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Pu Contamination in Snakes Inhabiting the  
Rocky Flats Plant Site

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and

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INTRODUCTION

Initially plutonium appears not to be a significant environmental hazard due to its low solubility and its few penetrating radiations. Existing knowledge of plutonium cycling in terrestrial ecosystems makes it difficult to answer questions pertaining to the hazards of long-term buildup, availability, transport mechanisms, and inhalation (Wh74).

Numerable studies have been reported in the past 15 years that have examined the physiological and toxicological properties of <sup>239</sup>Pu (Je72; Fo62; Co62; Ba61; La59). Although radioecological studies investigating the biological availability of plutonium contamination to indigenous species of plants and animals have also been reported (Wh75; Ha72; Ro70; Pa68; Pe64; Ol63), insufficient data are available to predict the fate and effects of this element in highly mobile carnivores in a terrestrial environment.

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For approximately four years studies have been underway at the Energy Research and Development Administration's (ERDA) Rocky Flats plant to determine contamination patterns and concentrations of plutonium in the biota (Wh75; Wh74). Contamination of the Rocky Flats environs has resulted from at least three incidents, a September 1957 fire (Hr71), a May 1969 fire (Ma70), and leaking barrels containing plutonium-laden cutting oil. The latter incident was considered by far the major source of the plutonium contamination (Kr70). This study was conducted to determine whether snake tissues of the area contained detectable amounts of  $^{239}\text{Pu}$  and, if so, at what concentrations.

#### MATERIALS AND METHODS

The Rocky Flats installation, located approximately 12 km (7.5 mi) northwest of Denver, Colorado, consists of habitat described as disturbed grassland. The study area covered approximately 21 ha and is immediately downwind from the area previously used for storage of barrels holding plutonium-laden cutting oil. This particular area was selected for several reasons: reasonably high soil plutonium concentrations, ( $> 2000 \text{ mCi}/\text{km}^2$ ), a relatively undisturbed vegetative community, continuing work on small mammal populations, the presence of several snake species, and the accessibility of area (Wh75).

Eastern yellow-bellied racers (Coluber constrictor flaviventris, n=3), bullsnakes (Pituophis melanoleucus sayi, n=2), and prairie rattlesnakes (Crotalus viridis viridis, n=3) were collected for  $^{239}\text{Pu}$  bioassay of three tissues. Snakes were captured using drift fences terminating in funnel traps and by opportunistic sampling. Lung (including trachea), liver, and bone (vertebrae) samples were taken from each of the nine specimens collected at Rocky Flats.

Lungs were selected as a tissue of interest because of their propensity to become contaminated through inhalation of resuspended soil particles

containing Pu. Previous work has suggested inhalation to be the principal potential route of entry of Pu into the body (La59). The snake's respiratory apparatus is comprised of a number of parts, the most important of which may be the glottis. This tube projects from the snake's mouth while prey is ingested, enabling the snake to breath and eat simultaneously. While the snake is feeding, the glottis is in close proximity to the food (e.g., adjacent to pelage of small mammals) increasing the possibility of the snake inhaling  $^{239}\text{Pu}$  particles.

Experimental data suggest that for  $\text{PuO}_2$  particles probably about one percent of the originally inhaled amount is absorbed through the lung and into the bloodstream with deposition primarily in bone and liver (Ca47; Ba58; La59; Ba61; Je72). Bone is usually considered the critical organ since it accumulates the majority of Pu and retains it essentially throughout an individual's lifetime (La59; Pe64; Je72). In fact, slower entry of Pu into the bloodstream (e.g., via absorption from the gastrointestinal tract and lungs) may give greater deposition in bone and less in liver (Ca47).

Pu assay was performed by LFE Environmental Analysis Laboratories whose procedure consisted of ashing, total dissolution, electrodeposition, and alpha spectrometry (We75; We71). Minimum detectable activity was 0.04 dpm/sample.

#### RESULTS

Concentrations (dpm/g) in tissues from the nine snakes collected at Rocky Flats are presented in Table 1. In 20 of 27 tissues sampled the activity was  $\leq 0.1$  dpm/g, and in no case did the concentration of any tissue analyzed exceed 1.0 dpm/g. No significant correlations were found between the distance the snake was caught from the presumed source of Pu contamination and the concentrations in the liver ( $r < 0.1$ ,  $df = 7$ ) or bone ( $r < 0.01$ ,  $df = 7$ ).

There was no detectable activity in eight of nine lung samples. Activities in the liver samples ranged from 0.01 to 0.44 dpm/g, while skeletal sample activities ranged from nondetectable (calculated as minimum detectable activity) to 0.56 dpm/g. Liver samples from each of two bullsnakes caught approximately 96 km (60 mi) north of the Rocky Flats site showed no detectable amounts of  $^{239}\text{Pu}$ . Although racers appeared to exhibit greater liver and bone concentrations than did either the bullsnakes or the rattlesnakes, a Kruskal-Wallis test showed no statistically significant difference between species with respect to either liver burdens ( $T = 3.28$ ;  $n = 2,3,4$ ;  $\alpha = 0.102$ ) or bone concentrations ( $T = 1.40$ ;  $n = 2,3,4$ ;  $\alpha = 0.102$ ) (Con71). This particular nonparametric test was performed because of the non-normal distribution suggested by the snake tissue concentrations.

Coefficients of variation (C.V.) of 156 percent and 176 percent (i.e., 1.56 and 1.76) were calculated for liver and bone tissues, respectively. This represents some of the lowest variability within biotic components of this study area at Rocky Flats (Wh74).

#### DISCUSSION

Snakes captured in the Rocky Flats area show detectable concentrations of  $^{239}\text{Pu}$ , primarily in the liver and bone tissues. No detectable activity was observed in the majority of the lung samples. Lungs have been considered important in assessing the biological hazards of  $^{239}\text{Pu}$  in the environment because of the chance of particle retention and absorption following inhalation (Ro72). The fate of inhaled  $\text{PuO}_2$  (probably the predominant chemical form of environmental plutonium) may be affected by any of several variables, the most important of which seems to be particle size. A reduction in particle size has been shown to result in a corresponding reduction in deposition. It is generally accepted that larger Pu particles, when deposited, are probably

cleared from the lungs and respiratory tract more readily than smaller particles (Ba62). Therefore, the fact that eight of the nine lung samples showed no detectable amounts of  $^{239}\text{Pu}$  may be explained either by the reduced deposition of Pu particles in the lungs (corresponding to a smaller particle size) or by the clearance of larger Pu particles that have been deposited. A third conceivable explanation of the non-detectable lung burdens may be the small mass of each lung sample. Present analytical procedures generally suggest compositing samples, especially those of low mass, to reduce the possibility of drawing erroneous conclusions (We71; We75). However, financial constraints restricted the sample size and, therefore, samples were treated individually rather than as a composite in this study.

Regardless of the concentrations, 17 of the 18 liver and bone samples showed detectable amounts of  $^{239}\text{Pu}$ . There are several ways that contamination of internal tissues can result. Absorption of  $^{239}\text{Pu}$  through the gut wall into the bloodstream is one possibility, however it is generally believed that this is not a major route of internal contamination. Gradual translocation of  $^{239}\text{PuO}_2$  has been observed from the lungs to other tissues, principally skeleton, muscle, liver, and spleen. Maximum translocation appears to be particle size dependent, increasing as smaller diameter particles are inhaled and deposited (Ba61; Ba62). The lack of any significant correlation between distance from the presumed source of contamination and  $^{239}\text{Pu}$  concentrations in the liver or bone may be partly due to the transiency of certain species of snakes. Low tissue concentrations may also be explained by the extensive movement patterns of some snakes (St47; Bar69; Hi69; Fi71). Small mammals which are important prey species of snakes, occupy smaller home ranges and exhibit higher tissue concentrations than snakes captured on the same site (T. Winsor, pers. comm.). Conversely, snakes, a very mobile predatory group with broader home ranges,

show some of the lowest  $^{239}\text{Pu}$  tissue concentrations of any Rocky Flats biota analyzed. Assuming that snakes do not feed exclusively in the more highly contaminated area of Rocky Flats, we would expect to see lower tissue concentrations. This phenomenon of decreasing tissue concentrations with increasing trophic position (i.e., biodiminution) may simply result from greater movement of animals occupying upper trophic levels.

Due to the high degree of variability in the data, interpretation of the tissue concentrations is complicated. Despite the fact that the coefficients of variation (C.V.) of snake tissues are relatively small when compared to those of other biological components of the same area, C.V.'s of 156 percent and 176 percent show considerable variability within each tissue and should not be regarded as insignificant.

From this preliminary data it appears that snakes are not an important organism in the redistribution of  $^{239}\text{Pu}$ . Insufficient data exists to draw valid conclusions concerning the mechanisms involved in the uptake of  $^{239}\text{Pu}$  by snakes. Further emphasis must be given to uptake and transport of transuranic nuclides from contaminated areas by vagile species to determine whether transient organisms constitute a major hazard in the redistribution of radionuclides in man's environment.

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Table 1. Mean  $^{239}\text{Pu}$  activities ( $\pm$  standard deviation) in tissues of nine snakes collected at Rocky Flats.

Species	Tissue (dpm/g)			Mean (all tissues)
	Lung	Liver	Bone	
Eastern yellow-bellied racer ( <i>Coluber constrictor flaviventris</i> )	< 0.07 < 0.11 < 0.15	0.12 $\pm$ 0.03 0.25 $\pm$ 0.05 0.44 $\pm$ 0.08	0.01 $\pm$ 0.01 0.34 $\pm$ 0.05 0.56 $\pm$ 0.07	< 0.07 < 0.23 < 0.38
mean	< 0.11	0.27	0.30	< 0.23
Bullsnake ( <i>Pituophis melanoleucus sayi</i> )	< 0.02 < 0.03	0.03 $\pm$ 0.02 0.03 $\pm$ 0.02	0.05 $\pm$ 0.01 0.06 $\pm$ 0.01	< 0.03 < 0.04
mean	< 0.02	0.03	0.05	< 0.03
Prairie rattlesnake ( <i>Crotalus viridis viridis</i> )	0.19 $\pm$ 0.09 < 0.05 < 0.02 < 0.07	0.31 $\pm$ 0.03 0.01 $\pm$ 0.00 0.02 $\pm$ 0.01 0.03 $\pm$ 0.02	0.05 $\pm$ 0.02 0.04 $\pm$ 0.02 < 0.01 0.03 $\pm$ 0.02	0.18 < 0.03 < 0.02 < 0.04
mean	< 0.08	0.09	< 0.04	< 0.07
Tissue means (all samples)	< 0.07	0.13	< 0.13	< 0.11