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A STUDY OF FISSION PRODUCT LEVELS AND
METABOLISM IN FOOD PRODUCING
ANIMALS AS DETERMINED BY WHOLE BODY COUNTING
PROGRESS REPORT

August 15, 1965 - August 15, 1966

Submitted by the
Department of Animal Science
College of Agriculture
and the
Department of Veterinary Physiology and Pharmacology
College of Veterinary Medicine

University of Illinois
Urbana, Illinois

RELEASED FOR ANNOUNCEMENT
IN NUCLEAR SCIENCE ABSTRACTS

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Summary

The Illinois Animal Science Counter (ILLASCO) has been calibrated for K-40 gamma detection efficiency in human beings by means of intravenous K-42 injection of 25 male volunteers. This calibration will be used in studies of the effects of exercise on body composition by the Department of Physical Education, University of Illinois. Calibrations for K-40 detection efficiency in swine have been continued, also by the use of K-42 administration. Potassium-40 measurements are being studied in relation to growth and as a means of selecting breeding stock in both sheep and swine. Several hundred animals are being used in these projects.

Cesium-137 retention has been measured in 3 cattle and 2 calves for 60 days after both oral and intravenous dosing, and in 4 sheep for 60 days after intravenous dosing. Retentions in both species can be described as 3-component sums of exponentials. For cattle after oral dosing, excretory processes having half-lives ($T_{1/2}$) of 1.3, 4.6, and 32 days accounted for 0.21, 0.33, and 0.43 of the administered dose, respectively. For 1 pregnant cow, compartment 3 had a $T_{1/2}$ of 42 days and accounted for 0.58 of the dose. Calves excreted Cs-137 more rapidly than cows, primarily because greater fractions of the dose were lost via the fast compartments 1 and 2. In cows, intravenous dosing appeared to accentuate loss by the first component at the expense of the second. In sheep, the average $T_{1/2}$'s were 1.2, 5.0, and 27 days, accounting for 0.28, 0.33, and 0.39 of the dose, respectively.

Methods for studying Ca-47 kinetics in dairy goats were developed. From November 15, 1965, to August 15, 1966, fifteen injections of Ca-47 were made in each of 5 animals. Blood specific activity-time

curves and whole-body retentions were obtained and exchangeable Ca pool sizes (E) and pool half-lives were calculated. Before pregnancy and during early pregnancy, E remained rather constant in individual animals and ranged between 3 gm Ca and 14 gm Ca in 5 goats. Pool half-lives varied from 20 hours to 32 hours. During late pregnancy, pool half-lives decreased as parturition approached to as low as 11 hours.

An investigation of Sr-85 retention in pigs has been initiated. The effects of age at which Sr-85 is administered and of dietary Ca and P content on retention are being studied.

Personnel

Persons employed with funds from this grant are:

(1) W. C. Crackel, B.S., Agriculture, University of Illinois, 1963; as a Research Assistant.

(2) D. Hartman, High School Graduate; as an Electronics Technician I until April 1, 1966.

(3) R. Kowalczyk, B.S., Agriculture, Western Illinois; as a Natural Science Technician II to replace D. Hartman.

(4) K. Bevans, half-time typist.

(5) Hourly Help: veterinary medical students hired as animal caretakers and laboratory assistants.

(6) J. J. B. Anderson, Ph.D. Cornell University, 1966; Assistant Professor. A portion of this salary from June 15 to September 1, 1966, from this grant. The university assumes responsibility for the entire salary September 1, 1966.

The addition of Dr. Anderson to our staff has greatly increased the level of Ph.D. participation, and his research experience is especially

suitable for the broad objectives of this project. He completed his Ph.D. under C. L. Comar, New York State Veterinary College, Cornell University, in February, 1966. The subject of his thesis research was Sr-85 retention in dogs as related to growth, exercise, and age at the time of isotope administration. He continued as a post-doctoral fellow under C. L. Comar until June, 1966, studying strontium absorption by the rat, dog and cow as affected by rations containing sodium alginate. He received his B.A. from Williams College (1956), M.A.T. from Harvard (1958), and M.A. from Boston University (1962). He was a participant in the Academic Year Institute on Radiation Biology at Cornell University, and subsequently joined the Department of Physical Biology as a graduate student.

Upon joining our staff in June, 1966, Dr. Anderson initiated a study of Sr-85 retention in pigs which is briefly described in this report. He also obtained 6 purebred beagles on loan from C. L. Comar which will be used to establish a beagle colony and for proposed research during the coming year. His participation in this project has been 50% of full time since July 1, 1966, and is proposed for 50% full time on future projects.

Persons participating in this project and funded from sources other than this project have been:

a. Project Director

A. R. Twardock, Ph.D.; Associate Professor of Veterinary Physiology and Pharmacology and Veterinary Pathology and Hygiene (30% of time to this project).

b. Associate Project Directors

(1) R. P. Link, Ph.D.; Professor and Head, Department of

Physiology and Pharmacology (5% of time to this project).

- (2) R. O. Nesheim, Ph.D.; Professor and Head, Department of Animal Science (5% of time to this project).

c. Staff Scientists

- (1) B. C. Breidenstein, Ph.D.; Associate Professor of Meats Technology (10% of time to this project).
- (2) U. S. Garrigus, Ph.D.; Professor of Animal Science (10% of time to this project).
- (3) K. A. Kendall, Ph.D.; Professor of Dairy Science (10% of time to this project).
- (4) H. W. Norton, Ph.D.; Professor of Statistical Design and Analysis, available for consultation as needed.

d. Other Personnel

- (1) T. G. Lohman, B.S.; Graduate Student; Assistant in Animal Science (5 to 10% of time to this project).
- (2) Y.-H. Kuo, M.S.; Graduate Assistant in Veterinary Physiology and Pharmacology (25% of time to this project).
- (3) R. H. Ball, M.S.; Graduate Assistant in Animal Science (10% of time to this project).
- (4) P. M. Ribisl, A.M.; Instructor in Physical Education (5 to 10% of time to this project).

Dr. B. C. Breidenstein terminated his appointment with the University of Illinois on August 15, 1966. His activities will be assumed by a member of the Animal Science Department to be named in the near future (tentatively, Dr. Joseph Kastelik, Professor of Nutritional Biochemistry; presently assigned to A.I.D., Sierra Leone, and due to return September, 1966).

Efficiency Calibrations and Body Composition Studies

During the past year, K-42 efficiency calibrations have been carried out in human beings, and additional calibrations have been obtained for swine. Twenty-five men volunteered to receive K-42 injections, most of whom were participants in a physical fitness training program of P. M. Ribisl, graduate student of Professor T. K. Cureton. The men were selected to represent a wide range of body types, weights, and heights. Anthropometric measurements were taken at the time of K-42 counting for possible correlation to counting results. Five subjects were measured for each of 5 evenings during the weeks of July 4th, 18th, and 25th. Results of these calibrations are not yet available.

R. H. Ball, graduate student in Animal Science under Prof. Breidenstein has been measuring the K-40 content of pigs during growth. Three to four pigs have been injected with K-42 at approximately monthly intervals and the project is still in progress. When it is complete, K-42 gamma-detection efficiencies will be known for pigs weighing from 50 to 350 pounds. These data will be compared with previous K-42 calibrations in swine (our progress report, 1964-65, p. 10) and will serve to increase the accuracy of K-40 measurements in that species.

These calibrations were also needed because a new loading system has been devised for swine. Rather than the previously used sling device, a capsule arrangement is now in use. Swine injected with K-42 for this study were measured in both the sling and capsule, which will allow data obtained using both loading devices to be directly compared. The new loading system permits the measurement of K-40 in more than 30 pigs during an 8-hour day by a two-man crew, a considerable improvement over the sling method in both numbers per day and labor required.

Although body composition studies are not among the primary objectives of the project at this time, the effects of fission products on whole-body K-40 measurements are continually of interest. Also, funds from the project are used to pay the counter operator who spends a significant portion of his time working in cooperation with scientists studying body composition in animals and human beings. Two papers dealing with whole-body K-40 measurements in cattle, including effects of fission products on such measurements, were recently accepted for publication by the Journal of Animal Science; copies of these papers are included with this report.

Outlines of Current Body Composition Studies:

Method of Measuring, and Factors Influencing, Body Composition in Swine

Project Leaders: B. C. Breidenstein and H. W. Norton

Objectives:

1. To determine the relative effectiveness of the following methods of determining proximate body composition.
 - a - Outcome groups according to breed, sex, and live weight
 - b - Live K-40 count
 - c - Whole body potassium
 - d - Body measurements
 - e - Carcass specific gravity
 - f - Carcass K-40 count
 - g - Partial and complete cut-out
2. To study the effects of dietary protein and energy levels on body composition from weaning to approximately 350 pounds live weight.
3. To study the effects of breed and sex on body composition in response to dietary treatments.

Justification:

Live weight has been used as a measure of animal performance for many years, although there seems to be general agreement that it is inadequate in market-weight animals. As a result, carcass data have been used as an attempt to improve the assessment of performance. There is, however, little evidence of unanimity of thought with regard to its usefulness. Carcass data are useful only when a clear-cut understanding of their role has been established. All traditional carcass criteria can be rather readily classified as either quantitative or qualitative.

It is the purpose of this proposed research to study only the quantity of lean meat produced. The very existence of the meat animal industry is dependent upon the capability of animals to convert feedstuffs to meat. The use of live weight as a measure of meat production is, in many cases, grossly inadequate and is often misleading. The use of a model in which constants have been fitted for normal outcome group criteria including breed, sex, age, live weight, and carcass weight accounts for 45% of the variance in weight of fat-free lean tissue in market-weight swine. The use of live K-40 count alone accounts for 81% ($r = .901$) of the variance (1). The addition of breed, sex, age, live weight and carcass weight add nothing to the variance accounted for. Other non-destructive methods of determining body composition in the living animal generally account for about 50 to 60% of the variance (2).

The primary limitation in the work thus far conducted at Illinois is in numbers of observations ($n = 30$) and the weight and age range of the observed animals. In addition, calibration curves for different weights of animals through the use of K-42 are needed to maximize effectiveness of the counter in determining body composition.

Planes of nutrition and their effect on body composition have been studied by Hammond (3) and McMeekan (4). These studies were, however, conducted in 1937 and 1940 respectively and on breeds of swine presumed to be quite different from modern American swine in terms of body composition at a given age. Moreover, because of the necessity of slaughtering and dissection of the animals, the numbers were small and, of course, an animal could not be retained on the experiment for the full age span through which the studies were made. The whole-body counter permits observation of each animal's composition during his own complete

age span being studied. The response of each individual animal can thus be observed, rather than assuming that one animal of a group properly represents the body composition of that group.

Procedure - Year 1

The animals will originate from, and be housed at, the Moorman Farm. Within breed and sex, animals will be allotted to slaughter weight groups of 50, 100, 150, 200, 250, 300 and 350 pounds. Boars, barrows, and gilts will be used.

Schematic Design

Slaughter Weights	Sex		
	Barrows	Gilts	Boars
50			
100			
150			
200			
250			
300			
350			

A minimum of 2 animals will be assigned to each cell. The animals will be lot-fed on appropriate conventional ration for their age and weight. The ration will contain 16% protein from weaning to 100 pounds and 12% from 100 pounds to 350 pounds.

Counting the animals for K-40 activity will be done one to three days prior to slaughter at the assigned weights. A portion of the animals will be injected with K-42 to supply needed calibration data.

These will be selected dependent upon the need at the time inasmuch as some K-42 calibration work is planned for June 1965.

Slaughter will be in accordance with normal practice. Conventional measures will be used for subcutaneous fat thickness, muscle areas and body length. Specific gravity will be by water displacement in vertical suspension at 48 hours post mortem. Carcass counting for K-40 activity will be done as soon thereafter as possible. One side of the carcass will be cut according to procedures recommended by the Reciprocal Meats Conference. The other side will be separated into skin, bone, and boneless mass. The boneless mass will be sampled for determination of moisture, ether extract and nitrogen.

In the second, third, and fourth years the animals will be fed on protein levels calculated in relation to muscle mass weight. Various protein levels will be used, based on the consumption/unit of muscle mass in the first year. The animals will be subjected to whole body counting every two to three weeks.

The method of least squares and (or) other appropriate statistical methods will be used in the interpretation of the data.

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University of Illinois
Animal Science Department
Ruminant Division Experiment R47

TITLE: The production and evaluation of the "large" lamb

JUSTIFICATION: Previous unpublished work from this Station indicated that lambs selected from relatively large-sized parent stock and grown to an average of 150 pounds live weight gained rapidly (0.686 pound per day average), efficiently (weaning to slaughter feed to gain ratio of 5.83), and produced more carcass and fat-free carcass lean per day of age than conventional lambs. Such lambs because of their large size could nearly double the pounds of lamb produced per year per ewe. The packer could anticipate the production of more pounds of lamb carcass per unit time and thus a reduction in the processing costs per pound of finished product. The wholesaler and retailer would have larger cuts and therefore greater flexibility in the manufacture of consumer products. In the final analysis the consumer would have a greater choice of more attractive cuts.

The following is a proposal for the first year's work directed toward establishing whether or not with lambs from parents of relatively large mature size, a "large" lamb can be rapidly, efficiently, and consistently produced. In the course of this work random samples of the animals on the individual studies will be slaughtered following K-40 counting to evaluate the composition of the carcass and thus the composition of the gain as well as the degree to which K-40 count can be used to estimate carcass composition. Although the K-40 counter has been used on slaughter animals within the normal range of weights found in the traditional finished slaughter lamb, at best, limited and in most cases no information is as yet available for lambs above 110 to 115 pounds live weight.

OBJECTIVE:

1. Study the rate and efficiency of growth of lambs fed to unusually heavy weights.
2. Study the composition of lamb gains using both the slaughter and K-40 technique.

PROCEDURE: Initially only wether lambs (castrated at about 30 days of age) will be used, and they will be weaned at 60 ± 3 days of age. For maximum numbers in one year and efficient use of facilities it is proposed that lambs from the Dixon Springs Agricultural Center (DSAC) ewe flock (lambling in February) as well as lambs from the Rambouillet flock in Urbana (lambling in July and November) be used. All lambs will be individually housed on elevated platforms and fed in Urbana.

The lambs will be housed in the metabolism unit only during periods of climatic extremes (December 15 to March 15), and the rest of the time housed in facilities such as the Jones barn.

For effective K-40 count lambs must be in "short fleece."

The tentative time periods and numbers of lambs involved will be as follows:

1. May, 1966. 36 lambs, the exact number depending on the number of lambs born, the sex ratio, the preweaning performance of the lambs, and the availability of facilities.
2. September, 1966. 12 lambs with adjustments made for the demands on Rambouillet lambs such as other research, classwork, ram replacements, et cetera, as well as the considerations listed in Item 1.
3. January, 1967. 18 lambs subject to adjustments as in Items 1 and 2.

Lambs will be slaughtered at 25-pound weight increments starting at 50 pounds live weight or about 75 days of age and continuing to and including 175 pounds live weight. The following is a schematic presentation of the foregoing:

Weight at slaughter (lb)	Number of lambs per lambing date		
	Feb. 1966	July 1966	Nov. 1966
50	6	2	3
75	6	2	3
100	6	2	3
125	6	2	3
150	6	2	3
175	6	2	3
Maximum date of termination	Oct. 1966	Feb. 1967	July 1967

Because not all animals can be expected to reach their designated final weights, particularly the higher weights, in a reasonable length of time or possibly never, a time limit will be imposed at which time all remaining lambs will be slaughtered. It is suggested lambs be maintained on study until 240 days of age or they reach the designated weight, whichever comes first.

The following data will be collected:

1. Initial body weight and body weight at 14-day intervals thereafter, except immediately prior to the assigned slaughter weight and then at 7-day intervals. Lambs will be changed from

the creep diet to the test diet over a four-day period; and about seven days, on the test diet and in the individual feeding units, will be allowed for the lambs to adjust prior to taking the initial weight and making the initial K-40 count.

2. Daily feed intake: The test diet will be fed to appetite. Samples of feed will be collected for analysis and counting.
3. K-40 count initially and at approximately 28-day intervals and immediately prior to slaughter. The lambs will be sheared initially and at about 56-day intervals thereafter unless more frequent shearing is required for accurate K-40 count. The protocol prior to slaughter will be:
 - a. Weigh off test on Monday
 - b. Weigh and count on Tuesday
 - c. Weigh and kill on Wednesday
4. Carcass observations
 - a. Detailed carcass cut-out as a means of reflecting tissue distribution.
 - b. Proximate chemical analysis of one side of each lamb as a means of deriving a meat yield value of a chemically constant lean-fat composition.
 - c. Loin-eye area as a reflection of display appeal.
 - d. Consumer acceptance on selected cuts from the side not used in proximate analysis.
 - e. Carcass K-40 count as a means of determining composition. If this proves successful, the scope of the project can be broadened to permit improved consumer evaluation.

Because variation in feed could markedly influence the performance of lambs it is further suggested that enough diet be mixed at one time from similar ingredients to last the duration of the study (See Table 1).

Table 1. Diet^{a/}

Ingredient	Pound
Corn, No. 2, shelled yellow, ground	355
Hay, ground	400
Soybean meal	180
Molasses, liquid	50
Limestone, feeding grade	10
Salt, plain	5
Vitamin A (1,000 IU per pound)	
Vitamin D (250 IU per pound)	
Antibiotic (10 mg per pound)	

^{a/}To be pelleted in 1/4-inch pellets and delivered in bulk.

Cesium-137 Metabolism in Cattle, Sheep, and Swine

Cattle: Three Guernsey cows and 2 calves were given Cs-137 by capsule and were whole-body counted for 60 days, after which they were injected intravenously with Cs-137 and counted for another 60 days. The oral Cs-137 dose was approximately 0.5 μc to each cow and 0.15 μc to each calf; intravenous doses were 0.36 μc per cow and 0.14 μc per calf. Prior to oral dosing the animals were fed a constant daily intake of ground feed and pelleted hay, and whole-body counted until Cs-137 body burdens reached a plateau. The same intake was maintained throughout the experiment except after calving, and the pre-dose body burden was subtracted from all subsequent measurements. Two cows were pregnant; at the end of the I.V. dose experiment, cow 1 was approximately 4 months pregnant and cow 3 was approximately 8 months pregnant. During the 120 day period, each cow gained about 100 lb and each calf about 200 lb. Initial body weights were: cow 1, 640 lb; cow 2, 680 lb; cow 3, 840 lb; calf 1, 122 lb; calf 2, 126 lb.

At the time of oral Cs-137 dosing, 0.5 μc of Cs-137 was put in a 55-gallon water-filled barrel. This standard was counted each time the animals were counted for the purpose of correcting for variations in instrument gamma-detection efficiency. Two measurements were made on the days of oral and I.V. dosing, the second being used as the 1.00 fractional body burden at zero time. Immediately before oral dosing, the cow's Cs-137 body burdens ranged from 38 to 55 nanocuries and the calves' body burdens were about 10 nanocuries. Oral dosing raised the animals' count-rates from less than half to 6 to 9 times background in the cows and to twice background in the calves. Sixty days after oral dosing, from 75 to 88% of the added body burden was excreted from cows, and 95% from the calves. Except for

the pregnant cow in which 25% of the dose remained, the balance of the oral dose should not have interfered to any great extent with the subsequent I.V. dose measurements.

Results from the cows and calves are given in tables 1, 2, and 3, and in figure 1. The retention curves for all the animals were similar in appearance, and data for only calf 1 are shown (figure 1). Retention appeared to be a single exponential function of time after 20 days, although continued counting might have shown that the semi-log plot was still curvilinear. All retention data were hand plotted and the time intervals after dosing during which semi-log plots appeared to be linear were determined. Regressions for log-nanocuries body burden against days after dosing were determined by computer, subtracted from the original retention curve, and regressions determined for the linear portions of the remainder. Repeated subtractions showed that retention could be described as a 3-component exponential function of time after dosing, as illustrated by figure 1. Half-lives and compartment sizes are given in table 1, and the equations in exponential form in table 2. Richmond et al. (8) found that a 3-component exponential model could be used to describe Cs-137 retention in mice.

Orally administered Cs-137 was excreted by three processes having half-lives of 1.3 days, 4.6 days, and 32 days, accounting for 0.21, 0.33, and 0.43 of the administered dose, respectively, in cows 1 and 2. Cow 3, which was in mid- to late pregnancy during the experiments, exhibited a different retention pattern, compartment 3 having a longer $T_{1/2}$ (42 days) and accounting for more (0.58) of the dose. The calves excreted Cs-137 more rapidly than cows, primarily because greater fractions of the dose were lost via the fast components

1 and 2. Half-lives of the fast components were similar in cows and calves, whereas the third component $T_{1/2}$ was shorter in calves, especially after oral dosing. Differences between oral and I.V. dose retention in the calves must be interpreted with the fact in mind that their body weights increased from 120 lb to almost 200 lb during the oral dose retention study. In cows, intravenous dosing appeared to accentuate the amount lost through the first component at the expense of the second, the third compartment size remaining relatively constant except in the pregnant cow (cow 3).

Hood and Comar (7) observed that cattle excreted orally administered Cs-137 in the feces within 24 hours after dosing, much more rapidly than rumen contents move through the intestinal tract. They suggested that Cs-137 was absorbed through the rumen wall and excreted further along the G.I. tract, probably in the large intestine. Compartment 1 in our data could be interpreted to represent fecal excretion on the basis that I.V. administration might be expected to increase fecal excretion as a result of bypassing absorption through the rumen. Compartment 3 might represent loss from soft tissues since its turnover is distinctly different in the mature and growing animal. These interpretations are only speculative and must be considered at greater length prior to publication.

The biological half-time ($B_T 1/2$), defined as the time required for excretion of one-half the initial dose, has been calculated for all animals for the purpose of comparison with similar values published in the literature (table 3). The $B_T 1/2$ for cows 1 and 2 averaged 7 days, and 16.5 days for pregnant cow 3. The $B_T 1/2$ for calves averaged 4.6 days, with evidence of an increase as body weight and age increased.

Hood and Comar (7) reported B_T $1/2$'s of 4 days and 9 days in cows dosed orally and intravenously, respectively.

Sheep: Procedures similar to those employed in cattle were used to observe intravenously injected Cs-137 retention in sheep. The 60-day retention study was terminated only recently and computer analysis of the data has not yet been carried out. For this report, the net animal count-rates were plotted and curve fitting was performed graphically. Shapes of the retention curves for all 4 sheep were similar; figure 2 shows the retention pattern of one-half a microcurie of Cs-137 for sheep 1. Prior to dosing, the sheep were individually fed a constant daily amount of grain and pelleted hay for 2 $1/2$ weeks until their body burdens reached a plateau. They received the same feed intake for the entire experimental period and gained from 8 lb to 16 lb each from initial weights of 82 lb to 97 lb. Sheep 1 and 3 were ewes; 2 and 4, wethers.

Results of the graphic analyses are given in table 4. As for cattle, a 3-component sum of exponentials provides a satisfactory model of retention. The 3 components had average half-lives in 4 sheep of 1.2 days, 5.0 days, and 27 days, and accounted for fractional excretions of the total dose of 0.28, 0.33, and 0.39, respectively. These half-lives and compartment sizes do not differ greatly from those observed in cows 1 and 2 injected I.V. (half-lives: 1.4 days, 5.3 days, and 34 days; compartment sizes: 0.30, 0.27, and 0.42). The biological half-time averaged 5.6 days in sheep as compared to 6.2 days for cows 1 and 2 dosed I.V.

Wasserman et al. (10) reported single oral dose Cs-137 excretion data for a 10-day period in goats. Their data were described by a 2-compartment sum of exponentials with half-lives of 0.7 and 3.5 days.

Over 90% of the dose was eliminated within 10 days, and they stated that a third component would account for only a small portion of total excretion if it were present. In our sheep however, only about 60% of the dose was excreted in 10 days, and the third component became evident. It is possible that a species difference exists, but Wasserman's goats were lactating and our sheep were not, which might also account for the difference. Shannon et al. (9) refer to observations in sheep indicating more than 50% excretion within 10 days of dosing, and a lower turnover in younger than in older animals. The latter observation differs from ours in calves as compared to cattle.

Some confusion appears to exist regarding the biological half-time of Cs-137 retention in the cow. Comar gives a figure of 20 days in one reference (2) but, as previously mentioned, Hood and Comar observed half-times of 4 and 9 days in cows dosed orally and I.V. (7) which is in better agreement with our data. The 20-day $B_T 1/2$ is given in the USDA Radiological Training Manual (2), without reference to its origin. Wasserman et al. (10) cite the same value and refer to ORO-91 (7). However, we can find no mention of a 20-day $B_T 1/2$ in either this report or the scientific publication taken from it (6), and the data in both publications indicate a $B_T 1/2$ close to 7 days (ORO-91, p. 17, figure 4, and p. 18). Cumulative excretion curves of single oral doses of Cs-137 in milk, urine, and feces of lactating cows were published by Cragle (3). His data also indicate a short $B_T 1/2$, between 3 and 4 days.

Swine: Four 120-lb Yorkshire-Duroc crossbreeds have recently been purchased and are being fed a standard ration in preparation for Cs-137 injection and whole-body counting, which should be initiated before September 1.

Calcium-47 Kinetics in Dairy Goats

Seven dairy goats (6 does and 1 buck) were purchased in September, 1965. One doe died shortly after purchase, apparently from parasitism and liver pathology. Beginning in November, 1965, 5 does have been injected intravenously with Ca-47 at 2- to 4-week intervals for the purpose of studying exchangeable Ca pool sizes and turnover rates. From November 15, 1965, to August 15, 1966, fifteen injections have been made in each animal. Injection schedules are dictated by the Ca-47 shipping schedules of Isoserve, Inc. The does were bred as they came into heat; two (1 and 7) delivered twins in February, 1966, and two (4 and 6) are now approximately 3 months pregnant (8/15/66). Thus it has been possible to observe changes in Ca kinetics as affected by gestation, parturition, and lactation. The short-lived Ca-47 is especially useful for this purpose because it allows repeated measurements to be made in the same animal within relatively short time intervals (easily within 2 weeks).

Methods: Each goat is given approximately 15 μ c Ca-47 by intravenous injection into the jugular vein and the same quantity is put in a 15-gallon water-filled barrel which serves as an instrument and decay standard for whole-body counting. Whole-body counts obtained 3-4 hours after injection are assumed to represent 100% retention. Whole-body counting is continued at twice-daily intervals for the first 5 days and at greater intervals thereafter. We anticipate that fewer whole-body counts will be required for accretion measurements once the preliminary data are analyzed and experimental procedures are refined. Heparinated blood samples are obtained during the 7 days after injection. Sampling times have varied as experience and data have accumulated. Plasma Ca-47 concentrations decrease very rapidly in lactating as compared

to non-lactating goats. Experience from the past 9 months has shown that sampling schedules must be adjusted according to the physiological status of the animal. Plasma concentrations fall so rapidly during lactation that counting results become statistically meaningless in samples taken later than 48 to 72 hours post-injection. Methods of following the plasma Ca-47 disappearance curve under such circumstances are being sought. Possible methods include increasing the size of dose administered, analyzing milk specific activity data, and more frequent sampling during the first 48 hours post-injection.

Dose size cannot be increased with the present whole-body counting procedure. Only 2 phototubes of the Illasco are used so as to decrease detector sensitivity to the large 15 μ c dose. Between 1 and 15 μ c, a plot of activity versus counting efficiency shows slight indications of nonlinearity at higher activities. A larger dose could be administered if we used the 5" x 4" NaI(Tl) detector as a whole-body counter. Cost of radioactivity is a prohibiting factor, however. Milk samples have been taken for the last 2 experiments and show promise as a means of performing kinetic studies during lactation. Counting statistics are much better due to the greater Ca content of milk than plasma, and repeated venipuncture can be avoided. The only question relates to the time lag between milk and plasma specific activity curves. Two or three more experiments will be performed using both plasma and milk sampling to determine the relationship between the specific activities of the two fluids and whether or not milk can be used as a partial or complete replacement for plasma.

Results: The first problem was to interpret the functional significance of semi-logarithmic plots of plasma Ca specific activity

data. Heaney and Whedon (5), Bauer et al. (1), and others state that, in human beings, plasma radiocalcium specific activity curves can be described as a sum of exponentials, the first 2 or 3 terms of which represent mixing in the "exchangeable Ca pool" (E) and the next term representing removal from E by accretion and excretion. Logarithmic removal after the mixing phase is observed until tracer deposited in bone by accretion starts to return to plasma as a result of resorption, at which time a break in the retention curve is observed at a time referred to by Heaney and Whedon as " θ " (theta). Some investigators (4) interpret the logarithmic removal phase and its change in slope as evidence of an exchange process rather than accretion and resorption. In human beings, θ is usually observed more than 100 hr after injection; the mixing phase is usually assumed to be complete at about 24 hr, the interval from 24 hr to more than 100 hr being useful for estimating E and accretion rates or bone formation rates.

Figure 8 illustrates plasma Ca specific activity as a function of time following I.V. injection of goat 6, the 7th experiment. These data are better than some we have obtained, but representative of most experiments. The first 6 points appear to follow a single exponential pattern, and at about 60 hours a definite break to a lesser slope is evident. This pattern is apparent in a majority of the curves, but is not always readily observed. Only recently, after many observations, has it seemed apparent that the pattern of radiocalcium disappearance in goat plasma is similar to that described in human beings. Interpreting the curves was difficult because of the relatively short time between mixing and θ , in comparison to human beings. Theta varies from 40 to 80 hours, occurring earlier during pregnancy and lactation.

We are particularly interested in measuring the size of E (the exchangeable Ca pool) and its turnover, with the hope that these might be used as parameters of the animals' ability to maintain normal Ca homeostasis during pregnancy, parturition and lactation. We plan, eventually, to apply the methods developed in dairy goats to a study of parturient paresis in dairy cattle. The data would also apply to changes in strontium retention and turnover in bone during the various stages of pregnancy and lactation. E was calculated by determining the intercept of the logarithmic removal phase (prior to θ) at zero hour. This intercept represents the plasma radiocalcium specific activity prior to any loss from E, had mixing been instantaneously complete at the time of injection. E is obtained by dividing the intercept specific activity (% dose/gm Ca) into the total dose (100%). Plasma specific activity data were recorded on IBM cards and regressions of log S.A. on time after dosing were determined by computer. Logs of intercepts and slopes were converted to E values and half-lives which are shown in figures 3 to 7.

Figure 3 shows kinetic data for goat 1, which was neither pregnant nor lactating during the time period illustrated. Exchangeable pool (E) values fluctuated between 3 and 5.5 gm, and half-lives of Ca removal from the pool between 20 and 32 hours. Data from goats 4 and 6 (figures 4 and 5) are similar, although pool sizes were larger than in goat 1. Half-lives were 20 hours or more, and ranged as high as 32 hours. Goats 4 and 6 were bred at approximately 180 days. In goat 4, in particular, E and $T_{1/2}$ appeared to decrease until breeding and may now be increasing. In all goats there seemed to be a direct relationship between E and $T_{1/2}$, small pool sizes and short half-lives generally being associated.

The data from goats 7 and 14 are most interesting. Parturition dates are indicated. The striking feature in both graphs is a marked drop in half-life beginning about 60 days before parturition. Pool size in goat 7 appeared to increase gradually until parturition, whereas the reverse may have been true in goat 14. Data from goat 14 were the most difficult to interpret; Ca-47 was removed so rapidly from her plasma that counting statistics deteriorated rapidly with increasing sampling times after injection, especially at parturition (1-hr counts on 15 ml plasma). Nevertheless, marked changes in Ca kinetics were evident, and can be measured more accurately with improved methods in the future.

We are now in the process of calculating accretion rates from the whole-body counts and plasma specific activity, but have not progressed to a point where the results can be reported. The approach shows considerable promise as a means of studying Ca kinetics in animals, and we are very interested in expanding this work to include additional animals and to investigate the effects of nutritional factors, as explained in the renewal proposal.

Strontium-85 Retention in Pigs

An experiment was recently initiated to study Sr-85 retention in pigs. The scientific background and objectives of this project are described in the current renewal proposal. Two litters of 8 and 9 baby pigs were obtained at 1 day of age after they received maternal colostrum. They are individually housed in specially constructed units designed for feces and urine collection, and are hand-fed until old enough to eat of their own accord.

Two pigs from each litter were injected intravenously with one-half microcurie Sr-85 between 1 and 3 days of age, and retention is being

determined by daily whole-body counting for the first week and weekly thereafter. Two more pigs from each litter will be injected at 2 months, 4 months, and 6 months of age. The ration of one litter contains 0.8% Ca and 0.6% P, and the other contains 1.2% Ca and 1.0% P. Strontium-85 whole-body retention as a function of age at the time of injection and dietary Ca and P levels will be observed.

Publications

Two papers have been accepted for publication by the Journal of Animal Science and are now in press:

1. Twardock, A. R., T. G. Lohman, G. S. Smith, and B. C. Breidenstein. Symposium on Atomic Energy in Animal Science: Part I. The Illinois Animal Science Center; Performance Characteristics and Animal Radioactivity Measurement Procedures.
2. Lohman, T. G., B. C. Breidenstein, A. R. Twardock, and H. W. Norton. Symposium on Atomic Energy in Animal Science: Part II. Estimation of Carcass Lean Muscle Mass in Steers by K-40 Measurement.

The subject matter of these papers was presented at the 56th Annual Meeting of the American Society of Animal Science, Knoxville, Tennessee. Six copies of each manuscript are included with this report.

The work on Cs-137 metabolism will be published when the retention studies in swine are finished this fall. Data from 3 species (cattle, sheep, and swine) can then be included in the paper.

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Table 1

Half-lives and Compartment Sizes for Three-Component Cesium-137
Retention Curves in Cattle and Calves for 60 Days Following Oral
and Intravenous Administration.

<u>Oral Dose</u>						
Cow No.	<u>Pool 1</u>		<u>Pool 2</u>		<u>Pool 3</u>	
	A ₁	T _{1/2}	A ₂	T _{1/2}	A ₃	T _{1/2}
1	.287	1.34	.332	4.62	.380	31.0
2	.166	1.31	.327	4.56	.480	32.2
3	.106	1.36	.316	4.25	.578	42.2
<u>Calf No.</u>						
1	.386	1.31	.284	4.35	.330	20.3
2	.143	1.13	.523	3.62	.334	25.70

<u>Intravenous Dose</u>						
Cow No.	<u>Pool 1</u>		<u>Pool 2</u>		<u>Pool 3</u>	
	A ₁	T _{1/2}	A ₂	T _{1/2}	A ₃	T _{1/2}
1	.298	1.31	.297	4.82	.404	34.2
2	.310	1.50	.252	5.78	.437	34.8
3	.151	1.46	.220	6.73	.629	46.3
<u>Calf No.</u>						
1	.392	1.50	.385	5.32	.222	29.0
2	.368	1.63	.360	6.22	.272	32.2

* Amount of injected dose removed by process having the half-life indicated, expressed as a fraction of the injected dose at day 0.

Table 2

Cesium-137 Retention Equations in Cattle and Calves Following Oral and Intravenous Administration.

Cow 1

$$\text{oral} \quad A^* = .287e^{-.518} + .332e^{-.150} + .380e^{-.0224}$$

$$\text{I.V.} \quad A = .298e^{-.530} + .297e^{-.144} + .330e^{-.0203}$$

Cow 2

$$\text{oral} \quad A = .166e^{-.529} + .327e^{-.152} + .480e^{-.0215}$$

$$\text{I.V.} \quad A = .310e^{-.463} + .252e^{-.120} + .437e^{-.0199}$$

Cow 3

$$\text{oral} \quad A = .106e^{-.511} + .316e^{-.163} + .578e^{-.0164}$$

$$\text{I.V.} \quad A = .151e^{-.476} + .220e^{-.103} + .629e^{-.0150}$$

Calf 1

$$\text{oral} \quad A = .386e^{-.530} + .284e^{-.159} + .330e^{-.0342}$$

$$\text{I.V.} \quad A = .392e^{-.463} + .385e^{-.130} + .222e^{-.0239}$$

Calf 2

$$\text{oral} \quad A = .143e^{-.615} + .523e^{-.191} + .334e^{-.0270}$$

$$\text{I.V.} \quad A = .368e^{-.426} + .360e^{-.111} + .272e^{-.0215}$$

* A = fraction of dose retained at day (t).

Table 3

Half-time of Removal for Orally and Intravenously Administered Cs-137 in Cattle and Calves.

<u>Animal</u>	<u>Oral</u> (days)	<u>I.V.</u> (days)
cow 1	7	5.5
cow 2	8.5	7
cow 3	14	19
calf 1	3.5	4
calf 2	4.5	6.5

Table 4

Retention of Intravenously Injected Cesium-137 in Sheep.

<u>Sheep No.</u>	<u>1</u>	<u>2</u>	<u>3</u>	<u>4</u>	<u>Average</u>
*A ₁	0.24	0.28	0.25	0.34	0.28
T _{1/2} (days)	28	24	29	28	27
A ₂	0.34	0.33	0.34	0.32	0.33
T _{1/2} (days)	5.3	5.0	4.6	5.3	5.0
A ₃	0.42	0.39	0.41	0.34	0.39
T _{1/2} (days)	1.5	1.0	1.0	1.2	1.2
**B _{T 1/2} (days)	6.5	5.5	5.8	4.8	5.6

* Fraction of total dose removed by processes with half-life indicated.

** Time required for one-half of body burden to be excreted.

Figure 1. Cs-137 Retention in Calf 1.

Figure 2. Cs-137 Retention in Sheep 1

Fig. 3.

GOAT #1 - POOL SIZE AND HALF-LIFE, POOL 1

Figure
GOAT #4 - POOL SIZE AND HALF-LIFE, POOL 1
X

Figure 5. 1
GOAT #6 - POOL SIZE AND HALF-LIFE, POOL 1

Fig 6.
GOAT #7 - POOL SIZE AND HALF-LIFE, POOL 1

Fig 7.

GOAT #14 - POOL SIZE AND HALF-LIFE, POOL 1

Figure 8. Plasma Ca-47 Specific Activity vs. Time; Coast 6, Experiment 7