last two columns. In evaluating the analytical results, it must be kept in mind that the methylpheophorbide preparations isolated from chlorella vulgaris were in all cases mixtures of the methyl esters of pheophorbide a and b, since we as yet have not attempted to separate these two compounds. It should also be pointed out that the ratio of methylpheophorbide a and b might be a function of the conditions of culturing. It is thus of interest to note that the analytical values obtained from material isolated from chlorella grown in the presence of acetate (columns 1 and 3) are in better agreement with each other than with values obtained when glycine instead of acetate was present in the culture medium (column 2). The somewhat elevated hydrogen values obtained are difficult to understand at the present stage of the work.

It should be pointed out that in the elementary analysis of chlorophyll-like compounds considerable difficulties are encountered, particularly with

respect to complete combustion when the ordinary Pregl methods are used. Nitrogen values are not shown because analytical procedures used up to now were found to be inadequate. However, we have recently been able to overcome this difficulty by carrying out combustions at 900°C*.

Results. In Table 2 below, the incorporation of the alpha-carbon atoms of acetate and of glycine in methylpheophorbide isolated from chlorella vulgaris is shown. The values are expressed as disintegrations per minute per micromole of carbon, using the values obtained by elementary analysis as the basis for calculation of total carbon content. The data presented indicate that the alpha-carbon atoms of glycine and acetate respectively are utilized by chlorella vulgaris for the synthesis of chlorophyll. It is also apparent that the alpha-carbon atom of acetate makes a greater contribution to the carbon skeleton of methylpheophorbide than the alpha-carbon atom of glycine.

Table 2

The Incorporation of $C^{14}H_2NH_2COOH$ and $C^{14}H_3COONa$ in Methylpheophorbide Isolated from Chlorella Vulgaris**

Isotopically Labeled Precursor	C^{14}-Activity Disintegrations per Min. per μM carbon	Co/C***
$C^{14}H_2NH_2COOH$	43	59
$C^{14}H_3COONa$	148	17

***Ratio of C^{14}-Activity of Compound Added (Co) to C^{14}-Activity of Compound Isolated (C), i.e. Dilution Constant.

*We are indebted to the Micro-Analytical Laboratory of the Eastman Kodak Company, and particularly to Mr. D. Ketchum, for performing these analyses for us.

**The carbon^{14} activities of the methylpheophorbide esters analyzed were in the order of at least five times backgrcund.

Discussion. At the present time only over-all biosynthetic reactions with respect to the tetra-pyrrol structure of hemin and chlorophyll are known. No statement can therefore be made with respect to the differences in synthetic pathways by which chlorophyll a and b might be formed. Such differences might actually be very slight since chlorophyll a and b differ only with respect to the chemical nature of the substituent in carbon atom 3, where, in the case of chlorophyll a, a methyl group is present instead of the formyl residue in the case of chlorophyll b. For this reason it did not seem essential at this stage of our knowledge to distinguish between chlorophyll a and b. It seemed important first to establish the fact that the alpha-carbon atoms of glycine and acetate are utilized directly in the biosynthesis of chlorophyll.

The low isotope dilutions as calculated in Table 2 (Page 18) support the conclusion that direct utilization of the two labeled carbon atoms has occurred. The spectroscopic as well as the analytical data indicate that a small impurity is present in our preparations although the method by which these preparations were obtained makes the presence of high activity contaminants unlikely. Because of the possible presence of small amounts of impurities, the activity data were calculated as indicated, namely, on the basis of the total amount of carbon present in the compounds analyzed.

Summary. From the data presented, we conclude that chlorella vulgaris is able to utilize the alpha-carbon atoms of glycine and acetate for the biosynthesis of chlorophyll.

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Problem Code: X.R.2 (Mechanism of Effects)

Section Code: 3150

Authors: L. T. Steadman and H. E. Thompson

Changes in Blood Lipids After Whole Body X-Irradiation.

Introduction. In the Quarterly Report for January to March 1949, No. UR-70, the above authors presented some initial results on the changes in plasma total lipids in rabbits following whole body x-irradiation. The radiation dose employed was 2000 r at 100 KV (peak) with no filtration other than the inherent filtration. The half value layer of this radiation was 1 mm of aluminum. It was found that on the first or second day after irradiation there was a transitory elevation in the total lipid, phospholipid, and cholesterol plasma levels which in some instances were as much as ten times the normal value. Also the frequently observed lipemic or opalescent plasmas were those showing the high lipid values. These lipid measurements have since been extended to other animals, the rat, dog and guinea pig; and the dose and quality of the radiation have been varied. The purpose has been to attempt to ascertain how important the lipid changes are as biological effects of radiation. Some of the results of the present experiments will be discussed in summary form in the following report.

Methods. The total lipids were measured as described before, by treatment of one ml amounts of plasma with alcohol-ether, followed by extraction with petroleum ether and weighing. The phospholipid and cholesterol were determined

on aliquots of the alcohol-ether extract of another sample. The micro oxidative method of Bloor was used to determine the phospholipid and Bloor's acetic anhydride-sulfuric acid procedure was used to measure the total cholesterol. Glucose was determined by the micro variation of the procedure of Folin and Wu.

Procedure. Since the radiation initially used was relatively soft and since Kultjugin found high lipid values after irradiation of rabbits with ultraviolet light, it might be supposed that the radiation effects were caused primarily by irradiation of the skin. It was of interest, therefore, to vary the quality of the radiation by increasing the peak voltage of the tube and also by introducing appropriate filtration. Furthermore, it is usually considered that the liver plays an important part in lipid metabolism. When the whole body, including the liver, is irradiated it is not easy to interpret the finding of elevated lipids in the blood stream in terms of radiation effects on the liver. The damaged liver may lose lipids or on the other hand it may not readily remove lipids from the blood which are mobilized from other locations. As a means of gaining more information on these points, it was decided to carry out a separate series of experiments wherein the liver was shielded from the radiation to a large extent. To do this a strip of lead 1.6 mm thick and 7.6 cm wide was placed across the abdomen over the region of the liver while the rabbit was being irradiated lying stretched out on its back on the animal board. It was determined that the liver was adequately covered by the lead and that the radiation below the lead was reduced to less than 5% of the intensity without the lead. However, with this arrangement it should be stated that a part of the stomach and small intestines, the adrenals, and the spleen are also protected to a considerable degree.

The irradiation periods were considerably longer when filtration was

used so the animals in these experiments were irradiated while in an upright position inside a wooden box.

Results. The data are shown in Table 1 (Page 23) as averages for the number of animals in each type of experiment. Although measurements were made at several times following irradiation, the values generally returned to the pre-irradiation levels after 3 days as shown in the earlier report and thus only the changes for the one and three day samples are given herewith. The per cent changes from the pre-irradiation value have been tabulated. For example, a value double the normal measurement is a + 100% change. The values just before death of the animal are also included since, as seen before, there is frequently an increase in lipids at this time even though it may be many days after the time of irradiation.

The data for the experiments using 2000 r at 100 KV unfiltered radiation are the most extensive. These do not include the previously reported data taken under the same conditions but are an addition to the earlier experiments and are in good agreement with the first results. Whereas the maximum changes on the first day may be as great as + 1000%, it is seen that the average increase is 330% for the total lipids, 140% for the phospholipid, and 84% for the cholesterol. The number of plasmas on the first day which are lipemic in appearance is 45%. In the earlier set of data where there were 2 day samples as well, the incidence of lipemic plasma was 70%. In other words, lipemic plasma was noted in 70% of the animals irradiated.

Comparing other data in Table 1 (Page 23) with the above figures, it is noted that with the liver shielded the increases in lipid are not as much but on the other hand the changes are not reduced to zero. It is concluded that although the irradiation of the liver makes some difference in the over-all

TABLE I

AVERAGE PER CENT CHANGE IN PLASMA TOTAL LIPIDS, PHOSPHOLIPID, AND CHOLESTEROL AFTER X-IRRADIATION OF RABBITS

Radiation No	Filter	Shield	No. Irrad.	Total Lipid Days 1	Total Lipid Days 3	Total Lipid Death	Phospholipid Days 1	Phospholipid Days 3	Phospholipid Death	Cholesterol Days 1	Cholesterol Days 3	Cholesterol Death
2000 r	200 KV*	none	3	+160		+88	+320	+12	+250	+130	+90	+120
		Liver	3	+170	+60		+160	+44	+320	+ 61	-28	+ 63
1500 r	200 KV	none	1	+140	-10		+230	-65	+130	+240	+37	+270
		Liver	1	+400	+55		+600	-45		+200	+140	
1000 r	200 KV	none	6	+220	+22		+ 45	-60	+230	+ 60	-15	+ 60
		Liver	5	+ 71	+10		+ 71	+ 0	+410	+ 46	+ 2	+ 87
500 r	200 KV	none	6	+ 80	+33		+120	-27		+ 54	+19	
		Liver	3	+ 2	-20		+120	+44		+ 0		
2000 r	100 KV**	none	14	+330	+70	+260	+140	+75	+100	+ 84	+45	+190
		Liver	10	+250	+55	+ 41	+150	+120	+520	+ 65	+50	+180
200 KV*** Filtered												
2000 r		none	3	+990		+990	+1060		+1060	+390		+390
1000 r		none	13	+170	+ 0	+130	+220	+83	+570	+150	+41	+260
500 r		none	8	+ 67	+10		+ 80	+ 8		+ 63	+18	
100 KV**** Filtered												
1000 r		none	2	+ 91	-20		+33	-33		+ 27	+27	
500 r		none	1	+190	- 3		+33	-22		+ 23	+ 6	

* HVL 0.16 mm Cu
** HVL 1 mm Al
*** Al parabolic +0.5 mm Cu HVL 1.5 mm Cu
**** 1 mm Al HVL 2 mm Al

metabolism of the lipids, the abnormal increases are not solely due to radiation damage to the liver.

With the unfiltered radiations the variation from 100 KV to 200 KV with the same dose does not show remarkable differences in the total lipids although the phospholipid and cholesterol changes are greater. If the radiation effects depend on the amount of tissue irradiated, then the higher kilovoltage radiation which would produce a greater depth dose might be expected to be more effective.

When one compares these data also with the figures obtained for the much harder radiation shown in the lower half of the table, it is noted that for the 500 and 1000 r doses again the phospholipid and cholesterol changes are greater and the total lipid values are about the same. It seems that the phospholipid and cholesterol therefore increase with increase in depth dose, while the total lipids which are for the most part neutral fat are not so influenced by change in depth dose. On the other hand the changes for 2000 r with the 200 KV filtered radiation are the most pronounced and are very large for all three of the measured quantities so that the total lipids may increase with the depth dose when the dose is large.

The lipid changes due to x-irradiation are therefore not simply effects in the skin but are associated with irradiation of the whole body.

The average survival time of the animals with the liver shielded is about twice that for the other rabbits.

Rat Experiments. In order to determine whether or not the lipid increases are peculiar to the rabbit which is believed to have a slow or poor mechanism for taking care of lipids, several rats were tested under similar conditions. The results are shown in Table 2 (Page 25) where the change in total lipid is recorded for each of the 11 rats. Although the experiment is not extensive

TABLE 2

RATS

Per Cent Change in Total Lipids One Day After Irradiation

Series 1. 2000 r 200 KV Unfiltered X-Rays HVL 0.16 mm Cu

Series 2. 2000 r 100 KV Unfiltered X-Rays HVL 1 mm Al

Series 3. 2000 r 100 KV Filter 1 mm Al HVL 2 mm Al

Series 1		Series 2		Series 3	
Animal Number	Plasma Lipid Change	Animal Number	Plasma Lipid Change	Animal Number	Plasma Lipid Change
1	-14%	4	-5%	9	+20%
2	-21	6	-24	15	-24
12	-60	7	+90	17	-20
13	+30	8	+20		

enough to prove that there is or is not a small effect due to radiation, there is no evidence that the rat behaves at all like the rabbit or that there is even an increase in lipids due to radiation.

Dog Experiments. In Table 3 (Page 26) are shown somewhat more data on 7 dogs. Here also there are no increases of the magnitude observed in the rabbits although there may be a small radiation effect noticeable in the total lipids.

Guinea Pig Experiments. Six guinea pigs were irradiated under similar conditions. It might be expected that the guinea pig would be more nearly like the rabbit in its lipid metabolism characteristics since they are both herbiverous

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TABLE 3

DOGS

Per Cent Change in Plasma Total Lipids, Phospholipid, and Cholesterol After Irradiation

Series 1. 2000 r 200 KV Unfiltered X-Rays HVL 0.16 mm Cu

Animal Number	One Day After Irradiation			Two Days After Irradiation		
	Total Lipid	Phospho-lipid	Choles-terol	Total Lipid	Phospho-lipid	Choles-terol
997	+31%	+12%	+0%	+71%	%	%
1466	-27	-22	+38	+5		
1467	+16	+3	-6	+5		
1468	+0	-6	+16	+10		
1469		-42	+14	+98	-27	+0
1471	+29	+28	+0	+0	-35	-15
1475	-10	+27	+14	+3	+41	-23
Average	+6.5	+0	+11	+27	-7	-13

animals whereas the rat and dog are not. Table 4 (Page 27) shows one instance of a fairly high total lipid and it may be that the one day period is not the best time to demonstrate a maximum change. There is a consistent increase in the lipids in the red blood cells for the guinea pigs, however, which compares favorably with a similar set of data on six rabbits shown in Table 5 (Page 28).

TABLE 4

GUINEA PIGS

Per Cent Change in Total Lipids and Glucose in Plasma and Red Cells One Day After Irradiation

2000 r 200 KV Unfiltered X-Rays HVL 0.16 mm Cu

Animal Number	Total Lipid		Glucose	
	Plasma	Cells	Plasma	Cells
1	-23%	+55%	-3%	+1%
2	-8	+52	-3	+60
3	+270	+83	-3	+11
5	+52	+16	-15	+0
7		+120		
8	+5	+150	-3	-11
Average		+79		

Conclusions.

1. The lipemia caused by x-irradiation is peculiar to the rabbit. No elevation of plasma lipids is found in the rat and only relatively small increases are detected in the dog. There is some evidence that the guinea pig may show effects similar to the rabbit but to a smaller degree.

2. The production of high lipid values is not simply a radiation effect on the skin but increases with depth dose.

3. Shielding the liver reduces the amount of lipemia appreciably, but damage to the liver is not the only factor in the production of the high blood lipids.

TABLE 5

RABBITS

Per Cent Change in Total Lipids and Glucose in Plasma and Red Cells One Day After Irradiation

1500 r 200 KV Unfiltered X-Rays HVL 0.16 mm Cu

Animal Number	Total Lipid		Glucose	
	Plasma	Cells	Plasma	Cells
680	+200%	+230%	+5%	-40%
686	+120	+120	-10	+35
703	+640	+110	+85	-50
706	+110	+140	+3	+43
727	+620	+10	+8	-44
728	+56	+150	+0	
Average	+290	+126		

PROGRAM U.

URANIUM

Problem Code: U.3 (Toxic Limits)

Section Code: 3210

Authors: H. B. Wilson, G. E. Sylvester, C. W. LaBelle, J. K. Scott and staff, F. A. Smith and staff, S. Laskin and staff, and K. E. Lauterbach

The Relation of Particle Size of Hydrated Uranium Trioxide Dust to Toxicity Following Inhalation by Rabbits and Rats

In an earlier study (1), particles of UO_2 of 0.5 μ mass-median size were found to be more toxic when inhaled by rabbits and rats than were particles of mass-median size greater than 1 μ.

Unlike UO_2, UO_3 is said to be soluble in body fluids. It is more toxic than UO_2 but less toxic than uranyl salts. In aqueous suspension, it slowly takes up water to give first the mono- and then the di-hydrate.

The method of preparation of $UO_3 \cdot 2H_2O$ of mass-median particle size below 1 μ and the toxicity following inhalation by rabbits and rats of a dust, generated by atomization of this suspension at a UO_3 concentration of 10 mg/m^3, has been reported (2).

Two experiments testing the effects of inhalation of hydrated UO_2 of mass-median size above 1 μ and UO_3 concentration of 10 mg/m^3 have since been completed. In the first, which will not be described in detail, the toxicity was significantly greater than that resulting from inhalation of hydrated UO_3 of mass-median size below 1 μ at the same concentration. The reason for this was thought to be chiefly the alkaline treatment used for the purpose of suspending and washing out fines at the end of each day's run. It was thought

that residual adsorbed NaOH probably reacted with CO_2 stirred into the suspension to give rise to significant amount of the more highly toxic sodium uranyl carbonate. The second "above 1 μ" experiment was designed to eliminate this probable cause of increased toxicity. Description of the second large-size (coarse) hydrated UO_3 experiment at a concentration of 10 mg/m^3 follows with comparisons with comparable results of the 'small' particle-size study.

Method. Orange UO_3 was freed from uranyl nitrate and converted to the yellow hydrate or mixture of hydrates ($UO_3 \cdot H_2O$ and $UO_3 \cdot 2H_2O$) by repeated washing over a period of several days. The suspension was atomized in the exposure chamber and rabbits and rats exposed 6 hours daily for 37 calendar days duplicating the schedule of the fine $UO_3 \cdot 2H_2O$ experiment. At the end of each day's run, NaOH solution was added to the contents of the chamber jar to suspend accumulated fines which were eliminated by decanting after a suitable settling period. After washing, adsorbed NaOH was eliminated by addition of dilute HCl followed by repeated washing until practically no UO_2^{++} remained in the supernate and the pH approximated that of the distilled water used.

The specific surface (Table 1 Page 31) of a typical sample of the coarse suspension was found by the ethane adsorption method to be 2.0 m^2/g. The porosity factor was 27.4. Corresponding values for the fine suspension were 5.14 m^2/g and the porosity factor 15.9. In applying the method, it was necessary to heat the compound to 110°C at pressure as low as 10^{-6} mm. Surface area and porosity values are therefore probably those of the partially dehydrated product, $UO_3 \cdot 1/2\ H_2O$.

Daily mean temperatures in the exposure chamber ranged from 74-77°F. The room in which animals were kept between exposure was not conditioned, however, and temperatures there were higher. Mean daily temperatures in the

TABLE 1

SPECIFIC SURFACE AND RELATED DATA FOR HYDRATED URANIUM TRIOXIDES USED FOR INHALATION TOXICITY STUDIES

Sample Description	Particle Size[a]				Specific Surface, M^2/g		Porosity
	Mass Median, μ	Geometric Standard Dev. of Mass Median	Count Median, μ	Surface Median, μ	Calc. From Particle Diam's.	By Ethane Adsorption	
Fine Suspension	1.48	1.56	0.42	1.13	0.32	5.1	15.9
Coarse Suspension	10.0	1.27	4.75	9.4	0.073	2.0	27.4

[a]Measurements of optical microscope

exposure chamber during the 0.6 μ experiment ranged from 70-76° F. Because of the nature of the experiment, matched groups of controls of both species were used on which all criteria necessary in evaluating toxicity of the uranium-exposed animals were employed.

Results. One rat died on the 20th calendar day, another on or about the 35th calendar day, and one rabbit on the 35th calendar day. Because of advanced autolysis, no estimate of the cause of death could be made in any case. It should be noted that these deaths occurred during the months of July and August, the last two immediately following a few days of exceptionally hot weather. The rabbit death was preceded by a sharp rise in blood NPN but the mean NPN of 6 control rabbits simultaneously rose to values significantly above normal. Results for survivors are compared in Table 2 (Page 32) with those following inhalation of hydrated UO_3 of smaller particle size.

TABLE 2

MEAN RESULTS OF EXPOSURE OF RABBITS AND RATS TO INHALATION OF HYDRATED UO_3[a]

Concentration $\frac{MgUO_3}{M^3}$	Particle Size Mg	Rabbit						Rat			
		Growth Retardation %	Urinary Protein Mg %	Blood NPN % Elevation	Urea N % Elevation	Terminal Renal Damage	Terminal U in Lung Mg U per g Fresh Tissue	Growth Retardation %	Serial Lung Damage	Terminal Renal Damage	Terminal U in Lung Mg U per g Fresh Tissue
9.9	0.6[b]	44	11	0	5	Moderate in 2 Minimal in 8	83	33	+7	Slight in 9 Trace in 1	69
10.0	3.7	40	13	4	5	Minimal in 9 of 9	36	28	+6	None	23

(a) Concentration of the hydrated UO_3 is the average of all daily mean concentrations. The particle size is the average of daily mass-median values determined by the Modified Cascade Impactor method. Growth retardation is expressed as the percentage difference between the mean percentage weight increase of controls and that of exposed from the 1st to the 30th calendar day (the 3rd to the 30th calendar day for the rabbit). Urinary protein is the mean of daily means for the first 3 weeks of exposure; elevations of blood NPN and urea N are percentage increases of exposed means over control means for the first 3 weeks of exposure. Terminal renal damage is the micropathologists estimate of damage, indicated by the presence of regenerated tubular epithelium of a type usually associated with uranium poisoning. Serial lung-damage values are derived from estimates of gross lung damage in serially sacrificed rats, and represent, in standard error units, the difference between exposed and control means. Any value greater than 3 has a high probability of being significant.

(b) Because of a recently discovered sampling error, the reported value of 0.45μ (2) must be revised upward. It is highly probable however, that the average mass-median value will not exceed 0.6 μ.

Weight response of "terminal" rats in the two studies are compared graphically in Figure 1 (Page 34). It will be observed that growth retardation occurred to about the same extent in both experiments. The somewhat greater loss in weight during the first week of exposure to the smaller size particles represents a typical weight response to acute U injury.

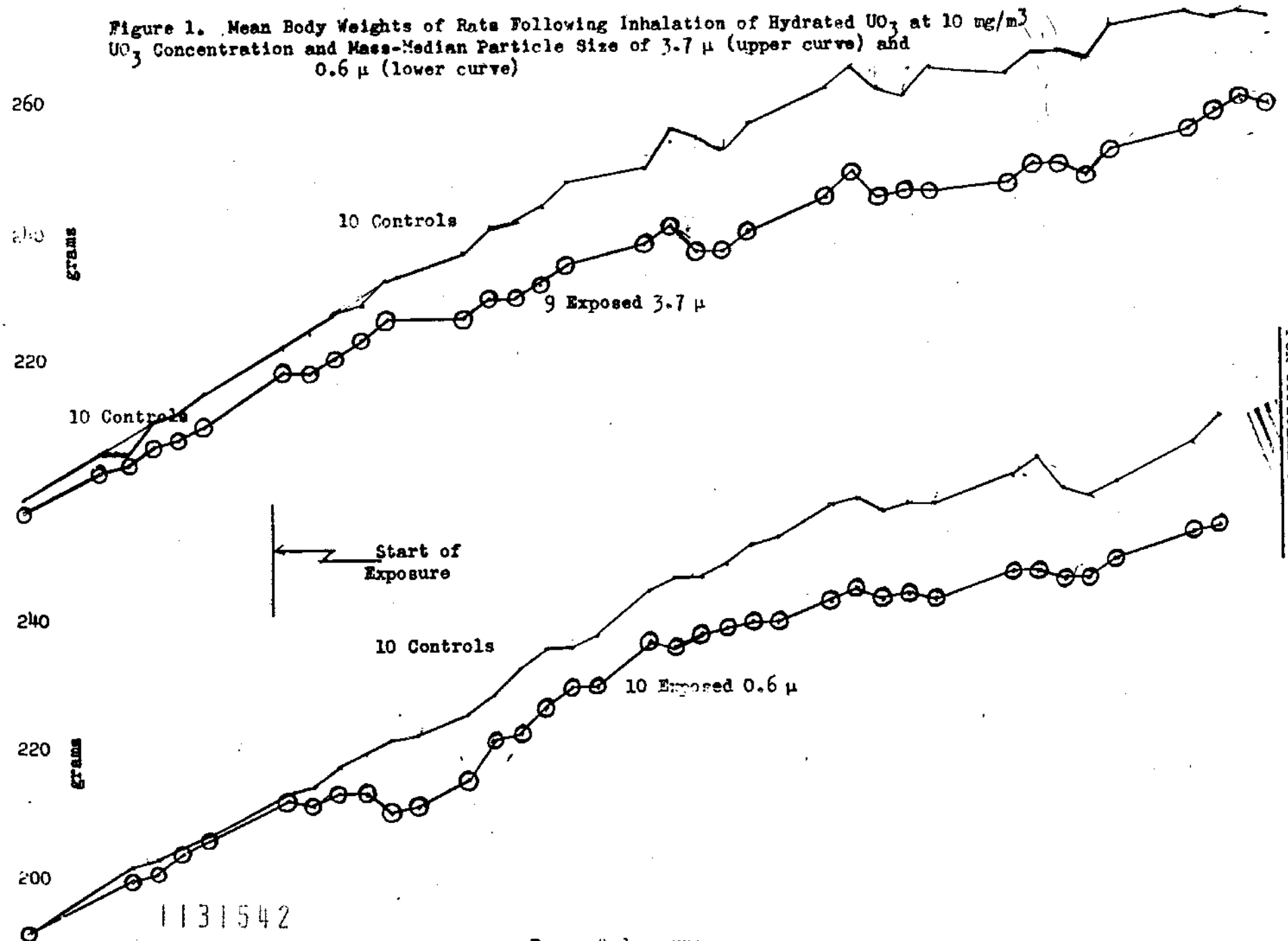
Discussion. The fact that three animals died following exposure to dust of 3.7 μ mass-median size, whereas at the same concentration none died following exposure to dust of less than 1 μ size seems to indicate a higher toxicity for the larger particle-size range. However, the exceptionally hot weather as a contributing cause of death and the elevation of blood NPN of control rabbits immediately preceding two of the deaths, must not be overlooked. It is doubtful that significant mortality would have been experienced if the large particle-size experiment had been performed not in July-August, but in October, the month during which the "<1 μ" study was undertaken.

With the possible exception of pathologic results, tabulated responses (Table 2 Page 32) of surviving rabbits show no significant difference whatever, whereas corresponding responses of rats indicate a slightly greater toxicity for particles of mass-median size below 1 μ.

In both species, the values for U in the lung (Table 2 Page 32) are more than twice as great following inhalation of the finer dust. A difference such as this was expected. On the other hand, if UO_3 is soluble in lung fluids, it is difficult to understand the retention of such relatively large amounts of U in the lung in either experiment.

Solubility of UO_3 in body fluids may be the factor because of which very little difference in toxicity resulted at such widely different average mass-median particle sizes. Surface area differences may, however, be a factor. It

Figure 1. Mean Body Weights of Rats Following Inhalation of Hydrated UO_3 at 10 mg/m^3 UO_3 Concentration and Mass-Median Particle Size of 3.7 μ (upper curve) and 0.6 μ (lower curve)



is seen (Table 1 Page 31) that porosity of the larger particles was nearly twice that of the smaller.

Conclusions. (1) Hydrated UO_3 at a UO_3 concentration of 10 mg/m^3 is slightly to moderately toxic when inhaled by rabbits and rats; and (2) particles of approximately 0.6 μ mass-median size were not significantly more toxic than those of 3.7 μ mass-median size. This second conclusion is made with the reservation, based on the responses of survivors (Table 2 Page 32) that, if susceptibility has not been aggravated by relatively high temperatures, the over-all toxicity of particles of 3.7 mass-median size might have been slightly but significantly lower than that of the finer particles.

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PROGRAM Be.

BERYLLIUM

Problem Code: Be.1 (Physical and Chemical Properties)

Section Code: 3220

Authors: W. F. Neuman, T. Y. Toribara, A. Underwood, J. Bonner, and I. Feldman

Analytical Chemistry. Having developed an elegant method for the determination of beryllium (1 to 20 micrograms) using alkanet or its parent-compound, naphthazarin, as the colorimetric reagent, a series of investigations were undertaken to elucidate the mechanism of the color reaction of beryllium with hydroxyanthraquinones. It has been shown that beryllium combines with the dye in stoichometric proportions, forming chelate-ring structures corresponding to the two types: Be-dye and Be(dye)$_2$. From the data at hand, it has been possible to assign structural formulae to these two compounds with a fair degree of certainty.

The most promising reagent for the microdetermination of beryllium in biological specimens is morin, as described by Sandell. It is the only agent thus far studied which possesses the extreme sensitivity required. The methods and procedures described to date have been unsatisfactory due primarily to the unavailability of pure morin. The commercial product contains only from 0.1 to 2% of the active material combined with a variable mixture of oil, resins, and other products. Culminating a series of studies on the technical grade product sold by Eastman Kodak Company, a procedure has been developed for the rapid isolation of pure morin. Preliminary experiments using this product are most promising and indicate that as little as 0.005 micrograms of Be may be detected.

All the chemical procedures for the analysis of beryllium are subject to interference by extraneous cations. Until recently it appeared difficult to

obtain an isolation method which did not involve many time consuming and laborous steps. To obviate these difficulties a series of investigations of the behavior of beryllium toward cation exchange resins was instituted. Under proper conditions, beryllium may be separated quantitatively from both calcium and phosphate by means of such resins.

Physical Chemistry. As part of a program of study designed to explain the metabolism and transport of beryllium in the animal body, the physical chemical properties of beryllium in solutions of physiological composition have been investigated. It appears that four types of substances are very active in complexing beryllium at physiological pH. These substances are; protein; bicarbonate (at high concentrations only), organic phosphates (in particular the polyphosphates derivatives), and complex organic acids related to citrate.

Problem Code: Be.3 (Toxic Limits)

Section Code: 3210

Author: R. H. Hall

Inhalation Toxicity of Beryllium Fluoride Mist. This report, in preliminary form, comprises the second of a series of inhalation toxicity studies delineating the character of beryllium fluoride poisoning in which the ultimate objective is the determination of safe limits of exposure to this compound. Beryllium fluoride is an intermediate in preparation of beryllium metal and which in plant operation constitutes a personnel exposure hazard. The first study, previously reported in this quarterly, was performed at a concentration of 10 mg/m^3. This concentration was so uniformly lethal to the majority of animals that the exposure was concluded after a period of 3 weeks; at this time an

LD50 obtained for dogs, rats and cats. The present study at 1 mg, although still highly toxic, was lethal only to rats and dogs after more than three months of exposure.

During exposure to a concentration of approximately 1 mg/m^3 of BeF_2 mist. the outstanding effects observed were a high mortality among full grown male rats and progressive development of a macrocytic anemia in dogs. A total of 141 animals comprising four species (5 cats, 6 dogs, 10 rabbits, and 120 rats) were exposed for a period of 102 calendar days (422 hours) beginning August 30th and ending December 9, 1949. The animals were exposed six hours daily for five days each week. One dog died on the 30th calendar day. The death of a second dog occurred on the 71st calendar day. Exposure of the remaining 4 dogs is being continued.

The data pertaining to mortality in two groups of adult male rats are summarized in Table 1 (Page 39). The initial group mean weights of the rats in these two groups were 290 and 300 grams, respectively. It will be noted that in the first group of rats, 37 of the original 40 animals were dead after accumulative exposure of 74 hours. The three surviving rats were exposed an additional 132 hours during a period of one month after which they were sacrificed. All of the rats in the second group of 30 animals were dead after accumulative exposure of 72 hours. In a third group of 40 rats having an initial group mean weight of 226 grams, only 8, or 20%, died during the period of exposure of 60 hours. The death of all rats was preceded by marked weight loss. The difference in mortality between the group of younger rats and the two groups of older rats was striking. The cumulative mortality in the two groups of older rats after 60 hours exposure was 63 and 73%, respectively.

The hematologic changes among the dogs were more striking than in previous experiments with inhaled hydrated $BeSO_4$ mist and BeO dust. All of the

TABLE I

CUMULATIVE MORTALITY AMONG RATS EXPOSED TO 1 mg/m^3 OF BeF_2 MIST

Group No.	Cumulative Hours Exposure	Cumulative Ct ($mg\text{-}min/m^3 \times 10^{-3}$)	Cumulative Mortality	
			No. deaths per No. exposed	%
I	48	2.9	8/40	20
	54	3.2	10/40	25
	57	3.4	13/40	32.5
	60	3.6	25/40	62.5
	66	4.0	29/40	72.5
	68	4.1	34/40	85.0
	74	4.4	37/40	92.5
II	42	2.5	6/30	20
	48	2.9	16/30	53
	54	3.2	19/30	63
	60	3.6	22/30	73
	66	4.0	28/30	93
	72	4.3	30/30	100

dogs received a dietary supplement of one teaspoonful of Lextron* daily for one month before exposure. At the beginning of exposure, the Lextron was discontinued in the diet of two of the dogs (#1537 and #1538). The red blood cell counts and the hemoglobin concentration began to decrease after 20 days exposure of all 6 dogs. At about the same time, the mean corpuscular volume began to increase. Neglecting the data for the two dogs that died and for which the data are therefore necessarily incomplete, there remained two groups of two dogs each, one group including dogs #1437 and #1518 which continued to receive the dietary supplement of Lextron throughout the exposure period and another group including dogs #1537 and #1538 for which the supplement was discontinued. It is noteworthy that the decrease in the red blood cell count of the latter group of dogs started

* A stomach and liver concentrate fortified with ferrous iron.

somewhat later and was slightly less than in the two dogs which continued to receive Lextron. In the latter group, the average decrease of the red blood cell count after 130 calendar days of exposure was 3.1 million cells whereas in the group in which the dietary supplement was discontinued, the average decrease was only 2 million cells. The average increase in mean corpuscular volume was the same for both groups of dogs.

Anorexia was observed in all 5 cats after 30 hours of exposure. Despite the loss of appetite, however, only minor fluctuations in body weight occurred and there were no deaths. All of the dogs developed cough after 30 hours of exposure and one dog exhibited anorexia at this time. Two other dogs showed a loss of appetite after 48 hours exposure. Improvement of all of the dogs' appetite was observed beginning after 22 calendar days (86 hours). Death was preceded by marked weight loss associated with anorexia, nasal hemorrhage and a hacking cough in the case of both dogs that died. Of the two dogs that continued to receive Lextron, #1518 gained in weight steadily throughout the period of exposure, while #1437 showed mark weight loss between the 8th and 25th calendar days followed by partial recovery. The two dogs whose Lextron supplement was discontinued showed only minor fluctuations in body weight. There were no deaths among the ten rabbits and all gained in weight slowly but steadily during the period of exposure. The rabbits and cats were sacrificed terminally and tissue samples were obtained both for chemical analysis and for histologic examination. The surviving rats are scheduled to be sacrificed serially to provide tissue samples for chemical and histologic examination.

Blood samples were obtained once each week from each of the six dogs, and were analyzed to determine serum protein and plasma fibrinogen concentrations, the A/G ratio, serum Ca and P concentrations and the Ca/P ratio, serum dilution

turbidity and serum alkaline phosphatase concentrations. No clear-cut trends have been observed in any of these clinical chemical indices in the blood of the four dogs which are still undergoing exposure. However, a sharp increase in serum alkaline phosphatase concentration occurred on the 21st calendar day in the blood of the dog that died on the 30th calendar day of exposure. Concomitantly, this same dog exhibited lowered serum Ca and P concentrations and an increase of Ca/P ratio due to greater lowering of serum P than serum Ca concentration. A trend toward decreasing serum albumin concentrations and increasing serum globulin concentrations, with consequent decrease in A/G ratio, was observed in the blood of the dog that died on the 71st calendar day of exposure. This same dog exhibited an initial decrease in Ca/P ratio, due to a marked decrease in serum Ca and a slight increase in serum P concentration, followed by prompt recovery. No trends were observed in other clinical chemical indices in the blood of either of these dogs.

Problem Code: Be.4 (Fate)

Section Code: 3210

Authors: H. E. Stokinger, R. E. Root and L. T. Steadman

Retention of Beryllium Tissues Following Inhalation. Considerable information on the rate of accumulation, distribution and retention of beryllium in the animal body has been obtained in ancillary studies performed during the past two years' work on the inhalation toxicity of beryllium compounds. This information contributes much to the understanding of the mechanism of beryllium poisoning. Out of the results, a hypothesis has been derived relating susceptibility to beryllium poisoning to the rate of Be elimination from the body.

In the course of exposure studies, deposition data on beryllium have included analyses of 16 tissues from 7 animal species that have inhaled dusts of the insoluble BeO mists of the soluble sulfate. This ad interim review presents information on 1) deposition and distribution of beryllium in the body, 2) accumulation and retention of Be, and 3) elimination and biologic half-time of Be in the lung and bone. Tissues for analysis were taken with due regard for possible contamination by performing autopsies a few days after exposure and by suitable cleansing and changing of instruments during autopsy. All analyses were performed by the spectrographic method and tissue samples prepared for analysis by an extraction procedure developed by Steadman (1).

Results. Deposition was confined chiefly to 5 sites in the animal body - lung, pulmonary lymph nodes, liver, bone and kidneys, in order of decreasing importance. Table 1 (Page 43) shows the mean amounts and range of beryllium deposition in 16 tissues of dogs exposed to 10 mg/m^3 of beryllium sulfate for 95 days and to the same concentration of the oxide for 40 days. It is seen that minute amounts of beryllium were deposited in other sites including the ductless glands; values in these sites ranged from 0-10% of those in the 5 principal sites.

The order of beryllium deposition in the several tissues was in general maintained in all species of animals exposed to the beryllium sulfate (Table 2, Page 44). Less distinction in this regard was noted with the insoluble BeO where the lung acts as a beryllium "reservoir" with only a minute fraction (1/200-1/3000 of that in the lung) transported from this site to the liver, kidney and bone (Table 2, Page 44).

A notable difference was seen in the distribution of beryllium in the various tissues of dogs at different exposure levels (Table 3, Page 45). At

Table 1. Deposition of Beryllium in Tissues of Dogs Exposed to Beryllium Dusts and Mists

Results expressed in μg Be/g fresh tissue

Tissue	$BeSO_4 \cdot 6H_2O$ 10 mg/m³ for 95 days		BeO 10 mg/m³ for 140 days	
	Mean	Range	Mean	Range
Lung	4.0	1.7 - 7.0	39.4	18.7 - 60
P.L.N.	1.99	0.7 - 4.3	156.3	32.6 - 280
Liver	1.8	1.5 - 2.2	0.24	0.14- 0.34
Bone	0.8	0.3 - 1.2	0.24	0.15- 0.32
Kidney	0.8	0.5 - 0.9	0.09	0.05- 0.12
Tooth	1.4	1.1 - 2.2		
Intestine	0.06	0.03- 0.1		
Gall Bladder	0.06	0.03- 0.1		
Spleen	0.04	0.03- 0.1	0.06	0.05- 0.07
Pancreas	0.03	0 - 0.04		
Heart	0.03	0.01- 0.5		
Stomach	0.3	0.04- 0.90		
Eye	0.02	0 - 0.03		
Gonad	-	-	0.015	0.01- 0.02
Thyroid	-	-	0.05	0.02- 0.08
Pituitary			0.005	0 - 0.01

Table 2. Showing Order of Deposition of Beryllium in Tissues From Inhalation of its Salts

Results expressed as µg Be/g fresh tissue

	Monkey	Cat	Dog	Rabbit	Dog	Dog	Rat	G.P.	Monkey
Concn. mg/m³	1	1	1	1	3.6	10	10	10	10
C.T. mg hr/m³	426	426	426	426	920	4,430	4,430	4,430	4,430
				$BeSO_4 \cdot 6H_2O$					
Lung	1.2	0.08	0.6	1.6	2.6	4	11	5.5	1.4
P.L.N.	1.3	-	0.7	-	-	2	-	-	-
Liver	0.5	0.02	0.006	0.004	0.5	1.8	-	-	-
Bone	0.1	0.03	0.03	0.02	0.8	0.8	0.3	1.1	0.2
Kidney	0.01	0.006	0.003	0.003	0.1	0.8	-	-	-

	DOG			CAT	MONKEY
Concn. mg/m³	82	10.2	86.5	86.5	86.5
C.T. mg hr/m³	7,400	2,400	9,160	9,160	9,160
BeO		400°C		Fluorescent	
Lung	256	39	432	474	656
P.L.N.	206	156	676	163	611
Liver	0.5	0.2	0.2	0.3	0.2
Bone	0.4	0.2	0.3	0.2	0.04
Kidney	0.3	0.1	0.02	-	0.02

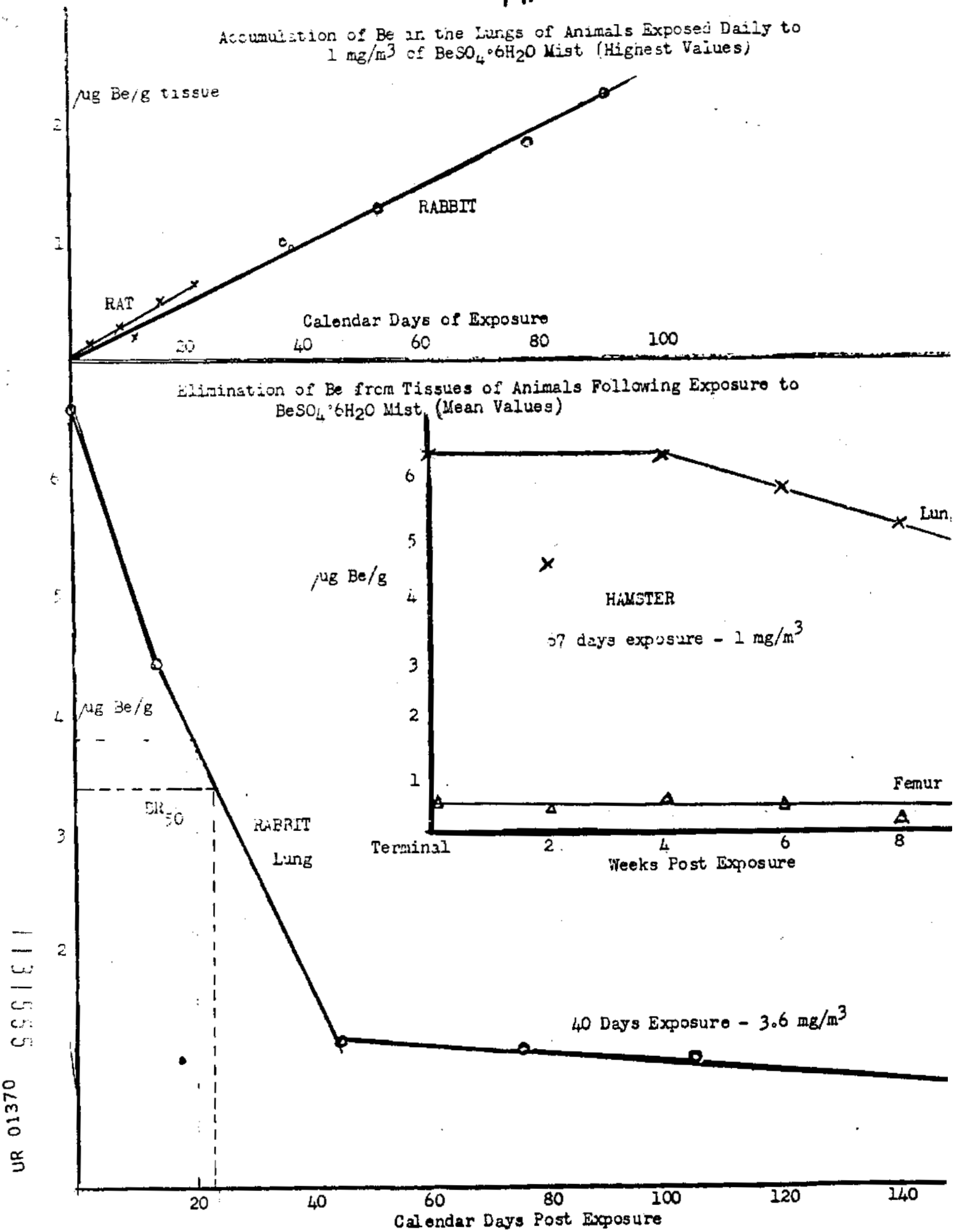
Table 3. The Comparison of Beryllium Content of Tissues of Dog at 1 and 10 mg/m^3 $BeSO_4 \cdot 6H_2O$

Tissue	5 Dogs 100 Days 1 mg/m3		Tissue	4 Dogs 95 Days 10 mg/m3	
	µg Be/g	%		µg Be/g	%
P.L.N.	0.7	115	Lung	4	100
Lung	0.6	100	P.L.N.	2	50
Femur	0.03	5	Liver	1.8	45
Spleen	0.008	1.3	Femur	0.8	20
Liver	0.006	1	Kidney	0.8	20
Kidney	0.003	0.5	Spleen	0.04	1

1 mg/m^3 but 1% of the beryllium found in the lung (taken as 100%) was found in the liver; at 10 mg/m^3, 45%, or 45 times that found at the lower level on a relative basis. Thus, it is noted that a different mode of transport must come into play at higher levels of exposure. This has tentatively been interpreted by Scott, Neuman and Allen (2) that at higher levels beryllium is transported in the colloidal form, as indicated by the greater retention of beryllium by the reticuloendothelial tissues, after the minute solubility product of the beryllium complex in the blood has been exceeded.

Accumulation and Retention. It is of greatest importance to determine the rate of accumulation of a toxic element, for this supplies needed information on the relation between response and exposure concentration. Moreover, the lowest level at which accumulation occurs - the 'threshold level of exposure' - is inherently important since this is the concentration below which negligible toxic response may be expected and above which toxic effects are almost certain to be found. Furthermore, from the knowledge of the exposure concentration, the toxic response and the degree of accumulation, the maximal quantity of the toxic element (beryllium) capable of being eliminated by the body during exposure may be obtained.

Some preliminary data on the rate of accumulation of beryllium in the lung and bone in rats and rabbits exposed to beryllium sulfate mist at 1 mg/m^3 have been obtained. Data on rats include a period of 21 days; on rabbits, for 90 days. Small groups of animals, two to five of each species, were killed at periodic intervals during daily, 6-hour exposures and the tissues were analyzed spectrographically. Results of a μg Be/g fresh tissue basis have been plotted in Figure 1 (Page 47) for the two species for the lung only. Plotted values represent the highest accumulated value in the lung obtained in each group at


Accumulation of Be in the Lungs of Animals Exposed Daily to 1 mg/m3 of BeSO4·6H2O Mist (Highest Values)
/ug Be/g tissue
RABBIT
RAT
Calendar Days of Exposure
20
40
60
80
100
Elimination of Be from Tissues of Animals Following Exposure to BeSO4·6H2O Mist (Mean Values)
/ug Be/g
HAMSTER
57 days exposure - 1 mg/m3
Lun
Femur
Terminal
2
4
6
8
Weeks Post Exposure
/ug Be/g
ER50
RABBIT
Lung
40 Days Exposure - 3.6 mg/m3
20
40
60
80
100
120
140
Calendar Days Post Exposure

the period of sacrifice. It is seen that even at the comparatively low level of exposure of 1 mg/m^3 (40 μg Be/m^3) both plots are linear over the time examined. The curve for rats had a slope of 0.78, that for the rabbit data somewhat less, 0.63. Averaged values (not shown) of beryllium content in the lung of rabbits gave evidence only at the 77th and 91st days that the rate of accumulation may be decreasing from daily exposure at this concentration. The femurs of these rabbits, the only other important site where accumulation occurs, showed a continued increase during the same period from a mean of 0.008 μg Be/g at day 10 to 0.03 μg/g at day 91, a four-fold increase compared with an 11-fold increase in the lung over this same period. Rats did not show appreciable amounts of beryllium in the femur at any time during the 21-day period of investigation.

A two-fold greater retention of BeO (400°C) was found in the lungs of dogs at 82 mg/m^3 than at 10 mg/m^3 than would be expected on the basis of CT values. CT values were 7,400 and 2,400 mg hr/m^3, resp., a three-fold difference, whereas the beryllium lung retention was respectively 256 and 39 μg/g, a six-fold difference. Beryllium retention in the lung of animals exposed to the sulfate however, showed values more generally in agreement with those expected from CT values.

Elimination and Half-Biologic-Retention Time (BR_{50}). Another factor of utmost importance in permitting an estimate of long-term effects from inhalation of a toxic element, is the rate of elimination of that element from the body from which may be calculated the BR_{50}. Information of preliminary sort is available on these factors in the rabbit, hamster and dog.

Rabbit. Members of this species, exposed to the soluble sulfate mist at 3.6 mg/m^3 for 40 days, were killed at regular intervals for 165 days after the termination of exposure. The average lung contents of beryllium per gram of

fresh tissue are plotted in Figure 2 (Page 47). The plot takes a form indicating rapid elimination with only slight change in slope for the first 45 days at which time less than 20% remains at that at the end of exposure. The BR_{50} value is shown to be somewhat more than 3 weeks (23 days). At 165 days (5.5 months) post-exposure, 10% on the average still remained, the rate of elimination having changed markedly after the 45th day. This may indicate that beryllium has now entered into a more stable tissue complex and thus is more permanently fixed in situ. No such evidence for the elimination of beryllium from bone in these animals was found; indeed, trends appear to show an increase at the 165th day.

Hamster. Similar plots of the elimination of beryllium from the lung and bone of this species showed evidence of an entirely different rate of elimination from that in the rabbit. Exposure of the hamster, however, was for a long period, 67 days, but to the same compound at 1 mg/m^3. The elimination was at least five times slower from the hamster lung than from the rabbit lung. The plot of the mean lung values is shown in Figure 2 (Page 47). An approximate calculation places BR_{50} at 14 weeks in the lung of this species. Inasmuch as inappreciable change occurred in the beryllium content of the femur of the hamster, no statement may be made as to the rate of elimination from this site over the 10-week period of investigation.

Dog. As yet no evidence of appreciable elimination of inhaled BeO from the lung and femur of this species has been obtained in a period greater than 1 year. Only a few animals have been examined.

Conclusions. Inhaled beryllium is deposited chiefly in the lungs and accumulates chiefly in four other tissues, namely, pulmonary lymph nodes, liver, bone and kidneys, in general in order of decreasing importance as named. The order of deposition was maintained in all species exposed. Less distinction in

this regard, however, was noted with the insoluble BeO where the lung acted as a reservoir and only minute amounts were transported to the other tissues. Absolute amounts, however, differed strikingly depending upon the level of exposure. At higher levels far greater amounts were found in the reticuloendothelial tissues indicating a colloidal method of transport at these levels not found at ten-fold lower levels (1 mg/m^3). Accumulation of beryllium appeared to be linear with time in the lung of both rats and rabbits in daily six-hour exposures at concentrations of 40 µg Be/m^3 up to 91 days for rabbits, the longest period studied. During the time that an 11-fold increase of beryllium occurred in the lung of the rabbit, a 4-fold increase in deposition occurred in the femur.

Elimination of beryllium was observed to vary so markedly among species that it is tempting to suggest that it is precisely this difference in capacity

Problem Code: Be.4 (Fate)

Section Code: 3220

Authors: W. F. Neuman, M. W. Neuman, B. J. Mulryan

Prior to work on the mechanism of beryllium's action in the skeletal system, it has been necessary to reinvestigate technics for the in vitro study of bone. The results obtained have stressed the dynamic nature of the inorganic phase of bone. It has been possible to demonstrate four separate processes which take place whenever powdered bone is in contact with buffered solutions. These processes are: lattice growth, recrystallization, adsorption and ionic exchange. Since these four variables are always present, the design of experiments is both important and difficult. Thus, the study of the mechanism of beryllium's action opens a field of fundamental importance which should prove useful in understanding the fixation in and effects on the skeleton of a number of project materials.

Problem Code: Be.5 (Mechanism of Toxic Effect)

Section Code: 3210

Author: C. J. Spiegl

Changes in Urinary Uric Acid:Creatinine Ratios as an Index of Be Poisoning.

In a previous experiment, an incidental observation disclosed that the ratio of uric acid to creatinine in the urine of dogs became elevated upon exposure of the animals to an atmosphere containing BeF_2. The changes induced were of a magnitude such that this index promised to be a criterion of beryllium poisoning. Consequently, the urinary uric acid and creatinine concentrations were investigated in more detail.

A. Control determinations of the ratio of uric acid to creatinine in the urine of 2 dogs gave values of from 7 to 10; this range agrees with that subsequently obtained with other animals. After 6 days of exposure by inhalation to 1 mg/m^3 of BeF_2, the ratio was increased by 75% and 55%, respectively. Elevated ratios persisted throughout the first 14 days of exposure; on the 14th day, a ratio greater than 460% of the control level was noted. At this time, the animal showing the highest ratio died and the other was temporarily removed from further exposure. After one week of nonexposure, the surviving dog again had a normal ratio of uric acid to creatinine in the urine. Re-exposure caused a 45% increase in the ratio in 6 days and has caused it to remain between 180% and 210% of the control value during the ensuing 51 days of exposure to BeF_2 (Table 1 below).

TABLE 1

URINARY URIC ACID/CREATININE RATIOS IN DOGS EXPOSED TO 1 mg/m^3 OF BeF_2

	Dog #1	Dog #2
Control Value	8.6	7.1
Days of Exposure		
6	14.9	11.0
8	13.8	11.1
12	17.1, 17.3	39.6
14	26.6	40*
One week without exposure	7.8	
Days of Re-exposure		
6	12.5	
9	17.7	
13	15.5	
20	16.3	
36	17.9	
57	16.7	

*This animal died after 14 days of exposure

B. Control ratios of urinary uric acid/creatinine obtained for 4 dogs in a total of 12 samples taken over a period of two weeks showed values ranging from 6.2 to 10.4 with a mean of 8.1 ± 1.3. After the dogs were exposed to 1 mg/m^3 BeF_2 for only 3 days, all animals gave uric acid/creatinine outside the range of normal values previously obtained with a mean elevation to 16.1 ± 2.7. After 10 days of exposure, the ratios had decreased somewhat but were all still considerably elevated from the normal (Table 2 below). Because this experiment is still in its early stages, however, no further conclusions can be drawn at this time.

TABLE 2

URINARY URIC ACID/CREATININE RATIOS IN DOGS EXPOSED TO 1 mg/m^3 OF BeF_2

	Dog #1	Dog #2	Dog #3	Dog #4
Control Values	7.4 8.2 9.2	8.3 7.0 6.5 6.2 6.8	10.4 7.0	9.7 10.1 4.6* 21.4*
Days of Exposure				
3	11.5	15.2	19.0	18.9
10	-	13.8	15.0	12.3

*These values are subject to doubt because of dilution and contamination of urine.

C. The urine of one dog undergoing exposure to 1 mg/m^3 of BeF_2 was sampled 9 times over a period of 10 weeks and gave ratios of urinary uric acid/creatinine between 10.1 and 21.0 with a mean of 16.0 ± 3.0. Although no control determinations are available for this particular animal, the mean of the values found during exposure to BeF_2 was 100% above the usual normal mean.

Discussion. In the experimental toxicology of beryllium compounds, a reliable clinical index of toxicity has long been desired. The ratio of urinary uric acid to creatinine may be such an index. Normally, both uric acid and creatinine concentrations vary greatly with the amount of water consumed; but the constant rate at which creatinine is excreted, regardless of urinary concentration, permits the use of the ratio of uric acid to creatinine. Changes in such a ratio, therefore, will not appreciably reflect urine dilution or concentration but a true metabolic alteration.

In the current experiments, significant increases were found in the ratio of uric acid to creatinine in dogs exposed by inhalation to 1 mg/m^3 of BeF_2. All 6 dogs in two experiments showed the elevated ratios within 3 to 6 days and maintained these high levels throughout the exposure period. It is further believed that this clinical manifestation is caused by the BeF_2 being inhaled, because one exposed dog, showing an elevated ratio, returned to normal within 6 days after removal from the BeF_2 exposure and again showed an elevated ratio within 6 days of re-exposure. Thus, the data suggest that the ratio of urinary uric acid to creatinine may be an index of beryllium poisoning. On the other hand, no information is at present available to show whether the causative agent is Be^{++}, BeF_2, or F^-, nor is the mechanism by which this change is produced understood. Furthermore, it would be desirable to have considerably more data on normal urines for comparison with experimental values. The acceptance of the uric acid to creatinine ratio as a clinical sign of beryllium poisoning must await further study.

PROGRAM Th.

THORIUM

Problem Code: Th.3 (Toxic Effects)

Section Code: 3210

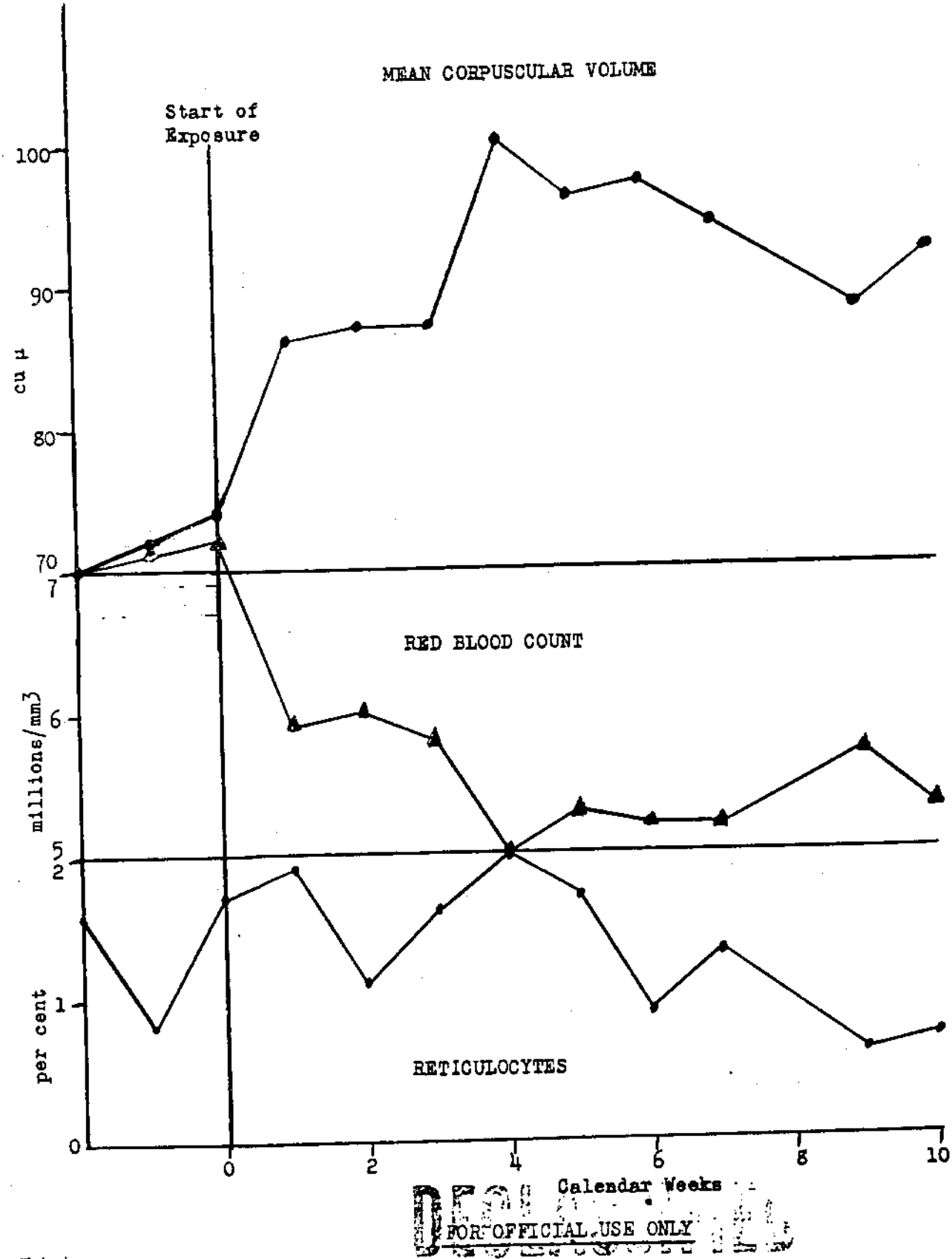
Authors: R. H. Hall, R. E. Root, and C. Stroud

Inhalation Toxicity of Thorium Fluoride. As a part of a program to define the type and degree of response to the inhalation of various industrially important dusts of thorium compounds used in the production of atomic energy, a short-term high level exposure, to an insoluble compound has been carried out.

During a 10-week period of exposure to ThF_4 dust by inhalation, the sole evidence of toxic effects observed in four female dogs was certain hematologic changes, including the development of a macrocytic anemia. The animals were exposed 6 hours daily, five days per week, to a concentration of approximately 15 mg/m^3. Beginning three weeks before the start of the exposure period, and continuing until the end, the dogs received a dietary supplement of one teaspoonful of lextron (a liver extract and iron concentrate) daily. Complete cell counts, hemoglobin concentrations, hematocrit values and coagulation times were determined upon blood samples obtained from each of the dogs once each week preceding and during exposure.

The red blood cell counts and hemoglobin concentration in the peripheral blood of all four dogs was decreased sharply after the first week of exposure. The average decrease in the RBC count was over a million cells per cu. min. At the same time, the mean corpuscular volume (MCV) was increased from an average value of 72 cu.μ. to 87 cu.μ. The maximal decrease in the RBC counts of three of the dogs occurred after 4 weeks exposure, as shown in Figure 1 (Page 56),

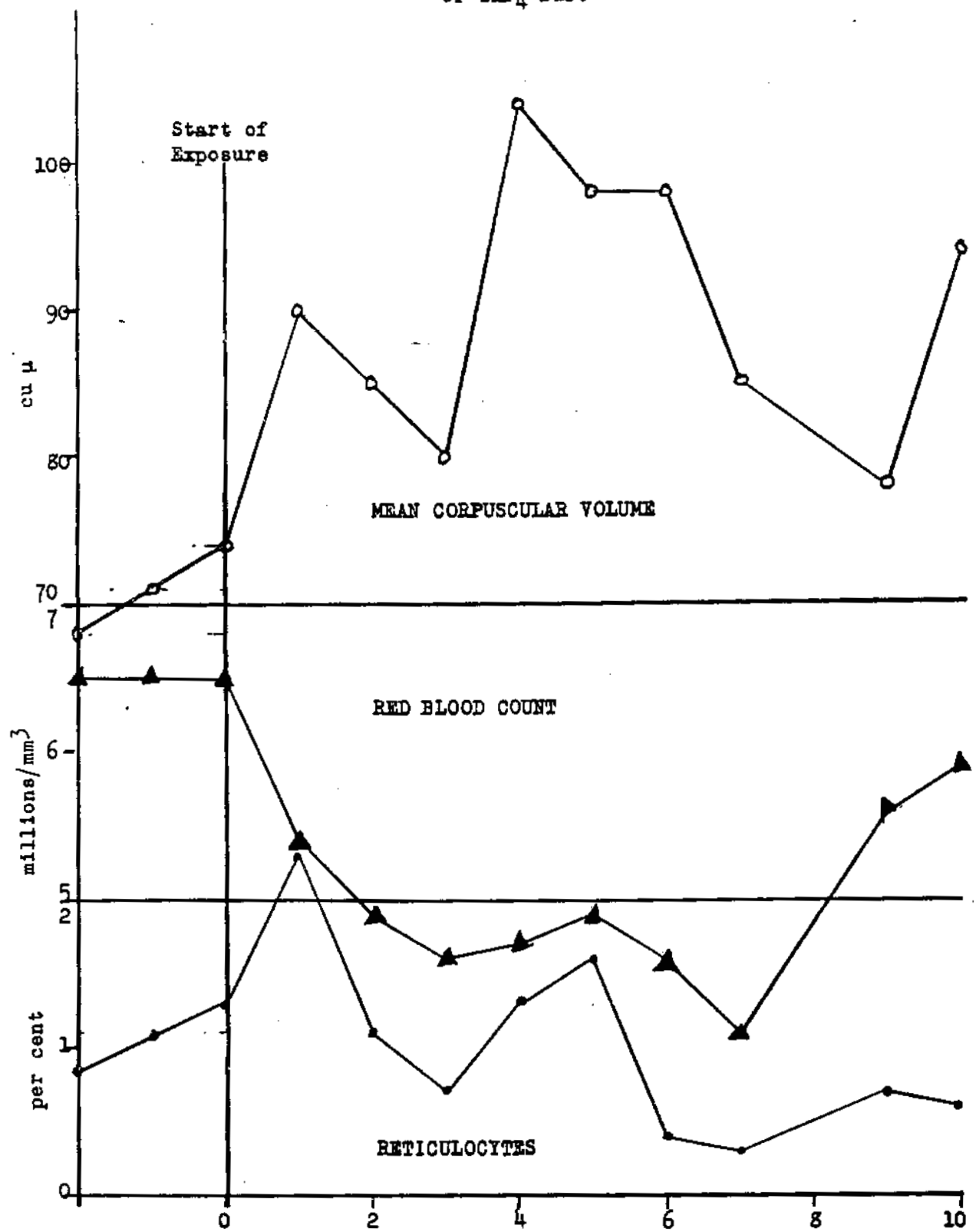
Figure 1. Mean Hematologic Changes in the Peripheral Blood of Dogs During Exposure to a Concentration of Approximately 15 mg/m^3 of ThF_4 Dust



after which the counts increased slightly. The changes in the RBC counts of these dogs were accompanied by an increase in MCV to an average value of 100 cu.μ. after 4 weeks' exposure, after which there was a slight decrease. There was a general trend toward a decrease in the reticulocyte counts of these dogs, but, as shown in Figure 1 (Page 56), the average values recorded weekly fluctuated in a pattern closely paralleling the changes in MCV. The fourth dog exhibited a maximal decrease of 2.4 million red blood cells after 7 weeks exposure. This represents a slightly greater change than occurred in the RBC counts of the other three dogs. As shown in Figure 2 (Page 58), the RBC counts of this fourth dog rose sharply after the 9th and 10th weeks of exposure. The mean corpuscular volume increased to 104 cu.μ. after 4 weeks exposure, then decreased steadily to 78 cu.μ. after the 9th week, only to rise sharply again after the 10th week of exposure. In this animal, likewise, there was a trend toward a decrease in the reticulocyte count during exposure, and, again, the fluctuations in this index tended to parallel those in MCV. In all four dogs the changes in hemoglobin concentration of the blood were such that the mean corpuscular concentration nearly always remained within normal limits throughout exposure.

The leukocyte counts of three of the dogs fluctuated in a normal manner during the exposure; however, these same three dogs exhibited a marked shift in the relative proportions of mature and immature polymorphonuclear neutrophils in their peripheral blood. With continued exposure there was a steady decrease in the percentage of mature, filamented granulocytes and a concomitant increase in the percentage of immature nonfilamented cells. This shift was not observed in the proportion of filamented and nonfilamented polymorphonuclear leukocytes in the blood of one dog which did, however, exhibit an initial decrease in its total WBC count. In this dog, the total leukocyte count decreased from an average of

Figure 2. Hematologic Changes in the Peripheral Blood of Dog #1561 During Exposure to a Concentration of Approximately 15 mg/m^3 of ThF_4 Dust



8,200 during the pre-exposure period of observation to a minimum of 4,300 cells per cu. mm. after 4 weeks exposure. There was perhaps some tendency for the WBC count to return to its pre-exposure level after the 4th week of exposure. The changes in the leukocyte counts of the dogs are summarized in Table 1 below.

All of the dogs gained in weight steadily during the period of exposure. Anorexia was not observed and only minimal signs of respiratory tract irritation were seen. There were no significant changes in the concentrations or distribution of serum proteins in the blood of any of the dogs.

TABLE 1

SUMMARY OF CHANGES IN THE LEUKOCYTE COUNTS IN THE PERIPHERAL BLOOD OF DOGS EXPOSED 10 WEEKS TO 15 mg/m^3 OF ThF_4

Number of Weeks Exposure	Average - Dogs #1454, 1543 and 1561			Dog #1533		
	Leukocyte Count x 10^{-3}	Neutrophils (%)		Leukocyte Count x 10^{-3}	Neutrophils (%)	
		F	NF		F	NF
-3	9.6	38	25	8.2	40	20
-2	10.2	27	33	8.7	27	29
-1	9.5	29	36	7.8	33	31
1	9.0	31	37	5.7	32	25
2	8.6	22	39	5.1	28	21
3	10.8	29	38	4.7	33	23
4	8.1	26	37	4.3	35	21
5	8.8	22	43	5.8	30	20
6	7.9	20	42	5.6	23	32
7	8.1	18	46	5.7	20	33
9	8.7	21	44	4.5	24	31
10	8.3	16	50	6.0	25	36

PROGRAM F.

FLUORIDE

Problem Code: F.3 (Toxic Limits)

Section Code: 3230

Authors: E. A. Maynard and W. L. Downs

Acute oral toxicity of NaF in albino rats. Single oral doses of sodium fluoride (NaF) in a concentration of 25 mg/ml of water were administered to a total of 98 mature female rats (body weights 174 to 225 g.). The calculated 24-hour LD50 was 80 mg/kg $\pm$ 5. The calculation was made with Winthrop probit-logarithm dose paper; the standard error was calculated following the method of Miller and Taintor (1944).

Problem Code: F.4 (Fate)

Section Code: 3210

Authors: F. A. Smith, D. E. Gardner and D. Wing

Fluoride Content in Human Bone. The customary procedure for the determination of fluoride in biologic material other than bone and teeth requires the preliminary destruction of organic matter by ashing the sample in the presence of a fluoride fixative such as calcium oxide. The ashing temperature routinely employed in different laboratories is approximately 575-600°C; at temperatures greater than 600°C appreciable quantities of the fluoride are lost due to the volatility of calcium fluoride at the elevated temperatures. To date, no reliable information has been available, however, regarding the effects of higher ashing temperatures upon the fluoride content of bones and teeth. Data of

this nature were required in order to permit an accurate estimate to be made of the fluoride content of a series of ashed samples obtained following the cremation, at approximately 950°C, of twenty cadavers. The tentative fluoride content of these samples was reported in a previous Quarterly Report (UR-70).

A large quantity of human ash was prepared at 575°C with the cooperation of Dr. John Hursh. Separate portions of this material were then ashed further at progressively increasing temperatures through 1000°C. A second series of samples was prepared independently by Dr. Hursh, using a different human ash as starting material. For both series, the samples prepared at temperatures higher than 575°C were ashed for two hour periods; this time interval corresponds to the total time required for the cremation of a cadaver. A series of control samples were prepared, in which known quantities of sodium fluoride were heated in the presence of calcium oxide for two-hour intervals at temperatures greater than 575°C. Fluoride analyses were then done on the various ashed samples and controls. The data are presented in Table 1 below.

TABLE 1

EFFECT OF ASHING TEMPERATURE ON THE DETERMINATION OF FLUORIDE IN HUMAN ASH AND SODIUM FLUORIDE

SODIUM FLUORIDE CONTROL			HUMAN BONE ASH			
			Series A		Series B	
Ashing Temp. °C.	µg F Added	% Recovered	Ashing Temp. °C.	PPM F in Ash	Ashing Temp. °C.	PPM F in Ash
575	100	97.9	400	1230		
850	100	83.7	500	1320	575	1410
925	100	75.4	600	1290	660	1460
			800	1350	800	1450
			1000	1330	1000	1460

The percentage recoveries obtained for the sodium fluoride controls show a progressive decrease as the ashing temperature is raised. Under these ashing conditions, the fluoride probably has been lost as volatile calcium fluoride. In contrast to these results, however, there is no loss of fluoride from human bone ash, even when the ashing temperature is increased to 1000°. The variations noted among these samples are well within the experimental error of the analytical procedure. The data demonstrate that no significant losses of fluoride in bone occur under the relatively severe ashing conditions of 1000°C for two hours. These results may be interpreted as indicating that little, if any, of the fluorid normally found in bone is present as simple calcium fluoride but must be combined as a more complex and stable salt of calcium (fluorohydroxyapatite).

Having established the fact that the process of cremation introduces no appreciable loss in fluoride content of the ash, it is now possible to correct the tentative figures reported previously for the fluoride content of total human ash (loc. cit.). The revised data are listed in Table 2 (Page 63).

The fluoride content of the total body ash ranges from 570 to 2360 ppm. These values are considered to be in excellent agreement with the values of 480-2100 ppm reported by Roholm (Fluorine Intoxication, p. 193, 1937) for the costae of eleven normal individuals 33-80 years of age.

TABLE 2

FLUORIDE CONTENT OF TOTAL HUMAN ASH (REVISED)

Sample No.	Sex	Age	Fluoride Content of Ash	
			PPM	Total (g)
1219	M	50	740	3.28
1228[a]	M	78	990	1.55
1230[b]	M	82	1490	3.86
1235[b,c]	M	55	1480	3.70
1237[a]	M	77	1190	2.29
1238[a]	M	77	2150	3.65
1253[b,c]	M	68	1020	2.55
1255[a]	M	57	2360	4.97
1260[a]	M	83	1510	3.31
1266[a]	M	---	1290	3.31
1267[a]	M	74	1490	3.36
1270[a]	M	74	720	1.11
1272[b,c]	M	54	660	1.65
1274[a]	M	60	300	1.51
1275[a]	M	75	330	2.39
1276[a]	M	66	1010	1.98
1278[a]	M	49	920	2.28
1281[d]	F	77	930	0.74
1283[b]	M	65	830	2.78
1294	F	85	570	0.85

a - Cadaver from Anatomy Department School of Medicine and Dentistry.

b - Ashes swept up from crematory floor; may contain small amounts of firebrick.

c - Estimated weight; ashes swept up from crematory floor.

d - Apparently this is not a complete sample.

UNCLASSIFIED

PROGRAM Zr.

ZIRCONIUM

Problem Code: Zr.1 (Physical and Chemical Properties)

Section Code: 3210

Authors: K. Lauterbach, J. Mitchell, S. Laskin

Particle-Size and Specific-Surface Measurements for Bulk Zirconium Compounds

The particle-size and specific-surface data in Table 1 (Page 65) have been determined for four zirconium compound bulk samples*. The samples show large differences in particle size as determined by optical microscope measurements, varying from a mass-median size of 2.4 μ for the pure $ZrCl_4$ to a value of 9.0 μ for the mass size of the zirconium cyanonitride. Since this latter value was so much higher than those of the other samples, the optical microscope measurement for cyanonitride was repeated. In this case a value of 8.8 μ was obtained for the mass median size, thus checking the original determination. Electron micrographs of these samples show no unusual structure and in general confirm the particle-size measurements by the optical microscope.

Specific-surface determinations by low-temperature adsorption of ethane are reported for all samples except $ZrCl_4$. The surface value of this compound could not be determined by the adsorption method because it decomposed under the high vacuum degassing treatment which is required prior to ethane adsorption. The specific-surface values of the remaining three zirconium compounds are in close agreement. It might be expected that the surface value for the Zr cyanonitr sample would be extremely low due to the large particle size, however, this sample

* Obtained from the Titanium Alloy Division of the National Lead Company, Niagara Falls, New York

TABLE 1

SPECIFIC SURFACE AND RELATED DATA FOR BULK ZIRCONIUM COMPOUNDS(a)

Sample Description	Particle Size(c)				Specific Surface, M^2/g		Porosity
	Mass Median, μ	Geo. Stand Dev. of Mass Median	Count Median, μ	Surface Median, μ	Calc. From Particle Diam.	By Ethane Adsorption	
TAM $ZrCl_4$, Z-135	2.4	2.1	0.38	1.4	0.29	(b)	
TAM ZrO_2, C, P, Z-135	3.5	1.9	0.90	2.3	0.13	3.8	29
TAM Superpax $ZrO.SiO_2$ Fines	3.0	1.9	0.73	2.0	0.20	3.2	16
Dust From Bag Filter(d)	9.0	2.2	0.86	5.7	0.047	5.2	111

(a) Samples obtained from Titanium Alloy Mfg. Div., National Lead Co., Niagara Falls, N. Y.

(b) This sample was found to decompose under the high vacuum degassing required prior to ethane adsorption.

(c) Measurements by optical microscope.

(d) Air from zirconium cyanonitride burning room.

shows the highest value of the three, indicating a relatively high "internal" surface. A porosity value of 111 for this sample indicates an internal surface at least 4 times as great as those of the ZrO_2 and $ZrO{\cdot}SiO_2$ samples which show porosities of 29 and 16, respectively.

PROGRAM S.M.

SPECIAL MATERIALS

Problem Code: S.M.2 (Toxic Effects)

Section Code: 3210

Authors: Sidney Laskin, Paul Frank and Robert H. Wilson

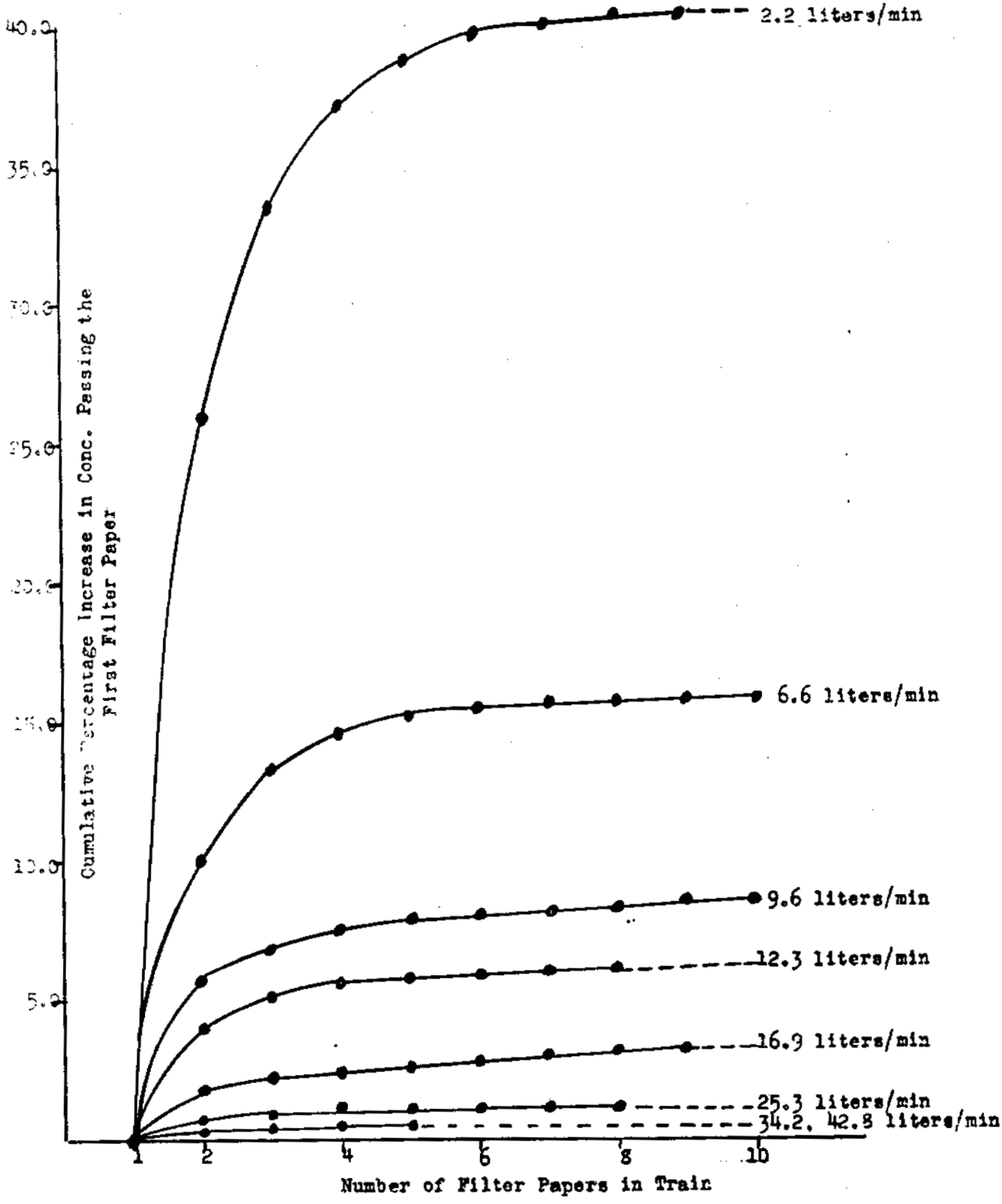
Efficiency of Sampling Methods for the Collection of Toxic Atmospheric Impurities.

Studies have continued on the investigation of the absolute efficiency of the filter paper dust sampler and the analysis of the factors determining its efficiency. Because of the complex nature of the problem discussed in previous reports, current studies have been limited to the effects of sampling velocity on the efficiency of Whatman #41 filter paper.

Fixed conditions were maintained with sodium chloride aerosol atmospheres dispersed and controlled as described in previous quarterly reports. Concentrations were analyzed by means of the flame photometer. Humidity was controlled to an average value of 82% to simulate a wet aerosol. Particle-size distributions were calculated from electron micrographs of samples collected with the oscillating thermal precipitator. The average particle size mass-median was 0.31 μ with a corresponding geometric standard deviation of 1.7.

A more detailed study of the multiple filter paper train as a method of determining absolute efficiency has been completed. These results are illustrated graphically in Figure 1 (Page 68) for a range of sampling velocities from 2.2 to 42.8 liters/min. An average concentration of sodium chloride aerosol of 29.8 mg/m^3 was maintained for all velocities. Trains of 8-10 filter paper elemen were used in these studies except for velocities above 30 liters/min where the resistance of the train became a limiting factor in obtaining the desired air

Figure 1. Effect of Increasing Number of Elements on the Collection Efficiency of a Filter Paper Sampling Train. Sodium Chloride Aerosol at an Average Concentration of 29.8 mg/m^3



rate. The concentrations collected on each of the papers of the train are expressed in terms of the cumulative percentage increase above the value of the first paper. In agreement with previously reported results, this family of curves illustrates an increase in efficiency with increasing elements of the filter-paper train and an increased efficiency obtained on the first filter paper with increased sampling velocity. All curves approach a limiting value asympotically which, on the basis of previous studies with the flame photometer, were assumed to be equivalent to the total amount of material passing the first paper. Parallelism of the curves and the agreement with previous results appear to justify this procedure. The number of elements required to obtain a value of absolute efficiency, however, is found to be related to the sampling velocity. Thus at 48.2 liters/min at which an efficiency of 99.6% was obtained for the first paper, only a minimum of 2 papers are required to obtain significant results This number increases with decrease in sampling velocity to a minimum of 7 at 2.2 liters/min where a sampling efficiency of 71.3% was obtained for the first paper. The previously reported use of the 6 paper train was shown to be satisfactory for the range of velocities explored in those studies (7.4-31.1 liters/min).

Studies of the effects of sampling velocity on the efficiency of Whatman #41 filter paper have also been completed at an average concentration of 31.0 mg/m^3 for a range of sampling velocities from 2.2 liters to 43.3 liters/min. The summarized results of these studies are shown in Table 1 (Page 70). In order to obtain statistically significant results, a total of 95 determinations for 8 sampling velocities were made. The mean efficiency results as a function of sampling velocity are illustrated graphically in Figure 2 (Page 71). The results indicate a rapid increase in efficiency with increase in sampling rate from

TABLE 1

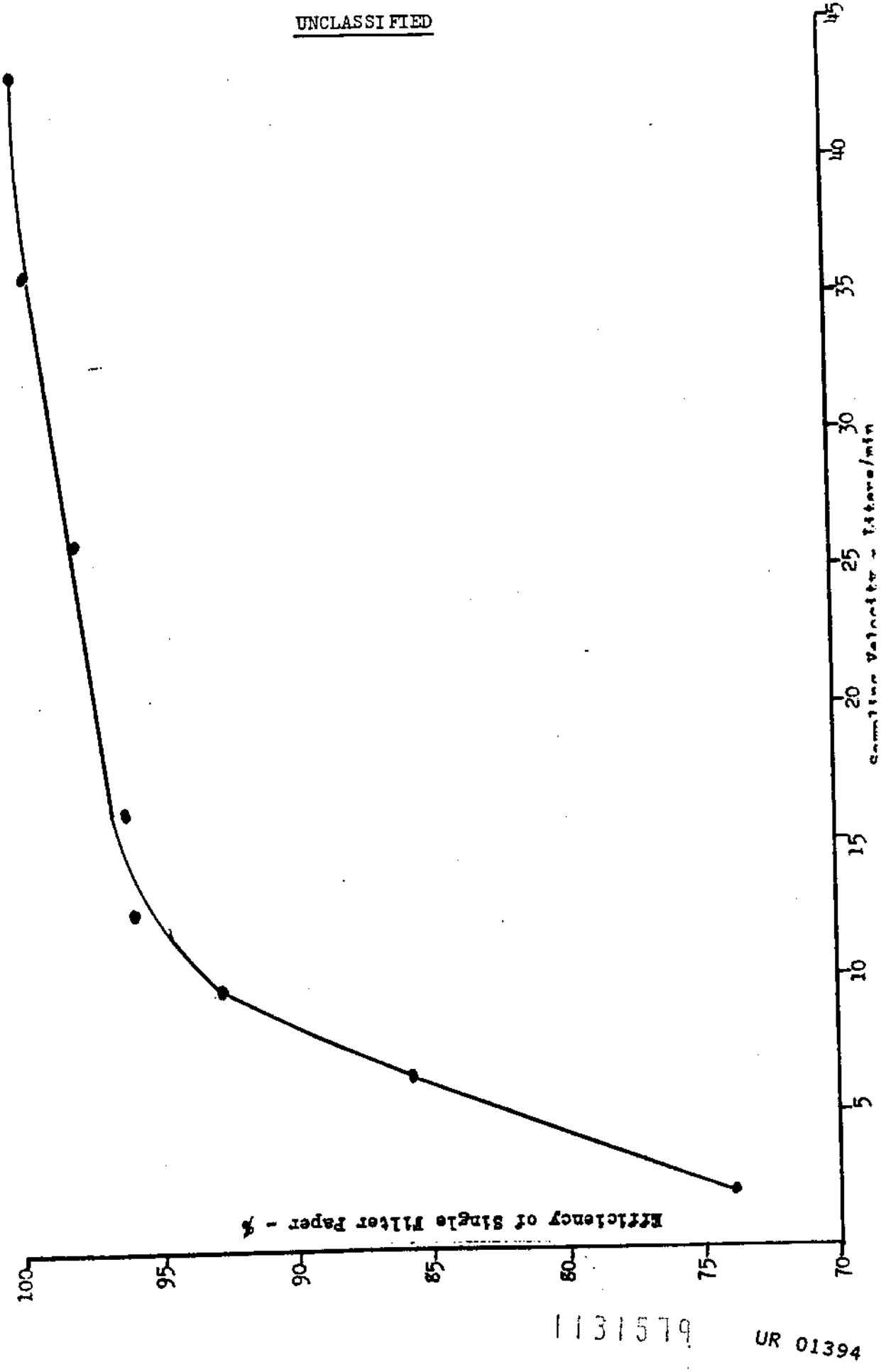
EFFICIENCY OF WHATMAN #41 FILTER PAPER AS A FUNCTION OF SAMPLING VELOCITY

NaCl AEROSOL AT AN AVERAGE CONCENTRATION OF 31.0 mg/m^3

Experiment No.	No. of Samples	Concentration		Sampling Velocity		% Retained by First Paper	
		Mean	Range	Mean	Range	Mean	Range
		MG NaCl/3		liters/min			
1	14	38.2	31.3-43.0	2.2	2.2- 2.3	73.3	69.9-76.3
2	11	26.9	22.4-29.2	6.4	5.6- 6.6	85.7	82.7-87.6
3	9	28.3	22.0-32.0	9.7	8.5-10.2	92.6	90.4-94.5
4	14	29.8	26.8-32.8	12.5	11.2-12.9	95.8	93.8-97.3
5	19	29.6	24.6-35.6	16.3	14.7-18.9	96.2	94.3-97.8
6	9	32.8	29.6-34.6	26.0	23.6-27.3	97.9	96.9-99.0
7	10	30.9	29.2-32.9	36.0	34.2-37.6	99.4	98.5-99.7
8	9	31.6	27.5-35.9	43.3	42.5-44.6	99.6	99.4-99.7

Figure 2. Effect of Sampling Velocity on the Efficiency of Whatman #41 Filter Paper

Sodium Chloride Aerosol at an Average Concentration of 31.0 mg/m^3



values of 2.2 liters/min up to 9.7 liters/min, mean efficiencies of 73.3 and 92.6% being found, respectively. With further increase in sampling rate, the efficiency increases more slowly, gradually approaching a 100% with the highest recorded value being 99.6% at 43.3 liters/min.

Problem Code: S.M.3 (Toxic Limits)

Section Code: 3210

Authors: S. Laskin, R. H. Wilson, K. E. Lauterbach, L. J. Leach, and D. W. Falcone*

Production Design of the Modified Cascade Impactor.

The experience of the Industrial Hygiene Section with the Modified Cascade Impactor has demonstrated its value as a practical field and laboratory instrument for sampling and characterizing atmospheric particle-size distributions (1,2,3,4). The design and results have aroused the interest of industrial hygiene and health physics groups resulting in numerous requests for standardized instruments. Interested groups include several areas under supervision of the Atomic Energy Commission and its subcontractors, the Army Chemical Center and several state industrial hygiene laboratories. On the basis of tentative requests for about 300 instruments, arrangements are being completed via the N. Y. Operations Office for the commercial production of the instruments by Mine Safety Appliances (M.S.A.), Pittsburgh, Pennsylvania.

To facilitate large-scale production by modern die-cast or machining methods, a new design of the Modified Cascade Impactor has been developed in cooperation with Donald W. Falconer of the Army Chemical Corps. The Assembly

* Biological Dept., Chemical Corps., M. Division, Camp Detrick, Frederick, Maryland

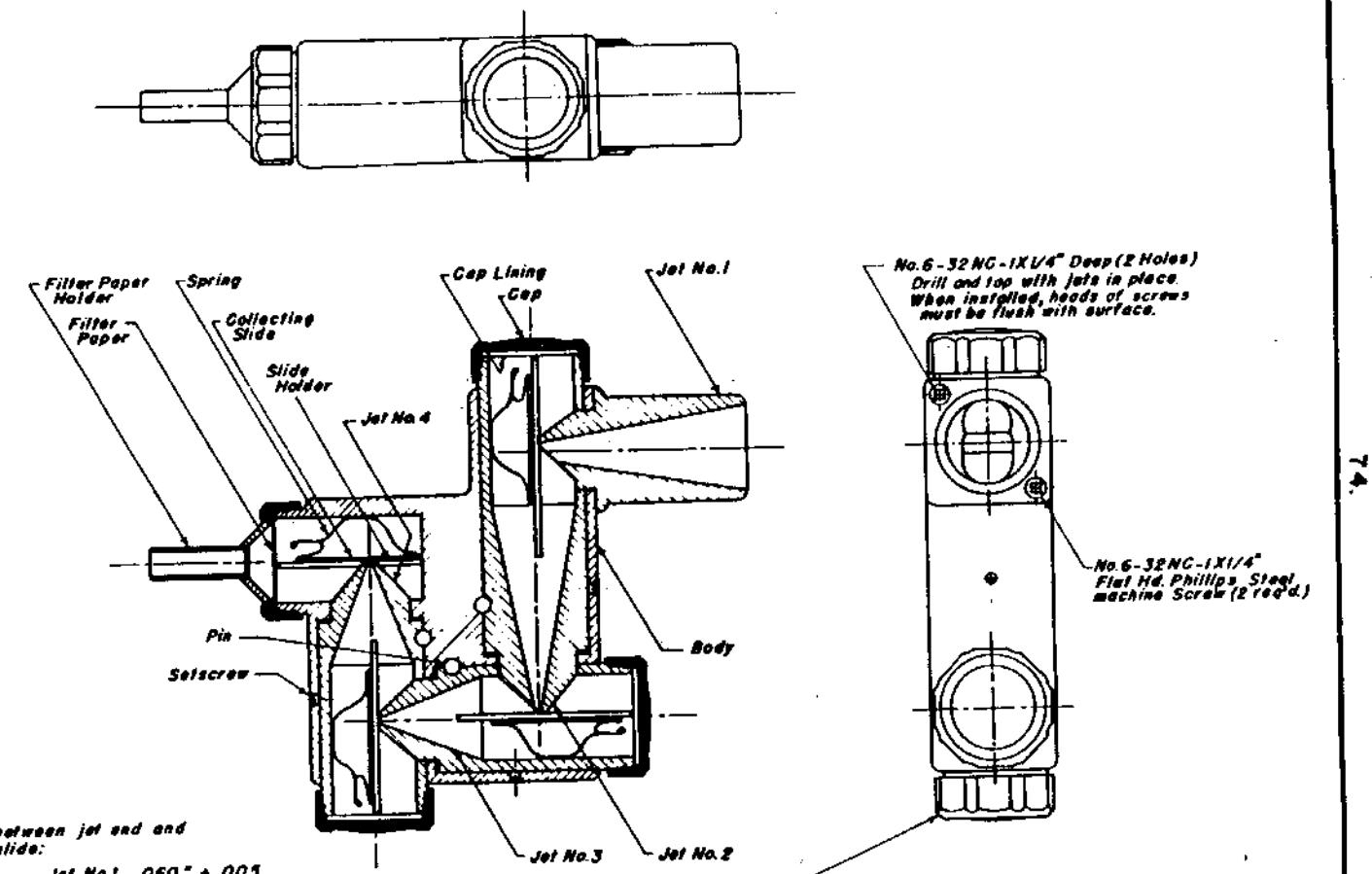
drawing for this instrument is illustrated in Figure 1 (Page 74). Detail construction drawings have been completed and tentatively approved by a M.S.A. representative (Mr. T. Connelly). Present status of progress is the experimental production of a shop model by M.S.A. to be completed in the near future. Approval of operating characteristics of this model will determine the date of production.

In the latest design, internal jets, orifice dimensions and jet-to-slide spacings have been held to their original specifications. Several revisions have been made in the assembly of the instrument which result in a stronger unit and a simplified assembly procedure. Each jet consists of a separate cylinder fitted into and held in position with respect to other stages by a main body piece. Internal surfaces will be smooth and without protruding edges. Construction will be of aluminum, magnesium or a light weight alloy. Other major revisions represent improvements based on our practical experience. These include such items as: (1) replaceable plastic caps requiring a minimum of effort to close the jet chamber; (2) a new type of spring to hold the collecting slide and facilitate handling of the samples; (3) provision of sufficient metal in the body of the instrument to provide for connection to mounting assembles.

Construction of this design to the rigid specifications should eliminate the need for complete calibration of the particle-size stage constants for each instrument. Standard values based on those obtained from the calibration of several master instruments will be furnished along with a comparative calibration for each instrument. Application of the instrument to most other dusts will then only require a simple recalculation according to the density of the material and sampling velocity. The calibration procedure and method of calculation for various dusts have been previously reported(2,4).

S. LADEIN K.E. LAUTERBACH
R.H. WILSON L.J. LEACH

FIG. 1



Clearance between jet end and collecting slide:

Jet No.1	.060" ± .005
Jet No.2	.022" ± .002
Jet No.3	.059" ± .002
Jet No.4	.024" ± .001

Caution In assembling jets no.1 and 2 care must be taken to insure proper positioning of 45° angle on orifice as shown. In assembling jets no.3 and 4 care must be taken to insure proper positioning of slide holders. These holders are not on center.

1 inch

CASCADE IMPACTOR ASSEMBLY

THE UNIVERSITY OF ROCHESTER
ATOMIC ENERGY PROJECT
DIVISION OF PHARMACOLOGY
SECTION OF INDUSTRIAL HYGIENE

DRAWN BY L.J.L.	DATE 8-29-49
CHECKED BY K.E.L. S.L.	DWG NO. DCCI-1

74.

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4. See also numerous Rochester Area Reports on Field and Laboratory Problems

Problem Code: S.M.5 (Mechansim of Toxic Effect)

Section Code: 3210

Authors: R. H. Wilson, S. Laskin, and D. W. Meier

A New Design for an Oscillating Thermal Precipitator.

On the basis of experience gained from former models of oscillating thermal precipitators, a new design for the instrument has been developed. A feature of this design is simplified construction, maintaining the desired characteristics for practical field use. The materials of construction are brass, steel and aluminum with external dimensions of 1" x 2" x 6". Better oscillation has been obtained through the use of a positive drive in both directions, accomplished by means of double-acting, ball-bearing roller followers on a heart-shaped cam. The basic construction elements are shown in Figure 1 (Page 76) which is an x-ray photograph of the instrument. The unit is powered with a Telechron type B-3, 1 rpm, 4 watt, synchronous motor directly coupled to the cam shaft.

Other design features include a simplified arrangement for sample preparation and an improved method for mounting electron microscope screens. Figure 2

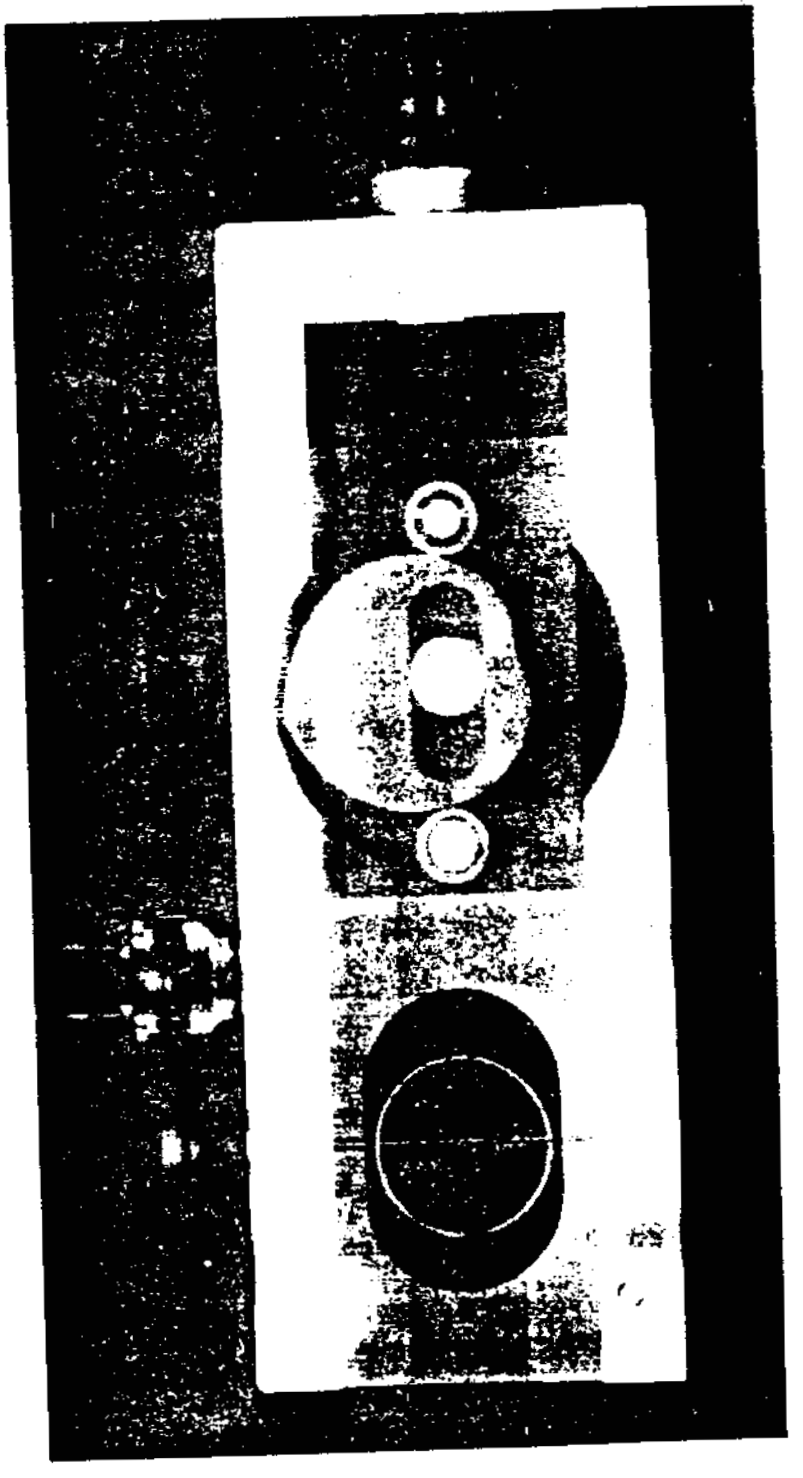

Figure 1. X-ray photograph of oscillating thermal precipitator showing drive mechanism.

(Page 77) shows the assembled instrument with sample holders for both electron and optical microscopy.

Double samples for comparative purposes are taken through the provision of two oscillating slides each of which carry a sample holder. Either or both types may be used in drawing a given sample. The collected sample is in the form of a rectangle measuring 5/8" x 1/2", the latter dimension being the length of stroke or oscillation. Twenty-two millimeter circular cover slips are used for the collection of samples for optical purposes. For electron microscopy the cover slip sample holders are replaced with screen holder plugs which place the screens in a position central with respect to the deposition area. Preliminary tests have shown satisfactory operation at collecting rates of 20 cc/min.

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PROGRAM I.S.

ISOTOPES

Problem Code: I.S.3 (Therapy)

Section Codes: 3310, 3340, 3350

Authors: Frank W. Furth, W. Burkett Mason, and J. Russell Hayes

In September of 1949 a cooperative program for the clinical use of radioisotopes was instituted between the Atomic Energy Project of the University of Rochester and the Genesee Hospital of Rochester, New York. Although the program was designed to employ any of the radioactive isotopes designated for diagnostic or therapeutic use, to date only radioactive iodine, I^{131}, has been used. Since September, 1949, twenty-one patients have received I^{131} for diagnosis or treatment of various types of thyroid disease. In conference with the clinicians in charge of the program at the Genesee Hospital, Drs. George Baron and Jacob Goldstein, a working arrangement for the program was designed. The principles of this arrangement are given below, together with a summary of the data on the patients treated through December 1949.

The selection of the patients to receive the I^{131} has been done mainly by Drs. Baron and Goldstein from patients referred by private physicians, or by the house staff of the Genesee Hospital. The patient's clinical history, physical examination, and laboratory work-up are carefully reviewed, and on the basis of this clinical appraisal the patient is accepted for radioiodine therapy. In general, all types of hyperthyroid and malignant thyroid disease, with one exception, have been accpeted. This one exception is the patient with a toxic nodular goiter who is at least a fair operative risk. Because of the higher incidence of thyroid malignancy arising in this type of goiter, operative

therapy would seem preferable, and has been recommended. Of the twenty-one patients accepted for therapy, nineteen had the clinical diagnosis of hyperthyroidism or suspected hyperthyroidism. Within this group there was a considerable variation in the degree of hyperthyroidism. Two patients have proven malignancies of the thyroid and will be discussed separately. The ages of the hyperthyroid patients ranged from 34 years to 76 years with an average of 57 years. Since a large percentage of patients were in the older age group the incidence of the degenerative diseases was appreciable and the presence of these associated conditions tended to make the clinical diagnosis of hyperthyroidism difficult to establish. Hypertensive cardiovascular disease and arteriosclerotic heart disease were the most commonly associated clinical syndromes, occurring in a mild to severe form in nine of the patients. Because of the high incidence of these complications at least 50% of the patients were poor operative risks. No patient with markedly diminished renal function, nor any patient with malignant exophthalmus was chosen for therapy. Approximately 60% of the patients had received antithyroid therapy with the thiouracils and/or iodine at sometime previous to the administration of the I^{131}, but it had been either totally ineffective or only temporary in its effect. An effort was made to see that no patient received any thiouracil derivative or iodine preparation during the month preceding radioiodine therapy. Three of the patients had previously had partial thyroidectomies with return of typical symptoms of hyperthyroidism at some time after operation.

Arrangements were made for each patient to have a certain minimum of laboratory work, such as blood counts, urinalysis, BMR, etc., before the I^{131} was given. In some patients a more extensive laboratory work-up was done including creatine and galactose tolerance tests. The details of these laboratory

findings and their correlation with the excretion of the I^{131} will be reported later.

As a general procedure, it was agreed that a therapeutic dose of radioiodine should be preceeded by a trace dose of approximately 50 microcuries, with determination of the urinary excretion during the first 48 hours following administration. Fourteen of the patients have received such a trace dose, and of these, four were subsequently given therapy doses. Five patients have received therapy doses without having been given previous trace doses. These exceptions to the general rule were for two reasons, the clinical picture of thyrotoxicosis was obvious, and/or the seriousness of the patient's condition demanded urgent therapy. For the purposes of interpreting the data on the excretion of the I^{131}, it was decided that if the patient excreted less than 55-60% of the administered dose in 48 hours, he would be considered hyperthyroid. The average excretion of the fourteen patients who received trace doses was 38%, with a range from 13% to 70%. Eleven patients had values less than 55%. The patient who excreted 70% of the trace dose had previously had two partial thyroidectomies and now has moderately severe hypertensive cardiovascular disease. A summary of the data on the patients given trace and therapy doses is shown in Table 1 (Pages 82 and 83).

Therapy doses of I^{131} were given to a total of nine patients. One of these patients has received two therapy doses. As noted above, only four of these patients had a prior trace dose. The excretion of the trace dose was not above 30% in any of these four patients, but in each case the percent excretion of the therapy dose was higher than the percent excretion of the trace dose. The highest percent excretion in these four patients was 42%, while the highest percent excretion in those five patients who did not receive a trace dose was

TABLE 1

Patient	Clinical Diagnosis	Other Diagnoses	Trace Dose		Trace Dose % Excretion in 48 hours	Therapy Dose		Therapy Dose % Excretion in 48 hours
			Date	Amount		Date	Amount	
R. W.	Hyper-thyroidism	HCVD	9/21/49	.050mc	70%			
M. E.	? Hyper-thyroidism	ASHD HCVD	9/21/49	.050mc	58%			
F. E.	Thyro-toxicosis	Thyroid Heart Disease				9/21/49 12/9/49	7.3mc 8.1mc	39% 52%
L.	? Hyper-thyroidism		10/15/49	.050mc	44.7%			
H. T.	? Hyper-thyroidism		10/15/49	.050mc	50.0%			
L. F.	? Hyper-thyroidism	Essential Hypertension HCVD Nephro-sclerosis	10/15/49	.050mc	12.9%			
W. A.	? Hyper-thyroidism		10/15/49	.050mc	56.7%			
B. F.	Hyper-thyroidism	? Neur-asthenia	10/15/49	.050mc	38.1%			
A. R.	Hyper-thyroidism	ASHD				10/15/49	4.0mc	50.2%
K. T.	Thyro-toxicosis					10/15/49	7.4mc	12.4%

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TABLE 1 (Cont'd.)

Patient	Clinical Diagnosis	Other Diagnoses	Trace Dose		Trace Dose % Excretion in 48 hours	Therapy Dose		Therapy Dose % Excretion in 48 hours
			Date	Amount		Date	Amount	
D. K.	Thyro-toxicosis		11/15/49	.053mc	23.9%	12/9/49	8.1mc	31.9%
M. S.	? Hyper-thyroidism		11/15/49	.053mc	32.0%			
R. H.	Thyro-toxicosis		11/15/49	.053mc	29.3%	12/9/49	7.0mc	42.4%
R.M.	Thyro-toxicosis		11/15/49	.053mc	23.1%	12/9/49	7.0mc	29.5%
J. V.	? Hyper-thyroidism		11/15/49	.053mc	22.5%			
T. B.	? Thyro-toxicosis	HCVD	11/15/49	.053mc	21.6%	12/9/49	6.0mc	37.7%
H. H.	? Hyper-thyroidism		11/15/49	.053mc	53.0%			
E. L.	Hyper-thyroidism	HCVD				11/15/49	6.28mc	87.0%
R. Hd.	Thyro-toxicosis	Congestive Ht. Failure				12/9/49	8.9mc	38.1%

87%. The average percent excretion for all the therapy patients was 42% with all but one of these patients excreting 50% or less of the administered dose. The computation of the therapy dose for each patient was done on a more or less empirical basis taking into consideration the percent excretion of the trace dose, when given, the degree of hyperthyroidism, and the size of the thyroid gland. No serious effort was made to estimate the gland weight. It was agreed that no dose for the treatment of hyperthyroidism should exceed 10 mc. The average dose was 7.0 mc. with a range from 4.0 mc. to 8.9 mc. These doses fall well within the range reported recently in the literature (1-7). No epithyroid counts were done, but a method of directly measuring the uptake of I^{131} by the gland is being devised. The dose given to each patient is listed in Table 1 (Pages 82 and 83).

A discussion of the effect of the I^{131} on the hyperthyroidism in these patients is not possible at this time since such a short period of time has elapsed since the therapy doses were given. One patient, F. E., was first seen in September, 1949, and at that time clinically had moderately severe hyperthyroidism associated with thyrotoxic and arteriosclerotic heart disease. He had rapid auricular fibrillation and mild congestive heart failure, which was not controlled by digitalis and the usual cardiac regime. He was given 7.3 mc. of radioiodine. During the two months following therapy he showed some improvement, but then the rapid heart rate and congestive heart failure returned, and it was felt that residual hyperthyroidism contributed to this. He was given a second therapy dose of 8.1 mc. of I^{131} on December 9, 1949. The ultimate results of the I^{131} therapy in this patient and all other patients will wait upon a longer period of observation and will be reported later.

Two patients with carcinoma of the thyroid have been treated. The first

of these, J. A., is a 17 year old white girl who at the age of ten years had an adenoma of the thyroid removed surgically. At age 14 years she noted that the thyroid gland was enlarging. This enlargement increased gradually over a period of three years. In August, 1949, she consulted her physician who recommended surgical exploration, which was done in October, 1949. At operation a very vascular invading tumor was found which proved to be a papillary adenocarcinoma of the thyroid on pathological examination. She was referred to the Genesee Hospital for I^{131} therapy, and on November 15, 1949, she was given a trace dose of 1.1 mc. of I^{131}. Exploration of the surface of the body with a Geiger counter following the administration of this dose revealed no evidence of localization of the I^{131} except in the thyroid gland. She excreted 51% of this dose. It was decided to give her a myxedema producing dose, and on December 9, 1949, she received 20 mc. of I^{131}. She excreted 76% of this dose in 48 hours. It is proposed at present to repeat this dose if no signs of hypothyroidism appear within six to eight weeks. The other patient is a 57 year old white woman who had noted a lump in her neck for many years. During the past year this lump had increased in size and because of this she consulted her physician. A biopsy was done in November 1949 which revealed a papillary and follicular type of carcinoma of the thyroid. On December 9, 1949, she was given a trace dose of 1.0 mc. of I^{131} of which she excreted 54%. It is proposed to give this patient a myxedema producing dose of I^{131}.

The standardization and measurement of dose of the I^{131} has been done, and will continue to be done in the Clinical Chemistry Section of the Medical Services Division. The urinary excretion measurements to date have also been done in the Clinical Chemistry Section, although it is planned to have these done at the Genesee Hospital in the future. The carrier-free I^{131} is obtained

from the Oak Ridge Laboratories and is administered to the patient in approximately 25 ml. of a slightly alkaline aqueous solution containing a small amount of sodium sulfite. The standardization of the radioiodine is done in triplicate by the procedure specified by the National Bureau of Standards in Circular 473 issued during June, 1949. The urine samples obtained at various intervals are asseyed for radioiodine by a comparison technique in which the activity of 0.050 ml. portions of urine are compared with the activity of 0.050 ml. portions of reference standard solutions prepared from aliquots of the same I^{131} solution administered to the patient. The strengths of the reference standards are so chosen that the activity of a 0.050 ml. sample would closely approximate that of a 0.050 ml. of urine. The final dilution of each reference standard is made with human urine. All other dilutions are made as recommended by the National Bureau of Standards in Circular 473. The comparison of reference standards and unknowns is made by measuring the mixed beta and gamma radiation given off during decay of I^{131} using a counter having a total equivalent window thickness of 6.7 mg/cm^3. The estimated error in these procedures approximates 10%.

Summary.

1. A cooperative program for the clinical use of radioisotopes has been instituted between the Atomic Energy Project of the University of Rochester and the Genesee Hospital of Rochester, New York. This program is described briefly.

2. The clinical studies, to date, have been limited to the use of I^{131} for the diagnosis and treatment of thyroid disease.

3. Nineteen patients with hyperthyroidism have received I^{131} during the period September-December 1949, and of these, nine patients have received therapy doses. The clinical diagnosis and 48 hour urinary excretion of I^{131} is

tabulated for each patient. A longer period of observation is necessary before the results of therapy can be evaluated.

4. Two patients with malignant disease of the thyroid have received I^{131}. Their cases are discussed briefly.

5. The methods for standardization of the I^{131}, and for measuring the excretion of I^{131} in the urine, are described.

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PROGRAM H. P.

HEALTH PHYSICS

Problem Code: H.P.1 (Research and Development)

Section Code: 3320

Author: Herbert Mermagen

Calculations of Radium Exposures for Photographic Sensitometry.

During a discussion on photo-sensitive emulsion densities produced by exposure to radium gamma radiation, the question of what value to assume when a change is made from a source filtered through 0.5 mm. platinum to a source filtered through 1.0 mm. platinum was considered.

Taking the basic figure of 8.47 r/hr/mg. at 1 cm. for a radium source filtered through 0.5 mm. platinum, it should be possible to compute the total intensity of a source filtered through 1.0 mm. platinum.

A reference (1) to a change in intensity by changes in filtration was found, and values for the difference in transmission of gamma rays from radium are reported in Table 1 below.

TABLE 1

FRACTIONS TRANSMITTED

Mm. of Pt.	.1	.5	1.0	2.0
Gamma Soft	.905	.605	.367	.135
Gamma Hard	.99	.95	.905	.818

From these values, it was thought interesting to calculate the total transmission intensities for the filters stated previously

Method #1

This total intensity can be expressed as the sum of the individual transmitted intensities:

(1) $$I_t = I_1 e^{-\mu_1 x} + I_2 e^{-\mu_2 x}$$

where I_t = total transmitted intensity

I_1 = intensity of the soft gamma components

I_2 = intensity of the hard gamma components

and the exponentials refer to the fractional decrease of either soft or hard gamma radiation. These values are stated in Table 1 (Page 88) as .605 and .95 for .5 mm. platinum filtration, and .367 and .905 for 1 mm. platinum filter. The equation (1) can now be re-written with numerical values for the exponential expressions and value for T_t 8.47 for .5 mm. platinum filter.

(1a) $$8.47 = .605\ I_1 + .95\ I_2$$

Similarly, the equation for the fractional values for .1 mm. platinum is:

(1b) $$x = .367\ I_1 + .905\ I_2$$

where x is the total transmitted intensity for 1 mm. platinum filters.

According to Failla (2) the ratio of hard to soft components has been found to be approximately 3:1, thus again from equation (1a)

(2) $$8.47 = .605\ I_1 + .95\ (3\ I_1)$$

When this is solved for I_1, one obtains

(3) $$I_1 = 2.45 \text{ and consequently } I_2 = 7.35$$

We can now calculate the total intensity for 1 mg. radium with 1 mm. platinum filter from equation (1b).

(4) $$x = .367\ (2.45) + .905\ (7.35)$$

$$x = 7.55 \text{ r/hr/mg. at 1 cm.}$$

The results of equation (3) also would indicate the total intensity from .1 mg.

radium without filtration to be

(5) $2.45 + 7.35 = 9.80$ r/hr/mg. at 1 cm.

This value for unfiltered gamma radiation from radium is comparable to the value of 9.69 r/hr/mg. at 1 cm. calculated by R. E. Evans (3).

Method #2

If one uses this value for 9.69 and by means of equation (1) re-calculates the total intensity for 1 mg. and 1 mm. platinum filter, than

(6) $8.47 = (9.69 - I_2)\ .605 + .95\ I_2$

and $I_2 = 7.53$

$I_1 = 2.16$

Again with aid of equation (1b) for 1 mm. platinum filter,

(7) $x = .367\ (2.16) + .905\ (7.53)$

$x = 7.61$ r/hr/mg. at 1 cm.

In comparing Failla's value for the ratio of hard to soft components with the latter values using Evans value, one finds

Failla----------3:1

Evans----------3.48:1

Method #3

Still another approach may be taken, and this was found in "Nucleonics" by G. W. Morgan (4). This author states that the total transmission intensity may be calculated from the formula

(8) $r/hr. = 8.98\ m\ (1 - .12\ t)$*

---

* This equation originates from G. C. Lawrence (5). This author obtained a mean value of 8.35 r/hr/mg. at 1 cm. for total intensity from radium filtered through 0.5 mm. platinum, and apparently a value of 8.98 r/hr/mg. at 1 cm. for unfiltered radiation. His general formula for the intensity of radiation as applicable to any filter thickness of platinum greater than 0.3 mm. is stated as

$(8.98 - 1.18\ mm^{-1}\ t)$ in r/hr/mg. at 1 m

It seems that a slight error is present in G. W. Morgan's formula, which should read $8.98\ (1 - .13\ t)$ and it was this value which led to the values of (9a) and (9b).

m = mass in mg. of radium

t = thickness of platinum in mm. not less than .3 mm. platinum

Substituting 1 mg. of radium and .5 mm. platinum and 1 mm. platinum respectively, one obtains

(9a) 8.396 r/hr/mg. at 1 cm. for .5 mm. platinum filter

(9b) and 7.81 r/hr/mg. at 1 cm. for 1 mm. platinum filter

Method #4

There is yet a further approach to calculate the total intensity after platinum filtration, based on a value of average photon energy of radium gamma rays. This average energy calculated from Evans' (3) values is .78 Mev. The absorption coefficient for platinum is found as 1.716 cm^{-1} and thus one may write the exponential absorption equation for 1 mm. platinum as

(10) $x = 8.47\ e^{-1.716\ (.05)}$

$= 7.77$ r/hr/mg. at 1 cm.

A comparative table (Table 2) has been prepared for intensity transmission values as calculated above.

TABLE 2

Method	0	.5	1.0 mm. platinum filter
1	9.8	8.47	7.55
2	9.69	8.47	7.61
3		8.40	7.81
4	9.23	8.47	7.77
5	9.51	8.47	7.54

Two series of film exposures were made with two radium sources, one with .5 mm. platinum, the other with 1.0 mm. platinum filter. The exposure calculations

were based on the accurately known content of a 10 mg. radium needle with .5 mm. platinum filter. For equal densities, it was found that the ratio of the r values for the two sources is .8905. Thus, a value for the true transmitted intensity was obtained as shown in Table 2 (Page 91) as Method #5, which is in good agreement with Methods #1 and #2, as calculated.

Bibliography

1. Medical Physics, Glasser, Otto, Year Book Publication, 1947, p. 1185.

2. American Jour. Roentg. and Ra. Ther., 44, 889, 1940.

3. Advances in Biological and Medical Physics, Vol. 1, 151-218, Academic Press, New York, 1948.

4. Nucleonics, 4, 24-36, 1948

5. Canadian Jour. Research, 15A, 67-78, 1937.

PROGRAM I. N.

INSTRUMENTATION (SPECTROSCOPY, ELECTRON MICROSCOPY, X-RAY AND NUCLEAR RADIATION DETECTORS, X-RAY DIFFRACTION, ELECTRONICS)

Problem Code: I.N.2 (Service)

Section Code: 3150

Author: L. T. Steadman

1. 139 chamber dust samples were analyzed for beryllium
2. 10 chamber dust samples were analyzed for zirconium
3. 12 air dust samples were analyzed for zirconium and silicon
4. 89 animal tissues were analyzed for beryllium
5. 8 uranium fume samples were analyzed for impurities
6. 3 animal food samples were analyzed for manganese
7. 4 cement samples were analyzed for iron
8. 1 skin granuloma was analyzed for beryllium
9. 4 human autopsy samples were analyzed for beryllium
10. 1 human lung sample was analyzed for uranium

EDUCATIONAL PROGRAM

Problem Code: None

Section Code: 3480

Author: J. N. Stannard

Radiological Physics Courses. During this quarter, the course in Instrumentation was completed (see outline pages 95-97) and the course in Radiation Biology begun. A detailed outline for this latter course, which will continue until February 17, is shown on pages 98-101. In addition to the ten A.E.C. Fellows in Radiological Physics, one A.E.C. post-doctoral Fellow is taking this course for credit and several graduate students from other departments of the Medical School are auditing.

Civil Defense Training. Preliminary plans were formulated during this quarter for a one week course in Medical Aspects of Civil Defense Measures, to be given in March, 1950. This course is part of a cooperative effort involving seven Universities and Atomic Energy Commission installations, and will be given to physicians selected by the National Securities Resources Board and the Commission. Detailed outlines of the subject matter discussed will be included in the next quarterly report.

COURSE IN RADIOLOGICAL PHYSICS

SCHEDULE FOR INSTRUMENTATION SECTION*

October 6 to December 2, 1949

DATE	**TYPE	SUBJECT
October 6 (A.M.)		Instrumentation Orientation.
October 11 (A.M.)	L	Introduction to Radiation.
October 11 (P.M.)	D	Scintillation and range of alpha particles.
October 13 (A.M.)	E	Range and penetration of gamma and beta radiation.
October 13 (P.M.)	E	Range and penetration of gamma and beta radiation.
October 14 (A.M.)	L	Ionization in gases.
October 17 (A.M.)	L	Ionization measuring devices.
October 18 (A.M.)	E	Demonstration and calibration of electrometers.
October 18 (P.M.)	E	Demonstration and calibration of electrometers.
October 20 (A.M.)	L	Vacuum tube electrometers.
October 20 (P.M.)	E	Vacuum tube electrometers.
October 21 (A.M.)	L	Vibrating reed and Ballistic Vane electrometers.
October 24 (A.M.)	L	Electronic components for counting equipment.
October 25 (A.M.)	E	Triode and pentode amplifiers.
October 25 (P.M.)	E	Triode and pentode amplifiers.
October 27 (A.M.)	L	Regulated power supplies.
October 27 (P.M.)	E	Regulated power supplies.

* Responsible Instructor: H. E. Mermagen
Assistants: R. G. Vyverberg, G. R. Hopkins. S. G. Sizzo

** L - Lecture
D - Demonstration
E - Experiment

DATE	TYPE	SUBJECT
October 28 (A.M.)	L	Multivibrators and Univibrators.
October 31 (A.M.)	L	Scaling circuits.
November 1 (A.M.)	E	Univibrators.
November 1 (P.M.)	E	Scaling circuits.
November 3 (A.M.)	L	Counting rate meter.
November 3 (P.M.)	E	Counting rate meter.
November 4 (A.M.)	L	Mechanical recording equipment.
November 7 (A.M.)		Quiz.
November 8 (A.M.)	L	Alpha counters.
November 8 (P.M.)	L	Proportional counters.
November 10 (A.M.)	D	Proportional counters.
November 10 (P.M.)	L	GM tubes and quenching circuits.
November 11 (A.M.)	E	GM tubes and quenching circuits.
November 14 (A.M.)	L	Scintillation and crystal counters.
November 15 (A.M.)	D	Scintillation and crystal counters.
November 15 (P.M.)	L	Measurement of radiation.
November 17 (A.M.)	E	Counter efficiency and geometry.
November 17 (P.M.)	E	Counter efficiency and geometry.
November 18 (A.M.)	L	International roentgen.
November 21 (A.M.)	L	Radiation units.
November 22 (A.M.)	E	Measurement of roentgen with standard ionization chamber.
November 22 (P.M.)	E	Measurement of roentgen with standard ionization chamber.
November 28 (A.M.)		Discussion period.
November 29 (A.M.)	L	Measurements of radiation with photographic emulsions.

DATE	TYPE	SUBJECT
November 29 (A.M.)	E	Measurements of radiation with photographic emulsions.
December 1 (A.M.)		Review.
December 1 (P.M.)		Final examination.
December 2 (A.M.)		Discussion of examination.

References:

1. Lapp and Andrews - "Nuclear Radiation Physics"

2. Korff, Serge A., - "Electron and Nuclear Counters"

3. Bureau of Ships, Navy Department - "Radar Electronic Fundamentals" - Navships 900,016

4. "Theory and Operation of Geiger-Muller Counters" - Nucleonics

   Part 1, Vol. 2, No. 6, Pages 10-22 (June 1948)
   Part 2, Vol. 3, No. 2, Pages 50-64 (August 1948)
   Part 3, Vol. 3, No. 4, Pages 46-61 (October 1948)

5. "Electronics", Vol. 1, Chapters 1-7, AECD-2208

UNCLASSIFIED

OUTLINE OF COURSE IN RADIATION BIOLOGY FOR 1949-50

LECTURE AND CONFERENCE SCHEDULE

Monday and Friday - 10:30 A.M., Tuesday and Thursday - 9:00 A.M.

December 5, 1949 to February 17, 1950

	Title	Instructor
1.	Introduction, Outline	Stannard
2.	Physical factors affecting biological action I.	Steadman
3.	Physical factors affecting biological action II.	Steadman
4.	Physical factors affecting biological action III.	Steadman
5.	Chemical effects I. Radiation effects on water and simple aqueous solutions	Mason
6.	Chemical effects II. Biochemistry of nucleic acids, proteins, and related substances	Altman
7.	Chemical effects III. Radiation effects on nucleic acids, proteins, and related substances	Salomon
8.	Theories of the mode of action of radiation	Steadman
9.	Theoretical and practical considerations in the measurement of x-ray dosage	Steadman
	Quiz and Conference	
10.	Dosage units and their measurement	Hursh
11.	Dosage measurements for external sources other than x-rays	Hursh
12.	General responses to radiation I. Total body effects	Casarett
13.	Genetics and Cytology I. Mitosis	Otis
14.	General responses to radiation II. Radiation sickness	Howland
15.	Concept of radiation sensitivity of tissues	Casarett
16.	Blood and hematopoietic system	Ingram
17.	Effects of radiation on gonadal tissue	Casarett

	Title	Instructor
18.	Genetics and Cytology II. Meiosis	Otis
	Quiz and Conference	
19.	Effects of radiation on skin and G.I. tract	Casarett
20.	Relatively resistent organs	Casarett
	Discussion of laboratory reports	
21.	Genetics and Cytology III.	Otis
	Examination	
22.	Internal emitters I. Survey	Stannard
23.	Internal emitters II. Dosage calculations	Hursh
24.	Internal emitters III. Alpha emitters	Stannard
25.	Internal emitters IV. Fission products and transuranium elements	Stannard
26.	Genetics and Cytology IV.	Otis
	Conference	
27.	Therapeutic uses of radioactive isotopes	Hursh
28.	Tolerance I. Basis for radium and radon M.P.D. figures	Hursh
29.	Tolerance II. Basis for x-ray, γ-ray M.P.D. figures	Stannard
30.	Genetics and Cytology V. (Tolerance III.), genetic aspects of the tolerance problem	Otis
	Quiz and Conference	
31.	Tolerance IV. Chronic vs. acute problems	Stannard
32.	Tracer Chemistry I. Isotope separation techniques	Miller
33.	Tracer Chemistry II. Choice of tracer, design of experiments	Miller
34.	Tracer Chemistry III. Isotopes as a tool in the study of radiation effects	Salomon
35.	Tracer Chemistry IV. Conclusion	Miller

Title	Instructor
Student reports	
Conference	Staff
Final examination	

LABORATORY AND DEMONSTRATION SCHEDULE

Tuesday and Thursday after lectures

December 6, 1950 to February 9, 1950

	Title	Instructor
1.	Survey of radiation sources (Hospital, Annex, River Campus)	Mermagen Steadman
2.	X-ray Physics, Part I.	Steadman VanSlyke
3.	Effects of radiation on water and simple aqueous solutions (Demonstration)	Mason
4.	Biochemical techniques (Demonstration)	Salomon Altman
5.	X-ray Physics, Part II.	Steadman VanSlyke
6.	X-ray dosage measurements	Steadman VanSlyke
7.	Neutron and other external dosage measurements	Hursh Mermagen
8.	Start LD_{50} experiment, study normal histology*	Casarett
9.	Genetics and Cytology I. Meiosis	Otis
10.	Hematology	Ingram
11.	Continue hematological observations; Slides (x-ray effects)	Ingram Casarett

* Experiments so marked will require frequent observations over several days time.

	Title	Instructor
12.	Genetics and Cytology II. Meiosis in gonads	Otis
13.	Slides (x-ray effects) Observations and correlations in LD_{50} experiment	Casarett Stannard
14.	Conclude LD_{50} experiment, reports and discussion	Casarett Stannard
15.	Genetics and Cytology III.	Otis
16.	Start internal emitter experiment*	Stannard
17.	Slides (special features of pathology with internal emitter)	Casarett
18.	Autoradiography (Lecture-Demonstration)	Boyd
19.	Genetics and Cytology IV.	Otis
20.	Localization of I^{131} in thyroid	Hursh
21.	Conclude internal emitter experiment	Stannard
22.	Genetics and Cytology V.	Otis
23.	Tracer Chemistry I.	Miller
24.	Tracer Chemistry II.	Miller
25.	Hemin synthesis in irradiated animals (Demonstration)	Salomon Altman

*Experiments so marked will require frequent observations over several days time.

TECHNICAL REPORTS ISSUED FOR DISTRIBUTION

October 1, 1949 thru December 31, 1949

Report No.	Title	Authors	Subject Category
UR-83	"The Therapeutic Use of BAL in Polonium Toxicity. I. The Effect of Dithiol Compounds on the Excretion of Polonium. II. Blood and Tissue Distribution of Polonium as Affected by BAL. III. Survival Experiments on Polonium Injected Rats as Affected by BAL Treatment." (RESTRICTED) Issued: 10/5/49	Hursh	Health and Biology
UR-85	"Spectrophotometric Studies of the Uranyl Citrate Complex" (RESTRICTED) Issued: 10/26/49	Feldman Neuman Havill	Chemistry General
UR-89	"Effects of X-Irradiation on Thyroid Function in Rats" (UNCLASSIFIED) Issued: 10/21/49	Hursh Mohney VanValkenburg	Health and Biology
UR-90	"Studies on the Absorption and Distribution of Vitamin A in X-Irradiated Rats" (UNCLASSIFIED) Issued: 10/11/49	Bennett Bennett Shaver Grachus	Health and Biology
UR-91	"Beta Track Autoradiography Using Carbon 14" (UNCLASSIFIED) Issued: 10/18/49	Boyd Levy	Health and Biology
UR-93	"Distribution of Selenium in Dog Serum Proteins and Red Blood Cells After Subcutaneous Injection of Sodium Containing Radio-Selenium" (UNCLASSIFIED) Issued: 11/25/49	McConnell Cooper	Health and Biology
UR-94	"Studies on Factors Effecting the Radiation Syndrome. I. The Effect of Aureomycin and Antibiotics on Whole Body Radiation" (UNCLASSIFIED) Issued: 11/2/49	Howland et al	Health and Biology

Report No.	Title	Authors	Subject Category
UR-96	Quarterly Technical Report (FOR OFFICIAL USE ONLY) Issued: 12/16/49		Health and Biology
UR-97	"The Oxidation of Carbon Monoxide to Carbon O_2 as shown by Experiment with C^{14}" (UNCLASSIFIED) Issued: 11/25/49	Stannard et al	Health and Biology
UR-98	"Relationship of the Cell Surface to Metabolism VI. Hydrolysis of Sucrose and Raffinose." (FOR OFFICIAL USE ONLY) Issued: 11/18/49	Rothstein Meier	Health and Biology