JUL 11 1966

COO-1184-4

THE METABOLISM OF NATURALLY OCCURRING RADIUM-226

BY SWINE

H.C. \$ 5.00 MN 1.00

MASTED

THE FINAL REPORT UPON A RESEARCH PROJECT
SUPPORTED BY THE
ATOMIC ENERGY COMMISSION
OF THE
UNITED STATES OF AMERICA

RELEASED FOR ANNOUNCEMENT
IN NUCLEAR SCIENCE ABSTRACTS

NEAL S. NELSON, DVM, Ph.D.

AND

JOHN H. RUST, DVM, Ph.D.

SECTION OF NUCLEAR MEDICINE

DEPARTMENT OF PHARMACOLOGY

THE UNIVERSITY OF CHICAGO

CHICAGO, ILL. (60637), USA

MAY 15, 1966

AEC DOCUMENT NO. 000-1184-4

LEGAL NOTICE

[illegible]

DISCLAIMER

This report was prepared as an account of work sponsored by an agency of the United States Government. Neither the United States Government nor any agency Thereof, nor any of their employees, makes any warranty, express or implied, or assumes any legal liability or responsibility for the accuracy, completeness, or usefulness of any information, apparatus, product, or process disclosed, or represents that its use would not infringe privately owned rights. Reference herein to any specific commercial product, process, or service by trade name, trademark, manufacturer, or otherwise does not necessarily constitute or imply its endorsement, recommendation, or favoring by the United States Government or any agency thereof. The views and opinions of authors expressed herein do not necessarily state or reflect those of the United States Government or any agency thereof.

DISCLAIMER

Portions of this document may be illegible in electronic image products. Images are produced from the best available original document.

PREFACE

The help of many people must be acknowledged, particularly we thank Mr. Henry F. Lucas, Jr., and his associates at the Radiological Physics Division, Argonne National Laboratory for their assistance in performing the first analyses and continuing to provide information and suggestions concerning the construction and operation of the equipment and the mathematical treatment of data. Mr. Arthur Jacobson did the calcium analyses of ashed samples. The Meat Inspection Service of the Department of Agriculture, Mr. John Thompson of the Reliable Packing Company and Kerber Packing Company have given us assistance in obtaining samples. The Swine Division of the College of Agriculture, University of Illinois, helped us in the metabolism study. Many others have contributed, notably Mrs. Shirley Danner and Mrs. Teresa Kirschner. For all of this help we are grateful.

Financial support was furnished by: the USPHS National Cancer Institute, the Rockefeller Foundation Fund for the Section of Nuclear Medicine, Department of Pharmacology, University of Chicago, the Atomic Energy Commission under AEC Contract AT (11-1) 1184 from the Chicago Operations Office.

Computation was accomplished with the assistance of the Biological Sciences Computation Center, University of Chicago, under USPHS grant FR 00013 from the Division of Research Facilities and Resources of the National Institutes of Health.

Portions of this final report appeared under the title "Skeletal Distribution of ²²⁶Ra in Swine Consuming Feed and Water with Naturally Occurring Radium" in Health Physics 10, 407-413 (1964) and other portions have been presented at the 1966 International Symposium on Radioecological Concentration Processes of the Swedish National Committee on Radiation Protection, Stockholm, Sweden,

April, 1966, and will appear in the published proceedings of that symposium under the title of "Distribution and Discrimination Against Naturally Occurring Radium-226 in Swine." Much of it also was used by Dr. Nelson in his dissertation submitted to the Faculty of the Division of Biological Sciences in his candidacy for the degree of doctor of philosophy in the Department of Pharmacology, The University of Chicago. The title of the dissertation was "Some Observations on the Metabolic Fate of Radium-226 in Swine" and its date was June, 1964.

TABLE OF CONTENTS

	Page
PREFACE	ii
LIST OF TABLES	vi
LIST OF ILLUSTRATIONS	viii
INTRODUCTION	1
MATERIALS AND ANIMALS	5
METHODS	9
DATA AND DISCUSSION	16
Preliminary Investigation Skeletal Distribution of Radium-226 in Swine Comparison of the Skeletal Radium-226 Burden in Overtly Normal and in Lymphosarcomatous Swine Metabolic Fate of Radium-226 in Swine	
CONCLUSIONS	55
SUMMARY	57
REFERENCES	59
APPENDICES	
A. RADON COUNTING BOTTLE	78
Construction Calibration	
B. STANDARDIZATION OF COUNTING EQUIPMENT	88
Reproducibility of Counts	
C. PREPARATION OF SAMPLES	95
Bone Tissue, Feed, and Feces (First Steps) Tissue (Second Steps) Feed and Feces (Second Steps) Urine and Blood (as Liquids) Blood and Milk (as Ashed Samples)	

TABLE OF CONTENTS--Continued

	Page
D. RADON COLLECTION SYSTEM	103
Generator Flasks	
Radon Traps	
Operation of the Radon Collection System	
Corrections for Dead Space	
Half Transfer Volume	
E. RADON TRANSFER SYSTEM	117
F. MATHEMATICAL TREATMENT OF DATA	123
G. SPECIAL ANALYTICAL PROBLEMS	129
H. SPECIAL MATERIALS	134

LIST OF TABLES

Table		Page
1.	Radium-226/Calcium Ratios in Some Southeastern Kansas Waters That Have Been Used to Water Livestock	18
2.	Distribution of ²²⁶ Ra in Swine Skeletons Normalized for Comparison	20
3.	Correlation Between Origin and Skeletal Burden of Radium-226 in Overtly Normal and in Lymphosarcomatous Swine	24
4.	Maximum and Minimum Skeletal Burden of Radium-226 in Swine in This Study	26
5.	Radium-226 Content of Feed and Water, Tissues, Secretions and Excretions for All Animals Assayed	29
6.	Average Per Cent Distribution of Excreted Radium-226 in Urine and Feces for a 7-Day Period	30
7.	Ratio of Radium-226 to Calcium in Feed and Water, Tissues, Secretions and Excretions for All Animals Assayed	37
8.	Ratio of Radium-226 to Phosphorous in Feed and Water, Tissues, Secretions and Excretions of All Animals Assayed	38
9.	Observed Ratios Based on Radium to Calcium Ratios	39
10.	Observed Ratios Based on Radium to Phosphorous Ratios	40
11.	Observed Ratios for Neonate, 2-Week Old and 4-Week Old Swine	41
12.	Comparison of Observed Ratios for Radium-226 in Swine, Based on Radium to Calcium Ratios, with Observed Ratios Reported for Strontium-90 in Several Species	42
13.	Comparison of Theoretical and Calculated OR's Based on Radium-226 to Calcium or Radium-226 to Phosphorous Ratios	50
14.	Transmission of Light (4500 Å) by Quartz Windows, Efficiency and Background Count of Radon Counting Bottles	82

LIST OF TABLES--Continued

Table		Page
15.	Examples of Primary Calibration of Counting Bottles	86
16.	Per Cent Equilibrium Between Radon-222 and Radium-226 for Various Time Intervals of Aging	112
17.	Correction for Dead Space for Various Systems	113
18.	Half Transfer Volumes for Large Solution Volumes	115
19.	Effect of Flow Rate on Sample Analysis	116

LIST OF ILLUSTRATIONS

Figure		Page
1.	Dimensions and Construction of Radon Counting Bottle	79
2.	Arrangement of Radon Counting Bottles and Photo-multiplier Tubes in the Counting Equipment	89
3.	Operational Plateaus for One Scaler	90
4.	Small Radon Generator Flask	97
5.	Large Radon Generator Flask	100
6.	Radon Collection System Showing a Generator Flask of the 500 cc. or 1,000 cc. Size in the System	104
7.	Radon Collection System Showing a Generator Flask of the 1 to 5 Gallon Size, the CO ₂ Trap, and Cold Finger Moisture Traps in the System	105
8.	Dimensions and Construction of Radon Trap	110
9.	Dimensions and Construction of the Radon Transfer System	118
10.	Equations for Calculation of Radon Content and Error of This Calculation	125
11.	Equation for Calculating Weighted Means	128

INTRODUCTION

Radiotoxicology and health physics have been of increasing interest because of the stimulation which came with the development of nuclear energy, the concern with fallout and the possible injury from use of radioisotopes in research and industry.

Radium and radon played a major role in the discoveries in nuclear physics and not unexpectedly in the development of radiotoxicology. The earliest evidence of radioactivity was by Becquerel (1) in 1896, when he discovered that photographic film, covered with black paper, would develop an image when exposed to uranium salts. Other sources of the rays, soon to be called "Becquerel rays," from naturally radioactive materials were rapidly discovered and reported. Radium was identified by the Curies and Bemont (2) in 1898, polonium by the Curies (3) in 1898, actinium by Debierne (4) in 1899, radon-220 (thoron) by Rutherford (5) in 1900, radon-222 (radon) by Dorn (6) in 1900, and radon-219 (actinon) by Giesel (7) in 1902. Radon-222 was originally known as niton. Other elements were rapidly added to this list. And in 1911 Soddy wrote the first of his books on the chemistry of the radioelements (8).

The first investigation of the toxicity of a radioactive compound was done by Bouchard et al. (9) in 1904, who exposed mice to an atmosphere containing radon. Even though the animals died in 6 to 9 hours and by autoradiography were shown to have deposits of radioactive materials in all tissues, radium and related compounds were soon to be used in injections, drinks, and baths. Medical literature from 1912 into the 1930's contains many case reports and recommendations for its use. However, even in 1912 there were some

investigators who had doubts of the safety of the use of radioactive materials for medical treatment. Bickel (10), Gudzent (11), and Klemperer (12) reported cases of poisoning which were believed to be due to thorium-X (radium-224) treatment. Tagle-Chile (13), in 1912, reported that damage could be caused to tissues by radiothorium (thorium-228). Little notice was taken of these observations and use of radium and related compounds proceeded unabated.

In the early 1920's, watch companies in Newark and Orange, New Jersey, employed a number of women to paint watch dials with a luminous paint containing radium-226 and radium-224. The women's habit of "tipping" the paint brushes with their tongues provided a good opportunity for ingesting radium. Blum (14), a New Jersey dentist, first reported a case observed in 1923, in which the patient had osteomyelitis of the mandible and maxilla similar to phosphorus necrosis. He attributed the condition to some radioactive substance used in the manufacture of luminous dial watches, and called the condition "Radium Jaw." In 1925, Hoffman (15) and Martland et al. (16), also of New Jersey, associated what they termed "Radium Necrosis" of the bone with the radioactivity of radium and thorium compounds used in industry. In 1929, St. George et al. (17) published the values for the radium content of the tissues of a female radium dial painter who had died in 1922. Her bones contained 2×10^{-8} gm. of radium-226/gm. of ash, about 10,000-fold more than normal values found today in human bones (see also references cited under Methods).

Looney (18), Looney et al. (19), Bloom and Bloom (20), and Dougherty et al. (21) have contributed important information on the effects of alpha particle emitters, including radium-226, on tissues. The review of pre-1945 literature on radium poisoning by Silberstein (22) and other available literature clearly show the important role that radium toxicity holds in all consideration of effects of internal-emitting and bone-seeking radioisotopes.

In fact, most of the values for maximum permissible concentration of internally lodged radioisotopes are derived from studies of radium toxicity.

Thompson (23) states, "The central position of radium in considerations of internal emitter hazards is the result of some hundreds of human exposures in which the metabolism and biological effects of radium may be studied." Dr. Katherine Williams (24) said that the International Commission on Radiological Protection (ICRP) considered "occupational and medical experience with radium offers much more justification for accepting the 0.1 μCi ^{226}Ra and for the RBE dose it delivers to the bone as a basic reference for permissible occupational exposure than any arbitrarily chosen dose rate to an individual organ." The 1954 ICRP recommendations (25) state, "In the case of α -emitting radioisotopes that localize in the bone the maximum permissible body burden, q , is determined from a direct comparison with radium. Man has had years of experience with radium, and it is believed that the long-established maximum permissible body burden for radium of 0.1 μCi should serve as the basis of choice of the maximum permissible body burden of other similar α -emitting radioisotopes that are deposited in the bone." This central position of radium-226 in radiotoxicology assures that additional knowledge of the metabolic fate of radium-226 in animals will be of significant value.

Further, Schraub et al. (26) have reported that radium-226, radon-222, and daughter products contribute between 37.5% and 71% of the total natural dose rate to body tissue in humans. Radium-226 and daughters might then play a large role in the total radiation exposure in all animals. Because of the physiological similarities between swine and human (27), this investigation into the metabolic fate of radium-226 was initiated in swine with the hope that by compiling such information it not only would add to general information about radium-226 but also could be correlated with the human case.

It was of special interest to us to know whether physiological boundaries presented any selective barrier to radium-226 when compared to other divalent cations.

An attempt was made to correlate radium-226 burden and lymphosarcoma in swine. The proximity of radium-226 deposited in bone to sensitive elements of the hematopoietic system suggested that radiation damage might conceivably play a part in the genesis of the disease. Three cases of myeloid leukemia have been associated with radium-226 poisoning in humans (28,29,30). There have also been reports of reduced euploidy in bone marrow cells (31), chromosomal aberrations in blood cells and abnormalities in lymphocytes (32) with whole body burdens as low as 10% of the Maximum Permissible Burden.

MATERIALS AND ANIMALS

With the exception of the glass systems which were specially constructed by the University of Chicago Glass Shop for this study and those special materials listed in Appendix H, all components and reagents were obtained from standard sources of supply.

The samples assayed for radium in this study were obtained from several sources. Samples from thirty-four mature, eleven immature, and eight neonatal animals were assayed, a total of fifty-three swine.

Four skeletons of mature swine (U of I No. III, No. IV, No. V, and No. VI) for study of the skeletal distribution of radium-226 were obtained through the cooperation of the College of Veterinary Medicine at the University of Illinois, Urbana, Illinois. Although the exact farm from which these swine skeletons were obtained is known, only the general geographic area of the source will be reported.

Skeletons of a normal mature swine (Normal No. 3) and of a leukemic mature swine (Leukemic No. 2) for study of the skeletal distribution of radium-226 were obtained from a local packing house. Samples of rib and/or humerus for study of the relationship of radium-226 bone burden and lymphosarcoma from nine leukemic swine (Leukemic No. 1 and Nos. 4 to 11) and three normal swine (Normal No. 1, No. 2, and No. 4) were secured through the cooperation of the Meat Inspection Service of the U. S. Department of Agriculture and local packing houses. The leukemic swine were diagnosed as leukemic provisionally during postmortem inspection at the packing house. Liver, spleen, and lymph nodes were taken with the bone samples from each animal. The tissue specimens were fixed then sectioned and stained with hemotoxylin and eosin for

histopathological examination.¹ All animals provisionally diagnosed as being leukemic at autopsy were later confirmed as being lymphosarcomatous by histopathological examination.

The animals used for the radium metabolism study were from two sources. Eight 6-month old males (Barrow 1-1, 1-2, 1-3, 1-4, 2-1, 2-2, 2-3, and 2-4) were obtained from the Swine Division of the College of Agriculture at the University of Illinois, Urbana, Illinois. These barrows were brothers or half-brothers from crossbred Hampshire sows bred to a Montana No. 1 boar. They were farrowed in February, 1962, and slaughtered in August, 1962. These animals had been kept in the same lot after weaning. They received the same "creep," "grower," and "finisher" feeds and were supplied with water from the same source.

All eight animals were removed from the lot at the same time. Four animals (Group 1) were placed in swine metabolism cages at the Swine Division barns; the other four (Group 2) were maintained on clean concrete and were later placed in the metabolism cages. Each group of animals was held in the metabolism cages for 7 days. During the time before they were placed in metabolism cages and while in the metabolism cages, all animals were maintained on feed and water obtained from the same source they had had when they were running together in the lot.

While in the metabolism cages, the animals were fed twice a day. One feeding was in the morning, the second about 8 hours later. Feed was fed as a slurry of dry feed and water mixed in a 1:1 ratio by weight. The consumption of dry feed, the weight of feces excreted, and the volume of urine voided were recorded daily for each animal. The urine and feces were collected daily and pooled at the end of the 7-day period. All maintenance of records, feeding,

¹The Clinical Pathology Laboratory, The University of Chicago.

and collection of excreta was done by University of Illinois, Swine Division, personnel using standard University of Illinois procedure for metabolism studies.

Random samples of feed and water were collected daily and pooled for each 7-day period. The water was obtained from the city of Urbana water system and had been subjected to water softening treatment by the city. The radium content of the pooled water sample for the first group of four barrows (Barrow 1-1, 1-2, 1-3, and 1-4) was about twice the radium content of the pooled water sample for the second group of four barrows (Barrow 2-1, 2-2, 2-3, and 2-4). In view of this difference, pooled water and pooled feed samples were treated as if each group of four barrows was a separate study. In contrast, the discrimination factors and observed ratios computed for each animal in the two groups are presented as averages of eight animals. Samples of blood, lean muscle, humerus, and rib were obtained from each animal at slaughter.

For investigation of placental and mammary discrimination, a group of eight mature females¹ (Sow Nos. 1 to 8) was obtained from a farm near Frankfort, Illinois. They were sisters or half-sisters farrowed in April, 1962, and were raised on that farm. They had received the same amount and type of feed, i.e., 2.56 kilograms of dent corn and cob plus some supplement per day. The test sows were bred to a Hampshire boar on or about August 30, 1963. It was the second litter for all sows. After the sows were bred, each then received 0.5 kilograms of a special sow supplement each day in addition to the normal feed ration.

The pregnant sows were transferred to a farm at Goodenow, Illinois, on October 21, 1963, where they were held until they farrowed. At Goodenow, each sow received 3.66 kilograms of ground feed containing corn, oats, and supplement plus 16 liters of water each day. Pooled water samples taken

¹Connor Prairie hybrids, a mixture of Hampshire, Landrace, and other unknown domestic breeds.

during the study had nearly identical radium-226 contents. The average of these pooled samples was used as the radium-226 content of the water.

The sows farrowed in the following order:

Nos. 1 and 2 on December 17, 1963,
No. 3 on December 19, 1963,
Nos. 4, 5, and 6 on December 21, 1963,
No. 7 on December 22, 1963, and
No. 8 on December 24, 1963.

No. 7 died during farrowing and was dropped from the study and replaced by No. 9. No. 9 was identical to the other seven sows with the exception of time and breeding. She was bred on or about September 19, 1963, and she farrowed on January 10, 1964.

Neonatal piglets were obtained from each sow at farrowing and killed before they could nurse. Samples of milk were obtained from five of the eight sows between 4 and 5 weeks after they farrowed. The sows were slaughtered on February 7, 1964. At the time of slaughter, samples of blood, skeleton, and organs were obtained from each sow.

METHODS

Many approaches have been used for determining the quantity of radium present in a sample. The problem is particularly difficult at the naturally occurring levels of radium-226. Natural levels of radium-226 may range from 10^{-9} to 10^{-15} gm. $^{226}\text{Ra}/\text{gm.}$ of vegetable products (33-36) and from 10^{-10} to 10^{-18} gm. $^{226}\text{Ra}/\text{gm.}$ of animal tissue and water (34-49). Since the range of naturally occurring levels is so great, it was believed best to adopt methods capable of detecting radium-226 levels of 5×10^{-15} gm. of radium-226 per gram of sample. The system selected, therefore, had to be able to provide high sensitivity and precision with a minimum of sample processing even for large dilute samples.

The techniques available could be divided into three broad categories: alpha particle track autoradiography, systems using electrometers or electroscopes and systems using scintillation counting.

Alpha particle track autoradiography has been used for measurements of α -emitting radioisotopes by Rotblat and Ward (45), Hindmarsh and Vaughan (47), Hoecker and Rooft (50), Rowland and Marshall (51), Lloyd (52), and Ward (53). With this technique about one out of every 4.5 alpha particle emissions will produce a recognizable track in an emulsion placed against the bone surface, within the range of the alpha particle emitted (51). This technique was not considered suitable, since good autoradiographs require either high levels of radioactivity in the sample or long periods of exposure of the sample-emulsion package. Rough calculations suggested there would be only one alpha track per square centimeter of surface per year for a burden of 1×10^{-12} gm. of radium-226 per gram of sample. A modification of this

technique has been developed by Snihs (54) which is applicable to emanation assay techniques. The method is rather complex though it is sensitive.

Various types of electrometers have been used by many investigators. Rutherford (5), Muth et al. (34), Krebs (41), Radford et al. (42), Hursh and Lovaas (43), Walton et al. (44), Spiers and Burch (48), Madam Curie (55), Becker and Stehberger (56), Cubeli and Stori (57), Stehney et al. (58), Evans (59), Rona and Urry (60), Curtiss and Davis (61), Hudgens et al. (62), and Stover et al. (63) have reported radium-226 measurements made with an electrometer. The electrometers used were of many types and included vibrating reeds, differential ion chambers and others. Some of the authors using these systems (61,62) have been able to measure as little as 10^{-13} to 10^{-15} gm. of ^{226}Ra . While this was sensitive enough for the objectives of this study, other problems such as the need for purification of radon prior to counting and an absolute sensitivity limit for ion chambers of 10^{-14} gm. ^{226}Ra (59) made it essential that a better technique be used.

The technique of alpha particle scintillation counting is one of the newest and probably the most satisfactory technique for quantitatively measuring alpha particle emitting radioisotopes. Turner et al. (39), Greig (64), Kaul and Muth (65), Rosholt (66), Rosholt and Dooley (67), Muth and Oberhausen (68), Kulp et al. (69), Van Dilla and Taysum (70), Van Dilla et al. (71), Sax and Rosi (72), Lucas (38,73), and Snihs (54) have reported on alpha particle scintillation counting. Some investigators (39,66-69,72) have mixed ash containing radium-226 or precipitated (or coprecipitated) radium-226 with zinc sulfide and then counted the emissions directly with a photomultiplier system. Other investigators (64,64,70,71,73) have used systems which trapped radon-222 gas and transferred it to a zinc sulfide coated vessel for counting. The value of zinc sulfide as a scintillator and methods of preparing optimum thickness layers for alpha counting has been discussed by Robinson and Arnold (74) and

Robinson et al. (75), who found that zinc sulfide layers about 20 mg. per cm² thick would give a maximum count rate. Further, they noted that count rates would reach a plateau as the operating voltage of the photomultiplier tube was increased. Finally, they observed that zinc sulfide was the most satisfactory inorganic scintillator tested for alpha emission counting. Damon and Hyde (76) constructed a scintillator lined glass tube with a clear window at one end. Radioactive gases were transferred to the tube by a vacuum transfer method and scintillations detected with a photomultiplier system.

Other investigators (38,58,64,75,73) have used activated charcoal to trap and concentrate radon-222, the daughter of radium-226. The phenomenal capacity of activated charcoal to trap radon, especially at temperatures below 0° C., has been noted by Becker and Stehberger (56), Gubeli and Stori (57), Stehney et al. (58), Siebert (77), and Jennings and Russ (78), who have given attention to some of the problems created by the various carrier gases.

Of the techniques considered, that of Lucas (38,73) seemed most appropriate for the objectives of this study. It combined the concentration of radon by trapping on charcoal with alpha particle scintillation counting. The system is sensitive over the range of values of radium concentrations we expected. The particular restriction imposed by the system is that the radium bearing sample must be soluble. A sample can be in an acid, alkaline, or neutral solution (79) if maintained in an ionic or chelated state. It later developed that there is another important restriction; excessive amounts of contaminating gas, e.g., CO₂ and H₂S, have to be removed before the radon reached the radon collection trap.

Radon counting bottles were constructed (see Appendix A) and electronic equipment standardized (see Appendix B), and calibrated (see Appendix A).

All solid samples, milk and some blood were ashed at 500° to 800° C. as the first step. The ash was dissolved in concentrated HCl or HNO₃ (see Appendix C) and diluted to the proper volume with 1 molar HCl or HNO₃. No correction was made for radium content of HCl or HNO₃ since Lucas (79) has found that volumes of acid (Baker, reagent grade) up to 3 liters contained no detectable radium-226. No correction was made for the distilled water used in dilution, since 200 ml. of water contributed only 1.54 fCi¹ of radium-226 to the radium content of a sample in a 500 cc. generator flask. This contribution was never more and usually much less than 10%. The same per cent contribution was observed in all other generator flask sizes.

Dissolved, ashed samples, and liquid samples (blood, urine, or water) were transferred to generator flasks, purged free of previously accumulated radon-222 and then allowed to age before analysis (see Appendix D). For the analysis, radon-222, which had reached a time-dependent equilibrium with radium-226, was flushed from the generator flask to the radon collection trap in the radon collection system (see Appendix D) where it was trapped on charcoal in the radon collection trap at -70° C. The radon collection trap was then placed in the radon transfer system and the radon transferred to the radon counting bottle (see Appendix E). The radon-222 filled counting bottles were then allowed to stand for 3 to 4 hours, until the radon-222 and its daughters had reached an equilibrium, and then were counted. A minimum of 1,600 counts were registered, if possible, in order to maintain counting errors at 2.5%. Some of the samples would have required counting periods of 160 hours or longer to reach 1,600 counts so these were counted for 48 hours rather than to 1,600 counts. In some cases, counts were extended to 2,500 and higher to reduce counting error.

¹One femtocurie (fCi) = 1×10^{-15} curies.

The raw data as total counts, times, and dates was analyzed by a computer which determined the radium-226 content and the error range of the analysis of the sample (see Appendix F). Error was expressed as the 95% confidence limits (1.96 times the standard deviation).

Samples of feed, water excretions, and milk were obtained during the course of the investigation. Tissue samples were obtained at slaughter.

Samples of milk were obtained from only five sows. Hartman and Pond (80) reported on the design, construction, and use of a milking machine for sows, stating that maximum milk production was reached at the fourth week of lactation and one to two pints of milk could be obtained per milking. A milking machine was constructed by following the instructions given by Hartman and Pond, and it was used as they suggested commencing 3 weeks after the start of lactation in order to get as large a milk sample as possible from each animal. The milking machine proved unsatisfactory. Fearing that all eight sows would stop lactating, since three had already ceased lactating during the unsuccessful attempts to use the machine, milk samples were then obtained by hand milking by one of the investigators (NSN). The hand milking was preceded by the injection of 25 to 30 units of oxytocin into the marginal ear vein. Within 30 seconds after injection of the oxytocin, milk would flow freely for about 15 minutes. Each sow was milked several times between the fourth and fifth weeks of lactation. One hundred ml. was chosen as the minimum quantity, since the anticipated radium-226 content of 100 ml. of sows milk was 30 fCi. Thirty fCi would give a count of about 1.5 times background which would have had the maximum counting error which could be tolerated in this investigation, i.e., about $\pm 30\%$.

Calcium determinations were made on all tissue and fluid samples. The calcium values were determined by the photometric technique of Kingsley and Robnett (81). This colorimetric method has a 4% reproducibility over a

range of 5 to 250 micrograms of calcium per milliliter. All samples analyzed for calcium content were diluted to fall within this range.

The high iron content of some samples interfered with this colorimetric method, so it was necessary to remove the iron. Although there are many possible ways to remove iron from solutions, many of them also remove calcium, e.g., precipitation reactions through adsorption. The approach using organic solvent extraction offered the most hope (82), since the extremely low pH of the solutions (pH 0 to 2) made it very difficult to use the other systems. Three different solvent extraction systems were tested:

1. Tri-iso-octylamine (TIOA)--20% in CHCl_3 ,
2. Di(2-ethylhexyl)phosphoric acid (D2EHPA)--10% in toluene,
3. 2,4-pentanedione (acetylacetone)--20% in CHCl_3 .

The acetylacetone- CHCl_3 system is similar to that used by Steinbach and Freiser (83,84), and was the best of the systems tried. Ten ml. samples (aliquots) were extracted with 5 ml. portions of the acetylacetone- CHCl_3 mixture by adequate mixing for 2 minutes, centrifuging, and then removing the colored iron complex with the CHCl_3 layer. The procedure was repeated four or more times with the extraction mixture, then given a final wash with 5 ml. of CHCl_3 by mixing for 1 minute, centrifuging and decanting the CHCl_3 layer. If the CHCl_3 layer was not practically colorless, the sample was again treated with the acetylacetone- CHCl_3 mixture and the final wash repeated.

Recovery of calcium from calcium "spiked" test samples was $100 \pm 4\%$. The error in the determination of calcium in bone solution standards was, over 80 replications, $\pm 1.30\%$. Distilled water used for dilutions contributed only 0.116 $\mu\text{gm./cc.}$ of water. Calcium values are reported as milligrams of calcium per gram of ash:

$$\left(\frac{\text{mg. Ca}}{\text{gm. ash}} \right)$$

The phosphorous content of all samples was also determined, using the Gomori modification of the method of Fiske and Subbarow (85,86). The analyses were made using aliquots of the same solutions used for calcium determinations. Test extractions of phosphorous from phosphorous "spiked" test samples was $100 \pm 3\%$. The error in determination of the phosphorous content of the bone solution standard used was, over six replications, $\pm 1.71\%$. Distilled water used for dilutions did not contain detectable amounts of phosphorous.

Phosphorous values are reported as milligrams of phosphorous per gram of ash:

$$\left(\frac{\text{mg. P}}{\text{gm. ash}} \right)$$

DATA AND DISCUSSION

Data and discussion are presented on: (1) the preliminary investigations, (2) the skeletal burden of radium-226 in swine, (3) comparison of radium-226 burden in the bone of lymphosarcomatous and overtly normal swine, and (4) the metabolism of and physiological discrimination against radium-226 in swine.

All data derived from assays of the radium-226 content are reported as picocuries (pCi) or femtocuries (fCi) per gram of ash or per gram of calcium where:

1 picocurie (pCi) = 1×10^{-12} curies = 1 micromicrocurie

1 femtocurie (fCi) = 1×10^{-3} picocuries = 1×10^{-15} curies

Preliminary Investigation

When the apparatus had been constructed and the counting chambers calibrated, some preliminary studies were made to determine the sensitivity of the system and to determine what the ranges of sample size required might be.

Eight samples of water were obtained from sources in Southeastern Kansas where the water has been used or is being used for livestock. It was known to have a high radium-226 content in some instances. Bone samples were obtained from a slaughter house in the Chicago area.

These samples were used to develop the techniques used during this study. The water samples were handled as large dilute samples, just adding enough hydrochloric acid to acidify the water. The bones were processed as indicated in Appendix C.

The radium-226 content of the eight Southeastern Kansas waters (Table 1) appears to be related to the depth of the well and the aquifer supplying the well as has been noted by other investigators (34,38,40,49,87,88). The calcium content of the water, however, does not appear to be very closely related to the depth of the well as is the radium-226. Therefore, the radium-226 calcium ratio varied considerably from sample to sample in a way that could not be predicted.

In the light of these observations, it seemed prudent to make the metabolism studies in areas where there was a source of water with a constant $^{226}\text{Ra}/\text{Ca}$ ratio from a well defined aquifer. In this way the variation introduced by fluctuations in the water table would be small. The Central Illinois, Central Indiana, and Eastern Iowa and Nebraska Area was most suitable.

The bone samples we obtained could be identified only on the basis of what portion of the state they originated from. Many of them are included and located in another portion of this report (Table 3).

During the ashing, a carbon residue was left in some of the samples. This residue could be driven off by extending the duration of ashing at 600°C . to 800°C . or by increasing the ashing temperature to 900°C . The ashing temperature has a profound effect on the per cent calcium per gram ash. As reported by Neuman and Neuman (89) and McLean and Budy (90) below 600°C . weight change is due to loss of water and hydroxyl ions. Between 600° and 800°C . there is a change in the x-ray diffraction pattern from that of hydroxyapatite to that of β -tricalcium phosphate. Hydroxyapatite $(\text{Ca}_{10}[\text{PO}_4]_6[\text{OH}]_2)$ contains 39.6% calcium while β -tricalcium phosphate $(\text{Ca}_3[\text{PO}_4]_2)$ contains 38.5% calcium.

Thus, it can be seen, that the temperature at which the bone was ashed in combination with any deficiencies in the lattice have a great influence on the amount of calcium which will be found in the subsequent

TABLE 1

RADIUM-226/CALCIUM RATIOS IN SOME SOUTHEASTERN KANSAS WATERS
THAT HAVE BEEN USED TO WATER LIVESTOCK

Source	Depth of Well	pCi ^{226}Ra /liter	mgm. Ca/liter	pCi ^{226}Ra /gm. Ca
Swilley	70-80 ft.	0.574 $\pm$ 0.016	126.2	4.55
Clark	65 ft.	0.186 $\pm$ 0.009	153.2	1.21
Rust	1,000 ft. "shot off" at 25 ft.	0.356 $\pm$ 0.011	74.1	4.81
Graybill	260+ ft.	0.510 $\pm$ 0.016	121.4	4.20
Pioneer	850 ft.	3.78 $\pm$ 0.19	21.8	173.00
Wagner	1,100 ft.	162.6 $\pm$ 2.8	1776.8	91.54
Brown	1,200 ft.	128.9 $\pm$ 3.1	665.8	193.62
Williams	1,220 ft.	1.08 $\pm$ 0.03	73.3	14.73

analysis. With this in mind, it was decided to use an ashing temperature of 600° to 700° C. for ashing bone.

Techniques for processing other types of samples were developed as the need arose.

Skeletal Distribution of Radium-226 in Swine

Individual bones or groups of bones from the skeletons of eight or more animals were analyzed for radium-226 content. In the assay homologous bones from each side of the skeleton were combined and aliquots of the mixed ash taken for analysis. For convenience all bones of the manus and pes¹ were combined.

The data obtained for radium-226 content per gram of ash are presented in Appendix . Since the radium-226 burdens of the individual skeletons varied widely, the values were normalized. In human studies (44), where the long bones constitute 60% or more of the human skeleton (48,91), the technique of normalizing radium-226 content against the mean value for the long bones has been used. However, this was not appropriate for swine since the long bones constituted slightly less than 50% of the skeleton in the three complete swine skeletons analyzed. Therefore, the mean value for the skeleton was used as the reference value rather than the mean value for the long bones of the skeleton. The normalization was done by dividing the value computed for an individual bone or group of bones by the computed mean value for that skeleton. The normalized values are presented in Table 2.

From the data presented in Table 2, it was shown that even though the skeletons varied greatly in radium content, the normalized radium-226 burden of the bones was within $\pm 20\%$ of the mean value for the skeleton. This $\pm 20\%$

¹The manus is composed of the carpals, metacarpals, and associated phalanges and the pes is composed of the tarsals, metatarsals, and associated phalanges.

TABLE 2
DISTRIBUTION OF ^{226}Ra IN SWINE SKELETONS
NORMALIZED^a FOR COMPARISON

Summary Table

Bone	N	Normalized Mean $\pm$ E ^b ^c
Skull	11	96.1 $\pm$ 13.9
Mandible	9	98.4 $\pm$ 7.0
Scapula	14	96.0 $\pm$ 12.6
Humerus	14	107.6 $\pm$ 11.8
Radius and ulna	14	104.1 $\pm$ 13.0
Mamms	8	110.0 $\pm$ 5.2
Spine	14	103.2 $\pm$ 15.5
Ribs	14	92.4 $\pm$ 10.9
Sternum	8	86.5 $\pm$ 6.3
Pelvis	14	97.1 $\pm$ 9.0
Femur	14	107.9 $\pm$ 12.0
Tibia and fibula	14	104.0 $\pm$ 15.6
Pes	8	109.4 $\pm$ 7.0
Tail	8	115.2 $\pm$ 5.6

N = Number of samples assayed.

^a Normalized value = $\frac{\text{individual bone value}}{\text{skeletal mean value}}$.

^b E = 95% confidence limits.

^c Computed as suggested by Dean and Dixon (99).

variation is in agreement with the report by Walton et al. (44) on the distribution of radium-226 in human bones. This value of $\pm 20\%$ is also in agreement with observations in young animals and human fetuses of $\pm 10\%$ to $\pm 30\%$ (92,93,94) and in adult sheep and goats of $\pm 16\%$ to $\pm 30\%$ (95). Both sets of findings, however, are at variance with the values reported by Schulert et al. (91) who show a variance of $\pm 90\%$ to $\pm 60\%$ in the distribution of strontium-90 in human bones and Kulp (96) and Kulp et al. (97) who found a variance of about $\pm 110\%$ to $\pm 55\%$. Dubrovina et al. (95) also found that the strontium-90 distribution in adult human bones was extremely variable both between and within bones. Kshirsagar et al. (98) showed that 10 minutes after the injection of strontium-90 in the rabbit, the distribution in the individual bones of the skeleton was $\pm 30\%$ to $\pm 70\%$. However, 460 days after the injection, the distribution was $\pm 7\%$ to $\pm 16\%$. Thus, the wide variations noted may be just the expression of the lack of equilibrium in the divalent cation homeostatic systems.

The discrepancies noted in distribution of radium-226 in the various bones of the swine skeleton might have resulted from earlier completion of calcification in the case of skull, manus, and pes. In this case it would reflect the radium-226 levels in the diet of the dam during pregnancy and/or their diet as newborn animals. These bones may have completed calcification while dietary radium-226 was low. Alternatively there is the possibility that skull, ribs, manus, and pes contain a higher proportion of trabecular bones to compact bone than do the other bones of the skeleton. Bauer (100) has reported that for strontium the turnover rate for trabecular bone is about four times as fast as for compact bone. A similar difference has been observed by many investigators (89,95,101,102). If this was the case for radium-226 in these swine, then the cancellous bones reflected the radium-226 levels of

the current diet. It is believed that the ribs, at least, with a turnover time of about 100 days (79), did more nearly reflect the radium-226 content of the current diet. However, as suggested by Dubrovina et al. (95):

1. The activity of a part of a bone cannot serve as a basis for estimation of the activity present in the entire skeleton, a whole bone must be used.

2. The ratio between radioisotope content per gram of calcium in individual bones and the radioisotope content of the skeleton varies with time (and diet) and, therefore, cannot serve to evaluate long term skeletal activity. A single whole bone can be used to estimate (within $\pm 20\%$) the total skeletal activity only at that moment in time when the bone was obtained.

On the basis of the above data, it was decided that samples of bone selected to determine the radium-226 burden should include: (1) a sample of humerus, radius, and ulna, femur or tibia and fibula, to indicate the skeletal mean value, and (2) a sample of rib to demonstrate the effect of the current diet.

Comparison of the Skeletal Radium-226 Burden
in Overtly Normal and in Lymphosarcomatous
Swine

The observation that radium-226 causes injury to cells of the hemato-poietic, lymphocytic, and myelocytic series has been reported by numerous investigators (103-106). Shiels (107) reported that doses of ionizing radiation of the order of 85 mr per week has significant effects on the blood of humans. Schraub et al. (26) have estimated a total body dose from radium-226 and its daughters in the body of 75 to 350 mr per year. This value is much lower than the maximum permissible dose of 300 mr per week suggested by the ICRP (25). The body burden in swine is probably similar to that of man, since the

radium-226 burden per gram of skeletal ash is similar (34-44). Therefore, the dose rate in man and swine is probably of the same order of magnitude.

To investigate the possibility of a correlation between radium-226 burden in bone and lymphosarcoma in swine, bone samples were obtained from normal and leukemic animals originating in different geographic areas of the state of Illinois. Animals from various sections of the state were chosen because aquifers of St. Peter and Galesville sandstones (Ordovician and Cambrian geological systems) reach to the surface or are near the surface in many widely distributed locations in the state. The radium-226 content of the water from these aquifers may run as high as 15 pCi/liter (49). The radium-226 skeletal burden of swine from areas with water derived from St. Peter or Galesville sandstone aquifers should be higher than those of swine from areas supplied with waters containing less radium-226. Observed differences could be ascribed to high radium-226 levels in the feed or water. A correlation between skeletal burden of radium-226 and lymphosarcoma in swine might have been expressed in those animals with the highest skeletal burdens.

The data gathered in this investigation is presented in Table 3, with some data obtained from apparently normal animals in the other studies.

There was an obvious correlation between the radium-226 burden and the geographic origin of the swine, but none between the radium-226 burden and the occurrence of lymphosarcoma. A comparison of the maximum and minimum skeletal radium-226 burden in all swine investigated (including one from Nebraska) was made in Table 4.

Only a whole skeleton value was available for sample Normal No. 2 and its value was expected to be within $\pm 20\%$ of the value for the humerus.

Since the maximum and minimum values for skeletal burden of radium-226 in lymphosarcomatous swine was within the range of values for normal swine, it

TABLE 3

CORRELATION BETWEEN ORIGIN AND SKELETAL BURDEN OF RADIUM-226
IN OVERTLY NORMAL AND IN LYMPHOSARCOMATOUS SWINE

Bone Burden in $\frac{\text{FCI } ^{226}\text{Ra}}{\text{gm. ash}} \pm E^a$

Bone Analyzed	Northern Illinois				
	Leukemic No. 1	Leukemic No. 2	Leukemic No. 5	Leukemic No. 9	Leukemic No. 11
Whole skeleton	64.4				
Humerus		47.0 $\pm$ 3.0	54.7 $\pm$ 0.6		40.1 $\pm$ 1.2
Rib		40.0 $\pm$ 2.3	46.2 $\pm$ 0.7	42.9 $\pm$ 1.1	
Mandible					31.3 $\pm$ 1.0
	Normal No. 1	Normal No. 3	Normal Sow No. 4	Normal Sow No. 2	Normal Sow No. 1
Whole skeleton	41.8				
Humerus		42.2 $\pm$ 0.8	54.4 $\pm$ 2.3	40.6 $\pm$ 1.8	59.7 $\pm$ 2.8
Rib		36.0 $\pm$ 0.7	48.7 $\pm$ 2.1	38.6 $\pm$ 1.7	46.7 $\pm$ 1.5
North Central Illinois					
	Leukemic No. 4	Leukemic No. 6	Leukemic No. 8	Leukemic No. 10	
Humerus	17.2 $\pm$ 0.4	9.0 $\pm$ 0.4			
Rib	15.4 $\pm$ 0.5	7.9 $\pm$ 0.7	8.8 $\pm$ 0.4	17.5 $\pm$ 0.7	

TABLE I
MAXIMUM AND MINIMUM SKELETAL BURDEN OF RADIUM-226
IN SWINE IN THIS STUDY

Bone Analyzed	Normal		Lymphosarcomatous	
	Maximum	Minimum	Maximum	Minimum
	Normal No. 2	Barrow 2-1	Leukemic No. 7	Leukemic No. 6
	North Western Illinois	North Central Illinois	Omaha, Nebraska	North Central Illinois
Whole skeleton	214 ± 6			
Humerus		4.4 ± 0.1	125 ± 2	9.0 ± 0.4
Rib		4.1 ± 0.1	123 ± 3	7.9 ± 0.7

Bone burden reported as $\frac{\text{fCi } ^{226}\text{Ra}}{\text{gm. ash}} \pm E.$

E = 95% confidence limits.

was concluded that the skeletal burden of radium-226 was not an obvious factor in the incidence of lymphosarcoma in swine. However, it must be recognized that unless the incidence of lymphosarcoma induced by radium-226 is very high it would not be shown in the small sample of the population available to us.

Metabolic Fate of Radium-226 in Swine

The metabolic fate of radium-226 is complex. It is a function of accretion, retention, exchange, and diffusion across physiological boundaries and is intimately related to the current calcium balance of the animal.

Stover et al. (63) have stated that the metabolism of radium is qualitatively the same as that of calcium, but quantitatively unique. This has also been claimed by Norris and Kisielewski (108) for strontium-89, 90 and radium-226.

Marshall (109) has proposed a power function model to explain alkaline earth metabolism, including radium-226. He postulates the domination of alkaline earth metabolism by a complex deposition system for calcium and calcium-like elements, especially in bone. Deposition of calcium occurs primarily in submicroscopic crystals ($25-75 \text{ \AA}$ in diameter) of hydroxyapatite in the bone. The deposition of calcium from blood will occur in any calcifiable matrix and there is mobilization of calcium whenever osteoclastic activity resorbs the matrix. Smaller amounts of calcium might be deposited or mobilized from extracellular fluids, crystal surfaces and even the crystals deep within the bone. Other tissues will also contribute calcium within the homeostatic system of the body. The various exchange processes involving calcium thus form a complex distribution system, the pathways of which have turnover times of various lengths. An alkaline earth cation, therefore, may be excreted or it

may leave the blood for another locus, to return to the blood in minutes, days, or even years.

Several investigators (110-115) have suggested the value of the power function model in studies of the metabolism of radium-226. They have also suggested that deviation from a power function during the first few weeks after administration of a tracer dose of an alkaline earth radioisotope, may be caused by the requirements for the equilibration of the homeostatic system exchanging alkaline earth cations with the skeleton. Certain matters should be discussed before the power function model is applied.

In this part of the study the animals used had been on the same diet for at least 56 days and the diet over their life time was alike and well known. With the exception of minor variations caused by uneven mixing of feed or variations in the radium-226 content of the water supply, the diet was held constant during the entire study period. The length of the period the animals were on the constant diet should have allowed the animals to attain a quasi-steady state equilibrium¹ in most alkaline earth exchange systems.

A summary of radium-226 content of feed, water, tissue, secretions, and excretions for eight barrows from the University of Illinois and eight sows from Frankfort, Illinois, are listed in Table 5. Table 6 lists the average per cent distribution of radium-226 in the excreta of the eight barrows over the 7-day collection period. Microscopic examination of the residue remaining after fecal ash samples were dissolved and filtered showed that this residue was composed of worn silicates commonly found in soil. Quantities as large as 75 gm. of these worn silicates were recovered. Since the amount of radium-226 biologically available in the soil was unknown, it was decided to treat all radium-226 recovered as being biologically available. For this

¹A complete steady state equilibrium is not attained since the rate of change of equilibrium is extremely slow over the period of the experiment.

TABLE 5

RADIUM-226 CONTENT OF FEED AND WATER, TISSUES, SECRETIONS
AND EXCRETIONS FOR ALL ANIMALS ASSAYED

Summary Table

Sample	fCi ^{226}Ra /gm. ash $\pm$ E ^a	Sample	fCi ^{226}Ra /gm. ash $\pm$ E ^a
Sows		Boars	
Skull	45.0 $\pm$ 5.8	Humerus	6.1 $\pm$ 1.7
Mandible	49.0 $\pm$ 5.8	Ribs	5.0 $\pm$ 1.4
Scapula	46.0 $\pm$ 5.2	Blood	7.9 $\pm$ 3.9
Humerus	51.5 $\pm$ 7.0	Muscle	3.0 $\pm$ 2.0
Radius and ulna	48.5 $\pm$ 6.1	Urine	36.5 $\pm$ 8.7
Mamms	52.5 $\pm$ 6.7	Feces	107.0 $\pm$ 42.3
Spine	46.5 $\pm$ 4.9		536.0 $\pm$ 166.8
Ribs	45.0 $\pm$ 5.5	Feed for Boar Groups	
Sternum	38.0 $\pm$ 6.1	Feed I	180.0 $\pm$ 5.0
Pelvis	46.0 $\pm$ 4.6	Water I ^b	136.0 $\pm$ 5.0
Femur	48.0 $\pm$ 4.9	Feed II	204.0 $\pm$ 5.0
Tibia and fibula	46.0 $\pm$ 5.5	Water II ^b	66.0 $\pm$ 2.0
Pes	50.0 $\pm$ 6.7	Feed for Sows	
Tail	53.5 $\pm$ 8.4	Goodenow feed	346.0 $\pm$ 15.0
Skeleton	45.6 $\pm$ 8.4	Goodenow water ^b	48.0 $\pm$ 2.0
Blood	7.0 $\pm$ 1.8	Frankfort feed	27.0 $\pm$ 1.0
Milk	11.0 $\pm$ 7.8	Frankfort supplement	1750.0 $\pm$ 64.0
Muscle	3.9 $\pm$ 2.3	Frankfort water ^b	400.0 $\pm$ 13.0
Skin and fat	17.0 $\pm$ 6.7		
Lung	26.0 $\pm$ 3.5		
Liver	1.7 $\pm$ 0.8		
Heart	2.4 $\pm$ 1.0		
Kidney	5.4 $\pm$ 4.7		

^aE = 95% confidence limits.

^bfCi ^{226}Ra /liter.

TABLE 6
 AVERAGE PER CENT DISTRIBUTION OF EXCRETED
 RADIUM-226 IN URINE AND FECES FOR A
 7-DAY PERIOD

Animal Assayed	Per Cent in Urine	Per Cent in Feces
Group 1		
Barrow 1-1	0.81	99.19
Barrow 1-2	1.24	98.76
Barrow 1-3	3.94	96.06
Barrow 1-4	2.44	97.56
Group 2		
Barrow 2-1	2.60	97.40
Barrow 2-2	3.24	96.76
Barrow 2-3	1.48	98.52
Barrow 2-4	1.07	98.93
Mean	1.96 ± 0.72	98.04 ± 0.72

reason the measured soil content of radium-226 was included in the consumption of radium-226 to balance consumption and excretion, since if the metabolism had equilibrated, there should be no net loss of radium-226. The total amount of radium-226 reported as consumed is a minimum value since an undetermined amount of radium-226 may have been absorbed and retained by each animal.

The mean values obtained of $98.04 \pm 0.72\%$ excreted in the feces and $1.96 \pm 0.72\%$ voided in the urine are closer to those reported for human (22) of $\approx 98\%$ in feces and $\leq 2\%$ in urine than to the values reported for the rat of 81-88% in feces, 5-10% in urine (108). This comparison shows one of the interspecies differences and similarities in metabolism of radium-226. Single dose intravenous studies would be required to further measure the difference.

In Table 5 the radium-226 content of feed, water, tissues, and secretions of the eight sows from Frankfort, Illinois, differs slightly from that of the eight barrows from the University of Illinois. In this group of eight sows the radium-226 per gram of ash in the bones appears to reflect more of the radium-226 content of the feed in the diet than the water. Lucas (38) has reported that the total body burden in humans may be approximately equal to 22 times the daily intake of radium-226 in food plus the radium-226 content of 50 liters of the water consumed. In these eight animals the average body burden was 214.8 pCi of radium-226. This was about two times the daily intake of radium-226 in feed or the radium-226 content of about 537 liters of water.

On the other hand, the eight barrows from the University of Illinois had an average total body burden of 15.07 pCi of radium-226. This corresponds to about one and one-half times the daily intake of radium-226 in feed or the radium-226 content of about 100 liters of water. If the availability of radium-226 is the same in swine as it is in humans where the radium-226 in

water is two times as available as that in food (79), then the feed contributes between 85% and 90% of the total body burden of these animals. On this basis the total body burden for the sows would be 1.70 times the daily intake in feed plus 4.19 times the radium-226 content of 1 liter of water. The radium-226 total body burden for the barrows would be 1.20 times the daily food intake plus 9.23 times the content of 1 liter of water.

A general equation for a power function model of retention of radium-226 can be derived as follows:

$$R_t = At^{-b} \quad \text{where } t \geq A^{1/n} \quad \text{and } 0 < b < 1$$

where

A = fraction of the assimilated dose of ^{226}Ra retained when $t = 1$ day,

t = time after the assimilation of a dose of ^{226}Ra ,

b = slope of the linear log-log transform of the retention curve,

R_t = fraction of assimilated dose retained t days after assimilation.

The total body burden of ^{226}Ra at the time of slaughter was computed for each animal and the results averaged to give an R_t which allowed us to develop a power function model. The total ^{226}Ra present in the food and water was also computed as follows:

1. For the barrows

2,100 gm. bone ash	12,571.2 fCi
250 gm. muscle ash	948.4 fCi
75 gm. blood ash	688.0 fCi
25 gm. fat ash	663.8 fCi
50 gm. pluck ¹ ash	310.7 fCi
Radium-226 total body burden	15,072.1 ± 3,032.8 fCi
Radium-226 consumed/day in feed	7.84 fCi
Radium-226 consumed/day in water	0.15 fCi

¹ Contents of the thoracic and abdominal cavities.

2. For the sows

4,270 gm. bone ash	211,098.1 fCi
300 gm. muscle ash	1,396.7 fCi
88 gm. blood ash	806.9 fCi
68 gm. fat ash	988.5 fCi
75 gm. pluck ash	492.1 fCi
Radium-226 total body burden	214,785.0 $\pm$ 29,253.4 fCi
Radium-226 consumed/day in feed	107.54 pCi
Radium-226 consumed/day in water	8.00 pCi

Data were not available from this study to employ the general power function model. However, it was possible to employ a modification developed by Halden et al. (116) from the power function model for radium-226 first described by Norris et al. (117). To use data developed in this study to calculate such a model, it was further assumed that:

1. Human and porcine radium excretion patterns are similar (in this study it appeared to be true).

2. Absorption of radium-226 is identical in human and porcine (we have no information on this assumption).

The expression for the power function retention model in swine then was:

$$R_t = C [1.12(t^x - 1) + 0.54] = 0.54 t^{-(1-x)}$$

where

R_t = pCi of radium-226 in the body,

t = time in days,

C = pCi of radium-226 metabolized per day (10% of radium-226 ingested per day),

x = slope of the linear log-log transform of the retention function.

A. For the borrows over a 180-day life span

$$15.07 \text{ pCi} = 0.80 \text{ pCi} [1.12 (180^x - 1) + 0.54]$$

$$180^x = 17.34$$

$$x = 0.55$$

$$R_t = 0.54 t^{-0.45}$$

B. For the sows over a 480-day period on sow diet

$$\begin{aligned} 214.8 \text{ pCi} &= 11.55 \text{ pCi} [1.12 (480^x - 1) + 0.54] \\ 480x &= 17.11 \\ x &= 0.46 \\ R_t &= 0.54 t^{-0.54} \end{aligned}$$

If all assumptions made are valid, then the power function model for retention in adult swine ($R_t = 0.54 t^{-0.54}$) is very similar to that of the adult human ($R_t = 0.54 t^{-0.52}$). The assumption of 0.54 as the initial retention at one day after the start of exposure to the radium-226 may well be incorrect. Norris and Kisielewski (108) report a value of 0.80 in rats, Miller and Finkel (115) a value of from 0.605 to 0.797 in mice, and Van Dilla et al. (71) a value of 0.787 in dogs. If a value of 0.80 is used as original retention, the power function model for radium-226 in swine is then $R_t = 0.80 t^{-0.54}$. This function still resembles the human function more closely than any other species. For example, $R_t = 0.787 t^{-0.198}$ in the dog (71), $0.605 t^{-0.233}$ to $0.797 t^{-0.306}$ in the mouse (115), and $0.80 t^{-0.229}$ in the rat (108). In young swine the models for the different initial retention values are: $R_t = 0.54 t^{-0.45}$ and $R_t = 0.80 t^{-0.45}$ respectively. It is probable that the reduced slope (reduced rate of excretion) observed in the young swine is a reflection of increased accretion due to the rapidly enlarging skeletal mass. Further studies using single intravenous doses of radium-226 would be required to test this assumption.

The presence of specific preferential barriers for radium-226 can be intimated from calculated "observed ratios." Comar et al. (118) proposed the use of the term "strontium-calcium observed ratio" (OR) in double tracer studies using calcium-45 and strontium-89. Comar et al. (119) previously had used strontium-90 and stable calcium fed in a constant ratio to study differential metabolism of strontium-90 and calcium. Finally, Comar et al. (120) extended the concept of OR's to include a single tracer in combination with

stable calcium in equilibrium. Hallden et al. (116) have described OR's for radium-226 in the human, where radium-226 was related to stable calcium.

In this study an analogous procedure was used with radium-226 at equilibrium where:

1. The number of picocuries of radium-226 per gram calcium in the sample is called the radium-calcium ratio (Ra/Ca).
2. The radium-calcium observed ratio (OR) is,

$$OR_{\text{sample-precursor}} = \frac{\text{Ra/Ca of sample}}{\text{Ra/Ca of precursor}} .$$

The OR is used as Comar first used it to denote the over-all discrimination observed in the movement of the two elements across one or more physiological boundaries or barriers. The use of this model to describe selective preferential barriers at physiological boundaries is valid where the calcium intake has been relatively constant and all calcium systems are in equilibrium.

Kornberg (121) points out some of the limitations of the use of a ratio of pairs of elements to predict concentrations of the pair at different points in a system. Of most interest is his observation that as the concentration of one member of the pair increases, the other member does not necessarily decrease proportionately. He also observed that time and mixing effects influence the ratio. His criticism is valid principally in the non-equilibrium condition.

In this study the ratio of radium-226 to calcium in feed and water had a minimum of variation. All rations were balanced and adequately mixed. The effect of the length of time on animal was on a specific diet cannot be directly measured nor can the effect of the age of the animal, but they may be reflected in the difference observed in the power function computed for the young barrows when compared to that of the more mature sows. It has been

reported that both of these factors influence strontium-90 OR's (122-126) and so might be expected to have some effect on radium-226 OR's.

As a preface to the discussion of OR's, several facts should be pointed out. Wasserman (127) states that there is an active transport system for calcium which does not transport strontium. Logically, this transport mechanism might be expected to discriminate against radium-226. Twardock and Comar (128) have observed an unidirectional barrier to strontium-90 being secreted by the mammary gland. There was a barrier against secretion of strontium-90 from blood to milk, but strontium-90 could enter the blood as freely as calcium-45 when both were introduced into the gland via the teat canal. Twardock (129) further found active transport of calcium and selective discrimination against strontium by the mammary gland. He also noted that strontium is 10 to 20 per cent more ultrafiltrable than calcium in cow and goat plasma. Since there is less bound strontium than calcium in the plasma, the mammary gland must be actively discriminating against the strontium.

It has been shown that calcium, strontium, and cesium readily cross the placenta (130-133), reaching a steady state 16 to 30 hours after administration. In this steady state the ratio of these elements in maternal blood to these elements in fetal blood approached 1 to 1. It is reasonable to assume that radium-226 will cross the placenta with equal ease. Therefore, the radium-226-calcium ratio of maternal blood has been considered to be the ratio for fetal blood in this study.

Ratios of radium-226 to calcium and to phosphorous are listed in Tables 7 and 8. OR's of sample radium-226 to precursor radium-226 are listed in Tables 9 and 10. OR's for young animals, neonate to age 4 weeks, are listed in Table 11. Finally, the OR's of radium-226 calculated for swine in this study are compared in Table 12 to the OR's for strontium-90 as calculated by several investigators.

TABLE 7

RATIO OF RADIUM-226 TO CALCIUM IN FEED AND WATER, TISSUES, SECRETIONS
AND EXCRETIONS FOR ALL ANIMALS ASSAYED

Summary Table

Sample	fCi $^{226}\text{Ra}/\text{gm. Ca} \pm E^a$	Sample	fCi $^{226}\text{Ra}/\text{gm. Ca} \pm E^a$
Sows		Boars	
Skull	116.7 $\pm$ 14.9	Humerus	15.1 $\pm$ 1.9
Mandible	121.7 $\pm$ 15.4	Ribs	13.3 $\pm$ 4.1
Scapula	112.9 $\pm$ 12.7	Blood	907.1 $\pm$ 551.6
Humerus	131.4 $\pm$ 17.9	Muscle	403.5 $\pm$ 212.0
Radius and ulna	121.0 $\pm$ 14.6	Gut	1345.0 $\pm$ 274.8
Manus	134.0 $\pm$ 17.5	Urine	262.0 $\pm$ 81.0
Spine	121.1 $\pm$ 13.3	Feces	2979.7 $\pm$ 783.0
Ribs	114.7 $\pm$ 14.2	Feed for Boar Groups	
Sternum	101.2 $\pm$ 17.0	Feed I	1293.1
Pelvis	118.0 $\pm$ 12.8	Water I	5619.8
Femur	124.7 $\pm$ 12.8	Diet I	1324.7
Tibia and fibula	117.3 $\pm$ 14.2	Feed II	1280.6
Pes	130.6 $\pm$ 17.0	Water II	2101.9
Tail	139.5 $\pm$ 19.5	Diet II	1287.5
Skeleton	118.4 $\pm$ 38.6	Feed for Sows	
Blood	963.5 $\pm$ 124.9	Goodenow feed	1551.6
Milk	42.1 $\pm$ 32.7	Goodenow water	625.0
Muscle	296.4 $\pm$ 129.1	Goodenow diet	1482.0
Skin and fat	477.9 $\pm$ 125.5	Frankfort feed	1708.9
Lung	494.8 $\pm$ 170.7	Frankfort	
Liver	107.1 $\pm$ 36.0	supplement	6515.3
Heart	55.4 $\pm$ 20.1	Frankfort water	3838.8
Kidney	161.2 $\pm$ 41.1	Frankfort diet	6156.3

^aE = 95% confidence limits.

TABLE 8

RATIO OF RADIUM-226 TO PHOSPHOROUS IN FEED AND WATER, TISSUES,
SECRETIONS AND EXCRETIONS OF ALL ANIMALS ASSAYED

Summary Table

Sample	fCi $^{226}\text{Ra/gm. Ca} \pm E^a$	Sample	fCi $^{226}\text{Ra/gm. Ca} \pm E^a$
Sows		Boars	
Skull	256.6 $\pm$ 32.6	Humerus	31.9 $\pm$ 3.7
Mandible	269.1 $\pm$ 34.2	Ribs	27.2 $\pm$ 8.1
Scapula	245.5 $\pm$ 30.5	Blood	213.7 $\pm$ 120.4
Humerus	281.9 $\pm$ 37.3	Muscle	15.0 $\pm$ 9.9
Radius and ulna	262.6 $\pm$ 31.2	Urine	206.0 $\pm$ 45.6
Manus	294.8 $\pm$ 39.7	Feces	138.0 $\pm$ 39.0
Spine	257.6 $\pm$ 27.7		3078.2 $\pm$ 569.6
Ribs	251.5 $\pm$ 32.4	Feed for Boar Groups	
Sternum	215.7 $\pm$ 35.4	Feed I	1,154.6
Pelvis	255.9 $\pm$ 28.1	Water I	340,000.0
Femur	264.4 $\pm$ 29.8	Diet I	1,180.1
Tibia and fibula	254.8 $\pm$ 32.0	Feed II	1,275.0
Pes	283.0 $\pm$ 38.7	Water II	165,000.0
Tail	294.8 $\pm$ 46.5	Diet II	1,289.4
Skeleton	256.0 $\pm$ 84.4	Feed for Sows	
Blood	164.9 $\pm$ 55.9	Goodenow feed	1,663.5
Milk	71.3 $\pm$ 42.1	Goodenow water	106,666.7
Muscle	20.1 $\pm$ 13.8	Goodenow diet	1,033.1
Skin and fat	150.8 $\pm$ 59.4	Frankfort feed	151.5
Lung	155.9 $\pm$ 18.8	Frankfort supplement	15,391.4
Liver	6.8 $\pm$ 3.1	Frankfort water	666,666.7
Heart	14.6 $\pm$ 6.8	Frankfort diet	9,577.0
Kidney	29.1 $\pm$ 27.9		

aE = 95% confidence limits.

TABLE 9
OBSERVED RATIOS BASED ON RADIUM TO CALCIUM RATIOS

Summary Table

OR	Mean $\pm$ E ^a	OR	Mean $\pm$ E ^a
	Sows		Boars
Humerus-diet	0.021 $\pm$ 0.003	Humerus-diet	0.012 $\pm$ 0.003
Rib-diet	0.018 $\pm$ 0.002	Rib-diet	0.010 $\pm$ 0.003
Blood-diet	0.638 $\pm$ 0.084	Blood-diet	0.704 $\pm$ 0.416
Muscle-diet	0.200 $\pm$ 0.088	Muscle-diet	0.304 $\pm$ 0.165
Skin and fat-diet	0.322 $\pm$ 0.085	Gut-diet	1.019 $\pm$ 0.203
Lung-diet	0.334 $\pm$ 0.120	Urine-diet	0.204 $\pm$ 0.061
Liver-diet	0.072 $\pm$ 0.025	Feces-diet	2.278 $\pm$ 0.579
Heart-diet	0.037 $\pm$ 0.014		
Kidney-diet	0.109 $\pm$ 0.028	Bone-blood	0.020 $\pm$ 0.014
Bone-blood	0.030 $\pm$ 0.008	Muscle-blood	0.330 $\pm$ 0.296
Muscle-blood	0.302 $\pm$ 0.136	Urine-blood	0.362 $\pm$ 0.471
Milk-blood	0.044 $\pm$ 0.022		
Skin and fat-blood	0.508 $\pm$ 0.172		
Lung-blood	0.598 $\pm$ 0.166		
Liver-blood	0.112 $\pm$ 0.069		
Heart-blood	0.062 $\pm$ 0.022		
Kidney-blood	0.165 $\pm$ 0.079		
Neonate-diet	0.016 $\pm$ 0.005		
Neonate-dam	0.882 $\pm$ 0.391		
Neonate-blood	0.028 $\pm$ 0.008		

^aE = 95% confidence limits.

TABLE 10
OBSERVED RATIOS BASED ON RADIUM TO PHOSPHOROUS RATIOS

Summary Table

OR	Mean $\pm$ E ^a	OR	Mean $\pm$ E ^a
	Sows		Boars
Humerus-diet	0.030 $\pm$ 0.004	Humerus-diet	0.025 $\pm$ 0.003
Rib-diet	0.026 $\pm$ 0.003	Rib-diet	0.023 $\pm$ 0.004
Blood-diet	0.160 $\pm$ 0.054	Blood-diet	0.176 $\pm$ 0.092
Muscle-diet	0.019 $\pm$ 0.005	Muscle-diet	0.016 $\pm$ 0.008
Milk-diet	0.069 $\pm$ 0.052	Gut-diet	0.163 $\pm$ 0.041
Skin and fat-diet	0.146 $\pm$ 0.057	Urine-diet	0.107 $\pm$ 0.033
Lung-diet	0.151 $\pm$ 0.018	Feces-diet	2.511 $\pm$ 0.533
Liver-diet	0.007 $\pm$ 0.003		
Heart-diet	0.014 $\pm$ 0.006	Bone-blood	0.196 $\pm$ 0.054
Kidney-diet	0.028 $\pm$ 0.024	Muscle-blood	0.069 $\pm$ 0.050
		Urine-blood	0.420 $\pm$ 0.327
Bone-blood	0.147 $\pm$ 0.059		
Muscle-blood	0.130 $\pm$ 0.044		
Milk-blood	0.327 $\pm$ 0.294		
Skin and fat-blood	0.750 $\pm$ 0.382		
Lung-blood	0.944 $\pm$ 0.348		
Liver-blood	0.036 $\pm$ 0.011		
Heart-blood	0.089 $\pm$ 0.040		
Kidney-blood	0.178 $\pm$ 0.119		
Neonate-diet	0.046 $\pm$ 0.013		
Neonate-dam	1.696 $\pm$ 0.938		
Neonate-blood	0.288 $\pm$ 0.087		

^aE = 95% confidence limits.

TABLE 11
OBSERVED RATIOS FOR NEONATE, 2-WEEK OLD AND 4-WEEK
OLD SWINE

	fCi $^{226}\text{Ra}/\text{gm. ash} \pm E^a$	fCi $^{226}\text{Ra}/\text{gm. Ca} \pm E^a$	Mean $\text{OR}_{\text{Neonate-diet}} \pm E^a$
For OR's Based on Radium to Calcium Ratio			
Neonate	9.0 $\pm$ 2.6	26.3 $\pm$ 7.1	0.018 $\pm$ 0.005
2 weeks	17.8 $\pm$ 8.6	50.1 $\pm$ 25.6	0.034 $\pm$ 0.018
4 weeks	36.0 $\pm$ 11.0	102.0 $\pm$ 32.2	0.069 $\pm$ 0.021
For OR's Based on Radium to Phosphorous Ratio			
Neonate	9.0 $\pm$ 2.6	47.4 $\pm$ 13.3	0.046 $\pm$ 0.013
2 weeks	17.8 $\pm$ 8.6	94.4 $\pm$ 43.7	0.092 $\pm$ 0.043
4 weeks	36.0 $\pm$ 11.0	189.0 $\pm$ 58.5	0.179 $\pm$ 0.052

aE = 95% confidence limits.

TABLE 12

COMPARISON OF OBSERVED RATIOS FOR RADIUM-226 IN SWINE, BASED
ON RADIUM TO CALCIUM RATIOS, WITH OBSERVED RATIOS
REPORTED FOR STRONTIUM-90 IN SEVERAL
SPECIES

Observed Ratio Considered	Strontium-90 Observed Ratios in Human, Dog, Cow, Sheep, Goat, Rabbit, and Rat (118-120, 129, 131, 134-137)	Radium-226 Observed Ratios in This Study
Bone-diet	0.21 - 0.35 0.17 - 0.19 in swine (122)	0.010 - 0.02 ^a
Plasma-diet	0.18 - 0.52	0.638 - 0.704
Milk-diet	0.074 - 0.16	0.028
Fetus-diet	0.08 - 0.17 0.03 in swine (138)	0.018 ^b
Urine-diet	1.1 - 4.3	0.204
Feces-diet	1.2	2.278
Milk-plasma	0.40 - 0.80	0.044
Fetus-plasma	0.49 0.12 in swine (138)	0.028 ^b
Urine-plasma	7.4 - 16.0	0.362
Fetus-dam	0.40 - 0.71	0.882 ^b

^aHalden et al. reported an $OR_{\text{bone-diet}}$ of 0.015 - 0.016 for radium-226 in humans (116).

^bNeonatal values were used. These newborn animals were killed immediately after birth.

The OR's listed for blood to diet are the most variable of those computed, and differ considerably from the values noted by Lucas (79) in humans. The OR_{blood-diet} for radium-226 is near that reported for strontium-90 and suggests a similarity in mode of intestinal absorption. For all animals the diet contained calcium in excess of minimum requirements (139) so body depots should not have been disturbed. The variation might be due to a difference in metabolism of radium by various species.

It has been shown, for example, that in humans on a constant diet containing a strontium-85 tracer, that there can be a two-fold difference between the plasma strontium levels of individuals on the same diet (142). This study also suggested that there is a difference in the availability of strontium to individuals fed the same diet. It has also been reported elsewhere (143) that in rats the blood calcium remains constant over a wide range of daily absorption of calcium.

In this study the magnitude of differences in radium-226 OR's could have been augmented by the fact that whole blood was assayed instead of plasma. Stover et al. (144) have noted in dogs and Fink (145) in rats, that radium-226 is distributed both in the cells and in the plasma of blood, reaching a 1 to 1 ratio after a period of time. With these variations, possibly independent of calcium intake levels, the observation of a wide variation in blood-diet OR's is not unexpected.

However, the mean blood-diet OR of 0.638-0.734 is of the same order of magnitude as that reported recently for strontium in humans of 0.3 to 0.5 (146) or in rats of 0.68 (147). The same report (147) indicated that the presence of various sugars or amino acids in the diet also influenced the strontium OR plasma-diet. Further study is required to determine if one or more of these factors might be influencing the radium-226 OR_{blood-diet} in swine.

The OR's for neonatal swine were based on the diet received by the dams at Goodenow. The Goodenow diet value was used because the greatest accretion of calcium by the fetus (93 to 96 per cent) is in the last trimester (132,139) and the sows were at Goodenow during the last trimester. Similar observations have been made for strontium-90 administered to rats (140,141). A comparison of the bone-diet ratios for neonates and sows will suggest whether there is a placental barrier to radium-226 in swine. If there is no barrier, then the ratio of the $OR_{\text{bone-diet}}$ in neonates and the $OR_{\text{bone-diet}}$ in dams will be unity. In this study the ratio was 0.882, indicating that there is little or no placental barrier.

Although placental discriminations with an OR of about 0.50 have been reported by many investigators in many animals including humans (93,101,102, 119,120,148,149) for both stable strontium and strontium-90, this value may not be presenting a true picture. In investigations of radium-226 in humans, Rajewsky et al. (150) found that the specific activity of radium-226 in human fetal bones (mens IV to mens X) is identical to the specific activity in the bones of adult humans. Stahlhofen (151) further reports that the specific activity of human fetal bone and soft tissue is equal to that found in adult humans. He states, ". . . preferential discrimination of radium-226 by the placental barrier may not be assumed in these cases of natural activity level."

The ten-fold placental discrimination against alpha emitters noted by Marsden (152) in cattle and sheep and the three-fold placental discrimination against radium-226 in cattle and the thirty-fold discrimination in swine observed by Di Ferrante (153) when comparing maternal and fetal bones are probably artifacts induced by a change in diet.

In this study, when the absolute values of radium-226 in fetal and maternal bones are compared, there is a four- to ten-fold apparent placental

discrimination. But if the bones are compared to the diet fed during the time of calcification, then the maternal and fetal bone-diet OR's are compared, the apparent placental discrimination vanishes and is shown to be an artifact which can only be related to the diet.

Kriegel (154) found that carrier-free strontium-90 injected into rats was not obstructed by any "preferential" barrier at the placenta.

The fact that different placental types are involved is of no great importance. Recent electron microscopic studies of placentae have led investigators to the conclusion that the grasser classification system, Epithelio-chorial (6 layers, e.g., Pig) to Hemochorial (3 layers, e.g., Human) is unacceptable as a basis for comparative placental physiology. There is evidence that where fluid, ions, or other moieties are to be transported, especially against an osmotic gradient, there is an increase in development of: (1) convolution of syncytium plasma membranes (fluid transport); (2) vesiculation of basement membranes (ion transport); or (3) microvilli, pinocytosis, vesiculation and convolution of membranes (transport against osmotic gradient). In fact, a thicker anatomical "barrier" may, through increased metabolic activity, facilitate rather than impede transport of metabolites not transferred by simple diffusion. Finally, intimate contact of maternal and fetal circulations has also been observed even in the 6-layered placenta of the swine (155-161).

These observations indicate that the placental barrier "observed" at least at low levels of activity, is an artifact. Mole (148) has stated, "The ratio of perinatal bone to maternal bone will be a biased estimate of placental discrimination to the extent to which maternal plasma and maternal bone are not in equilibrium during pregnancy." Mole further places in evidence a human case where the $OR_{\text{fetal bone-maternal bone}}$ was 0.9 for stable strontium, while it was 2.5 for strontium-90.

The increase in OR's observed as the young piglets grow and increase in size is due to a number of factors. Since the animals assayed were all 4 weeks of age or less, they were still in a period where there was probably little or no intestinal barrier against radium. It has been shown that in many animals there is little or no intestinal barrier against strontium-90 at birth, but an increasingly active barrier as the animal ages (125,126,149,162-166). As has been mentioned by several investigators (147,163,167-169), many of the components of milk, especially lactose, stimulate the absorption of calcium and strontium from the intestine. If radium behaves as does calcium and strontium, the combination of a reduced discrimination with a stimulation of absorption over a varying diet of both milk and dry feed led to the increasing OR's by increasing the rate of accretion of radium-226 in bone while the rate of loss remained constant.

Milk is separated from plasma by a barrier against radium-226 which is much stronger than that against strontium-90. It has been shown that strontium-90 is rapidly secreted into the milk in swine (170) and equilibrium of excretion is rapidly attained in cattle (123,134,171). Other investigators have found that the $OR_{\text{milk-plasma}}$ for strontium-90 was about 0.40 in humans (172) and 0.33 in sheep (173), while that for radium-226 was about 0.20 in sheep (173). The fact that secretion of radium-226 into milk in this study was approximately an order of magnitude less than what might be expected based on strontium-90 OR's, suggests that there may be a species difference in secretion of radium-226 into milk.

In the soft tissues, it is quite possible that the OR's have no great significance. Calcium may be required for metabolic processes where another ion can replace it. It is also possible that $^{226}\text{RaSO}_4$ is formed in certain tissues. Since, as Schubert and Conn (174) have shown, $^{140}\text{Ba}^{++}$ ions can form

a radiocolloid with SO_4^{2-} ions even though the concentrations of ions do not exceed the solubility product for BaSO_4 . This is quite probably the reason for the high OR's observed in this study for skin and fat and lungs, and the report by Atherton et al. (175) that skin and trachea (i.e., the cartilage therein) retain relatively large quantities of radium-226. The SO_4^{2-} in the chondroitin sulfuric acid present in cartilage can react with some of the radium to form a radiocolloid and cause an increased retention of radium-226.

The much smaller OR's observed for muscle, heart, liver, and kidney may be but varying facets of the same simple mechanism. Bligh and Taylor (176) have shown that calcium, strontium, and barium are concentrated in the submaxillary salivary gland, barium being accumulated more than strontium. The accumulation of calcium by the submaxillary gland is known to be a process not dependent on oxidative phosphorylation (177) so not an active process. Again, if radium is handled in an analogous way, it is possible that phosphorylation reactions play a part in control of accumulation of radium.

If all cellular and subcellular membranes are considered to be both functionally and structurally similar, but with specific modifications or capacities, and both membranes are freely permeable to phosphate (178), then under physiological conditions calcium and phosphate are transported across the membrane as an ion pair.

Lehninger (179) suggests that in transport across the membrane of mitochondria, it is possible that during accumulation of Ca^{++} , Mg^{++} , Mn^{++} , Ba^{++} , and Sr^{++} , inorganic phosphorous (P_i) is the moiety that is being actively accumulated. Since P_i is taken up at the same time as the cations are being accumulated, the uptake of the cations may be a passive process related to the ability of the ions to penetrate, the specificity of ion-binding sites and the propensity of these ions to form insoluble phosphates within the mitochondrion.

Approximately 1.7 Ca^{++} molecules are accumulated for every 1.0 P_i molecule accumulated per pair of electron equivalents traversing a phosphorylation site. The 1.0 value for P_i is obviously in a 1:1 integral ratio with the phosphorylation sites, while the nonintegral value of 1.7 for Ca^{++} accumulation may be a reflection of the formation of a CaPO_4 precipitate within the mitochondrion. This stoichiometric relationship of P_i has also been reported in the case of Mg^{++} and Mn^{++} accumulation (180-185).

It has also been observed (186) that Mn^{++} binding at the surface of the mitochondrion is competitive with Ca^{++} and Mg^{++} binding and in the absence of P_i accumulation of Mn^{++} can be reversed by Ca^{++} or Mg^{++} . It is believed that in the presence of P_i $\text{Mn}_3(\text{PO}_4)_2$ precipitates, whereas in the absence of P_i the Mn^{++} is bound reversibly to phospholipids. The accumulation of Mg^{++} is inhibited by high concentrations of Ca^{++} (187) and the accumulation of Ca^{++} is faster and uses up more ATP than the accumulation of Mg^{++} .

The uptake is not specific for Ca^{++} or Mg^{++} as both Ba^{++} and Sr^{++} are accumulated under the same conditions. In calcium free medium, strontium is accumulated to the same extent as calcium (178). But, if calcium, strontium, and barium are present in equimolar concentrations, three times more calcium is taken up than strontium and five times more calcium than barium (174,178). However, magnesium, zinc, and cadmium are not accumulated to any significant extent under the same conditions (178).

It is of interest to note that magnesium, calcium, strontium, and barium have their two valence electrons in the "s" orbit of the outer shell and the electrons in the next innermost shell split two in the "s" orbit and six in the "p" orbital. On the other hand, zinc and cadmium, while having the two valence electrons in the "s" orbital, have the electrons in the next innermost shell split two in the "s", six in the "p" and ten in the "d" orbital

(188). Radium has the same terminal electron configuration as the magnesium, calcium, strontium, and barium group. There are similarities of resonance energy and ionization energy within the group also, with the exception of magnesium (188).

With these facts to form a rationale, radium/calcium and radium/phosphorous OR's were calculated and, in Table 13, compared with theoretical OR's. The theoretical OR's were based on a unit barrier postulation, where the unit barrier was the hindrance offered by intervening tissues, to the passage of an ion from an epithelial mucosal surface to the space enclosed by vascular endothelium or from the latter space into tissues (into the blood stream or out of it). When a unit barrier with 6.5-fold hindrance to the passage of radium is postulated, most of the Ra/P OR's fall near this value.

Specific differences at the local level can explain OR's that do not fit, i.e.: (1) skin and fat or lung have local precipitation or retention of radium in sulfate complexes; (2) heart and liver have abnormally high phosphate levels due to the constant high metabolic activity of these organs; and (3) urine is an ultrafiltrate rather than being due to active transport processes.

Although this is only a postulation at present, further investigation and experiment may prove that phosphate transport is the unifying principle for all cation accumulation. Differences observed between different tissues would then be due to specific local conditions influencing availability of ions or their binding for transport. It seems more reasonable that there is a single basic mechanism with local modifications, rather than a separate mechanism for each tissue.

When bone is considered, nearly all investigators agree that the strontium-90 bone-diet OR is 0.25 (93,94,96,97,101,125,126,189), but there is some difference of opinion as to whether the bone-plasma OR is 1, 0.5, or

TABLE 13

COMPARISON OF THEORETICAL^a AND CALCULATED OR'S BASED ON RADIUM-226 TO
CALCIUM OR RADIUM-226 TO PHOSPHOROUS RATIOS

Tissue-Precursor	Number of Unit Barriers	Theoretical ^a OR Based on Ra/Ca Ratios with 1.56 Fold Discrimi- nation per Unit Barrier	Calculated OR's Based on Ra/Ca Ratios	Theoretical ^a OR Based on Ra/Ca Ratios with 5.75 Fold Discrimi- nation per Unit Barrier	Calculated OR's Based on Ra/Ca Ratios	Theoretical OR if Each Unit Barrier Ra/P is	
						6.5-Fold	7.0 Fold
Bone-diet	2	0.422	0.015 [±] 0.003	<u>0.030</u>	0.026 [±] 0.006	<u>0.023</u>	<u>0.020</u>
Milk-diet	2	0.422	0.028 [±] 0.015	<u>0.030</u>	0.069 [±] 0.052	<u>0.023</u>	<u>0.020</u>
Muscle 1-diet	2	<u>0.422</u>	0.330 [±] 0.196	0.030	0.016 [±] 0.008	<u>0.023</u>	<u>0.020</u>
Muscle 2-diet	2	0.422	0.200 [±] 0.088	0.030	0.019 [±] 0.005	<u>0.023</u>	<u>0.020</u>
Lung-diet	2	<u>0.422</u>	0.334 [±] 0.120	0.030	0.151 [±] 0.018	0.023	0.020
Skin and fat-diet	2	0.422	0.322 [±] 0.085	0.030	0.146 [±] 0.057	0.023	0.020
Liver-diet	2	0.422	0.072 [±] 0.025	0.030	0.007 [±] 0.003	0.023	0.020
Heart-diet	2	0.422	0.037 [±] 0.014	0.030	0.014 [±] 0.006	0.023	<u>0.020</u>
Kidney-diet	2	0.422	0.109 [±] 0.028	<u>0.030</u>	0.028 [±] 0.024	<u>0.023</u>	<u>0.020</u>
Urine-diet	1	0.422	0.204 [±] 0.061	0.030	0.107 [±] 0.033	0.023	0.020
Feces-blood		0.649		0.174			
Blood 1-diet	1	<u>0.649</u>	0.704 [±] 0.416	<u>0.174</u>	0.176 [±] 0.092	<u>0.153</u>	<u>0.143</u>
Gut-diet	1	0.649	1.019 [±] 0.203	<u>0.174</u>	0.163 [±] 0.041	<u>0.153</u>	<u>0.143</u>
Urine-blood	<1	0.649	0.362 [±] 0.471	<u>0.174</u>	0.410 [±] 0.327	<u>0.153</u>	<u>0.143</u>

TABLE 13--Continued

Tissue-Precursor	Number of Unit Barriers	Theoretical ^a OR Based on Ra/Ca Ratios with 1.56 Fold Discrimination per Unit Barrier	Calculated OR's Based on Ra/Ca Ratios	Theoretical ^a OR Based on Ra/Ca Ratios with 5.75 Fold Discrimination per Unit Barrier	Calculated OR's Based on Ra/Ca Ratios	Theoretical OR if Each Unit Barrier Ra/P is	
						6.5 Fold	7.0 Fold
Muscle-blood	1	0.649	0.330 [±] 0.196	0.174	0.069 [±] 0.050	0.153	0.143
Bone-blood	1	0.649	0.025 [±] 0.008	<u>0.174</u>	0.177 [±] 0.054	<u>0.153</u>	<u>0.143</u>
Blood 2-diet	1	<u>0.649</u>	0.638 [±] 0.084	<u>0.174</u>	0.160 [±] 0.054	<u>0.153</u>	<u>0.143</u>
Muscle 2-diet	1	0.649	0.302 [±] 0.136	<u>0.174</u>	0.130 [±] 0.044	<u>0.153</u>	<u>0.143</u>
Milk-blood	1	0.649	0.044 [±] 0.022	<u>0.174</u>	0.327 [±] 0.294	<u>0.153</u>	<u>0.143</u>
Skin and fat-blood	1	<u>0.649</u>	0.508 [±] 0.172	0.174	0.750 [±] 0.382	0.153	0.143
Lung-blood	1	<u>0.649</u>	0.598 [±] 0.166	0.174	0.944 [±] 0.348	0.153	0.143
Heart-blood	1	0.649	0.062 [±] 0.022	0.174	0.089 [±] 0.040	0.153	0.143
Liver-blood	1	0.649	0.112 [±] 0.069	0.174	0.036 [±] 0.011	0.153	0.143
Kidney-blood	1	0.649	0.165 [±] 0.079	<u>0.174</u>	0.178 [±] 0.119	<u>0.153</u>	<u>0.143</u>
Neonate-diet	1	0.422	0.018 [±] 0.005	0.030	0.046 [±] 0.013	0.023	0.020
Neonate-blood	1	0.649	0.028 [±] 0.009	0.174	0.288 [±] 0.078	0.153	0.143

Numbers underlined are within the 95% confidence limits calculated for the corresponding sample OR.

^aTheoretical OR is average discrimination for the blood-diet unit barrier, the first unit barrier.

0.33 (98,101,146,189). In this study, even though the blood-diet OR is about 0.5, as it is for strontium, the bone-blood OR is about 0.025, nearly 20-fold lower than that observed for strontium. The absolute quantity of radium-226 per gram of ash in the bone is about the same as that for blood, or what blood is estimated to have been, at the time of formation of the bone. This is not a contradiction with Bryant and Loutit's observation (101) that discrimination is possible only during osteogenesis, while in primary uptake by established bone (e.g., exchange, remodeling, etc.) ions are taken up according to their ratio in the interstitial fluid. The calcium deposition in bone may be considered a two-stage process. The first stage is deposition in a nucleation site, an extremely rapid process where about 70 per cent of the calcium is fixed to the matrix as soon as the osteon is formed (191). Here there is opportunity for discrimination. Further mineralization during the second stage carries the osteon from its initial density of 1.7 to a final density of 2.2 to 2.3. The rate of deposition of calcium during this mineralization is relatively independent of diet, but the rate of resorption is related to the calcium level of the diet (141). There is much opportunity for changes in specific activity in bone, since the bone in the immature animal has a high rate of turnover (149) and the adult animal has only a compartment, estimated to be about 10 per cent of the skeleton, which has a relatively high rate of turnover (101). The diffuse turnover of calcium has been estimated at from 5 to 20 per cent in adult dogs (191) to about 50 per cent in young rats (192) per year.

With these observations as a background, it is obvious that if the Ra/Ca based OR is correct, nearly all of the discrimination against radium in bone is during the passage of the radium from blood to bone. This is in complete disagreement with the observation of Lucas (79) that the $OR_{\text{bone-blood}}$ for radium-226 in humans is approximately 1.0. However, if an OR based on

Ra/P is used as mentioned earlier, no especially postulated highly preferential barrier against radium is required. The OR is of the same order of magnitude as would be predicted for passage through two unit barriers.

The OR's calculated for excreted radium may be in error since they were calculated on the assumption that the divalent cation systems were in equilibrium. In fact, since the animals were in metabolism cages, they were probably in some stage of negative calcium balance. The exact magnitude of error introduced by this is unknown, but it is certainly in the direction of reducing the difference observed in urinary excretion.

The fecal to urinary excretion ratio was about 49 to 1, more nearly like that reported for humans of 49 to 1 (22) or rats of 10 to 1 or 20 to 1 (108), than that reported for dogs of 2.5 to 1 (193). However, comparison of results obtained by single injections with those from continuous feeding is not valid, since in the case of a single injection it is obvious that there is no equilibrium reached.

The $OR_{\text{urine-blood}}$ in the swine is interesting in that it may reflect a basic difference in the way the kidney handles radium. The variations in urinary output or individual OR's is not surprising since daily variations of $\pm 30\%$ in an individual and two-fold differences in urinary output between individuals fed a constant diet containing strontium-90 have been observed in humans (140). The $OR_{\text{urine-diet}}$ for strontium has been reported as ranging from 0.75 in humans (194) to 4.3 in lower animals (see references for Table 12).

Calcium is known to form electrostatic complexes with SO_4^{2-} and PO_4^{3-} , which are biologically inert and handled by the renal tubule in a manner different from that used for free ions (178). It has also been reported that some chelating agents have opposite effects on renal excretion of calcium and strontium (195). Further, in the rat, it has been observed that in one

strain the urinary excretion of ^{90}Sr was 1.9 times greater than that of ^{140}Ba in young animals and 1.4 times greater in adults (176).

Taking into consideration both the similarities and differences in the properties of the divalent cations, it is possible to hypothesize that neither barium nor radium can form the same types of electrostatic or chelate complexes as strontium or calcium, possibly because of their ionic diameter. Since they do not form the same types of biologically inert complexes, they reach the renal tubules as free ions and are resorbed. There are, however, many other possibilities, but it should be possible to resolve the question with perfusion, microperfusion, and stop-flow studies in combination.

The other differences in OR's of radium-226 and strontium-90 found can be due to several factors but were not within the scope of this study. These include differences in solubility and/or diffusion constants, inability of biological chelating agents to chelate radium, differences in binding capacity of proteins for divalent cations of different ionic size, specific hormonal regulation, etc. (196). But there are definite differences in the physiological handling of all divalent cations in spite of their many similarities.

This study has shown the existence of selective barriers at physiological boundaries for radium-226 when it is compared to other divalent cations, i.e., calcium and strontium. The mechanism controlling these barriers is not known. Additional techniques and methodologies will be needed to understand the comparative mechanisms of divalent cation metabolism, especially the metabolic fate of ingested radium-226, which have been raised in this report.

CONCLUSIONS

It is concluded that:

1. If radium-226 is associated with calcium in the metabolism of divalent cations, it is confronted by a selective barrier at physiological boundaries. In comparison with published data on strontium-90, it is noted that:

- a. Radium-226 passes through the porcine placenta with little or no discrimination much as do calcium, strontium, and cesium;
- b. Radium-226 passing from diet to bone is discriminated against 10- to 20-fold more than strontium-90 and 50- to 100-fold more than calcium;
- c. Radium-226 going from diet to milk is discriminated against 3 to 8 times more than strontium-90 and 33-fold more than calcium;
- d. Radium-226 crossing the intestinal wall from diet to plasma is not discriminated against more than strontium-90;
- e. Radium-226 passing from diet to feces is not discriminated against more than strontium-90;
- f. Radium-226 crossing from diet to urine is favored 5- to 20-fold more than strontium-90.

2. The distribution of radium-226 in the porcine skeleton is relatively uniform with the exception of the skull, ribs, manus, and pes. With the exceptions noted, the radium-226 burdens of the individual bones of the skeleton lie within $\pm 15\%$ of the mean burden of the skeleton.

3. A power function model for retention of radium-226 can be calculated with a value resembling that for the human, i.e., porcine $0.54t^{-0.54}$, human $0.54t^{-0.52}$.

4. There is no evident correlation between radium-226 burden and incidence of lymphosarcoma in swine in the small sample of the population that was examined.

SUMMARY

The study reported here investigated some aspects of the metabolic fate of radium-226 in swine. Samples of bone were obtained from ten leukemic and eight normal animals. Samples of feed and water, and tissues, and excretions were obtained from eight normal barrows. In addition, samples of feed, water, tissues, and secretions were obtained from eight normal sows. In addition, eleven immature swine and eight neonatal swine were assayed. These samples were assayed for radium-226 content by the radon-222 emanation method.

In fourteen skeletons assayed all bones had a gram-of-radium-226-per gram-of-ash-burden lying within $\pm 20\%$ of the mean burden for the entire skeleton.

Of ten leukemic and twenty-four normal animals, the bone burden in the normal swine ranged from 4.10 ± 0.11 to 214.50 ± 5.65 pCi of radium-226 per gram ash, while burdens for the leukemic swine ranged from 7.90 ± 0.68 to 124.60 ± 2.23 pCi per gram of ash. There was no evidence of correlation between radium-226 burden and the incidence of leukemia in these animals. However, there was a correlation between geographical area of origin and radium-226 burden for these animals.

Observed ratios, comparing pCi of radium-226 per gram of calcium in a product to the pCi of radium-226 per gram of calcium in the precursor of that product, suggested a selective preferential barrier for radium-226 at physiological boundaries.

There was no discrimination observed at the placenta. Discrimination of the same order of magnitude as that reported for strontium-90 was observed for radium-226 between feces and diet and between plasma and diet.

Discrimination 2- to 20-fold greater than that for strontium-90 was observed between urine and diet, bone and diet, and milk and diet.

If a unit barrier, consisting of all tissues which impede either entry into or exit from the blood vascular system, and association of radium with phosphorous rather than calcium during transport is assumed; then any differences in observed ratios can be explained on the basis of local influences on a basic mechanism. In this way, postulating a 6.5-fold hinderance to the passage of radium ions at each unit barrier, it is not necessary to describe specific or unique discrimination mechanisms for every tissue.

However, much further study using other methods and techniques is required to determine the mechanism of action for the selective discrimination observed in the metabolism of divalent cations.

REFERENCES

1. H. Becquerel, Sur les radiations emises par phosphorescence. *Compt. Rend.*, 122: 420-421 (1896).
2. P. Curie (Mme.), P. Curie, and M. G. Bemont, Sur une nouvelle substance fortement radioactive, contenue dans la pechblende. *Compt. Rend.* 127: 1215-1217 (1898).
3. P. Curie and (Mme.) S. Curie, Sur une substance nouvelle radio-active, contenue dans la pechblende. *Compt. Rend.* 127: 175-178 (1898).
4. A. Debierne, Sur une Nouvelle Matiere Radio-active. *Compt. Rend.* 129: 593-595 (1899).
5. E. Rutherford, A Radio-active Substance Emitted from Thorium Compounds. *Phil. Mag.* 49: 1-14 (1900).
6. F. Dorn, Versuche über Sekundarstrahlen und Radiumstrahlen. *Abh. Naturforsch. Ges. Halle* 22: 37-43 (1900).
7. F. Giesel, Ueber Radium und Radioactive Stoffe. *Chem. Ber.* 35: 3608-3611 (1902).
8. F. Soddy, The Chemistry of the Radioelements. Longmans Green and Co., London (1911).
9. Ch. Bouchard, P. Curie, and V. Baltazard, Action physiologique de l'emanation du radium. *Compt. Rend.* 138: 1384-1387 (1904).
10. A. Bickel, Beitrag zur Thorium X-Dehandlung der perniciosen Anämie. *Klin. Wochschr.* 49: 1322-1323 (1912).
11. F. Gudsent, Zur Frage der Vergiftung mit Thorium X. *Klin. Wochschr.* 49: 933-935 (1912).

12. G. Klemperer, Demonstration von Präparaten von perniziöser Anämie nach Thorium X - Behandlung. *Klin. Wochschr.* 49: 1290 (1912).
13. E. Prado Tagle-Chile, Zur Kenntnis der durch Radiothorium erzeugten Gewebeveränderungen. *Klin. Wochschr.* 49: 1557-1560 (1912).
14. T. Blum, Osteomyelitis of the Mandible and Maxilla. *J. Am. Dental Assoc.* 11: 802-805 (1924).
15. F. L. Hoffman, Radium (Mesothorium) Necrosis. *J. Am. Med. Assoc.* 85: 961-965 (1925).
16. H. S. Martland, P. Conlon, and J. P. Knaf, Some Unrecognized Dangers in the Use and Handling of Radioactive Substances. *J. Am. Med. Assoc.* 85: 1769-1776 (1925).
17. A. V. St. George, A. O. Gettler, and R. H. Muller, Radioactive Substances in a Body 5 Years After Death. *Arch. Pathol.* 7: 397-405 (1929).
18. W. B. Looney, Late Clinical Changes Following the Internal Deposition of Radioactive Materials. *Ann. Internal Med.* 42: 378-387 (1955).
19. W. B. Looney, R. J. Hasterlik, A. M. Brues, and E. Skirmont, A Clinical Investigation of the Chronic Effects of Radium Salts Administered Therapeutically (1915-1931). *Am. J. Roentgen. Radium Therapy Nucl. Med.* 73: 1006-1037 (1955).
20. M. A. Bloom, and W. Bloom, Late Effects of Radium and Plutonium on Bone. *Arch. Pathol.* 47: 494-511 (1949).
21. T. F. Dougherty, B. J. Storer, J. H. Dougherty, W. S. S. Jee, C. W. Mays, C. E. Rehfeld, W. R. Christensen, and H. C. Goldthorpe, Studies of the Biological Effects of Ra^{226} , Pu^{239} , $Ra^{228}(MsTh)$, $Th^{228}(RdTh)$, and Sr^{90} in Adult Beagles. *Radiation Res.* 17: 625-681 (1962).
22. H. E. Silberstein, Radium Poisoning A E C D-2122, Technical Information Branch AEC, Oak Ridge, Tennessee (1945).

23. R. C. Thompson, Vertebrate Radiology: Metabolism of Internal Emitters. *Ann. Rev. Nucl. Sci.* 10: 531-560 (1960).
24. K. Williams, Industrial Medicine and Toxicology Lecture Series, Department of Pharmacology, University of Chicago (1962).
25. Recommendations of the International Commission on Radiological Protection. *Brit. J. Radiol. Suppl.* 6 (1955).
26. A. Schraub, K. Aurand, and W. Jacobi, The Importance of Radon and Its Decay Products in Relation to the Normal Radiation Dose in Humans. *Brit. J. Radiol. Suppl.* 7, 114-119 (1957).
27. L. K. Bustad, and R. O. McClellan, Use of Pigs in Biomedical Research. *Nature* 208: 531-535 (1965).
28. P. Emile-Weil, and A. Lacassagne, IV. Anémie pernicieuse et leucémie myéloïde mortelles provoquées par la manipulation de substances radioactives. *Bull. Acad. Nat. Med. (Paris)* 93: 237-241 (1955).
29. J. D. Abbatt, Investigation of the Radioactivity of a Radium and Radon Worker. *Brit. J. Radiol. Suppl.* 7, 67-70 (1957).
30. C. R. Hill, Polonium-210 in Man. *Nature* 208: 423-428 (1965).
31. R. Tuscany, and V. Klenner, Decrease of Euploidia in Bone Marrow Cells in Persons with Internal Contamination by Some Radioisotopes. *Cesk. Fysiol* 12: 391 (1963) in Czech.
32. R. D. Evans, Radium and Mesothorium Toxicity in Human Beings, pp. 1-112. Radium and Mesothorium Poisoning and Dosimetry and Instrumentation Techniques in Applied Radioactivity. MIT-952-1. MIT Cambridge Radioactivity Center, 1964.
33. R. C. Turner, J. M. Radley, and W. V. Mayneard, The Naturally Occurring α -Ray Activity of Foods, *Health Phys.* 1: 268-275 (1958).

34. H. Muth, B. Rajewsky, H. J. Hantke, and K. Aurand, The Normal Radium Content and the Ra^{226}/Ca Ratio of Various Foods, Drinking Water and Different Organs and Tissues of the Human Body, *Health Phys.* 2: 239-245 (1960).
35. H. Muth, A. Schraub, K. Aurand, and H. J. Hantke, Measurements of Normal Radium Burdens, *Brit. J. Radiol. Suppl.* 7, 54-66 (1957).
36. J. B. Hursh, Natural Occurrence of Radium in Man and in Waters and in Food. *Brit. J. Radiol. Suppl.* 7, 45-53 (1957).
37. R. C. Turner, J. M. Radley, and W. V. Mayneord, Alpha-Ray Activities of Humans and Their Environment. *Nature* 181: 518-521 (1958).
38. H. F. Lucas, Jr., Correlation of the Natural Radioactivity of the Human Body to that of Its Environment: Uptake and Retention of Ra^{226} from Food and Water. Argonne National Laboratory, Radiological Physics Division Semi-annual Report, ANL-6297, pp. 55-65 (1960).
39. R. C. Turner, J. M. Radley, and W. V. Mayneord, The Alpha-Ray Activity of Human Tissues. *Brit. J. Radiol.* 31: 397-406 (1958).
40. A. F. Stehney and H. F. Lucas, Jr., Retention of Environmental Radium by Young Human Males. Argonne National Laboratory, Radiological Physics Division Semi-annual Report, ANL-5456, pp. 95-98 (1955).
41. A. Krebs, Über die normale und Anormale Radioaktivität Menschlichen und Tierischen Gewebs. *Fundam. Radiol.* 5: 89-112 (1939).
42. E. P. Radford, Jr., V. R. Hunt, and D. Sherry, Analysis of Teeth and Bones for Alpha-Emitting Elements. *Radiation Res.* 19: 298-315 (1963).
43. J. B. Hursh, and A. Lovas, Radium-226 in Bone and Soft Tissue of Man. *Nature* 198: 265-268 (1963).
44. A. Walton, R. Kologrivov, and J. L. Kulp, The Concentration and Distribution of Radium in the Normal Human Skeleton. *Health Phys.* 1: 409-416 (1959).

45. J. Rotblat, and G. Ward, Autoradiograph Measurements of the Bones of a Radium Worker. Brit. J. Radiol. Suppl. 7, 90-91 (1957).
46. R. C. Turner, and W. Anderson, Measurements of Radioactivity in a Radium and Radon Worker. Brit. J. Radiol. Suppl. 7, 71-80 (1957).
47. M. Hindmarsh, and J. Vaughan, The Distribution of Radium in Certain Bones from a Man Exposed to Radium for Thirty-four Years. Brit. J. Radiol. Suppl. 7, 71-80 (1957).
48. F. W. Spiers, and P. R. J. Burch, Measurements of Body Radioactivity in a Radium Worker. Brit. J. Radiol. Suppl. 7, 81-89 (1957).
49. T. E. Larson, and R. L. Weatherford, Natural Radioactivity in Illinois Water Resources. Illinois State Water Survey Circular No. 81, Urbana, Illinois (1960).
50. F. E. Hoscker, and P. G. Roope, Studies of Radium in Human Bone. Radiology, 56: 89-98 (1951).
51. R. E. Rowland, and J. H. Marshall, Calibration of the Quantitative Alpha-Track Autoradiographic Technique. Argonne National Laboratory, Radiological Physics Division Semi-annual Report, ANL-6049, pp. 23-29 (1959).
52. E. Lloyd, The Distribution of Radium in Human Bone. Brit. J. Radiol. 34: 521-528 (1961).
53. G. Ward, The Use of Nuclear Tract Autoradiography in the Study of the Distribution of Thorium and Its Products in Living Organisms (Ph.D. thesis, Physics as Applied to Medicine, University of London, June, 1955).
54. J. O. Snihs, Nuclear Emulsion Technique in Measurement of Radon. Arkiv för Fysik 25: 65-85 (1963).
55. (Mme.) S. Curie, Radioactive Substances. Chem. Prod. 88: 85-86, 97-99, 134-135, 145-146, 159-160, 169-171, 175-177, 187-188, 199-201, 211-212, 223-224, 235-236, 247-249, 258-261, 271-272 (1903).

56. A. Becker, and K. H. Stehberger, Über die Adsorption der Radiumemanation, Ann. Physik 1: 529-555 (1929).
57. O. Gföbeli und M. Störi, Zur Mischadsorption von Radon an Aktivkohle mit verschiedenen Trägergasen, Helv. Chim. Acta 37: 2224-2230 (1954).
58. A. F. Stehney, W. P. Norris, H. F. Lucas, Jr., and W. H. Johnston, A Method for Measuring the Rate of Elimination of Radon in Breath. Am. J. Roentgen. Radium Therapy Nucl. Med. 73: 774-784 (1955).
59. R. D. Evans, Apparatus for the Determination of Minute Quantities of Radium, Radon and Thoron in Solids, Liquids and Gases. Rev. Sci. Instr. 6: 99-112 (1935).
60. E. Rona, and W. D. Urry, Radioactivity of Ocean Sediments. VIII. Radium and Uranium Content of Ocean and River Waters. Am. J. Sci. 250: 243-262 (1952).
61. L. F. Curtiss, and F. J. Davis, A Counting Method for the Determination of Small Amounts of Radium and of Radon. J. Res. Nat. Bur. Std., C. 31: 181-195 (1943).
62. J. E. Hudgens, R. O. Benzing, J. P. Cali, R. C. Meyer, and L. C. Nelson, Determination of Radium or Radon in Gases, Liquids or Solids. Nucleonics 9: 14-21 (1951).
63. B. J. Storer, D. R. Atherton, and J. S. Arnold, Comparative Metabolism of Ca^{45} and Ra^{226} . Proc. Soc. Exp. Biol. Med. 94: 269-272 (1957).
64. J. D. Greig, The Determination of Low Radon and Radium Concentration in Liquids and Slurries. Intern. J. Appl. Radiation Isotopes 11: 101-107 (1961).
65. A. Kaul und H. Muth, Messungen sehr kleiner Radonkonzentrationen mit Szintillationsanordnungen. In 9th International Congress of Radiology, pp. 1391-1396, George Thieme Verlag, Stuttgart (1960).

66. J. N. Rosholt, Jr., Quantitative Radiochemical Methods for Determination of the Sources of Natural Radioactivity. *Anal. Chem.* 29: 1398-1408 (1957).
67. J. N. Rosholt, Jr., and J. R. Dooley, Jr., Automatic Measurements and Computations for Radiochemical Analysis. *Anal. Chem.* 32: 1093-1098 (1960).
68. H. Muth, and E. Oberhausen, Die gesamte natürliche Alphastrahlenaktivität des Menschen. In 9th International Congress of Radiology, pp. 1140-1145, Georg Thieme Verlag, Stuttgart (1960).
69. J. Laurence Kulp, Heinrich D. Holland, and Herbert L. Volchok, Scintillation Alpha Counting of Rocks and Minerals. *Trans. Amer. Geophys. Un.* 33: 101-113 (1952).
70. M. A. Van Dilla, and D. H. Taysum, Scintillation Counter for Assay of Radon Gas. *Nucleonics* 13: 68-69 (1955).
71. M. A. Van Dilla, B. J. Stover, R. L. Floyd, D. R. Atherton, and D. H. Taysum, Radium (Ra^{226}) and Radon (Rn^{222}) Metabolism in Dogs. *Radiation Res.* 8: 417-437 (1958).
72. N. Sax, and D. Rosi, 4-P: Zinc Sulfide Alpha Counting. *Anal. Chem.* 32: 1900 (1960).
73. H. F. Lucas, Jr., Improved Low-Level Alpha Scintillation Counter for Radon. *Rev. Sci. Instr.* 28: 680-683 (1957).
74. L. B. Robinson, and J. R. Arnold, The Scintillation Counter. I. The Existence of Plateaus. *Rev. Sci. Instr.* 20: 549-553 (1949).
75. L. B. Robinson, C. S. Cook, and D. E. Jefferson, The Scintillation Counter. II. The Preparation of Transparent Inorganic Phosphor Screens. *Rev. Sci. Instr.* 21: 148-149 (1950).
76. P. E. Damon, and H. I. Hyde, Scintillation Tube for the Measurement of Radioactive Gases. *Rev. Sci. Instr.* 23: 766 (1952).

77. W. Siebert, Absorptionsuntersuchungen mit Ra-Emanation. Z. Physik. Chem. (Frankfurt) 180: 169-184 (1937).
78. W. A. Jennings, and S. Russ, Radon: Its Technique and Use. Published for the Middlesex Hospital Press by John Murray, London (1948).
79. H. F. Lucas, Jr., Personal communication.
80. D. A. Hartman, and W. G. Pond, Design and Use of a Milking Machine for Sows. J. Animal Sci. 19: 780-785 (1960).
81. G. H. Kingsley, and O. Robnett, Further Studies on a New Dye Method for the Direct Photometric Determination of Calcium. Am. J. Clin. Path. 29: 171-175 (1958).
82. C. F. Coleman, G. A. Blake, and K. B. Brown, Analytical Potential of Separations by Liquid Ion Exchange. Talanta 9: 297-323 (1962).
83. J. F. Steinbach, and H. Freiser, Acetylacetone in the Dual Role of Solvent and Reagent in Extraction of Metal Chelates. Anal. Chem. 25: 881-884 (1953).
84. J. F. Steinbach, and H. Freiser, Acetylacetone as an Analytical Extraction Agent--Extraction of Aluminum, Gallium and Indium. Anal. Chem. 26: 375-379 (1954).
85. J. S. Annino, Clinical Chemistry, Principles and Procedures. 2nd ed. Little, Brown and Co., Boston, Toronto (1961).
86. P. B. Hawk, B. L. Oser, and W. H. Summerson, Practical Physiological Chemistry. McGraw-Hill, New York (1954).
87. R. L. Morris, and J. W. Klinsky, Radiochemistry and Removal Characteristics of Radium Isotopes in Iowa Well Waters. Proc. Iowa Acad. Sci. 69: 396-399 (1962).
88. A. B. Tanner, Physical and Chemical Controls on Distribution of Radium-226 and Radon-222 in Ground Water Near Great Salt Lake, Utah, pp. 253-276.

- The Natural Radiation Environment, ed. J. A. S. Adams and W. M. Lowder, University of Chicago Press, Chicago (1964).
89. W. F. Neuman, and H. W. Neuman, The Chemical Dynamics of Bone Mineral. University of Chicago Press, Chicago (1958).
 90. F. C. McLean, and A. M. Eudy, Radiation, Isotopes and Bone. Academic Press, London (1964).
 91. A. R. Schulert, E. A. Peets, D. Laszlo, H. Spencer, M. Charles, and J. Samachson, Comparative Metabolism of Strontium and Calcium in Man. Intern. J. Appl. Radiation Isotopes 4: 144-153 (1959).
 92. A. Morgan, and J. E. Wilkins, Distribution of Fallout Radiostrontium in a Sheep Skeleton. Biochem. J. 71: 419-422 (1959).
 93. S. Haggroth, and O. Högglund, Determination of ^{90}Sr and Stable Strontium in Bones from Sheep Ewes and Their Fetuses. Exptl. Cell Research 24: 80-87 (1961).
 94. J. L. Kulp, A. R. Schulert, and E. J. Hodges, Strontium-90 in Man, III. Science 129: 1249-1255 (1959).
 95. Z. V. Dubrovina, S. Ya. Zaidman, I. Ya. Panchenko, I. A. Sarapul'tsev, and A. M. Skryabin, The Evaluation of Skeleton Activity as a Function of Strontium-90 Content in Individual Bones. Radiobiologiya 111: 773-777 (1963) in AEC-tr-5438, pp. 233-241 (1963).
 96. J. L. Kulp, Strontium-90 in Man. Strahlenschutz 18: 3-11 (1961).
 97. J. L. Kulp, A. R. Schulert, and E. J. Hodges, Strontium-90 in Man, IV. Science 132: 448-454 (1960).
 98. S. G. Kahriragar, E. Lloyd, and J. Vaughan, Discrimination Between Strontium and Calcium in Bone and the Transfer from Blood to Bone in the Rabbit. Brit. J. Radiol. 39: 131-140 (1966).
 99. R. B. Dean, and W. J. Dixon, Simplified Statistics for Small Numbers of Observations. Anal. Chem. 23: 636-638 (1951).

100. G. C. H. Bauer, Radioisotopes in the Skeleton: Observations on Distribution in Man, pp. 354-365. In Radioisotopes in the Biosphere (ed. R. S. Caldecott and L. A. Snyder), University of Minnesota Center for Continuation Study, Minneapolis (1960).
101. F. J. Bryant, and J. F. Loutit, The Entry of Strontium-90 into Human Bone. Proc. Royal Soc., B, 159: 449-465 (1964).
102. J. L. Kulp, and A. R. Schulert, Strontium-90 in Man. V. Science 136: 619-632 (1962).
103. M. A. Bloom, Bone Marrow, pp. 162-242. In Histopathology of Irradiation from External and Internal Sources (ed. William Bloom), McGraw-Hill Book Co., Inc., New York (1948).
104. R. G. Murray, The Spleen, pp. 243-347. In Histopathology of Irradiation from External and Internal Sources (ed. William Bloom), McGraw-Hill Book Co., Inc., New York (1948).
105. P. P. H. De Bruyn, Lymph Node and Intestinal Lymphatic Tissue, pp. 348-445. In Histopathology of Irradiation from External and Internal Sources (ed. William Bloom), McGraw-Hill Book Co., Inc., New York (1948).
106. R. G. Murray, The Thymus, pp. 446-501. In Histopathology of Irradiation from External and Internal Sources (ed. William Bloom), McGraw-Hill Book Co., Inc., New York (1948).
107. D. O. Shiels, Effect on Lymphoid Cells of Occupational Exposure to Low Doses of Ionizing Radiation. Australasian Ann. Med. 11: 267-269 (1962).
108. W. P. Norris, and W. Kisielleski, Comparative Metabolism of Radium, Strontium, and Calcium. Cold Spring Harbor Symp. Quant. Biol. 13: 164-172 (1948).
109. J. H. Marshall, Theory of Alkaline Earth Metabolism. J. Theoret. Biol. 6: 386-412 (1964).

110. S. M. Saunders, Jr., Power Functions Relating Excretion to Body Burden. *Health Phys.* 2: 295-303 (1960).
111. J. Anderson, S. B. Osborn, R. W. W. Tomlinson, and I. Weinbren. Some Applications of Power Law Analysis to Radioisotope Studies in Man. *Phys. Med. Biol.* 8: 287-295 (1963).
112. R. E. Rowland, Retention and Plasma Clearance of the Alkaline Earth Elements. Argonne National Laboratory, Radiological Physics Division Semi-annual Report, ANL-6104, pp. 34-37 (1959).
113. G. W. Dolphin, and I. S. Eve, Some Aspects of Radiostrontium Dosimetry. *Phys. Med. Biol.* 8: 205-214 (1963).
114. W. P. Norris, S. A. Tyler, and A. M. Brues, Retention of Radioactive Bone-Seekers. *Science* 128: 456-462 (1958).
115. C. E. Miller, and A. J. Finkel, Radium Retention in Mice After a Single Intravenous Injection. *Radiation Res.* 26: 269-286 (1965).
116. N. A. Halliden, I. M. Fiscenne, and J. H. Harley, Radium-226 in Human Diet and Bone. *Science* 140: 1327-1329 (1963).
117. W. P. Norris, T. W. Speckman, and P. F. Gustafson, Studies of the Metabolism of Radium in Man. *Am. J. Roentgen. Radium Therapy Nucl. Med.* 73: 785-802 (1955).
118. C. L. Comar, R. H. Wasserman, and M. M. Nold, Strontium-Calcium Discrimination Factors in the Rat. *Proc. Soc. Exp. Biol. Med.* 92: 859-863 (1956).
119. C. L. Comar, I. B. Whitney, and F. W. Lengemann, Comparative Utilization of Dietary Sr^{90} and Calcium by Developing Rat Fetus and Growing Rat. *Proc. Soc. Exp. Biol. Med.* 88: 232-236 (1955).
120. C. L. Comar, R. S. Russell, and R. H. Wasserman, Strontium-Calcium Movement from Soil to Man. *Science* 126: 485-492 (1957).

121. H. A. Kornberg, Radioisotopes and Environmental Circumstances: The Passage of Pairs of Elements Through Food Chains, pp. 225-228. In Radioisotopes in the Biosphere (ed. R. S. Caldecott and L. A. Snyder), Center for Continuation Study, University of Minnesota, Minneapolis (1960).
122. D. E. Hogue, W. G. Pond, C. L. Comar, L. T. Alexander, and E. P. Hardy, Comparative Utilization of Dietary Calcium and Strontium-90 by Pigs and Sheep. *J. An. Sci.* 20: 514-517 (1961).
123. B. Kahn, I. R. Jones, C. R. Porter, and C. P. Straub, Transfer of Radiostrontium from Cow's Feed to Milk. *J. Dairy Sci.* 48: 1023-1030 (1965).
124. R. F. Palmer, R. C. Thompson, and H. A. Kornberg, Factors Affecting the Relative Deposition of Strontium and Calcium in the Rat. *Science* 128: 1505-1506 (1958).
125. R. O. McClellan, Calcium-Strontium Discrimination in Miniature Pigs as Related to Age. *Nature* 202: 104-106 (1964).
126. R. F. Palmer, and R. C. Thompson, Strontium-Calcium Interrelationships in the Growing Rat. *Am. J. Physiol.* 207: 561-565 (1964).
127. R. H. Wasserman, Radioisotope Absorption and Methods of Elimination: A Critical Review, pp. 564-573. In Radioisotopes in the Biosphere (ed. R. S. Caldecott and L. A. Snyder), Center for Continuation Study, University of Minnesota, Minneapolis (1960).
128. A. R. Twardock, and C. L. Comar, Calcium and Strontium Secretion from Blood to Milk. *Am. J. Physiol.* 201: 645-650 (1961).
129. A. R. Twardock, Studies on the Movement of Calcium and Strontium Across the Bovine Mammary Gland, pp. 327-339. In The Transfer of Calcium and Strontium Across Biological Membranes (ed. R. H. Wasserman). New York: Academic Press, 1963.

130. N. S. MacDonald, D. L. Hutchinson, and M. Helper, The Bidirectional Transport of Radiostrontium Across the Primate Placenta. *Radiation Res.* 17: 752-766 (1962).
131. W. Moore, Jr., C. L. Comar, and R. H. Wasserman, Cesium, Strontium, and Calcium Metabolism: Some Recent Developments, pp. 117-136. In The Use of Radioisotopes in Animal Biology and the Medical Sciences, New York, Academic Press, 1962.
132. C. L. Comar, Radiocalcium Studies in Pregnancy. *Ann. N. Y. Acad. Sci.* 64: 281-298 (1956).
133. J. W. Bawden, and J. W. Osborne, Tracer Study on the Effect of Dietary Calcium Deficiency During Pregnancy in Rats. *J. Dental Res.* 41: 1349-1358 (1962).
134. C. L. Comar, Factors Influencing the Biological Availability of Fallout Radionuclides for Animals and Man. *Federation Proc.* 22: 1402-1409 (1963).
135. C. L. Comar, and R. H. Wasserman, Radioisotope Absorption and Methods of Elimination: Differential Behavior of Substances in Metabolic Pathways, pp. 526-540. In Radioisotopes in the Biosphere (ed. R. S. Caldecott and L. A. Snyder), Center for Continuation Study, University of Minnesota, Minneapolis, 1960.
136. C. L. Comar, and R. H. Wasserman, Strontium-Calcium Metabolism in Man and Animals as Studied by Radioisotope Methods. UNESCO International Conference on Radioisotopes in Scientific Research, UNESCO/NS/RIC/176. Pergamon Press Ltd., London, 1957.
137. C. L. Comar, Some Over-all Aspects of Strontium-Calcium Discrimination, pp. 405-417. In The Transfer of Calcium and Strontium Across Biological Membranes (ed. R. H. Wasserman), New York, Academic Press, 1963.

138. R. C. Thompson, Factors and Conditions Modifying the Absorption and Retention of Chronically Ingested Radiostrontium, pp. 393-423. In The Transfer of Calcium and Strontium Across Biological Membranes (ed. R. H. Wasserman), New York, Academic Press, 1963.
139. L. A. Maynard, Animal Nutrition. New York, McGraw-Hill Book Co., Inc. 1951.
140. T. V. Vorozheika and Yu. D. Parfenov, The Passage of Strontium-90 Through the Placenta. Bull. Exptl. Biol. Med. USSR (English translation), 54: 907-910 (1963).
141. V. G. Kulikova, The Transition of Radioisotopes from Mother to Rat Progeny. Med. Radiol. 9: 61-64 (1963) in Russian.
142. T. E. F. Carr, G. E. Harrison, J. F. Loutit, and A. Sutton, Relative Availability of Strontium in Flour and Milk. Proc. Nutr. Soc. 24: 120-126 (1965).
143. F. Bronner, and J.-P. Aubert, Bone Metabolism and Regulation of the Blood Calcium Level in Rats, Am. J. Physiol. 209: 887-890 (1965).
144. B. J. Stover, D. R. Atherton, F. W. Bruenger, and C. W. Mays, Distribution of ^{226}Ra Between Blood Cells and Plasma. Health Phys. 11: 617-622 (1965).
145. R. M. Fink, Biological Studies with Plutonium, Radium and Polonium. New York, McGraw-Hill Book Co., Inc., 1950.
146. J. L. Kulp, W. P. Eckelman, and A. R. Schulert, Strontium-90 in Man. Science 125: 219-225 (1957).
147. C. S. Marcus, and W. H. Wasserman, Comparison of Intestinal Discrimination Between Calcium-47, Strontium-85, and Barium-133. Am. J. Physiol. 209: 973-977 (1965).
148. R. H. Mole, Stable Strontium in Human Bones: Geographical and Age Differences in the United Kingdom and Their Correlation with Levels of Strontium-90. Brit. J. Nutr. 19: 13-33 (1965).

149. J. F. Loutit, Calcium, Strontium and Osteogenesis. *Proc. Nutr. Soc.* 22: 140-145 (1963).
150. B. Rajewsky, V. Belloch, E. Lohr, and W. Stahlhofen, in preparation.
151. W. Stahlhofen, Measurement of the Natural Content of Th^{228} , Ra^{226} and Their Daughters in the Human Body, pp. 505-519. In Assessment of Radioactivity in Man, International Atomic Energy Agency, Vienna, 1964.
152. E. Marsden, Radioactivity of Soils, Plant Ashes, and Animal Bones. *Nature* 183: 924-925 (1959).
153. E. R. D. Ferrange, The Natural Concentration of Ra^{226} in Bovine Bones and Teeth. *Health Phys.* 10: 259-264 (1964).
154. Heins Kriegel, Das biologische Verhalten radioaktiver Spaltprodukte trächtigen Säugetieren, *Verh. d. Dtsch. Zool. Gesellschaft*, Suppl. 24, 238-244 (1960).
155. R. M. Wynn, and J. Davies, Comparative Electron Microscopy of the Hemochorial Placenta. *Am. J. Obst. and Gynecol.* 91: 533-549 (1965).
156. H. S. Bennett, The Concepts of Membrane Flow and Membrane Vasiculation as Mechanisms for Active Transport and Ion Pumping. *J. Biophys. Biochem. Cytol.* 2: 99-104 (1956 Supplement).
157. D. C. Pease, Infolded Basal Plasma Membranes Found in Epithelia Noted for Their Water Transport. *J. Biophys. Biochem. Cytol.* 2: 203-208 (1956 Supplement).
158. A. C. Enders, Electron Microscopic Observations on the Villous Hemochorial Placenta of the Nine-banded Armadillo (*Dasypus novem cinctus*). *J. Anat.* 94: 205-215 (1960).
159. J. A. G. Rhodin, and J. A. Terzakis, The Ultrastructure of the Human Full-term Placenta. *J. Ultrastruct. Research* 6: 88-106 (1962).

160. E. W. Dempsey, G. B. Wislock, and E. C. Amoroso, Electron Microscopy of the Pig's Placenta, with Special Reference to the Cell Membranes of the Endometrium and Chorion. *Am. J. Anat.* 96: 65-101 (1955).
161. R. M. Wynn, and J. Davies, Ultrastructure of Hydatidiform Mole; Correlative Electron Microscopic and Functional Aspects. *Am. J. Obst. and Gynecol.* 90: 293-307 (1964).
162. R. O. McClellan, J. R. McKenney, and L. K. Bustad, Changes in Calcium- Sr^{90} Discrimination with Age in Young Miniature Swine. *Life Sci.* 1: 669-675 (1962).
163. D. M. Taylor, P. H. Bligh, and M. H. Duggan, The Absorption of Calcium, Strontium, Barium, and Radium from the Gastrointestinal Tract of the Rat. *Biochem. J.* 83: 25-29 (1962).
164. S. A. Lough, J. Rivera, and C. L. Comar, Retention of Strontium, Calcium, and Phosphorus in Human Infants. *Proc. Soc. Exptl. Biol. Med.* 112: 631-636 (1963).
165. F. C. Gran, Calcium and Strontium-90 Metabolism in Rats. *Acta Physiol. Scand.* 48, Suppl. 167 (1960).
166. R. J. Della Rosa, M. Goldman, and A. C. Andersen, Discrimination Against Strontium in Growing Rats Continually Ingesting Sr^{90} . *Radiation Res.* 16: 582 (1962).
167. R. H. Wasserman, C. L. Comar, and M. M. Nold, The Influence of Amino Acids and Other Organic Compounds on the Gastrointestinal Absorption of Calcium-45 and Strontium-89 in the Rat. *J. Nutr.* 59: 371-383 (1956).
168. R. H. Wasserman, and C. L. Comar, Carbohydrates and the Gastrointestinal Absorption of Radiostrontium and Radiocalcium in the Rat. *Proc. Soc. Exptl. Biol. Med.* 101: 314-317 (1959).

169. R. H. Wasserman, Lactose-stimulated Intestinal Absorption of Calcium: A Theory. *Nature* 201: 997-998 (1964).
170. R. O. McClellan, and L. K. Bustad, Transfer of Ingested Sr^{90} and Calcium to Milk of Miniature Swine, *Radiation Res.* 16: 581-582 (1962) Abstract.
171. C. L. Comar, R. H. Wasserman, and R. A. Twardock, Secretion of Calcium and Strontium into Milk. *Health Phys.* 7: 69-80 (1961).
172. C. P. Straub, and G. K. Murthy, A Comparison of Sr^{90} Component of Human and Cow's Milk. *Pediatrics* 36: 732-735 (1965).
173. R. O. McClellan, H. W. Casey, and L. K. Bustad, Transfer of Some Radio-nuclides to Milk, pp. 98-108. Hanford Atomic Products Operation Report, HW-76000 (1963).
174. J. Schubert, and E. E. Conn, Radiocolloid Behavior of Some Fission Products, *Nucleonics* 4: 2-11 (1949).
175. D. R. Atherton, B. J. Stover, and C. W. Mays, Soft Tissue Retention of ^{226}Ra in the Beagle. *Health Phys.* 11: 101-108 (1965).
176. P. H. Eligh, and D. M. Taylor, Comparative Studies of the Metabolism of Strontium and Barium in the Rat. *Biochem. J.* 87: 612-618 (1963).
177. D. Schachter, E. B. Dowdle, and H. Schenker, Accumulation of Ca^{45} by Slices of the Small Intestine. *Am. J. Physiol.* 198: 275-279 (1960).
178. H. Rasmussen, and H. F. DeLuca, Calcium Homeostasis. *Ergebn. Physiol.* 53: 108-173 (1963).
179. A. L. Lehninger. The Mitochondrion. W. A. Benjamin, Inc., New York, 1965.
180. G. P. Brierley, E. Bachmann, and D. E. Green, Active Transport of In-organic Phosphate and Mg^{44} by Beef Heart Mitochondria. *Proc. Natl. Acad. Sci. U. S.* 48: 1928-1935 (1962).

181. F. D. Vasington, Ca^{40} Uptake by Mitochondrial Subfragments and Its Dependence on Electron Transport. *J. Biol. Chem.* 238: 1841-1847 (1963).
182. C. S. Rossi, and A. L. Lehninger, Stoichiometric Relationships Between Mitochondrial Ion Accumulation and Oxidative Phosphorylation. *Biochem. Biophys. Res. Commun.* 11: 441-446 (1963).
183. C. S. Rossi, and A. L. Lehninger, Stoichiometric Relationships Between Accumulation of Ions by Mitochondria and the Energy-Coupling Sites in the Respiratory Chain. *Biochem. Zeit.* 338: 698-713 (1963).
184. H. F. De Luca, G. W. Engstrom, and H. Rasmussen, The Action of Vitamin D and Parathyroid Hormone in Vitro on Calcium Uptake and Release by Kidney Mitochondria. *Proc. Natl. Acad. Sci. U. S.* 48: 1604-1609 (1962).
185. J. B. Chappell, G. D. Greville, and K. E. Bicknell, Stimulation of Respiration of Isolated Mitochondria by Manganese Ions. *Biochem. J.* 84: 61 (1962).
186. G. P. Brierley, Ion Accumulation in Heart Mitochondria, pp. 237-251. Energy-Linked Functions of Mitochondria, 1st Johnson Research Foundation Colloquium (ed. B. Chance), Academic Press, New York, 1963.
187. J. B. Chappell, M. Cohn, and G. D. Greville, The Accumulation of Bivalent Ions by Isolated Mitochondria, pp. 219-235, 1st Johnson Research Foundation Colloquium (ed. B. Chance), Academic Press, New York, 1963.
188. Documenta Geigy 6th Edition Scientific Tables, p. 267. J. R. Geigy, S.A., Basle, 1962.
189. W. A. Muller, Untersuchungen zum Strontiumstoffwechsel im Säugetierskelett, *Atompraxis* 10: 1-4 (1963).
190. A. Asconsi, E. Bonucci, and D. S. Bocciarelli, An Electron Microscope Study of Osteon Calcification. *J. Ultrastruct. Research* 12: 287-303 (1965).

191. J. H. Marshall, R. E. Rowland, and J. Jowsey, Microscopic Metabolism of Calcium in Bone V. The Paradox of Diffuse Activity and Long-term Exchange. *Radiation Res.* 10: 258-270 (1959).
192. D. H. Tomlin, K. M. Henry, and S. K. Kon, Interstitial Metabolism of Calcium in the Bones and Teeth of Rats. *Brit. J. Nutr.* 9: 144-156 (1955).
193. B. J. Stover, D. R. Atherton, and C. W. Mays, Studies of the Retention and Distribution of Ra^{226} , Pu^{239} , Ra^{228} , $(MsTh)$, Th^{228} , $(RdTh)$, and Sr^{90} , pp. 7-25. Some Aspects of Internal Radiation (ed. T. F. Dougherty), Pergamon Press, New York, 1962.
194. W. Czosnowska, The Relationship Between the Sr^{90}/Ca Urinary/Food Ratio and Urinary Calcium in Humans. *Nukleonika* 9: 757-758 (1964) in Polish.
195. A. Schmid, and K. Zipf, ⁸Über die Wirkung Verschiedener Chelatbildner auf die Renale Ausscheidung von Strontium und Calcium. *Zentralblatt für Veterinar Medizin, B*, 12: 558-562 (1965).
196. The Transfer of Calcium and Strontium Across Biological Membranes (ed. R. H. Wasserman), Academic Press, New York, 1963.

APPENDIX A

RADON COUNTING BOTTLE

Construction

The construction and calibration of the radon counting bottle was the most difficult problem in the construction of the radon counting system.

The basic unit of the counting bottle was a "Kovar" cup seal (see list of materials, Appendix H). The "Kovar" cups were turned and "faced-off" on a lathe to a length of 6.46 cm. (see Figure 1). A 1.8 mm. hole was then drilled in the center of the cup and a length of brass tubing 6.25 mm. in diameter by 1.3 cm. long was centered and soft-soldered to the cup around the 1.8 mm. hole.

A glass micro stop-cock with a 12/5 male ball joint at one end was cut to a length of 5.2 cm. and inserted into the brass tube. It was then sealed in place with R 313 adhesive (see list of materials, Appendix H). When the R 313 adhesive had cured, the cup weight was determined and the cup placed in a jig and rotated at 30 revolutions per minute. Silver activated zinc sulfide scintillation mixture (see list of materials, Appendix H) was then sprayed into the rotating cup until a coating of zinc sulfide 21 milligrams or more per square centimeter was built up. Since the internal area of the cup was about 100 cm., this required at least 2.1 grams of zinc sulfide. The spraying was done in the following manner: a spray gun was mounted about 6 inches from the open end of the cup, 2 inches above and 2 inches to one side. The gun was aimed at the intersection of the curved top and cylindrical wall of the cup, using an "Air Brush" (see list of materials, Appendix H) as the spray gun.

Fig. 1.—Radon collection system showing a generator flask of the 500 cc. or 1,000 cc. size in the system.

To make the scintillation mixture, 10 grams of zinc sulfide, 4 to 5 ounces of ethylene dichloride and 10 to 15 drops of DEK-adhese (see list of materials, Appendix H) were mixed thoroughly by shaking in a flask. The mixture was then allowed to settle for 10 to 15 seconds. The supernatant was poured into a 150 cc. beaker and allowed to settle for 4 minutes. The resulting clear supernatant was decanted. The mixture of zinc sulfide, ethylene dichloride, and DEK-adhese was again shaken thoroughly and 1/4 cc. of the mixture was added to the spray gun cup. The zinc sulfide mixture was sprayed into the "Kovar" cup using a full air flow through the spray gun. The air flow was continued after all of the mixture was coated upon the "Kovar" cup. Blowing air onto the material on the cup walls dried the coating. Slowly an increasing amount of the mixture was then alternately sprayed and dried until 2 cc. volumes of the mixture were being sprayed. Ten to 12 sprayings coated the walls of the cup with at least 2.1 grams of zinc sulfide. This was determined by weighing the cups after each spraying and then comparing the new weight with the original weight of the cup.

In spraying the cups, care was taken not to spray too large a volume of the mixture at any one time. Excessive amounts of mixture would run and cause uneven coats of the zinc sulfide. When this happened, the coating had to be cleaned out, using ethylene dichloride as the solvent. The cup was then dried, weighed, and recoated. The "Air Brush" was cleaned with ethylene dichloride.

Care had to be observed in the amount of DEK-adhese used as binder. Too much DEK-adhese would cause a reduction in pulse height of the alpha particle interaction and a lessened efficiency of the counting bottle (1).

The open end of the cup was closed with a specially prepared quartz disk (see list of materials, Appendix H), 5.08 cm. diameter by 4 mm. thick and optically flat. The disks were cleaned in hot chromic acid and were washed

repeatedly with hot distilled water and acetone. The disks were not touched with fingers at any time. The clean disks were placed on asbestos sheet in a muffle furnace and heated to 520° to 600° C. for 10 to 20 minutes. A pyrex chimney about 3 inches high was placed around each disk to reduce convection currents. The next step was to coat one surface of the quartz disk with stannic oxide, an electrical conductor.

A generator flask was prepared by putting 10 cc. of fuming stannic chloride in a round bottom flask. Moist air (prepared by passing it through water at the rate of 1 to 3 bubbles per second) was passed into the generator through a glass tube which reached to 1-1/2 inch above the surface of the stannic chloride. A second tube starting 1 inch above the surface of the stannic chloride carried the stannic hydroxide fumes formed by the generator. The fumes were carried by tygon and glass tubing into the pyrex chimney protecting the quartz disk, for 20 to 30 seconds. One minute later the disk was removed from the furnace. This operation produced a clear, electrically conductive, relatively uniform coat of stannic oxide on the surface of the quartz disk. When the disks cooled, they were checked for electrical conductivity. If the resistance from edge to edge, checked at several points, was 100,000 ohms or less, the disks were acceptable.

Non-uniform coatings resulted whenever the quartz was dirty or there were convection currents present about the quartz disks. A poor optical coating and high resistance resulted if the temperature in the furnace was too low or if the disks were not heated sufficiently before coating. Tests were made on pyrex fragments to determine whether the temperature was satisfactory prior to coating the disks.

The transmission of the disks both before and after coating was checked with $4500 \text{ \AA}$ light using a Beckman DU Spectrometer (see Table 14).

TABLE 14
TRANSMISSION OF LIGHT (4500 Å) BY QUARTZ WINDOWS, EFFICIENCY
AND BACKGROUND COUNT OF RADON COUNTING BOTTLES

Counting Bottle Number	% Transmission of Quartz Windows	Efficiency of Bottle (Counts per Minute per pCi of Radium-226)	Background Count (Counts per Hour)
1	88.5	5.25	5.3 ± 0.67
2	92.7	5.34	4.2 ± 0.31
3	88.5	4.98	7.2 ± 0.65
4	84.6	5.34	5.6 ± 0.66
5	84.1	5.30	4.5 ± 0.45
6	84.8	5.34	5.4 ± 0.74
7	83.4	5.34	6.4 ± 0.62
8	93.7	5.34	4.8 ± 0.64
9	87.1	5.40	4.3 ± 0.43
10	84.1	5.34	8.1 ± 0.66
11	76.0	4.85	6.8 ± 0.81
12	80.0	5.41	7.7 ± 0.66
13	74.6	5.34	6.7 ± 0.83
14	76.9	5.34	8.5 ± 0.81
15	83.0	5.37	6.8 ± 0.97
16	77.8	5.54	5.2 ± 0.81
17	81.0	5.60	5.5 ± 0.72
18	79.9	5.77	5.6 ± 0.74
19	86.5	5.42	8.4 ± 0.74
20	81.0	5.34	8.8 ± 0.90
21	86.0	5.52	7.0 ± 0.94
22	77.4	5.53	5.0 ± 0.84
23	88.1	5.19	7.8 ± 0.71
24	78.1	5.60	7.9 ± 0.83
25	77.3	5.54	5.4 ± 0.12

Per cent transmission of quartz windows before coating with tin
oxide, 98.74 ± 0.34 .

All procedures of coating the "Kovar" cups with zinc sulfide and the quartz disks with tin oxide was done in a fume hood. Ethylene dichloride vapors produced in coating the "Kovar" cups are toxic and the gases produced by fuming stannic chloride include hydrochloric acid and other toxic products which had to be vented.

In the construction of the first ten counting bottles, the conductive surface of the quartz disk was rimmed with a silver loaded paint, of the type used to repair electronic printed circuits, to insure an electrically conductive bond between the tin oxide on the quartz disk and the coated walls of the "Kovar" cup. The quartz disk was then externally bonded to the "Kovar" cup with R 313 adhesive. Paint occluding the window portion of the quartz disk would reduce efficiency. In the second group of fifteen counting bottles, the quartz disk was bonded directly to the "Kovar" cup with R 321 adhesive (see list of materials, Appendix H). R 321 is a silver loaded, conductive variety of R 313 adhesive. The R-321 provided the necessary conductive path from the stannic oxide on the quartz disk to the coated walls of the "Kovar" cup without the necessity for rimming the disks with a conductive paint and avoided the problem of occlusion of the quartz window.

The coated walls of the "Kovar" cup and the stannic oxide coat on the inner face of the quartz disk formed a conductive shield which insulates the contents of the counting bottle from the effects of the potential on the photocathode of the photomultiplier tube (6).

After the quartz windows were bonded to the "Kovar" cups, the counting bottles were evacuated and checked for leaks. Typically, bottles leaked very slightly going from 0.050 mm. Hg to 1.00 mm. Hg in 24 hours. Following the leak check, counting bottles were placed in a vacuum desiccator, with the stopcocks left open. The vacuum desiccator was evacuated to 0.10 mm. Hg and the

counting bottles allowed to remain in that reduced atmosphere for 24 hours in order to remove the remaining ethylene dichloride (7).

Calibration

The counting bottles were then calibrated. Radium-226 standards from the National Bureau of Standards were used in the calibration. One was a 100 cc. standard which contained 10.01 pCi (10,010 fCi) of radium-226 as a chloride. This standard was transferred to a 500 cc. generator flask (see Appendix D) washing with 50 cc. of a special water, also obtained from the National Bureau of Standards, which contained 0.02 pCi (20 fCi) of radium-226. Thus the first standard contained 10.03 pCi (10,030 fCi) of radium-226.

The second standard was prepared by transferring, volumetrically, 5 ml. of a standard which in this case contained 9.99 pCi (9,990 fCi) of radium-226 per ml. to a 500 ml. generator flask. This standard was washed into the flask with 100 ml. of special water from the National Bureau of Standards which contained 0.04 pCi (40 fCi) of radium-226. Therefore, the second standard contained 49.99 pCi (49,990 fCi) of radium-226. These standards for convenience were designated as containing 10 pCi (10,000 fCi) and 50 pCi (50,000 fCi) of radium-226 respectively.

The background count of the radon counting bottles was determined by counting by the photomultiplier 24 hours or more. It ranged from 4.2 to 8.8 counts per hour (see Table 14). The backgrounds were rechecked at frequent intervals and the values readjusted as they changed.

The radium-226 standards were flushed free of radon, using dry radon free air. They were then allowed to stand until some radon had formed and assayed by the standard procedures used with all samples, as described elsewhere (see Appendix D).

In the calibration procedure, the radon-222 content was computed by starting with an arbitrarily assigned efficiency (E) of one count per minute per picocurie (see Appendix F). In primary calibration, since the number of picocuries contained in the standard was known, the efficiency of the counting bottle was:

$$E = \frac{\text{pCi as computed}}{\text{pCi present in the standard}} \quad (\text{see Table 15}).$$

Not all counting bottles were calibrated by using the radium-226 primary standards. Most were done by intercalibration or secondary calibration with all other counting bottles by comparing count rates for individual samples with analyses of the same sample in the standard or primary calibrated counting bottle, i.e., sample X was run a number of times using counting bottles A and B and calibrated counting bottle No. 1; sample Y was run a number of times using counting bottles A and C and calibrated counting bottle No. 2, etc.

During intercalibration, the radon-222 content for each assay was computed using an E of 1 count per minute per picocurie for the bottles being calibrated. The radon-222 content of the sample was determined using a bottle which had been calibrated using a radium-226 primary standard and had a known E. The efficiency of the counting bottle to be determined was equal to the computed radon-222 content for an E of 1 count per minute per picocurie divided by the radon-222 content computed for the same sample using a counting bottle of known efficiency for the analysis. Efficiencies for most counting bottles were within 5% of the mean value for all counting bottles.

TABLE 15

EXAMPLES OF PRIMARY CALIBRATION OF COUNTING
BOTTLES

50 pCi Standard	10 pCi Standard
Bottle No. 2 for a computed efficiency of 5.34 counts per minute per picocurie (pCi) of radium-226	
<p>a. Computed pCi</p> <p>267.0 ± 7.0</p> <p>$E = \text{computed pCi/pCi in standard}$</p> <p>$E = 5.34 \text{ counts per minute per pCi}$</p>	<p>a. Computed pCi</p> <p>55.0 ± 2.0</p> <p>$E = \text{computed pCi/pCi in standard}$</p> <p>$E = 5.50 \text{ counts per minute per pCi}$</p> <p>b. Computed pCi</p> <p>50.0 ± 1.0</p> <p>$E = \text{computed pCi/pCi in standard}$</p> <p>$E = 5.00 \text{ counts per minute per pCi}$</p>
Bottle No. 3 for a computed efficiency of 4.98 counts per minute per picocurie (pCi) of radium-226	
<p>a. Computed pCi</p> <p>249.0 ± 7.0</p> <p>$E = \text{computed pCi/pCi in standard}$</p> <p>$E = 4.98 \text{ counts per minute per pCi}$</p> <p>b. Computed pCi</p> <p>249.0 ± 8.0</p> <p>$E = \text{computed pCi/pCi in standard}$</p> <p>$E = 4.98 \text{ counts per minute per pCi}$</p>	

TABLE 15--Continued

50 pCi Standard	10 pCi Standard
Bottle No. 15 for a computed efficiency of 5.37 counts per minute per picocurie (pCi) of radium-226	
<p>a. Computed pCi</p> <p>253.0 $\pm$ 7.0</p> <p>E = computed pCi/pCi in standard</p> <p>E = 5.16 counts per minute per pCi</p>	<p>Some of the 10 pCi standard was lost due to a pressure blowback on one run. The standard was restandarized using counting bottle No. 2 at:</p> <p>9.00 $\pm$ 0.49 pCi radium-226</p> <p>Computed pCi</p> <p>48.0 $\pm$ 2.0</p> <p>E = computed pCi/pCi in standard</p> <p>E = 5.33 counts per minute per pCi</p>
<p>b. Computed pCi</p> <p>272.0 $\pm$ 8.0</p> <p>E = computed pCi/pCi in standard</p> <p>E = 5.44 counts per minute per pCi</p>	
<p>c. Computed pCi</p> <p>278.0 $\pm$ 9.0</p> <p>E = computed pCi/pCi in standard</p> <p>E = 5.56 counts per minute per pCi</p>	

APPENDIX B

STANDARDIZATION OF COUNTING EQUIPMENT

The electronic counting equipment used in this study was of the conventional decade scaler type. The critical requirement for the electronic equipment was the instrumental noise. The maximum allowable instrumental noise at the operating point could not exceed three counts per 1,000 minutes. Scalers with non-overloading linear amplifiers and ten turn helipot discriminators using cathode follower or emitter follower preamplifiers and EMI type 9536B photomultiplier tubes in shielded probes provided high sensitivity with low instrumental noise.

The probes were mounted in a vertical position so that the face of the photomultiplier tube was readily exposed. In use, counting bottles were placed on the photomultiplier tube with the quartz window in direct contact with the face of the photomultiplier tube. No special optical coupling medium was used between the quartz window and the face of the photomultiplier tube, just direct air coupling. The counting bottle and the photomultiplier tube photo cathode had to be shielded from light. A spun aluminum can served as a light shield which completely enclosed the counting bottle and fit into a grooved ring in the support for the shielded probe (see Figure 2). The grooved ring served as a light baffle to prevent light from entering around the bottom of the light shield.

Operational plateaus (see Figure 3) were determined for each scaler-probe combination by counting a sample from a 499.5 pCi (499,500 pCi) radium-226 source. These samples ordinarily had a count rate of 750 to 800 counts per

Fig. 2.—Radon collection system showing a generator flask of the 1 to 5 gallon size, the CO₂ trap, and cold finger moisture traps in the system. This arrangement is suitable for CO₂ producing sample solutions.

RADON GENERATOR FLASK
 (Small solution volume)
 Volume of solution limited to 20% to 80%
 of flask volume

Figure 3.—Small Radon generator flask usually of 500 cc. or 1,000 cc. capacity, suitable for a maximum sample solution volume of 400 cc. or 800 cc. respectively.

minute when drawn. The sample was allowed to decay for 2 days before counting when the rate of decay was at 3% over a 6-hour counting period. Counting the sample at this rate of decay introduced the least possible error that could be obtained in the establishment of the operational plateau. The counts for establishing the operational plateau could usually be run in less than 6 hours. At the time the sample was counted, the variation due to the rate of decay was of the same order of magnitude as the uncertainty in count due to variations in distribution of the time of disintegration of the atoms being counted.

A series of 3-minute counts was made at 50 volt increments of the high voltage applied to the photomultiplier tube at each of several discriminator settings. This series of determinations of count rate was plotted as a family of curves (see Figure 3). From this family of curves an optimal operational point was chosen by selecting the best high voltage-discriminator setting combination. The discriminator setting chosen was the one providing 20 millivolts of discrimination, i.e., all pulses with less than 20 millivolts magnitude were not counted. The high voltage setting was then adjusted so that the operating point was at least 100 volts beyond the beginning of the constant count rate plateau of the curve.

Operational plateaus were determined both for direct coupling of the counting bottle and the photomultiplier tube and for coupling through 1/32 inch of plexiglass plus 1/8 inch of air. The plastic-air gap occurred in an automatic sample changer designed for one of the counting systems. The 1/32 inch of plexiglass and an 1/8 inch air gap was to protect the quartz window of the counting bottle and the face of the photomultiplier tube during rotation of the sample changer table (see Figure 2). The addition of the 1/32 inch of plexiglass and 1/8 inch air gap in the sample changer did not appear to influence the count rate of a sample (see Figure 3).

Although all scalers were standardized by using radon samples in counting bottles to determine the optimal operational points which would give identical counts (within counting error), a secondary standard had to be constructed. This secondary standard was used to monitor and correct drift in the operating point caused by day to day variations in line voltage. All scalers were operated on a regulated 110 volt line. However, even this regulated 110 volt line had an appreciable variation in line voltage over an extended period of time. The drift in optimal operating point could be corrected by adjusting the high voltage setting.

The secondary standard used to monitor and correct drift in the optimal operating point was constructed by spreading a few drops of plutonium-239 on a stainless steel planchet 2 inches in diameter. Enough plutonium-239 was used to provide a total activity of 40,000 disintegrations per minute. A 1/2 inch thick section of plexiglass rod 2 inches in diameter was "squared off" and "faced off" in a lathe, then polished as flat as possible at both ends. A thin layer of zinc sulfide scintillator was sprayed on one end of the plastic disk. The planchet was then mounted on the disk with the plutonium-239 facing the zinc sulfide. The planchet was secured to the disk by three stainless steel, 1-72, screws in holes drilled and tapped in the plastic.

Alpha particles emitted by the plutonium-239 would cause measurable scintillations in the zinc sulfide scintillator. However, the thin layer of zinc sulfide would not completely absorb all alpha particles. The variation in absorbed energy from the alpha particles tended to create a pseudo-spectrum of energies registering on the photomultiplier tube. Because of this pseudo-spectrum of energies, the count rate for the secondary standard never reached a plateau as did the count rate for a sample of radon in a counting bottle. Changing either the high voltage setting or the discriminator setting would

change the count rate of the secondary standard. To calibrate the secondary standard, the optimal operating point was located using a sample of radon in a counting bottle. As soon as the optimal operating point was located, a series of 3-minute counts of the secondary standard were made with no change in either high voltage setting or discriminator setting. The mean value of this series of counts was then used as the count rate for the secondary standard at the optimal operating point of the counting equipment.

Before counting a sample, the secondary standard was counted for 3 minutes. If the count rate was too high, the high voltage setting would be reduced; if too low, the high voltage setting increased. Then another 3-minute count of the secondary standard would be made. When any drift in the optimal operating point had been corrected by adjusting the high voltage setting until the count rate of the secondary standard indicated the optimal operating point had been located, the sample could be counted. It was not possible to correct for any drift of the optimal operating point that might occur during long counts.

Reproducibility of Counts

Repetitive counts were made of several samples to determine how much error would be introduced by counting variations. The counts were made as follows: a sample would be counted to a specific number, usually 2,500 counts, and the times required for the measurements noted. Then, without changing the sample, the scaler would be reset and the sample counted again. This was repeated five times. Counts were made on both relatively high and relatively low activity samples. The total time used to make the repetitive counts was not so long that the rate of radon-222 decay would introduce errors.

Standard error for the repetitive counts was then computed. Twice the standard error of the mean (95% confidence limits) ranged from 3% to 6%. It was, therefore, decided that 95% of the counts should be within $\pm 5\%$ of the

mean. Any counts (outliers) of a series of analyses of a sample that did not agree with previous counts to within 5% or less were discarded.

In some samples with extremely low activity, it was necessary to use overlapping of the 95% confidence limits of the samples as the criterion for discarding individual analyses in a series. This breakdown of normal procedure in the case of these low-activity samples was caused by the indeterminacy of the precision with which the background count of the counting bottle used could be determined. The 95% confidence limits for repetitive counts ranged as high as - 80% in these cases.

Usually counts that were outside of the 95% confidence limits were due to mechanical difficulties or problems in collection and transfer of the radon to counting bottles. In most cases, both the existence of an erroneous count and the direction of error, high or low, could be observed and/or predicted by close observation during collection and transfer of the radon. Though the errors could often be seen or predicted, it was impossible to correct them at the time the sample was being analyzed.

In all cases the measurements were repeated until the operator was satisfied that the data were in agreement within the precision of the method and equipment used.

APPENDIX C

PREPARATION OF SAMPLES

The chief problem in sample preparation for determination of radium-226 content by the emanation technique is getting the sample into solution. For example, the radon in respired air can be trapped directly, after the moisture is removed, on activated charcoal in a radon trap. However, in all samples except gaseous ones, the radon must be retained during a period of growth while the sample is freely in solution and held in a sealed vessel. Quantitative recovery of radon from a solution is possible only if the solution is maintained in a sealed vessel and then the solution and vessel are washed with a sufficient quantity of carrier gas at the time the radon is transferred to a radon trap (see Appendix D).

Since the mean path of the recoiling radon atom following alpha emission by a radium atom is of the order of $200 \text{ \AA}$ (8), it is doubtful that the radon atom can recoil free of all solid or crystal structures in which it might be born. Therefore, the sample must be dissolved and the solution must be maintained at the proper level of acidity or alkalinity or have had the proper chelating agents introduced to keep all atoms in solution--free of imprisonment in solids or crystals. Radon-222 determinations by the emanation technique, as done in this study, can be made on any substance which can be dissolved.

The determination of radon using the emanation technique is particularly useful when the concentration of radium in the sample is very low. It is possible, for example, to concentrate the radon from a large or dilute sample by trapping the radon on activated charcoal at a low temperature. However,

the concentration of the radon from a large or dilute sample can create problems, especially in sample preparation.

Techniques of sample preparation which work adequately on small samples often break down when they are applied to large samples. Techniques of sample preparation, therefore, had to be developed or modified so that various types and sizes of samples encountered could be used in radon-222 determination by the emanation technique.

Bone

Bone samples were prepared by first removing as much flesh as possible by dissection. The clean bones were then ashed in fused quartz trays at 600-800° C. for 18 hours. When the ashed samples had cooled, 75 gram aliquots of ash were weighed and put in 600 ml. pyrex beakers. If the total weight of the sample was less than 75 grams, the whole sample was used.

Concentrated nitric acid was added to the ashed bone aliquot at the rate of 1.85 cc. of acid for every gram of ash. This ash-acid mixture was then heated until all of the bone ash dissolved. The solution of bone ash was diluted to a volume of 250 ml. using 1 molar nitric acid.

The diluted solution was filtered while hot through "Whatman No. 41" filter paper into a pyrex generator flask (see Figure 4). The beaker and filter paper were washed with 50 ml. of 1 molar nitric acid, so that the total volume of bone ash solution was 300 ml.

Tissue, Feed, and Feces (First Steps)

Part of the procedure for preparing ashed samples of tissue, feed, and feces was identical. Because of the large volumes involved in each sample (tissue 16 to 17 kg., feed 2 to 3 kg., feces 2 to 4 kg.), wet ashing techniques were not appropriate. High temperature ashing was also considered inappropriate

Fig. 4—Large radon generator flask usually of 1 to 5 gallon capacity, suitable for sample solution volumes of 1 to 16 liters.

because of the creation of "fly-ash," and the subsequent loss of portions of the sample, associated with the high temperature ashing.

A modified high temperature ashing was developed. The sample was first charred by heating at 800° to 850° F. for 18 hours in a closed vessel. In this case, the vessel was a 5-gallon stainless steel stock kettle and lid. Later samples were charred in covered "in conel" vessels constructed for that purpose. At this temperature with the lid on the kettle, oxidation was incomplete because of the restricted air supply. All of the volatile hydrocarbons boiled off leaving a charcoal-like mass. This mass was heated in a muffle furnace at 650° C. in a fused quartz tray. As the carbon was oxidized, more of the charcoal-like mass was added to the tray until all of the carbon was gone leaving only the mineral ash. This approach to high temperature ashing minimized or prevented the problem with "fly-ash."

The muffle furnace had a 12-liter volume and air was released into the muffle at the rate of 1.2 liters per minute, providing a turnover of 14.4 moles of O_2 per day. Therefore, about 172.8 grams of carbon could be oxidized per day. This approximation was used as an estimate of the period of time required to finish ashing each sample. After the samples of tissue, feed, and feces were ashed, the preparation of each category of sample required special procedures.

Tissue (Second Steps)

The total volume of tissue ash was weighed and placed in a 600 ml. pyrex beaker. The tissue ash was dissolved by adding 1.5 ml. of aqua regia per gram of ash and heating. Next concentrated hydrochloric acid was added until NO_2 was no longer evolved, converting the ash to chlorides. The chlorides, especially potassium chloride, are more soluble below 30° C. than

are the nitrates. After the hydrochloric acid was added to the hot solution and all NO_2 driven off, the solution was diluted to 500 ml. with 1 molar hydrochloric acid. The tissue ash solution was then filtered hot through "Whatman No. 41" filter paper into a pyrex generator flask (see Figure 4). The beaker and filter paper were washed with 100 ml. of 1 molar hydrochloric acid so that the total volume of tissue ash solution was 600 ml.

Feed and Feces (Second Steps)

The total volume of a sample of ash of feed or feces was weighed and placed in a 600 ml. pyrex beaker. The ash was then dissolved by heating in 1.5 ml. of aqua regia per gram of ash, then adding concentrated hydrochloric acid until NO_2 was no longer evolved. The hot solution was diluted to 500 ml. with 1 molar hydrochloric acid, and filtered hot through "Whatman No. 41" filter paper in a Buchner funnel into a pyrex generator bottle (see Figure 5). The residue on the filter paper was washed with 100 ml. of hot concentrated hydrochloric acid, then with 600 ml. of hot 1 molar hydrochloric acid, and finally with 1,000 ml. of hot distilled water. The washes with hydrochloric acid dissolved any ferric phosphate which may have formed and precipitated.

The final volume of feed or feces ash solution in the generator bottle was 2.2 liters.

Urine and Blood (as Liquids)

Urine collected with a urine feces separator was measured and transferred to a generator flask (see Figure 5) of appropriate size.

Blood was collected with EDTA (ethylenediaminetetracetic acid) solution, measured, and transferred to a generator flask of appropriate size. The EDTA solution was used to chelate the blood calcium and prevent coagulation. Twelve and five-tenths grams of tetrasodium EDTA in solution, buffered to physiological

Fig. 5.—Dimensions and construction of radon trap

pH was used for every gram of calcium anticipated. This is about 12.5 grams of tetrasodium EDTA per 8 liters of blood.

These liquid samples could be used with a little further preparation as dilute, large volume samples.

The only further preparation required was the addition of 10 to 20 ml. of octanoic (caprylic) alcohol to urine samples and 3 to 6 ml. to blood samples to prevent foaming by reducing surface tension.

Blood and Milk (as Ashed Samples)

Samples of blood, with anticoagulants, or milk were transferred 500 ml. at a time to "pyro-ceram" or "vicor" dishes and heated until carbonized at 350° C. That is, each 500 ml. of sample was heated until dry, then the next 500 ml. of sample was added until all of the sample had been measured and heated until carbonized and dry in a single dish.

The dishes containing dry carbonized samples were placed in a muffle furnace and heated for 24 hours at 500° C. to ash the sample. The total volume of sample was weighed and dissolved in a minimum of concentrated hydrochloric acid (about 2 ml. per gram of ash) while heating, then diluted to 250 ml. with 1 molar hydrochloric acid. The diluted solution was filtered hot through "Whatman No. 41" filter paper into a generator flask (see Figure 4) and washed with 50 ml. of 1 molar hydrochloric acid, making the total volume of blood or milk ash solution 300 ml.

The carbonizing was done in vessels which would hold at least 1-3/4 quarts. The largest vessel compatible with the size of the muffle should be used. The large size was required since the blood and milk, especially the blood, expand during the first phase of carbonizing and might overflow. Also during the ashing in the muffle furnace, the carbonized material will foam and overflow if the vessel is too small. The 1-3/4 quart size mentioned was

adequate for samples up to 3 liters in volume, added and carbonized in 500 cc. aliquots.

The diluted solutions were filtered into the generator flasks to remove any unoxidized carbon, or other insoluble material. The weight of insoluble residue on the filter paper was subtracted from the total weight of the sample. The difference obtained was used as the weight of sample for determining the amount of radon per aliquot (total radon/sample weight).

The residue from samples of feces was surprising in its magnitude. A kilogram sample of feces contained up to 75 grams of residue. Microscopic examination indicated it was silicates. This microscopic examination together with gross observation of pebbles of various sizes in the residue was clear evidence that much sand and soil was consumed by the animals. In all other cases, except the feces, the residue contained less than 0.5% of the weight of the sample. Usually the residue in these samples contained a small amount of carbon and possibly some silica chips acquired during ashing procedures.

Concentrated nitric acid was avoided when dissolving any samples except bone. The samples other than bone which were dissolved in concentrated nitric acid showed a tendency to foam, even after they were diluted with 1 molar nitric acid. This tendency makes the samples more difficult to work with and necessitated the addition of a few drops of octanoic acid when the samples were transferred to generator flasks.

APPENDIX D

RADON COLLECTION SYSTEM

The radon collection system is the first in the chain of systems for "milking" the radon-222 produced by radium-226 in a sample and for transferring the radon-222 to a radon counting bottle for quantitative measurement. Simply described, the system provides a supply of dry radon-free air with which radon is flushed from a radon generator flask to a radon collection trap.

The radon collection system contains, from beginning to end, these components: a tank of dry compressed air, a molecular sieve, a moisture trap, a radon trap, a generator flask, a pair of moisture traps, a radon collection trap, and a manostat flow meter (see Figure 6). When the radium-226 was in a large volume of liquid, a pair of cold finger moisture traps could be inserted between the generator flask and the pair of moisture traps. When urine or liquid blood, serum or plasma samples were to be measured, a CO₂ trapping column was inserted between the generator flask and the pair of cold finger moisture traps (see Figure 7). Of the various components, only the pair of moisture traps and the manostat flow meter were rigidly mounted in place. The first moisture trap was mounted so that it would swing out of the way and allow insertion of a freshly activated radon trap at the start of each day's series of analysis. Generator flasks and radon collection traps were changed for each sample analyzed.

The molecular sieve (Linde, type 5A, see list of materials, Appendix H) was enclosed in a 3-foot length of 3-inch diameter pipe and all joints were sealed with pipe joint sealing compound. One end of the enclosure was connected

Fig. 6.—Dimensions and construction of the radon transfer system.

Fig. 7.—Dimensions and construction of radon counting bottle.

to the pressure regulator on the tank of dry air and the other was filled with a needle valve. A length of flexible copper tubing connected the needle valve to the inlet arm of the moisture trap, the copper tubing being joined to the glass inlet arm by a short length of gum rubber tubing.

The molecular sieve acted as a prefilter for the system, trapping water vapor and scavenging oil vapors, chemical contaminants, and other particulate materials. The sieve assured that only very dry air would enter the system.

Radon traps were replaced each day at the start of a series of analyses. Although each charcoal radon trap had trapping capacity of 500,000 millicuries or more of radon (5), full trapping capacity was provided each day by a fresh radon trap. Use of a fresh trap also assured that no contaminants would be carried in the system for more than 1 day. The radon traps were activated by heating and flushing with helium as described in Appendix B.

The generator flask and radon collection trap were bypassed by gum rubber tubing bypasses. These were built into the system line so that, with the exception of the generator flask and the radon collection trap, the system could be flushed free of radon-containing ambient air by allowing a flow of dry radon-free air to purge the system. This approach to removing ambient air provided an alternative to evacuation of the system followed by filling the system with dry radon-free air as suggested by Lucas (2). Originally the system was constructed without the CK-2 stopcocks in the bypasses. Screw clamps were used to close the bypasses. Analyses made with this original system were very erratic and completely irreproducible. On the assumption that the initial surge of air through the generator flask into the rest of the system forced some radon-laden air past the screw clamps, the system was modified so that CK-2 stopcocks closed the bypasses. When this modification was made, assays became readily reproducible. Successive assays of the same sample often agreed to within 0.5% and most agreed within 6%.

The twin cold finger moisture traps were employed in the system line for samples exceeding 1 liter in volume. The large volumes of air required to transfer the radon from these large samples to the radon collection trap carried more moisture than the two permanent moisture traps could remove. The twin cold finger traps reduced the load on the permanently installed moisture traps to manageable proportions. They were also used whenever the CO_2 trap was in the system.

The CO_2 trap was constructed to hold 300 cc. of 4.2 molar potassium hydroxide solution. Dispersion of air passing through the CO_2 trap was provided by one-eighth pound of 4 mm. glass beads, supported by a No. 30 coarse glass frit at the bottom of the trap (see Figure 7). The trap was filled with the potassium hydroxide solution immediately before use and emptied, washed, and dried immediately after use. This procedure was required in order to reduce the possibility of the potassium hydroxide solution etching any radium-226 from the glass walls of the trap.

The CO_2 trap was flushed free of radon during purging of the system line, by allowing the dry radon-free air to flow through the potassium hydroxide solution at a rate of 2 liters per minute for 20 minutes. Following this, the solution in the CO_2 trap would contribute only 1.11 fCi of radon-222 per hour to the total amount of radon-222 trapped.

The CO_2 trap was employed when samples of urine, liquid blood, serum, or plasma were assayed. The slow breakdown of organic materials in these solutions produced enough CO_2 to poison the activated charcoal in the radon collection trap. This was empirically and dramatically observed. Whenever an excessive amount of CO_2 was collected in the radon collection trap, it would blow apart at the 29/42 pyrex ground joint during heating to 450° to 500° C. in the radon transfer system.

The radon collection system was constructed from 8 mm. pyrex tubing, 12/5 pyrex ball and socket joints and 29/42 pyrex ground joints so that as far as possible parts were interchangeable. The dimensions of the system were not critical but were governed by the space available. The system was constructed so the 500 cc. and 1,000 cc. generator flasks would fit into a space with a constant distance between inlet and outlet arms. When the cold finger moisture traps and/or the CO₂ trap were used, the distance between inlet and outlet arms was constant but not identical with the constant distance space provided for the 500 cc. and 1,000 cc. generator flasks. The cold finger moisture traps and CO₂ trap were placed behind the plane of the rest of the system so that the larger generator flasks could be employed. A minimum of pure gum rubber tubing was used to connect components, since Lucas (1) states that if more than 30 cm. of rubber tubing is used, a significant contribution by radon in the ambient air can occur through the tubing. The rubber tubing used in the bypasses need not be included in this 30 cm. limit.

Generator Flasks

Two varieties of generator flasks were used in this study. The size and variety used were governed by requirements for interchangeability of components and sample size. The first was a specially constructed 500 cc. or 1,000 cc. round bottom pyrex flask as shown in Figure 4. Dimensions were not critical, but adjusted so the flasks would fit into the system. The stopcock on the outlet arm was placed 6.35 cm. away from the wall of the flask so that bubbles formed while air was flowing through the flask would not be blown past the stopcock and sample solution lost. The flasks were filled, emptied, and cleaned through the 14/35 joint on the inlet arm.

The second variety of generator flask was a pyrex bottle of either 1, 2.5, 3.5, or 5 gallon capacity adapted as shown in Figure 5. These generator

flasks were always used with the CO_2 trap and/or the cold finger moisture traps. Short lengths of gum rubber tubing in the inlet and outlet arms allowed maximum interchangeability and flexibility of positioning. On some occasions, teflon-coated, magnetic stirring bars were used in these large generators to facilitate gaseous exchange between sample solution and air in the free air space above the solution. Stirring for 24 hours before analysis of a sample yielded erratic results but appeared to reduce the half transfer volume (see discussion of half transfer volume, page 114) by $1/3$. Because such stirring was undependable, this practice was omitted in subsequent samples.

Radon Traps

Radon traps (see Figure 8) were constructed to specifications suggested by Lucas (1). A radon trap could be used in the radon collection system in either of two ways: (1) the trap designated the "radon trap" was to remove radon from dry air entering the system, and (2) the trap designated the "radon collection trap" was to trap radon produced by the sample and flushed from the generator flask for assay. The only difference between these two uses was the location and purpose of the trap.

Operation of the Radon Collection System

The radon collection system used in this study was a modification of that first described by Lucas (2). The system described by Lucas was evacuated, then filled with dry radon-free air; however, the system employed in this study was only purged with dry radon-free air before use. Had the radon in residual air which was left in the dead air spacer of the system during the purging operation caused an appreciable error when added to the extremely low radon content of a sample, this system also could have been evacuated. In most

Fig. 8.—Arrangement of radon counting bottles and photomultiplier tubes in the counting equipment showing: (A) Direct contact coupling of radon counting bottle and photomultiplier tube, (B) 1/32" plexiglass-1/8" air gap coupling of radon counting bottle and photomultiplier tube in an automatic sample changer, (C) general layout of the automatic sample changer.

cases no error could have been introduced because the evacuation step was omitted.

The radon collection system was used for two purposes: (1) to purge a generator flask of all radon when a sample was prepared for the first analysis, and (2) to flush all radon from a generator flask to a radon collection trap for analysis of the radium-226 content of the sample. In both cases the sequence of operations was similar: The system was cleaned and assembled as shown in Figures 1 or 2. All radon traps were cooled to -70° C. in dry ice-alcohol baths. Valves and stopcocks, exclusive of the generator flask and radon collection trap, were opened and dry radon-free air allowed to purge the system. After the system had been purged with 5 liters of dry radon-free air, the radon collection trap bypass was closed and the radon collection trap opened to the flow of air. If the flow rate indicated by the manostat flow meter showed that there was a patent airway through the radon collection trap, the generator flask bypass was closed and the generator flask opened. When the necessary volume of air required to quantitatively transfer radon produced in the generator flask (ten or more half transfer volumes, see discussion of half transfer volume, page 114) had passed through the generator flask, both the radon collection trap and the generator flask were closed as nearly simultaneously as possible. The radon collection trap, still in its dry ice-alcohol bath was transferred to the radon transfer system for further processing (see Appendix E). The generator flask was placed aside to "age" until time for the next analysis.

If the generator flask was being purged free of radon, prior to the first collection of the sample for analysis, the sequence of operations was identical with one exception--the radon collection trap bypass was not closed nor the radon collection trap opened. No quantitative transfer of radon was

made, but the manostat flow meter was still used to determine the total volume of air passing through the generator flask.

All generator flasks were allowed to "age" before a quantitative analysis for radon content was attempted. The term "age" is used here to indicate the time during which radium-226 in the sample solution was permitted to decay to radon-222. The per cent equilibrium that radon-222 would reach with respect to the radium-226 present is related directly to the time elapsed after the generator flask was closed after being purged or flushed free of radon-222. The rate of increase of per cent equilibrium slows rapidly after 14 days (see Table 16) so no sample was "aged" for more than 14 days.

TABLE 16

PER CENT EQUILIBRIUM BETWEEN RADON-222 AND
RADIUM-226 FOR VARIOUS TIME INTERVALS
OF AGING

Days	Per Cent Equilibrium	Days	Per Cent Equilibrium
0.5	8.66	8.5	78.57
1.0	16.57	9.0	80.42
1.5	23.80	9.5	82.12
2.0	30.40	10.0	83.67
2.5	36.43	10.5	85.08
3.0	41.94	11.0	86.38
3.5	46.97	11.5	87.56
4.0	51.56	12.0	88.63
4.5	55.76	12.5	89.62
5.0	59.59	13.0	90.52
5.5	63.09	13.5	91.34
6.0	66.29	14.0	92.09
6.5	69.21	15.0	93.40
7.0	71.88	20.0	97.33
7.5	74.31	25.0	98.92
8.0	76.54	38.25	99.44

Since all aspects of the analysis are extremely time-dependent, the exact time and date of each critical operation was carefully noted during the

analysis. A card was attached to each generator flask with positions to note the following: (1) sample number and analysis number, (2) radon counting bottle used, (3) time and date generator flask was closed after being purged free of radon, (4) time and date generator flask was closed after radon was flushed quantitatively to the radon collection trap, (5) time and date radon collection trap was placed in the heater on the radon transfer system, (6) time and date of start and stop of counting, and (7) total count for this analysis (see Appendix F).

Corrections for Dead Space

As noted before, the use of pressure purging with dry radon-free air instead of evacuation plus purging introduced a small but not significant error in the total radon-222 absorbed in the radon collection trap. Dead spaces in the lines of the system are created largely by the location of the stopcocks in the system. It is not possible to purge the residual air from these spaces. Assuming the radon content of ambient air to be 276.0 fCi per liter (3) then the system will contain the following residual radon content (Table 17).

TABLE 17
CORRECTION FOR DEAD SPACE FOR VARIOUS SYSTEMS

Type of Generator Flask	Dead Space in Liters	Residual Radon in fCi
1. 500 cc or 1,000 cc.	0.009048	2.50
2. 1 gallon		
a. CO ₂ trap and cold finger moisture traps in use	0.01627	4.49
b. Only cold finger moisture traps in use	0.01664	4.59
3. 2.5, 3.5, or 5.0 gallon		
a. CO ₂ trap and cold finger moisture traps in use	0.01188	3.28
b. Only cold finger moisture traps in use	0.01239	3.40

It was decided that when the residual radon was $\leq 1\%$ of the radon content of the sample, no correction would be made for residual radon in the system.

Half Transfer Volume

The volume of dry radon-free air required to flush 50% of the radon present from a given volume of solution was called the half transfer volume. The half transfer volume was based on the quantity of radon-containing solution rather than the size of the generator flask even though 25% or less of the radon was held in solution in the liquid volume (4.5). The radon in the free air space above the solution was readily swept clear of radon by the air passing through the space, primarily by the process of dilution.

Flushing radon from the solution was more complex since it was the product of the coefficient of the solubility of radon in water and the surface area of the air-water interface within the bubbles of air passing through the solution. The rate of flow of air through the solution, as will be shown later (Table 19), did not appear to change the half transfer volumes.

Six or more half transfer volumes were required to flush a minimum of 98% of the radon from the generator flask. It was the policy to use at least fifteen half transfer volumes for all collections. Although Lucas (2) used a 30-liter flush at the rate of 1 liter per minute for a 500 cc. generator flask, he has said (1) that as a rule of thumb the half transfer volume should be equal to twice the volume of solution with the exception of whole blood, plasma, or serum. The half transfer volume for the latter samples was equal to six times the volume to be flushed.

Half transfer volumes were determined for several combinations (Table 18); the solution used was water "spiked" with 28 pCi of radium-226 per liter.

TABLE 18

HALF TRANSFER VOLUMES FOR LARGE SOLUTION VOLUMES

Volume of Solution in Liters	Number of Analyses	Generator Flask in Gallons	Half Transfer Volume $\pm$ C.L. 95 in Liters
18	5	5.0	30.8 $\pm$ 5.1
12	3	5.0	20.5 $\pm$ 9.6
6	2	3.5	11.1 $\pm$ 2.3

The large spread in the 95% confidence limits, especially in the sample with 12 liters of solution in a 5.0 gallon generator flask, is due to variations in the rate of flow of dry radon-free air through the flask. In analyzing these samples the total volume of air which passed through the system was computed by multiplying the rate of flow indicated on the manostat flow meter by the time during which air was allowed to flow through the system. This agreed within 1% of total air volume that was measured by a wet test flow meter if there was no obstruction of the airway to cause reduction in flow rate. The best control of the total volume of air flushed through the system line was obtained by using a wet test flow meter along with the manostat flow meter. By always using a total flushing volume of at least fifteen half transfer volumes, it was assured that any transitory blockage of the line would not reduce the total flushing volume to less than the equivalent of six half transfer volumes (98% transfer).

The rate of flow of the air flushing radon from the generator flask to the radon collection trap did not appear to influence the transfer of radon. Flow rates between 0.5 liters per minute and 2.0 liters per minute were used both on large and small generator flasks. Values obtained for individual sample analyses were within the 95% confidence limits for the mean value for

the sample or within 5% of the mean value, irrespective of differences in rate of flow. Typical observations (Table 19) are:

TABLE 19
EFFECT OF FLOW RATE ON SAMPLE ANALYSIS

Sample (Bone Ash)	Analysis Number	Flow Rate Through the System (liter/minute)	Radon Content Computed for This Sample Analysis (fCi)	Mean Radon Content Measured in Sample (fCi)
7-8	6	2	4.4	4.4 $\pm$ 0.1
5-1	3	1.5	48.3	46.6 $\pm$ 0.8
1-3	4	1.2	15.6	15.6 $\pm$ 0.5
7-14	3	2	6.8	6.6 $\pm$ 0.2
1-2	4	2	10.2	10.3 $\pm$ 0.2

The rate of flow of air through the system line was limited more by the ability of the moisture traps to dry the moist air from the generator flask than by the capacity of the air moving at that flow rate quantitatively transfer radon to the radon collection trap. Excessive water vapor in the air competed with the radon for absorption on the charcoal in the radon collection trap. This in turn interfered with quantitative transfer of radon from the radon collection trap to the radon counting bottle (see Appendix G).

APPENDIX E

RADON TRANSFER SYSTEM

After the radon-222 had been trapped and concentrated on activated charcoal in a dry ice-alcohol bath, it was transferred to a radon counting bottle (see Appendix A) to be counted.

The radon transfer system used (see Figure 9) was modeled after the apparatus described by Lucas (6). The system was constructed of 6 mm. ID pyrex tubing with the following exceptions: (1) the mercury manometer tube was of 1 mm. bore, pyrex, capillary tubing, (2) the stems of the stopcocks were of 3 mm. ID pyrex tubing, and (3) the pure gum rubber tubing passing through the peristaltic pump was of 6.35 mm. ID.

The volume of the tubing on the radon trap side of the peristaltic pump was 19.14 cc. and volume of tubing on the counting bottle side of the peristaltic pump was 8.40 cc. This was a total dead air space of 27.54 cc.

The system was simple in operation. The stopcocks of the radon collection trap was closed and the trap removed from the radon collection system still in its dry ice-alcohol bath. The radon collection trap was then connected to the radon transfer system at the point where the radon counting chamber is shown in Figure 9. The radon collection trap, still in the dry ice-alcohol bath was opened to the vacuum line of the radon transfer system and evacuated to 0.200 mm. Hg or less. Lucas (6) evacuated to 0.020 Hg. in 5 minutes, but the vacuum pump used with this system took 15 minutes to evacuate the radon collection trap to 0.200 mm. Hg. After the radon collection trap was evacuated to ≤ 0.200 mm., it was then closed and removed from the radon transfer system.

Fig. 9.—Operational plateaus for one scaler for:
 (A) direct coupling of photomultiplier tube and radon counting bottle (1) count rate, directed coupled, 593.6 ± 5.5 cpm, (2) count rate, coupled through $1/8$ " air gap and 1.32 " of plexiglass, 585.2 ± 13.6 cpm; (B) $1/32$ " plastic- $1/8$ " air gap coupling of photomultiplier tube and radon counting bottle in an automatic sample changer (1) count rate, direct coupled, 508.0 ± 7.1 cpm, (2) count rate, coupled through 1.8 " air gap and $1/32$ " of plexiglass, 506.8 ± 7.1 cpm.

Next, a counting bottle was connected at the point where the radon collection trap had just been removed, and opened to the vacuum line and evacuated. It was left open to the vacuum line.

The evacuated radon collection trap was next removed from the dry ice-alcohol bath, wiped, inserted into the heater and connected to the radon transfer system. The remainder of the radon transfer system, with the exception of the radon collection trap and the helium line, were then opened to the vacuum line. All portions of the radon transfer system opened to the vacuum line were evacuated to ≤ 0.200 mm. Hg.

When the radon transfer system had been evacuated to 0.200 mm. Hg, the stopcocks to the helium reservoir and to the vacuum line (see Figure 9) were closed.

The helium reservoir was filled to a pressure of 1.5 pounds as shown on the vacuum-pressure gauge connected to the reservoir and the valve to the helium line closed. The evacuated radon collection trap was opened to the radon transfer system and the stopcock between the radon transfer system and the helium reservoir opened momentarily to allow a 15 cc. portion of helium into the radon transfer system and radon collection trap.

Finally the peristaltic pump was started and run for one minute, transferring helium and radon into the evacuated counting bottle on the other side of the pump. This cycle of filling the helium reservoir, bleeding it into the radon transfer system, and transferring the helium and radon to the counting bottle was repeated, until on the counting bottle side of the radon transfer system, atmospheric pressure was reached as indicated by the mercury manometer.

The peristaltic pump was run at 380 rpm for the first two cycles and 575 rpm for all subsequent cycles to allow maximum action of the radon transfer system moisture trap for the first two cycles.

When atmospheric pressure was reached, the newly filled counting bottle was closed, removed from the radon transfer system, and set aside to age prior to counting. The exhausted radon charcoal trap was cleaned by filling it with helium then evacuating it to ≤ 0.200 mm. Hg three or more times. Then the radon charcoal trap was filled with helium to 500 mm. Hg, closed, removed from the radon transfer system, and stored for subsequent use.

The heater used was a "Hoskins" combustion tube furnace liner and heating element. These were covered with a 1-inch layer of asbestos paper, a layer of aluminum foil, then another 1-inch layer of asbestos paper. The completed heater was coated with sodium silicate solution to produce a hard outer shell.

The heater was maintained at an operational temperature of 450° to 500° C. by a control unit that regulated the per cent of time during which line voltage was furnished to the heating element. The temperature was monitored with a pyrometer.

Pressure in the transfer system was monitored with a thermocouple vacuum gauge.

The dead air space of 27.54 cc. introduced some ambient radon into the system. However, after evacuation of 0.200 mm. Hg, this dead space contains only 7.85×10^{-3} cc. of air and would introduce about 2.166×10^{-3} fCi of radon. This was considered to be a negligible amount and was ignored in the final calculations.

The dead air space on the counting bottle side of the peristaltic pump introduced some error since not all of the radon and helium mixture pumped from the radon trap would enter the counting bottle. About 2 cc. of the 8.40 cc. of dead space on the counting bottle side of the peristaltic pump was not on a line to the counting bottle. Lucas (1) calculates an absolute loss of approximately

5% of the radon due to dead air space and mixing and other loss in the transfer system. This problem is not of any concern in the use of the transfer system described here, since the process of calibration of counting bottles compensates for and includes these losses in the transfer system.

The pure gum rubber tubing used in the peristaltic pump and the helium used as a carrier gas can introduce radon into the system. To minimize this error, two procedures were followed. First, all helium was aged before use. The tank of helium was stored for at least 40 days before being used. This allowed all radon trapped in the helium during processing to decay through ten half lives, reducing its concentration to about 0.5% of the original value. The final radon content of the helium was then dependent on the release of radon from the natural radium-226 in the walls of the cylinder. The slow release of radon from the cylinder walls was considered to have maintained the radon content of the helium at a relatively constant level.

Second, all rubber tubings were pre-cut into standard lengths to fit the peristaltic pump. These lengths of rubber tubing were stored for at least 40 days in closed containers to allow all absorbed radon to decay to a negligible amount. After storage the lengths of rubber tubing contained only the radon released by the natural radium-226 in the rubber. The lengths of rubber tubing in the peristaltic pump were changed each day before the radon transfer system was used to minimize adsorption of ambient radon-222. The error introduced by the helium and rubber tubing was determined to be 2.2 fCi of radon for a complete transfer cycle.

The inability of the vacuum pump to evacuate the radon trap to below 0.200 mm. Hg in the pre-transfer evacuation procedure required a modification of transfer sequence in many cases. In some assays, in order to get six flushes of the radon trap and at the same time fulfill the PV (pressure-volume)

restrictions, flush aliquots were reduced to 12 or 13 cc. of helium. Checks made by placing a second counting bottle on the transfer system and flushing the exhausted radon charcoal trap again after the first counting bottle had been filled showed that 99.5% or more of the radon was transferred even with the reduced volume of the flushing aliquot. Also, the volume of water vapor or carbon dioxide retained on the charcoal in the radon trap was noted to be increased because a pressure of 0.200 mm. Hg rather than 0.020 mm. Hg was reached. However, the volume transferred to the counting bottle never exceeded 15 cm. of pressure as indicated by the mercury manometer. Lucas (1) reports that over 20 cm. of pressure must be transferred before there is any noticeable effect on the counting plateau. Therefore, the amount of water vapor or carbon dioxide transferred was not considered to have a significant effect in this system as long as the volume transferred did not exceed 15 cm. of pressure.

APPENDIX F

MATHEMATICAL TREATMENT OF DATA

The data obtained from analyses of individual samples was in the form of counts per unit time. This raw data needed to be converted to a more useful coordinate system.

The transformation of raw data to the coordinates of grams ^{226}Ra per aliquot of sample was done using a computer program developed by Lucas (1). The program took into consideration the laws of decay for radioisotopes in transient or secular equilibrium. The transformation was based on the following:

In a genetic sequence of radioisotopes that is in secular equilibrium:

1. Each radioisotope is present in a constant but different amount,
2. The numbers of atoms of the various radioisotopes in the sequence are proportional to their half-lives,
3. Every radioisotope is decaying and, except for the first member of the sequence, being produced at the same rate, $\dots N_1 \propto 1$.

Although the actual working fluid of the procedure was the gas radon, knowledge of the amount of radon present at equilibrium was sufficient to determine the amount of radium present. Since the radon was in secular equilibrium with its parent radium, radium-226 present was considered to be identical in value to radon-222 present. This introduced a small error, since 1 gram of radium equals 0.9878 curies of radium (9). Therefore, in secular equilibrium inasmuch as the radium was the limiting factor, 1 gram of radium-226 was in equilibrium with 0.9878 curies of radon-222.

The final value of femtocuries of radon-222 ($\approx$ femtocuries of radium-226) per aliquot was found by dividing the total content of radon-222 ($\approx$ radium-226) in femtocuries in the sample by the size of the sample in the desired units, i.e., grams, liters, etc. This value in femtocuries of radon-222 was considered identical to the value in femtograms of radium-226 and the 1.0122 conversion factor was not used to convert femtocuries of radon-222 to femtograms of radium-226.

The "J" factor adjustment proposed by Lucas and Woodward (10) was needed because the alpha particles emitted by radon daughters and counted during the equilibrium period in the counting bottles were not randomly distributed. In a Poisson distribution, each event must be independent of all others. The "J" factor corrected for the deviation from Poisson statistics.

Since the appearance of the radon daughters was dependent on the decay of the preceding radioisotopes and since they had a short half-life relative to the duration of the counting period, the distribution of alpha counts from the daughters did not satisfy all conditions for validity of a Poisson distribution (11,12).

The equations for computing the radon-222 content of the sample and the error in the determination of the radon-222 content (expressed as the 95% confidence limits) are shown in Figure 10, where:

RN = quantity of radon-222 in the sample measured in picocuries.

ERN = error in radon-222 determinations in picocuries.

$RC = RN^2$.

CK = confidence interval.

$LJ(t, t, Plan)$ = J factor.

R = fraction of equilibrium attained at time of count.

TC = total count.

$$RN = \frac{\left[\frac{\left(\frac{TC/t - BKGND}{E \cdot R \cdot EPN1} - C_2 + C_2 \cdot EPN2 \right)}{EPN3} - C_3 \right]}{1.0 - EPN4}$$

$$ERN = RN \cdot CK \sqrt{\frac{\left[\frac{\left(XJ(t_s, t, Plam) \left(\frac{TC}{t^2} - \frac{BKGND}{t} \right) + \frac{BKGND}{t} + E4^2 \right)}{E^2 \cdot R^2 C} \right] + E1}{R^2 \cdot EPN1^2} + \frac{EC2(1-EPN2)^2}{R^2 C} + \frac{EC3}{R^2 C}}{(1-EPN4)^2}}$$

Fig. 10.—Equations for calculation of radon content and error of this calculation.

t = duration of count in hours.

BKND = background of counting bottle in counts per hour.

E_b = variance in background in counts per hour.

E = fractional efficiency of counting bottle in counts per minute per picocurie.

E_L = variance of calibration of counting bottle in counts per minute per picocurie.

C_2 = radium-226 content of charcoal in the radon trap in picocuries.

EC_2 = variance of C_2 in counts per hour.

C_3 = amount of radon-222 entering counting bottle from helium and through the rubber tubing in the transfer system in picocurie.

EC_3 = variance in C_3 in counts per hour.

$EPN_1 = e^{-\lambda t_1}$ = decay of radon in counting bottle.

$EPN_2 = e^{-\lambda t_2}$ = growth and trapping of radon from the charcoal of the radon trap.

$EPN_3 = e^{-\lambda t_3}$ = decay of radon in radon trap.

$EPN_4 = e^{-\lambda t_4}$ = growth of radon in generator flask.

t_1 = duration of time from the time the first of the radon was transferred into the counting bottle to half way through the period of counting in hours.

t_2 = duration of time from the time the radon trap was heated and flushed with helium until the time the first of the radon was transferred into the counting bottle in hours.

t_3 = duration of time from the time the last of the radon is flushed into the radon trap until the time the first of the radon is transferred into the counting bottle in hours.

t_4 = duration of time from the time of the end of the flush of the sample generator on the last run to the time of the end of the flush of the sample generator on the current run in hours.

Determination of Weighted Mean for a Group of
Runs of the Same Sample

After radium-226 determinations had been obtained for a sample by making a number of sequential analyses for that sample, the mean value for these individual determinations was derived. The mean value obtained was then used as the radium content of the sample.

Since the values of individual determinations were influenced by (1) the fraction of equilibrium obtained in each phase of the analysis, (2) by the duration and absolute value of the count, and (3) by counting statistics, the mean value was expressed as a weighted mean. By using the method of determining a weighted mean suggested by Brownlee (13), the statistical variations introduced in each analysis could be minimized.

A computer program was designed so that the output from the program for determining radon-222 content, as proposed by Lucas, could be combined directly with the program for determining the weighted mean of a group of assays of the same sample.

The equation used for computing the weighted means is shown in Figure 11, where

$\bar{X}_w$ = weighted mean,

V_{X_1} = variance of X_1 .

S_{X_w} = standard deviation of $\bar{X}_w$.

X_1 = the value of the 1 th observation of X .

$$\bar{X}_\omega = \frac{\sum_{i=1}^i \omega_i x_i}{\sum_{i=1}^i \omega_i}, \text{ where } \omega_i = \frac{1}{V_{x_i}}$$

$$S_{\bar{X}_\omega} = \sqrt{\frac{1}{\sum_{i=1}^i \omega_i}}$$

Fig. 11.—Equation for calculating weighted means.

APPENDIX G

SPECIAL ANALYTICAL PROBLEMS

Inaccurate analyses of a sample would often be observed and the direction of the error could be predicted before the sample was counted. Assays which produced high radon-222 values occurred less frequently than those which produced low values.

High values were usually encountered for one of three reasons.

First, early in our studies the contamination of a radon collection trap by residual radon from a prior run occasionally produced high values. If the prior analysis was from a source of extremely high activity, of say 100 pCi (100,000 fCi) or higher, then there would be residual radon-222 of 0.01 remaining in the radon collection trap or about 10 fCi of radon-222. When the next sample run using this trap was a low activity sample, of the order of 500 fCi or less, the radon-222 residual from the prior sample could then produce an apparently greater radon-222 content for the low activity sample of 2% or more. When this source of error was eventually identified, all radon traps were thereafter numbered and a record of their use maintained. From that time on radon collection traps were only used for samples of a similar magnitude of activity. It was also found that if traps were flushed six times with helium while being held at 450° to 500° C. and then allowed to rest for 4 days, they could again be used with the low activity samples. The 4-day rest allowed 50% decay of the radon-222 remaining after the heated flush. This technique eventually alleviated the error problem created by the residual contamination with radon-222.

A second source of high values was the drift in high voltage to a higher voltage. This was caused by upward fluctuations in line voltage. A frequent check of the operating point with the secondary standard and the readjustment to optimal operating point (see Appendix B) was required to control this problem.

A third reason for high values was the light excitation of the zinc sulfide scintillator or the photocathode of the photomultiplier. Whenever the radon counting chamber was exposed to light, as it was when it was being placed on the photomultiplier tube for counting, the light caused photoexcitation of the zinc sulfide scintillator. The excitation was manifest as a delayed phosphorescence with a very short half life. For about 3 minutes after the radon counting chamber was placed in a counter, the delayed phosphorescence would cause an extremely high count rate. The count rate would then rapidly drop to background or to the level of the sample being counted. To control this problem all samples were kept within the darkened counter shield for at least 5 minutes before the timed count was initiated.

Photoexcitation of the photocathode of the photomultiplier would also occur if excessive light were allowed to fall upon the photocathode while counting bottles were being changed. This photoactivation would occur even though the high voltage to the photomultiplier tube had been turned off. Shielding the photocathode from direct light and restricting the duration of its exposure to light was the only method to control this problem.

Most unexplained high radon values could be ascribed to this since it was very difficult to prove that photoexcitation had not persisted beyond the 5 minutes holding time before the initiating of count. A high count rate was observed to persist for 2 hours in one case. The count was stopped, the scaler reset, and a new count initiated. The new count proceeded at the

expected rate. The reason for this persistent high count rate was never determined, but it was believed to be associated with the photoexcitation phenomena.

Assays producing low radon-222 values were the most frequent type of error. One cause of low values was easily diagnosed. The contamination of the radon collection trap with a gas or substance that was volatile at 450° or lower would interfere with transfer of radon-222 from the trap to the radon counting bottle. Since the radon counting bottle had a volume of about 90 cc., any contaminating gas which would fill this volume would prevent quantitative transfer of the radon with the helium flushes (see Appendix E).

This was observed with CO_2 and H_2S , as well as with a silicone anti-foam agent used in the generator flask, which were trapped by the charcoal. This problem was controlled by using special traps between the generator flask and the radon collection trap to remove the contaminating materials from the radon-222 laden air.

Air leaks at joint and stopcocks in either the radon collection system or the radon transfer system would also produce low values. If a number of runs were suspected to be low, then both systems were evacuated and tested for pinhole leaks with a high frequency vacuum tester (Tesla coil). It was usually easy to detect an imperfect seal at a stopcock (greased with Apiezon-N) since it would appear as if minute refractive striations had suddenly developed in the seal, though some leaks could be detected only with a high frequency vacuum tester. On these occasions radon counting bottles or radon traps could not be evacuated to below 500 microns of vacuum. The checks that were made with the high frequency vacuum tester showed leaks in the seal at the stopcock even though the striations characteristic of an imperfect seal were not observed.

There was one site of leakage in the radon transfer system which would develop after prolonged operation of the system. The heater used to activate the charcoal and drive the radon out of the charcoal in the radon collection traps occasionally warmed the nearest ball and socket joint (see Figure 9) to such an extent that the viscosity of the Apiezon-N grease changed. Tests with a high frequency vacuum tester would show air flowing through the partially liquefied Apiezon-N in the joint. The additional volume of gas which leaked into the system resulted in an incomplete transfer of the radon from the radon collection trap to the radon counting bottle.

Suddenly changing the radon collection trap from its dry ice-alcohol bath at -70° C. to the heater in the radon transfer system at 150° to 500° C. produced stains which occasionally caused cracks to appear in the wall of the radon collection trap. Usually these cracks could be easily found by the visual inspection of the radon traps just prior to their use, but occasionally one would be so small that it would be missed.

Leaks at the joints of the radon collection system occurred infrequently. Early in the study it had been noted that at pressures exceeding 5 psi, as indicated by the pressure regulator on the cylinder of dry air, joints were liable to rupture. Therefore, the internal radon collection system pressures were not allowed to exceed 4 psi during the operation of the system. Even so, occasionally abrupt decreases in flow rate to near zero flow were observed. Inspection of the system immediately after one of these events would usually reveal a ruptured seal at some joint.

Another, rather sporadic, source of low values was due to the interruption of the operation of the electronic counting systems. In the first scaler used, whenever line voltage was interrupted the equipment would, of course, stop, and when line voltage returned the equipment would resume its

counting. When the scaler was turned off by interruption of line power, the total count accumulated until that time was lost, since the scaler had no memory. When the scaler was started again, it would show an accumulated total count on the decade display, but that count would be a series of random digits caused by electronic noise as the scaler started and warmed up. The scaler would then proceed to add counts to that display of random digits. This could produce either a high count or a low count, but never the correct count. Two scalers acquired later would not resume counting if the power was interrupted. They were then used as line voltage monitors for the older scaler.

On the basis of the possible sources of variations in counts, it was decided that identification of radon collection trap, counting bottle, and counter used should be included in the information on each analysis of a sample. By comparing this information, it was possible to determine if low or high values occurred consistently in a specific trap or counting bottle or on a specific scaler. Whenever the distribution of high and low values was not at random but occurred consistently in a specific component, then that component was subjected to tests to determine the source of the error.

APPENDIX H

SPECIAL MATERIALS

Linde Molecular Sieve, Type 5A, 1/16" size

Linde Company Division
Union Carbide Corporation
230 North Michigan Avenue
Chicago, Illinois 60601

Vicor Trays and Pyroceram Dishes

Corning Glass Works
Corning, New York

ZnS (Ag) Scintillator (Hoecon Cat. No. 2205)

United States Radium Corporation
535 Pearl Street
New York 7, New York

Kovar Cups (Cat. No. 942003) No. Kc-5A

Carborundum Company
Refractories Division
3335 West 47th Street
Chicago, Illinois 60632

R-313 Adhesive and R-321 Adhesive

Carl H. Biggs Company
1547 14th Street
Santa Monica, California

DEK-adhese

Mr. Donald Tulloch, Jr.
Box 17
Gadsds Ford, Pennsylvania

2" x 1/8" Quartz disks ground and polished

General Electric
Lamp Glass Department
Euclid Avenue and Campbell Road
Willoughby, Ohio

Fused Quartz Trays

Amersil Quartz, Division of
Englehard Industries
685 Ramsey Avenue
Hillside 5, New Jersey

Aprezon N. grease

Aprezon Products Limited
England
(distributed by Shell Oil Company)

Air Brush, Model C

Thayer and Chandler
910 West Van Buren Street
Chicago, Illinois

REFERENCES

1. Henry F. Lucas, Jr., Personal communication.
2. Henry F. Lucas, Jr., Correlation of the Natural Radioactivity of the Human Body to That of Its Environment: Uptake and Retention of Ra^{226} from Food and Water, Argonne National Laboratory, Radiological Physics Division Semi-annual Report, ANL-6297, pp. 55-66 (July-December, 1960).
3. H. Moses, A. F. Stehney, and H. F. Lucas, Jr., The Effect of Meteorological Variables Upon the Vertical and Temporal Distributions of Atmospheric Radon. *J. Geophys. Res.* 65: 1223-1238, No. 4 (1960).
4. W. A. Jennings, and S. Russ. Radon: Its Technique and Use. Published for the Middlesex Hospital Press by John Murray, London, 1948.
5. J. H. Lawrence, W. F. Loomis, C. A. Tobias, and F. H. Turpin, Preliminary Observations on the Narcotic Effect of Xenon with a Review of Values for Solubilities of Gases in Water and Oils. *J. of Physiol.* 105: 197-204 (1946).
6. Henry F. Lucas, Jr., Improved Low-Level Alpha Scintillation Counter for Radon. *The Rev. of Scient. Instru.* 28: 680-683, No. 9 (1957).
7. Henry F. Lucas, Jr., Radon Scintillation Counter, ANL-5596, pp. 15-18, January-June, 1956.
8. R. E. Rowland, J. Jowsey, and J. H. Marshall, Radon Escape from Bone Mineral. *Radiation Res.* 8: 298-306 (1958).
9. G. R. Martin, and D. G. Tuck, The Specific Activity of Radium. *International J. of Appl. Radiation and Isotopes* 5: 141-145 (1959).

10. H. F. Lucas, Jr., and D. A. Woodward, Effect of Long Decay Chains on the Counting Statistics in the Analysis of Radium-224 and Radon-222, Argonne National Laboratory, Radiological Physics Division, ANL-6646, pp. 78-91 (January-June, 1962).
11. Norman I. Adams, Jr., An Application of Probabilities to the Counting of Alpha Particles. Physical Review 44: 651-653 (1933).
12. Robley D. Evans, The Atomic Nucleus. McGraw-Hill Book Company, Inc., New York, 1955, p. 818.
13. K. A. Brownlee, Statistical Theory and Methodology in Science and Engineering. John Wiley and Sons, Inc., New York, 1960, p. 73.

APPENDIX I

DATA DETERMINED OR CALCULATED FOR EACH INDIVIDUAL ANIMAL IN THE STUDY

During this study determinations were made for the ^{226}Ra , calcium, and phosphorous content; wet weight; ashed weight and per cent ash for a number of samples from each animal. In the body of the text, resume values were presented as the mean $\pm$ 95% confidence limits for each group analyzed. In this appendix, the data are determined for each animal and then the various ratios calculated from these data are presented for comparison. All values enclosed in a box have been considered "outliers" and not used in calculation of the mean and 95% confidence limits. Criteria suggested by Dean and Dixon (14) have been used in all cases whenever a determination was thought to be a candidate to drop from the calculations.

The main and unique feature of the data is the wide variation between similar samples from different animals. This individual "bio-variation" was several times greater than the error for any given determination even though we tried to limit genetic variation as much as possible.

The wet weight of all tissues was determined before they were ashed and prepared for assay (see Appendix E). The ashed weight was used as the reference for all calculations. Although the arguments presented by Holtzmann (15) for relating bone-seeking minerals to ash weight were for bone only, they can be applied to other tissues. The difficulties we encountered in obtaining wet weights, dry weights, fat free weights, etc., which truly represent the weight of the tissue of interest, suggest that ashed weight

might profitably be used for most, if not all, types of determinations when tissue weight is of prime interest.

Ashing samples also has the advantage that the element to be determined can be very dilute, since ashing concentrates mineral elements. The rather dilute ^{226}Ra we encountered was concentrated; a constant weight was easily obtained, processing was facilitated, and fortunately the loss of all elements during ashing was minimal.

Tables 20 to 34 present the ^{226}Ra , calcium and phosphorous contents per gram of ash, determined for each animal. They also give the radium-calcium, radium-phosphorous, calcium-phosphorous and OR's (observed ratios) calculated for each animal as calculated from the chemical determinations made for each of the individual elements. Table 35 gives the wet weight, ashed weight and per cent ash determined for all soft tissues analyzed. Table 36 gives the same information for sow's milk. Table 37 lists the ^{226}Ra , calcium and phosphorous contents of bone samples from eight leukemic animals, obtained from abattoirs.

Since the ^{226}Ra content of most tissues had been determined, it was possible for us to estimate the total body burden of ^{226}Ra for the sows and barrows. The skeletal weights, but not all soft tissue weights, were known for all animals. The skeleton contributed the major portion of the burden in both cases, so we believe that the uncertainty introduced by estimating ^{226}Ra content of unknown soft tissues was probably less than 2% in the sows and 5% in the barrows. Our calculated total body burdens are given in Table 38.

Table 39 is a comparison of the per cent ash, mgm. of calcium and phosphorous per gram of ash and calcium to phosphorous ratio as determined in this study with similar data available in the literature (16,17). In most cases there is good agreement between the values obtained in this study and the published values. Differences that we noted may be due to any number of

TABLE 20
 INDIVIDUAL DISTRIBUTION OF ²²⁶Ra PER GRAM OF ASH IN BONES NORMALIZED^a
 AGAINST THE SKELETAL AVERAGE

Skeletal Bone	Sows								Mean $\pm$ 95% Confidence Limits
	No. 1	No. 2	No. 3	No. 4	No. 5	No. 6	No. 8	No. 9	
Skull	1.004	0.981	0.993	1.059	0.943	0.934	1.023	0.982	0.987 $\pm$ 0.036
Mandible	0.984	0.902	1.060	0.980	0.985	1.010	0.984	0.896	0.984 $\pm$ 0.048
Scapula	0.925	1.008	0.993	1.000	0.901	0.884	0.927	0.954	0.940 $\pm$ 0.036
Humerus	1.181	1.088	1.148	1.039	1.069	1.111	1.004	1.010	1.078 $\pm$ 0.051
Radius and ulna	1.122	0.955	1.038	1.020	0.985	1.035	0.965	1.098	1.028 $\pm$ 0.048
Carpals	1.220	1.061	1.170	1.078	1.090	1.111	1.042	1.127	1.100 $\pm$ 0.052
Spine	0.925	1.114	1.015	1.000	1.111	1.162	1.062	1.098	1.080 $\pm$ 0.069
Ribs	0.925	1.034	0.949	0.961	0.985	0.909	0.907	0.867	0.937 $\pm$ 0.018
Sternum	0.768	0.981	0.773	0.863	0.901	0.858	0.984	0.867	0.865 $\pm$ 0.063
Pelvis	1.024	0.981	0.993	0.980	0.985	0.960	1.023	1.010	0.989 $\pm$ 0.023
Femur	1.122	1.088	1.060	0.941	1.006	1.086	1.042	1.156	1.073 $\pm$ 0.062
Tibia and fibula	1.063	0.955	1.015	1.039	0.964	0.909	0.946	1.012	0.988 $\pm$ 0.045
Tarsals	1.201	1.061	1.192	0.961	1.090	1.111	0.984	1.098	1.094 $\pm$ 0.070
Tail	1.083	1.220	1.192	1.196	1.111	1.060	1.255	1.069	1.152 $\pm$ 0.056

TABLE 20--Continued

Skeleton	Normal No. III	Leukemic No. II	U of I No. XII	U of I No. IV	U of I No. V	U of I No. VI	Mean $\pm$ 95% Confidence Limits
Bone							
Skull			0.906		0.791	0.980	0.906 $\pm$ 0.2
Mandible			0.789				
Scapula	1.002	1.074	0.935	0.934	0.845	1.051	0.966 $\pm$ 0.09
Humerus	1.037	0.971	1.169	1.040	1.080	1.074	1.057 $\pm$ 0.08
Radius and ulna	1.000	0.962	1.169	1.045	1.114	1.064	1.054 $\pm$ 0.08
Spine	1.000	1.022	1.081	0.883	0.946	1.025	1.011 $\pm$ 0.06
Ribs	0.906	0.826	0.964	0.959	0.852	0.894	0.900 $\pm$ 0.06
Pelvis	0.974	0.929	0.877	0.898	0.959	0.968	0.944 $\pm$ 0.02
Femur	1.040	1.074	1.169	1.081	1.147	1.009	1.085 $\pm$ 0.05
Tibia and fibula	1.060	1.111	1.169	1.091	1.167	1.056	1.101 $\pm$ 0.04
Carpals, tarsals	1.015	1.053	1.198	1.203	1.167	0.856	1.110 $\pm$ 0.24

Normalized value = $\frac{^{226}\text{Ra content of bone}}{^{226}\text{Ra skeletal average}}$

TABLE 21

RADIUM CONTENT OF ^{226}Ra fCi OF ^{226}Ra PER GRAM OF ASH FOR ALL SAMPLES FROM SOWS
FROM FRANKFORT, ILLINOIS

^{226}Ra fCi gm. ash	Sows							
	No. 1	No. 2	No. 3	No. 4	No. 5	No. 6	No. 8	No. 9
Skeleton	50.8 $\pm$ 0.4	37.7 $\pm$ 0.4	45.3 $\pm$ 0.4	51.0 $\pm$ 0.5	47.7 $\pm$ 0.5	47.7 $\pm$ 0.5	39.6 $\pm$ 0.4	34.6 $\pm$ 0.4
Skull	51 $\pm$ 2	37 $\pm$ 1	45 $\pm$ 2	54 $\pm$ 2	45 $\pm$ 2	37 $\pm$ 1	53 $\pm$ 1	34 $\pm$ 1
Mandible	50 $\pm$ 2	34 $\pm$ 1	48 $\pm$ 2	50 $\pm$ 2	47 $\pm$ 2	40 $\pm$ 2	51 $\pm$ 1	31 $\pm$ 1
Scapula	47 $\pm$ 2	38 $\pm$ 1	45 $\pm$ 2	51 $\pm$ 2	43 $\pm$ 1	35 $\pm$ 1	48 $\pm$ 2	33 $\pm$ 2
Humerus	60 $\pm$ 3	41 $\pm$ 2	52 $\pm$ 2	54 $\pm$ 2	51 $\pm$ 2	44 $\pm$ 2	52 $\pm$ 2	36 $\pm$ 2
Radius and ulna	57 $\pm$ 2	36 $\pm$ 2	47 $\pm$ 2	52 $\pm$ 2	47 $\pm$ 2	41 $\pm$ 2	50 $\pm$ 2	38 $\pm$ 2
Manus	62 $\pm$ 2	40 $\pm$ 2	53 $\pm$ 2	55 $\pm$ 2	52 $\pm$ 2	44 $\pm$ 2	54 $\pm$ 2	39 $\pm$ 1
Spine	47 $\pm$ 2	42 $\pm$ 1	46 $\pm$ 2	51 $\pm$ 2	53 $\pm$ 2	46 $\pm$ 2	55 $\pm$ 2	38 $\pm$ 1
Ribs	47 $\pm$ 2	39 $\pm$ 2	43 $\pm$ 1	49 $\pm$ 2	47 $\pm$ 2	36 $\pm$ 1	47 $\pm$ 2	30 $\pm$ 1
Sternum	39 $\pm$ 1	37 $\pm$ 2	35 $\pm$ 1	44 $\pm$ 2	43 $\pm$ 2	34 $\pm$ 2	51 $\pm$ 1	30 $\pm$ 1
Pelvis	52 $\pm$ 2	37 $\pm$ 1	45 $\pm$ 2	50 $\pm$ 2	47 $\pm$ 2	38 $\pm$ 1	53 $\pm$ 1	36 $\pm$ 2
Femur	57 $\pm$ 2	41 $\pm$ 2	48 $\pm$ 2	48 $\pm$ 2	48 $\pm$ 2	43 $\pm$ 2	54 $\pm$ 2	40 $\pm$ 2
Tibia and fibula	54 $\pm$ 2	36 $\pm$ 1	46 $\pm$ 2	53 $\pm$ 2	46 $\pm$ 2	36 $\pm$ 2	49 $\pm$ 2	35 $\pm$ 1
Pes	61 $\pm$ 2	40 $\pm$ 1	54 $\pm$ 2	49 $\pm$ 2	52 $\pm$ 2	44 $\pm$ 1	51 $\pm$ 2	38 $\pm$ 2
Tail	54 $\pm$ 2	45 $\pm$ 2	54 $\pm$ 2	62 $\pm$ 2	53 $\pm$ 2	42 $\pm$ 2	66 $\pm$ 2	37 $\pm$ 1
Long bones	52.2 $\pm$ 0.5	37.3 $\pm$ 0.4	46.8 $\pm$ 0.5	51.4 $\pm$ 0.6	47.1 $\pm$ 0.6	39.3 $\pm$ 0.5	52.0 $\pm$ 0.5	35.1 $\pm$ 0.4

TABLE 21--Continued

^{226}Ra fci gm. ash	Sows							
	No. 1	No. 2	No. 3	No. 4	No. 5	No. 6	No. 8	No. 9
Neonate	10.4 $\pm$ 0.6	7.4 $\pm$ 0.3	6.5 $\pm$ 0.4	6.4 $\pm$ 0.4	10.2 $\pm$ 0.5	12.8 $\pm$ 0.6	7.7 $\pm$ 0.5	15.5 $\pm$ 0.7
2 weeks		27.8 $\pm$ 0.6			21.7 $\pm$ 0.8	15.1 $\pm$ 0.6		
4 weeks	20 $\pm$ 0.8	26 $\pm$ 1	58 $\pm$ 1	50 $\pm$ 1	33 $\pm$ 1	21.6 $\pm$ 0.8	57 $\pm$ 2	39 $\pm$ 1
Milk		7.7 $\pm$ 0.7			23 $\pm$ 6	14 $\pm$ 1	11 $\pm$ 2	10 $\pm$ 1
Blood	6.2 $\pm$ 0.4	7.2 $\pm$ 0.3	8.2 $\pm$ 0.4	4.8 $\pm$ 0.3	6.8 $\pm$ 0.4	6.6 $\pm$ 0.4	8.8 $\pm$ 0.5	10.9 $\pm$ 0.5
Muscle	1.9 $\pm$ 0.1	3.8 $\pm$ 0.1	1.1 $\pm$ 0.1	4.1 $\pm$ 0.2	4.0 $\pm$ 0.2	4.1 $\pm$ 0.2	3.6 $\pm$ 0.2	9.0 $\pm$ 0.3
Skin and fat	27 $\pm$ 1	9.9 $\pm$ 0.4 ^a	18 $\pm$ 1	17 $\pm$ 1	3.8 $\pm$ 0.3	7.2 $\pm$ 0.4	6.9 $\pm$ 0.5	17 $\pm$ 1
Lung	32 $\pm$ 2	30 $\pm$ 2	22 $\pm$ 1	31 $\pm$ 2	31 $\pm$ 2	21 $\pm$ 2	21 $\pm$ 2	20 $\pm$ 1
Heart	2.4 $\pm$ 0.9	2.5 $\pm$ 0.8	3.8 $\pm$ 0.8	2.1 $\pm$ 0.6	1.9 $\pm$ 0.9	4.9 $\pm$ 1.1	1.3 $\pm$ 1.0	3.0 $\pm$ 0.9
Kidney	4.7 $\pm$ 1.3	17 $\pm$ 2	20 $\pm$ 2	4.7 $\pm$ 0.9	5.6 $\pm$ 1.5	3.7 $\pm$ 2.6	5.3 $\pm$ 1.9	5.9 $\pm$ 2.5

Frankfort feed 27 $\pm$ 1
 Frankfort supplement 1750 $\pm$ 64
 Frankfort water^a 400 $\pm$ 13

Goodenow feed 346 $\pm$ 15
 Goodenow water^a 48 $\pm$ 2

^afci ^{226}Ra /liter.

TABLE 22

CALCIUM CONTENT IN MCM. OF CALCIUM PER GRAM OF ASH FOR ALL
SAMPLES FROM SOWS FROM FRANKFORT, ILLINOIS

MCM. Ca gm. Ash	Sows							
	No. 1	No. 2	No. 3	No. 4	No. 5	No. 6	No. 8	No. 9
Skull	385.2	376.9	382.7	385.4	388.4	393.0	383.3	383.2
Mandible	392.4	390.0	387.2	390.1	393.4	390.5	382.7	385.9
Scapula	391.3	390.3	386.6	395.6	393.0	384.0	381.0	387.1
Humerus	390.7	399.9	394.4	389.7	389.2	394.2	390.1	391.7
Radius and ulna	396.6	386.0	390.3	384.4	386.9	385.5	385.2	390.8
Manus	384.5	381.0	397.9	388.6	385.8	384.0	388.1	386.6
Spine	384.4	400.2	393.6	387.1	398.5	383.5	382.7	388.4
Ribs	395.1	388.2	389.5	387.9	392.1	387.6	384.5	388.4
Sternum	373.1	378.3	368.1	381.1	377.1	375.2	364.9	369.3
Pelvis	387.6	387.2	391.9	383.8	387.7	387.6	384.7	384.9
Femur	387.4	386.5	381.4	386.5	383.3	390.6	381.0	388.4
Tibia and fibula	387.6	387.2	394.8	385.2	389.6	388.1	382.3	387.2
Pes	389.4	378.8	401.6	383.2	390.3	390.5	382.2	387.2
Tail	379.0	368.1	382.9	380.4	384.0	382.9	384.7	386.1
Neonate	340.8	346.9	322.1	332.4	334.3	333.2	347.9	355.3
2 weeks		355.5			325.5	323.4		
4 weeks	358.7	365.1	372.2	344.4	351.4	341.4	341.8	354.4
Milk		326.2			333.2	289.5	311.4	237.4
Blood	7.72	6.83	8.41	7.70	6.65	7.01	8.49	11.45
Muscle	20.92	10.79	12.91	13.68	12.36	15.18	12.98	16.97
Skin and fat	42.20	40.63	38.41	34.90	18.34	15.49	32.61	34.20
Lung	61.12	40.08	57.75	66.53	31.97	51.81	37.30	56.17
Liver	16.25	26.89	66.26	19.96	26.08	19.35	14.28	22.81
Heart	48.22	39.02	62.25	63.89	44.41	47.97	38.45	33.29
Kidney	24.62	145.70	167.11	21.04	38.05	21.11	25.93	72.22

Frankfort feed 15.8
Frankfort supplement 268.6
Frankfort water^a 104.2

Goodenow feed 223.0
Goodenow water^a 76.8

^amgm. Ca/liter.

TABLE 23

PHOSPHOROUS CONTENT IN MCM. OF PHOSPHOROUS PER GRAM OF ASH FOR ALL
SAMPLES FROM SOWS FROM FRANKFORT, ILLINOIS

mgm. P gm. Ash	Sows							
	No. 1	No. 2	No. 3	No. 4	No. 5	No. 6	No. 8	No. 9
Skull	175.2	173.1	175.5	177.7	175.2	179.0	176.3	177.7
Mandible	175.5	180.0	176.5	176.3	176.5	180.0	175.7	180.0
Scapula	183.2	173.9	179.0	176.6	179.5	177.6	176.3	179.6
Humerus	182.2	181.6	182.7	180.6	182.7	184.0	181.1	179.5
Radius and ulna	180.0	168.6	177.9	181.6	180.0	182.3	182.3	181.6
Manus	176.0	173.9	179.0	175.0	177.1	179.0	180.3	181.1
Spine	180.0	181.1	182.1	181.0	178.4	181.0	181.0	182.3
Ribs	179.9	178.9	177.9	179.9	178.4	179.5	169.7	181.6
Sternum	174.9	177.6	179.6	174.8	179.6	179.6	175.5	178.0
Pelvis	177.3	179.0	178.4	181.0	181.1	181.0	179.7	181.7
Femur	177.9	179.0	179.5	178.7	183.7	182.3	181.0	183.7
Tibia and fibula	177.9	170.3	181.1	178.3	180.0	181.7	180.3	181.0
Pes	176.0	171.8	177.9	176.0	177.3	179.7	177.3	178.3
Tail	177.3	181.0	181.7	178.4	181.2	181.5	179.8	178.9
Neonate	189.6	184.6	187.6	185.0	187.8	190.0	190.3	192.7
2 weeks		188.5			192.7	191.1		
4 weeks	189.1	186.5	188.6	189.2	188.5	193.8	191.8	192.2
Milk		159.7			175.8	139.2	154.2	174.6
Blood	39.79	41.69	38.98	41.32	43.38	42.00	40.39	35.30
Muscle	195.72	192.01	191.75	200.84	179.79	175.80	185.63	169.21
Skin and fat	107.87	105.71	111.33	121.48	83.51	62.20	111.87	93.61
Lung	182.57	171.91	166.70	168.46	173.19	152.88	175.96	166.52
Liver	248.38	253.39	212.22	253.48	250.52	234.28	235.25	241.86
Heart	171.57	173.44	170.44	142.15	181.48	179.66	192.91	177.38
Kidney	199.44	202.20	193.73	164.17	173.14	197.67	179.43	236.66

Frankfort feed 178.2
Frankfort supplement 113.7
Frankfort water^a 0.6

Goodenow feed 208.0
Goodenow water^a 0.45

^amgm. P/liter.

TABLE 24

RADIUM TO CALCIUM RATIOS AS ^{226}Ra PER GRAM OF CALCIUM FOR ALL SAMPLES
FROM SOWS FROM FRANKFORT, ILLINOIS

$\frac{\text{FCI } ^{226}\text{Ra}}{\text{gm. Ca}}$	Sows								Mean $\pm$ 95% Confidence Limits
	No. 1	No. 2	No. 3	No. 4	No. 5	No. 6	No. 8	No. 9	
Skull	132.40	97.65	117.58	140.11	115.86	94.15	138.27	88.73	116.7 $\pm$ 14.9
Mandible	127.10	87.18	123.97	126.17	119.47	102.43	133.26	80.33	121.7 $\pm$ 15.4
Scapula	120.11	99.92	116.40	128.92	109.11	91.14	125.98	85.25	112.9 $\pm$ 12.7
Humerus	153.57	102.52	131.84	138.57	131.04	111.62	133.30	91.91	131.4 $\pm$ 17.9
Radius and ulna	143.72	93.26	120.42	135.28	121.48	106.36	129.80	97.24	121.0 $\pm$ 14.6
Manus	161.25	104.99	133.20	141.53	134.78	114.58	139.14	100.88	134.0 $\pm$ 17.5
Spine	122.27	104.95	116.87	131.75	133.00	119.95	143.72	97.84	121.1 $\pm$ 13.3
Ribs	118.96	100.46	110.40	126.32	119.88	92.88	122.24	77.24	114.7 $\pm$ 14.2
Sternum	104.53	97.60	95.08	115.46	114.03	90.62	139.76	81.23	101.2 $\pm$ 17.0
Pelvis	134.16	95.56	114.82	130.28	121.23	98.04	137.77	93.53	118.0 $\pm$ 12.8
Femur	147.13	106.08	125.85	124.19	125.23	110.09	141.73	102.99	124.7 $\pm$ 12.8
Tibia and fibula	139.32	92.98	116.51	137.59	118.07	92.76	128.17	90.39	117.3 $\pm$ 14.2
Feet	156.65	105.60	134.46	127.87	133.23	112.68	133.44	98.14	130.6 $\pm$ 17.0
Tail	142.48	115.95	141.03	162.99	138.02	109.69	171.56	95.83	139.5 $\pm$ 19.5
Avg.	135.98 $\pm$ 16.23	100.35 $\pm$ 7.16	121.32 $\pm$ 11.68	133.50 $\pm$ 11.03	123.91 $\pm$ 8.75	103.36 $\pm$ 9.99	137.01 $\pm$ 11.70	91.54 $\pm$ 8.05	118.37 $\pm$ 38.63

HIS

TABLE 24--Continued

FCI 226 _{Na} gm. Ca	Sows								Mean $\pm$ 95% Confidence Limits
	No. 1	No. 2	No. 3	No. 4	No. 5	No. 6	No. 8	No. 9	
Neonate	30.52	21.33	20.18	19.25	30.51	38.42	22.13	43.62	26.3 $\pm$ 7.1
2 weeks		50.07			66.67	46.69			50.1 $\pm$ 25.6
4 weeks	55.76	71.21	155.83	145.18	93.91	63.27	166.76	110.04	102.0 $\pm$ 32.2
Milk		23.60			69.03	43.36	35.32	42.12	42.1 $\pm$ 32.7
Blood	803.11	1054.17	975.03	623.38	1022.56	941.51	1036.51	951.96	963.5 $\pm$ 124.9
Muscle	94.90	352.18	85.20	315.38	323.62	270.09	277.35	530.35	296.4 $\pm$ 129.1
Skin and fat	639.61	549.69	468.63	487.11	207.20	464.82	211.59	497.08	477.9 $\pm$ 125.5
Lung	523.56	746.50	380.95	465.96	969.66	405.33	563.00	356.06	494.6 $\pm$ 170.7
Liver	104.62	70.66	166.01	185.37	61.35	62.02	112.04	109.60	107.1 $\pm$ 36.0
Heart	49.77	64.07	61.04	32.87	42.78	102.15	33.81	90.12	55.4 $\pm$ 20.1
Kidney	190.90	116.68	119.68	223.38	147.17	175.27	204.40	81.69	161.2 $\pm$ 42.1

Frankfort feed 1708.86
 Frankfort supplement 6515.26
 Frankfort water 3838.77

Goodenow feed 1551.57
 Goodenow water 625.00

TABLE 25

RADIUM TO PHOSPHOROUS RATIOS AS FCI OF ^{226}Ra PER GRAM OF PHOSPHOROUS FOR ALL
SAMPLES FROM SOWS FROM FRANKFORT, ILLINOIS

$\text{FCI } ^{226}\text{Ra}$ gm. P	Sows								Mean $\pm$ 95% Confidence Limits
	No. 1	No. 2	No. 3	No. 4	No. 5	No. 6	No. 8	No. 9	
Skull	291.10	213.75	256.41	303.88	256.85	206.70	300.62	191.33	256.6 $\pm$ 32.6
Mandible	284.90	188.89	271.95	283.61	266.29	222.22	290.27	172.22	269.1 $\pm$ 34.2
Scapula	256.55	218.52	251.40	288.79	239.55	197.07	272.26	183.74	245.5 $\pm$ 30.5
Humerus	329.31	225.77	284.62	299.00	279.15	239.13	287.13	200.56	251.9 $\pm$ 37.3
Radius and ulna	316.67	213.52	264.19	286.34	261.11	224.90	274.27	209.25	262.6 $\pm$ 31.2
Manus	352.27	230.02	296.09	314.28	293.62	245.81	299.50	215.35	294.8 $\pm$ 39.7
Spine	261.11	231.92	252.61	281.77	297.08	254.14	303.87	208.45	257.6 $\pm$ 27.7
Ribs	261.26	218.00	241.71	272.37	263.45	200.56	276.96	165.20	251.5 $\pm$ 32.4
Sternum	222.98	208.33	194.88	251.72	239.42	189.31	290.60	168.54	215.7 $\pm$ 35.4
Pelvis	293.29	206.70	252.24	276.24	259.52	209.94	294.94	198.13	255.9 $\pm$ 28.1
Femur	320.40	229.50	267.41	268.61	261.30	235.87	298.34	217.75	264.4 $\pm$ 29.8
Tibia and fibula	303.54	211.39	254.00	297.25	255.56	198.13	271.77	193.37	254.8 $\pm$ 32.0
Pes	346.59	232.83	303.54	278.41	293.29	244.85	287.65	213.12	283.0 $\pm$ 38.7
Tail	304.57	248.62	297.19	347.53	292.49	231.40	367.07	206.82	294.8 $\pm$ 46.5
Avg.	296.04 $\pm$ 36.62	219.84 $\pm$ 14.65	263.44 $\pm$ 27.80	289.27 $\pm$ 23.04	268.48 $\pm$ 19.54	221.43 $\pm$ 21.12	293.95 $\pm$ 23.68	195.99 $\pm$ 17.67	256.05 $\pm$ 84.39

TABLE 25--Continued

FCI 226 _{Ra} gm. P	Sows								Mean $\pm$ 95% Confidence Limits
	No. 1	No. 2	No. 3	No. 4	No. 5	No. 6	No. 8	No. 9	
Neonate	54.79	40.09	34.65	34.59	54.31	67.37	40.46	80.44	47.4 $\pm$ 13.3
2 weeks		94.43			112.61	79.02			94.4 $\pm$ 43.7
4 weeks	105.76	139.41	307.53	264.27	175.07	111.46	297.18	202.91	189.0 $\pm$ 58.5
Milk		48.22			130.83	100.57	71.34	57.27	71.3 $\pm$ 42.1
Blood	155.82	172.70	210.36	116.17	156.75	157.14	217.88	308.78	164.9 $\pm$ 55.9
Muscle	9.71	19.79	5.74	20.41	22.25	23.32	19.39	53.19	20.1 $\pm$ 13.8
Skin and fat	250.30	211.27	161.68	139.94	45.50	115.76	61.68	181.60	150.8 $\pm$ 59.4
Lung	175.28	174.51	131.97	184.02	178.99	137.36	119.34	120.10	155.9 $\pm$ 18.8
Liver	6.84	7.50	51.83	14.60	6.39	5.12	6.80	10.34	6.84 $\pm$ 3.10
Heart	13.99	14.41	22.30	14.77	10.47	27.27	6.74	16.91	14.6 $\pm$ 6.8
Kidney	23.57	84.08	103.24	28.63	32.34	18.72	29.54	24.93	29.1 $\pm$ 27.9

Frankfort feed 151.52
 Frankfort supplement 15,391.38
 Frankfort water 666,666.67

Goodenow feed 1,663.46
 Goodenow water 106,666.67

TABLE 26

CALCIUM TO PHOSPHOROUS RATIOS AS GRAMS OF CALCIUM PER GRAM OF PHOSPHOROUS FOR ALL
SAMPLES FROM SOWS FROM FRANKFORT, ILLINOIS

Ca/P	Sows								Mean $\pm$ 95% Confidence Limits
	No. 1	No. 2	No. 3	No. 4	No. 5	No. 6	No. 8	No. 9	
Skull	2.199	2.189	2.181	2.169	2.217	2.196	2.174	2.156	2.18 ⁺ 0.02
Mandible	2.242	2.167	2.194	2.213	2.229	2.169	2.178	2.144	2.19 ⁺ 0.03
Scapula	2.136	2.187	2.160	2.210	2.189	2.162	2.161	2.155	2.16 ⁺ 0.03
Humerus	2.144	2.202	2.159	2.158	2.130	2.142	2.154	2.182	2.15 ⁺ 0.02
Radius and ulna	2.203	2.289	2.194	2.117	2.119	2.115	2.113	2.152	2.15 ⁺ 0.05
Manus	2.185	2.191	2.223	2.221	2.178	2.145	2.152	2.135	2.18 ⁺ 0.02
Spine	2.136	2.210	2.161	2.139	2.234	2.119	2.114	2.131	2.14 ⁺ 0.04
Ribs	2.196	2.170	2.189	2.146	2.196	2.159	2.266	2.139	2.18 ⁺ 0.04
Sternum	2.137	2.130	2.050	2.180	2.100	2.089	2.079	2.075	2.09 ⁺ 0.04
Pelvis	2.186	2.163	2.197	2.120	2.141	2.141	2.141	2.118	2.14 ⁺ 0.02
Femur	2.178	2.159	2.122	2.163	2.087	2.143	2.105	2.114	2.13 ⁺ 0.03
Tibia and fibula	2.179	2.274	2.180	2.160	2.164	2.136	2.120	2.139	2.16 ⁺ 0.04
Pes	2.212	2.205	2.257	2.177	2.201	2.173	2.156	2.172	2.19 ⁺ 0.03
Tail	2.138	2.144	2.107	2.132	2.119	2.110	2.140	2.158	2.14 ⁺ 0.02
Avg.	2.18 ⁺ 0.03	2.19 ⁺ 0.04	2.17 ⁺ 0.05	2.17 ⁺ 0.04	2.17 ⁺ 0.05	2.14 ⁺ 0.03	2.15 ⁺ 0.04	2.14 ⁺ 0.03	2.16 ⁺ 0.08

150

TABLE 26--Continued

Ca/P	Sows								Mean $\pm$ 95% Confidence Limits
	No. 1	No. 2	No. 3	No. 4	No. 5	No. 6	No. 8	No. 9	
Neonate	1.796	1.879	1.717	1.797	1.780	1.754	1.820	1.844	1.80 $\pm$ 0.05
2 weeks		1.886			1.689	1.692			1.70 $\pm$ 0.26
4 weeks	1.897	1.958	1.973	1.820	1.864	1.762	1.782	1.844	1.85 $\pm$ 0.06
Milk		2.043			1.895	2.080	2.019	1.360	2.02 $\pm$ 0.21
Blood	0.194	0.164	0.216	0.186	0.153	0.167	0.210	0.324	0.186 $\pm$ 0.021
Muscle	0.102	0.056	0.052	0.068	0.069	0.086	0.070	0.100	0.070 $\pm$ 0.014
Skin and fat	0.391	0.384	0.345	0.287	0.220	0.249	0.291	0.365	0.318 $\pm$ 0.050
Lung	0.335	0.233	0.346	0.395	0.184	0.339	0.212	0.337	0.336 $\pm$ 0.061
Liver	0.065	0.106	0.312	0.079	0.104	0.082	0.061	0.094	0.082 $\pm$ 0.015
Heart	0.281	0.225	0.365	0.449	0.245	0.267	0.199	0.188	0.256 $\pm$ 0.076
Kidney	0.123	0.720	0.863	0.128	0.220	0.107	0.144	0.305	0.182 $\pm$ 0.219

Frankfort feed 0.09
 Frankfort supplement 2.362
 Frankfort water 173.7

Goodenow feed 1.072
 Goodenow water 170.7

TABLE 27

RADIUM CONTENT IN PGI OF ^{226}Ra PER GRAM OF ASH FOR ALL SAMPLES FROM
BARROWS FROM THE UNIVERSITY OF ILLINOIS

^{226}Ra PGI gm. Ash	Barrows							
	Group I				Group II			
	I-1	I-2	I-3	I-4	II-1	II-2	II-3	II-4
Humerus	5.2 ± 0.2	6.4 ± 0.2	4.4 ± 0.1	10.3 ± 0.2	4.4 ± 0.1	5.8 ± 0.2	6.5 ± 0.2	6.6 ± 0.2
Ribs	4.4 ± 0.1	5.0 ± 0.2	5.0 ± 0.1	9.4 ± 0.2	4.1 ± 0.1	7.1 ± 0.2	4.8 ± 0.1	6.2 ± 0.1
Blood	5.0 ± 0.4	2.8 ± 0.3	12.8 ± 0.6	9.2 ± 0.4	11.5 ± 0.4	4.8 ± 0.3	16.4 ± 0.7	6.5 ± 0.4
Muscle	5.5 ± 0.2	1.8 ± 0.1	5.4 ± 0.2	3.5 ± 0.2	7.8 ± 0.2	1.4 ± 0.1	4.5 ± 0.1	1.0 ± 0.1
Urine ^a	172 ± 5	219 ± 5	99 ± 4	81 ± 3	152 ± 5	98 ± 3	114 ± 3	73 ± 2
Feces	776 ± 13	676 ± 11	917 ± 15	806 ± 15	476 ± 15	342 ± 10	495 ± 13	475 ± 12
Urt	48 ± 2	25 ± 1	38 ± 2	43 ± 2	35 ± 1	18 ± 1	40 ± 2	21 ± 1
Feed	180 ± 5				204 ± 5			
Water	136 ± 5				66 ± 2			

^a PGI ^{226}Ra /liter.

TABLE 28
CALCIUM CONTENT IN MCM. OF CALCIUM PER GRAM OF ASH FOR ALL
SAMPLES FROM BARROWS FROM THE UNIVERSITY OF
ILLINOIS

mgm. Ca gm. Ash	Barrows							
	Group I				Group II			
	I-1	I-2	I-3	I-4	II-1	II-2	II-3	II-4
Humerus	388.1	386.5	384.4	380.4	381.0	385.2	393.6	385.4
Ribs	375.7	374.7	377.9	379.9	390.8	381.2	389.8	387.0
Blood	9.25	10.06	5.87	6.48	9.43	12.79	8.41	10.93
Muscle	11.46	13.19	9.61	10.70	9.15	11.53	6.85	7.89
Urine ^a	517.8	476.7	526.2	409.5	502.4	499.7	474.6	277.1
Feces	185.9	203.4	200.7	206.2	193.2	183.0	202.7	180.2
Uter	35.07	26.75	23.80	32.30	24.99	20.10	28.64	32.36
Feed Water ^a	139.2 24.2				159.3 31.4			

^a mgm. Ca/liter.

TABLE 29

PHOSPHOROUS CONTENT IN MGM. OF PHOSPHOROUS PER GRAM OF ASH
FOR ALL SAMPLES FROM BARROWS FROM THE UNIVERSITY
OF ILLINOIS

mgm. P gm. Ash	Barrows							
	Group I				Group II			
	I-1	I-2	I-3	I-4	II-1	II-2	II-3	II-4
Humerus	188.6	185.6	184.7	184.9	182.5	181.7	187.1	189.1
Ribs	198.4	188.1	193.3	184.0	176.9	182.6	185.3	185.8
Blood	38.24	46.20	32.02	34.10	32.88	30.45	34.47	44.38
Muscle	202.12	193.07	199.57	186.03	200.66	191.64	205.97	207.18
Urine ^a	1045.0	1031.3	920.0	1045.0	1065.0	667.5	858.1	782.0
Feces	188.7	196.2	209.1	189.5	196.6	131.0	182.6	189.8
Gut	190.74	147.05	173.57	178.29	181.22	190.29	176.41	169.09
Feed Water ^a	155.9 0.4				160.0 0.4			

^a mgm. P/liter.

TABLE 30

RADIUM TO CALCIUM RATIO AS $\text{fCi } ^{226}\text{Ra}$ PER GRAM OF CALCIUM FOR ALL SAMPLES
FROM BARROWS FROM THE UNIVERSITY OF ILLINOIS

fCi ²²⁶ Ra gm. Ca	Barrows								Mean $\pm$ 95% Confidence Limits	
	Group I				Group II					
	I-1	I-2	I-3	I-4	II-1	II-2	II-3	II-4		
Humerus	13.40	16.56	11.45	27.08	11.55	15.06	16.51	17.12	15.1 $\pm$	1.9
Ribs	11.71	13.34	13.23	24.74	10.49	18.62	12.31	16.02	13.3 $\pm$	4.1
Blood	539.96	278.33	2180.58	1119.75	1219.51	375.29	1950.06	594.69	907.1 $\pm$	551.6
Muscle	479.93	136.47	561.91	327.10	852.46	121.42	656.93	126.74	403.5 $\pm$	212.0
Urine	349.1	478.0	200.2	205.0	319.3	215.2	251.1	274.0	262.0 $\pm$	81.0
Feces	4174.29	3323.50	4569.01	3908.83	2463.77	1868.85	2442.03	2635.96	2979.7 $\pm$	783.0
Out	1368.69	934.57	1596.64	1331.26	1400.56	695.52	1396.64	648.95	1345.0 $\pm$	274.8
Feed Water	1293.10 5619.83				1280.60 2101.91				1286.8 3860.8	

TABLE 31

RADIUM TO PHOSPHOROUS RATIO AS fci OF ^{226}Ra PER GRAM OF PHOSPHOROUS FOR ALL
SAMPLES FROM BARROWS FROM THE UNIVERSITY OF ILLINOIS

fci ²²⁶ Ra gm. P	Barrows								Mean ± 95% Confidence Limits	
	Group I				Group II					
	I-1	I-2	I-3	I-4	II-1	II-2	II-3	II-4		
Humerus	27.57	34.48	23.82	55.70	24.11	31.92	34.74	34.90	31.9 ±	3.7
Ribs	24.44	26.58	27.78	51.09	23.18	38.88	25.90	33.37	27.2 ±	8.1
Blood	130.75	60.61	399.75	269.79	349.76	157.64	475.78	146.46	213.7 ±	120.4
Muscle	27.21	9.32	27.06	18.81	38.87	7.31	20.60	4.83	15.0 ±	9.9
Urine	165.0	212.0	108.0	78.0	143.1	147.2	133.0	93.1	138.0 ±	39.0
Feces	4112.35	3445.46	4385.46	4253.30	2421.16	2610.69	2710.84	2502.63	3078.2 ±	569.6
Gut	251.65	170.01	218.93	241.18	193.14	94.59	226.74	124.19	206.0 ±	45.6
Feed Water	1,154.59 340,000.0				1,275.00 165,000.0				1,214.8 252,500.0	

TABLE 32

CALCIUM TO PHOSPHORUS RATIOS AS GRAMS OF CALCIUM PER GRAM
OF PHOSPHORUS FOR ALL SAMPLES FROM BARROWS FROM
THE UNIVERSITY OF ILLINOIS

Ca/P	Barrows								Mean $\pm$ 95% Confidence Limits
	Group I				Group II				
	I-1	I-2	I-3	I-4	II-1	II-2	II-3	II-4	
Humerus	2.058	2.082	2.081	2.057	2.088	2.120	2.104	2.038	2.08 $\pm$ 0.02
Ribs	2.087	1.992	2.099	2.065	2.209	2.088	2.104	2.083	2.09 $\pm$ 0.06
Blood	0.242	0.218	0.296	0.190	0.287	0.420	0.244	0.246	0.245 $\pm$ 0.067
Muscle	0.057	0.068	0.048	0.058	0.046	0.060	0.033	0.038	0.052 $\pm$ 0.010
Urine	0.472	0.444	0.539	0.377	0.448	0.694	0.530	0.341	0.460 $\pm$ 0.102
Feces	0.985	1.037	0.960	1.088	0.983	1.397	1.110	0.949	1.01 $\pm$ 0.13
Gut	0.184	0.182	0.137	0.181	0.138	0.106	0.162	0.191	0.172 $\pm$ 0.025
Feed Water	0.893 60.5				0.996 78.5				0.944 69.5

TABLE 33

RADIUM TO CALCIUM OBSERVED RATIOS FOR ALL SAMPLES ANALYZED

Observed Ratios	Barrows								Mean $\pm$ 95% Confidence Limits
	I-1	I-2	I-3	I-4	II-1	II-2	II-3	II-4	
Humerus-diet	0.010	0.012	0.009	0.020	0.009	0.012	0.013	0.013	0.012 $\pm$ 0.003
Rib-diet	0.009	0.010	0.010	0.019	0.008	0.014	0.010	0.012	0.010 $\pm$ 0.003
Blood-diet	0.408	0.210	1.646	1.072	0.947	0.291	1.515	0.462	0.704 $\pm$ 0.416
Muscle-diet	0.362	0.103	0.424	0.247	0.662	0.094	0.510	0.098	0.304 $\pm$ 0.165
Urine-diet	0.263	0.361	0.151	0.155	0.248	0.167	0.195	0.213	0.204 $\pm$ 0.061
Feces-diet	3.151	2.509	3.449	2.951	1.914	1.452	1.897	2.047	2.278 $\pm$ 0.579
Feces-water	0.743	0.591	0.813	0.696	1.172	0.889	1.162	1.254	0.851 $\pm$ 0.192
Out-diet	1.033	0.705	1.205	1.005	1.089	0.696	1.085	0.504	1.019 $\pm$ 0.203
Urine-blood	0.646	1.717	0.092	0.144	0.262	0.573	0.129	0.461	0.362 $\pm$ 0.471
Muscle-blood	0.889	0.490	0.258	0.231	0.699	0.324	0.337	0.213	0.330 $\pm$ 0.196
Bone-blood	0.023	0.054	0.006	0.018	0.009	0.045	0.007	0.028	0.020 $\pm$ 0.014
	Sows								
	No. 1	No. 2	No. 3	No. 4	No. 5	No. 6	No. 8	No. 9	
Humerus-diet	0.025	0.016	0.021	0.022	0.021	0.018	0.022	0.015	0.021 $\pm$ 0.003
Rib-diet	0.019	0.016	0.018	0.020	0.020	0.015	0.020	0.012	0.018 $\pm$ 0.002
Blood-diet	0.542	0.711	0.658	0.421	0.690	0.635	0.699	0.642	0.638 $\pm$ 0.084
Milk-blood		0.022			0.067	0.050	0.033	0.044	0.044 $\pm$ 0.022
Milk-diet		0.016			0.046	0.032	0.024	0.028	0.028 $\pm$ 0.015
Muscle-diet	0.064	0.238	0.057	0.213	0.219	0.183	0.188	0.359	0.200 $\pm$ 0.088
Skin and fat-diet	0.432	0.371	0.316	0.329	0.140	0.324	0.143	0.335	0.322 $\pm$ 0.085

TABLE 33--Continued

Observed Ratios	Sows								Mean $\pm$ 95% Confidence Limits
	No. 1	No. 2	No. 3	No. 4	No. 5	No. 6	No. 8	No. 9	
Lung-diet	0.353	0.505	0.257	0.314	0.654	0.274	0.380	0.240	0.334 $\pm$ 0.120
Liver-diet	0.070	0.048	0.112	0.126	0.041	0.042	0.075	0.074	0.072 $\pm$ 0.025
Heart-diet	0.033	0.043	0.041	0.022	0.029	0.069	0.023	0.061	0.037 $\pm$ 0.014
Kidney-diet	0.129	0.078	0.080	0.151	0.099	0.119	0.139	0.055	0.109 $\pm$ 0.028
Neonate-diet	0.020	0.014	0.014	0.013	0.020	0.026	0.015	0.029	0.018 $\pm$ 0.005
Neonate-dam	0.905	0.859	0.711	0.599	0.995	1.548	0.676	1.946	0.882 $\pm$ 0.391
Bone-blood	0.041	0.023	0.030	0.052	0.029	0.026	0.032	0.023	0.030 $\pm$ 0.008
Muscle-blood	0.118	0.334	0.087	0.506	0.316	0.287	0.268	0.557	0.302 $\pm$ 0.136
Skin and fat-blood	0.797	0.521	0.481	0.781	0.203	0.494	0.204	0.522	0.508 $\pm$ 0.172
Lung-blood	0.652	0.710	0.391	0.748	0.948	0.430	0.543	0.374	0.598 $\pm$ 0.166
Liver-blood	0.130	0.067	0.170	0.297	0.060	0.066	0.108	0.115	0.112 $\pm$ 0.069
Heart-blood	0.062	0.061	0.063	0.053	0.042	0.108	0.033	0.095	0.062 $\pm$ 0.022
Kidney-blood	0.238	0.111	0.123	0.358	0.144	0.186	0.197	0.086	0.165 $\pm$ 0.070

U of I diet No. I 1324.7
 U of I diet No. II 1287.5

Goodenow diet 1482.0
 Frankfort diet 6156.3

RADIUM TO PHOSPHOROUS OBSERVED RATIOS FOR ALL SAMPLES ANALYZED

TABLE 34

Observed Ratios	Batches								Mean $\pm$ 95% Confidence Limits
	I-1	I-2	I-3	I-4	II-1	II-2	II-3	II-4	
Humerus-diet	0.023	0.029	0.020	0.0471	0.019	0.025	0.027	0.027	0.025 $\pm$ 0.003
Rib-diet	0.021	0.022	0.024	0.043	0.018	0.030	0.020	0.026	0.023 $\pm$ 0.004
Blood-diet	0.111	0.051	0.339	0.229	0.271	0.122	0.369	0.114	0.176 $\pm$ 0.092
Muscle-diet	0.023	0.008	0.023	0.016	0.030	0.006	0.016	0.004	0.016 $\pm$ 0.008
Urine-diet	0.140	0.160	0.092	0.066	0.111	0.114	0.103	0.072	0.107 $\pm$ 0.033
Feces-diet	3.465	2.920	3.716	3.604	1.878	2.025	2.102	1.941	2.511 $\pm$ 0.533
Feces-water	0.012	0.010	0.013	0.012	0.015	0.016	0.016	0.015	0.014 $\pm$ 0.002
Gut-diet	0.213	0.144	0.186	0.204	0.150	0.073	0.176	0.096	0.163 $\pm$ 0.041
Urine-blood	1.261	3.529	0.271	0.288	0.410	0.934	0.279	0.632	0.410 $\pm$ 0.327
Muscle-blood	0.207	0.157	0.068	0.070	0.111	0.049	0.043	0.035	0.069 $\pm$ 0.050
Bone-blood	0.207	0.510	0.065	0.196	0.066	0.230	0.065	0.228	0.196 $\pm$ 0.054
Humerus-diet	0.034	0.024	0.030	0.031	0.029	0.025	0.030	0.021	0.030 $\pm$ 0.004
Rib-diet	0.027	0.023	0.025	0.028	0.028	0.021	0.029	0.017	0.026 $\pm$ 0.003
Blood-diet	0.151	0.167	0.204	0.112	0.152	0.152	0.211	0.299	0.160 $\pm$ 0.054
Milk-blood	0.231	0.231	0.047	0.127	0.760	0.638	0.327	0.184	0.327 $\pm$ 0.294
Milk-diet	0.047	0.047	0.006	0.020	0.022	0.097	0.069	0.055	0.069 $\pm$ 0.052
Muscle-diet	0.009	0.019	0.006	0.020	0.022	0.022	0.019	0.051	0.019 $\pm$ 0.005
Skin and fat-diet	0.242	0.204	0.156	0.135	0.044	0.112	0.060	0.176	0.146 $\pm$ 0.057

Sows

Batches

TABLE 34--Continued

Observed Ratios	Sows								Mean $\pm$ 95% Confidence Limits
	No. 1	No. 2	No. 3	No. 4	No. 5	No. 6	No. 8	No. 9	
Lung-diet	0.170	0.169	0.128	0.178	0.173	0.133	0.116	0.116	0.151 $\pm$ 0.018
Liver-diet	0.007	0.007	0.050	0.014	0.006	0.005	0.007	0.010	0.007 $\pm$ 0.003
Heart-diet	0.014	0.014	0.022	0.014	0.010	0.026	0.007	0.016	0.014 $\pm$ 0.006
Kidney-diet	0.023	0.081	0.100	0.028	0.031	0.018	0.028	0.024	0.028 $\pm$ 0.024
Neonate-diet	0.053	0.039	0.034	0.033	0.052	0.065	0.039	0.078	0.046 $\pm$ 0.013
Neonate-dam	1.766	1.625	1.214	1.100	1.857	2.826	1.300	4.333	1.696 $\pm$ 0.938
Bone-blood	0.199	0.138	0.137	0.268	0.184	0.151	0.142	0.064	0.147 $\pm$ 0.059
Muscle-blood	0.060	0.114	0.029	0.178	0.145	0.145	0.090	0.170	0.130 $\pm$ 0.044
Skin and fat-blood	1.602	1.222	0.764	1.205	0.289	0.737	0.284	0.589	0.750 $\pm$ 0.382
Lung-blood	1.126	1.012	0.627	1.589	1.138	0.875	0.550	0.388	0.944 $\pm$ 0.348
Liver-blood	0.046	0.042	0.245	0.125	0.040	0.033	0.033	0.033	0.036 $\pm$ 0.011
Heart-blood	0.093	0.084	0.108	0.125	0.066	0.171	0.033	0.054	0.089 $\pm$ 0.040
Kidney-blood	0.152	0.185	0.490	0.250	0.204	0.118	0.133	0.080	0.178 $\pm$ 0.119

U of I diet No. I 1180.1
 U of I diet No. II 1289.4

Goodenow diet 1033.1
 Frankfort diet 9577.0

TABLE 35

WET WEIGHT, ASHED WEIGHT, AND PER CENT ASH FOR ALL SOFT
TISSUES ANALYZED

Animal	Wet Weight	Ashed Weight	% Ash	Wet Weight	Ashed Weight	% Ash
Sow No.	Kidney			Lung		
1	495	3.213	0.635	1045	7.231	0.692
2	395	3.415	0.865	1045	8.593	0.822
3	444	3.710	0.836	995	8.505	0.855
4	495	4.441	0.891	945	7.536	0.798
5	495	3.417	0.690	1240	9.008	0.726
6	395	2.408	0.610	695	8.853	1.274
8	445	2.898	0.651	695	7.176	1.032
9	395	2.154	0.545	795	8.954	1.126
Mean			0.670 ±0.083			0.838 ±0.151
Sow No.	Liver			Heart		
1	2350	29.348	1.249	595	6.154	1.034
2	2350	26.213	1.115	595	6.402	1.076
3	2900	34.277	1.182	695	7.218	1.038
4	2500	28.163	1.126	595	7.653	1.286
5	2100	24.548	1.169	595	5.741	0.965
6	2100	23.664	1.127	545	4.905	0.900
8	2100	25.663	1.222	595	4.759	0.800
9	2350	28.407	1.209	545	5.212	0.956
Mean			1.175 ±0.035			1.000 ±0.126
Sow No.	Blood			Muscle		
1	515.33	6.82	1.323	20,350	134.76	0.662
2	516.36	7.882	1.526	20,130	144.13	0.716
3	520.16	6.87	1.321	20,620	154.89	0.751
4	3851.52	48.013	1.246	19,850	139.29	0.702
5	3998.73	47.272	1.182	20,150	154.17	0.756
6	518.32	6.703	1.293	20,750	153.26	0.738
8	3522.60	44.562	1.180	20,400	146.21	0.717
9	2947.57	36.457	1.237	20,550	163.44	0.795
Mean			1.270 ±0.100			0.727 ±0.035

TABLE 35--Continued

Animal	Wet Weight	Ash Weight	% Ash	Wet Weight	Ash Weight	% Ash
Barrow	Blood			Muscle		
I-1	3333.50	38.88	1.166	16,000	131.30	0.821
I-2	3210.50	38.13	1.188	15,000	124.31	0.829
I-3	3983.50	44.37	1.114	15,000	125.64	0.838
I-4	3841.70	42.93	1.118	15,000	120.89	0.806
II-1	3317.70	43.72	1.318	17,000	133.62	0.786
II-2	3033.60	42.80	1.431	17,000	132.86	0.782
II-3	3179.60	42.96	1.351	17,000	134.16	0.789
II-4	2933.60	33.61	1.146	17,000	124.07	0.730
Mean			1.177 ±0.086			0.796 ±0.031
Sow No.	Fat and Skin					
1	20,270	22.11	2.109			
2	20,620	25.57	0.124			
3	20,330	25.15	0.124			
4	19,920	20.48	0.103			
5	20,260	26.87	0.133			
6	20,320	26.85	0.132			
8	19,910	21.32	0.107			
9	19,930	35.38	0.176			
Mean			0.124 ±0.021			
Barrow	Gut					
I-1	2,375	19.32	0.813			
I-2	2,365	26.87	1.136			
I-3	2,460	20.37	0.828			
I-4	2,320	18.85	0.812			
II-1	2,380	21.40	0.899			
II-2	2,420	18.36	0.759			
II-3	2,360	24.33	1.031			
II-4	2,430	26.88	1.106			
Mean			0.864 ±0.109			

Mean per cent ash of all bloods, 1.26 - 0.11%.

Mean per cent ash of all muscles, 0.76 - 0.05%.

TABLE 36

WET WEIGHT, ASHED WEIGHT AND PER CENT
ASH OF SOW'S MILK ANALYZED

Animal	Wet Weight	Ashed Weight	% Ash
Sow No.			
2	1080	15.16	1.40
5	100	1.23	1.23
6	560	8.46	1.46
8	280	4.49	1.60
9	500	4.51	0.90
Mean			1.40 $\pm$ 0.30

TABLE 37

RADIUM CONTENT IN Pb^{226} PER GRAM OF ASH, CALCIUM CONTENT IN MGM. OF CALCIUM PER GRAM OF ASH AND PHOSPHOROUS CONTENT IN MGM. OF PHOSPHOROUS PER GRAM OF ASH FOR SAMPLES FROM SOME LEUKEMIC ANIMALS

	Animal							
	No. 4	No. 5	No. 6	No. 7	No. 8	No. 9	No. 10	No. 11
Pb^{226} Ra/gm. Ash								
Humerus	17.2 - 0.4	54.4 - 0.8	87.9 - 0.3	124.7 - 2.2				40.1 - 1.2
Ribs	15.6 - 0.5	46.6 - 0.8	79.0 - 0.7	122.9 - 2.9	8.8 - 0.4	41.9 - 1.1	17.5 - 0.7	
Mandible								31.3 - 1.0
mgm. Ca/gm. Ash								
Humerus	389.4	389.2	383.9	390.8				394.6
Ribs	392.5	384.5	390.6	389.0	377.3	384.6	386.3	
Mandible								390.8
Teeth								386.3
mgm. P/gm. Ash								
Humerus	179.5	185.8	182.4	188.0				187.1
Ribs	179.5	198.8	183.6	186.2	183.1	188.1	188.8	
Mandible								185.1
Teeth								191.2

TABLE 38

BODY BURDENS OF ANIMALS ANALYZED AS COMPUTED FROM
DATA OBTAINED

Total FCI 226Ra	Barrows							
	I-1	I-2	I-3	I-4	II-1	II-2	II-3	II-4
Skeleton	10,084	12,466	10,137	20,726	9,130	13,630	12,677	13,695
Blood	500	251	1,131	815	1,257	561	1,763	516
Muscle	1,050	452	1,367	846	2,084	361	1,207	241
Fat and skin	735	317	957	592	1,459	253	845	168
Pluck ^a	280	197	234	245	226	99	290	170
Gut	932	657	781	816	753	330	967	565
Total	13,583	14,310	14,608	24,040	14,909	15,235	17,750	15,385

Mean Body Burden for Barrows from the University of Illinois
15,072 - 3,033 FCI 226Ra

	Sows							
	No. 1	No. 2	No. 3	No. 4	No. 5	No. 6	No. 8	No. 9
Skeleton	213,979	160,310	236,734	228,318	237,984	137,025	208,217	139,552
Blood	711	806	859	580	808	730	920	1,196
Muscle	644	1,369	409	1,424	1,542	1,555	1,327	3,677
Fat and skin	1,471	1,427	1,107	870	255	483	367	1,530
Pluck ^a	311	382	666	375	348	247	237	278
Gut	478	2,475	264	1,022	1,000	1,015	908	2,245
Total	217,594	166,769	240,038	235,412	241,938	141,064	211,976	148,479

Mean Body Burden for Sows from Frankfort, Illinois
214,785 - 29,253 FCI 226Ra

^aOrgans of thoracic cavity and abdominal cavity with the exception of the gastrointestinal tract.

TABLE 39

COMPARISON OF PER CENT ASH, CALCIUM AND PHOSPHOROUS CONTENTS AND CALCIUM TO PHOSPHOROUS
RATIO CALCULATED IN THIS STUDY WITH SOME PUBLISHED VALUES

Species	Human	Bovine	Ovine	Equine	Rabbit	Rat	Caprine	Swine	Canine	This Study mgm./gm. ash
<u>Bone</u>										
% Ash	68.8	75.0	61-64	70.2	72.0					64.1
% Ca	25.6	25.8	22.2-23.0	26.2	27-31					24.8-3.9
P	12.3	11.9	10.8	11.4	3.6-4.2					11.4-1.7
Ca/P	2.08	2.17	2.06-2.13	2.30	7.4-7.5					2.16-0.08
<u>Milk</u>										
% Ash	0.2	0.7	0.9	0.5	2.3	1.4	0.8	1.1		1.40-0.30
Ca	150-165	178	215	204			162	236		311.4-48.9
P	375	137	105	126			132	88		159.7-18.7
Ca/P	2.00	1.30	1.94	1.62			1.23	2.57		2.02-0.21
<u>Blood</u>										
% Ash	1.2							0.92		1.26-0.11
Ca	9.6							11-12		8.5-2.6
P	33.4							54-54.3		37.6-6.6
Ca/P	0.29							0.201- 0.220		0.22-0.07
<u>Heart</u>										
% Ash	0.87-1.1	0.92			1.1					1.00-0.13
Ca	4.4-15.1	11							9.3	46.2-8.9
P	113-235	174-255							217	175.4-14.7
Ca/P	0.033- 0.64	0.043- 0.062							0.043	0.25-0.08
<u>Muscle</u>										
% Ash	0.93-1.1	0.8-1.0	0.85		1.0-1.4			0.65		0.76-0.05
Ca	6.5-9.9	11	11-22		18			13.8		11.8-2.7
P	116-220	210	195-230		245			258		194.5-10.1
Ca/P	0.032- 0.085	0.052	0.054- 0.095		0.073			0.054		0.061-0.014

TABLE 39--Continued

Species	Human	Bovine	Ovine	Equine	Rabbit	Rat	Caprine	Swine	Canine	This Study mg./gm. ash
<u>Liver</u>										
% Ash	0.88-1.4	1.4			1.2-2.0	1.4-1.6				1.18±0.04
Ca	7.2-12	6			2.5-10	1.9-2.5				20.0 ±4.2
P	148-240	267				190-250				245.1 ±12.0
Ca/P	0.039- 0.069	0.022				0.010				0.082±0.015
<u>Lung</u>										
% Ash	1.0				1.0-1.3					0.84 ±0.15
Ca	17									54.0 ±10.0
P	95-120									170.2 ±8.6
Ca/P	0.179- 0.242									0.336 ±0.051
<u>Kidney</u>										
% Ash	0.8	1.1	1.3		1.3	1.3-1.5		1.2		0.67±0.08
Ca	24.2	12.7	10		5	4.3-7.9		5-9		32.0 ±4.4
P	212	237	214			207		205		195.7±21.0
Ca/P	0.113	0.054	0.057			0.021- 0.038		0.044- 0.024		0.182±0.019
<u>Fat</u>										
% Ash	0.51							0.15		0.12 ±0.02
Ca	23							25		34.6 ±7.7
P	96							255		106.8 ±17.2
Ca/P	0.242							0.098		0.318 ±0.050
<u>Out</u>										
% Ash	0.86-0.90									0.864 ±0.109
Ca	14.0									27.7 ±4.4
P	128.0									177.4 ±12.7
Ca/P	0.109									0.172 ±0.025

reasons, not the least of which is the difficulty in determining exact wet weights of the tissue being examined.

The scarcity of comparative data and lack of uniform techniques for determining and expressing data, indicate to us that there is need of comprehensive study of comparative biochemistry in all aspects of the field of ^{226}Ra metabolism in non-human mammals.