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ABSORPTION OF PLUTONIUM FED CHRONICALLY TO RATS

PART I

EFFECT OF SEX ON FRACTION ABSORBED AFTER HIGH DOSAGE LEVELS

MARCH 13, 1951

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ABSORPTION OF PLUTONIUM FED CHRONICALLY TO RATS
PART 1.
EFFECT OF SEX ON FRACTION ABSORBED AFTER HIGH DOSAGE LEVELS

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March 13, 1951

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ABSTRACT

Preliminary investigations undertaken to determine the percentage absorption from the rat's gastro-intestinal tract of plutonium administered chronically and to find whether the sex of the animal plays a role in the rate of absorption are described. Five male and five female rats each received an average of 810,000 d/m (5.9 μg) of Pu^{239} (+4) as the nitrate, per os, over a period of 90 days. Radiochemical analyses indicated that an average of 0.0095 per cent of the administered dose was deposited in the total carcass. The pelt contained negligible amounts, but the gastro-intestinal tract and contents retained considerable quantities of plutonium even after a fast of 3-5 days. There was no evidence of any sex difference in plutonium retention.

INTRODUCTION

The preliminary investigation reported here was undertaken to determine the percentage of absorption of plutonium into the body under certain conditions. We sought to find (a) the extent of absorption of plutonium from the gastro-intestinal tract of rats subjected to prolonged oral administration of this radio-element, and (b) whether sex of the experimental rats plays a role in the absorption of plutonium present in drinking water. The present work, which will serve as a guide for future experiments, was deemed essential in order to establish within narrower limits than those already published in current literature the absorption of plutonium from the gastro-intestinal tract. A search of the literature (1) had established the fact that no data were available on whether the gastro-intestinal absorption of plutonium was influenced by the sex of the experimental animal. Such a sex differentiation possibly would be useful in future gastro-intestinal absorption studies.

In order to deposit detectable quantities of Pu^{239} in the rats during a 90-day feeding period, the level of the radioelement fed in the present investigation was much higher than that currently accepted as the maximum permissible for drinking water. In future long-term feeding experiments, Pu^{238}, which has a specific activity about 260 times greater than that of Pu^{239}, will be administered at much lower mass levels in order to determine whether the percentage of plutonium absorbed from the gastro-intestinal tract is a function of mass and concentration.

EXPERIMENTAL

Fourteen young adult Sprague-Dawley albino rats, seven males and seven females, were used in this experiment. All animals were housed in individual cages and were maintained on a standard diet of Purina dog chow pellets, fresh lettuce, and carrots. Water was supplied ad libitum by means of individual water-bottles. Half-inch mesh wire floors in the cages allowed excreta to drop into pans lined with paper which was changed twice weekly and discarded with contaminated wastes. Five male and five female rats each received a total of about 810,000 d/m (5.9 μg) of Pu^{239} during the experimental period of 90 days. Two male and two female rats were selected as controls.

A solution of Pu^{239} nitrate at pH 2 containing from 80 to 90 per cent of the radioisotope in the tetravalent state, with the remainder hexavalent, was used. Each experimental animal received 0.4 ml (400 lambda) of the plutonium solution (0.065 μg) daily by gavage for 90 consecutive days. Similarly, each control rat received 0.4 ml of water daily per os.

The equipment utilized to administer the solutions to the rats consisted of a 1 ml tuberculin syringe attached to a 0.4 ml pipette by means of a rubber sleeve and a rubber catheter (French # 8) approximately 16 cm long. One gavage unit was used for administering the plutonium to all the rats. Another gavage unit was used to administer the water to all the control animals.

In order to determine the amount of plutonium administered to the rats and also to determine the uniformity of volume of solution of plutonium expelled by above method, after the solution was administered to every two rats, one equal volume, (0.4 ml) was delivered onto a flamed counting plate. Five plates were thus prepared daily, allowed to air-dry overnight, and counted after being flamed again. Reliability of counting technique was checked by preparing two reference plates which were counted daily along with plates described above. To minimize the amount of plutonium which might adhere to rubber and glass, the lumina of the pipettes and rubber catheters were coated with a silicone preparation (GE "Drifilm" 9999).

At the end of 90 days the rats were fasted for three to five days and then sacrified by a sharp blow on the head. The pelt, gastro-intestinal tract, and carcass of each rat were separated, weighed, and analyzed. Tissues were digested in new beakers with concentrated nitric acid. After partial evaporation, the mixtures were transferred to 250 ml Vycor beakers and then muffled at 600°C. to a white ash. The ash was dissolved in nitric acid and the radiochemical analysis for plutonium was completed by following the standard procedure now in use in our laboratory (2). Complete details of this method are described by Case and Miller (3). Final solutions were spread on 1-1/2 inch flamed stainless steel plates, dried under an infrared heat lamp, and flamed again. Counting was done in parallel-plate standard alpha counters operating at the 90 per cent confidence level and with a 52 per cent geometry. Recoveries

on rat carcasses spiked with plutonium nitrate before ashing and analyzed by this method gave an average recovery of 96.50 per cent with a standard deviation of 4.50 per cent. Spiked rat GI tracts and rat pelts gave recoveries of 92.90 (standard deviation of 1.34) and 74.78 (standard deviation of 20.06) per cent, respectively.

RESULTS AND DISCUSSION

Summaries of results are presented in Tables 1 and 2. A statistical analysis of our data (4) reveals that the average amount of plutonium retained in the carcasses of the 10 experimental animals was 77.1 d/m ± 39.8 d/m (5.6×10^{-4} μg) at the 99 per cent confidence level. Since a total of 810,000 d/m (5.9 μg) had been administered during the 90 days, the average gastro-intestinal absorption was at least 0.0095 per cent of the amount fed. In spite of a fasting period of 3 to 5 days, considerable quantities of plutonium were found in the lumen of the gastro-intestinal tract. A major portion of the radioelement was probably either adsorbed on the gastro-intestinal mucosa or on solid particles present in the stomach or intestines. The plutonium content of the pelt was negligible and may have partly originated from contact with excreta. In calculating percentages of plutonium actually absorbed, only the amounts present in the carcasses were used. No correction was made for the amount of plutonium which was absorbed from the gastro-intestinal tract and which was excreted in the urine during the experimental period. Our coefficient of absorption is in close agreement with the value found by Carritt, et al (5) although we fed a total of 5.9 μg over a period of 90 days, while Carritt administered 488 μg in only 5 days.

The experimental error which included any actual differences in plutonium retention among the rats was ± 96.7 d/m for a single rat carcass (99 per cent level of confidence). The observed difference between male and female rats with respect to the plutonium found in the carcass (88.0 and 66.1 d/m, respectively) is well within the experimental error. This means that there is insufficient evidence of any real difference in plutonium retention between male and female rats. There was no definite relationship between weight of rat and plutonium retention.

TABLE 1

RADIOCHEMICAL ANALYSES - TISSUE CONTENTS OF EXPERIMENTAL RATS

Rat No.	Sex	Carcass d/m	Carcass % Total Administered*	Skin d/m	Skin % Total Administered*	GI Tract Plus Contents d/m	GI Tract Plus Contents % Total Administered*	Whole Rat d/m	Whole Rat % Total Administered*
1	F	60.8 ± 3.5	7.5	2.3 ± 0.7	0.3	35.8 ± 2.7	4.4	98.9	12.2
3	F	101.0 ± 4.2	12.5	1.9 ± 0.7	0.2	27.5 ± 2.3	3.4	130.4	16.1
5	F	58.8 ± 3.5	7.3	3.3 ± 1.0	0.4	334.0 ± 9.2	41.2	396.1	48.9
7	F	65.6 ± 3.5	8.1	2.4 ± 0.8	0.3	5.5 ± 1.2	0.7	73.5	9.1
9	F	44.4 ± 2.9	5.5	0.6 ± 0.6	0.1	25.0 ± 2.3	3.1	70.0	8.7
Averages		66.1	8.2	2.1	0.3	85.6	10.6	153.8	19.0
2	M	60.5 ± 3.5	7.5	3.1 ± 0.9	0.4	13.7 ± 1.7	1.7	77.3	9.6
4	M	92.7 ± 3.8	11.5	2.7 ± 0.9	0.3	4.9 ± 1.1	0.6	100.3	12.4
6	M	122.2 ± 4.6	15.1	6.3 ± 1.3	0.8	305.7 ± 8.7	37.8	434.2	53.7
8	M	81.2 ± 3.7	10.0	1.9 ± 0.8	0.2	2.6 ± 1.0	0.3	85.7	10.5
10	M	83.6 ± 3.8	10.3	2.6 ± 0.8	0.3	3.1 ± 1.0	0.4	89.3	11.0
Averages		88.0	10.9	3.3	0.4	66.0	8.2	157.4	19.4

* Per cent figures have been multiplied by 10^3.

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TABLE 2

RADIOCHEMICAL ANALYSES -- CONTROL RATS

Rat No.	Sex	d/m per Total Tissue: Carcass	Skin	GI Tract Plus Contents	Whole Rat
A	F	2.50 ± 0.87	1.31 ± 0.67	---	3.81
B	F	0.62 ± 0.58	0.87 ± 0.87	0.06 ± 0.67	1.55
Averages		1.56	1.09	0.06	2.68
C	M	3.00 ± 0.86	0.44 ± 0.44	0.00	3.44
D	M	0.38 ± 0.62	0.06 ± 0.50	---	0.44
Averages		1.69	0.25	0.00	1.94

The random error in counting was normal and in control, as confirmed by the variation between triplicate half-hour counts made on each carcass. This is substantial evidence that the counting data from carcasses were reliable. With respect to the counting of the plates which were prepared to determine dosage of plutonium which was administered, it was calculated that the best available estimate of the reproducibility of a single dose is ± 343 d/m. The best estimate of the reliability of the total 90 doses is ± 3272 d/m.

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