

ENVIRONMENTAL PLUTONIUM ON THE NEVADA TEST SITE AND ENVIRONS

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NEVADA APPLIED ECOLOGY GROUP
UNITED STATES
ENERGY RESEARCH AND DEVELOPMENT ADMINISTRATION
LAS VEGAS, NEVADA

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ENVIRONMENTAL PLUTONIUM ON THE NEVADA TEST SITE AND ENVIRONS

JUNE 1977



EDITED BY

**M. G. White, P. B. Dunaway,
And W. A. Howard**

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LAS VEGAS, NEVADA**

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PREFACE

PREFACE

The Nevada Applied Ecology Group (NAEG) annual information conference held in Las Vegas, Nevada, in February, 1976, resulted in some interesting reports. This publication is a compilation of research reports presented at that meeting, as well as those presentations concerning support to the group field studies at the Nevada Test Site.

The objectives of the NAEG are restated here:

1. Delineate locations of contamination.
2. Determine concentrations in ecosystem components.
3. Quantify rates of movement among ecosystem components.
4. Evaluate radiological hazards of plutonium.
5. Identify areas which need to be cleaned up or treated.
6. Develop techniques for cleanup or treatment.

Work in the NAEG safety-shot intensive study sites is nearing final stages; the planning for NAEG studies in the nuclear event sites at the Nevada Test Site is progressing to an early field operations beginning. Synthesis and evaluation of the data generated during the 3 years of safety-shot plutonium studies has been accelerated in recent months so as to provide guidance for the nuclear site projects. Several authors of papers in this publication have made efforts at synthesis of data under their respective investigative responsibility.

The cover of this report is a view from Rainier Mesa (6,000 feet elevation) at the Nevada Test Site overlooking a playa, Groom Lake. Pahute Mesa, Buckboard Mesa, and Rainier Mesa are NTS Great Basin desert environment locations under study by the Nevada Applied Ecology Group.

We should like to thank the following persons for continued support and encouragement: G. C. Facer, DMA/HQ; M. E. Gates (Manager), R. Ray, and R. L. Hitechew, of ERDA/NV. A special note of appreciation goes to A. E. Bicker, D. L. Wireman, D. N. Brady, L. M. Rakow, the NAEG field crew, and laboratory personnel, Reynolds Electrical & Engineering Co., Inc. (REECo); to H. B. Gayle, P. G. Noblitt, Timothy M. Catt, and the Word Processing Center of Holmes & Narver, Inc., Las Vegas; and to Willie B. Morris and Ruby A. Ramstad, secretaries, Bioenvironmental Sciences Division, ERDA/NV.

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SOILS

PLUTONIUM DISTRIBUTION IN A DESERT PAVEMENT-DESERT MOUND

SOIL SYSTEM IN AREA 11

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ABSTRACT

This interim report presents data on plutonium distribution in a desert pavement desert-mound soil system in Area 11 of the Nevada Test Site. Total plutonium soil concentrations with depth, plutonium concentrations in the different particle size classes, and preliminary information on plutonium distribution as a function of mineral density fractions are included.

Analyses of plutonium in the desert-pavement sample showed that 95% of the activity is in the surface layer (0 - 2.5 cm). Evidence of plutonium migration downward into the soil profile is suggested by higher contributions to the plutonium concentration in deeper layers by the silt and clay size particles. In the desert-mound sample, 60% of the activity is in the surface layer. Significant amounts of plutonium occur down to the 7.5-cm depth, but there is a possibility that this depth distribution is due to the method of sampling.

The soil particle size classes bearing the highest plutonium activity are the coarse silt (53 - 20 μm) in the desert pavement and the medium silt (20 - 5 μm) in the desert mound. The high activity associated with soil particles less than 20- μm particle size in the desert mound sample suggests that the fine-particle activity is from a source close to ground zero. Density gradient segregation of the silt fractions in the surface layer showed that the highest activity is in the heaviest fraction (greater than 2.9 g/cm³ density); contributions to the activity in the heaviest fraction decrease with finer particle sizes.

INTRODUCTION

A common feature in the desert landscape of the Nevada Test Site (NTS) is the occurrence of sandy desert mounds formed under desert shrubbery. The mounds are formed by the filtering action of the vegetation in intercepting saltation and creep particles. In contrast to the desert mound, the adjacent bare area consists of desert pavement soils, frequently with a gravelly surface texture.

These coexisting areas show morphologic and topographic differences; and in the plutonium-contaminated areas, the plutonium-activity levels, as measured by field survey instruments (FIDLER), are higher in the desert mound than in the contiguous desert pavement.

Since these mounds serve not only as the "host" soil for the desert shrubbery but as sites for other biological activity, it is important that the character and behavior of the plutonium in these two related soil systems be investigated and understood. The objective of this study is to define the differences in characteristics of the plutonium in these two related soil systems. Included in this report are data on plutonium concentration as a function of depth, particle size, and particle density on two soil samples from a desert mound in Area 11 and a desert pavement sample 5 ft from the mound.

MATERIALS AND METHODS

The samples were collected by personnel of the Reynolds Electrical and Engineering Company (REECO) in June 1975. Two samples were taken in Area 11; both samples were in a northwesterly direction from ground zero (GZ) of "C" site. One desert pavement--desert mound sample was located 102.4 ft (31.2 meters) from GZ, and the other was 30.7 ft (9.4 meters) from GZ. This report covers results obtained to date on the sample located 30.7 ft from GZ. The samples were taken in slices representing 2.5-cm depth increments, and four vertical depth samples were taken down to 10 cm. The samples were placed in double plastic bags without further treatment and placed in 1-gal aluminum cans which were sealed for shipment.

Upon arrival, the samples were placed in glove boxes, opened, air-dried, and the samples passed through a 2-mm sieve to determine the gravel content. Aliquots of the less than 2-mm size fraction were taken for total plutonium analysis as well as for particle size segregation. Plutonium was analyzed by the HASL-LASL technique using HF-HNO₃ digestion, with ²³⁶Pu as internal tracer for recovery efficiency correction (Tamura, 1975a). Sample size for analysis was 10 g for the unsegregated soil; for segregated size fractions, 10 g were used where sample size permitted; otherwise, one-half of the sample was used for plutonium analysis.

Particle size segregation to obtain sand, silt, and clay fractions was performed as described earlier (Tamura, 1975a). No dispersing agent or mechanical dispersive technique was applied prior to segregation; hence, the reported particle size reflects "existing" sizes rather than ultimate particle sizes as normally determined.

Density gradient segregation, when performed on selected size fractions, utilized gradient solutions ranging from 1.8 to 2.90 g/cm³. Tetrabromoethane (TBE) was the heavy density solution; ethanol containing 10% polyvinylpyrrolidone (PVP) was the dispersant, and mixed with appropriate amount of TBE was the light solution. The samples were treated for 2 minutes with ultrasonic vibration to induce mineral disaggregation. For more detailed description of the technique, readers are referred to papers by Francis *et al.*, 1972a, and Francis and Tamura, 1972b.

RESULTS AND DISCUSSION

Plutonium Concentration in the Soils

The concentrations of plutonium in the less than 2-mm soil particles are given in Table 1. In the desert pavement, approximately 95% of the activity is found in the 0 - 2.5 cm surface layer. The remaining 5% is distributed in the lower horizons with indications of a higher concentration in the 7.5 - 10 cm layer than in the two intermediate layers. By comparison, the 0 - 2.5 cm surface layer of the desert mound contains about 60% of the activity with 30% and 7% contributions in the 2.5 - 5.0 and 5.0 - 7.5 cm layers, respectively. This distribution would suggest a sizable buildup of the mound subsequent to the detonation event. However, this interpretation regarding buildup should be reviewed with caution since the observed distribution may be a reflection of the sampling technique used.

In sampling the mound, the layers were separated at 2.5-cm increments; the 0 level was measured from the higher elevation point of the mound along the sampling slope. This point is illustrated in Figure 1, a schematic representation of a desert mound. The first increment of 2.5 cm then represents the depth measured from the highest point (0 level) on the mound. At lower sections of the mound, the depth of penetration would be less than 2.5 cm, depending on the slope. Sampling in this manner evidently resulted in a lower mass of sample from the surface layer since the gross weight of this sample was 233 grams in the 0 - 2.5 cm layer as compared with a mean weight of 315 ± 3.6 grams for the 2.5 increments of the three lower layers.

If one assumes that the buildup of the mound was relatively even, then sampling the mound perpendicular to the sloping surface (Fig. 1) rather than perpendicular to the ground level might better reflect the depth distribution. Assuming that 315 grams represent the mass of soil 10 cm x 10 cm x 2.5 cm (sampling dimensions), then 233 grams represent 74% of the mass based on 250 cm³. Thus, approximately 26% of the 0 - 2.5 cm layer may be present in the 2.5 - 5.0 cm layer. If this 26% contributed the same activity to the 2.5 - 5.0 cm layer as found in the surface layer, then the calculated activity in the remaining portion of the 2.5 - 5.0 cm layer would be 5250 dpm/g compared to the measured 7492 dpm/g. Similar calculations of the 5.0 - 7.5 cm layer show that the activity of this layer would be 392 dpm/g compared to the measured 1655 dpm/g; and the 7.5 - 10.0 cm layer would be 97 compared to 174 dpm/g. If the distribution occurred in the manner postulated above, then the calculated results of plutonium distribution would suggest that a small amount existed prior to detonation. It appears worthwhile to sample desert mounds by both methods as well as at different locations around the shrubbery to establish the activity distribution and activity buildup beneath the shrubbery.

Table 1. Plutonium concentration in two soils from Area 11 (activity in dpm/g of < 2 mm soil particles; $\pm$ value is one standard deviation)

Depth (cm)	Desert Pavement	Desert Mound
0 - 2.5	7,520 $\pm$ 1,976	13,875 $\pm$ 2,325
2.5 - 5.0	126 $\pm$ 107	7,492 $\pm$ 591
5.0 - 7.5	53 $\pm$ 55	1,655 $\pm$ 183
7.5 - 10.0	227 $\pm$ 108	174 $\pm$ 88

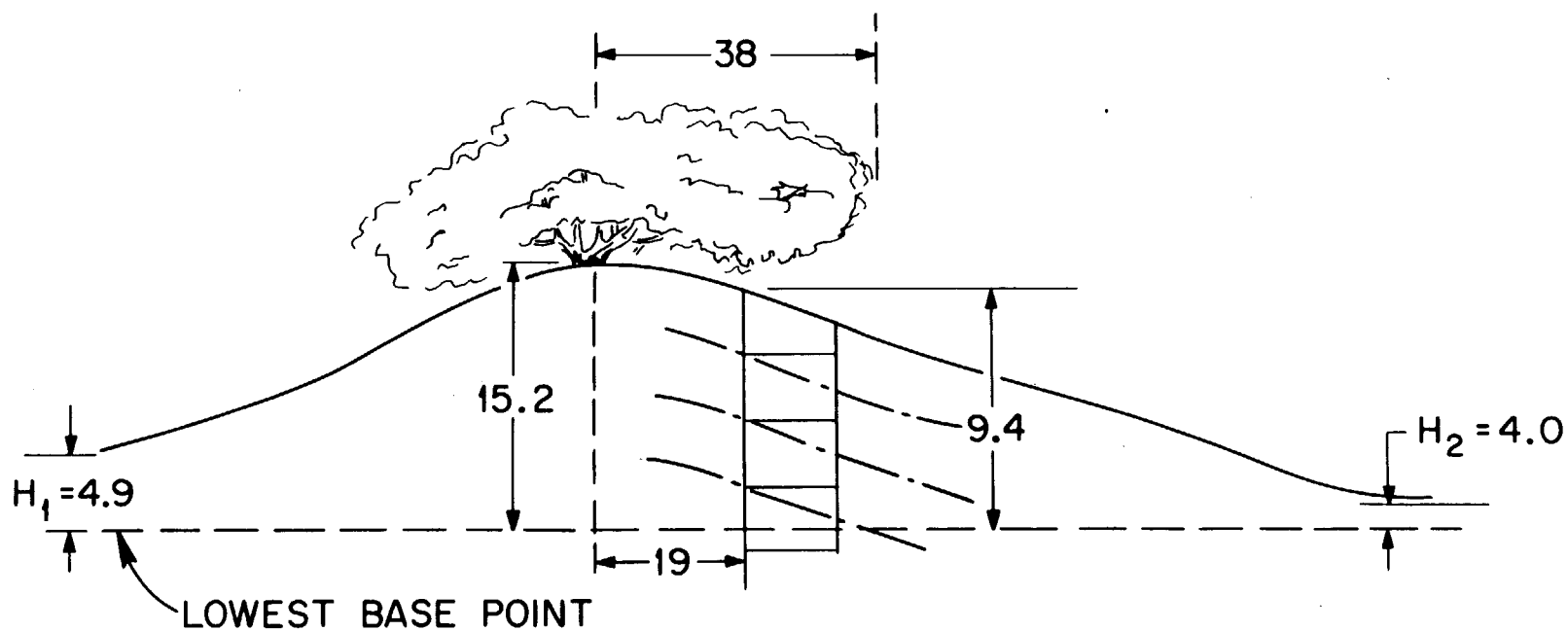


Fig. 1. Schematic representation of a desert mound. Samples were taken at middistance from stem of shrubbery and vertical to the lowest base point plane. Curve line suggests an alternative sampling mode. H_1 = highest point of mound base; H_2 = height of mound base at lowest point from sampled area of mound. Values are in centimeters.

Particle Size Distribution

Table 2 includes data on the particle size distribution of the desert pavement and desert mound samples. The high gravel content ($> 2000 \mu\text{m}$) in the surface of the desert pavement is typical of eroded surfaces in the desert. According to Leavitt (1974), the desert pavement soil possesses an A_2 horizon which extends down to 10 cm; thus, the lower three depth increments which show similar particle size distribution reflect the more normal A_2 horizon. Inspection of the data suggests that the finer silt and clay size ($< 20 \mu\text{m}$) particles were eroded from the surface preferentially as evidenced by the low content of these particle sizes in the 0 - 2.5 cm layer.

The desert mound, lacking gravel particles, shows a very high content of sands which dominate the texture throughout the samples. With depth, the sand content decreases from 93% at the surface to 77% in the 7.5 - 10.0 cm depth; the silt content increases from 4% to 15%; and the clay content increases from 0.5% to 3%. A comparison of the particle size distribution in the 7.5 - 10 cm sample from the desert mound with the size distribution in lower layers of the desert pavement suggests that the textural composition of the 7.5 - 10.0 cm sample is approaching that of the original residual soil.

Plutonium in Particle Sizes

Tables 3 and 4 include data on the plutonium content of the different particle sizes. The activity in each particle size fraction is presented in row A. In terms of activity per unit mass, the coarse silt ($53 - 20 \mu\text{m}$) contains the highest activity in the desert mound. Since the activity in the finer particles (less than $20 \mu\text{m}$) of the surface layer in the desert mound is higher than the corresponding particle sizes in the neighboring desert pavement, the data would suggest that the source of the plutonium associated with the finer sizes in the surface layer of the mound is not the adjacent pavement soil but a source closer to GZ.

Row B presents the plutonium concentrations in disintegrations per minute (dpm) per gram of soil of each particle size class. This contribution takes into account the mass contribution of each size class to the soil particles less than 2 mm. Row C is the percentage contribution of plutonium for each size class to the soil. Thus, in the desert pavement, the $125 - 53 \mu\text{m}$ size class contributes about one-third of the total soil activity as compared to 25% by the $53 - 20 \mu\text{m}$ size class, although the latter has a higher activity per unit mass. The $125 - 53 \mu\text{m}$ fraction contributes 16.28%* of the less than 2 mm soil, whereas the $53 - 20 \mu\text{m}$ fraction contributes 3.54%*. This mass difference compensates for the activity difference noted in Table 3. Similarly, in the desert mound, the $20 - 5 \text{ m}$ size class has the highest activity per unit mass (229, 400 dpm/g), but the small weight contribution (0.86%*) lowers this fraction's contribution to about 20%.

*Percentage values in Table 2 recalculated excluding gravel ($> 2 \text{ mm}$) particles.

Table 2. Particle size distribution of desert pavement and desert mound soils from Area 11 (results expressed in percentage by weight)

Size Range (μm)	Soil Depth (cm)							
	0 - 2.5		2.5 - 5.0		5.0 - 7.5		7.5 - 10.0	
	A*	B*	A	B	A	B	A	B
> 2000	18.40	0.81	2.33	1.19	2.98	1.59	5.58	3.69
2000-840	3.94	1.59	2.19	1.90	2.47	1.52	2.78	2.84
840-250	30.13	32.74	29.42	26.72	27.94	24.35	27.34	26.23
250-125	29.22	42.22	27.52	40.81	24.69	37.64	29.62	31.66
125- 53	13.75	16.60	13.22	21.51	12.68	22.47	13.79	16.09
53- 20	3.36	2.65	3.82	3.59	3.67	3.85	3.39	3.99
20- 5	2.37	0.86	10.93	1.72	11.09	3.12	11.10	6.08
5- 2	0.62	0.41	4.56	0.82	6.38	1.90	4.16	4.88
< 2	0.67	0.47	4.23	1.13	5.00	1.98	3.84	3.20
Total	102.46	98.35	98.52	99.39	96.90	98.42	101.60	98.66

*A refers to desert pavement; B refers to desert mound.

Table 3. Distribution of plutonium in size fractions and contribution of sizes to total soil plutonium in desert pavement

Sample Depth (cm)	Activity*	Size Range (μm)							
		2,000-840	840-250	250-125	125-53	53-20	20-5	5-2	< 2
0 - 2.5	A	2,007	3,017	3,945	22,295	76,271	58,338	30,099	13,143
	B	94	1,077	1,365	3,634	2,700	1,633	217	104
	C	0.8	10.0	12.6	33.6	24.9	15.1	2.0	1.0
2.5- 5.0	A	16	23	291	73	638	369	266	196
	B	0	7	82	10	25	41	12	8
	C	0	3.8	44.3	5.4	13.5	22.2	6.5	4.3
5.0- 7.5	A	7	3	33	38	15	78	72	172
	B	0	1	8	5	1	9	5	9
	C	0	2.6	21.0	13.2	2.6	23.7	13.2	23.7
7.5-10.0	A	7	20	23	78	725	711	215	640
	B	0	6	7	11	26	83	9	26
	C	0	3.6	4.2	6.6	15.5	49.4	5.4	15.5

*A = dpm/g in size fraction; B = contribution in dpm/g of soil, activity rounded off to nearest whole number; C = percentage contribution of activity in size fraction to soil.

Table 4. Distribution of plutonium in size fractions and contribution of sizes to total soil plutonium in desert mound

Sample Depth (cm)	Activity*	Size Range (μm)							
		2,000-840	840-250	250-125	125-53	53-20	20-5	5-2	< 2
0 - 2.5	A	10,523	4,003	1,221	21,653	74,058	229,401	56,653	29,221
	B	169	1,320	522	3,622	1,977	1,972	232	137
	C	1.7	13.3	5.2	36.4	19.9	19.8	2.3	1.4
2.5- 5.0	A	6,597	1,553	5,354	6,193	64,525	70,809	25,517	11,098
	B	127	419	2,211	1,350	2,368	1,232	212	126
	C	1.6	5.2	27.5	16.8	29.4	15.3	2.6	1.6
5.0- 7.5	A	1,069	541	851	1,117	15,496	16,484	4,834	2,547
	B	16	133	325	255	606	522	93	51
	C	0.8	6.6	16.2	12.7	30.3	26.1	4.6	2.6
7.5-10.0	A	11	24	51	74	1,042	898	699	144
	B	0	6	17	12	43	57	35	5
	C	0.1	3.4	9.7	6.9	24.6	32.6	20.0	2.9

*A = dpm/g in size fraction; B = contribution in dpm/g of soil, activity rounded off to nearest whole number; C = percentage contribution of activity in size fraction to soil.

The distribution of plutonium in the different particle size classes and in depth shows an interesting trend. Consider the desert pavement, for example, where the bulk of the activity in the first two depths is in the sand sizes and the activity in the second layer is about 2% of that in the surface layer. In the lower two layers, the silt and clay sizes contain the major fraction of the soil activity. In Table 1, the plutonium concentrations in the depth increments of the desert pavement suggest a possible accumulation in the 7.5 - 10.0 cm layer. The change in percentage contribution by the finer sizes in the lower depths also suggests downward migration of activity, providing further support for this hypothesis. The movement of plutonium downward into the lower soil horizons at the Nevada Test Site has also been observed by Essington *et al.* (1975), including several soils in Area 11.

The distribution of plutonium with depth in the desert mound sample is more difficult to explain. Unlike the desert pavement, which shows an abrupt decrease in activity below the surface layer, the desert mound shows a gradual decrease in activity with depth. Furthermore, unlike the desert pavement, which shows an abrupt change in particle size distribution above and below the 2.5-cm depth, the desert mound shows a gradual change in the particle size distribution with depth, with the deepest sample approaching a particle size distribution similar to that in the lower layers of the desert pavement. This gradual change in particle size distribution with depth in the desert mound suggests that considerable mixing of the original soil has occurred in the secondary deposit. Considering the closeness (30.7 feet) of the mound to GZ, the possible mechanical disturbance during preparation of the site, and the fact that these data are the result of a single sampling of one mound, further interpretations or speculations about the depth distribution of plutonium in Area 11 is not warranted.

Density Gradient Segregation and Plutonium Distribution

In order to further characterize the plutonium in the desert pavement and desert mound samples, selected size fractions were subjected to density gradient segregation. The technique separates minerals with different densities by allowing them to accumulate at their respective mineral density positions in the linear density gradient solution. To date, the three silt sizes of the surface layers have been separated in this manner. The results presented in Table 5 include the density range or bands in which the particles concentrated, the weight of the particles, and the percent activity in each of the bands. The activity measurements were obtained with a portable alpha survey meter placed over the dry powder; the activity percentages listed in the table, therefore, should be considered as approximate indications of the distribution and not as definitive values.

Results show that as the particles get smaller, their densities become lighter. This observation is consistent with mineral hydration (in these cases, the polyvinylpyrrolidone substitutes for water), which increases the effective diameter and reduces the particle density. There is an indication that the desert mound particles in the 20 - 5 and 5 - 2 μm size classes are lower in density than the corresponding size classes in the desert pavement. In the study of a desert pavement-desert mound system in Area 13 (600 feet from GZ) reported earlier (Tamura, 1975b), the 53 - 20 μm size particles of the two

Table 5. Density gradient segregation of the coarse, medium, and fine silt fractions of the 0- to 2.5-cm-depth samples of the desert pavement and desert mound soils

Band No.	Desert Pavement			Desert Mound		
	Density Range (g/cm ³)	Weight Fraction (%)	Activity (%)	Density Range (g/cm ³)	Weight Fraction (%)	Activity (%)
53 - 20 μ m						
1	< 1.8	0.3	0	< 1.8	1.4	2
2	1.98-2.14	2.8	2	1.93-2.04	2.7	3
3	2.26-2.32	12.1	2	2.20-2.30	15.6	8
4	2.46-2.54	13.0	3	2.45-2.52	9.9	2
5	2.56-2.62	43.0	2	2.54-2.61	45.4	6
6	> 2.9	18.3	90	> 2.9	19.0	80
20 - 5 μ m						
1	< 1.8	1.0	2	< 1.8	3.9	5
2	2.07-2.16	9.2	2	1.92-2.02	8.0	9
3	2.24-2.35	12.7	3	2.06-2.11	8.6	6
4	2.38-2.43	25.4	4	2.19-2.33	32.9	11
5	2.55-2.64	40.6	9	2.36-2.56	33.6	12
6	> 2.9	4.1	78	> 2.9	3.4	57
5 - 2 μ m						
1	< 1.8	2.3	6	< 1.8	8.1	7
2	2.10-2.20	14.9	15	1.94-1.99	8.9	10
3	2.20-2.26	21.4	14	2.08-2.17	15.7	10
4	2.26-2.39	21.5	13	2.17-2.27	39.7	24
5	2.48-2.58	32.9	17	2.27-2.36	22.4	23
6	> 2.9	1.9	36	> 2.9	0.8	26

soils (desert pavement and desert mound) showed a very different distribution of the plutonium. In the desert pavement of Area 13, the plutonium was concentrated in the greater than 2.90 g/cm² band, whereas in the desert mound it was concentrated in the 2.5 - 2.7 g/cm³ band. In the surface samples from Area 11, the distribution of plutonium in both soils occurs in the greater than 2.90 g/cm³ band. However, in the smaller size classes, the data suggest that plutonium is distributed in the lighter fractions as well as the heaviest fraction.

Implications of Findings on Plutonium Distribution

Evidence indicates that plutonium is moving downward into deeper horizons in the desert pavement soils. The association of plutonium with the finer particle sizes in the deeper horizons supports this hypothesis of downward movement. What is not established is (are) the mechanism(s) of movement. Density gradient segregation of mineral particles from the different size classes and depths may provide some insight into the mechanism(s).

The higher activity associated with the less than 20 μ m size particles in the desert mound as compared to the adjacent desert pavement indicates that the finer sizes of plutonium are derived from a source close to GZ. The distribution of mineral particle sizes with depth in the mound suggests considerable mixing of the secondary aeolian material with the original substrate. Important questions are raised about the mode of sampling desert mounds. Further studies are needed to establish the distributional pattern of plutonium in desert mounds.

It must be stressed that the results presented in this progress report are preliminary and subject to change with additional information presently being obtained. It is also expected that the results from these studies, compared and collated with those being obtained by other participants in the soils program, will help to evaluate further the situation existing in these NTS test areas.

FUTURE PLANS

The desert pavement-desert mound samples taken at 102 feet from GZ in Area 11 will be investigated in a manner similar to the sample reported herein. Results of studies on samples previously taken in Area 11 as part of the preliminary mound study (Brady, 1975) will be examined, and samples which show promise of providing further insight into the plutonium distribution in the desert mounds will be investigated.

Additional fractionation of the organic material from the inorganic fraction will be made of several desert mound samples. The separated organic fraction will be analyzed for plutonium to obtain the plutonium concentration, and studies will be made to compare differences in characteristics of the organic-versus inorganic-associated plutonium.

Depending on the findings of the samples from 30 and 102 feet from GZ in Area 11, another sample will be taken either between the two sampled distances or beyond 102 feet. The three samples, together with information already obtained from a pavement-mound sample from Area 13, should provide useful information on the accumulation of plutonium in desert mounds.

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SOILS ELEMENT HISTORY, SAMPLING, ANALYSES, AND RECOMMENDATIONS

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INTRODUCTION

The Soils Element of the Nevada Applied Ecology Group (NAEG) had its beginnings in 1970. The Soils Element contributed to the objectives of NAEG in such areas as soil sampling, processing, radiochemical analysis, referee, and special interpretive aspects regarding the distributions of plutonium and other radionuclides at the Nevada Test Site (NTS).

Since the inception of NAEG, with the exception of the sampling and sample preparation experimental techniques used in the small microplot experimental area, and the recent soil mound study, sampling and sample preparation techniques have remained the same for all of the safety-shot study areas.

The original missions were developed by various committees to meet their specific needs and hence were discipline oriented. In the absence of detailed guidance from the literature, missions were developed largely by intuition. The mission of the Soils Element (initially Soil Sampling and Analyses Committee) simply stated was "to develop methods for sampling and analyses of soils for the determination of plutonium content." Results were to be used in the calculation of an inventory characteristic of the study site.

Discussed in this report are the efforts of the Soils Element from the standpoint of (1) history and initial charges relative to soil sampling and analyses, (2) development of sampling and analytical procedures, (3) assigned referee activities, (4) special studies relating to particular sampling and analytical problems, and (5) recommendations for new or for completing past areas of soils study.

HISTORY

A chronology of past activities is presented as background to an understanding of the present status of the Soils Element studies.

The NAEG Soil Sampling and Analyses Committee was established in 1970. Data obtained relative to the concentration and distribution of plutonium (and later, other radionuclides of interest) were to provide basic source terms to be used by other elements in the interpretation of their results. Samples were predominately surface samples taken to 5 cm depth. However, results from some preliminary profile samples, taken at 2.5 cm increments, indicated the importance of plutonium at depths greater than 2.5 cm and a percentage of profile samples was factored into the sampling program. The committee devised general procedures for sampling, sample preparation, and analyses of soils. Based on the preliminary objectives, certain exploratory experiments were conducted as an aid to design and development of methods. One of the early experiments was designed to test sampling techniques and was conducted in a microplot. In early 1971, in conjunction with Dr. Evan Romney, Chairman of the Field Ecology Studies Committee, a series of microplots was selected near ground zero of Project 57, at Area 13. The soil sampling methods which had been proposed were tested on the microplots; also tested was the proposed method for sampling vegetation.

With respect to analyses, a number of methods for determination of plutonium, the radionuclide of prime interest, were available. However, a consideration of the possible number of samples and variety of matrices limited consideration of methods to those which would be rapid and inexpensive yet accurate and precise for the anticipated matrices. Methods from the literature and committee members' experiences were compared and in some cases tested on "standard" samples.

Two methods were chosen for final comparison by five cooperating laboratories. The two methods were those of Talvitie (1972) and what has since become known as the LASL-HASL digestion (Essington and Fowler, 1975). A definitive comparison of methods could not be made since in several instances, necessary information requested relative to man-hours and total cost per sample was not obtained. In some cases, only one method was tested or the cooperating laboratory used neither of the two methods. The exercise proved to be academic since contractors for analytical work preferred to use the method prevalent in their own laboratory and since subsequent cross calibrations proved results to be comparable with all methods used. However, based on results which were obtained from analysis of standard samples, the LASL-HASL digestion was the method of choice, and it was recommended by the Soil Sampling and Analyses Committee. The LASL does use the recommended method; it has been modified to accommodate various matrices (Fowler and Essington, 1975).

In July, 1971, the assistance of statisticians Dr. L. L. Eberhardt and Dr. R. O. Gilbert of Battelle Northwest, Hanford, was obtained by NAEG to provide statistical input to the experimental design for sampling of various matrices and to interpret results of analyses.

Some funding was made available for the Los Alamos Scientific Laboratory (LASL) activities in 1972 and in January, 1973, a full-time staff member joined the LASL efforts. The committee concluded that its initial assignment had been completed, and in June, 1972, a detailed discussion of the Soil Sampling and Analyses Committee's activities was submitted in writing to the Office of Effects Evaluation. The discussion served as a step-by-step rationale of committee recommendations and procedures and as an informational document to serve as a guide to the NAEG program (private communication, 1972).

In July, 1972, by a request from Nevada Operations Office (ERDA) to LASL, the Soil Sampling and Analyses Committee was changed to the Soils Element, retaining the chairman of the former as Manager of the new Element. (Editor's Note: In spring, 1976, it was determined that the most effective operational management unit of NAEG studies had been that of principal investigator, plus co-investigators as necessary, for specific NAEG environment studies. At that time, the "elements" of the Nevada Applied Ecology Group were dissolved, with notification to the former program element managers involved.)

Since that date, the Soils Element activities included (1) development of special sampling methods such as for mounds, (2) development of analytical methods, (3) quality assurance studies, (4) consultations relative to site selection, and definition of related missions and experimental design, (5) assistance to the statistician in the interpretation of data, (6) preparation of reports, including annual progress reports, (7) analysis of routine samples as a contributing laboratory, and (8) assignment as referee laboratory for NAEG soil samples.

SOIL SAMPLING AND ANALYSIS METHODS

The sampling methods used at NTS were developed as modifications of the trench method for sampling of profiles and of Alexander's "cookie cutter" method (Alexander *et al.*, 1960) for sampling surface soils. Both methods were successfully tested on the Area 13 microplot; they were applicable to sandy or loamy soils where stones or highly cemented soil horizons did not present a problem. More recently, areas other than Area 13 have been sampled using the above techniques with no difficulty.

Microplot Study

The microplot study was initiated to test (1) proposed surface sampling methods, (2) proposed profile sampling methods, (3) proposed field radiation measurement methods with the FIDLER instrument (FIDLER--field instrument for the detection of low-energy radiation), and (4) proposed methods for sampling matrices other than soils.

A microplot area 3.7 x 2.5 meters was selected at random approximately 250 m west of Area 13, Project 57 ground zero. The plot was covered by a structure to minimize movement of soil by wind into and out of the plot. Before soil samples were collected, shrubs were removed by cutting at ground level and the microtopography was mapped. The plot was laid out in a grid pattern and the level of radioactivity of each grid section was determined by readings taken with a FIDLER instrument. Three different FIDLER geometries were investigated: (1) 30.5 cm lead collimator, (2) 15.3 cm lead collimator, and (3) detector placed on a sampling ring at 2.5 cm above the soil surface. The collimator seriously degraded the spectrum and its use was discontinued for determination of inventories. At the time of the microplot study, the FIDLER instrument represented the state of the art for survey instruments. The FIDLER [and Ge(Li) spectrometer] data are based on the 60 keV ^{241}Am gamma emission. The ^{241}Am is the daughter product of ^{241}Pu present in plutonium nuclear fuels, and plutonium concentrations can be estimated from those data if the ^{241}Am to plutonium ratio in the sample is known.

Part of the mission for the microplot study was to determine whether a good correlation existed between FIDLER measurements and data obtained from radiochemical determinations and to determine whether FIDLER data could be used to calculate inventories.

Plutonium radiochemical analytical results typical of the area near the microplot were compared with their respective determinations made in situ by the FIDLER, and made by Ge(Li) spectrometry. Table 1 presents a comparison of those data obtained from the survey. The points to be noted relate to comparisons of activities in isopleth 2 with those in isopleth 6 and the ratios listed in the last two columns. Activities determined by radiochemistry were used as a basis for comparison. There is an apparent general increase in radioactivity among samples as determined by the FIDLER and Ge(Li) spectrometry, which is not necessarily true for the case of radiochemical analysis. If the FIDLER and Ge(Li) spectrometer were detecting the same source of radioactivity, their ratios to radiochemistry should be relatively constant. That condition is more nearly approached in the case of the Ge(Li) and radiochemistry determinations for higher activity samples. The lack of consistent agreement between FIDLER results and results from radiochemical analysis performed on the soil samples is partially due to the fact that the FIDLER senses radioactivity from an area about 1 m dia and about 1 cm deep, whereas Ge(Li) spectrometry and radiochemical analyses are performed on a limited sized sample.

In order for the FIDLER to serve as an instrument for the determination of inventory, the following requirements would have to be met:

1. The radioactivity must be located in a very thin layer at the surface or at a known, and constant, depth,
2. the radioactivity must be evenly distributed locally,
3. Americium-241 and plutonium must be equally distributed horizontally and vertically,
4. the ratio of ^{241}Am to plutonium must be known if ^{241}Am concentrations are to be used to estimate plutonium concentrations,
5. vegetation present in the area of measurement must have no influence on the radioactivity-detector geometry, and
6. the measurement area must not have quantities of radionuclides which would interfere with the 60 keV ^{241}Am gamma emission.

All of the above conditions are rarely met in practice.

The FIDLER could serve as a survey instrument to define isopleths but would not provide acceptable data for the determination of inventories of surface-deposited materials at the safety shot site areas. The FIDLER also can be used in some instances to indicate differences in activities of importance in grouping samples for analysis. The Ge(Li) spectrometer can be used to estimate plutonium where a specific sample to be analyzed is small, contains no interfering radioisotopes, has sufficiently high levels of ^{241}Am , and the ratio of ^{241}Am to plutonium is known; under many conditions, Ge(Li) spectrometry has been used successfully for the determination of ^{241}Am on samples from the safety shot sites.

Table 1. Comparison of Instrumental and Wet Chemical Analyses for Plutonium in Field Soils

Isopleth and Samp. No.	FIDLER 60 keV (Gross c/m)	Ge(L1) nCi/g (As $^{239-240}\text{Pu}$) *	Radiochemistry nCi/g ($^{239-240}\text{Pu}$)	Ratio Ge(L1) Radiochem.	Ratio FIDLER Radiochem. ($\times 10^4$)
Isopleth 2 #20	6000	.01	.1	.08	6.2
#26	8000	.02	.08	.22	9.5
Isopleth 6 #40	3500	2.5	2.5	.99	1.4
#21	100000	14.0	11.	1.2	.88

*60 keV ^{241}Am $\times 10$

Based on more recent knowledge, changing criteria, and further FIDLER development, the FIDLER may prove to be of greater value than was believed originally. For example, if a "level of no concern" is set within the detection range of the FIDLER, the number of samples subjected to radiochemistry could be substantially reduced if the FIDLER were used and hence there would be a resultant reduction in analytical costs.

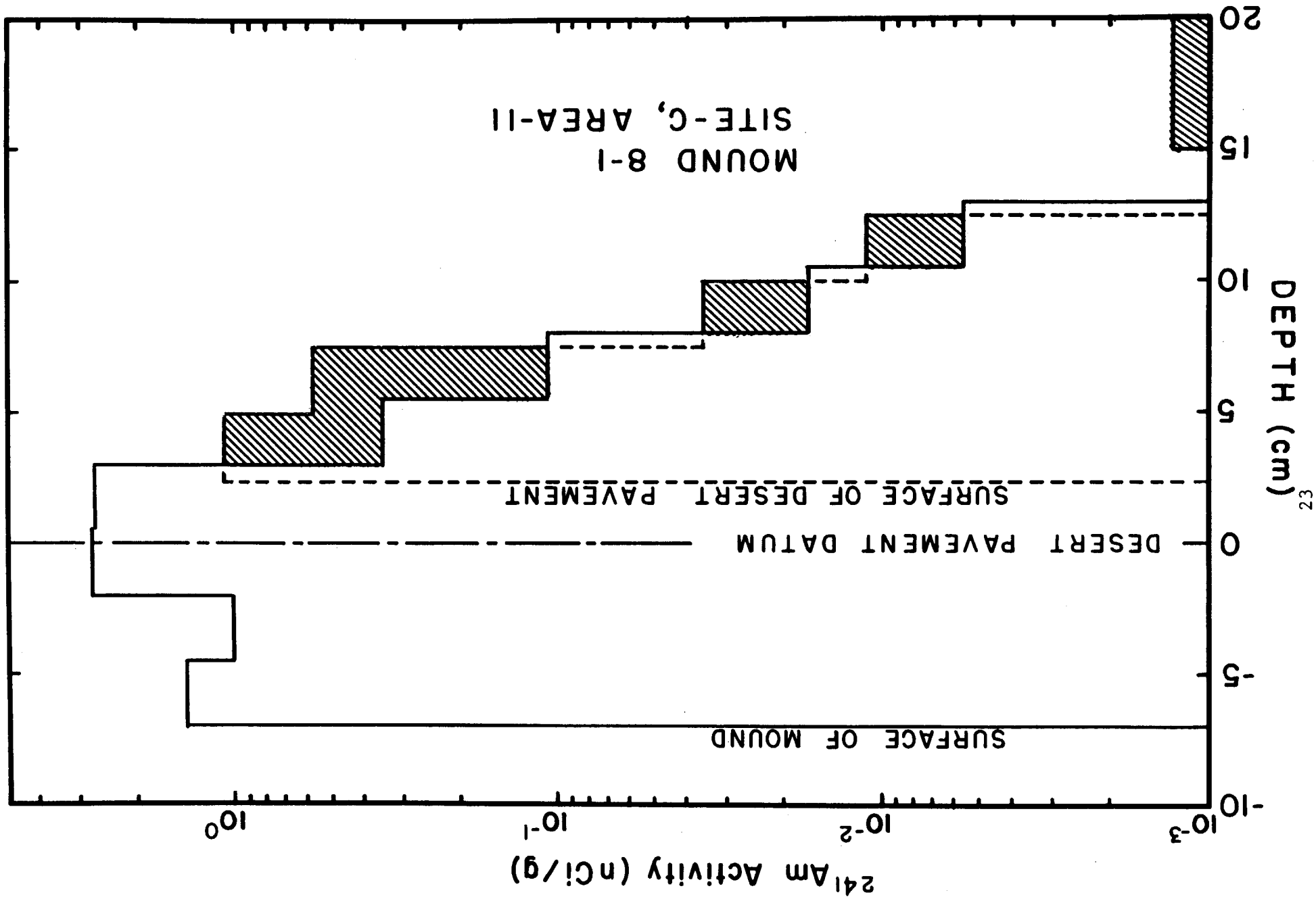
Mound Sampling

Certain studies present unique sampling requirements such as those associated with blow-sand mounds. Blow-sand mounds may constitute as much as 20-30% of a study area. The mound may be considered as composed of layers which have been intermittently deposited by wind. The structure is complicated by the dynamic nature of the mound in that deposition and removal are coexistent. Many mounds have been created or altered by animal activities. The mound may be an accumulator of windblown radionuclides; a knowledge of associated concentrations is necessary to a calculation of inventory. The location and concentration of radionuclides within the mound and the proximity of radionuclides to the root zone are also important.

The Soils Element submitted a procedure for mound sampling to NAEG in March, 1974. The procedure was developed such that simple or complex missions could be satisfied. Procedures were submitted by other Elements, and an integrated version of the proposed methods was used in the summer of 1974 in a pilot study titled Mound Study #1. Profile samples were obtained from a series of mounds and adjacent desert pavement in Area 11 and assayed by Ge(Li) spectrometric techniques for ^{241}Am to predict the distribution of plutonium within the mound. Figure 1 presents one typical pair of mound and desert pavement profiles. It will be noted that in this mound, the surface of the surrounding desert pavement is below the desert pavement datum found under the mound, possibly indicating erosion of the surrounding desert pavement during and after mound formation. Further, radioactivities below the desert pavement datum within the mound and those in the desert pavement surrounding the mound correlate very well below the 2 cm depth. Based on the data obtained, two possible explanations for the observed ^{241}Am distribution are (1) that the radioactivity was deposited prior to mound formation and (2) that the mound may have dynamically moved in the direction of the sampling point covering an already contaminated surface with additional contaminated material. A second point to note is the higher radioactivity associated with the mound than that associated with the surrounding desert pavement. Since the mounds are formed from a relatively narrow range of particles, it can be assumed that ^{241}Am and consequently plutonium is associated with soil particles which fall within that range.

It was concluded from Mound Study #1 that mounds are an important sink for radioactivity; hence, their contribution to inventory may be important. A second study related to mound sampling (Mound Study #2) was initiated in FY 1976 to determine the contribution of plutonium and ^{241}Am in the mound to total inventory. Sampling and analysis are progressing; results are not available at this time.

Fig. 1. ^{241}Am Activity - mound vs surrounding desert pavement



Several extensions of the original mission of the Soils Element as previously described relate to in-depth studies of problems which became evident as the program developed. These studies include soil radionuclide profile evaluations, quality assurance, determination of isotopes of uranium by means other than mass spectrometry, and study site selection.

Soil Radionuclide Profiles

Profile samples were obtained initially as a part of the sampling program for inventory. As profile data accumulated, it became evident that a considerable fund of information was available which might be used to indicate possible mechanisms of soil-radionuclide interactions.

Essington *et al.* (1976) have reported on profile data as related to soil structure and soil anomalies. Figure 2 indicates one type of concentration profile found and the soil structure associated with plutonium concentrations. The points of interest are the two peaks in radioactivity at the 10 and 17 cm depths. Those peaks are at the depths where the profile changes from the A to the B horizon and the B changes to the C horizon. Apparently, the physical nature and/or the chemical changes occurring at the horizon interfaces act upon plutonium, thus causing deposition.

All soil profiles from the safety shot sites have been taken to a depth of 25 cm. No information is available to indicate the maximum depth of penetration of plutonium. In fact, several of the profiles indicate that plutonium may have migrated to depths greater than 25 cm as shown in Fig. 3. Relatively high and constant concentrations of plutonium were found between 7 and 25 cm and there is no indication of a substantial decrease in plutonium between the two depths. Presumably, similar levels of plutonium could be found to greater depths.

Quality Assurance

The quality assurance program was established to provide needed data for comparison of results from different laboratories which used different analytical methods for the estimation of plutonium.

The Soils Element has prepared and submitted for analysis "standard" samples of soil, meat, and vegetation to participating laboratories. Where available, results have been reviewed and used to indicate problem areas which might exist. Results of analyses performed on dosed meat samples are presented in Tables 2 and 3; three isotopes and two levels of radioactivity were investigated. With respect to the high activity samples on Table 2 and the low activity samples on Table 3, results are considered generally satisfactory, although there are anomalies.

Variations observed do indicate the need for extreme care to avoid cross contamination when low activity samples are processed. Such quality control comparisons can be used to alert those involved to potential problems which might not be recognized otherwise.

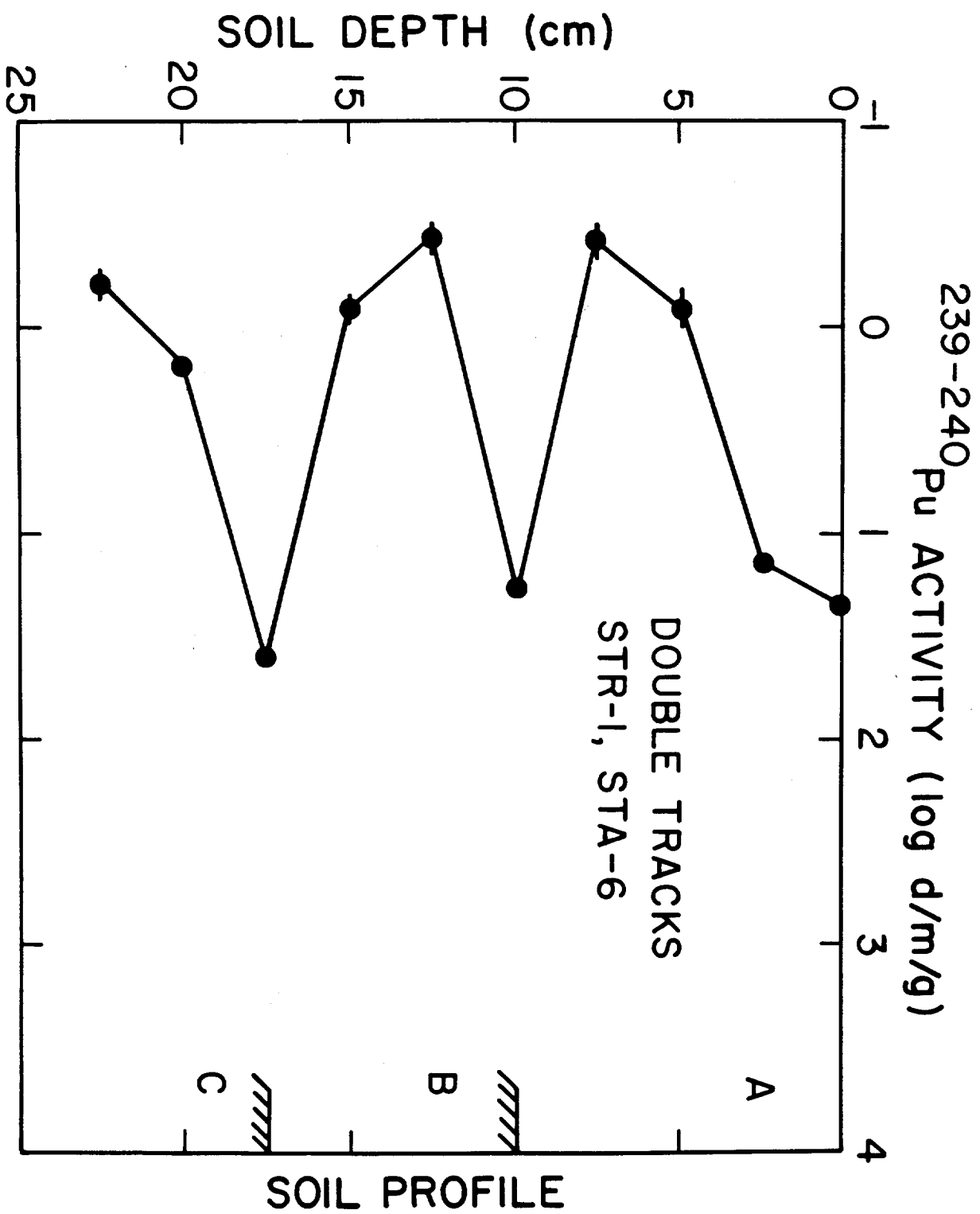


Fig. 2. Distribution of plutonium in Double Tracks soil profile with silicon/iron traces

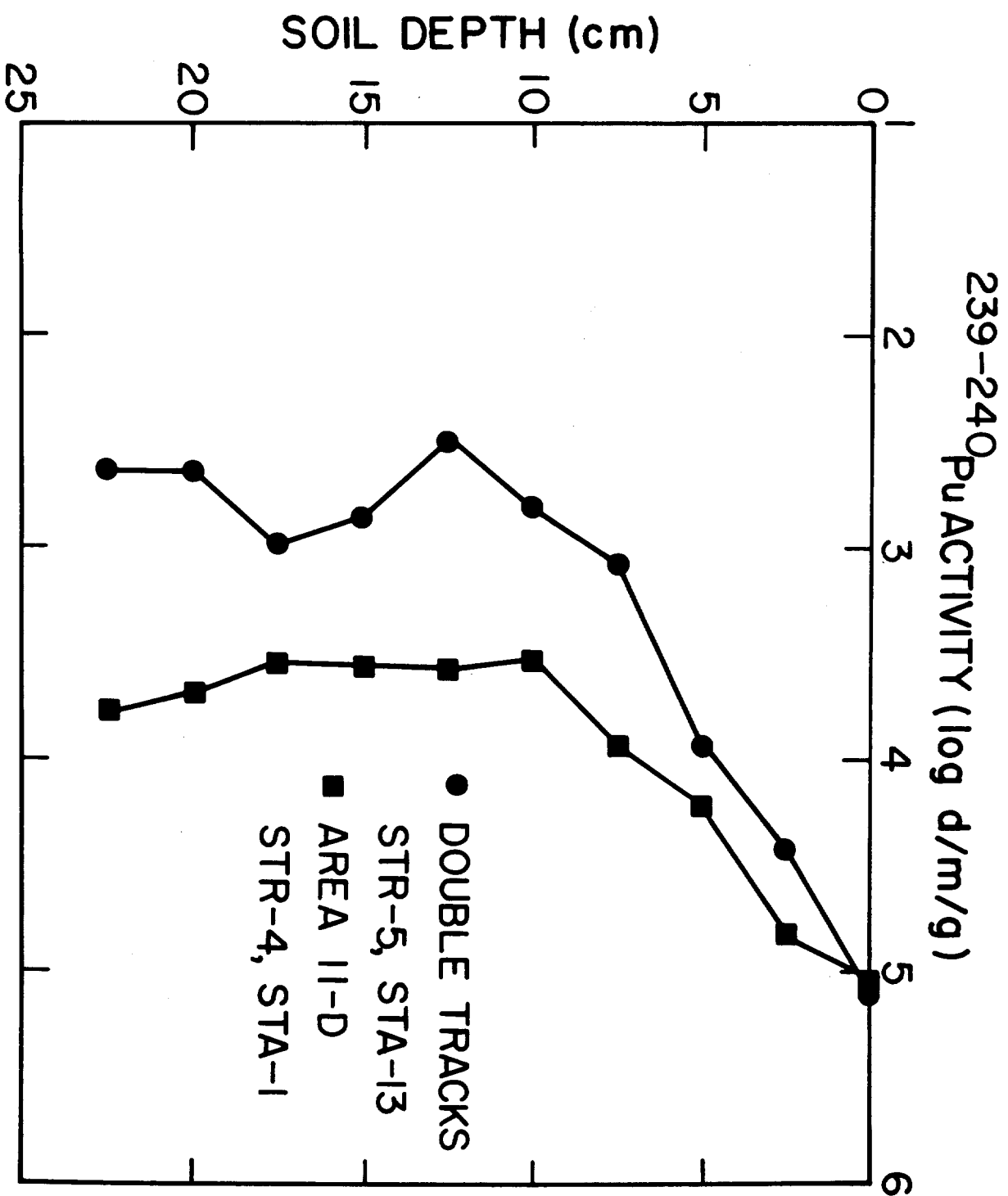


Fig. 3. Distribution of plutonium in top 25 cm of soil.

Table 2. Plutonium and Americium in Dosed Meat Samples

	High Activity (dis/min/g)		
	^{238}Pu	$^{239-240}\text{Pu}$	^{241}Am
Dose	55.6	2590.	1160.
Samp 1	41.9	2750.	1110.
Samp 2	42.6	2580.	1240.
Samp 1	46.0	2400.	1100.
Samp 2	56.0	2830.	1320.
Samp 1	NR	50.	SL
Samp 2	54.0	2440.	960.
Samp 1	NR	2460.	1160
Samp 2	54.0	2690.	1370.

SL = Sample lost
NR = Results not reported

Table 3. Plutonium and Americium in Dosed Meat Samples

	Low Activity (dis/min/g)			
	^{238}Pu	$^{239-240}\text{Pu}$	^{241}Am	
Dose	1.85	86.4	37.5	
Samp 1	3.15	90.6	40.0	SL
Samp 2	4.32	93.7	37.	39.0
Samp 1	2.2	82.		SL
Samp 2	1.82	89.9		33.
Samp 1	NR	2770.		51.5
Samp 2	1.3	74.		40.6
Samp 1	NR	81.3		
Samp 2	1.96	77.9		

SL = Sample lost
NR = Results not reported

Quality assurance associated with the preparation and analysis of samples of vegetation is now in progress for ^{90}Sr , $^{239,240}\text{Pu}$, ^{241}Am , and isotopes of uranium.

A sample plate of electrodeposited plutonium has been circulated among eight laboratories. Results are presented in Table 4. Since the sample was a "real life" sample from NTS, its absolute concentrations are unknown and only data means can be given for purposes of comparison. The exercise checks the counting and calculation procedures; results are equally divided between those which might be considered high and those which might be considered low, yet all results except for one ^{238}Pu analysis fall within plus or minus two standard deviations of the mean. For very precise analytical determinations, those data would indicate that recalibration at several of the laboratories should be accomplished. However, when considering the experience of the large degree of variability among aliquots of safety shot site soil, all laboratories are producing adequate results. Each laboratory used its own counting equipment and methods of calculation. Although differences are small among laboratories, it should be noted that good agreement for one radionuclide does not assure good agreement for a different radionuclide analyzed by the same laboratory. The conclusion is reached that such tests for quality assurance should include all radionuclides of project interest and not just a single radionuclide.

Isotopic Determination of Uranium

It is recognized that the mass spectrometer is the ultimate tool for analysis for the isotopes of uranium; however, for large numbers of samples where the high precision of the mass spectrometer is not required, a rapid, radiochemical method is of value. A radiochemical method has been developed and is now under extensive testing for the determination of the uranium isotopes ^{238}U , $^{235,236}\text{U}$, $^{233,234}\text{U}$, in soil and vegetation.

Due to the low concentrations of deposited device uranium present relative to the concentrations of natural uranium in the soil, it is presently difficult to obtain reliable data for the former. If the concentration of device uranium in the soil is considered of sufficient import, more data are needed which relate to the concentration of natural uranium in the soils of interest so that the amount of device uranium can be determined adequately. The problem has been recognized by earlier investigators and is addressed by Essington et al. (1976) in their recent paper on profile studies.

Site Selection

Originally, the isotope of interest was plutonium; therefore, recommendations had related to safety shot sites and experiments were designed around measurements of plutonium. Following early recommendations, Area 13, Area 5-GMX, and Area 11 were selected as intensive study sites. The Tonopah Test Range (TTR) was later added as a fourth intensive study site (in reality, there are four discrete sites at TTR and Area 11). Although plutonium was the element of prime interest, it was pointed out by the Soils Element in 1973 (Talvittie, 1972) that the ingrowth of ^{241}Am could result in ^{241}Am concentrations being as important as those of plutonium at some future date.

Table 4. Plutonium Activity of Single Electroplated Source
Determined by eight Laboratories
Activity Reported dis/min/g

^{242}Pu	% Dev*	$^{239-240}\text{Pu}$	% Dev*	^{238}Pu	% Dev*
8.35	2.3	4.34	1.4	.73	1.4
8.83	3.3	4.64	8.1	.83	15.
8.45	1.2	4.57	6.8	.72	0.
8.63	.94	3.93	8.2	.68	5.6
8.24	3.6	5.00	17.	.69	4.2
9.38	9.7	3.65	15.	.64	11.
7.97	6.8	3.92	8.4	.72	0.
8.57	.40	4.24	.90	.73	1.4
Mean 8.55		4.28		.72	
CV (%) 4.9		10.		7.	

*Percent deviation from mean
CV = Coefficient of variation

Meetings have been held over the past six months relative to the selection of nuclear sites for intensive study. Preliminary samples from the nonclassified areas are under analysis.

AREAS OF CONCERN AND RECOMMENDATIONS

The LASL soil study personnel have reviewed past and recent activities as they relate to initial assignment. A primary purpose for the review has been to indicate major problems which require further study. By way of summary, recognized gaps in our knowledge are presented as questions, answers, and recommendations from the LASL soil studies personnel and other NAEG principal investigators who have indicated to us related problems.

Sampling

For the soil types sampled, data indicate that if cross contamination has occurred, it has not contributed significantly to the results presented. For the types of soil sampled, the two methods presented, surface and profile sampling, have been successfully used for the safety shot site soils. For stony or rock soils as will be encountered in the nuclear sites, for sampling into highly cemented soil horizons, or for special matrices such as asphalt, concrete, steel, or wood, the LASL soil study personnel recommend development and testing of special sampling techniques. Groundwater, mucks, and sludges will present problems, particularly if profile samples are to be required.

Testing procedures should consist of (1) determination of physical uniformity of samples collected, (2) measurement of degree of cross contamination due to sampling, and (3) comparison of methods statistically.

Analytical Methods for Radionuclides of Interest

Of certain radioisotopes listed in the original charter, the major effort has been placed on plutonium, americium, and uranium. As related to the original charge, are the analytical methods now in use or available adequate for the determination of low levels of activity of future radionuclides of interest?

The methods for plutonium analysis used to date have proved adequate on standard samples; however, comparison of samples is difficult where the "hot particle problem" exists. A concentrated effort should be expended on testing of present radiochemical procedures for low levels of ^{241}Am , especially where plutonium to americium ratios are of interest. The work on such methods now in progress could serve as a useful base.

Quality Assurance

The LASL soil study personnel view quality assurance as a most essential part of any sampling and analysis program. A question is posed as to the adequacy of the present quality assurance program.

The original program design for analyses recommended a 15% replication of samples. Historically, due to economic considerations, the replication has been about 9% for the total number of samples from Areas 13, 5-GMX, and Double Tracks. Experience has shown that the quality assurance program recommended in the original program design was unrealistic for the safety shot site studies due to the "hot particle problem." The particle problem was recognized early in the safety shot site studies, and much of the data has confirmed the importance of the particle problem. Gilbert reviewed the data on replicate samples provided by the LASL Soils Element Referee and concluded that the number of replications necessary to detect a significant difference among laboratories is prohibitive. Thus, quality assurance cannot be based on real life samples from the safety shot sites for economic reasons. For the safety shot site studies, this type of quality control should be discontinued or adjusted. One adjustment might be to follow a recommendation of the Soil Sampling and Analyses Committee made in September, 1971, that each batch of samples could contain a known or "standard" (prepared) sample with results of analyses to be reviewed by the Referee. One such sample was provided by the EPA early in 1972.

The quality assurance program will be expanded to include other radioisotopes of interest as they are defined and in matrices encountered on NTS of interest to the NAEG program.

Soils Element Data

The Soils Element provides a basic source term necessary to all other Elements. NAEG vegetation studies personnel and the NAEG statistician indicated the need for better NAEG ^{241}Am data, specifically more precise analyses (or more numerous samples) at low levels of ^{241}Am and determination of ^{241}Am on the same aliquot as used for $^{239,240}\text{Pu}$. There are indications of disparity in the plutonium-ameridium ratios among study sites. It is not known whether this is real, but it is suspected that it is due in part to difficulty in americium determinations.

The NAEG vegetation study personnel are also interested in concentrations of radionuclides in the rhizosphere, since that concentration is important to plant uptake. A more detailed study of mounds is indicated which is designed to determine the location, amount, and form of the plutonium and ^{241}Am with respect to the rhizosphere.

The importance of some profile sampling to greater depths than the originally recommended 25 cm has been indicated. The referenced reduction of such profile data as were available shows anomalous distribution of Pu and Am which should be studied further to provide data for the prediction of future distribution. Anomalies at greater depths may exist but information is not available at this time. Although some safety shot site profiles have been resampled to greater depths, the samples have not yet been analyzed. It is strongly recommended that future soil sampling efforts include a sufficient number of deep profile samples to determine the maximum depth of penetration of the radionuclides of interest and possible presence of anomalies.

The mechanisms of transport of radionuclides through the soil profile are almost completely unknown. Plutonium and ^{241}Am may be transported as particles, ions, complex ions, colloids, or combinations of those states to loci of concentration. There is a large gap in our knowledge at this point; the mechanisms should be investigated. The presence of inorganic and organic

complexes will have an effect on those mechanisms. Some effort should be directed toward complete chemical analyses, including organic matter, of some soils to determine possible relationships. The organic matter within a mound is of immediate importance in that respect.

Dr. Tamura (ORNL) has performed particle sizing on some samples and determined the plutonium concentration associated with the various sized fractions. This information will be of value for profile and mound studies. However, from the standpoint of hazard evaluation, the NAEG resuspension study personnel need specific data relative to plutonium concentration in the respirable fraction, i.e., that fraction about 4 μm or less in size. Further investigations related to that need should be initiated.

There are microplot data which should be assembled and summarized in a final report. We would recommend that a person or persons be assigned that task and the necessary data supplied to them.

A considerable volume of FIDLER data exists. An attempt should be made to synthesize the data into a report. We have no knowledge of its true value; however, an effort should be made to evaluate the data. As with the microplot data, a person or persons should be assigned this task.

With respect to inventory, there were questions as to whether mound samples were collected at random since the number of mound samples is exceedingly small when compared to the number of desert pavement samples. Field data notes are inadequate on this subject. The data should be reviewed in light of the known mound to desert pavement ratio; mound data should also be reviewed to determine any possible bias which might result if a disparity exists between numbers of mounds sampled and numbers which should have been sampled.

As has been pointed out previously, concentrations of plutonium calculated from ^{241}Am data often show a disparity with data obtained for plutonium concentrations determined by radiochemical analysis. If funds are available, it is recommended that radiochemical analyses be performed on Mound Study #1 samples for $^{239,240}\text{Pu}$ as well as ^{241}Am on the low activity samples to confirm the Ge(Li) scans and to obtain direct data for $^{239,240}\text{Pu}$ to ^{241}Am ratios.

It would be desirable to determine the absolute concentration of ^{241}Pu so that future ^{241}Am concentrations can be projected and so that the absolute relationship between ^{241}Pu and ^{241}Am can be determined.

A procedure for the determination of low levels of uranium has been developed recently. We recommend a meeting of analysts who will be involved for the purpose of reviewing the procedure and designing a testing program commensurate with possible NAEG requirements.

Interim Report

As the effort at the safety shot sites nears completion and NAEG prepares to move into the nuclear sites, it becomes increasingly apparent that synthesized interim reports collating and coordinating those data available should be prepared. The reports should review past NAEG field studies and contain reduced data from each study. Every attempt should be made to interrelate needs and results from each study and thus define "gaps" where further work is indicated. Possible errors in technique and/or philosophy should be examined and alternate recommendations made to provide a guide for further studies.

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VEGETATION

ESTIMATED INVENTORY OF PLUTONIUM AND URANIUM RADIONUCLIDES
FOR VEGETATION IN AGED FALLOUT AREAS

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ABSTRACT

This interim report is third in a series reporting data pertinent to the contamination of vegetation by plutonium and other radionuclides in aged fallout areas on the Nevada Test Site (NTS) and the Tonopah Test Range (TTR).

The standing biomass of vegetation estimated by nondestructive dimensional methods varied from about 200 to 600 g/m² for the different fallout areas. Estimated ^{239,240}Pu inventories (in millicuries) for vegetation of sites located at NTS were 0.47 for Area 5 (GMX); 0.098, 2.2, 3.8, and 6.7 for Area 11, Sites A, B, C, and D, respectively; and 28.2 for Area 13 (Project 57). The inventory estimates for sites at TTR were 0.39, 0.54, 2.6, and 5.7 for DT, CS1, CS2, and CS3, respectively. Estimated standard errors for these inventory estimates are given in this report. Comparisons of soil and vegetation inventory estimates indicate that the standing vegetation contributes an insignificant portion of the total amount of ^{239,240}Pu present in these aged fallout areas. The amounts of plutonium available for vegetation-transport to animals grazing on-site would appear to be relatively small in comparison to the total amounts deposited upon soil. Findings indicate that most of the contaminant found on vegetation probably is attributable to resuspendable materials. For those sites presently under investigation, the contamination level on vegetation amounts, in almost all cases, to less than one-thousandth of that which is present on soil. It is important to recognize that the standing vegetation of these aged fallout areas acts as a windbreak and probably reduces the amount of contaminant that otherwise would move by resuspension should these fallout areas ever become denuded.

Inventory estimates of total uranium for vegetation varied among the different fallout areas from about three grams for Site A in Area 11 to more than 200 grams for the large (4.7 x 10⁶ m²) ABCD region in Area 11. Too few samples have been analyzed to ascertain the significance of the uranium results and their relationship to original fallout depositions and natural background levels at this point in time.

INTRODUCTION

An important objective of the Nevada Applied Ecology Group (NAEG) program is to estimate the total amount (inventory) and geographical distribution of radionuclides in vegetation of fallout areas resulting from "safety tests" in which a chemical explosive was detonated in close proximity to assemblies of plutonium and/or uranium. Estimates of the total amounts of plutonium and uranium for vegetation at each site are given here. These were obtained using stratified random sampling wherein vegetation samples were collected adjacent to random soil locations within strata (subregions) at each site. The inventory estimates for vegetation reported here for plutonium represent data from about 60 percent of the samples collected and analyzed from the fallout areas. Inventory estimates for uranium are preliminary in the sense that only about 20 percent of the total number of vegetation samples collected for estimating inventory have been analyzed for uranium. Updated estimates for plutonium and uranium will be given in future progress reports when these data become available.

METHODS

Attention is directed to earlier progress reports for details of methods concerning the collection and processing of soil and vegetation samples (Gilbert and Eberhardt, 1974; Gilbert *et al.*, 1975a; Romney *et al.*, 1974, 1975) for determining $^{239,240}\text{Pu}$ and ^{241}Am contents (Major *et al.*, 1974) in order to derive inventory estimates for the aged fallout areas. Estimates of inventory were obtained using the corrected stratum area estimates (Gilbert, 1977).

Estimates of the standing biomass of perennial vegetation were made for each test area using nondestructive, dimensional measurements. Procedural details and calculations involving this method have been previously reported (Romney *et al.*, 1974; Wallace and Romney, 1972).

Some data for uranium radionuclides in soil and vegetation samples collected from Area 11, Site A, were reported previously (Gilbert and Eberhardt, 1976; Gilbert *et al.*, 1975b). Results reported herein include data from analyses of additional vegetation samples from other study sites by the contract analytical laboratories.

RESULTS AND DISCUSSION

Estimated Inventory of Plutonium for Vegetation of Fallout Areas

Estimates for the vegetational biomass of the different study areas are given in Table 1. Highest estimates were obtained from the study sites located on

Table 1. Estimated Biomass for Perennial Vegetation of Aged Fallout Areas

Fallout Area	n	Biomass (g/m ²) + S.E.
<u>Nevada Test Site</u>		
Area 5 (GMX)	5	220 ± 36
Area 11A	2	520 ± 150
Area 11B	2	580 ± 24
Area 11C	3	450 ± 22
Area 11D	3	420 ± 7
Area 13 (Project 57)	8	290 ± 34
<u>Tonopah Test Range</u>		
Double Track	6	170 ± 17
Clean Slate 1	6	210 ± 6.8
Clean Slate 2	6	260 ± 57
Clean Slate 3	10	210 ± 11

NTS in Area 11. The floristic composition for these sites generally is more complex than it is for most of the other study sites, partly because Area 11 is located within the transition zone between the Great Basin and Mojave Deserts. The vegetation of Area 5 (GMX) is typically Mojave Desert, whereas that in Area 13 is more representative of the Great Basin Desert flora. The vegetation of study sites located on the Tonopah Test Range is common to the Great Basin Desert.

The data presented in Table 2 have been taken from the corrected stratum areas given in an earlier progress report (Gilbert *et al.*, 1975a). These areas are presented here so that the reader can relate the inventory estimates more easily to the land area represented by the activity strata of each fallout area.

Table 3 gives the estimated inventory of $^{239,240}\text{Pu}$ for vegetation in the aged fallout areas under investigation. It should be understood that these vegetation inventories are only approximate, since the average biomass data used in the calculations were approximated using dimensional analyses (linear regression of biomass on shrub volume) based upon small numbers (see Table 1) of 2 x 50 meter sampling quadrants randomly selected within large areas. Furthermore, all plant species have been lumped together to get an average radionuclide concentration; again the formula for S.E. for inventory is only approximate, being based upon a Taylor series expansion. Nevertheless, these results should give a general impression of the relative magnitude of vegetation contamination within each of these aged fallout areas.

The standing biomass of vegetation varied from about 200 to 600 g/m² (2,000 to 6,000 kg/hectare). Estimates of new leaf and stem production for vegetation of the kind found in these study areas normally are much less than 10 percent of the standing biomass (Kaaz, 1972; Turner and McBrayer, 1974). These estimates represent very low levels of new primary productivity when compared to good grassland or cultivated forage production. However, it should be understood that the standing biomass of these fallout areas is mainly in the form of woody shrubs containing stem material which can be many years old, depending upon the species involved. Data reported previously (Romney *et al.*, 1974, 1975) indicate that most of the contaminant found on this kind of vegetation is attributable to resuspended material deposited upon the external surface of foliage. The estimated vegetation-to-soil inventory ratios given in Table 3, last column, indicate that a greater proportion of the deposited source material can move onto the vegetation at locations farther away from ground zero. This is thought to reflect an initial partitioning in the particle size of the fallout material originally deposited within the fallout pattern. The mean particle size of the fallout material generally decreased at greater distances downwind from ground zero (Romney *et al.*, 1963). Inasmuch as the resuspendable source material entrapped upon plant foliage represents only a limited particle size range, that which is deposited upon foliage contributed less activity in proportion to the total amount of fallout contamination deposited on soil at points nearer to ground zero compared to points farther away.

The estimates of $^{239,240}\text{Pu}$ inventory (in millicuries) for vegetation in the fallout areas located on NTS were $0.47 \pm .073$ (S.E.) for Area 5 (GMX); $0.098 \pm .045$, $2.2 \pm .028$, $3.8 \pm .70$, and $6.7 \pm .73$ for Area 11, Sites A, B, C, and D,

Table 2. Land Area Represented by the Activity Strata Within Aged Fallout Areas

Activity strata	Size of area*		Activity strata	Size of area*		
	m ²	%		m ²	%	%
<u>NTS Area 5 (GMX)</u>			<u>NTS Area 11, ABCD Overlap</u>			
5	3,800	3.0	1	4,672,000**		96.7
1	111,300	88.8				
2	8,400	6.7	<u>NTS Area 11, CD Overlap</u>			
3	800	0.6	6	62,200		1.3
4	1,000	0.8				
Total	125,300	99.9				
<u>NTS Area 13</u>			<u>NTS Area 11, Site B</u>			
1	1,245,000	31.0	2	8,200	46.9	
2	2,547,000	63.4	3	6,000	34.3	
3	108,000	2.7	4	3,300	18.9	
4	74,000	1.8	Total	17,500	100.1	0.4
5	19,000	0.5				
6	24,000	0.6	<u>NTS AREA 11, Site C</u>			
Total	4,017,000	100.0	2	16,400	63.6	
			3	5,600	21.7	
<u>TTR Double Track</u>			4	3,500	13.6	
1	176,000	98.3	5	300	1.2	
2	1,600	0.9	Total	35,800	100.1	0.5
3	800	0.4				
4	600	0.3	<u>NTS Area 11, Site D</u>			
Total	179,000	99.9	2	32,300	60.5	
			3	13,300	24.9	
<u>TTR Clean Slate 1</u>			4	4,900	9.2	
1	157,000	88.9	5	2,900	5.4	
2	10,000	5.7	Total	53,400	100.0	1.1
3	8,400	4.5				
4	1,700	1.0				
Total	177,100	100.1	Grand Total for Area 11	4,840,900		100.0
<u>TTR Clean Slate 2</u>			*Data in this table are taken from Gilbert (1977) as corrected from Gilbert <i>et al.</i> , 1975a.			
1	351,000	74.7				
2	82,300	17.4	**Includes Site A of Area 11 (see Figure 10 in Gilbert <i>et al.</i> , 1975a. Site A is approximately 134,000 m ² in size with 93.9%, 5.8%, and 0.4% in strata 1, 2, and 3, respectively.			
3	26,200	5.5				
4	11,000	2.3				
Total	470,500	99.9				
<u>TTR Clean Slate 3</u>						
1	1,615,000	93.2				
2	61,000	3.5				
3	40,000	2.0				
4	16,000	0.8				
Total	1,732,000	99.9				

Table 3. Estimated Inventory of $^{239-240}\text{Pu}$ for Vegetation in Aged Fallout Areas

Activity strata	n	Mean $\pm$ S.E. (nCi/g dry)	Mean ^a $\pm$ S.E. ^b (nCi/m ²)	Inventory $\pm$ S.E. (millicuries)	Percent by strata	Veg. invent. $\pm$ S.E. ^c Soil invent. $\pm$ S.E. ^c
<u>NTS Area 5--GMX (1954-1955)</u>						
5	13	.0083 $\pm$.0016	1.8 $\pm$.47	.007 $\pm$.002	1	.00038 $\pm$.000091
1	47	.0092 $\pm$.0020	2.1 $\pm$.56	.23 $\pm$.061	49	.00067 $\pm$.00016
2	24	.064 $\pm$.014	14 $\pm$ 3.9	.12 $\pm$.032	26	.00034 $\pm$.00010
3	12	.26 $\pm$.090	58 $\pm$ 22	.046 $\pm$.018	10	.00022 $\pm$.000099
4	17	.31 $\pm$.045	69 $\pm$ 15	.069 $\pm$.015	15	.00014 $\pm$.000034
Total	113			.47 $\pm$.073	101	.00034 $\pm$.000046
<u>NTS Area 11, Site A (1956)</u>						
1	12	.0015 $\pm$.00057	.76 $\pm$.35	.095 $\pm$.045	97	.0029 $\pm$.0018
2	18	.00064 $\pm$.000099	.33 $\pm$.11	.0023 $\pm$.0007	2	.0050 $\pm$.0020
3	6	.0010 $\pm$.00038	.54 $\pm$.24	.00026 $\pm$.00011	0.3	.00031 $\pm$.00033
Total	36			.098 $\pm$.045	99.3	.0028 $\pm$.0017
<u>NTS Area 11, Site B (1956)</u>						
3	11	.10 $\pm$.024	60 $\pm$ 14	.50 $\pm$.12	22	.0020 $\pm$.00085
2	14	.19 $\pm$.053	110 $\pm$ 31	.65 $\pm$.19	29	.00049 $\pm$.00019
4	19	.57 $\pm$.087	330 $\pm$ 52	1.1 $\pm$.17	49	.00023 $\pm$.000055
Total	44			2.2 $\pm$.28	100	.00039 $\pm$.000058
<u>NTS Area 11, Site C (1956)</u>						
2	12	.14 $\pm$.026	61 $\pm$ 12	1.0 $\pm$.20	27	.0018 $\pm$.0014
3	14	.36 $\pm$.054	160 $\pm$ 25	.89 $\pm$.14	24	.0018 $\pm$.00052
4	17	1.1 $\pm$.41	490 $\pm$ 180	1.7 $\pm$.65	45	.00035 $\pm$.00013
5	5	1.2 $\pm$.38	530 $\pm$ 170	.16 $\pm$.052	4	.000088 $\pm$.000049
Total	48			3.8 $\pm$.70	100	.00048 $\pm$.000089

Table 3. (Cont.)

Activity strata	n	Mean $\pm$ S.E. (nCi/g dry)	Mean ^a $\pm$ S.E. ^b (nCi/m ²)	Inventory $\pm$ S.E. (millicuries)	Percent by strata	Veg. invent. $\pm$ S.E. ^c Soil invent. $\pm$ S.E. ^c
<u>NTS Area 11, Site D (1956)</u>						
2	12	.17 $\pm$.033	72 $\pm$ 14	2.3 $\pm$.44	34	.0016 $\pm$.00034
3	13	.24 $\pm$.079	99 $\pm$ 33	1.3 $\pm$.44	19	.00045 $\pm$.00025
4	20	.72 $\pm$.14	300 $\pm$ 58	1.5 $\pm$.28	22	.00031 $\pm$.00011
5	11	1.3 $\pm$.21	550 $\pm$ 89	1.6 $\pm$.26	24	.00020 $\pm$.000088
Total	56			6.7 $\pm$.73	100	.00039 $\pm$.000084
<u>NTS Area 13 (1957) Project 57</u>						
1	36	.0052 $\pm$.00068	1.5 $\pm$.27	1.9 $\pm$.34	7	.00076 $\pm$.00016
2	25	.013 $\pm$.0020	3.8 $\pm$.73	9.6 $\pm$ 1.9	34	.00064 $\pm$.00016
3	15	.17 $\pm$.055	49 $\pm$ 17	5.3 $\pm$ 1.9	19	.0021 $\pm$.00069
4	18	.077 $\pm$.018	22 $\pm$ 5.8	1.6 $\pm$.43	6	.00040 $\pm$.00013
5	10	.28 $\pm$.077	81 $\pm$ 24.3	1.5 $\pm$.46	5	.00077 $\pm$.00026
6	37	1.2 $\pm$.35	348 $\pm$ 109	8.3 $\pm$ 2.6	29	.00043 $\pm$.00012
Total	141			28.2 $\pm$ 3.8	100	.00060 $\pm$.000076
<u>TTR Double Track (1963)</u>						
1	17	.010 $\pm$.0035	1.7 $\pm$.63	.30 $\pm$.11	78	.00026 $\pm$.000096
2	9	.072 $\pm$.029	12 $\pm$ 5.2	.020 $\pm$.0084	5	.000035 $\pm$.000012
3	11	.11 $\pm$.036	19 $\pm$ 6.5	.015 $\pm$.0052	4	.00010 $\pm$.000014
4	11	.49 $\pm$.16	84 $\pm$ 29	.051 $\pm$.018	13	.000031 $\pm$.000015
Total	48			.39 $\pm$.12	100	.00012 $\pm$.000036
<u>TTR Clean Slate 1 (1963)</u>						
1	20	.014 $\pm$.0080	2.9 $\pm$ 1.7	.45 $\pm$.27	84	.00019 $\pm$.00011
2	12	.027 $\pm$.011	5.7 $\pm$ 2.3	.057 $\pm$.024	11	.000088 $\pm$.000056
3	9	.012 $\pm$.0033	2.5 $\pm$.69	.021 $\pm$.0058	4	.000023 $\pm$.000010
4	12	.028 $\pm$.0078	6.0 $\pm$ 1.7	.010 $\pm$.0028	2	.000050 $\pm$.000024
Total	53			.54 $\pm$.27	101	.00013 $\pm$.000078

Table 3. (Cont.)

Activity strata	n	Mean $\pm$ S.E. (nCi/g dry)	Mean ^a $\pm$ S.E. ^b (nCi/m ²)	Inventory $\pm$ S.E. (millicuries)	Percent by strata	Veg. invent. $\pm$ S.E. ^c Soil invent.
<u>TTR Clean Slate 2 (1963)</u>						
1	18	.0066 $\pm$.0013	1.7 $\pm$.49	.59 $\pm$.17	23	.00042 $\pm$.00019
2	13	.047 $\pm$.0055	12 $\pm$ 3.0	1.0 $\pm$.25	39.1	.00017 $\pm$.000075
3	12	.090 $\pm$.019	23 $\pm$ 7.0	.58 $\pm$.18	23	.000087 $\pm$.000021
4	20	.14 $\pm$.032	35 $\pm$ 11	.39 $\pm$.13	15	.00014 $\pm$.000050
Total	63			2.6 $\pm$.38	100.1	.00015 $\pm$.000030
<u>TTR Clean Slate 3 (1963)</u>						
1	10	.013 $\pm$.0042	2.7 $\pm$.89	4.3 $\pm$ 1.4	75	.00023 $\pm$.000074
2	11	.020 $\pm$.0038	4.2 $\pm$.82	.26 $\pm$.05	5	.000072 $\pm$.000028
3	10	.068 $\pm$.015	14 $\pm$ 3.3	.57 $\pm$.13	10	.000067 $\pm$.000028
4	10	.17 $\pm$.055	36 $\pm$ 12	.58 $\pm$.19	10	.000097 $\pm$.000051
Total	41			5.7 $\pm$ 1.4	100	.00016 $\pm$.000037
^a Mean (nCi/m ²) = (mean biomass in g/m ²) x mean ²³⁹⁻²⁴⁰ Pu concentration in nCi/g ^b S.E. = $\pm(\text{mean g/m}^2)^2 \times \text{Var}(\text{mean nCi/g}) + (\text{mean nCi/g})^2 \times \text{Var}(\text{mean g/m}^2) - \text{Var}(\text{mean nCi/g}) \times \text{Var}(\text{mean g/m}^2)$ ¹ ^c S.E. = $R \pm \frac{s_x^2}{3} + \frac{s_y^2}{2} - \frac{2 r s_x s_y}{xy}$ ^{1/2} where R = x/y; r = estimated correlation between x, y.						

respectively; and 28.2 ± 3.8 for Area 13 (Project 57). The four sites located on TTR contained inventory estimates of $0.39 \pm .12$, $0.54 \pm .27$, $2.6 \pm .38$, and 5.7 ± 1.4 for Double Track, and Clean Slate 1, 2, and 3, respectively. Comparisons of soil and vegetation inventory estimates (Table 3, last column) indicate that the standing vegetation contributes an insignificant portion of the total amount of $^{239,240}\text{Pu}$ present in these aged fallout areas. With the exception of an anomalous condition represented by the test of Area 11, Site A (which in view of the large S.E. may be unreliable), the level of contamination attributable to standing vegetation accounted for only one to six ten-thousandths of that estimated for soil. It seems safe to conclude, therefore, that the amounts of $^{239,240}\text{Pu}$ in or on vegetation which are available for grazing animals is relatively small compared to the total amounts deposited in these aged fallout areas.

In view of findings that the vegetation of these fallout areas is being contaminated by resuspendable materials, it seems important to consider the role that this same vegetation also plays in limiting this contamination to the low levels actually encountered. There is no doubt that vegetation acts as a windbreak against wind-driven erosional processes. It is safe to assume that the level of contamination for vegetation in these stabilized fallout areas is much less now than it would be in surrounding areas, should these sites ever become denuded. One of the best safeguards for keeping the soil contaminant in its present location within each area is to maintain the integrity of the standing vegetation.

Preliminary Estimate of Uranium Inventory for Vegetation of Aged Fallout Areas

Some data are given in Tables 4 and 5 concerning the average concentrations and estimated inventory of total uranium for different fallout areas. These data are preliminary in the sense that they represent only about 20 percent of the total number of vegetation samples collected for estimating inventory at the "safety test" sites. Samples to ascertain natural background have not been analyzed, so the data now presented have not been adjusted to account for that portion which might have been derived from the source material of the test devices. Updated estimates and discussion of findings will appear in future progress reports since too few data presently are available to ascertain the significance of these preliminary results and their relationship to original fallout deposition.

Table 6 lists inventory estimates of the different uranium radionuclides for vegetation of Area 11, Site A. The details of the work done at this site and some data for soil and vegetation have been reported earlier by Gilbert and Eberhardt (1975) and Gilbert *et al.* (1975b). The area sampled for Site A consisted of three strata defined on the basis of Ge(Li) scans for ^{235}U on soil collected on a grid system. The land area represented by these three strata is about 480 m² for stratum 3; 7,700 m² for stratum 2; and 126,000 m² for stratum 1. The total land area involved is about 134,000 m².

The contamination of vegetation samples with any radionuclide was highest for samples collected nearest ground zero in stratum 3; lowest levels of contamination were found on vegetation located farther away from ground zero in stratum 1. Summation of the amounts estimated for the different strata give

Table 4. Estimated Average Concentrations of Total Uranium for Vegetation in Aged Fallout Areas

Activity strata	n	Mean $\pm$ S.E. ($\mu\text{g/g dry}$) ^a	Minimum concentration	Maximum concentration	c ^b
<u>NTS Area 5--GMX (1954-1955)</u>					
5	3	.13 $\pm$.029	.097	.19	.38
1	2	.18 $\pm$.063	.11	.24	.50
2	1	.15			
3	1	.37			
4	3	.56 $\pm$.070	.45	.69	.21
Total	10				
<u>NTS Area 11, Site A (1956)^c</u>					
1	12	.046 $\pm$.0066	.015	.084	.50
2	20	.12 $\pm$.020	.021	.32	.75
3	6	.14 $\pm$.027	.074	.25	.47
Total	38				
<u>NTS Area 11, Site B (1956)</u>					
2	4	.15 $\pm$.050	.024	.27	.68
3	6	.20 $\pm$.035	.082	.32	.42
4	9	.25 $\pm$.034	.087	.39	.40
Total	19				
<u>NTS Area 11, Site C (1956)</u>					
2	8	.18 $\pm$.032	.059	.28	.50
3	5	.29 $\pm$.097	.13	.66	.74
4	7	.23 $\pm$.094	.026	.75	1.1
5	1	.99			
Total	21				
<u>NTS Area 11, Site D (1956)</u>					
2	4	.16 $\pm$.021	.13	.22	.25
3	3	.072 $\pm$.029	.015	.11	.69
4	5	.11 $\pm$.041	.0079	.22	.83
5	1	.11			
Total	13				
<u>NTS Area 11, CD Overlap (1956)</u>					
6	3	.18 $\pm$.038	.13	.26	.36
<u>NTS Area 11, ABCD (1956)</u>					
1	29	.10 $\pm$.012	.014	.24	.60

Table 4. (Cont.)

Activity strata	n	Mean $\pm$ S.E. ($\mu\text{g/g dry}$) ^a	Minimum concentration	Maximum concentration	c ^b
<u>NTS Area 13 (1957) Project 57</u>					
1	2	.14 $\pm$.093	.076	.21	.66
2	1	.16			
3	0				
4	1	.12			
5	2	.16 $\pm$.088	.15	.17	.11
6	2	.13 $\pm$.030	.10	.16	.33
Total	8				
<u>TTR Double Track (1963)</u>					
1	5	.15 $\pm$.059	.023	.36	.86
2	2	.14 $\pm$.13	.0092	.26	1.3
3	0				
4	4	.19 $\pm$.035	.11	.22	.37
Total	12				
<u>TTR Clean Slate 1 (1963)</u>					
1	6	.13 $\pm$.038	.019	.27	.73
2	6	.26 $\pm$.066	.077	.55	.63
3	2	.25 $\pm$.13	.12	.38	.75
4	5	.30 $\pm$.041	.19	.39	.30
Total	19				
<u>TTR Clean Slate 2 (1963)</u>					
1	10	.077 $\pm$.024	.020	.25	.98
2	5	.13 $\pm$.018	.062	.17	.32
3	3	.15 $\pm$.049	.058	.22	.56
4	7	.32 $\pm$.037	.21	.46	.30
Total	25				
<u>TTR Clean Slate 3 (1963)</u>					
1	7	.19 $\pm$.020	.11	.27	.27
2	3	.15 $\pm$.011	.13	.17	.13
3	8	.35 $\pm$.079	.070	.73	.64
4	6	.40 $\pm$.12	.088	.88	.70
Total	24				

^a Total uranium measured by fluorimetry method for which the analysis error is about = 20 percent.

^b $c = s/\bar{x}$ = coefficient of variation.

^c Data taken from Gilbert and Eberhardt, 1976.

Table 5. Estimated Inventory of Total Uranium for Vegetation in Aged Fallout Area

Activity strata	Size of area (% of total)	n	Mean $\pm$ S.E. ($\mu\text{g/g dry}$) ^a	Mean ^b $\pm$ S.E. ^c ($\mu\text{g/m}^2$)	Inventory $\pm$ S.E. (grams)	% veg. invent. by strata
<u>NTS Area 5--GMX (1954-1955)</u>						
1,2,5	99	6	.15 $\pm$.023	33 $\pm$ 7.4	4.1 $\pm$ 0.91	95
3,4	1	4	.52 $\pm$.069	110 $\pm$ 24	.20 $\pm$.043	5
Total	100	10			4.3 $\pm$ 0.91	100
<u>NTS Area 11, Site A (1956)^d</u>						
1	93.9	12	.046 $\pm$.0066	24 $\pm$ 7.4	3.0 $\pm$.93	85
2	5.8	20	.12 $\pm$.020	62 $\pm$ 20	.48 $\pm$.16	14
3	.2	6	.14 $\pm$.027	73 $\pm$ 25	.034 $\pm$.012	1
Total	100 2.8	38			3.5 $\pm$.95	100 1.4
<u>NTS Area 11, Site B (1956)</u>						
2,3	84	10	.18 $\pm$.029	100 $\pm$ 17	1.4 $\pm$.24	74
4	16	9	.25 $\pm$.034	150 $\pm$ 21	.50 $\pm$.069	26
Total	100 .4	19			1.9 $\pm$.25	100 0.8
<u>NTS Area 11, Site C (1956)</u>						
2,3	84	13	.22 $\pm$.043	100 $\pm$ 20	2.2 $\pm$.44	79
4,5	14	8	.32 $\pm$.12	150 $\pm$ 57	.57 $\pm$.22	21
Total	100 .6	21			2.8 $\pm$.49	100 1.1
<u>NTS Area 11, Site D (1956)</u>						
2,3,4,5	100 1.2	13	.12 $\pm$.019	50 $\pm$ 8.0	2.7 $\pm$.43	100 1.1
<u>NTS Area 11, CD Overlap (1956)^e</u>						
6	1.3	3	.18 $\pm$.038	80 $\pm$ 17	5.0 $\pm$ 1.1	2.0

Table 5. (Cont.)

Activity strata	Size of area (% of total)	n	Mean $\pm$ S.E. ($\mu\text{g/g dry}$) ^a	Mean ^b ₂ $\pm$ S.E. ^c ($\mu\text{g/m}^2$)	Inventory $\pm$ S.E. (grams)	% veg. invent. by strata
<u>NTS Area 11, ABCD (1956)^f</u>						
1	93.6	29	.10 $\pm$.012	50 $\pm$ 6.4	230 $\pm$ 30	93.5
Total Area 11	99.9	85			246 $\pm$ 30	99.9
<u>NTS Area 13 (1957) Project 57</u>						
1-6	100	8	.14 $\pm$.015	41 $\pm$ 6.5	165 $\pm$ 26	100
<u>TTR Double Track (1963)</u>						
1,2,3,4	100	12	.22 $\pm$.060	37 $\pm$ 11	6.6 $\pm$ 2.0	100
<u>TTR Clean Slate 1 (1963)</u>						
1,2	93	12	.19 $\pm$.041	40 $\pm$ 8.7	6.7 $\pm$ 1.5	92
3,4	7	7	.29 $\pm$.041	61 $\pm$ 8.9	.62 $\pm$.09	8
Total	100	19			7.3 $\pm$ 1.5	100
<u>TTR Clean Slate 2 (1963)</u>						
1	75	10	.077 $\pm$.024	20 $\pm$ 7.4	7.0 $\pm$ 2.6	60
2,3	23	8	.14 $\pm$.020	35 $\pm$ 9.2	3.8 $\pm$ 1.0	34
4	2	7	.32 $\pm$.037	83 $\pm$ 21	0.91 $\pm$.23	8
Total	100	25			12.0 $\pm$ 2.8	100
<u>TTR Clean Slate 3 (1963)</u>						
1,2	96	10	.18 $\pm$.016	38 $\pm$ 6.3	61 $\pm$ 10	94
3,4	4	14	.37 $\pm$.065	78 $\pm$ 14	4 $\pm$.8	6
Total	100	24			65 $\pm$ 10	100

Table 5. (Cont.)

^a Measured by fluorimetric methods for which analysis error is about $\pm 20\%$.

^b Mean ($\mu\text{g}/\text{m}^2$) (mean biomass in g/m^2) x (mean total U conc. in $\mu\text{g}/\text{g}$ dry).

^c S.E. $[(\text{mean } \text{g}/\text{m}^2)^2 \times \text{Var} (\text{mean } \mu\text{g}/\text{g dry}) + (\text{mean } \mu\text{g}/\text{g dry})^2 \times \text{Var} (\text{mean } \text{g}/\text{m}^2) - \text{Var} (\text{mean } \mu\text{g}/\text{g/dry}) \times \text{Var} (\text{mean } \text{g}/\text{m}^2)]^{1/2}$

^d Area 11, Site A data obtained by alpha spectrometry methods; data from other locations obtained using fluorimetric methods.

^e Average biomass (g/m^2) estimated using data from C and D sites (Table 1).

^f Average biomass (g/m^2) estimated using data from all Area 11 sites.

total estimates of 0.0084 g ^{234}U , 0.48 g ^{235}U , 0.0025 g ^{236}U , and 3.0 g ^{238}U for Area 11, Site A. Comparison of the vegetation and soil inventory of the ^{235}U source material (Table 6, last column) indicates that a greater portion of the activity deposited farther out from ground zero is on the vegetation. Here again, as mentioned above for plutonium data in Table 3, we apparently are still seeing evidence, at this point in time, of the original partitioning which occurred during fallout deposition, wherein the mean fallout particle size decreased with increasing distance from ground zero. The resuspendable source material consists of a limited size range; consequently, these small particles contribute less activity in proportion to the total amount of contamination present in the soil at points nearer to ground zero.

Based upon the data now available for ^{235}U and ^{238}U , a comparison of the vegetation and soil inventory estimates for Area 11, Site A (Table 6, last column), shows that the contamination attributable to vegetation represents an insignificant portion of that which is present in surface (0-5 cm) soil. Again we may conclude that less than one-thousandth of the uranium activity present is potentially available to on-site grazing animals via the vegetation transport route.

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Table 6. Estimated Inventory of Uranium Radionuclides for Vegetation of Area 11, Site A (1956)

Activity strata	n	Mean $\pm$ S.E. (ng/dry)	Mean ^a ₂ $\pm$ S.E. ^b ($\mu\text{g}/\text{m}^2$)	Inventory $\pm$ S.E. (grams) ^c	Percent by strata	Veg. invent. $\pm$ S.E. ^d Soil invent. $\pm$ S.E.
<u>234Uranium</u>						
1	7	.093 $\pm$.027	.048 $\pm$.019	.0060 $\pm$.0024	72	Soil data not available
2	20	.55 $\pm$.14	.28 $\pm$.10	.0022 $\pm$.00081	26	
3	6	.82 $\pm$.17	.42 $\pm$.15	.00021 $\pm$.000069	2	
Total	31			.0084 $\pm$.0025	100	
<u>235Uranium</u>						
1	12	4.1 $\pm$ 1.3	2.1 $\pm$.88	.26 $\pm$.11	55	.0015 $\pm$.0012
2	20	49 $\pm$ 12	25 $\pm$ 9.4	.20 $\pm$.072	41	.00060 $\pm$.00025
3	6	74 $\pm$ 16	38 $\pm$ 13	.018 $\pm$.0063	4	.000034 $\pm$.000019
Total	38			.48 $\pm$.13	100	.00046 $\pm$.00012
<u>236Uranium</u>						
1	7	.025 $\pm$.0082	.013 $\pm$.0055	.0016 $\pm$.00070	64	Soil data not available
2	20	.20 $\pm$.050	.10 $\pm$.039	.00081 $\pm$.00030	33	
3	6	.31 $\pm$.065	.16 $\pm$.056	.000076 $\pm$.000027	3	
Total	31			.0025 $\pm$.00075	100	
<u>238Uranium</u>						
1	12	41 $\pm$ 5.7	21 $\pm$ 6.7	2.7 $\pm$.84	90	.00028 $\pm$.000091
2	20	72 $\pm$ 8.8	37 $\pm$ 11	.29 $\pm$.088	10	.00039 $\pm$.00012
3	6	81 $\pm$ 11	42 $\pm$ 13	.020 $\pm$.0063	1	.000058 $\pm$.000027
Total	38			3.0 $\pm$.84	101	.00029 $\pm$.000085

^aSee footnote b, Table 5.^bSee footnote c, Table 5.^cIncludes natural uranium and uranium from safety shot device.^dSee footnote c, Table 3.

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PLANT UPTAKE OF $^{239,240}\text{Pu}$ AND ^{241}Am THROUGH ROOTS
FROM SOILS CONTAINING AGED FALLOUT MATERIALS

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ABSTRACT

Several species of plants were grown under glasshouse conditions in pot cultures containing soil from some of the aged plutonium fallout areas on the Nevada Test Site (NTS) and the Tonopah Test Range (TTR). This interim report contains results for alfalfa, barley, and soybean plants from experiments designed to test the influence of soil amendments on plant uptake of $^{239,240}\text{Pu}$ and ^{241}Am through live root systems. Growth conditions were such that foliage surface contamination could be prevented with confidence.

Results from an experiment with Area 13 soil showed that additions of nitrogen fertilizer and organic matter amendments did not alter the uptake of $^{239,240}\text{Pu}$ and ^{241}Am through roots of barley and alfalfa plants. On the other hand, acidulation of this soil with agricultural grade sulfur significantly increased root uptake of these radionuclides ($P = .05$ level), especially when applied in combination with the chelating agent, diethylenetriaminepentaacetic acid (DTPA). A second experiment involving soil from eight of the different NAEG study sites also showed that the DTPA chelate amendment significantly increased root uptake of these radionuclides by soybean plants.

In these experiments, the Pu/Am ratios for plants generally were much lower than the Pu/Am ratios for soils in which they were grown, indicating much greater uptake of ^{241}Am through roots in proportion to $^{239,240}\text{Pu}$ uptake. Vegetation to soil concentration ratios (CR), which serve as an index of uptake through roots, were low for each plant species and for all soils tested. The CR values for $^{239,240}\text{Pu}$ ranged from 10^{-5} to 10^{-3} for barley straw, and 10^{-6} to 10^{-3} for barley fruit heads; 10^{-5} to 10^{-4} for alfalfa forage; 10^{-4} to 10^{-3} for soybean leaves and stems; and 10^{-6} to 10^{-4} for soybean fruit pods. The effect of increasing uptake by DTPA chelate amendment was generally within one order of magnitude for Pu and Am radionuclides. The CR values for ^{241}Am ranged from 10^{-5} to 10^{-3} for barley straw and fruit heads; 10^{-4} to 10^{-2} for alfalfa forage; 10^{-3} to 10^{-1} for soybean leaves and stems; and 10^{-4} to 10^{-2} for soybean fruit pods. These CR values obtained under glasshouse conditions were low compared to the vegetation to soil activity ratios of 10^{-2} to 10^0 .

which have been observed for samples collected from the aged fallout areas where conditions encouraged resuspension and surface contamination.

INTRODUCTION

Two important mechanisms of incorporation are involved in the vegetation transport of radionuclides from contaminated soil to grazing animals. Findings in earlier progress reports (Romney *et al.*, 1974; 1975) indicate that superficial entrapment of resuspendable material on plant foliage is probably the most important process whereby vegetation becomes contaminated with Pu and Am radionuclides in aged fallout areas on the Nevada Test Site and the Tonopah Test Range. The other process of incorporation by root uptake is difficult to assess under natural conditions involving resuspension. Therefore, samples of soil were removed from the aged fallout areas and used in pot culture experiments to test for root uptake under glasshouse conditions where foliage contamination could be prevented with confidence.

This interim report contains data on the uptake of $^{239,240}\text{Pu}$ and ^{241}Am through roots of alfalfa, barley, and soybean plants from experiments designed to test the effects of nitrogen fertilizer, acidulation, and organic matter amendments with and without additions of diethylenetriaminepentaacetic acid (DTPA). This chelating agent has the ability to increase uptake by plants through roots of several metals and is widely used as a practical means of correcting iron deficiency in plants. It has been shown to greatly increase the uptake from soil of ^{241}Am by plants (Wallace, 1972). Another motivation for testing the ability of chelating agents to modify plant uptake of Pu and Am radionuclides stems from their wide use in chemical processing and storage of transuranic elements.

MATERIALS AND METHODS

Soil was collected from stratum 3 of the Area 13 fallout area (Gilbert and Eberhardt, 1974) for use in experiments to investigate the effects of soil amendments on plant uptake of $^{239,240}\text{Pu}$ and ^{241}Am with and without additions of DTPA chelate. The experimental design involved three sets, each containing four soil amendments with and without DTPA, in triplicate (3 sets x 4 amendments x 2 DTPA x 3 replicates = 72 pots). Soil collected from the field was subdivided into twelve 20 kg lots and mixed thoroughly with given amendments in a P-K blender for one hour before subdividing into six 3200-g lots for potting. An 800-g sample of soil from each mixing was submitted to the contract laboratory for radiochemical analysis. The soil amendments consisted of a control soil treatment (unmodified except for mixing), nitrogen fertilization (applied as

Table 1. $^{239,240}\text{Pu}$ and ^{241}Am Contents of Area 13 Soil Amended for Alfalfa and Barley Experiments

Soil amendments*	$^{239,240}\text{Pu}$ (pCi/g)				^{241}Am (pCi/g)			
	Pu	Mean	S.D.	S.E.	Am	Mean	S.D.	S.E.
Control	601				99.3			
	649	706	142.2	82.2	94.1	107	18.3	10.5
	868				128			
Nitrogen	628				92.8			
	606	664	81.6	47.1	85.1	95.0	11.1	6.4
	757				107			
Sulfur	505				70.1			
	761	644	129.4	74.7	118	98.0	24.9	14.4
	666				106			
Organic matter	(lost)				(lost)			
	548	645	137.2	97	78.5	93.8	21.6	15.3
	742				109			
Overall		666	106.4	32.1		98.9	17.1	5.2

*No significant difference ($P=.05$) in Pu or Am contents due to soil amendments. Average Pu/Am ratio $\pm$ S.E. = 6.74 ± 0.11 (see footnote, Table 2)

NH₄NO₃ equivalent to 200 kg N per hectare), 2 percent agricultural grade sulfur (reduced pH from 7.6 to 5.4), and 5 percent organic matter (alfalfa meal). The mixing and potting of soil was done in closed systems within a full containment dust hood. Soil was potted in 22-cm diameter plastic pots which were sleeved inside of 10-liter plastic buckets, and then covered with 5 cm of #16 silica sand to prevent soil particle resuspension. Seeds were planted in the sand immediately above the soil surface. Moisture was supplied by irrigation with deionized water. Cooling air for the glasshouse was drawn through medical filter media and activated charcoal filters.

Results from radiochemical analysis of Area 13 soil subsamples are given in Table 1. Reasonable uniformity in ^{239,240}Pu and ²⁴¹Am contents was obtained by the mixing process; however, the activity levels turned out to be much lower than had been anticipated. As a result, it was necessary to combine the plant material grown on similar treatments of the three sets into just one set of three pooled replicates in order to obtain an adequate sample size for radioassay. Barley plants were grown first on the Area 13 soil and harvested in the dough stage by cutting them at a point about 5 cm above the top of the sand layer and dividing them into straw and fruit head samples. Alfalfa was grown next on these soils. Three successive cuttings of forage were harvested in the quarter-bloom stage and combined to form one set of three pooled replicates for radiochemical analysis. As with barley, the alfalfa plants were cut about 5 cm above the sand surface as an added precaution to prevent contamination from soil particles.

Results from the first experiment showed need for working with soil containing higher levels of contamination. In addition, the response from the soil amendment treatments indicated a practical influence from only the DTPA chelating agent. Consequently, a second series of experiments was run using soil collected from eight of the NAEG study areas. For these experiments, the soil was collected from a site within each fallout area at which FIDLER activity readings ranged from 20,000 to 30,000 cpm. The design of each soil experiment consisted of three sets, with and without DTPA chelate, replicated three times (3 sets x 2 DTPA x 3 replicates = 18 pots). Mixing and potting methods were the same as described above for Area 13 soil. The ^{239,240}Pu and ²⁴¹Am contents and ratios for these soils are given in Table 2. Wheat and soybean plants were grown on these soils, harvested, and pooled into one set of three replicates each, in order to provide an adequate sample of plant tissue for radiochemical analysis. Results for the wheat crop have not yet been returned from the contract analytical laboratory.

RESULTS AND DISCUSSION

Data from the first experiment with barley plants are given in Table 3. The results for both Pu and Am radionuclide uptake were very erratic and reminiscent of the kinds of variability often encountered during our earlier investigations (Nishita *et al.*, 1961) of plant uptake of radionuclides from soils contaminated with fallout material from aboveground nuclear weapon tests at NTS. In this

Table 2. $^{239,240}\text{Pu}$ and ^{241}Am Contents and Ratios for Soils
Used in Soybean Experiment

Soil source	$^{239,240}\text{Pu}$ (nCi/g)			^{241}Am (nCi/g)			$^{239,240}\text{Pu}/^{241}\text{Am}$		
	Mean	±	S.E.*	Mean	±	S.E.*	Ratio	±	S.E.**
Area 11 B	2.8	±	0.41	0.44	±	0.068	6.5	±	0.17
Area 11 C	9.6	±	1.3	1.8	±	0.20	5.3	±	0.17
Area 11 D	4.6	±	0.24	0.87	±	0.062	5.2	±	0.12
Area 13	6.0	±	0.93	1.1	±	0.11	5.6	±	0.27
Clean Slate 1	4.3	±	0.57	0.23	±	0.033	19	±	0.33
Clean Slate 2	15	±	3.6	0.72	±	0.13	21	±	1.9
Clean Slate 3	11	±	1.6	0.59	±	0.073	19	±	0.67
Double Track	5.9	±	0.79	0.28	±	0.033	21	±	0.88

*Standard error of mean - $[\text{Var.}/n]^{1/2}$

**S.E. = $\left[\left(\frac{\sum Y_i^2}{X_i} - \frac{(\sum Y_i)^2}{\sum X_i} \right) / (n-1) \right]^{1/2}$ (Snedecor and Cochran, 1967)

Table 3. ^{239}Pu and ^{241}Am Contents and Ratios for Barley Plants Grown on Area 13 Soil Treated With Agricultural Amendments

Soil amendment	^{239}Pu (pCi/g)		^{241}Am (pCi/g)		$^{239}\text{Pu}/^{241}\text{Am}$		Veg./Soil C.R. ⁺⁺	
	Mean	± S.E. ⁺	Mean	± S.E. ⁺	Ratio	± S.E. ⁺	Pu	Am
<u>Barley straw</u>								
Control	0.14	± 0.12	0.052	ND	2.6	ND	1.9 E-4	4.8 E-4
C + DTPA	0.16	± 0.099	0.14	± 0.014	1.2	± 0.77	2.3 E-4	1.3 E-3
Nitrogen	0.012	± 0.0010	0.016	± 0.0028	0.76	± 0.20	1.8 E-5	1.6 E-4
N + DTPA	0.15	± 0.062	0.18	± 0.045	0.85	± 0.28	2.2 E-4	1.8 E-3
Sulfur	0.050	± 0.023	0.057	± 0.011	0.87	± 0.46	2.8 E-4	5.8 E-4
S + DTPA	3.7	± 1.1**	0.42	± 0.27**	8.9	± 4.8	5.7 E-3	4.2 E-3
Organic matter	0.016	± 0.0054	0.0056	± 0.0010	2.8	± 0.49	2.4 E-5	6.0 E-5
O.M. + DTPA	0.56	± 0.20	0.27	± 0.15	2.1	± 0.26	8.6 E-4	2.8 E-3
<u>Barley fruit heads</u>								
Control	0.29	± 0.14	0.051	± 0.040	5.5	± 4.2	4.1 E-4	4.8 E-4
C + DTPA	0.0042	± 0.0030	0.0075	ND	0.56	ND	5.9 E-6	7.0 E-5
Nitrogen	0.0026	± 0.0014	ND	ND	ND	ND	3.8 E-6	ND
N + DTPA	0.010	± 0.0023	0.028	± 0.012	0.36	± 0.10	1.6 E-5	3.0 E-4
Sulfur	0.14	± 0.095	0.050	± 0.0042	2.7	± 2.2	2.1 E-4	5.2 E-4
S + DTPA	2.0	± 0.54	0.35	± 0.050	5.8	± 0.86	3.1 E-3	3.5 E-3
Organic matter	0.0032	± 0.0050	0.013	± 0.0079	0.25	± 0.17	5.0 E-6	1.4 E-4
O.M. + DTPA	0.15	± 0.059	0.11	± 0.0098	1.3	± 0.44	2.4 E-4	1.2 E-3

⁺See footnotes, Table 2. Average Pu/Am ratio for Area 13 soil = 6.7 ± 0.11

ND = Not detected in some replicates

⁺⁺C.R. = vegetation to soil activity ratio (pCi per g Veg./pCi per g soil)

**Significant differences (P = .05)

particular instance, however, much of the variability is attributable to the insensitivity of the analytical methods to accurately measure the low levels of Pu and Am radionuclides present in the small samples of vegetation available for radioassay. In spite of the high degree of variability, these results gave some interesting findings. First, the uptake of Pu and Am radionuclides through plant roots was relatively low compared to the levels of contamination present in the soil. CR values for $^{239,240}\text{Pu}$ ranged from 10^{-5} to 10^{-3} for barley forage and from 10^{-6} to 10^{-3} for the fruit heads. In several cases, the CR values for ^{241}Am were within an order of magnitude higher than for Pu, indicating greater uptake of Am in proportion to Pu uptake through plant roots. Second, the nitrogen fertilizer and organic matter amendments had no significant influence on root uptake of these radionuclides. The DTPA chelate treatments showed a tendency for increased uptake, but the high variability masked any test for significance. Third, the acidulation effect of sulfur in combination with DTPA chelate significantly increased ($P = .05$) plant uptake of $^{239,240}\text{Pu}$ and ^{241}Am through roots. This might imply greater solubility of the source materials under more acidic edaphic conditions, but such conditions are unlikely to occur in the soils of aged fallout areas at NTS and TTR because of their high buffering capacity.

Results from the second experiment with alfalfa plants are given in Table 4. Inasmuch as three different cuttings of forage were pooled together, the sample size available for radioassay was as much as 10 times that obtained from the single barley crop. Consequently, more reliable results were obtained with less variability in the analytical data. The results for alfalfa again verified relatively low plant uptake through roots of $^{239,240}\text{Pu}$ and ^{241}Am from Area 13 soils. There were also no significant effects of the soil amendments applied without DTPA chelate, with the exception of an increase in ^{241}Am uptake attributable to acidulation by sulfur. For treatments where DTPA chelate had been applied, a significant increase ($P = .05$) in root uptake of Pu and Am radionuclides was induced by acidulation with sulfur. This particular effect was more pronounced for Pu than for Am uptake. The organic matter amendment in combination with DTPA chelate also significantly increased $^{239,240}\text{Pu}$ uptake through alfalfa roots. However, part of this particular increase might have resulted from the slight acidulation effect caused by the high organic matter treatment (pH 7.6 to 6.5). Even though certain modifying effects of soil amendments mentioned above were significant, from a practical standpoint, the CR values for Pu were altered only from 10^{-5} to 10^{-4} and from one to two orders of magnitude higher for Am.

Results are given in Table 5 for the third experiment wherein root uptake of Pu and Am from different fallout area soils was tested with soybean plants. (Analytical data for wheat grown in this experiment were not available to include in this progress report.) The $^{239,240}\text{Pu}$ and ^{241}Am contents of plant tissues varied among the different soils according to differences in the contamination levels present. Inasmuch as a common level of contamination had not been achieved, comparisons between data can be made only for the DTPA chelate and nonchelate treatments for a given soil. A simple test of these data indicated that, in virtually all cases, the addition of DTPA chelate significantly increased Pu and Am radionuclide uptake through soybean roots. Among the different soils tested, higher levels of Pu and Am activity appeared to be taken up from the Area 11C and D soils in proportion to the contamination

Table 4. $^{239,240}\text{Pu}$ and ^{241}Am Contents and Ratios for Alfalfa Grown on Area 13 Soil Treated With Agricultural Amendments

Soil amendment	$^{239,240}\text{Pu}$ (pCi/g)			^{241}Am (pCi/g)			$^{239,240}\text{Pu}/^{241}\text{Am}$		Veg./Soil C.F. ⁺	
	Mean	±	S.E. ⁺	Mean	±	S.E. ⁺	Ratio	± S.E. ⁺	Pu	Am
Control	0.027	±	0.010	0.052	±	0.012	0.53	± 0.27	3.8 E-5	4.8 E-4
C + DTPA	0.042	±	0.024	0.098	±	0.014	0.43	± 0.25	5.9 E-5	9.2 E-4
Nitrogen	0.043	±	0.030	0.0556	±	0.014	0.77	± 0.34	6.4 E-5	5.8 E-4
N + DTPA	0.042	±	0.015	0.090	±	0.026	0.47	± 0.19	6.5 E-5	9.4 E-4
Sulfur	0.047	±	0.013	0.10	±	0.016**	0.47	± 0.19	7.2 E-5	1.1 E-3
S + DTPA	0.23	±	0.062	0.19	±	0.52**	1.2	± 0.0022	3.6 E-4	1.9 E-2
Organic matter	0.080	±	0.047	0.046	±	0.0032	1.7	± 0.92	1.3 E-4	4.9 E-4
O.M. + DTPA	0.22	±	0.022	0.094	±	0.045	1.7	± 1.0	3.4 E-4	1.0 E-3

⁺See footnote, Tables 2 and 3. Average Pu/Am ratio for Area 13 soil = 6.7 ± 0.11

**Significant differences (P = .05)

Table 5. $^{239,240}\text{Pu}$ and ^{241}Am Contents and Ratios for Soybean Plants Grown on Soils Containing Aged Fallout Materials

Soil source and treatment	$^{239,240}\text{Pu}$ (pCi/g)			^{241}Am (pCi/g)			$^{239,240}\text{Pu}/^{241}\text{Am}$			Veg./Soil C.R. ⁺	
	Mean	±	S.E. ⁺	Mean	±	S.E. ⁺	Ratio	±	S.E. ⁺	Pu	Am
<u>Soybean leaf and stem</u>											
Area 11 B	0.43	±	0.046	0.52	±	0.11	0.83	±	0.11	1.5 E-4	1.2 E-3
11 B + DTPA	4.6	±	0.67	23	±	4.1	0.20	±	0.016	1.6 E-3	5.2 E-2
Area 11 C	1.7	±	0.15	4.9	±	1.8	0.35	±	0.13	1.8 E-4	2.7 E-3
11 C + DTPA	36	±	5.2	162	±	41	0.22	±	0.10	3.7 E-3	8.9 E-2
Area 11 D	5.0	±	1.3	15	±	3.5	0.33	±	0.12	1.1 E-3	1.7 E-2
11 D + DTPA	30	±	3.6	329	±	34	0.92	±	0.018	6.6 E-3	3.7 E-1
Area 13	0.66	±	0.077	3.6	±	1.1	0.19	±	0.035	1.1 E-4	3.3 E-3
A13 + DTPA	24	±	9.7	29	±	14	0.89	±	0.17	3.9 E-3	2.7 E-2
Clean Slate 1	1.8	±	0.38	3.2	±	1.6	0.57	±	0.36	4.3 E-4	1.4 E-2
CS1 + DTPA	3.0	±	0.057	4.8	±	2.2	0.62	±	0.65	7.1 E-4	2.1 E-2
Clean Slate 2	11	±	5.6	1.8	±	0.81	6.3	±	1.3	7.6 E-4	2.5 E-3
CS2 + DTPA	18	±	5.8	29	±	3.1	0.63	±	0.14	1.2 E-3	4.0 E-2
Clean Slate 3	6.3	±	1.5	3.3	±	0.34	1.9	±	0.26	5.5 E-4	5.5 E-3
CS3 + DTPA	8.9	±	0.46	5.2	±	0.94	1.7	±	0.43	7.8 E-4	8.6 E-3
Double Track	1.5	±	0.52	1.6	±	0.81	0.97	±	0.72	2.6 E-4	5.7 E-3
DT + DTPA	2.5	±	0.17	1.9	±	0.30	1.3	±	0.29	4.2 E-4	6.7 E-3
<u>Soybean fruit pods</u>											
Area 11 B	0.023	±	0.0017	0.071	±	0.038	0.32	±	0.19	7.8 E-6	1.6 E-4
11 B + DTPA	0.074	±	0.037	0.52	±	0.058	0.14	±	0.062	2.6 E-5	1.2 E-3
Area 11 C	0.11	±	0.017	0.35	±	0.081	0.30	±	0.090	1.1 E-5	1.9 E-4
11 C + DTPA	1.2	±	0.074	3.6	±	2.2	0.31	±	0.22	1.2 E-4	2.0 E-3
Area 11 D	0.19	±	0.023	0.67	±	0.23	2.9	±	0.10	4.2 E-5	7.6 E-4
11 D + DTPA	1.0	±	0.074	20	±	13	0.050	±	0.039	2.2 E-4	2.3 E-2
Area 13	0.014	±	0.0010	0.085	±	0.0065	0.17	±	0.015	2.4 E-6	8.1 E-5
A13 + DTPA	0.92	±	0.49	1.3	±	0.18	0.73	±	0.31	1.5 E-4	1.2 E-3
Clean Slate 1	0.072	±	0.012	0.046	±	0.0092	1.6	±	0.086	1.7 E-5	2.0 E-4
CS1 + DTPA	0.096	±	0.0030	0.24	±	0.022	0.41	±	0.052	2.2 E-5	1.0 E-3
Clean Slate 2	0.22	±	0.015	0.097	±	0.0076	2.2	±	0.19	1.4 E-5	1.3 E-4
CS2 + DTPA	0.67	±	0.070	0.90	±	0.090	0.74	±	0.048	4.4 E-5	1.2 E-3
Clean Slate 3	0.20	±	0.015	0.20	±	0.011	1.0	±	0.066	1.8 E-5	3.4 E-4
CS3 + DTPA	0.30	±	0.053	0.34	±	0.075	0.88	±	0.088	2.7 E-5	5.8 E-4
Double Track	0.058	±	0.016	0.16	±	0.061	0.36	±	0.51	9.8 E-6	5.8 E-4
DT + DTPA	0.080	±	0.017	0.24	±	0.031	0.33	±	0.028	1.3 E-5	8.6 E-4

⁺ See footnotes. Tables 2 and 3 for soils are in Table 2

levels present (compare with soil data in Table 2). Additional work is under way to determine the extent to which this finding is related to physical and chemical differences in source material and to edaphic characteristics. The activity levels for these radionuclides in fruit pods were from one-tenth to one-hundredth of the levels found in the forage tissue. The CR values for Pu in soybean leaf and stem tissues ranged from 10^{-4} to 10^{-3} , and the fruit pod values ranged from 10^{-6} to 10^{-4} . For Am, the CR values ranged from 10^{-3} to 10^{-1} for leaf and stem tissue and 10^{-1} to 10^{-2} for fruit pods.

It should be understood that the CR value is a ratio of the vegetation to soil activity level which expresses the relative uptake of a radionuclide through the plant root system. It should not be misconstrued to be an index representing contamination hazards even though it may give an indication, for a given contamination condition, of the relative proportion of that contaminant which might be expected to be incorporated in vegetation through the plant root system.

We believe that perhaps the most important finding from these experiments pertains to the greater uptake of ^{241}Am through plant roots in proportion to the uptake of $^{239,240}\text{Pu}$ demonstrated by these experiments. The magnitude of this differential uptake can be ascertained by comparing the Pu/Am ratios for vegetation (Tables 3, 4, and 5) with the Pu/Am ratios for the soils on which the plants were grown (Tables 1 and 2). In spite of the erratic data for barley (Table 3), the Pu/Am ratios for plant tissues were, in many cases, much less than the average Pu/Am ratio of 6.74 for Area 13 soil. Data for alfalfa (Table 4) conclusively showed greater uptake of ^{241}Am over $^{239,240}\text{Pu}$ uptake by a factor of at least ten for the unamended soil. In those cases where the acidulation amendment had markedly enhanced $^{239,240}\text{Pu}$ uptake, this effect was still around a factor of 5. Even higher factors indicating preferential uptake of ^{241}Am through roots are evident from the soybean data in Table 5. We believe that this differential uptake characteristic for Am probably contributes to the slightly lower Pu/Am ratios often encountered for vegetation samples collected in the aged fallout areas (Romney *et al.*, 1974, 1975) compared to Pu/Am ratios for the soils. The impact of this finding for ^{241}Am uptake assumes greater importance when coupled with the fact that this radionuclide is an ingrowth product of the source material in the aged fallout areas (Fowler and Essington, 1974). About 50 years of time still must elapse before the ingrowth of this radionuclide will peak out. It appears, therefore, that potential problems from Am are equally as important, if not of greater concern, as the aged plutonium source material in these fallout areas.

FUTURE PLANS

As the result of experimental findings, work on the soil amendments has been discontinued, except for chelating agents. The acidulation effects of sulfur have no practical value other than to raise the question of whether or not these transuranic radionuclides might be more available to plants in the event of future contamination problems at sites involving acidic soils.

Some earlier work (Romney *et al.*, 1970) raised concern that Pu may become more available with passing time under continued cropping conditions. In order to further investigate this problem and the comparable fate of Am, long-term cropping experiments are under way with these fallout area soils being cropped with alfalfa and native desert plants.

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INITIAL LAND RECLAMATION PROCEDURES RELATED TO
POSSIBLE Pu-CLEANUP ACTIVITIES AT THE TONOPAH TEST RANGE

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ABSTRACT

If areas of the Tonopah Test Range (TTR) are to be used for experimental tests of procedures for cleanup of Pu contamination, there are experiences in the Great Basin Desert portions of the Nevada Test Site (NTS) which can serve as guides to reclamation and revegetation of such arid lands. Procedures which will encourage development of the grasses *Hilaria jamesii* and *Oryzopsis hymenoides*, as well as the perennial shrubs *Eurotia lanata* and *Atriplex canescens*, would greatly improve the area as rangeland.

FIELD OBSERVATIONS

It is possible that small portions of areas which have been contaminated with ^{239}Pu during testing activities at the Nevada Test Site (NTS) and the Tonopah Test Range (TTR) will need to be subjected to cleanup procedures. Disturbed areas are also possibly subject to the National Environmental Policy Act of 1969, Executive Order 11514, and ERDA Manual Chapter 0510 regulating posttreatments of any areas disturbed by activities of man. The best procedures for handling contaminated land areas under desert conditions are not yet known (Wallace and Romney, 1974), nor are the problems relative to potential hazard fully understood (Stannard, 1973).

Consideration is being given to at least some experimental cleanup of study sites at the TTR which are located in the Great Basin Desert at an elevation of about 1,958 meters. Average annual precipitation over a 9-year period is 13 cm (Schaeffer, 1970). Precipitation usually occurs during every month of the year. Peak temperature conditions are milder at TTR than in the Mojave Desert where the elevation is typically less than 1,000 meters. This results in lower evapotranspiration rates at TTR. Summers are cooler and winters are colder with more snow and freezing in the Great Basin Desert as compared with the Mojave Desert (Table 1).

Table 1. Average meteorological data for study sites at TTR (Great Basin Desert) and Rock Valley (Mojave Desert).

Month	Tonopah Test Range (1961-1969)				Rock Valley (1963-1970)		
	Precipitation	T ^o C		Snow	Rainfall	T ^o C	
	cm	max	min	cm	cm	max	min
Jan.	0.6	7	-10	5.1	1.8	20	-8
Feb.	1.7	9	-6	17.0	1.5	21	-5
Mar.	0.4	12	-4	7.9	0.9	27	-3
Apr.	1.1	17	-1	7.9	1.4	29	-1
May	1.1	22	4	5.1	0.4	35	4
June	1.7	27	9	0.0	0.5	39	8
July	1.0	32	12	0.0	1.3	41	17
Aug.	2.4	31	12	0.0	1.4	41	15
Sept.	1.2	26	7	0.0	0.6	36	10
Oct.	0.3	21	2	0.3	0.3	32	4
Nov.	1.0	12	-4	4.1	1.8	25	-2
Dec.	0.5	7	-8	4.6	1.3	19	-7
Totals	13.0	-	-	52.0	13.2	-	-

Nights are cool throughout the year at the TTR. For all these reasons, restoration of disturbed land, although difficult because of low rainfall, would be somewhat easier to achieve at the TTR than in the Mojave Desert.

The revegetation experiences at the Palanquin study site on Pahute Mesa on the NTS (Wallace *et al.*, 1973) are of some relevance to the conditions at the TTR. Both sites are in the Great Basin Desert. Both have less than 20 cm of rainfall per year on the average. Both are shrublands, although with somewhat different vegetation (Wallace *et al.*, 1972; Leavitt, 1974; Rhoads, 1974; Rhoads and Mullen, 1974). After the native ground cover of the Palanquin close-in fallout pattern had been killed by radiation from a nuclear test, the area rapidly became a grassland. Measurements taken 4 years after this event showed very little grass in the undisturbed areas compared to a grass cover of 28% in an area in which the shrubs had been killed by the radiation (Wallace *et al.*, 1972). The grasses were typically *Sitanion jubatum* and *Hilaria jamesii*, although *Oryzopsis hymenoides* and *Poa sandbergii* also were abundant.

Important considerations arise from these effects. For one, when the shrubs were killed, they could no longer compete with seedlings for soil moisture. Such plants then survive. However, since few grass seeds were available in the area at the time the vegetation was killed, it took about 4 years to build up a sufficient supply of grass seed to complete the process of grassland development. This is typical when shrub competition minimizes seed production of nonshrub species. It is expected then that any revegetation process can be hastened by planting grass seed into the disturbed areas resulting from cleanup procedures.

Soil conditions differ markedly at Pahute Mesa and on the TTR, the more productive soils being at Pahute Mesa. In addition, the radiation kill at Pahute Mesa did not involve any soil disturbance such as removal of topsoil containing seeds and soil organic matter. Any cleanup operation which removes topsoil containing the majority of the soil organic matter will result in far more drastic effects than the radiation kill at Pahute Mesa. The results then at the Palanquin site could not be expected to be completely duplicated at the TTR without careful land manipulation.

The limiting factors for reestablishment of vegetation for disturbed areas will be the same in any desert. These are: first, low soil moisture; second, unfavorable soil structure resulting from very low soil organic matter; third, harsh chemical soil environment related to salinization and high CaCO_3 ; fourth, the invasion of disturbed areas by unwanted annual plant species such as *Salsola*; and fifth, animal activity which destroys many new seedlings. These five factors result not only in difficulty of plant reestablishment, but also in soil instability ending in wind and water erosion.

Principles and practices for land restoration and revegetation which may be useful at the TTR and elsewhere have been outlined previously (Wallace and Romney, 1974). It is generally considered that revegetation is extremely difficult to achieve with less than 25 cm annual precipitation (Bleak *et al.*, 1965). The 13 cm at TTR falls well below this level. Since the TTR is in a cattle-grazing area, revegetation and land manipulation should be done with future grazing in mind. Initially, however, the restored area should be

protected from grazing, and possibly from rodents, in order to facilitate establishment and growth of new seedlings.

If at all possible, grass species which are capable of high grazing intensity should be encouraged, and even some new or introduced species could be used (Asay, 1975). The grass, *Hilaria jamesii* (galleta), an extremely valuable grazing plant, will be an important grass species to use for reseeding the area, since it is abundant there under natural conditions (Rhoads, 1974; Leavitt, 1975). Galleta seed production is low and poor establishment has been experienced for seeding operations on rangeland (West *et al.*, 1972); however, the amplitude of galleta is wide. The following discussion is quoted from West *et al.* (1972) because of relevance to revegetation work at the TTR.

"Throughout most of its range, especially in the northern portion, galleta endures some periods of freezing temperatures and snow cover. At all northern locations investigated, the mean daily minimum temperatures are normally below freezing from November through March. In their report on studies in the Escalante Desert where galleta is a prominent forage species, Cook and Hurst (1962) state that about 60% of the annual precipitation is snow. At the extreme northern end of its range, galleta is found only on the exposed south-facing slopes (Gibbens and Fisser, 1970) which indicates that low winter temperatures might be quite important in limiting its northward distribution.

"At the southern extremes of its range, quite the opposite situation exists. Pinkney (1969) stated that *H. jamesii* apparently is better adapted to cooler environments than the other *Hilaria* species he studied. Of the four species investigated (*H. jamesii*, *H. belangeri*, *H. mutica*, and *H. rigida*), he found galleta to grow on sites with the lowest monthly mean temperatures of 12°C and the lowest mean monthly maximum temperatures 22°C.

"From its common occurrence in various desert communities, it is apparent that galleta can endure arid environments. According to Knight *et al.* (1908) it is 'distinctively a desert grass.' Several references have been made in the literature to galleta growing well in areas where the mean annual precipitation is less than 25 cm (Bleak *et al.*, 1965; Cook and Hurst, 1962; Bridges, 1941; Hickey and Garcia, 1964; Vallentine, 1961). At Hanksville, Utah, galleta is abundant in certain plant communities where the mean annual precipitation is less than 25 cm (Bleak *et al.*, 1965; Cook and Hurst, 1962), galleta survives and appears to be in good vigor although the mean annual precipitation is only about 9.4 cm. Bridges (1941) seeded galleta on the Jornada Plain in New Mexico where precipitation during the growing season of July 1 to October 31 is under 15 cm and the potential evaporation is about 94 cm.

"Galleta is found in a wide variety of precipitation patterns. Its habitat varies from areas where most of the moisture comes during the winter and summer droughts are common (Bleak *et al.*, 1965; Cook and Hurst, 1962; Vallentine, 1961) to areas where as much as 60% of the moisture comes during the growing season (Hickey and Garcia, 1964; Jameson, 1962, 1965; Lotspeich and Everhart, 1962; Vallentine, 1961). In many of

the areas of spring and fall precipitation, separated by a summer drought, galleta displays a tendency toward two separate growing seasons with a semi-dormant period in the summer. In much of Arizona and New Mexico, there is an early spring growth period then semi-dormancy in May and June with growth starting again in July or August.

"Evidently galleta can tolerate a wide variety of topographic conditions. It has been reported on deserts, dry upland plains, sandy mesas, rocky benches and flats, canyons and open valleys, and rocky shale slopes (Gould, 1951; Graham, 1937; Hitchcock, 1951; Kearney and Peebles, 1960; Knight *et al.*, 1908; McCorkle and Heerwagen, 1951; U.S. Forest Service, 1937; Vallentine, 1961; West and Ibrahim, 1968)."

Indian ricegrass (*Oryzopsis hymenoides*) is another valuable grass species adapted to the TTR which should be experimentally encouraged in any revegetation endeavor. Scarification requirements of seed also make it difficult to reseed naturally. Seed from this species, however, can be readily collected, scarified, and planted. Irrigation may be necessary to ensure success, since Stuart *et al.* (1973) failed to establish both tall wheatgrass and Indian ricegrass in a somewhat similar area to the TTR which had only 9.4 cm of rainfall.

In addition to the grass species, the perennial shrubs *Atriplex canescens* (four winged saltbush) and *Eurotia lanata* (winterfat) have considerable grazing advantage. If these two species are used, however, it is advisable that seed collections be made from the area since genetic variability within the species has resulted in the development of distinct ecotypes adapted especially for each local environment (Wallace *et al.*, 1972). At present, *E. lanata* is rather sparse in the immediate area of the study sites at the TTR, but this species is present and can be encouraged.

Once an area such as the TTR, which is primarily an *Atriplex* shrubland, has been disturbed by scraping or plowing, it tends to revert to a grassland until reinvaded by shrubs (Wallace and Romney, 1974). Competition for water by shrubs decreases, allowing grasses to grow. It also follows that, because grasses grow first, the subsequent competition for water prevents, or retards, the establishment of new perennial shrubs. It has been postulated that the establishment of new shrubs from seed requires several consecutive years of favorable rainfall (Wallace *et al.*, 1972).

As mentioned, native animal activity is one of the big deterrents to successful revegetation in deserts. Another is overgrazing by livestock. The existing fenced study areas at the TTR present excellent examples of what can happen. Good grassland conditions now exist inside the fences even where land had been disturbed. Outside the fences, the cattle have destroyed the grass as well as much of the ability of the land to produce grass. The ecological principles involved include nitrogen fixation processes as well as competition.

The first recommendation for any mechanically disturbed land then is that it be fenced to prevent grazing for a sufficient time period to develop a grassland. The land will thus be better protected from erosion, becoming far more valuable for grazing land over a long period of time.

Three large tracts of land were bladed free of vegetation to serve as balloon launching sites for suspending vertical arrays of air sampling equipment during the 1963 Clean Slate tests at the TTR. The recovery of vegetation on these areas after 11 years was described by Wallace and Romney (1974). Again, this is Great Basin Desert, but with relatively sparse rainfall. The recovery resulted in an increased percentage of new grasses. The blading did result in some *Halogeton* invasion. This weed is poisonous to livestock and usually invades overgrazed areas. All these effects continue to be studied.

Of special interest in the deliberations concerning alternate procedures for cleanup action are the decontamination tests made 18 years ago (in 1957) in connection with the ^{239}Pu safety test conducted in Area 13 (Great Basin Desert) at the NTS (Dick and Baker, 1967). These tests provide excellent background information concerning treatment effectiveness as well as land recovery 18 years following disturbance. Even though records of long-time effect were not kept, the information available is of vital importance to the present decontamination deliberations. These areas were described in some detail by Wallace and Romney (1974). The reported cleanup effectiveness of the various treatments appears in Table 2. The water-spray treatment was designed to mix the surface-deposited ^{239}Pu into the soil sufficiently deep to decrease its resuspension hazard before the application of other decontamination operations. The test areas, which were plots 50 feet by 100 feet (15.3 x 30.6 m), were not replicated. There were 11 treatments in all.

The environs of Area 13 are described as Great Basin Desert (Beatley, 1965). Because of greater rainfall and somewhat lower evapotranspiration, as mentioned earlier, revegetation under Great Basin conditions is generally more favorable than under Mojave Desert conditions (Wallace *et al.*, 1972). The Area 13 site has somewhat less rainfall than most Great Basin Desert locations. For this reason, its recovery after 18 years has not been as great as might have been experienced elsewhere. At the present time, there is about 25% as much vegetation on the plowed site as on the undisturbed area, even less on some of the scraped sites. After 18 years, these sites are no longer grasslands, although they may have been at some intervening time. This interesting and important area continues to be studied.

Some vegetation studies have been conducted in areas disturbed by contractors in preparations for underground nuclear detonations in Hot Creek Valley, Nevada (Central Nevada Supplemental Test Area). The climate of Hot Creek Valley is semiarid. The elevation of the valley floor is about 1,700 meters. Mean yearly precipitation over a 20-year period was 13.5 cm, occurring as both snow and rain. Precipitation is distributed almost evenly throughout the year (Tueller *et al.*, 1972). Species and three seeding method trials were evaluated over a 5-year period in order to make revegetation recommendations. Findings showed that grass seeding should be made as soon as possible in the spring, preferably February or March. The reseeding studies under natural soil wetting resulted in some success, but considerable failure. Secondary invading species appeared on the disturbed areas, the most persistent of which were skeleton weed (*Erigonum deflexum*) and Russian thistle (*Salsola kali*). Survival counts after 4 years indicated that pubescent wheat-grass (*Agropyron trichophorum*) is a most successful species of grass for use in the big sagebrush habitat type. Among the native grasses which reseeded naturally were *Hilaria jamesii*, *Oryzopsis*

Table 2. Land area fixation and/or decontamination efficiencies.*

Method	Mean initial (dpm/m ²)	Mean final (dpm/m ²)	Efficiency (percent)
Plowing	2630	55	97.9
Oiling and scraping	1240	55	95.6
0.3-inch water leaching and scraping	205	15	92.7
0.3-inch water FeCl ₂	1405	118	91.6
Disking	500	54	89.2
1.0-inch water leaching	515	65	87.4
Scraping	79	11	86.0
Oiling (RC-0 road oil)	121	37	69.4
0.3-inch water leaching	8133	3660	55.0
0.3-inch water-Alconox leaching	380	309	18.7
*From Dick and Baker (1967)			

hymenoides, and *Sitanion hystrix*. Outplanting tests showed that planting of four winged saltbush (*Atriplex canescens*) in early spring can be highly successful, but irrigation is necessary if long dry periods develop before the new transplants are firmly rooted. Protection of the test plots with rabbit-proof fencing was essential for successful establishment of new seedlings (Tueller *et al.*, 1974).

If land disturbance equivalent to plowing or scraping is necessary in future land cleaning on a major scale, agricultural and engineering procedures must be considered. Adequate regard for ecosystem stability and maintenance is necessary to avoid creating new disasters which may take decades or even hundreds of years to overcome (Wells, 1961). If land must be disturbed, it is possible, and even necessary, to stabilize the remaining soil to prevent dust or water erosion. Rapid cure (RC) road oil has been used extensively at the NTS for soil stabilization and for decontamination (Brown *et al.*, 1964). Geo Tech foam (Southwest Consultants) is a commercial spray of plastic-type material which can coat soil particles and prevent erosion. While both of these materials can be used, it should be remembered that they will not replace the vegetation removed from the soil.

Although apparently discouraging, the past decontamination and soil disturbance experiences in the Great Basin Desert do indicate some tendency for the disturbed areas to mend slowly by themselves. Perhaps even more important, however, is an awareness that these experiences indicate the need for further experimentation to determine how careful application of agricultural, engineering, and ecological principles can hasten the recovery process.

RECOMMENDATIONS

In experiments conducted to help ascertain the best procedures for handling of land areas at the TTR for cleanup and restoration, the following measures should be utilized or tested further:

- a. Seeding with various species of grass and *Atriplex canescens* and *Eurotia lanata*.
- b. Fencing for grazing control.
- c. Soil manipulation which could concentrate water.
- d. Stabilization of soil with road oil.
- e. Native animal control.
- f. Partial removal of existing vegetation only.

Monitoring programs must be developed to determine efficiency of cleanup, magnitude of land erosion, and recovery of the vegetation on the land.

Some of our general recommendations on this subject continue as follows:

- a. The Pu-contaminated areas in Nevada should continue to be used for much needed research.
- b. Fencing only should be seriously considered as the accepted control measure for the most highly contaminated areas.
- c. The environmental consequences of plowing followed by oiling of the Great Basin ecosystems seem to be relatively mild after 18 years. Research on feasibility and costs for this procedure should be undertaken after experimental work is done to ascertain the true effects of plowing and oiling on revegetation. This should be done before any large areas are plowed if deemed necessary.
- d. Feasibility of developing small microcatchment basins to concentrate moisture for plant revegetation after land has been scraped should be studied further before any large areas are denuded. Various procedures for growing plants can be used. It should be possible to develop close to a dry-farm procedure for revegetation on most of the land areas under investigation.
- e. Extensive and intensive studies should be made on uses of, effect of, and the physics and chemistry of road oil stabilization of the NTS soil since it appears that road oil is a highly useful and relatively inexpensive material to decrease resuspension.
- f. Studies should be made of partial cleanup procedures which would leave from 50 to 100 perennial plants per acre, preferably grasses, reasonably undisturbed. Soil stabilization studies should be made of the soil which is disturbed.
- g. Seed collection, germination studies, and plant propagation trials should be intensified. Reasonable facilities for this work should be developed at the Nevada Test Site.
- h. Alternate methods of seeding should be evaluated in revegetation programs.
- i. Alternate means of soil stabilization should be investigated together with their impact on plant and animal activity.

CONCLUSIONS

Test areas in Nevada that have been contaminated with sufficient ^{239}Pu to cause serious consideration of some cleanup procedures should continue to be studied. The potential hazards, if any, are not yet fully understood, nor have the best methods for decontamination and land reclamation been determined. The total area where the surface contamination level is above, roughly,

1,000 pCi/cm² is about 300 acres. Only a very few acres are above 7,000 pCi/cm², the level where cleanup in the past has been done (Langham, 1968). Much has been learned about Pu contamination during the 6 years that NAEG has been studying these areas, but much more can yet be learned. These contaminated areas present an opportunity for solving many problems and, if at all possible, they should be preserved at least until the necessary research is accomplished and perhaps permanently if no cleanup is deemed necessary.

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NAEG AREA 11 SITES A, B, C, AND D,

REPORT OF VEGETATION RESULTS

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ABSTRACT

This memorandum documents the final McClellan Central Laboratory (MCL) results from plutonium, americium, and uranium analyses of Nevada Applied Ecology Group (NAEG) Area 11 vegetation samples. Specific activities of ^{238}Pu , $^{239,240}\text{Pu}$, ^{241}Am , and ^{235}U are reported for each sample.

GENERAL

A total of 86 Nevada Test Site (NTS) Area 11 vegetation samples were provided to MCL for analyses of their plutonium, americium, and uranium content. Included among these samples were overlap collections between Sites B and C, and A, B, C, and D, as well as collections from each individual site. Each sample was additionally identified by its type (genus and species) and the soil activity stratum (isopleth) from which it was collected. Vegetation types represented among the 86 samples included *Atriplex confertifolia*, *Atriplex canescens*, *Tetradymia glabrata*, *Chrysothamnus viscidiflorus*, *Lycium andersonii*, *Hymenoclea salsola*, *Grayia spinescens*, *Grayia spinosa*, *Eurotia lanata*, *Ambrosia dumosa*, and *Coleogyne ramosissima*. Soil activity strata (isopleths) were numbered from 1 to 6, with each representing a specific net FIDLER ^{241}Am cpm range. Table 1 summarizes the soil isopleths and FIDLER count rates associated with each sampling site or overlap. In the case of Site A, additional isopleths were established from Ge(Li) measurement of the ^{235}U activity (dpm) in collected soils. The ^{235}U isopleths were numbered from 1 to 3 and represented activity ranges of 0-1, 1-25, and greater than 25 dpm, respectively.

Table 1. Area 11 Soil Activity Strata (Isopleths)

SITE	ISOPLETH	^{241}Am cpm RANGE (FIDLER)
A	1	<5,000
B	2	5,000-25,000
	3	25,000-100,000
	4	100,000-500,000
C and D	2	5,000-25,000
	3	25,000-100,000
	4	100,000-500,000
	5	>500,000
CD OVERLAP	6	5,000-10,000
ABCD OVERLAP	1	<5,000

ANALYTICAL PROGRAM

All vegetation samples were ashed and dissolved at LFE. Upon receipt at MCL, each solution was quantitatively transferred to a 500 ml volumetric flask and diluted to volume. Analyses for ^{238}Pu , $^{239,240}\text{Pu}$, ^{241}Am , and ^{235}U were performed from aliquots of these diluted vegetation solutions. As the initial step, a 100 ml aliquot of each vegetation solution was provided to Ge(Li) for measurement of its ^{241}Am content. For those solutions exhibiting less than 100 dpm of ^{241}Am (Ge(Li) limit), alpha-PHA of traced americium samples was required. Estimates of the plutonium levels were made by assuming a $^{239,240}\text{Pu}/^{241}\text{Am}$ dpm ratio of 10. This assumption was subsequently confirmed by mass spectrometric measurement of the $^{239,240}\text{Pu}$ levels in four randomly selected vegetation solutions. Quantification of the ^{238}Pu and $^{239,240}\text{Pu}$ in all vegetations was achieved by alpha-PHA of ^{242}Pu traced plutonium samples. All uranium analyses were accomplished via mass spectrometric measurement of ^{233}U spiked samples. Specific activities were calculated on a dpm/gram of ash basis using the appropriate LFE ash weights and volumetric dilution factor corrections.

During the development of the MCL analytical program, the original NAEG uranium analyses requirements were modified and expanded. Initially, the uranium analyses were to be constrained to 52 vegetation samples; 39 for measurement of atoms ratios and 13 for quantification of ^{235}U . However, since mass spectrometric analyses satisfied these two analytical requirements concurrently, atom ratios and ^{235}U content were reported for each vegetation sample. In addition to this modification, the uranium analyses requirements were expanded to

include the balance of the vegetation samples. With the inclusion of these samples, a complete examination of $^{239,240}\text{Pu}$ correlation with ^{235}U was possible.

RESULTS AND DISCUSSION

Accurate interpretation of the measurement results demands that all reported specific activities be representative of the extent of sample (vegetation) contamination, i.e., necessary corrections for local background have been applied. Since Area 11 is also known as "Plutonium Valley," it was not at all surprising to find that the plutonium atom ratios from the four selected vegetations showed no requirement for background corrections. Such was not the case for the uranium. The observed $^{238}\text{U}/^{235}\text{U}$ atom ratios were influenced by the natural uranium background in degrees varying from slight to significant. In order to eliminate these natural uranium influences, mathematical expressions were derived which provided for resolution of the local background (b) and contaminant (c) fractions via known atom ratios. Detailed discussion of this mathematical technique may be found in MCL-TM-76-26 dated April 20, 1976. Briefly, the atom ratio of ^{235}U in the contaminant to ^{235}U in the background was calculated from the following general expression:

$$\frac{^{235}\text{U}_c}{^{235}\text{U}_b} = \frac{\text{U}_b - \text{U}_o}{\text{U}_o - \text{U}_c}, \quad (1)$$

where

U_b = atom ratio of isotope X to ^{235}U in the local background.

U_o = atom ratio of isotope X to ^{235}U in the observed sample, and

U_c = atom ratio of isotope X to ^{235}U in the contaminant or extraneous source.

Although values of the $^{235}\text{U}/^{235}\text{U}_b$ atom ratio may be calculated from Eq. (1) for each of the uranium isotopic atom ratios ($^{238}\text{U}/^{235}\text{U}$, $^{236}\text{U}/^{235}\text{U}$, and $^{234}\text{U}/^{235}\text{U}$), only the $^{238}\text{U}/^{235}\text{U}$ ratios were used. The reason for this decision was based solely on an inability to accurately define the $^{236}\text{U}/^{235}\text{U}$ and $^{234}\text{U}/^{235}\text{U}$ atom ratios of the contaminant. Such was not the case for the contaminant $^{238}\text{U}/^{235}\text{U}$ atom ratio. For the type of uranium assumed to have contaminated the sampling area, an uncertainty in the possible range of $^{238}\text{U}/^{235}\text{U}$ atom ratios of no greater than 2% can be accurately predicted. Once the $^{235}\text{U}/^{235}\text{U}_b$ ratios were determined, the atoms of ^{235}U per gram associated with the contaminant, $^{235}\text{U}_c$, were determined from

$$^{235}\text{U}_c = \frac{^{235}\text{U}_o \frac{^{235}\text{U}_c}{^{235}\text{U}_b}}{\frac{^{235}\text{U}_c}{^{235}\text{U}_b} + 1} \quad (2)$$

where $^{235}\text{U}_o$ is the atoms of ^{235}U in the observed sample. Conversion of the $^{235}\text{U}_o$ values to specific activities was accomplished using a ^{235}U decay constant of $1.8558 \times 10^{-15} \text{ min}^{-1}$. Values defined for U_b and U_c were $137.83 \pm 0.3\%$ and $0.0558 \pm 2.0\%$, respectively.

Individual measurement results have been arranged by sampling site and vegetation type. The specific activities of ^{238}Pu , $^{239,240}\text{Pu}$, and ^{241}Am and observed uranium atom ratios may be found in Appendix I. Appendix II contains the results of the ^{235}U calculations. Associated soil isopleths and sampling stake numbers, when known, have also been included with each sample entry.

APPENDIX I

NAEG Vegetation Results

Site A

MCL No. 7570	NAEG Number	239-40 _{Pu} dpm/g	238 _{Pu} dpm/g	241 _{Am} dpm/g	238/235	Uranium 236/235	234/235	240/239	Plutonium 241/239	Stake (ISO)
<i>ATRIPLEX CONFERTIFOLIA</i>										
5077	L13592	1.84×10^1 ± 1.5%	<0.27	2.69×10^0 ± 3.5%	31.0 ± 0.2%	0.00313 ± 1.8%	0.00991 ± 2.5%			UNK (1)
5078	L13596	5.70×10^0 ± 5.6%	1.3×10^{-1} ± 11%	8.29×10^{-1} ± 2.7%	1.56 ± 0.2%	0.00401 ± 0.5%	0.0112 ± 0.3%			UNK (2)
5079	L13597	1.06×10^1 ± 5.3%	2.34×10^{-1} ± 9.5%	1.36×10^0 ± 3.9%	1.42 ± 0.2%	0.00408 ± 0.4%	0.0113 ± 0.2%			UNK (2)
5081	L13610	7.28×10^0 ± 7.0%	2.0×10^{-1} ± 30%	9.3×10^{-1} ± 11%	1.98 ± 0.2%	0.00386 ± 1.4%	0.0111 ± 0.9%			UNK (2)
5083	L13621	9.15×10^0 ± 1.8%	2.3×10^{-1} ± 13%	9.8×10^{-1} ± 10%	4.09 ± 0.2%	0.00393 ± 1.0%	0.0110 ± 0.8%			UNK (2)
5084	L13624	4.50×10^0 ± 2.9%	1.64×10^{-1} ± 21%	6.62×10^{-1} ± 5.5%	2.47 ± 0.2%	0.00412 ± 0.7%	0.0111 ± 0.4%			UNK (2)
5086	L13634	1.18×10^1 ± 4.3%	2.64×10^{-1} ± 7.4%	1.51×10^0 ± 7.8%	1.10 ± 0.1%	0.00411 ± 0.4%	0.0113 ± 0.2%			UNK (3)
<i>TETRADYMIA GLABRATA</i>										
5080	L13605	2.90×10^1 ± 2.2%	6.3×10^{-1} ± 18%	3.17×10^0 ± 11%	0.988 ± 0.3%	0.00399 ± 0.3%	0.0112 ± 0.2%			UNK (2)
5082	L13617	1.23×10^1 ± 1.7%	2.72×10^{-1} ± 7.2%	2.2×10^0 ± 11%	2.52 ± 0.3%	0.00394 ± 0.4%	0.0112 ± 0.3%			UNK (2)

APPENDIX I (Continued)

NAEG Vegetation Results
Site A

MCL No. 7570	NAEG Number	$^{239-40}\text{Pu}$ dpm/g	^{238}Pu dpm/g	^{241}Am dpm/g	238/235	Uranium 236/235	234/235	240/239	Plutonium 241/239	Stake (ISO)
<i>ATRIPLEX CANESCENS</i>										
5076	L13585	2.46×10^1 $\pm 1.7\%$	6.99×10^{-1} $\pm 8.2\%$	6.56×10^0 $\pm 2.1\%$	21.8 $\pm 0.2\%$	0.00356 $\pm 3.3\%$	0.0107 $\pm 1.2\%$			UNK (1)
<i>CHRY. VISCIDIFLORUS</i>										
5085	L13630	9.18×10^0 $\pm 2.9\%$	1.8×10^{-1} $\pm 17\%$	1.2×10^0 $\pm 11\%$	1.10 $\pm 0.3\%$	0.00406 $\pm 0.4\%$	0.0112 $\pm 0.2\%$			UNK (3)
<i>LYCIUM ANDERSONII</i>										
5075	L13582	1.44×10^1 $\pm 3.0\%$	1.99×10^{-1} $\pm 25\%$	4.05×10^0 $\pm 5.0\%$	35.7 $\pm 0.3\%$	0.0025 $\pm 11\%$	---			UNK (1)
<i>EUROTIA LANATA</i>										
5074	L13575	7.24×10^1 $\pm 0.9\%$	1.84×10^0 $\pm 6.7\%$	7.04×10^0 $\pm 5.2\%$	24.5 $\pm 0.2\%$	0.00361 $\pm 2.1\%$	0.0105 $\pm 1.2\%$			UNK (1)

APPENDIX I (Continued)

NAEG Vegetation Results
Site B

MCL No. 7570	NAEG Number	239-40 _{Pu} dpm/g	238 _{Pu} dpm/g	241 _{Am} dpm/g	238/235	Uranium 236/235	234/235	240/239	Plutonium 241/239	Stake (ISO)
<i>ATRIPLEX CONFERTIFOLIA</i>										
5002	V12736	6.30×10^2 ± 2.4%	1.16×10^1 ± 5.2%	7.2×10^1 ± 24%	27.6 ± 0.3%	0.00355 ± 2.0%	0.0105 ± 1.9%			S 29 (2)
5005	V12752	3.80×10^3 ± 7.3%	7.34×10^1 ± 7.8%	4.22×10^2 ± 5.2%	12.0 ± 0.3%	0.00408 ± 1.0%	0.0110 ± 0.6%			O 25 (2)
5006	V12753	4.06×10^3 ± 4.5%	8.5×10^1 ± 13%	4.96×10^2 ± 3.3%	5.06 ± 0.2%	0.00429 ± 2.0%	0.0110 ± 1.0%	0.0550 ± 0.4%	0.00157 ± 0.7%	G 19 (3)
5008	V12763	1.81×10^3 ± 1.1%	3.76×10^1 ± 5.5%	2.15×10^2 ± 6.0%	8.61 ± 0.2%	0.00420 ± 0.9%	0.0110 ± 0.8%			M 25 (3)
5009	V12774	1.58×10^4 ± 1.7%	3.28×10^2 ± 5.9%	1.79×10^3 ± 2.3%	1.32 ± 0.3%	0.00416 ± 1.8%	0.0111 ± 1.0%	0.0546 ± 0.4%	0.00152 ± 0.7%	K 17 (4)
5012	V12785	4.05×10^3 ± 1.1%	8.13×10^1 ± 4.8%	4.48×10^2 ± 2.3%	3.00 ± 0.3%	0.00435 ± 0.5%	0.0112 ± 0.3%			K 17 (4)
<i>TETRADYMIA GLABRATA</i>										
5003	V12742	4.74×10^3 ± 2.9%	9.23×10^1 ± 3.7%	5.15×10^2 ± 5.3%	7.56 ± 0.2%	0.00419 ± 1.2%	0.0110 ± 0.7%			K 9 (2)
5007	V12762	3.12×10^3 ± 1.3%	6.73×10^1 ± 4.4%	3.82×10^2 ± 5.7%	10.8 ± 0.2%	0.00397 ± 1.1%	0.0110 ± 0.9%			G 9 (3)
5011	V12783	2.08×10^4 ± 1.6%	4.65×10^2 ± 7.4%	2.38×10^3 ± 5.4%	---	---	---			I 13 (4)

APPENDIX I (Continued)

NAEG Vegetation Results
Site B

MCL No. 7570	NAEG Number	$^{239-40}\text{Pu}$ dpm/g	^{238}Pu dpm/g	^{241}Am dpm/g	238/235	Uranium 236/235	234/235	240/239	Plutonium 241/239	Stake (ISO)
<i>CHRYS. VISCIDIFLORUS</i>										
5001	V12734	5.89×10^2 $\pm 1.5\%$	1.24×10^1 $\pm 6.8\%$	6.50×10^1 $\pm 7.0\%$	33.0 $\pm 0.4\%$	0.00335 $\pm 3.9\%$	0.0101 $\pm 2.2\%$			S 29 (2)
5014	V12794	1.37×10^4 $\pm 1.6\%$	2.78×10^2 $\pm 7.0\%$	1.55×10^3 $\pm 1.6\%$	2.78 $\pm 0.2\%$	0.00434 $\pm 0.6\%$	0.0112 $\pm 0.3\%$			I 13 (4)
<i>LYCIUM ANDERSONII</i>										
5004	V12744	5.19×10^2 $\pm 2.2\%$	9.6×10^0 $\pm 13\%$	6.0×10^1 $\pm 14\%$	18.2 $\pm 0.2\%$	0.00379 $\pm 3.8\%$	0.0108 $\pm 0.6\%$			C 5 (2)
5013	V12792	1.48×10^4 $\pm 1.3\%$	3.5×10^2 $\pm 16\%$	1.65×10^3 $\pm 3.3\%$	2.00 $\pm 0.3\%$	0.00437 $\pm 0.5\%$	0.0112 $\pm 0.3\%$			I 15 (4)
<i>HYMENSACLEA SALSOLA</i>										
5010	V12776	4.45×10^3 $\pm 2.4\%$	8.44×10^1 $\pm 9.9\%$	5.7×10^2 $\pm 10\%$	---	---	---			M 19 (4)

APPENDIX I (Continued)

NAEG Vegetation Results
Sites C, D

MCL No. 7570	NAEG Number	$^{239-40}\text{Pu}$ dpm/g	^{238}Pu dpm/g	^{241}Am dpm/g	238/235	Uranium 236/235	234/235	240/239	Plutonium 241/239	Stake (ISO)
<i>ATRIPLEX CONFERTIFOLIA</i>										
5051	V12805	2.58×10^2 $\pm 0.7\%$	6.55×10^0 $\pm 3.7\%$	4.61×10^1 $\pm 4.0\%$	69.6 $\pm 0.3\%$	0.00124 $\pm 6.8\%$	0.00859 $\pm 4.8\%$			UNK (6)
5016	V12808	1.81×10^2 $\pm 1.9\%$	5.53×10^0 $\pm 8.6\%$	3.0×10^1 $\pm 18\%$	66.4 $\pm 0.4\%$	0.0017 $\pm 13\%$	0.00848 $\pm 9.2\%$	0.0650 $\pm 0.3\%$	0.00204 $\pm 0.8\%$	UNK (6)
5017	V12814	3.14×10^2 $\pm 1.1\%$	8.36×10^0 $\pm 4.1\%$	4.76×10^1 $\pm 4.3\%$	---	---	---			UNK (6)

APPENDIX I (Continued)

NAEG Vegetation Results
Site C

MCL No. 7570	NAEG Number	239-40 _{Pu} dpm/g	238 _{Pu} dpm/g	241 _{Am} dpm/g	238/235	Uranium 236/235	234/235	240/239	Plutonium 241/239	Stake (ISO)
ATRIPLEX CONFERTIFOLIA										
5018	V12816	9.43×10^2 ± 4.9%	2.5×10^1 ± 11%	1.46×10^2 ± 6.6%	16.0 ± 0.2%	0.00357 ± 1.2%	0.0104 ± 0.8%			M 16 (2)
5019	V12825	7.17×10^2 ± 5.0%	2.4×10^1 ± 20%	1.08×10^2 ± 3.2%	---	---	---			G 15 (2)
5024	V12846	1.13×10^3 ± 1.8%	3.52×10^1 ± 6.7%	1.77×10^2 ± 9.5%	11.7 ± 0.2%	0.00374 ± 1.4%	0.0102 ± 1.2%			G 16 (3)
5025	V12853	1.54×10^3 ± 3.9%	4.7×10^1 ± 17%	2.60×10^2 ± 9.8%	8.82 ± 0.2%	0.00382 ± 1.1%	0.0106 ± 0.8%			G 16 (3)
5029	V12874	2.11×10^4 ± 4.0%	5.2×10^2 ± 10%	3.68×10^3 ± 3.8%	2.98 ± 0.3%	0.00402 ± 0.6%	0.0104 ± 1%			G 16 (4)
5031	V12882	9.81×10^3 ± 0.5%	2.46×10^2 ± 7.7%	1.57×10^3 ± 7.7%	4.28 ± 0.3%	0.00416 ± 1.4%	0.0107 ± 1.3%			G 16 (4)
5032	V12886	8.85×10^3 ± 1.0%	2.40×10^2 ± 4.2%	1.97×10^3 ± 2.2%	4.49 ± 0.3%	0.00403 ± 0.9%	0.0104 ± 0.8%			G 16 (4)
5033	V12893	1.08×10^4 ± 6.4%	2.85×10^2 ± 8.1%	1.91×10^3 ± 6.1%	1.87 ± 0.8%	0.00411 ± 1.2%	0.0109 ± 3.5%	0.0684 ± 0.4%	0.00216 ± 1.4%	G 15 (5)
5034	V12895	1.98×10^4 ± 2.0%	4.7×10^2 ± 10%	3.52×10^3 ± 0.6%	2.81 ± 3.4%	0.00418 ± 3.9%	0.0105 ± 1.9%			G 15 (5)

APPENDIX I (Continued)

NAEG Vegetation Results
Site C

MCL No.	7570	NAEG Number	239-40Pu dpm/g	238Pu dpm/g	241Am dpm/g	238/235	Uranium 236/235	234/235	240/239	Plutonium 241/239	Stake (ISO)
<i>TETRADYMIA GLABRATA</i>											
5022		V12833	$2.92 \times 10^3 \pm 1.2\%$	$7.80 \times 10^1 \pm 4.9\%$	$4.56 \times 10^2 \pm 5.3\%$	$20.9 \pm 0.2\%$	$0.00343 \pm 1.7\%$	$0.0102 \pm 0.8\%$			H 16 (2)
5023		V12845	$5.05 \times 10^3 \pm 2.2\%$	$1.62 \times 10^2 \pm 8.4\%$	$8.40 \times 10^2 \pm 3.7\%$	$6.36 \pm 0.2\%$	$0.00387 \pm 0.7\%$	$0.0104 \pm 0.7\%$			G 15 (3)
5030		V12876	$1.10 \times 10^4 \pm 1.4\%$	$3.21 \times 10^2 \pm 6.2\%$	$2.48 \times 10^3 \pm 4.2\%$	$6.45 \pm 0.2\%$	$0.00381 \pm 1.0\%$	$0.00979 \pm 0.9\%$			G 15 (4)
<i>ATRIPLEX CANESCENS</i>											
5020		V12826	$2.15 \times 10^3 \pm 3.2\%$	$5.93 \times 10^1 \pm 4.1\%$	$2.95 \times 10^2 \pm 2.7\%$	$15.1 \pm 0.2\%$	$0.00367 \pm 1.3\%$	$0.0101 \pm 0.8\%$			G 16 (2)
5027		V12862	$2.73 \times 10^4 \pm 3.3\%$	$7.0 \times 10^2 \pm 14\%$	$4.44 \times 10^3 \pm 1.8\%$	$3.33 \pm 0.1\%$	$0.00392 \pm 0.7\%$	$0.0103 \pm 0.7\%$			G 15 (4)
5028		V12864	$1.05 \times 10^4 \pm 1.5\%$	$2.85 \times 10^2 \pm 5.9\%$	$1.75 \times 10^3 \pm 2.7\%$	$7.87 \pm 0.2\%$	$0.00397 \pm 0.8\%$	$0.0102 \pm 1.1\%$			G 15 (4)
<i>LYCIUM ANDERSONII</i>											
5026		V12856	$3.77 \times 10^3 \pm 1.5\%$	$1.01 \times 10^2 \pm 6.3\%$	$6.45 \times 10^2 \pm 5.5\%$	$8.25 \pm 0.4\%$	$0.00426 \pm 3.4\%$	$0.0105 \pm 3.7\%$			G 15 (3)

APPENDIX I (Continued)

RESULTS
Site C

MCL No. 7570	NAEG Number	$^{239-40}\text{Pu}$ dpm/g	^{238}Pu dpm/g	^{241}Am	$^{238}/^{235}$	Uranium $^{236}/^{235}$	$^{234}/^{235}$	$^{240}/^{239}$	Plutonium $^{241}/^{239}$	Stake (ISO)
GRAYIA SPINESCENS										
5021	V12832	7.94×10^2 $\pm 2.0\%$	2.2×10^1 $\pm 13\%$	1.04×10^2 $\pm 9.3\%$	32.7 $\pm 0.3\%$	0.00299 $\pm 7.3\%$	0.00989 $\pm 2.6\%$			G 14 (2)

APPENDIX I (Continued)

NAEG Vegetation Results
Site D

MCL No. 7570	NAEG Number	$^{239-40}\text{Pu}$ dpm/g	^{238}Pu dpm/g	^{241}Am dpm/g	238/235	Uranium 236/235	234/235	240/239	Plutonium 241/239	Stake (ISO)
<i>ATRIPLEX CONFERTIFOLIA</i>										
5038	V12916	1.09×10^3 $\pm 6.2\%$	2.2×10^1 $\pm 20\%$	1.94×10^2 $\pm 5.2\%$	22.5 $\pm 0.2\%$	0.00177 $\pm 6.1\%$	0.0104 $\pm 2.0\%$			G 27 (2)
5039	V12923	2.09×10^3 $\pm 2.4\%$	4.7×10^1 $\pm 14\%$	3.77×10^2 $\pm 6.6\%$	13.0 $\pm 0.4\%$	0.00125 $\pm 2.6\%$	0.0100 $\pm 3.8\%$			G 25 (3)
5043	V12942	4.85×10^3 $\pm 1.3\%$	1.27×10^2 $\pm 6.0\%$	9.85×10^2 $\pm 2.4\%$	5.20 $\pm 0.1\%$	0.00145 $\pm 1.4\%$	0.0115 $\pm 0.5\%$			K 25 (4)
5044	V12946	1.44×10^4 $\pm 4.9\%$	4.09×10^2 $\pm 8.4\%$	2.33×10^3 $\pm 3.0\%$	3.27 $\pm 1.0\%$	0.00146 $\pm 8.5\%$	0.0113 $\pm 1.0\%$			G 17 (4)
5046	V12959	3.06×10^3 $\pm 2.2\%$	7.89×10^1 $\pm 7.2\%$	5.47×10^2 $\pm 4.9\%$	5.13 $\pm 2.1\%$	0.0024 $\pm 27\%$	0.0098 $\pm 15\%$			O 23 (4)
5047	V12963	2.55×10^3 $\pm 1.6\%$	6.38×10^1 $\pm 7.0\%$	4.46×10^2 $\pm 3.9\%$	16.2 $\pm 0.2\%$	0.00137 $\pm 2.5\%$	0.0111 $\pm 0.8\%$			K 27 (4)
5050	V12978	1.61×10^4 $\pm 1.6\%$	4.85×10^2 $\pm 7.7\%$	3.75×10^3 $\pm 2.0\%$	2.92 $\pm 0.2\%$	0.00151 $\pm 0.8\%$	0.0116 $\pm 0.3\%$			I 19 (5)
5051	V12984	6.04×10^4 $\pm 2.6\%$	1.5×10^3 $\pm 15\%$	1.12×10^4 $\pm 6.9\%$	1.54 $\pm 0.2\%$	0.00150 $\pm 1.4\%$	0.0116 $\pm 0.3\%$			I 17 (5)
5052	V12987	2.63×10^4 $\pm 2.3\%$	6.9×10^2 $\pm 10\%$	4.95×10^3 $\pm 4.3\%$	2.34 $\pm 0.3\%$	0.00156 $\pm 1.7\%$	0.0116 $\pm 0.6\%$			K 17 (5)

APPENDIX I (Continued)

NAEG Vegetation Results

Site D

MCL No. 7570	NAEG Number	$^{239-40}\text{Pu}$ dpm/g	^{238}Pu dpm/g		238/235	Uranium 236/235	234/235	240/239	Plutonium 241/239	Stake (ISO)
<i>ETRADYMIA GLABRATA</i>										
5035	V12902	2.64×10^3 $\pm 1.5\%$	7.33×10^1 $\pm 6.6\%$	4.31×10^2 $\pm 3.4\%$	21.6 $\pm 0.3\%$	0.00267 $\pm 3.4\%$	0.00970 $\pm 3.4\%$			Y 09 (2)
5036	V12906	7.70×10^3 $\pm 1.1\%$	1.64×10^2 $\pm 7.2\%$	10^2 $\pm 0.2\%$	12.7 $\pm 0.2\%$	0.00140 $\pm 5.8\%$	0.0112 $\pm 0.5\%$			S 03 (2)
5037	V12914	1.74×10^3 $\pm 3.3\%$	4.6×10^1 $\pm 12\%$	2.97×10^2 $\pm 9.4\%$	34.5 $\pm 0.2\%$	0.00141 $\pm 4.1\%$	0.0103 $\pm 1.7\%$			H 12 (2)
5040	V12927	8.47×10^2 $\pm 1.5\%$	2.00×10^1 $\pm 7.1\%$	1.26×10^2 $\pm 7.5\%$	52.3 $\pm 0.2\%$	---	---			E 05 (3)
5042	V12935	1.54×10^4 $\pm 1.8\%$	4.13×10^2 $\pm 8.4\%$	2.39×10^3 $\pm 2.3\%$	3.69 $\pm 0.3\%$	0.00144 $\pm 3.4\%$	0.0118 $\pm 1.9\%$			O 02 (3)
<i>ATRIPLEX CANESCENS</i>										
5048	V12965	8.49×10^3 $\pm 2.0\%$	2.28×10^2 $\pm 8.9\%$	1.33×10^3 $\pm 3.4\%$	5.72 $\pm 0.2\%$	0.00142 $\pm 3.4\%$	0.0113 $\pm 0.7\%$			Q 17 (4)
5049	V12976	9.36×10^3 $\pm 1.4\%$	2.71×10^2 $\pm 4.8\%$	1.70×10^3 $\pm 1.8\%$	2.87 $\pm 0.2\%$	0.00148 $\pm 1.6\%$	0.0116 $\pm 0.6\%$			M 23 (5)

APPENDIX I (Continued)

NAEG Vegetation Results
Site D

MCL No.	NAEG Number	239-40Pu dpm/g	238Pu dpm/g	241Am dpm/g	238/235	Uranium 236/235	234/235	240/239	Plutonium 241/239	Stake (ISO)
<i>CHRYS. VISCIDIFLORUS</i>										
5045	V12952	1.19×10^4 ± 1.1%	3.29×10^2 ± 7.0%	1.90×10^3 ± 6.0%	4.97 ± 0.3%	0.00150 ± 4.3%	0.0115 ± 1.8%			I 21 (4)
<i>LYCIUM ANDERSONII</i>										
5041	V12932	1.32×10^3 ± 2.1%	3.27×10^1 ± 9.5%	2.0×10^2 ± 19%	20.3 ± 0.9%	0.0015 ± 21%	0.0114 ± 3.3%			I 03 (3)

APPENDIX I (Continued)

NAEG Vegetation Results
Sites A, B, C, D

MCL No. 7570	NAEG Number	239-40 _{Pu} dpm/g	238 _{Pu} dpm/g	241 _{Am} dpm/g	238/235	Uranium 236/235	234/235	240/239	Plutonium 241/239	Stake (ISO)
<i>ATRIPLEX CONFERTIFOLIA</i>										
5057	V13012	2.19×10^1 ± 13%	5.17×10^{-1} ± 16%	3.90×10^0 ± 18%	126 ± 0.3%	0.00076 ± 16%	0.00840 ± 3.9%			D 11 (1)
5058	V13015	1.43×10^1 ± 4.6%	3.39×10^{-1} ± 9.1%	2.3×10^0 ± 13%	122 ± 0.5%	<0.0008	0.0050 ± 18%			J 24 (1)
5061	V13033	1.96×10^1 ± 2.8%	5.05×10^{-1} ± 8.2%	3.28×10^0 ± 4.0%	117 ± 0.4%	<0.0002	0.0060 ± 15%			K 22 (1)
5062	V13035	3.51×10^0 ± 4.0%	9.04×10^{-2} ± 9.4%	5.59×10^{-1} ± 3.1%	135 ± 2.7%	0.00060 ± 28%	0.00627 ± 9.7%			A 08 (1)
5065	V13056	3.19×10^0 ± 4.0%	8.5×10^{-2} ± 10%	4.05×10^{-1} ± 9.3%	10.9 ± 2.5%	0.00355 ± 2.3%	0.0111 ± 0.6%			D 25 (1)
5070	V13079	2.91×10^1 ± 1.6%	6.82×10^{-1} ± 3.2%	5.55×10^0 ± 2.7%	120 ± 0.2%	0.00036 ± 15%	0.00917 ± 2.1%			I 08 (1)
5072	V13083	1.03×10^1 ± 9.7%	2.1×10^{-1} ± 19%	2.05×10^0 ± 4.2%	126 ± 0.4%	0.00053 ± 45%	0.00866 ± 3.6%			M 24 (1)
<i>TETRADYMIA GLABRATA</i>										
5053	V12993	3.83×10^2 ± 2.4%	1.29×10^1 ± 6.6%	3.68×10^1 ± 9.5%	104 ± 0.2%	0.00071 ± 15%	0.00879 ± 3.8%			I 24 (1)
5059	V13025	2.51×10^2 ± 3.3%	6.29×10^0 ± 7.3%	4.00×10^1 ± 2.8%	84.2 ± 0.3%	0.0015 ± 13%	0.00885 ± 2.7%			H 20 (1)

APPENDIX I (Continued)

NAEG Vegetation Results

Sites A, B, C, D

MCL No. 7570	NAEG Number	$^{239-40}\text{Pu}$ dpm/g	^{238}Pu dpm/g	^{241}Am dpm/g	238/235	Uranium 236/235	234/235	240/239	Plutonium 241/239	Stake (ISO)
5064	V13046	7.49×10^1 $\pm 2.4\%$	2.1×10^0 $\pm 17\%$	1.18×10^1 $\pm 4.8\%$	118 $\pm 0.2\%$	0.00052 $\pm 25\%$	0.00857 $\pm 3.4\%$			B 20 (1)
5069	V13076	2.00×10^1 $\pm 2.4\%$	4.6×10^{-1} $\pm 19\%$	2.83×10^0 $\pm 7.1\%$	50.0 $\pm 1.0\%$	0.00250 $\pm 5.8\%$	0.0100 $\pm 1.5\%$			C 25 (1)
5073	V13094	1.25×10^1 $\pm 3.4\%$	3.29×10^{-1} $\pm 9.4\%$	2.03×10^0 $\pm 5.0\%$	134 $\pm 0.3\%$	<0.0002	0.0055 $\pm 10\%$			C 15 (1)
<i>ATRIPLEX CANESCENS</i>										
5054	V12997	2.02×10^2 $\pm 1.4\%$	5.12×10^0 $\pm 5.7\%$	2.31×10^1 $\pm 3.0\%$	82.4 $\pm 2.2\%$	0.0042 $\pm 33\%$	<0.009			T 20 (1)
<i>CHRY. VISCIDIFLORUS</i>										
5060	V13027	1.76×10^1 $\pm 5.1\%$	5.14×10^{-1} $\pm 7.5\%$	2.03×10^0 $\pm 3.9\%$	135 $\pm 0.3\%$	<0.0002	0.00760 $\pm 3.1\%$			A 09 (1)
5063	V13042	1.26×10^1 $\pm 4.5\%$	3.4×10^{-1} $\pm 11\%$	1.76×10^0 $\pm 5.0\%$	136 $\pm 1.1\%$	0.00062 $\pm 20\%$	0.00901 $\pm 4.9\%$			G 05 (1)
<i>EUROTIA LANATA</i>										
5055	V13005	2.48×10^1 $\pm 2.8\%$	6.79×10^{-1} $\pm 5.4\%$	4.94×10^0 $\pm 1.8\%$	129 $\pm 0.2\%$	0.00036 $\pm 21\%$	0.00841 $\pm 5.3\%$			I 05 (1)

APPENDIX I (Continued)

NAEG Vegetation Results
Sites A, B, C, D

MCL No. 7570	NAEG Number	$^{239-40}\text{Pu}$ dpm/g	^{238}Pu dpm/g	^{241}Am dpm/g	238/235	Uranium 236/235	234/235	240/239	Plutonium 241/239	Stake (ISO)
5056	V13008	1.57×10^1 $\pm 2.9\%$	3.90×10^{-1} $\pm 5.6\%$	2.24×10^0 $\pm 4.0\%$	136 $\pm 0.5\%$	0.00051 $\pm 25\%$	0.00879 $\pm 8.7\%$			D 01 (1)
5066	V13057	1.91×10^2 $\pm 2.6\%$	4.63×10^0 $\pm 5.1\%$	3.25×10^1 $\pm 2.6\%$	65.5 $\pm 8.1\%$	0.00190 $\pm 6.9\%$	0.00953 $\pm 3.0\%$			G 19 (1)
<i>GRAYIA SPINOSA</i>										
5067	V13062	8.72×10^0 $\pm 2.0\%$	2.9×10^{-1} $\pm 15\%$	1.13×10^0 $\pm 2.7\%$	122 $\pm 0.8\%$	---	---			E 13 (1)
<i>AMROSIA DUMOSA</i>										
5068	V13064	5.44×10^1 $\pm 1.5\%$	1.45×10^0 $\pm 9.5\%$	9.62×10^0 $\pm 3.1\%$	113 $\pm 0.4\%$	0.00042 $\pm 32\%$	0.00769 $\pm 9.4\%$			L 21 (1)
<i>COLEOGYNE RAMOSISSIMA</i>										
5071	V13082	5.45×10^1 $\pm 3.0\%$	1.24×10^0 $\pm 9.8\%$	8.74×10^0 $\pm 4.8\%$	126 $\pm 0.2\%$	0.00031 $\pm 22\%$	0.00738 $\pm 7.8\%$			M 23 (1)

APPENDIX II

NAEG Vegetation Results
Site A

MCL No. 7570	NAEG Number	^{235}U atoms/g (0)	$^{235}\text{U}/^{235}\text{C}$	^{235}U atoms/g (C)	^{235}U dpm/g (C)	$^{239+40}\text{Pu}$ dpm/g	Stake (ISO)
<i>ATRIPLEX CONFERTIFOLIA</i>							
5077	L13592	1.89×10^{13} $\pm 0.3\%$	3.46 $\pm 0.4\%$	1.47×10^{13} $\pm 0.3\%$	2.72×10^{-2} $\pm 0.3\%$	1.84×10^1 $\pm 1.5\%$	UNK (1)
5078	L13596	8.87×10^{14} $\pm 0.3\%$	90.6 $\pm 0.3\%$	8.78×10^{14} $\pm 0.3\%$	1.63×10^0 $\pm 0.3\%$	5.70×10^0 $\pm 5.6\%$	UNK (2)
5079	L13597	6.08×10^{14} $\pm 0.4\%$	99.9 $\pm 0.3\%$	6.02×10^{14} $\pm 0.4\%$	1.12×10^0 $\pm 0.4\%$	1.06×10^1 $\pm 5.3\%$	UNK (2)
5081	L13610	6.36×10^{14} $\pm 0.4\%$	70.4 $\pm 0.3\%$	6.28×10^{14} $\pm 0.4\%$	1.16×10^0 $\pm 0.4\%$	7.28×10^0 $\pm 7.0\%$	UNK (2)
5083	L13621	2.06×10^{14} $\pm 0.3\%$	33.2 $\pm 0.4\%$	2.00×10^{14} $\pm 0.3\%$	3.71×10^{-1} $\pm 0.3\%$	9.15×10^0 $\pm 1.8\%$	UNK (2)
5084	L13624	2.78×10^{14} $\pm 0.3\%$	56.0 $\pm 0.3\%$	2.73×10^{14} $\pm 0.3\%$	5.07×10^{-1} $\pm 0.3\%$	4.50×10^0 $\pm 2.9\%$	UNK (2)
5086	L13634	1.15×10^{15} $\pm 0.3\%$	131 $\pm 0.3\%$	1.15×10^{14} $\pm 0.3\%$	2.13×10^0 $\pm 0.3\%$	1.18×10^1 $\pm 4.3\%$	UNK (3)
<i>TETRADYMIA GLABRATA</i>							
5080	L13605	9.19×10^{15} $\pm 0.6\%$	147 $\pm 0.5\%$	9.13×10^{15} $\pm 0.6\%$	1.69×10^1 $\pm 0.6\%$	2.90×10^1 $\pm 2.2\%$	UNK (2)
5082	L13617	1.07×10^{15} $\pm 0.5\%$	55.0 $\pm 0.4\%$	1.05×10^{15} $\pm 0.5\%$	1.94×10^0 $\pm 0.5\%$	1.23×10^1 $\pm 1.7\%$	UNK (2)

APPENDIX II (Continued)

NAEG Vegetation Results

Site A

MCL No. 7570	NAEG Number	^{235}U atoms/g (0)	$^{235}\text{U}/^{235}\text{B}$	^{235}U atoms/g (C)	^{235}U dpm/g (C)	$^{239+40}\text{Pu}$ dpm/g	Stake (ISO)
<i>ATRIPLEX CANESCENS</i>							
5076	L13585	1.83×10^{13} $\pm 0.4\%$	5.33 $\pm 0.4\%$	1.54×10^{13} $\pm 0.4\%$	2.86×10^{-2} $\pm 0.4\%$	2.46×10^1 $\pm 1.7\%$	UNK (1)
<i>CHRY. VISCIDIFLORUS</i>							
5085	L13630	1.63×10^{15} $\pm 0.4\%$	131 $\pm 0.4\%$	1.62×10^{15} $\pm 0.4\%$	3.00×10^0 $\pm 0.4\%$	9.18×10^0 $\pm 2.9\%$	UNK (3)
<i>LYCIUM ANDERSONII</i>							
5075	L13582	1.32×10^{13} $\pm 0.5\%$	2.87 $\pm 0.6\%$	9.76×10^{12} $\pm 0.5\%$	1.81×10^{-2} $\pm 0.5\%$	1.44×10^1 $\pm 3.0\%$	UNK (1)
<i>EUROTIA LANATA</i>							
5074	L13575	4.91×10^{13} $\pm 0.3\%$	4.64 $\pm 0.4\%$	4.04×10^{13} $\pm 0.4\%$	7.49×10^{-2} $\pm 0.4\%$	7.24×10^1 $\pm 0.9\%$	UNK (1)

APPENDIX II (Continued)

NAEG Vegetation Results

Site B

MCL No. 7570	NAEG Number	^{235}U atoms/g (0)	$^{235}\text{U}/^{235}\text{B}$	^{235}U atoms/g (C)	^{235}U dpm/g (C)	$^{239+40}\text{Pu}$ dpm/g	Stake (ISO)
<i>ATRIPLEX CONFERTIFOLIA</i>							
5002	V12736	2.26×10^{13} $\pm 0.5\%$	4.00 $\pm 0.5\%$	1.80×10^{13} $\pm 0.6\%$	3.35×10^{-2} $\pm 0.6\%$	6.30×10^2 $\pm 2.4\%$	S 29 (2)
5005	V12752	1.38×10^{14} $\pm 0.5\%$	10.6 $\pm 0.4\%$	1.26×10^{14} $\pm 0.5\%$	2.34×10^{-1} $\pm 0.5\%$	3.80×10^3 $\pm 7.3\%$	O 25 (2)
5006	V12753	1.52×10^{14} $\pm 0.4\%$	26.5 $\pm 0.4\%$	1.46×10^{14} $\pm 0.4\%$	2.72×10^{-1} $\pm 0.4\%$	4.06×10^3 $\pm 4.5\%$	G 19 (3)
5008	V12763	5.93×10^{13} $\pm 0.3\%$	15.1 $\pm 0.3\%$	5.56×10^{13} $\pm 0.3\%$	1.03×10^{-1} $\pm 0.3\%$	1.81×10^3 $\pm 1.1\%$	M 25 (3)
5009	V12774	5.41×10^{14} $\pm 0.7\%$	108 $\pm 0.4\%$	5.36×10^{14} $\pm 0.7\%$	9.95×10^{-1} $\pm 0.7\%$	1.58×10^4 $\pm 1.7\%$	K 17 (4)
5012	V12785	1.38×10^{14} $\pm 0.4\%$	45.8 $\pm 0.4\%$	1.36×10^{14} $\pm 0.4\%$	2.52×10^{-1} $\pm 0.4\%$	4.05×10^3 $\pm 1.1\%$	K 17 (4)
<i>TETRADYMIA GLABRATA</i>							
5003	V12742	1.57×10^{14} $\pm 0.4\%$	17.4 $\pm 0.4\%$	1.48×10^{14} $\pm 0.4\%$	2.75×10^{-1} $\pm 0.4\%$	4.74×10^3 $\pm 2.9\%$	K 9 (2)
5007	V12762	1.30×10^{14} $\pm 0.3\%$	11.8 $\pm 0.3\%$	1.19×10^{14} $\pm 0.3\%$	2.22×10^{-1} $\pm 0.3\%$	3.12×10^3 $\pm 1.3\%$	G 9 (3)
5011	V12783	---	---	---	---	2.08×10^4 $\pm 1.6\%$	I 13 (4)

APPENDIX II (Continued)

NAEG Vegetation Results
Site B

MCL No. 7570	NAEG Number	^{235}U atoms/g (O)	$^{235}\text{U}/^{235}\text{B}$	^{235}U atoms/g (C)	^{235}U dpm/g (C)	$^{239+40}\text{Pu}$ dpm/g	Stake (ISO)
<i>CHRY. VISCIDIFLORUS</i>							
5001	V12734	2.27×10^{13} $\pm 0.5\%$	3.18 $\pm 0.6\%$	1.73×10^{13} $\pm 0.6\%$	3.20×10^{-2} $\pm 0.6\%$	5.89×10^2 $\pm 1.5\%$	S 29 (2)
5014	V12794	5.20×10^{14} $\pm 0.3\%$	49.6 $\pm 0.3\%$	5.10×10^{14} $\pm 0.3\%$	9.46×10^{-1} $\pm 0.3\%$	1.37×10^4 $\pm 1.6\%$	I 13 (4)
<i>LYCIUM ANDERSONII</i>							
5004	V12744	3.06×10^{13} $\pm 0.9\%$	6.59 $\pm 0.4\%$	2.65×10^{13} $\pm 0.9\%$	4.93×10^{-2} $\pm 0.9\%$	5.19×10^2 $\pm 2.2\%$	C 5 (2)
5013	V12792	5.59×10^{14} $\pm 0.5\%$	69.9 $\pm 0.4\%$	5.51×10^{14} $\pm 0.5\%$	1.02×10^0 $\pm 0.5\%$	1.48×10^4 $\pm 1.3\%$	I 15 (4)
<i>HYMENSCLEA SALSOLA</i>							
5010	V12776	---	---	---	---	4.45×10^3 $\pm 2.4\%$	M 19 (4)

APPENDIX II (Continued)

NAEG Vegetation Results
Sites C, D

MCL No. 7570	NAEG Number	^{235}U atoms/g (0)	$^{235}\text{U}/^{235}\text{B}$	^{235}U atoms/g (C)	^{235}U dpm/g (C)	$^{239+40}\text{Pu}$ dpm/g	Stake (ISO)
<i>ATRIPLEX CONFERTIFOLIA</i>							
5015	V12805	8.00×10^{12} $\pm 0.4\%$	0.983 $\pm 0.7\%$	3.96×10^{12} $\pm 0.6\%$	7.35×10^{-3} $\pm 0.6\%$	2.58×10^2 $\pm 0.7\%$	UNK (6)
5016	V12808	5.25×10^{12} $\pm 0.5\%$	1.08 $\pm 1.0\%$	2.72×10^{12} $\pm 0.9\%$	5.05×10^{-3} $\pm 0.9\%$	1.81×10^2 $\pm 1.9\%$	UNK (6)
5017	V12814	---	---	---	---	3.14×10^2 $\pm 1.1\%$	UNK (6)

APPENDIX II (Continued)

NAEG Vegetation Results
Site C

MCL No. 7570	NAEG Number	^{235}U atoms/g (0)	$^{235}\text{U}/^{235}\text{B}$	^{235}U atoms/g (C)	^{235}U dpm/g (C)	$^{239+40}\text{Pu}$ dpm/g	Stake (ISO)
ATRIPLEX CONFERTIFOLIA							
5018	V12816	5.20×10^{13} $\pm 0.4\%$	7.66 $\pm 0.4\%$	4.60×10^{13} $\pm 0.5\%$	8.54×10^{-2} $\pm 0.5\%$	9.43×10^2 $\pm 4.9\%$	M 16 (2)
5019	V12825	---	---	---	---	7.17×10^2 $\pm 5.0\%$	G 15 (2)
5024	V12846	3.04×10^{13} $\pm 0.3\%$	10.8 $\pm 0.3\%$	2.79×10^{13} $\pm 0.3\%$	5.17×10^{-2} $\pm 0.3\%$	1.13×10^3 $\pm 1.8\%$	G 16 (3)
5025	V12853	5.47×10^{13} $\pm 0.4\%$	14.7 $\pm 0.4\%$	5.12×10^{13} $\pm 0.4\%$	9.51×10^{-2} $\pm 0.4\%$	1.54×10^3 $\pm 3.9\%$	G 16 (3)
5029	V12874	3.75×10^{14} $\pm 4.7\%$	46.1 $\pm 0.4\%$	3.67×10^{14} $\pm 4.7\%$	6.81×10^{-1} $\pm 4.7\%$	2.11×10^4 $\pm 4.0\%$	G 16 (4)
5031	V12886	1.47×10^{14} $\pm 0.9\%$	31.6 $\pm 0.4\%$	1.42×10^{14} $\pm 0.9\%$	2.64×10^{-1} $\pm 0.9\%$	9.81×10^3 $\pm 0.5\%$	G 16 (4)
5032	V12886	1.59×10^{14} $\pm 0.3\%$	30.1 $\pm 0.4\%$	1.53×10^{14} $\pm 0.3\%$	2.85×10^{-1} $\pm 0.3\%$	8.85×10^3 $\pm 1.0\%$	G 16 (4)
5033	V12893	3.35×10^{14} $\pm 2.4\%$	74.8 $\pm 0.8\%$	3.30×10^{14} $\pm 2.4\%$	6.13×10^{-1} $\pm 2.4\%$	1.08×10^4 $\pm 6.4\%$	G 15 (5)
5034	V12895	2.12×10^{14} $\pm 0.4\%$	49.1 $\pm 0.4\%$	2.07×10^{14} $\pm 0.8\%$	3.85×10^{-1} $\pm 0.8\%$	1.98×10^4 $\pm 2.0\%$	G 15 (5)

APPENDIX II (Continued)

NAEG Vegetation Results
Site C

MCL No. 7570	NAEG Number	^{235}U atoms/g (0)	$^{235}\text{U}/^{235}\text{B}$	^{235}U atoms/g (C)	^{235}U dpm/g (C)	$^{239+40}\text{Pu}$ dpm/g	Stake (ISO)
<i>TETRADYMIA GLABRATA</i>							
5022	V12833	7.45×10^{13} $\pm 0.3\%$	5.60 $\pm 0.4\%$	6.32×10^{13} $\pm 0.3\%$	1.17×10^{-1} $\pm 0.3\%$	2.92×10^3 $\pm 1.2\%$	H 16 (2)
5023	V12845	1.50×10^{14} $\pm 0.3\%$	20.9 $\pm 0.3\%$	1.43×10^{14} $\pm 0.3\%$	2.66×10^{-1} $\pm 0.3\%$	5.05×10^3 $\pm 2.2\%$	G 15 (3)
5030	V12876	2.05×10^{14} $\pm 0.3\%$	20.5 $\pm 0.3\%$	1.95×10^{14} $\pm 0.3\%$	3.62×10^{-1} $\pm 0.3\%$	1.10×10^4 $\pm 1.4\%$	G 15 (4)
<i>ATRIPLEX CANESCENS</i>							
5020	V12826	4.51×10^{13} $\pm 0.3\%$	8.17 $\pm 0.3\%$	4.01×10^{13} $\pm 0.3\%$	7.45×10^{-2} $\pm 0.3\%$	2.15×10^3 $\pm 3.2\%$	G 16 (2)
5027	V12862	4.03×10^{14} $\pm 0.3\%$	41.1 $\pm 0.3\%$	3.94×10^{14} $\pm 0.3\%$	7.31×10^{-1} $\pm 0.3\%$	2.73×10^4 $\pm 3.3\%$	G 15 (4)
5028	V12864	1.14×10^{14} $\pm 1.4\%$	16.6 $\pm 0.3\%$	1.07×10^{14} $\pm 1.4\%$	1.99×10^{-1} $\pm 1.4\%$	1.05×10^4 $\pm 1.5\%$	G 15 (4)
<i>LYCIUM ANDERSONII</i>							
5026	V12856	5.98×10^{13} $\pm 0.6\%$	15.8 $\pm 0.5\%$	5.63×10^{13} $\pm 0.6\%$	1.04×10^{-1} $\pm 0.6\%$	3.77×10^3 $\pm 1.5\%$	G 15 (3)

APPENDIX II (Continued)

NAEG Vegetaion Results
Site C

MCL No. 7570	NAEG Number	^{235}U atoms/g (0)	$^{235}\text{U}/^{235}\text{B}$ $_{\text{C}}$	^{235}U atoms/g (C)	^{235}U dpm/g (C)	$^{239+40}\text{Pu}$ dpm/g	Stake (ISO)
<i>GRAYIA SPINESCENS</i>							
5021	V12832	1.62×10^{13} $\pm 0.4\%$	3.22 $\pm 0.5\%$	1.24×10^{13} $\pm 0.5\%$	2.30×10^{-2} $\pm 0.5\%$	7.94×10^2 $\pm 2.0\%$	G 14 (2)

APPENDIX II (Continued)

NAEG Vegetation Results
Site D

MCL No.	NAEG Number	^{235}U atoms/g (O)	$^{235}\text{U}/^{235}\text{C}$ B	^{235}U atoms/g (C)	^{235}U dpm/g (C)	$^{239+40}\text{Pu}$ dpm/g	Stake (ISO)
ATRIPLEX CONFERTIFOLIA							
5038	V12916	$1.82 \times 10^{13} \pm 0.3\%$	$5.14 \pm 0.4\%$	$1.53 \times 10^{13} \pm 0.3\%$	$2.83 \times 10^{-2} \pm 0.3\%$	$1.09 \times 10^3 \pm 6.2\%$	G 27 (2)
5039	V12923	$3.65 \times 10^{13} \pm 0.6\%$	$9.62 \pm 0.6\%$	$3.31 \times 10^{13} \pm 0.6\%$	$6.14 \times 10^{-2} \pm 0.6\%$	$2.09 \times 10^3 \pm 2.4\%$	G 25 (3)
5043	V12942	$1.29 \times 10^{14} \pm 0.3\%$	$25.8 \pm 0.3\%$	$1.24 \times 10^{14} \pm 0.3\%$	$2.30 \times 10^{-1} \pm 0.3\%$	$4.85 \times 10^3 \pm 1.3\%$	K 25 (4)
5044	V12946	$1.62 \times 10^{14} \pm 3.0\%$	$41.8 \pm 1.0\%$	$1.59 \times 10^{14} \pm 3.1\%$	$2.94 \times 10^{-1} \pm 3.1\%$	$1.44 \times 10^4 \pm 4.9\%$	G 17 (4)
5046	V12959	$8.11 \times 10^{13} \pm 3.3\%$	$26.1 \pm 2.2\%$	$7.82 \times 10^{13} \pm 3.4\%$	$1.45 \times 10^{-1} \pm 3.4\%$	$3.06 \times 10^3 \pm 2.2\%$	O 23 (4)
5047	V12963	$5.41 \times 10^{13} \pm 0.4\%$	$7.51 \pm 0.4\%$	$4.77 \times 10^{13} \pm 0.4\%$	$8.86 \times 10^{-2} \pm 0.4\%$	$2.55 \times 10^3 \pm 1.6\%$	K 27 (4)
5050	V12978	$2.60 \times 10^{14} \pm 0.4\%$	$47.1 \pm 0.4\%$	$2.55 \times 10^{14} \pm 0.4\%$	$4.73 \times 10^{-1} \pm 0.4\%$	$1.61 \times 10^4 \pm 1.6\%$	I 19 (5)
5051	V12984	$9.69 \times 10^{14} \pm 0.3\%$	$91.6 \pm 0.3\%$	$9.59 \times 10^{14} \pm 0.3\%$	$1.78 \times 10^0 \pm 0.3\%$	$6.04 \times 10^4 \pm 2.6\%$	I 17 (5)
5052	V12987	$4.24 \times 10^{14} \pm 0.5\%$	$59.4 \pm 0.4\%$	$4.17 \times 10^{14} \pm 0.5\%$	$7.73 \times 10^{-1} \pm 0.5\%$	$2.63 \times 10^4 \pm 2.3\%$	K 17 (5)

APPENDIX II (Continued)

NAEG Vegetation Results

Site D

MCL No. 7570	NAEG Number	^{235}U atoms/g (0)	$^{235}\text{U}/^{235}\text{B}$ ^{235}C	^{235}U atoms/g (C)	^{235}U dpm/g (C)	$^{239+40}\text{Pu}$ dpm/g	Stake (ISO)
<i>TETRADYMIA GLABRATA</i>							
5035	V12902	4.83×10^{13} $\pm 0.4\%$	5.40 $\pm 0.5\%$	4.08×10^{13} $\pm 0.4\%$	7.56×10^{-2} $\pm 0.4\%$	2.64×10^3 $\pm 1.5\%$	Y 09 (2)
5036	V12906	8.40×10^{13} $\pm 2.4\%$	9.94 $\pm 0.4\%$	7.63×10^{13} $\pm 2.4\%$	1.42×10^{-1} $\pm 2.4\%$	7.70×10^3 $\pm 1.1\%$	S 03 (2)
5037	V12914	3.34×10^{13} $\pm 0.4\%$	3.00 $\pm 0.4\%$	2.51×10^{13} $\pm 0.4\%$	4.65×10^{-2} $\pm 0.4\%$	1.74×10^3 $\pm 3.3\%$	H 12 (2)
5040	V12927	2.11×10^{13} $\pm 2.7\%$	1.64 $\pm 0.6\%$	1.31×10^{13} $\pm 2.8\%$	2.43×10^{-2} $\pm 2.8\%$	8.47×10^2 $\pm 1.5\%$	E 05 (3)
5042	V12935	1.83×10^{14} $\pm 0.6\%$	36.9 $\pm 0.4\%$	1.78×10^{14} $\pm 0.6\%$	3.30×10^{-1} $\pm 0.6\%$	1.54×10^4 $\pm 1.8\%$	O 02 (3)
<i>ATRIplex CANESCENS</i>							
5048	V12965	1.80×10^{14} $\pm 0.5\%$	23.3 $\pm 0.4\%$	1.73×10^{14} $\pm 0.5\%$	3.21×10^{-1} $\pm 0.5\%$	8.49×10^3 $\pm 2.0\%$	Q 17 (4)
5049	V 12976	1.75×10^{14} $\pm 0.3\%$	47.9 $\pm 0.3\%$	1.72×10^{14} $\pm 0.3\%$	3.18×10^{-1} $\pm 0.3\%$	9.36×10^3 $\pm 1.4\%$	M 23 (5)

APPENDIX II (Continued)

NAEG Vegetation Results
Site D

MCL No. 7570	NAEG Number	^{235}U atoms/g (O)	$^{235}\text{U}/^{235}\text{B}$	^{235}U atoms/g (C)	^{235}U dpm/g (C)	$^{239+40}\text{Pu}$ dpm/g	Stake (ISO)
<i>CHRY. VISCIDIFLORUS</i>							
5045	V12952	2.04×10^{14} $\pm 0.6\%$	27.0 $\pm 0.4\%$	1.97×10^{14} $\pm 0.6\%$	3.65×10^{-1} $\pm 0.6\%$	1.19×10^4 $\pm 1.1\%$	I 21 (4)
<i>LYCIUM ANDERSONII</i>							
5041	V12932	1.89×10^{13} $\pm 6.6\%$	5.80 $\pm 1.1\%$	1.62×10^{13} $\pm 6.6\%$	3.00×10^{-2} $\pm 6.6\%$	1.32×10^3 $\pm 2.1\%$	I 30 (3)

APPENDIX II (Continued)

NAEG Vegetation Results
Sites A, B, C, D

MCL No. 7570	NAEG Number	^{235}U atoms/g (0)	$^{235}\text{U}/^{235}\text{C}$ $^{235}\text{C}/^{235}\text{B}$	^{235}U atoms/g (C)	^{235}U dpm/g (C)	$^{239+40}\text{Pu}$ dpm/g	Stake (ISO)
<i>ATRIPLEX CONFERTIFOLIA</i>							
5057	V13012	7.38×10^{12} $\pm 0.4\%$	0.0926 $\pm 4.7\%$	6.26×10^{11} $\pm 4.5\%$	1.16×10^{-3} $\pm 4.5\%$	2.19×10^1 $\pm 13\%$	D 11 (1)
5058	V13015	1.65×10^{12} $\pm 0.7\%$	0.134 $\pm 4.9\%$	1.95×10^{11} $\pm 4.9\%$	3.62×10^{-4} $\pm 4.9\%$	1.43×10^1 $\pm 4.6\%$	J 24 (1)
5061	V13033	2.09×10^{12} $\pm 0.6\%$	0.182 $\pm 2.9\%$	3.22×10^{11} $\pm 2.8\%$	5.96×10^{-4} $\pm 2.8\%$	1.96×10^1 $\pm 2.8\%$	K 22 (1)
5062	V13035	3.99×10^{12} $\pm 9.4\%$	<0.03	$<9.0 \times 10^{10}$	$<1.7 \times 10^{-4}$	3.51×10^0 $\pm 4.0\%$	A 08 (1)
5065	V13056	4.43×10^{13} $\pm 4.5\%$	11.7 $\pm 2.7\%$	4.08×10^{13} $\pm 4.6\%$	7.57×10^{-2} $\pm 4.6\%$	3.19×10^0 $\pm 4.0\%$	D 25 (1)
5070	V13079	5.04×10^{12} $\pm 0.4\%$	0.146 $\pm 2.8\%$	6.42×10^{11} $\pm 2.6\%$	1.19×10^{-3} $\pm 2.6\%$	2.91×10^1 $\pm 1.6\%$	I 08 (1)
5072	V13083	2.12×10^{12} $\pm 0.5\%$	0.0960 $\pm 5.6\%$	1.85×10^{11} $\pm 5.5\%$	3.44×10^{-4} $\pm 5.5\%$	1.03×10^1 $\pm 9.7\%$	M 24 (1)
<i>TETRADYMIA GLABRATA</i>							
5053	V12993	1.30×10^{13} $\pm 0.3\%$	0.321 $\pm 1.5\%$	3.15×10^{12} $\pm 1.3\%$	5.84×10^{-3} $\pm 1.3\%$	3.83×10^2 $\pm 2.4\%$	I 24 (1)
5059	V13025	9.32×10^{12} $\pm 0.4\%$	0.638 $\pm 1.0\%$	3.63×10^{12} $\pm 0.9\%$	6.74×10^{-3} $\pm 0.9\%$	2.51×10^2 $\pm 3.3\%$	H 20 (1)

APPENDIX II (Continued)

NAEG Vegetation Results

Sites A, B, C, D

MCL No. 7570	NAEG Number	^{235}U atoms/g (0)	$^{235}\text{U}/^{235}\text{B}$	^{235}U atoms/g (C)	^{235}U dpm/g (C)	$^{239+40}\text{Pu}$ dpm/g	Stake (ISO)
5064	V13046	1.04×10^{13} $\pm 0.3\%$	0.170 $\pm 2.5\%$	1.51×10^{12} $\pm 2.3\%$	2.81×10^{-3} $\pm 2.3\%$	7.49×10^1 $\pm 2.4\%$	B 20 (1)
5069	V13076	2.41×10^{13} $\pm 2.4\%$	1.76 $\pm 1.7\%$	1.54×10^{13} $\pm 2.7\%$	2.85×10^{-2} $\pm 2.7\%$	2.00×10^1 $\pm 2.4\%$	C 25 (1)
5073	V13094	8.04×10^{12} $\pm 0.4\%$	0.028 $\pm 15\%$	2.2×10^{11} $\pm 15\%$	4.0×10^{-4} $\pm 15\%$	1.25×10^1 $\pm 3.4\%$	C 15 (1)
<i>ATRIPLEX CANESCENS</i>							
5054	V12997	5.44×10^{12} $\pm 2.4\%$	0.673 $\pm 5.5\%$	2.19×10^{12} $\pm 5.5\%$	4.06×10^{-3} $\pm 5.5\%$	2.02×10^2 $\pm 1.4\%$	T 20 (1)
<i>CHRYS. VISCIDIFLORUS</i>							
5060	V13027	1.93×10^{13} $\pm 0.4\%$	0.024 $\pm 18\%$	4.6×10^{11} $\pm 18\%$	8.5×10^{-4} $\pm 18\%$	1.76×10^1 $\pm 5.1\%$	A 09 (1)
5063	V13042	9.37×10^{12}	<0.02	$<1.4 \times 10^{11}$	$<2.6 \times 10^{-4}$	1.26×10^1 $\pm 4.5\%$	G 05 (1)
<i>EUROTIA LANATA</i>							
5055	V13005	9.57×10^{12} $\pm 0.5\%$	0.0663 $\pm 5.3\%$	5.95×10^{11} $\pm 5.1\%$	1.10×10^{-3} $\pm 5.1\%$	2.48×10^1 $\pm 2.8\%$	I 05 (1)

APPENDIX II (Continued)

NAEG Vegetation Results
Sites A, B, C, D

MCL No. 7570	NAEG Number	^{235}U atoms/g (0)	$^{235}\text{U}/^{235}\text{B}$ ^{235}C	^{235}U atoms/g (C)	^{235}U dpm/g (C)	$^{239+40}\text{Pu}$ dpm/g	Stake (ISO)
5056	V13008	1.46×10^{13} $\pm 0.8\%$	0.014 $\pm 41\%$	2.1×10^{11} $\pm 41\%$	3.8×10^{-4} $\pm 41\%$	1.57×10^1 $\pm 2.9\%$	D 01 (1)
5066	V13057	1.89×10^{13} $\pm 1.3\%$	1.1 $\pm 15\%$	9.9×10^{12} $\pm 13\%$	1.8×10^{-2} $\pm 13\%$	1.91×10^2 $\pm 2.6\%$	G 19 (1)
GRAYIA SPINOSA							
5067	V13062	2.95×10^{12} $\pm 1.1\%$	0.131 $\pm 7.2\%$	3.42×10^{11} $\pm 7.2\%$	6.34×10^{-4} $\pm 7.2\%$	8.72×10^0 $\pm 2.0\%$	E 13 (1)
AMROSIA DUMOSA							
5068	V13064	6.58×10^{12} $\pm 0.5\%$	0.219 $\pm 2.6\%$	1.18×10^{12} $\pm 2.5\%$	2.19×10^{-3} $\pm 2.5\%$	5.44×10^1 $\pm 1.5\%$	L 21 (1)
COLEOGYNE RAMOSISSIMA							
5071	V13082	1.11×10^{13} $\pm 0.4\%$	0.0940 $\pm 4.1\%$	9.57×10^{11} $\pm 3.9\%$	1.78×10^{-3} $\pm 3.9\%$	5.45×10^1 $\pm 3.0\%$	M 23 (1)

INVESTIGATION OF POSSIBLE CYTOLOGICAL EFFECTS ON SHRUBS
FROM CHRONIC LOW-LEVEL RADIATION AT NTS (PROGRESS REPORT)

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ABSTRACT

Evidence of radiation damage to vegetation at NTS has been found only at the morphological or phenological levels and in the vicinity of nuclear cratering experiments, or in fallout patterns of accidental ventings of radioactive debris. Some effects have been noted in controlled experiments around large gamma radiation sources, also. Possible effects at the cytological level at lower radiation doses are also of interest. Because there have been annual species which have been irradiated in all stages of their life cycles for a number of generations, these species are of special interest; however, unfavorable precipitation did not produce sufficient annuals in either 1974 or 1975 for examination. Shrub species are also of interest, and one shrub, *Artemisia spinescens*, was found to have cells at the proper stage for examination (chromosomes number after meiosis equal nine). At Site D, Area 11, irradiated since 1954-1955 to estimated doses of 35 to 140 R, 5 percent of the cells producing pollen were aberrant compared with 1.7 percent of the cells from plants outside the irradiated area. Because of the small numbers of cells examined, these numbers were not, however, considered adequate to provide more than an indication that there were more aberrants in the irradiated areas than in the nonirradiated.

INTRODUCTION

The search for radiation effects within areas contaminated with radioactive materials at the Nevada Test Site (NTS) has been carried out in the field almost from the start of testing of nuclear devices there. To date, evidence of radiation damage to vegetation in Nevada has been found only at the morphological or phenological level, and these radiation effects have all been in the vicinities of nuclear cratering experiments, or in the fallout patterns of accidental ventings of radioactive debris. Some vegetation effects have also been noted adjacent to large gamma radiation sources set up in controlled experiments around the test site (Kaaz *et al.*, 1971; Rhoads *et al.*, 1969).

Radioactivity-contaminated areas where morphological or phenological effects are not observable are of interest from another point of view, however, particularly where there have been low-level doses for one to two decades. In some areas, these accumulated doses approach levels associated elsewhere with morphological effects. Site D, Area 11, NTS, which is the subject of this study, is such an area. It is contaminated by plutonium spread primarily by a nonnuclear high explosive, although there was also a small component of nuclear fission debris accompanying the distribution of plutonium.

In the absence of obvious phenological or morphological changes, it would be informative to look for possible radiation effects at the cytological level or, more precisely, at the chromosomal level of organization in cells. At this level, radiation damage can sometimes be inferred from possible hindrance of production or germinability of seeds. However, a more direct method of investigating possible radiation damage is examination of the chromosomes themselves under a light microscope, where certain radiation effects have been noted for some decades prior to the development of nuclear energy or the use of nuclear explosives.

At Area 11, NTS, where safety tests were conducted in 1954 and 1955, dosimetry was set up and doses were estimated to range from 35 to 140 R in the particular area of interest to this study (Rhoads and Franks, 1975). In this area, a large but uncertain number of generations of annual plant species have been irradiated at all stages of their life cycle. For this reason, the annuals were, and continue to be, of special interest. However, due to the time of precipitation, there were almost no annual species produced in either 1974 or 1975, even though many species of annuals occur there in large numbers in more favorable years. Radiation effects on native shrubs at the cytological level have not been investigated either. Area 11 is dominated by the shrub species *Atriplex confertifolia*, although several other species also occur, among them *Artemisia spinescens*, *Grayia spinosa*, and *Eurotia lanata*.

METHODS

Cytological investigation requires that cells be collected at precisely the right time in order to see chromosomes with a light microscope, that is, at a time when chromosomes occur in a nondiffused condition within cell nuclei. These conditions are met at Metaphase I, before final division in the production of pollen cells found in the anthers of developing flower buds. Since none of the NTS species appear to have been the subject of cytological investigations, other than chromosome counts on some, there are no guidelines for flower bud collection to ensure finding the proper developmental stage. Among the five species of shrubs whose flowers were collected, only one, a Compositae, *Artemisia spinescens*, had cells at the proper stage for examination, with N (number of chromosomes after meiosis) equal nine.

This report is restricted to that species and plant material collected in Plutonium Valley, Site D, in Area 11, NTS. Ten year radiation doses to shrubs

at 25 cm above ground were estimated to be from 35 to 140 R, with a gamma exposure about 21 percent of the total dose. "Nonirradiated" material was taken south of the exclosure gate on the assumption, since reconsidered, that the area was not irradiated. In any event, that area does not now have radiation reading above what is considered normal background at NTS. This matter will be given further discussion subsequently.

At other places on NTS, there were a few annual species (compared to more favorable years) which germinated and reached the flowering stage. Such was the case at Rock Valley, NTS, where UCLA has had a long-term environmental radiation experiment using a large ^{137}Cs source (Kaaz *et al.*, 1971; French *et al.*, 1974). Doses were known for all parts of the irradiated area, which allows an estimate of total doses to both annuals and shrubs. Moreover, these doses are within the range of vegetation in the Pu-contaminated areas. For this reason, we collected annual species from the Rock Valley area to develop the technology for both collecting and examining annuals. Ten species were collected, of which only one, *Phacelia fremontii*, contained cells at the proper developmental stage for examination. *Phacelia fremontii* has N equal 13.

Flower buds of both shrubs and annuals were collected and kept in an ethyl alcohol-acetic acid fixative for 24 hours, then transferred to 70 percent ethyl alcohol in which they were returned to the laboratory for examination. In the laboratory, all collections were examined by the Beeks method (Beeks, 1955), which requires that anthers are squashed in acetocarmine and Hoyer's medium on a microscope slide. Slides were examined and photographs taken of both normal cells and those which provided possible evidence of abnormality for both irradiated and nonirradiated areas.

Detection of radiation damage to chromosomes depends, in visual methods, on observing particular chromosome configurations at or near Metaphase I, for certain conditions which are not considered normal. Among these are (1) any synaptic configuration larger than a bivalent pair, that is a trivalent or quadrivalent configuration which may appear as a ring or chain, (2) lagging bivalent chromosomes when other chromosomes have already moved to the poles, and (3) incomplete pairing among otherwise paired bivalents. Another condition is the production of dicentric chromosomes which produce "bridges" between nuclei and chromosomal fragments, but these have not been observed so far. There are still other conditions. Chromosome "stickiness" may result from radiation damage, but may also be an artifact of preparation methods. Other preparation artifacts may also appear essentially the same as radiation effects. Conditions which were observed will be described more fully in the succeeding paragraphs.

Figure 1 shows two nuclei of cells of *Artemisia spinescens* at Metaphase I. Nine bivalent chromosomes are clearly visible, consisting of four bivalent rings, that is chromosomes having two chiasmata near the ends of each bivalent pair, and five other bivalents, with only one chiasma on each bivalent pair. These are considered typical normal cells and were chosen because of the ease with which individual chromosomes can be distinguished. It should be pointed out perhaps that analysis of chromosome conditions are not possible solely on the basis of photographs and that much additional information can be gained

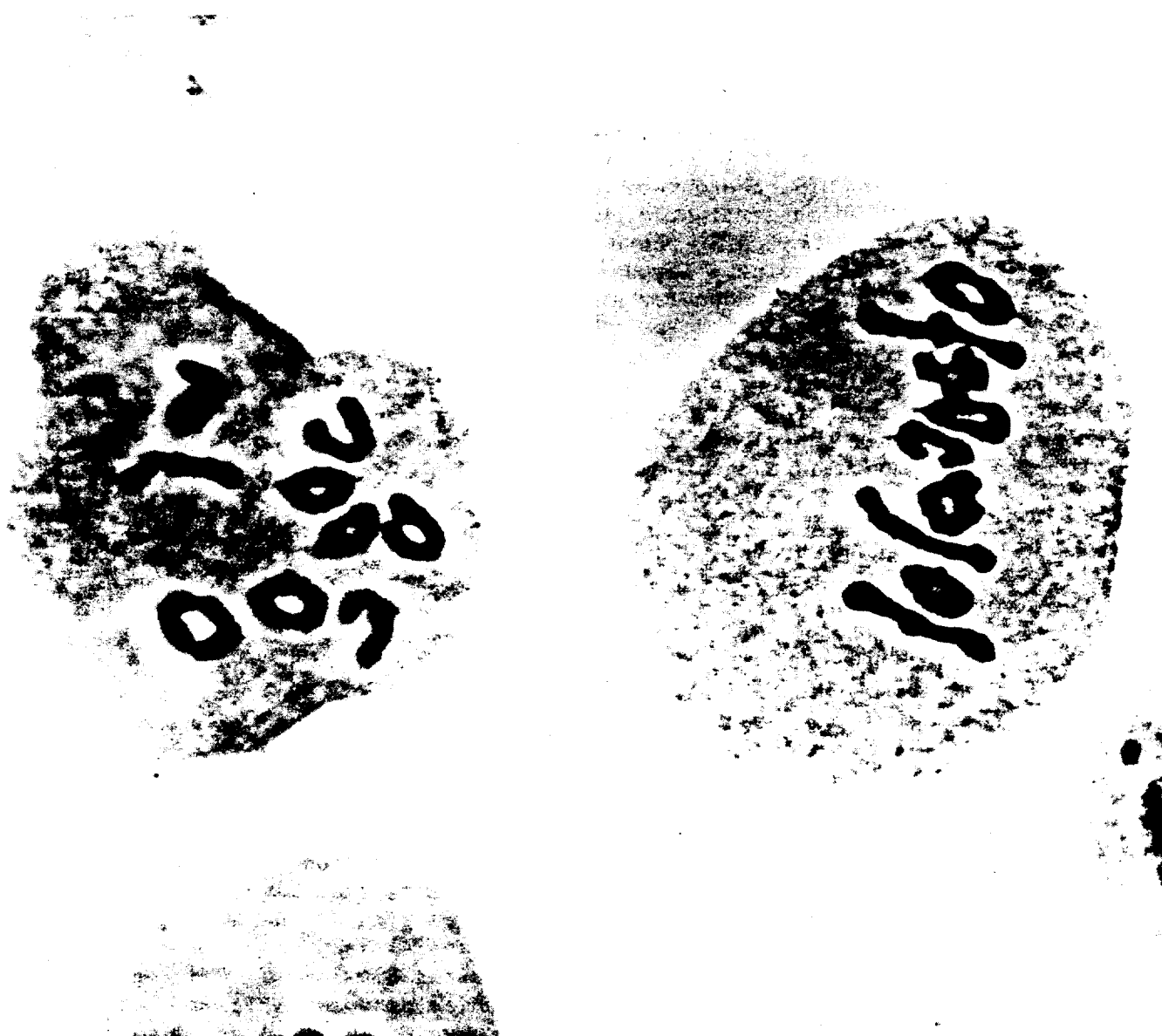


Figure 1. Two cells with the nine bivalent Metaphase I chromosome complement of *Artemisia spinescens* without evidence of abnormalities. The cell on the right has chromosomes lined up along the metaphase plate. Microscope magnification 1,250X

directly from the microscope which allows viewing a third dimension by focusing vertically on the slide preparation.

Figure 2 shows cells considered abnormal. The cell on the left has a ring of four chromosomes forming a figure "8" on the lower right. The cell on the right has both a ring of four chromosomes (upper left) and a chain of four chromosomes (upper right).

Figure 3 shows still two other cells with what are considered abnormalities. The cell on the left is in Metaphase I and has eight bivalents which appear normal and two univalents which do not appear to be properly paired. The cell on the right is in Telophase I. The bivalent chromosomes in the center belong to the group of chromosomes on the left and do not appear to be moving normally to that pole. In the subsequent nucleus which is to be formed, this bivalent pair would probably be omitted and the genetic components of those chromosomes would be lost to that cell which normally divides again meiotically to form pollen grains.

DISCUSSION

In addition to detection of possible abnormalities by visual examination of the chromosomes themselves, an attempt was made to determine whether there might be a change in pollen production between irradiated and nonirradiated areas. Pollen grains per anther were counted, but their very large numbers coupled with changes in anthers attributable to insect damage or pathological conditions due to unknown factors caused this approach to be abandoned. It should be noted in passing that the germinability of pollen grains in artificial media serves in other botanical investigations and may be also applicable to the objectives of this work. Beatley (unpublished) has looked at the germinability of the pollen of *Larrea tridentata* around the Sedan event.

The possibility of artifacts resulting from chromosomal preparations is difficult to assess for several of the abnormal conditions noted. For others, notably the occurrence of quadrivalent or hexavalent rings, there is little likelihood of preparational artifacts. These conditions have been shown to result from breakage with exchange of segments among nonhomologous chromosomes, conditions remote to those attributable to preparational artifacts.

The frequency of occurrence of abnormalities among irradiated and nonirradiated populations offers hope in resolving this problem. The abnormalities observed here occurred in both irradiated and nonirradiated populations, but there appears to be more occurring in the irradiated population. Of the 12 total aberrant cells noted, six occurred among 119 cells which were examined from the irradiated population. Six others occurred among 360 cells examined from nonirradiated shrubs. It is apparent that abnormalities are infrequent. On this basis, 5 percent of the irradiated population of cells appeared abnormal, and 1.7 percent of the nonirradiated cells appeared abnormal. Because of the nature of cytological investigations, these numbers are probably not adequate

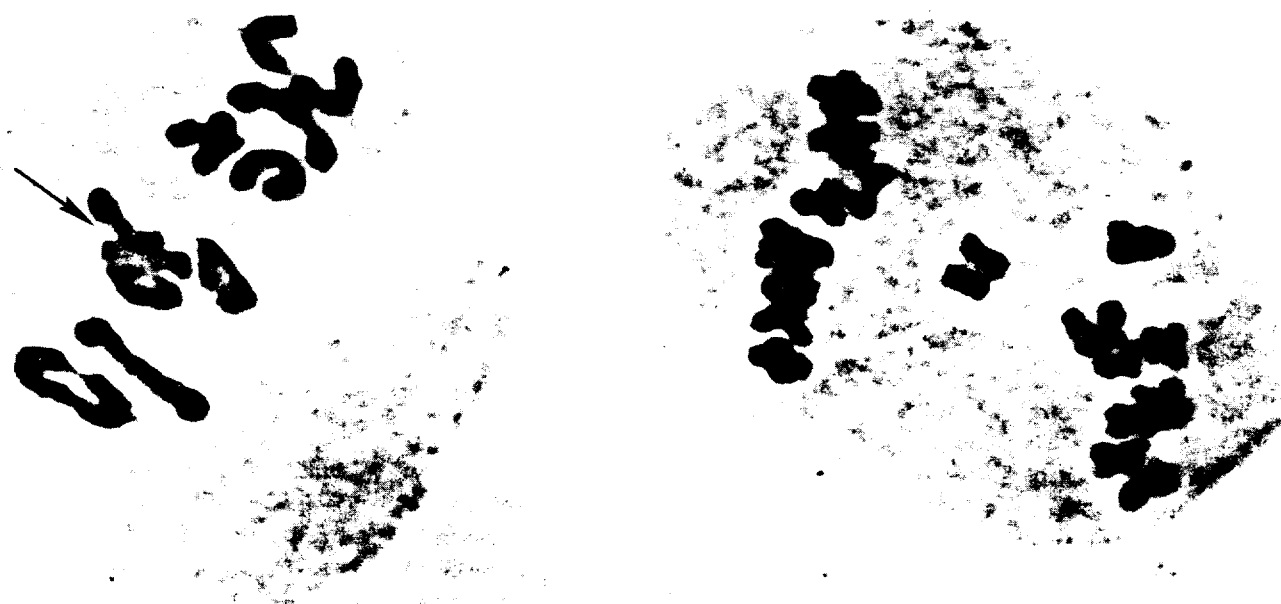


Figure 2. Abnormal chromosome configurations in *Artemisia spinescens*. Cell on the left has a chain of four in the form of a figure "8" in the lower part of the cell. Cell on the right has both a ring of four and a chain of four in the upper right and left of the cell respectively.

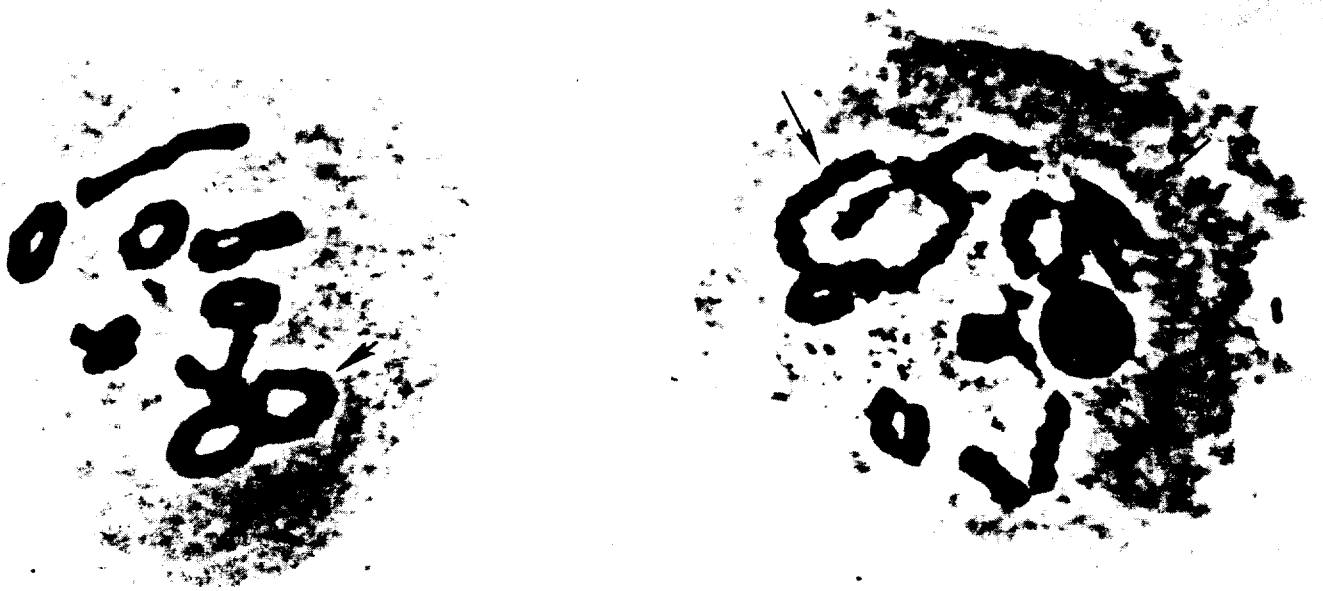


Figure 3. Two cells of *Artemisia spinescens* with abnormal chromosome configurations. Cell on the left is Metaphase I; cell on the right in Telophase I. Left cell contains two univalents which appear not to be properly paired. Cell on the right contains a lagging bivalent pair.

to provide more than an indication that there are more aberrant chromosomes among irradiated shrubs than among nonirradiated.

There is also the problem of finding nonirradiated populations of shrubs with certainty. Much of the area east of both Yucca and Frenchman Flats, even though they may presently show essentially background radiation levels, may have been irradiated by fallout from the early aboveground tests. Because of the long lives, most of the individual shrubs present today are those that were present 20 to 30 years ago. If, by going outside the localities, one tries to avoid the possibility that "nonirradiated" populations might have been irradiated in the past, then there is the possibility that genetic variability may become a factor. Despite this, it appears worthwhile to look at other populations also, and this will be attempted in the future.

Annuals continue to be of primary interest to this project, but unfavorable growing conditions appear to prevail, and consequently there probably will continue to be a dearth of material for examination.

SUMMARY AND CONCLUSIONS

Chromosomal abnormalities within populations of *Artemisia spinescens* were found with a low frequency in both irradiated and "nonirradiated" plants (selected plants outside the presently known radiation areas) from Plutonium Valley, Area 11, NTS. The frequency of abnormalities appears to be somewhat higher in the irradiated population than in the nonirradiated. More cells need to be examined before this conclusion can be considered firm. Despite the possible complication of variation due to genetic variability among different localities, other populations of *Artemisia* should also be evaluated. Because of the likelihood of radiation exposure to many generations, annual species remain a prime objective of this investigation. CY 1976 appears to be another year in which annuals will be sparse or absent from some areas of NTS.

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LARGE VERTEBRATES

SOLUBILITY OF PLUTONIUM AND AMERICIUM-241 FROM RUMEN CONTENTS
OF CATTLE GRAZING ON PLUTONIUM-CONTAMINATED DESERT VEGETATION IN
IN VITRO BOVINE GASTROINTESTINAL FLUIDS--NOVEMBER, 1974, TO MAY, 1975

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ABSTRACT

Rumen contents collected from cattle grazing at Area 13 of the Nevada Test Site were incubated in simulated bovine gastrointestinal fluids to study the alimentary solubility of plutonium and americium-241. The solubility of americium-241 was found to be similar to that of plutonium-239. In most cases, plutonium-238 was more soluble than plutonium-239. The solubility of plutonium-238, plutonium-239, and americium-241 was markedly greater when the rumen contents were collected from a cow which had grazed in a more intensively grazed enclosure located a greater distance downwind from ground zero.

INTRODUCTION

This is the second progress report of an ongoing study to determine factors affecting the solubility of selected field-ingested transuranics in an artificial rumen and simulated abomasal and intestinal fluids. An earlier report by Barth (1975) covered the period from the initiation of the study in November, 1973, through August, 1974, and was confined to plutonium-238 and plutonium-239. This report covers trials from November, 1974, through May, 1975, and includes plutonium-238, plutonium-239, and americium-241.

The earlier report indicated that field-ingested plutonium-238 was generally more soluble than plutonium-239 in these fluids. The differences were, however, quite variable. During the late summer, fall, and winter trials, low plutonium solubilities were observed; these were accompanied by comparatively high concentrations of plutonium in the rumen contents. An analysis of the vegetal composition of the rumen contents indicated that these conditions were associated with high levels of *Eurotia lanata* (winterfat). In the simulated duodenal fluid, which is presently believed to be the most important digestive stage in

regard to plutonium absorption, the minimum and maximum solubilities were 0.49% and 13.5% for plutonium-238. For plutonium-239, they were 0.14% and 0.76%.

During the spring and midsummer trials, a large increase in plutonium solubility was observed; this increase was accompanied by a marked reduction in total plutonium concentration in the rumen contents. Analyses of the vegetal composition of the rumen contents indicated a reduction in the proportion of *Eurotia lanata* and an increase in the proportion of *Oryzopsis hymenoides* (Indian ricegrass) or *Sitanion jubatum* (squirreltail grass). The minimum and maximum solubilities in the simulated duodenum were 95.1% and 96.6% for plutonium-238; for plutonium-239, they were 44.5% and 90.0%.

Boyd *et al.* (1974) reported that samples of a polydisperse aerosol of $^{241}\text{AmO}_2$ treated successively at 300° C and 1050° C were relatively insoluble with a fractional dissolution rate of less than 10^{-4} per day in a standard solvent prepared to simulate the chemical constituents of lung fluid. Repeatedly the dissolution rate proved to be inversely related to the total phosphate concentration in the standard solvent.

Raabe *et al.* (1974) collected samples of aerosols present in a glove box during a plutonium oxide and uranium oxide powder mixing operation. Solubility studies were run at 37° C using lung fluid simulant with and without the phosphate component. Preliminary indications were that the cumulative plutonium solubility was higher in the standard solvent, both with and without phosphate, than was observed for laboratory aerosols of $^{239}\text{PuO}_2$. The cumulative solubility of americium in the phosphate-free solvent was at least two orders of magnitude higher than that of plutonium, but was much less in the solvent with phosphate.

MATERIALS AND METHODS

The procedure described briefly here is essentially that of Barth (1975). Rumen-fistulated Hereford cattle were allowed to graze periodically in the inner enclosure and nonfistulated cattle were allowed to graze continually in the outer enclosure of Area 13 of the Nevada Test Site (Smith, 1975). The inner enclosure directly surrounds the ground zero of a high-explosive detonation of an atomic device during a safety test. The outer enclosure, a less heavily contaminated area, completely surrounds the inner enclosure, and most of it is located a greater distance downwind from ground zero than the inner enclosure. Area 13 has been described by Tamura (1975).

Samples of whole rumen contents were collected from the fistulated cattle following a 48-hour grazing period. Samples were collected from the nonfistulated cattle at the time of slaughter.

Samples of whole rumen contents were added to four Erlenmeyer digestion flasks with simulated abomasal juice consisting of HCl and pepsin, and the pH was adjusted to 3.0. Abomasal incubation in a water bath at 39.5° was allowed to

proceed for 3 hours. The contents of the flasks were then converted to simulate the duodenum by the addition of NaOH to adjust the pH to 4.5, and were incubated for approximately 10 minutes. The upper jejunum was simulated by the addition of bile, pancreatin, trypsin, and erypsin and adjustment of the pH to 6.0, followed by a 2-hour incubation period. The lower small intestine was simulated by adjustment of the pH to 7.5, followed by incubation for 2 more hours.

Following each incubation period, one of the digestion flasks was selected, and the entire contents were separated into solid and liquid fractions by preliminary filtration through several layers of cheesecloth. The resulting filtrate was centrifuged and the supernate collected while the sediment was added to the solid fraction. The entire solid and liquid fractions were analyzed for plutonium-238, plutonium-239, and americium-241 by LFE Environmental Analysis Laboratories Division, Richmond, California (Major *et al.*, 1975). Since a 100% recovery of the liquid phase is not possible, this procedure is "semiquantitative."

RESULTS

During trials 6, 7, 8, 10, and 11, the rumen contents were collected from fistulated steers allowed to graze in the inner enclosure. During trial 9, the rumen contents were collected at the time of slaughter from a cow which had grazed in the outer enclosure.

The percentages of soluble plutonium-238 and plutonium-239 ingested by grazing cattle in simulated bovine abomasal and intestinal fluids are shown in Table 1. These data indicate a moderate increase in the solubility of plutonium in the rumen contents collected during trials 7 and 10 and a very large increase in trial 9. In most cases, plutonium-238 was more soluble than plutonium-239. The ratios of the percentage of soluble plutonium-238 to soluble plutonium-239 varied with a range of 0.83 to 6.0.

There was a rise in plutonium solubility when the pH was increased to 4.5 in order to simulate the duodenum. This initial increase in solubility was followed by an additional increase during the jejunal incubation period following the addition of bile and enzymes and adjustment of the pH to 6. There was a sharp increase in plutonium-238 and plutonium-239 solubility in trial 9 and in plutonium-238 solubility in trial 10 during the lower intestinal incubation period.

The percentages of soluble americium-241 in these fluids are shown in Table 2. These data indicate a moderate increase in the solubility of americium-241 in the rumen contents collected during trials 7 and 10, and a very large increase in trial 9. The 6.5% americium-241 solubility shown in the jejunal phase of trial 6 is anomalous and is not considered in the interpretation of results. The solubility of americium-241 was generally similar to that of plutonium-239. However, the ratios of the percentage of soluble americium-241 to the percentage of soluble plutonium-239 were variable with a range from 0.18 to 6.1.

Table 1. Solubility of Ingested Plutonium in *In vitro* Bovine Gastrointestinal Fluids

	Trial-6 11/05/74	Trial-7 01/19/75	Trial-8 01/21/75	Trial-9** 01/29/75	Trial-10 03/12/75	Trial-11 05/29/75
<u>Abomasum, End pH 3</u>						
Plutonium-238	0.048%	*	*	*	0.32%	*
Plutonium-239	0.058%	0.25%	0.12%	0.19%	0.11%	0.054%
$^{238}\text{Pu}/^{239}\text{Pu}$	0.83	*	*	*	3.1	*
<u>Duodenum, pH 4.5</u>						
Plutonium-238	1.3%	2.0%	*	3.2%	*	0.22%
Plutonium-239	0.24%	0.69%	0.15%	0.64%	0.88%	0.13%
$^{238}\text{Pu}/^{239}\text{Pu}$	5.4	3.0	*	4.9	*	1.7
<u>Jejunum, pH 6</u> <u>Bile & Enzymes, 2 hrs</u>						
Plutonium-238	3.0%	10.6%	1.7%	8.5%	*	1.7%
Plutonium-239	0.64%	1.8%	0.75%	2.6%	1.7%	0.97%
$^{238}\text{Pu}/^{239}\text{Pu}$	4.7	5.8	2.3	3.3	*	1.7
<u>Lower Intestine,</u> <u>pH 7.5</u> <u>Bile & Enzymes, 2 hrs</u>						
Plutonium-238	1.0%	*	0.75%	62.8%	8.0%	*
Plutonium-239	0.76%	1.9%	0.73%	64.2%	1.3%	0.68%
$^{238}\text{Pu}/^{239}\text{Pu}$	1.4	*	1.0	0.98	6.0	*

*Below ^{238}Pu detection limit.

**Outer enclosure.

Table 2. Solubility of Ingested Americium-241 in *In vitro* Bovine Gastrointestinal Fluids

	Trial-6 11/05/74	Trial-7 01/19/75	Trial-8 01/21/75	Trial-9* 01/29/75	Trial-10 03/12/75	Trial-11 05/29/75
<u>Abomasum, End pH 3</u>						
Americium-241 $^{241}\text{Am}/^{239}\text{Pu}$	0.066% 1.1	0.31% 1.2	0.08% 0.66	1.2% 6.1	0.11% 1.0	0.06% 1.2
<u>Duodenum, pH 4.5</u>						
Americium-241 $^{241}\text{Am}/^{239}\text{Pu}$	0.43% 1.7	1.1% 1.6	0.44% 3.0	0.75% 1.2	1.4% 1.5	0.10% 0.80
<u>Jejunum, pH 6</u> <u>Bile & Enzymes, 2 hrs</u>						
Americium-241 $^{241}\text{Am}/^{239}\text{Pu}$	6.5** 10.2	0.89% 0.49	0.13% 0.18	2.3% 0.88	0.98% 0.57	0.23% 0.24
<u>Lower Intestine,</u> <u>pH 7.5</u> <u>Bile & Enzymes, 2 hrs</u>						
Americium-241 $^{241}\text{Am}/^{239}\text{Pu}$	0.58% 0.77	0.43% 0.23	1.2% 1.6	63.7% 0.99	3.3% 2.5	0.54% 0.80

*Outer enclosure.

**Anomalous: Not considered in interpretation of results.

There was usually an increase in americium-241 solubility when the pH was adjusted to pH 4.5 to simulate the duodenum. However, the change in americium-241 solubility during the jejunal incubation period was not consistent. There was a sharp increase in americium-241 solubility in trial 9 and a moderate increase in trial 10 during the lower intestinal incubation period.

The high levels of plutonium and americium-241 solubility during trial 9 were accompanied by the lowest concentrations of plutonium and americium-241 in the rumen contents (Tables 3, 4, 5, and 6). The rumen contents were taken from a cow grazing in the outer enclosure. The minimum concentration of plutonium and americium-241 in rumen contents collected from the steers grazing in the inner enclosure occurred during trial 7 (January 19, 1975), while the maximum concentration occurred during trial 6 (November 5, 1974).

Analyses of the vegetative composition of the rumen contents of the cattle indicate that *Eurotia lanata* was the predominant species present during trials 6, 8, 9, and 11. *Atriplex confertifolia* (shadscale) was the predominant species present during trials 7 and 10 (Table 7).

DISCUSSION

Since this study is still in progress, all interpretations are subject to reconsideration as additional data become available. The results presented here are in general agreement with the earlier NAEG report of Barth (1975).

Since the materials and reactions in the digestive fluids and plant residues form an extremely complex mixture, the term "solubility" refers only to the fraction of plutonium or americium-241 in solution at the termination of a specific digestion phase and disregards all potential intermediate reactions. Particulate dissolution, which is an initial reaction, could be greater than the final solubility following secondary or further reactions.

The greater solubility of plutonium-238 as compared with plutonium-239 reported here and earlier by Barth (1975) is consistent with a preliminary report by Smith *et al.* (1975) that the 239/238-plutonium ratios in tissues are lower than those found in the ingesta, indicating that plutonium-238 is more readily absorbed and retained than is plutonium-239.

The similarity of americium-241 to plutonium-239 solubility reported here is not in agreement with Raabe *et al.* (1974), cited earlier, who reported a greater solubility of americium. The disagreement in results could be due to the different chemical composition of the fluids used, especially phosphate, and perhaps because in this study decay of plutonium-241 had resulted in ingrowth of americium-241 within the plutonium particles.

The behavior of americium-241 differed from that of plutonium in respect to response to the jejunal incubation period. There was a consistent increase in plutonium solubility during the jejunal incubation period, while this was not

Table 3. Total Plutonium Present in 250 g of Rumen Contents (Radioactivity)

	Trial-6 11/05/74	Trial-7 01/19/75	Trial-8 01/21/75	Trial-9* 01/29/75	Trial-10 03/12/75	Trial-11 05/29/75
<u>Abomasum, End pH 3</u>						
Plutonium-238 (pCi)	173.7	13.3	47.0	4.0	75.4	135.3
Plutonium-239 (pCi)	7518.8	473.2	2005.9	139.4	2748.9	6264.1
$^{239}\text{Pu}/^{238}\text{Pu}$	43.4	35.7	42.7	34.8	36.5	46.3
<u>Duodenum, pH 4.5</u>						
Plutonium-238 (pCi)	182.3	15.4	36.1	9.2	104.0	147.3
Plutonium-239 (pCi)	7005.9	560.1	1684.4	362.0	2844.4	5050.6
$^{239}\text{Pu}/^{238}\text{Pu}$	38.4	36.3	46.7	39.3	27.4	34.3
<u>Jejunum, pH 6</u> <u>Bile & Enzymes, 2 hrs</u>						
Plutonium-238 (pCi)	173.0	7.7	43.0	5.3	40.9	122.2
Plutonium-239 (pCi)	7126.4	271.7	1467.4	129.1	1504.4	4795.3
$^{239}\text{Pu}/^{238}\text{Pu}$	41.2	35.3	34.1	24.3	36.8	39.2
<u>Lower Intestine,</u> <u>pH 7.5</u> <u>Bile & Enzymes, 2 hrs</u>						
Plutonium-238 (pCi)	168.6	3.9	57.5	9.9	72.5	159.6
Plutonium-239 (pCi)	7395.4	192.3	2021.3	130.3	2688.8	6888.6
$^{239}\text{Pu}/^{238}\text{Pu}$	43.9	48.2	35.2	13.1	37.1	43.2

*Outer enclosure.

Table 4. Total Plutonium Present in 250 g of Rumen Contents (Mass Basis)

	Trial-6 11/05/75	Trial-7 01/19/75	Trial-8 01/21/75	Trial-9* 01/29/75	Trial-10 03/12/75	Trial-11 05/29/75
<u>Abomasum, End pH 3</u>						
$^{238}\text{Pu} \times 10^{-9} \text{ (}\mu\text{g)}$	10,090.9	771.0	2,730.7	233.0	4,379.7	7,860.9
$^{239}\text{Pu} \times 10^{-6} \text{ (}\mu\text{g)}$	122,405.6	7,703.4	32,656.2	2,266.1	44,752.6	101,979.1
$^{239}\text{Pu}/^{238}\text{Pu} \times 10^3$	12.1	10.0	12.0	9.7	10.2	13.0
<u>Duodenum, pH 4.5</u>						
$^{238}\text{Pu} \times 10^{-9} \text{ (}\mu\text{g)}$	10,591.0	896.8	2,095.1	535.7	6,043.0	8,559.3
$^{239}\text{Pu} \times 10^{-6} \text{ (}\mu\text{g)}$	114,056.2	9,118.7	27,421.7	5,893.5	46,306.8	82,387.4
$^{239}\text{Pu}/^{238}\text{Pu} \times 10^3$	10.8	10.2	13.1	11.0	7.7	9.6
<u>Jejunum, pH 6</u> <u>Bile & Enzymes, 2 hrs</u>						
$^{238}\text{Pu} \times 10^{-9} \text{ (}\mu\text{g)}$	10,051.9	447.3	2,500.2	308.7	2,375.1	7,099.2
$^{239}\text{Pu} \times 10^{-6} \text{ (}\mu\text{g)}$	116,017.5	4,423.4	23,888.5	2,101.1	24,492.0	78,066.8
$^{239}\text{Pu}/^{238}\text{Pu} \times 10^3$	11.5	9.9	9.6	6.8	10.3	11.0
<u>Lower Intestine,</u> <u>pH 7.5</u> <u>Bile & Enzymes, 2 hrs</u>						
$^{238}\text{Pu} \times 10^{-9} \text{ (}\mu\text{g)}$	9,793.9	228.9	3,339.1	578.1	4,209.9	9,272.8
$^{239}\text{Pu} \times 10^{-6} \text{ (}\mu\text{g)}$	120,397.4	3,131.5	32,907.1	2,121.6	43,773.3	112,146.1
$^{239}\text{Pu}/^{238}\text{Pu} \times 10^3$	12.3	13.7	9.8	3.7	10.4	12.1

*Outer enclosure.

Table 5. Total Americium-241 Present in 250 g of Rumen Contents (Radioactivity)

	Trial-6 11/05/74	Trial-7 01/19/75	Trial-8 01/21/75	Trial-9* 01/29/75	Trial-10 03/12/75	Trial-11 05/29/75
<u>Abomasum, End pH 3</u>						
Americium-241 (pCi) $^{239}\text{Pu}/^{241}\text{Am}$	1146.5 6.6	75.6 6.3	416.1 4.8	16.7 8.3	396.1 6.9	946.5 6.6
<u>Duodenum, pH 4.5</u>						
Americium-241 (pCi) $^{239}\text{Pu}/^{241}\text{Am}$	1388.3 5.0	89.0 6.7	235.8 7.0	53.3 6.8	407.3 7.0	703.5 7.2
<u>Jejunum, pH 6</u> <u>Bile & Enzymes, 2 hrs</u>						
Americium-241 (pCi) $^{239}\text{Pu}/^{241}\text{Am}$	1629.8 4.4	158.0 1.7	241.7 6.1	19.2 6.7	319.0 4.7	750.4 6.4
<u>Lower Intestine,</u> <u>pH 7.5</u> <u>Bile & Enzymes, 2 hrs</u>						
Americium-241 (pCi) $^{239}\text{Pu}/^{241}\text{Am}$	1106.8 6.2	350.0 0.55	249.4 8.1	21.9 6.0	391.1 6.9	954.8 7.2

*Outer enclosure.

Table 6. Total Americium-241 Present in 250 g Rumen Contents (Mass Basis)

	Trial-6 11/05/74	Trial-7 01/19/75	Trial-8 01/21/75	Trial-9* 01/29/75	Trial-10 03/12/75	Trial-11 05/29/75
<u>Abomasum, End pH 3</u>						
$^{241}\text{Am} \times 10^{-6} \text{ (}\mu\text{g)}$	333.6	22.0	121.1	4.9	115.3	275.4
$^{239}\text{Pu}/^{241}\text{Am}$	366.9	350.0	269.7	465.3	388.2	370.3
<u>Duodenum, pH 4.5</u>						
$^{241}\text{Am} \times 10^{-6} \text{ (}\mu\text{g)}$	404.0	24.5	68.6	15.5	118.5	204.7
$^{239}\text{Pu}/^{241}\text{Am}$	282.3	372.8	399.7	380.0	390.7	402.4
<u>Jejunum, pH 6</u> <u>Bile & Enzymes, 2 hrs</u>						
$^{241}\text{Am} \times 10^{-6} \text{ (}\mu\text{g)}$	474.3	46.0	70.3	5.6	92.8	218.4
$^{239}\text{Pu}/^{241}\text{Am}$	244.6	96.2	339.6	375.9	263.9	357.5
<u>Lower Intestine,</u> <u>pH 7.5</u> <u>Bile & Enzymes, 2 hrs</u>						
$^{241}\text{Am} \times 10^{-6} \text{ (}\mu\text{g)}$	348.3	101.8	72.6	6.4	113.8	277.8
$^{239}\text{Pu}/^{241}\text{Am}$	345.7	30.7	453.3	333.6	384.6	403.6

*Outer enclosure.

Table 7. Predominant Vegetative Species Present in Rument Contents as Percent of Total²

	Trial-6 11/05/74	Trial-7 01/19/75	Trial-8 01/21/75	Trial-9 ¹ 01/29/75	Trial-10 03/12/75	Trial-11 05/29/75
<i>Eurotia lanata</i> *	62%	10%	61%	36%	32%	46%
<i>Atriplex canescens</i> †		4%	4%	15%		
<i>Oryzopsis hymenoides</i> **	10%		4%	17%		17%
<i>Sitanion jubatum</i> ††						28%
<i>Atriplex confertifolia</i> ***	27%	86%	28%	16%	61%	

*Winterfat

†Four-winged saltbush

**Indian ricegrass

††Squirreltail grass

***Shadscale

¹Outer enclosure.²Data used by permission of D. D. Smith.

true for americium-241 solubility. This increase in plutonium solubility also occurred during the earlier studies of Barth (1975) and Barth and Mullen (1974). The data of Barth and Mullen (1974) indicated that the immediate increase in plutonium solubility during the jejunal stage was due specifically to the presence of bile. Although biliary excretion of americium-241 has been suggested by Scott (1948) (as reported by Durbin, 1973), indicating that bile has the ability to complex americium-241, there was no consistent increase in the solubility of americium-241 during the jejunal incubation period.

In making comparisons of radionuclide concentrations in rumen contents between trials, only very large differences are meaningful since the cattle may graze at will throughout the enclosures. Also, the plant type ingested and the resuspension of radionuclides may vary. However, in the available data, a trend concerning peak activity of plutonium in the rumen contents appears to be developing.

Results to date (Tables 2 or 3 of Barth, 1975, and Tables 3 or 4 of this report) indicate that the highest concentrations of plutonium in the rumen contents of cattle grazing at Area 13 have occurred during the late summer or fall, followed by reduced concentrations during the winter and early summer. An exception to this occurred during trial 11.

Unfortunately, observations were not made of the field conditions of the *Eurotia lanata* during these trials. However, visual observations were made of the field conditions of *Eurotia lanata* at Area 13 during the summer and fall of 1975 and the winter of 1975/1976 (data not included in this report). The reproductive phase, characterized by the long-haired fruiting involucre, appeared during October, 1975. During January, 1976, some fruiting involucres remained. The onset of the reproductive phase will vary depending on rainfall and other conditions. This suggests that with the presence of involucres, the entrapment of resuspended particles would be greatly increased, resulting in higher concentrations of plutonium and americium-241 in the rumen contents. This plant is described, with a photograph of the reproductive phase, by Wallace and Romney (1972).

The moderate increases in plutonium and americium-241 solubility during trials 7 and 10, compared to other trials which used rumen contents collected from cattle grazing in the inner enclosure, were associated with a reduction in the intake of *Eurotia lanata* and an increase in the intake of *Atriplex confertifolia*.

Barth and Mullen (1974) reported average minimum and maximum solubilities of 1.5% and 8.4% for plutonium-238 dioxide in an artificial rumen and simulated abomasal and intestinal fluids procedure. However, during trial 9 of the present study, solubilities of over 62% for both plutonium-238 and plutonium-239 were shown, while in the earlier report of Barth (1975), the solubility reached 90% and above during spring and midsummer trials. This suggests the possibility that during these specific trials, most of the ingested plutonium was in a form other than plutonium dioxide and that most of this plutonium represented the low level internally incorporated in plant tissue.

Primary factors which may contribute to the marked increase in plutonium and americium-241 solubility during trial 9 (outer enclosure) are a greater distance

of the grazing area from ground zero and grazing intensity. Romney *et al.* (1975) stated that, as a general rule, the mean fallout particle size decreases with increasing distance downwind from the point of detonation. Hence, a reduced mean particle size is expected in the outer enclosure which would, in turn, result in an increased dissolution rate. Increasing plutonium availability with decreasing particle size in animals was demonstrated by Bair and Willard (1963) who reported that the rates of pulmonary clearance, translocation, and excretion in both urine and feces were greatest for the aerosol with particles of the smallest median diameters.

During the period that these rumen contents samples were collected, the outer enclosure was more intensely grazed than the inner enclosure. Nonfistulated cattle were allowed to graze continually in the outer enclosure, while fistulated cattle were allowed to graze quarterly, for periods of about 4 days, in the inner enclosure. This allowed for regrowth of the edible vegetation in the inner enclosure, while in the outer enclosure, it was continually eaten back. It appeared by visual observation that there were fewer of the fruiting involucres on the *Eurotia lanata* in the outer enclosure. This suggests a greatly reduced plant surface entrapment of resuspended particulate material, thereby increasing the proportion of plutonium and americium-241, presumably, internally incorporated into the plant tissue. Under these experimental conditions, the effects of distance from ground zero and grazing intensity cannot be differentiated.

The amount of soluble plutonium and americium-241 in each digestion phase is shown in picocuries in Table 8. In these trials, the highest amounts of radionuclide in solution, on a radioactivity basis, came from plutonium-239 followed by americium-241. Although on a mass basis minor amounts of plutonium-238 were present in the rumen contents collected, on a radioactivity basis the plutonium-238 in solution made up a considerable portion of the soluble plutonium present and in some cases approached or exceeded the levels of soluble americium-241. This can be accounted for by the generally greater solubility of plutonium-238 in these fluids and the specific activity of plutonium-238, which is approximately 280 times greater than that of plutonium-239 and 5 times greater than that of americium-241.

It is emphasized that the data and interpretations presented here and in the earlier report of Barth (1975) apply to the specific field conditions prevailing at the time that rumen contents were collected at Area 13 of the NTS. The field conditions at Area 13, observed to date, that may affect plutonium and americium-241 concentrations in rumen contents and their solubility in the digestive fluids described here are season, predominant vegetative species ingested, vegetative stage of growth, presence of fruiting involucres on *Eurotia lanata*, grazing intensity, and distance of grazing area from ground zero.

Table 8. Picocuries of Soluble Plutonium and Americium-241 in *In vitro* Bovine Gastrointestinal Fluids

	Trial-6 11/05/74 (pCi)	Trial-7 01/19/75 (pCi)	Trial-8 01/21/75 (pCi)	Trial-9** 01/29/75 (pCi)	Trial-10 03/12/75 (pCi)	Trial-11 05/29/75 (pCi)
<u>Abomasum, End pH 3</u>						
Plutonium-238	0.083	*	*	*	0.24	*
Plutonium-239	4.4	1.2	2.5	0.27	2.9	3.4
Americium-241	0.75	0.23	0.35	0.20	0.44	0.59
<u>Duodenum, pH 4.5</u>						
Plutonium-238	2.4	0.32	*	0.29	*	0.32
Plutonium-239	17.1	3.4	2.5	2.31	25.0	6.6
Americium-241	5.9	0.92	1.0	0.40	5.5	0.74
<u>Jejunum, pH 6</u> <u>Bile & Enzymes, 2 hrs</u>						
Plutonium-238	5.2	0.82	0.75	0.45	*	2.1
Plutonium-239	45.9	4.9	11.0	3.4	26.0	46.4
Americium-241	106.6***	1.4	0.32	0.44	3.1	1.7
<u>Lower Intestine,</u> <u>pH 7.5</u> <u>Bile & Enzymes, 2 hrs</u>						
Plutonium-238	1.8	*	0.43	6.2	5.8	*
Plutonium-239	56.2	3.6	14.7	83.7	35.7	47.2
Americium-241	7.0	1.5	3.0	13.9	12.9	5.2

*Below ^{238}Pu detection limit.

**Outer enclosure

***Anomalous: Not considered in interpretation of results.

FUTURE PLANS

The study described here will continue.

Future soil and microorganisms-artificial rumen joint studies have been designed with the following objectives:

1. To compare the biological availability of biologically incorporated plutonium with nonbiological plutonium.
2. To determine whether biological organic binding protects plutonium from being removed from solution by competing chemical reactions, such as adsorption, formation of insoluble salts, or polymerization.
3. To determine the effects of soil microbiological activity on the solubility of plutonium-238, plutonium-239, and americium-241 in contaminated soil in an artificial rumen and simulated abomasal and intestinal fluids.
4. To compare the *in vitro* behavior of plutonium-238, plutonium-239, and americium-241 in soil contaminated with nuclear debris.

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GRAZING STUDIES ON A CONTAMINATED RANGE OF THE NEVADA TEST SITE

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ABSTRACT

Actinide data from gonad, muscle, liver, lung, and femur samples collected from five cattle (three aged cows and two yearling bulls) that grazed a plutonium-contaminated range on the Nevada Test Site are discussed. Comparisons are made with data from similar tissues collected from a group of cattle that grazed in the area of the Rocky Flats Plant in Jefferson County, Colorado.

Plutonium-239 levels in the gonads from both groups were higher than those reported for muscle. The medium plutonium-239 value from the gonads of the Nevada Test Site cattle was 25 times higher than the muscle, approximately equal to the femur, and one-half to one-third of the liver and lungs. Uranium levels in the gonads of both groups of cattle were greater than in all of the other tissues discussed. Little difference was noted in the activity levels in the gonads of the males versus the females.

An estimate of soil ingestion by grazing cattle was made by weighing the sediment from washed ingesta collected from fistulated steers that had grazed for a 24-hr period on both ungrazed range and heavily grazed range. Sediment was also collected from the entire gastrointestinal tract of a sacrificed cow. It was estimated that less than 0.5 kilograms of soil would be ingested for a 24-hr period by cattle grazing this range.

INTRODUCTION

A grazing study on the plutonium-contaminated range of Area 13 of the Nevada Test Site was initiated in May, 1973 and is continuing. The objectives, protocol, sampling, and analytical methods used in this study were previously described (Smith, 1974, 1975, 1976). These reports included data on the botanical composition of rumen ingesta collected from both sacrificed animals and rumen-fistulated steers.

Since the study began, tissue samples have been collected from three cows, two calves, and one fetus in October, 1973; three cows and two fetuses in July, 1974; one cow and two yearling calves in January, 1975; and two cows and two calves in January, 1976. Other animals sampled from the study area included three goats, three foxes, one jackrabbit, one coyote, and one yearling beef animal that wandered into the area in December, 1975. One aged cow and two 2-year-old bulls are still grazing the study area.

Since the Nevada Applied Ecology Group meeting in May, 1975, analytical data have been received on all tissues collected through January, 1975. Much of these data were summarized and presented at the International Atomic Energy Agency/Energy Research and Development Administration symposium in November, 1975 (Smith *et al.*, 1976).

In January, 1976, an additional data printout was received from the analytical laboratory which included data on the gonads and four other tissues from five animals (three aged cows and two yearling bulls). The vital statistics of these animals are presented in Table 1. All animals grazed the outer compound of two compounds established in Area 13. The cows were sacrificed in July, 1974, approximately 14 months after their entry into the area. The bulls were sacrificed in January, 1975, 15 and 19 months after their birth within the outer compound.

RESULTS AND DISCUSSION

Table 1 also shows the data on the ash percentages of the tissues analyzed. Data are frequently expressed in reports as activity per unit of ash, or as per unit of dry weight, or as per unit of wet weight. Frequently there are wide variations in percent ash reported for similar tissues by the analytical laboratory, e.g., the gonads of cow number 4 and the femur of cow number 6 in Table 1. Also note the sixfold variation in other gonad data, fourfold in muscle, threefold in lung, and fourfold in liver. If the data are reported on these samples as activity per gram of ash, wide variation may result. Reporting the data as activity per kilogram of wet weight tends to level out these variations since water loss between sampling and analysis should not cause such a large variation.

For example, if two muscle samples each weighing 2 kilograms (kg) and each containing 100 picocuries (pCi) of activity, are analyzed and the ash percentages are reported as 3 and 9%, then the activity per gram (g) of ash would be reported as 1.67 pCi/g ash and 0.55 pCi/g ash. This is a 300% difference. When these values are converted back to wet weight, the correct value of 50 pCi/kg is obtained. Also, dose estimates from ingestion of tissues require that the activity be expressed on a wet weight basis. Therefore, in the following discussion, all data are reported on the wet weight basis.

Table 1. Vital Statistics and Tissue Ash Percentages of Area 13 Cattle

Animal Number	Sex	Age	Period in Outer Compound	Percent of Ash				
				Gonads	Muscle	Lungs	Liver	Femur
1	F	12 yrs	05/02/73-07/09/74	6.2	5.2	4.4	1.4	24.5
4	F	11 yrs	05/02/73-07/09/74	22	3.4	4.1	6.1	27.9
6	F	12 yrs	05/02/73-07/09/74	5.7	5.7	2.1	5.1	12.6
13	M	15 mos	11/02/73-01/29/75	0.89	1.3 & 4.4	3.9	3.4 & 5	25.4 & 24.4
15	M	19 mos	07/02/73-01/29/75	1.5	5.5 & 3.6	1.5	3.4 & 6.6	25.3 & 26.5

The plutonium-238, -239, americium-241, and uranium values reported in the gonad, muscle, liver, lung, and femur samples collected from each animal are listed in Table 2. Duplicate samples were collected of the muscle, liver, and femur of animals numbers 13 and 15 and are also shown in Table 2.

Comparison of the values for the two groups of cattle (three aged cows and two yearling bulls) reveals no major differences in concentrations, with the possible exception of plutonium in the femurs (see Fig. 1) which appears to be slightly higher in the younger animals. If this difference is real, it may be because of the more rapid growth rate of the bones of young animals, resulting in increased incorporation of plutonium in the bone. Also, as these animals were born within the area, they received a certain contribution from their dams prior to birth. More information relative to these observations will become available when data from the animals sacrificed in January, 1976, are obtained.

On the basis of these five animals, there appears to be little difference in actinide concentrations between male and female gonads. The male gonads (~ 500 g wet weight) should certainly provide adequate ash for analysis. At present, the only comparable group of cattle for which gonadal data are available is from the Rocky Flats area of Colorado where five aged cows and five yearling heifers were sampled in November, 1973, (Smith and Black, 1975). Values from these animals are compared to those reported from the Area 13 cattle in the following discussion and are displayed graphically in Figs. 2 and 3.

As shown in Fig. 2, the median plutonium-239 concentration in the gonads from Area 13 cattle was approximately 25 times higher than that found in muscle. An even greater median concentration variation was found between these two tissues in the Rocky Flats cattle. The plutonium-239 median concentration in the gonads from the Area 13 cattle was approximately equal to that of the femurs and one-half to one-third that of liver and lungs. The concentrations in gonads of Rocky Flats cattle were higher than those in muscle, lung, liver, and femur. However, the concentrations in these tissues were generally less than those reported for similar tissues for Area 13 cattle.

As shown in Fig. 3, the uranium concentration in gonads of Area 13 cattle was about ten times the median concentration in the other four tissue types. This increased concentration of uranium in the gonads was observed in the Rocky Flats cattle, also shown in Fig. 3.

The median plutonium-238 concentration in gonads from Area 13 cattle was approximately equal to that in lung, liver, and femur tissues and approximately 100 times greater than that reported in muscle tissue. The americium-241 concentration in gonads of the Area 13 cattle was approximately equal to that of the liver and lung tissues, but twice as great as reported in the femur and approximately 50 times that found in the muscle.

As shown in Fig. 4, the plutonium-239/americium-241 ratios were calculated for the five tissues from the two herds. These ratios were similar for both locations, but generally a smaller range of variation in concentrations was found in tissues from Area 13.

Table 2. Actinide Activities in Selected Tissues from Area 13 Cattle (wet weight)

Tissue	Radionuclide/ Units	Animal Number						
		1	4	6	13	13 (duplicate)	15	15 (duplicate)
Gonads	^{238}Pu pCi/kg	< MDA	< MDA	3.04 $\pm$ 1.85	0.163 $\pm$ 0.099			
	^{239}Pu pCi/kg	1.78 $\pm$ 1.33	6.9 $\pm$ 3.7	< MDA	2.5 $\pm$ 0.28		4.2 $\pm$ 0.9	
	^{241}Am pCi/kg	< MDA	2.9 $\pm$ 2.6	< MDA	2.1 $\pm$ 0.036		4.5 $\pm$ 0.3	
	U $\mu\text{g/kg}$	109	241	36	37.1		65.7	
Muscle	^{238}Pu pCi/kg	< MDA	0.036 $\pm$ 0.017	Lost	< MDA	< MDA	< MDA	< MDA
	^{239}Pu pCi/kg	0.05 $\pm$ 0.15	0.195 $\pm$ 0.03	Lost	< MDA	0.22 $\pm$ 0.04	0.272 $\pm$ 0.04	0.23 $\pm$ 0.05
	^{241}Am pCi/kg	0.02 $\pm$ 0.013	0.06 $\pm$ 0.036	0.04 $\pm$ 0.031	0.15 $\pm$ 0.07	0.04 $\pm$ 0.22	0.1 $\pm$ 0.048	Loss
	U $\mu\text{g/kg}$	4.1	1.01	3.64	4.7	0.89	3.6	9.7
Lungs	^{238}Pu pCi/kg	2.08 $\pm$ 0.17	1.5 $\pm$ 0.19	0.59 $\pm$ 0.059	0.52 $\pm$ 0.068		0.55 $\pm$ 0.16	
	^{239}Pu pCi/kg	74.4 $\pm$ 2.14	51.4 $\pm$ 2.57	18.2 $\pm$ 0.55	17.1 $\pm$ 0.51		5.4 $\pm$ 1.38	
	^{241}Am pCi/kg	10.6 $\pm$ 1.06	7.5 $\pm$ 0.53	2.5 $\pm$ 0.15	2.66 $\pm$ 0.17		2.63 $\pm$ 0.13	
	U $\mu\text{g/kg}$	15.8	5.5	5.3	5.9		14.1	
Liver	^{238}Pu pCi/kg	0.43 $\pm$ 0.19	0.6 $\pm$ 0.18	0.3 $\pm$ 0.19	0.35 $\pm$ 0.07	0.26 $\pm$ 0.029	0.29 $\pm$ 0.12	0.17 $\pm$ 0.03
	^{239}Pu pCi/kg	14.5 $\pm$ 0.88	13.8 $\pm$ 0.95	10.9 $\pm$ 0.87	12.8 $\pm$ 0.51	10.1 $\pm$ 0.7	7.3 $\pm$ 0.55	7.8 $\pm$ 0.23
	^{241}Am pCi/kg	2.5 $\pm$ 0.48	4.76 $\pm$ 0.43	0.79 $\pm$ 0.22	1.5 $\pm$ 0.5	1.2 $\pm$ 0.19	0.76 $\pm$ 0.01	0.33 $\pm$ 0.1
	U $\mu\text{g/kg}$	1.6	1	1.5	12.2	7.7	5.5	5.1
Femur	^{238}Pu pCi/kg	0.134 $\pm$ 0.11	< MDA	0.18 $\pm$ 0.11	< MDA	0.1 $\pm$ 0.19	< MDA	2 $\pm$ 0.2
	^{239}Pu pCi/kg	1.4 $\pm$ 0.25	3 $\pm$ 0.6	0.5 $\pm$ 0.17	6.3 $\pm$ 0.66	8.2 $\pm$ 0.65	2.9 $\pm$ 0.55	2.4 $\pm$ 0.26
	^{241}Am pCi/kg	0.43 $\pm$ 0.14	1.55 $\pm$ 0.45	0.28 $\pm$ 0.11	0.69 $\pm$ 0.52	1.4 $\pm$ 0.32	1.53 $\pm$ 0.55	< MDA
	U $\mu\text{g/kg}$	17.2	13	7.9	6.5	9.7	10.7	7.9

< MDA = less than minimum detectable activity.

COMPARISON OF ^{239}Pu AND ^{238}Pu VALUES IN THE FEMURS OF ADULT AND JUVENILE CATTLE

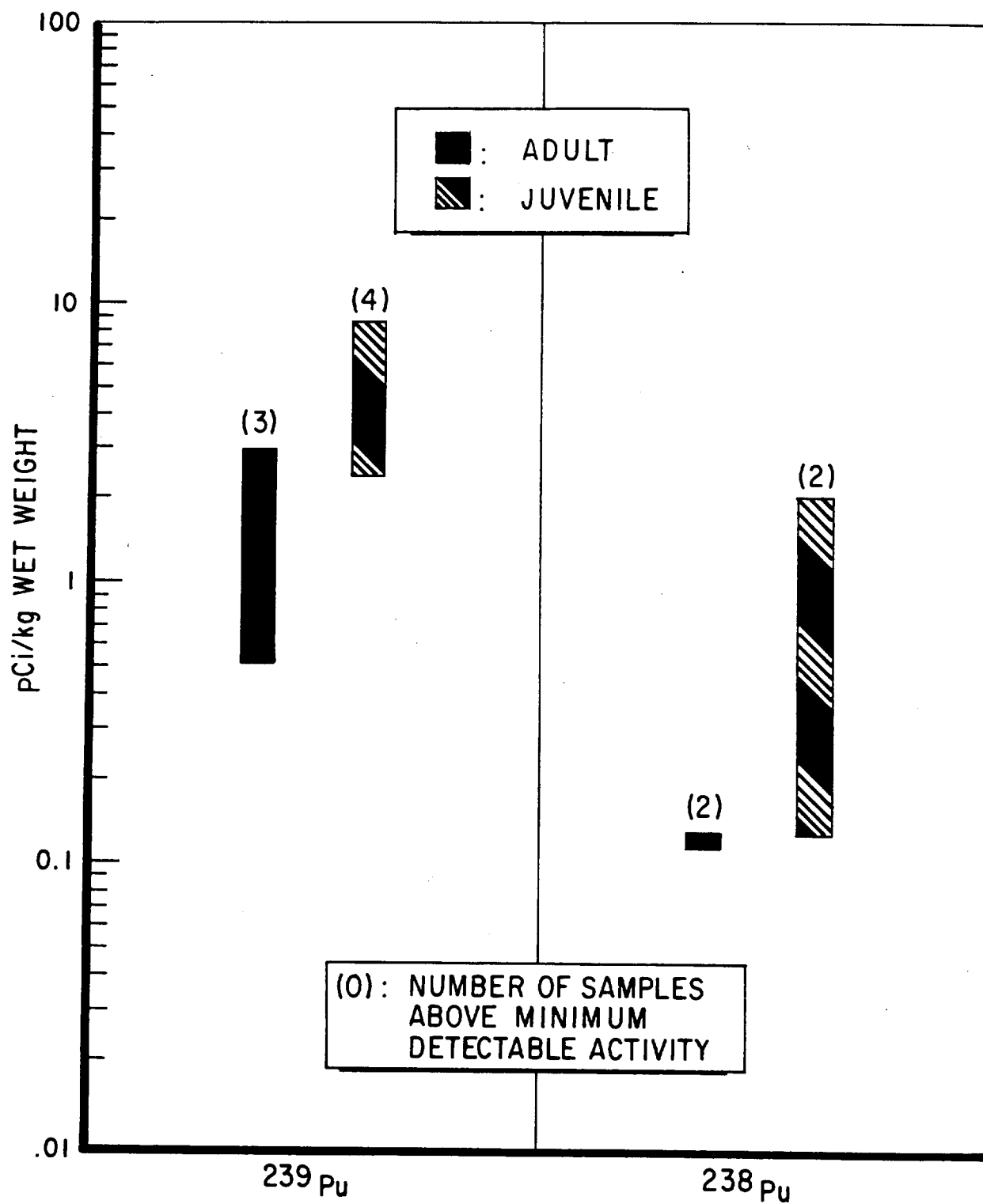


FIGURE 1

^{239}Pu ACTIVITIES IN CATTLE TISSUES

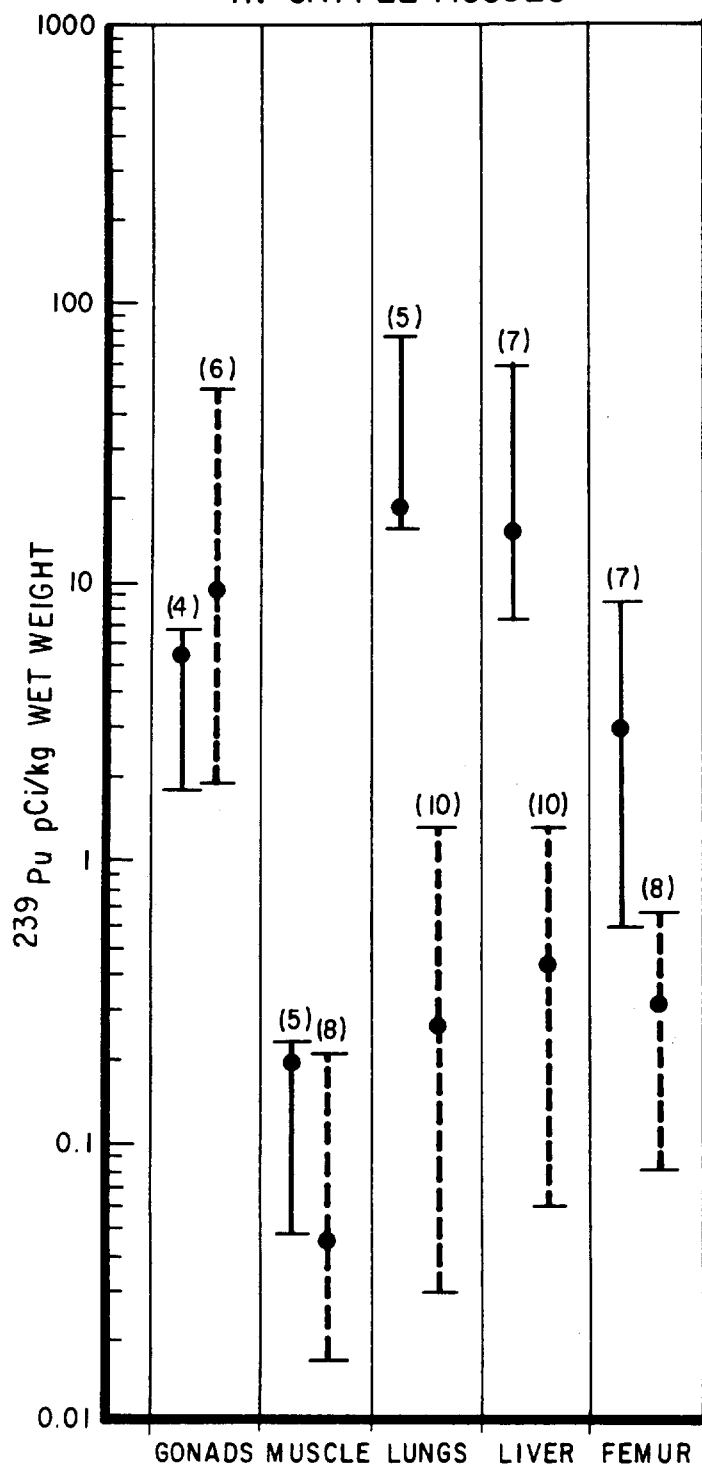


FIGURE 2

URANIUM ACTIVITIES IN CATTLE TISSUE

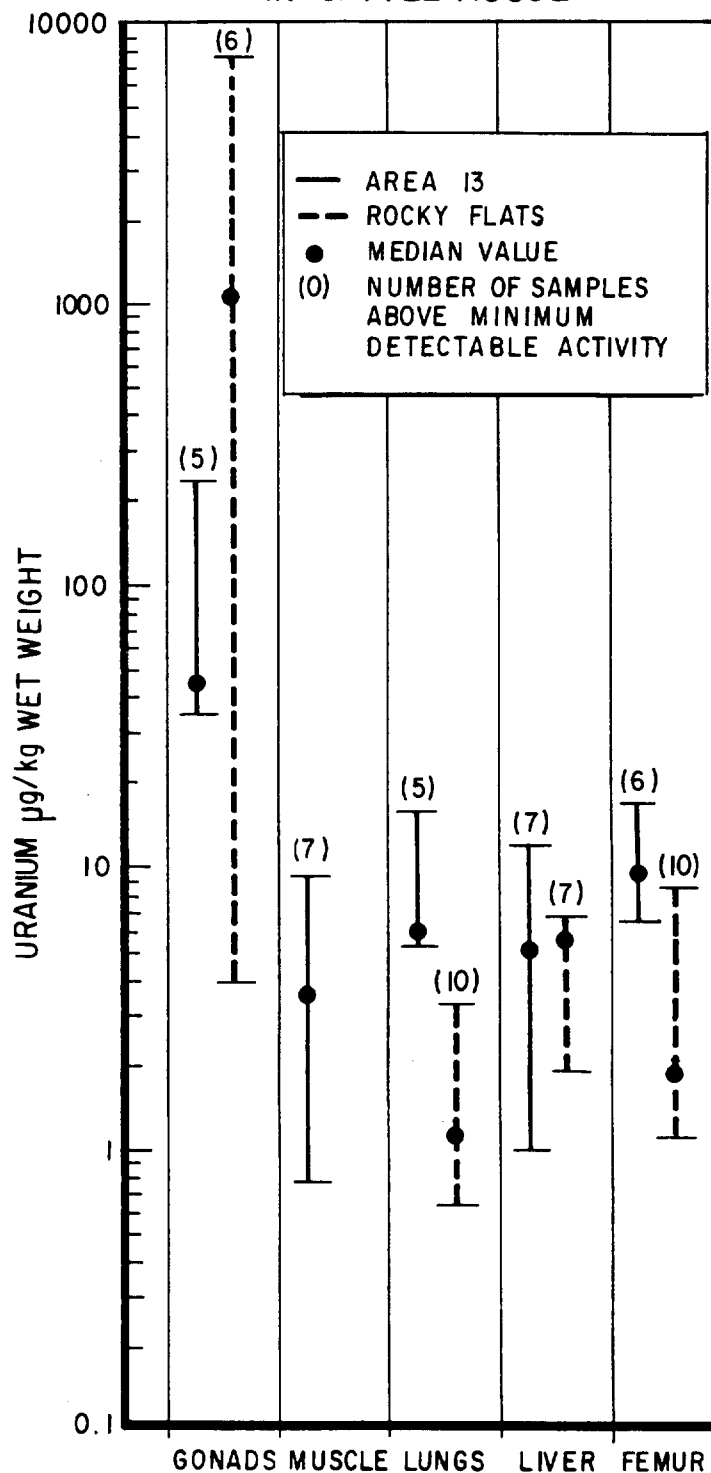


FIGURE 3

RANGES OF $^{239}\text{Pu} / ^{241}\text{Am}$ RATIOS IN SELECTED TISSUES FROM
CATTLE GRAZING AREA 13 vs THOSE GRAZING ROCKY FLATS

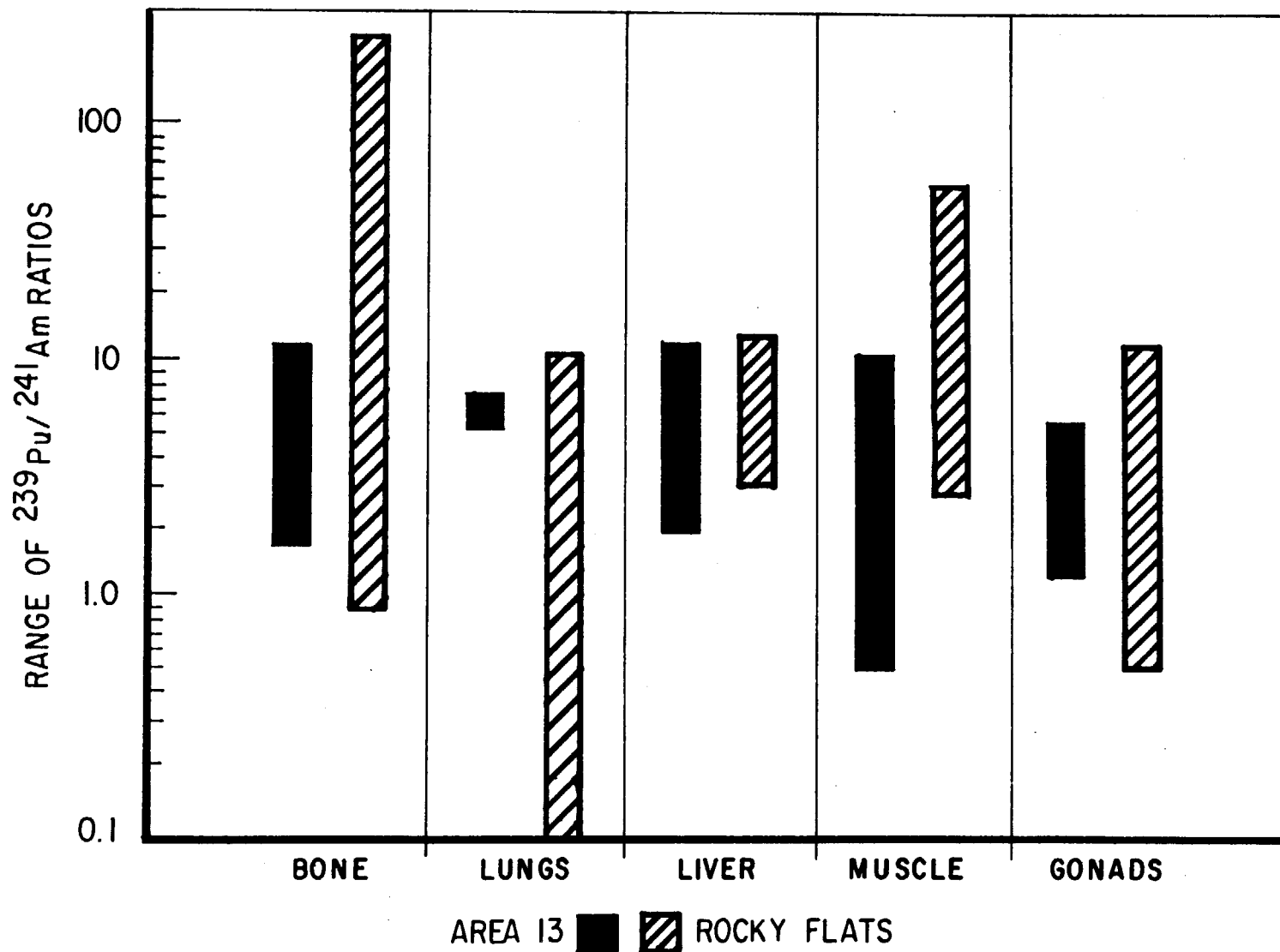


FIGURE 4

At previous NAEG meetings there frequently have been questions and discussions about the amount of soil a cow ingests during the grazing processes. Estimates of soil ingestion have been as high as 2 kg per day. A simple study was devised to provide an indication of the amount of soil ingested.

After the rumens of two fistulated steers were completely emptied of all ingesta, the rumen and reticulum were rinsed with water and the wash water bailed out to remove any residual sediments. This cleansing process was repeated three times. The animals were allowed to graze for 24 hrs on the selected range. Areas grazed included the well-grazed inner compound of Area 13 and an ungrazed range near White Rock Spring.

Following the grazing period, the ingesta was removed and agitated with water. The ingesta was washed through a screen (1.2- X 1.6-mm mesh) and the wash water saved. The rumen and reticulum were flushed with water three times and this wash water added to the ingesta wash water. The supernatant liquid was poured off and the sediments collected and heated at 450° C for 3 hr. This procedure oxidized any residual bits of vegetation. The residues were then weighed.

The same procedures were followed in the examination of the entire gastrointestinal tract from a cow sacrificed on January 28, 1976. This cow had grazed the outer compound of Area 13 for her entire life. The findings from both the fistulated steers and the sacrificed cow are summarized in Table 3.

It must be recognized that the sediment weights are approximate in that only those soil particles heavier than water were collected and undoubtedly some particles were entrapped in the villi or vegetation and were not released during the washing processes. Moreover, the data apply only to the individual animals on the day collected. However, the data from the permanent resident of the area (cow number 10) are considered to be significant as there would be no reason to believe that her grazing patterns would change significantly from day to day.

These data suggest that the total amount of soil ingested is much less than 2 kg per day, and that a reasonable estimate would be between 0.25 to 0.5 kg. This is still a significant amount, as some of the sand particles ingested could remain in the gastrointestinal tract for long periods of time. If such particles should contain relatively insoluble transuranic elements, this period would provide more time for reactions involved in gastrointestinal absorption, so uptake could be greater than would be derived from conventional studies carried out in ruminant digestion investigations.

When the fistulated steer data are examined, it is obvious that more soil is ingested from a heavily grazed area than from an ungrazed area. Also, the amount of soil increased with the amount of vegetation ingested. Neither of these observations is surprising. That more sediment is found in the rumens and reticula of the fistulated steers probably results from overeager grazing to fill the empty rumens. That is, they start out empty while the resident cow's rumen always contains some ingesta from the day before.

Table 3. Soil Sediments in the Ingesta

Animal Number	Date Sampled	Wet weight Area Grazed	Location of of Ingesta	Weight of Sediment	Sediment
761	01/21/76	Inner compound Area 13	14 kg	Reticulum & Rumen	57.3 g
774	01/21/76	Inner compound Area 13	22 kg	Reticulum & Rumen	278 g
729	01/30/76	White Rock Springs	9.3 kg	Reticulum & Rumen	2 g
707	01/30/76	White Rock Springs	6.4 kg	Reticulum & Rumen	28.9 g
10	01/28/76	Outer compound Area 13	Not weighed ~25 kg	Reticulum & Rumen	8.5 g
10	01/28/76	Outer compound Area 13	Not weighed	Omasum	8.6 g
10	01/28/76	Outer compound Area 13	Not weighed	Abomasum	8.2 g
10	01/28/76	Outer compound Area 13	Not weighed	Intestines	8.9 g

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PASSAGE OF SAND PARTICLES
THROUGH THE GASTROINTESTINAL TRACT OF DAIRY COWS

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ABSTRACT

A study was performed to determine whether the passage rate of particles through the bovine gastrointestinal system is related to particle size. Silica sand of four graded size ranges was obtained, and each size range was labeled with a different gamma-emitting radioisotope. The nominal diameters for the sand particle batches were 20, 80, 200, and 450 μm . A dose of each sand size was orally administered to each of four lactating Holstein dairy cows maintained in metabolism stalls. Fecal material from each cow was collected and analyzed by gamma-ray spectroscopy to determine the excretion rates for each particle size.

The data show that the passage time for soil particles through the gastrointestinal tract of dairy cows varies significantly with particle size. The smallest particles were passed rapidly by all cows; half of these were excreted within 1.7 days and 90 percent within 2.8 days of dosing. The three larger size particle groups were passed more slowly and with large variations among cows. Up to 8 days (average about 4 days) were required to excrete 50 percent of these larger particles and up to 12 days (average about 9 days) to excrete 90 percent of the particles in feces.

INTRODUCTION

The objective of this study was to determine whether the rates of passage or residence times for particles in the ruminant digestive tract are related to particle size.

Dairy cattle ingest moderate amounts of particulates during normal foraging. These particles are ingested after becoming deposited on forage plants by air currents, or because they are associated with plant roots ingested by cattle. In addition, soil that becomes deposited on the snouts of cattle is licked off and ingested. The gastrointestinal absorption efficiency (solubility) of material in the particles is related to the residence time of particles in the gut, and the residence time could vary for different sizes of particles which the animal may ingest.

Transuranic elements are usually in almost insoluble chemical forms when found in environmental media. However, it has been shown that relatively insoluble forms of plutonium are solubilized to a considerable degree in the gastrointestinal environment of cattle (Barth, 1975; Stanley *et al.*, 1975). Information on the gastrointestinal passage rates for various soil particle sizes is, therefore, relevant to the potential absorption efficiency for substances entrained in debris ingested by animals grazing in contaminated areas.

MATERIALS AND METHODS

Dose Material

The isotopes used as tracers were selected on the basis of noninterfering gamma-ray energies in order to facilitate analyses. The silica sand particles were sized and labeled with radionuclides at the Stanford Research Institute, Menlo Park, California (Lane, 1971). The radioisotopes were fused onto the sand and the particles were washed in dilute hydrochloric acid (pH of 1) prior to dose preparation. Less than 1% of the activity was removed by the acid wash. The particles appeared to be nearly spherical upon microscopic examination. Each of four size ranges of particles was labeled with a specific radionuclide as a tracer for a unique particle size. The labeled particles and associated parameters are described in Tables 1 and 2. The sand was weighed into gelatin capsules and administered orally via a balling gun. Each cow received individual doses of all four sand sizes. Weighed aliquots of each dose batch were used in preparation of gamma counting standards.

Facilities

The study was performed at the experimental farm operated by the U.S. Environmental Protection Agency at the Nevada Test Site. The cows were maintained in metabolism stalls located in a specially designed metabolism room. Water and hay (approximately 20 kg/day) were provided *ad libitum*. Pelleted dairy ration (2.3 kg/feeding) was provided at each milking, morning and evening. Animals were placed in metabolism stalls 24 hours before dose administration. An indwelling inflatable catheter was used for separating urine from feces. Fecal samples were collected in a grid-covered pan lined with polyethylene sheeting. The cows were milked morning and evening using a milking machine. Animal descriptions are given in Table 3.

Table 1. Description of Radioisotope-Labeled Sand Fed to Dairy Cows

Nominal Size* (μm)	Isotope Label	Weight per Particle** (mg)	Dose per Cow	
			(g)	(μCi)
20	^{141}Ce	9.7×10^{-6}	19	173
80	^{85}Sr	6.2×10^{-4}	15	116
200	^{54}Mn	9.7×10^{-3}	16	85
450	^{46}Sc	1.5×10^{-1}	40	96

*The nominal size is used in discussing the results. The size descriptions are:

20 μm - 95% of mass between 15 and 20 μm , and no particles greater than 25 μm ; separated by liquid sedimentation.

80 μm - pass 88 μm and retained on 74 μm sieves.

200 μm - pass 246 μm and retained on 175 μm sieves.

450 μm - pass 495 μm and retained on 417 μm sieves.

**Weight of nominal spherical size particle with specific gravity of 2.3.

Table 2. Characteristics of Isotopes Used to Label Sand Particles

Isotope	Half-Life (Days)	Gamma Energy (MeV)	Branching Ratio
^{141}Ce	32.5	0.145	48%
^{85}Sr	64.0	0.514	100%
^{54}Mn	312.5	0.835	100%
^{46}Sc	83.9	0.889	100%
		1.120	100%

Table 3. Holstein Dairy Animals Used to Study Rate of Passage for Sand Particles

Cow No.	Age (Years)	Weight (kg)		Days of Lactation	Average Milk Production (kg/day)**	Average Output (kg/day)***	
		Start	End*			Feces	Urine
123	6	891	848	227	9.6-5.3	27.3	21.8
153	5	832	766	32	22.3-19.1	35.1	32.0
184	4	739	682	159	12.7-8.9	33.2	28.8
241	3	580	553	156	14.6-12.1	28.4	23.1
*Weight after 8 days in metabolism stalls and 9 days in outside pens. **First number listed is the average milk production before study started, and the second is for the 7 days in the metabolism stalls. ***Average output while confined in metabolism stalls.							

Sampling

Fecal samples were collected every 4 hours for the first 40 hours after dosing in order to determine the time at which particles first appeared in feces. After a small aliquot was removed for analysis, the remainder of the fecal material was saved for compositing into a 24-hour sample. The 4-hour samples were mixed for 10 minutes with a Hobart mixer before aliquots were taken. The 24-hour samples, made up of 4-hour collections during the first 40 hours and fecal collections made at milking time thereafter, were also mixed with a Hobart mixer. Three aliquots of each 24-hour fecal sample were taken for analyses.

The cows were transferred to individual outdoor pens with concrete floors 7 days after dosing. Since some activity was still detectable in feces, grab samples of fresh fecal material were collected for 9 additional days. During this period, the daily particle output was estimated from the grab sample concentrations and the average fecal output rates for each cow.

Samples of urine, milk, and blood were collected 24 hours after dosing in order to check leaching from the particles and subsequent gastrointestinal uptake of the radionuclides.

Analyses

All samples were analyzed by gamma-ray spectrometry using a Ge(Li) detector (germanium-lithium drifted) and a pulse-height analyzer. Samples were counted so that the 2σ counting error was less than 10 percent or the counting time exceeded 80 minutes. The minimum detectable activity in the samples with <10% error at the 95% confidence level following 80-minute counting times was 2.2 nCi for ^{46}Sc , the isotope with the lowest counting efficiency. This amount is less than $3 \times 10^{-3}\%$ of the administered dose and represents less than 0.4% of the administered dose in a typical 30-kg 24-hour fecal collection. About 10% of the low activity samples were counted for 1,000 minutes. These samples included feces taken when isotope levels had decreased to very low levels, plus the blood, milk, and urine samples taken at 24 hours after dosing. The minimum detectable ^{46}Sc activity following 1,000-minute counts was 0.68 nCi at the 95% confidence level with 10% error. This amount is less than $10^{-3}\%$ of the administered dose and represents less than 0.1% of the administered dose in a typical 24-hour fecal collection.

Data given in the results section for 24-hour fecal samples represent the radioactive-decay-corrected average for the three aliquots from each collection.

RESULTS AND DISCUSSION

Initial Excretion of Particles

Particles, of each size were detected in feces from all cows at 12 to 16 hours after dosing. For the 20- μm particles, the highest concentrations in feces

occurred between 24 and 36 hours after dosing. The other three sizes generally reached maximum concentrations in feces at 36 to 64 hours after dosing.

Percent of Dose Retained vs. Time

Although considerable variation in excretion rates was observed among the four cows for the various sized particles, slower excretion rates were consistently associated with the larger particles.

The average percent of dose retained for the various particle sizes is given in Table 4 as a function of days after dosing. An indication of variability among cows is noted by the standard deviations. Data for the first 7 days, while the cows were in metabolism stalls, are shown in Figure 1. The three larger size particles tend to be excreted at similar rates, while the smallest particles are excreted much more rapidly. Data for each individual cow exhibited this tendency. However, as indicated in Table 4, the excretion rates for the three larger sized particles varied widely among the individual cows.

The average half-times for fecal excretion are given in Table 5. The half-times for particle excretion were obtained from the slopes of the best fit lines shown in Figure 1. These data indicate the excretion rate after excretion starts and neglect the initial period of retention from ingestion of initial excretion. While not of particular interest in this experiment, these data are included to facilitate comparison of results with those of other investigators. Such comparisons are given in the discussion section of this report.

The variability in excretion rates is obvious from the range of values for each particle size. Average rates were determined from a computed exponential best fit for all data points from the four cows during the 7 days in metabolism stalls.

Results of analyses on urine, blood, and milk showed that no significant *in vivo* leaching of the radionuclides from the particles occurred. The detection limits in these samples were less than $10^{-3}\%$ of the administered dose.

An important finding in this experiment was that essentially all the particles were recovered in feces. This suggests that very little of the sand was trapped in the fine structures of the GI tract.

Average half-time data from Table 5 are plotted vs. particle size in curve 1 of Figure 2. This information can be used to make an initial estimate of dairy cow excretion rates for ingested particles. However, the information on variability among animals, Tables 4 and 5, should also be considered when making such estimates.

Percent of Dose Retained vs. Fecal Output

Intuitively, it would appear that the particle elimination rate should be directly influenced by the fecal production rate for the cows. Data in Table 6 suggest little relationship between cumulative fecal output and the percent of particles excreted.

Table 4. Average Percent of Labeled Sand Dose Retained by Four Cows Following Oral Administration

Days After Dosing	Average Percent Retained $\pm$ One Standard Deviation			
	20 μ m	80 μ m	200 μ m	450 μ m
1	75 $\pm$ 8	97 $\pm$ 1	99 $\pm$ 1	99 $\pm$ 0.4
2	23 $\pm$ 7	80 $\pm$ 11	90 $\pm$ 7	93 $\pm$ 3
3	8 $\pm$ 6	60 $\pm$ 17	69 $\pm$ 24	76 $\pm$ 18
4	3 $\pm$ 4	46 $\pm$ 20	54 $\pm$ 32	62 $\pm$ 29
5	0.8 $\pm$ 0.8	34 $\pm$ 20	43 $\pm$ 33	53 $\pm$ 32
6	0.1 $\pm$ 0.2	24 $\pm$ 20	34 $\pm$ 33	44 $\pm$ 36
7	ND	17 $\pm$ 16	25 $\pm$ 24	34 $\pm$ 30
8*	ND	9 $\pm$ 9	15 $\pm$ 15	20 $\pm$ 17
9	ND	6 $\pm$ 5	9 $\pm$ 10	14 $\pm$ 12
10	ND	4 $\pm$ 3	7 $\pm$ 8	10 $\pm$ 8
11	ND	2 $\pm$ 2	5 $\pm$ 6	6 $\pm$ 6
12	ND	1 $\pm$ 1	3 $\pm$ 3	4 $\pm$ 4
13	ND	0.6 $\pm$ 0.5	1 $\pm$ 2	3 $\pm$ 3
14	ND	0.2 $\pm$ 0.2	0.5 $\pm$ 0.9	2 $\pm$ 3
15	ND	<0.1	0.2 $\pm$ 0.3	1 $\pm$ 2
16	ND	<0.1	<0.1	<0.1

*Estimates from grab samples were made after day 7.

ND--not detected.

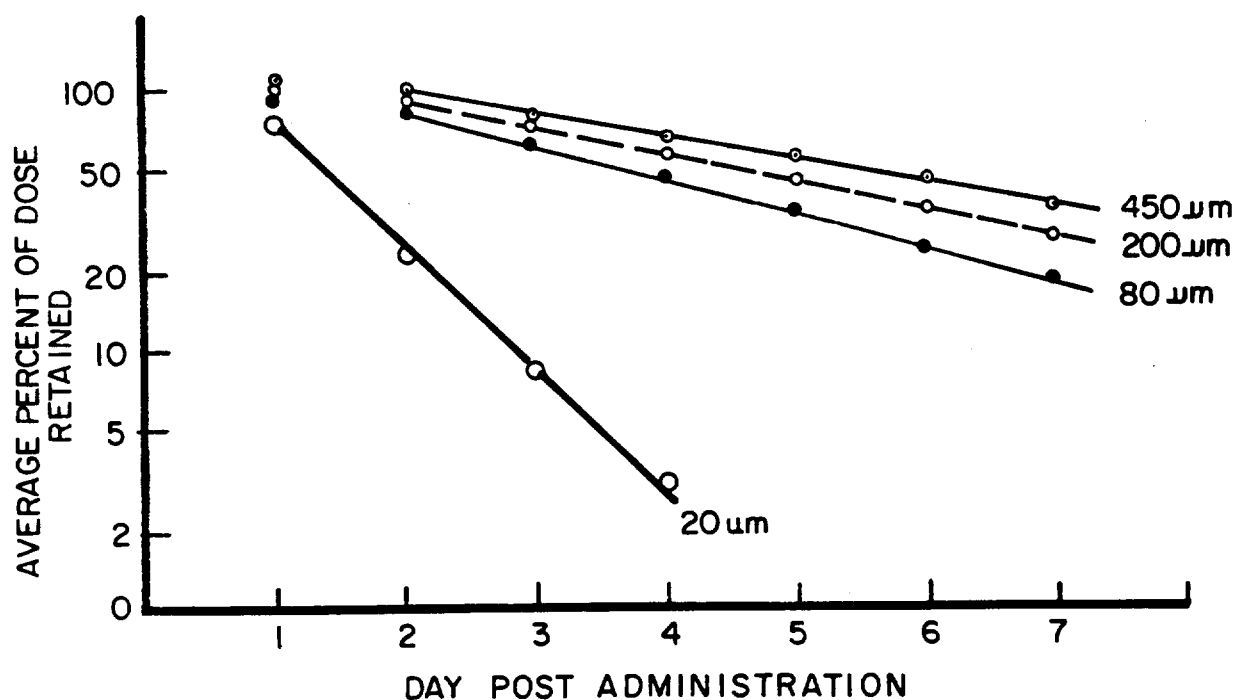


FIGURE 1. AVERAGE PERCENT OF VARIOUS SIZE SAND PARTICLES RETAINED BY COWS AFTER ORAL ADMINISTRATION.

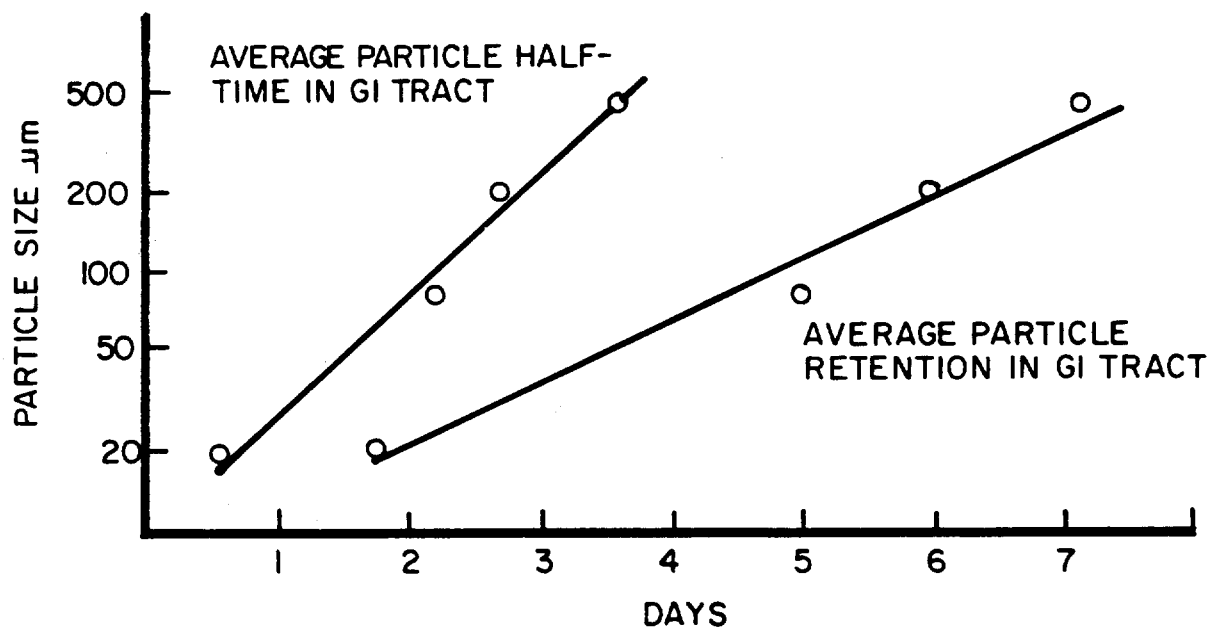


FIGURE 2. HALF-TIMES AND RETENTION TIMES FOR VARIOUS SIZE SILICA-SAND PARTICLES IN THE DAIRY COW GI TRACT.

Table 5. Fecal Excretion Half-Times for Particles

Particle Size (μm)	Excretion Half-Time in Days	
	Average	Range
20	0.55	0.4-0.6
80	2.2	1.0-4.6
200	2.7	1.1-7.7
450	3.6	1.5-13.9

Table 6. Cumulative Fecal Output for Excretion of One-Half the Particle Dose

Particle Size (μm)	Kg of Cumulative Fecal Output for 50% Excretion of Particles			
	Cow 123	Cow 153	Cow 184	Cow 241
20	25	40	40	26
80	100	82	192	72
200	205	83	330	74
450	450	64	260	93

The fecal material from all cows appeared to be similar in terms of apparent moisture content and gross consistency. While the approximate variation in daily fecal output between experimental animals was 25 percent, excretion rates for labeled sand particles were not related to the quantity of material that passed through the gastrointestinal tract.

Retention Time

The *mean retention time* can be calculated very simply if the cow gut is considered as a one-compartment model where mixing of ingesta is very rapid and emptying is by first-order kinetics. Under this assumption, the mean retention time would be the reciprocal of the first-order rate constant, or 1.44 times the biological half-time (Johnson and Lovaas, 1971). For these data, however, the area under the curve before excretion begins (Figure 1) is a considerable portion of the total area (especially for the 20- μ m particles), and such a calculated retention time would be useful mainly in comparison of particle kinetics. The biological half-time is equally useful in this regard. Other methods for calculating a mean retention time have been proposed (Balch and Campling, 1965; Phillipson, 1970; Johnson and Lovaas, 1971); however, variation among animals in this experiment indicates that exhaustive manipulation of these data is not warranted. A summary of retention time values from this experiment is given in Table 7. The definitions of the various terms are given in the footnotes to Table 7. The *average retention time*, or average time required for experiment cows to excrete 67% of the particles, is shown as curve 2 of Figure 2. This information is of value for initial estimates of dairy cow retention times for ingested particles. The information on variability among animals, Tables 4 through 7, should be noted in making such estimates. The average retention time is more useful than the biological half-time in estimating the period during which particles are subjected to digestive solubilization and absorption processes because it includes the initial period of retention in the rumen.

Discussion

Ruminant stomachs are of a compound nature and consist of four compartments, designated as rumen, reticulum, omasum, and abomasum. The anatomy and physiology of these compartments and the remainder of the gastrointestinal tract are thoroughly described in the literature (Annison and Lewis, 1959; Balch and Campling, 1965; Dukes, 1955; and Phillipson, 1970). Food materials remain in the cow rumen longer than in other segments. Once food material is passed to the fourth compartment, the abomasum, about 90% is excreted within 30 hours; from the rumen, about 100 hours are needed for 90% excretion (Phillipson, 1970). Digestive processes tend to allow accumulation of foreign debris in the reticulum and abomasum, while very little accumulates in the rumen and omasum. The material in the reticulum is liquid, so heavy foreign matter tends to settle out. Because the inlet and outlet of the reticulum are well above the fundus, heavy foreign matter is expelled with difficulty (Dukes, 1955).

A fairly wide variety of markers (e.g., dyed hay, plastic beads, and soluble unabsorbed radionuclides) have been used frequently to provide information for nutritional studies. Typical results for cattle fed marked food materials

Table 7. Retention Times for Sand Particles in Dairy Cows (Days)

Retention Time	Particle Size (μm)			
	20	80	200	450
Mean Retention Time*	0.8	3.2	3.9	5.2
Average Time for 50% Excretion**	1.7	3.5	4.2	5.1
Range	1.4-1.8	2.5-6.0	2.7-7.0	3.0-7.5
Average Retention Time***	1.8	5.0	6.0	7.3
Average Time for 90% Excretion**	2.8	7.6	8.7	10.0
Range	2.2-3.2	5.5-9.7	5.5-11.2	6.4-11.5

*Calculated as 1.44 times the average half-time given in Table 5.

**Time required after dosing to excrete X% of the particles, average and range for experimental data.

***Time required after dosing to excrete 67% of the particles, from experimental data.

show an initial appearance of the marker in feces at 12 to 24 hours after feeding, 80% excretion in 70 to 90 hours, and then slow excretion rates with complete excretion in 7 to 10 days. Experiments with other markers, e.g., chromic oxide and rubber or plastic particles, show that indicators are likely to pass at a different rate than the food with which they were ingested (Dukes, 1955). Moreover, it has been found that there is no consistent relationship between the nature of a cow's diet (e.g., straw, hay, or grass) and the length of time it is retained in the rumen (Annison and Lewis, 1959). The specific gravity of ingested particles is directly related to GI retention time, and the longer retention for higher specific gravities appears to be wholly accounted for by longer retention times in the reticulo-rumen, although the cow diet regime (e.g., feeding schedule and size of food particles) strongly influences rumen retention times (Balch and Campling, 1959). A nearly empty rumen has a short retention time for newly added material, and finely divided material tends to "float" through the system in a short time (Balch, 1950).

The results from this experiment indicate that the sand administered (specific gravity of about 2.3 and particle weights ranging from about 10^{-5} to 0.2 mg) was excreted about as would be expected from dyed-food feeding studies. The smallest particles appeared to "float" through the rumen, while the larger particles showed some relative delay in the excretion pattern. Plutonium oxide particles have a specific gravity of 11.46 and should be retained longer in the rumen than sand particles of comparable size. Plutonium particles or atoms in the environment may be bound or fused to siliceous materials and be excreted in a manner similar to sand.

The smallest particles first appeared in the feces at 8 to 12 hours after dosing, and the highest fecal concentration occurred at about 30 hours after dosing, which is about the same as reported for stained ground-hay particles (Dukes, 1955; Phillipson, 1970) and for small (25- to 30- μ m mean size) fallout particles (Potter *et al.*, 1971). The entire dose of these 20- μ m particles was excreted very rapidly, and the *average retention time* (Table 7, 1.8 days) was about that reported for stained ground-hay (Dukes, 1955; Phillipson, 1970) and for small fallout particles (Potter *et al.*, 1971). The *mean retention time* for soluble radionuclides in the dairy cow GI tract was reported to be 0.9 days (Osanov *et al.*, 1974), which is about that found for the 20- μ m particles in this experiment.

The three larger-sized particles, excreted in a similar fashion from the cows, were first detected in feces at 12 to 16 hours after dosing. However, the highest fecal concentrations occurred later than for the smallest particles, at 36 to 64 hours after dosing. An average of 7 to 10 days was required to excrete 90% of the larger particles. These times are similar to those reported for coarse roughage materials (e.g., long hay) fed to cows (Balch and Campling, 1965; Dukes, 1955; Phillipson, 1970).

The *mean retention time* for 88- to 175- μ m simulated-fallout particles was reported as 4.8 days in cattle (Johnson and Lovaas, 1971). This is slightly longer than the 3.2- and 3.9-day average values (Table 7) obtained in this experiment for 80- and 200- μ m particles, respectively.

Many of the characteristics of the cows used in this study are listed in Table 3. The variation among cows in sand particle excretion times shows no indication of correlation with:

- a. cow age,
- b. cow weight,
- c. cow weight losses during the study,
- d. days of lactation,
- e. fecal output rates,
- f. milk production rates,
- g. urine output rates,
- h. water consumption rates (as indicated by urine, milk, and feces production), or
- i. gross food consumption rates (as indicated by fecal output).

Results from this experiment indicate that the excretion patterns and retention times for various sizes of debris particles which cattle ingest are roughly predictable from previously reported observations for fine and coarse food materials. Although considerable variation was observed in retention times among the study animals on similar diet regimes, similar variation would be expected from results of conventional studies using marked food materials.

Since transuranic elements should be absorbed subsequent to the reticulorumen (Barth and Mullen, 1974), where larger particles are retained longer than small particles, the longer retention of large particles may offset somewhat the higher solubility rates for the smaller. The net result should be to cause the biological availability or absorption of transuranic elements from large and small particles to be less different than solubility rates (e.g., as a function of particle size) would indicate.

Because cattle are not normally maintained in very close proximity to nuclear events, most particles which could be ingested should be very small. By either air or water transport mechanisms, the large heavy particles tend not to be transported over large distances. Therefore, the rapid passage of the 20- μ m particles observed in this experiment is particularly pertinent to generally assess transuranic-particle passage through cattle.

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BIOLOGICAL TRANSPORT OF CURIUM-243 IN LACTATING DAIRY GOATS

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ABSTRACT

Six lactating dairy goats (average weight 43.5 kg) were given either intravenous or oral doses of curium-243 chloride. Three animals each received a single 20.8- μ Ci intravenous citrate-buffered injection and the remaining three goats were given a single oral dose of 200 μ Ci per animal. Milk, urine, fecal, and blood samples were collected daily over a 144-hour period after dosing and the animals were sacrificed approximately 162 hours post-treatment. Selected tissues were collected and analyzed for curium content.

Curium concentrations in the sample material were determined by counting the 228-keV and the 278-keV gamma rays of curium-243 using a NaI(Tl) detector and pulse height analyzer. Analytical results to date have shown that at least 99 percent of the oral dose was excreted in the feces with low to nondetectable concentrations in the milk and tissues. However, approximately 2 percent of the intravenous dose was transferred to milk during the 144-hour collection period. The average transfer to urine and feces in these injected goats was 4.6 and 4.5 percent of the administered dose, respectively. Curium retention in the tissues collected for analysis totaled approximately 64 percent of the injected dose, with essentially half of this retained curium found in the liver.

Comparisons between the transport of plutonium and curium in dairy goats are discussed.

INTRODUCTION

An increased research and development effort is currently being directed toward the use of radiofissionable fuels as power sources. Current projections of future energy requirements suggest that isotopes of uranium and plutonium

may need to be recovered from spent reactor fuels and refabricated into new fuel elements. As a by-product of fuel reprocessing, substantial amounts of curium will be available. This likelihood of increased curium production has, therefore, elevated concern for knowledge of its potential environmental transport following accidental contamination.

Most of the previous conclusions on the biological transport of curium have strongly indicated that actual curium data must be obtained directly, rather than by making extrapolations from previously collected plutonium data. Retention differences between plutonium and curium have been noted in bone following intramuscular nuclide doses. One month after an intramuscular dose of curium-242 nitrate was given to rats, 20 percent of the total injected dose was retained in bone, compared to 41 percent for plutonium nitrate (Nenot *et al.*, 1972). Plutonium was also apparently bound more firmly by protein in many molecular situations (Taylor, 1972), and it has been suggested that perhaps complex formation with protein may be more important in plutonium transport and tissue deposition than is the case with curium.

McClellan *et al.* (1972) exposed 24 beagle dogs to aerosols of curium-244 which were inhaled as either the chloride or the oxide. The rapid loss of curium from liver previously observed in rats (Hamilton, 1947) was not noted in dogs. Urinary clearance and tissue retention patterns were somewhat similar between dogs exposed to the relatively soluble chloride and those exposed to the relatively insoluble oxide. Information obtained from this beagle study differed somewhat from the transport characteristics of various plutonium forms (Bair, 1970), but it has also been recognized that conclusions derived from inhalation studies necessitate a consideration of particle size (Bair, 1970; McClellan, 1972) as well as the anatomic location of particle deposition (Stather and Howden, 1975).

Results from another study using beagle dogs (Lloyd *et al.*, 1974) have indicated that intravenously injected curium is excreted primarily in the urine. These investigators injected five beagles with approximately 2.6 μCi Cm-citrate/kg and noted that the average urinary curium output one day after injection was approximately five times greater than the average fecal excretion of curium. Furthermore, tissue concentrations one week after injection revealed that approximately 39 and 37 percent of the curium dose had been retained in the canine liver and bone, respectively.

Curium metabolism in dairy animals has not been extensively investigated. However, it is particularly important to establish the metabolic retention and excretion patterns of curium in those domestic animals maintained as a source of food for the human population. The current experiment using dairy goats was designed to examine the metabolic pattern of curium following oral and intravenous exposure. Additional objectives were to compare the biological transport of curium and plutonium in the goat for relative hazard evaluation and to help establish an appropriate oral curium dose for more elaborate future studies using the major milk producers, i.e., dairy cows.

MATERIALS AND METHODS

Six lactating goats were maintained in specially constructed stalls which allowed for the separate collection of urine and feces. Three of these goats received acute oral doses of curium-243 (200 μCi per animal), and the other three goats received intravenous curium-243 doses of 20.8 μCi per animal. Table 1 provides some general information on the animals used during this study. The goats were between 1 and 3 years of age, and had an average pre-study weight of 43.5 kg. Oral and intravenous doses were approximately 4.7 $\mu\text{Ci}/\text{kg}$ and 0.47 $\mu\text{Ci}/\text{kg}$, respectively, but as noted, no dose adjustments were made for individual variations in animal weight. Composite daily collections of milk, urine, and feces were sampled from each goat over a 144-hour period after dosing. Blood samples were also taken during this period.

Urine was collected with in-dwelling inflatable catheters which drained into polyethylene bottles. The goats were milked by hand twice daily, and milk from AM and PM milkings was mixed in plastic bottles. All urine, milk, and fecal collections were weighed. Three weighed aliquots were then taken from the respective composites and placed in individual aluminum cans with formaldehyde added as preservative. Addition of formaldehyde prior to mixing the fecal collections was advantageous due to the pellet nature of goat feces. Blood samples, collected by jugular venipuncture, were centrifuged and the plasma and cells separated using a disposable pipette. The packed cells were washed two times with physiological saline. Samples of plasma and cells were individually diluted with distilled water. Formaldehyde was then added as a preservative and the sample containers were sealed.

The animals were sacrificed using intravenously administered euthanasia solution approximately 162 hours post-treatment. Sacrifice collections included samples of bone, liver, kidney, gall bladder, bile, lung, gonads, spleen, muscle, lymph nodes, heart, thymus, adrenals, gastrointestinal tract, and mammary gland. Ten ml of formaldehyde was added to each tissue sample before the samples were individually sealed in aluminum cans. At time of sacrifice, total weights were taken on most organs so that the percentage of administered dose retained by a specific tissue or organ could be calculated. This obviously was not practical in the case of some tissues, e.g., muscle, bone, etc. Under these conditions, total curium content was based on calculated organ weights using the respective percentage of body weight reported by Davis *et al.* (1975). Curium concentrations from the femur (diaphysis and epiphysis) and sternum were averaged to estimate the osseous retention values.

The curium-243 stock material was obtained as a nitrate salt from Oak Ridge National Laboratory, and contained curium-244 as a major impurity (55.9 atom % curium-243, 42.1 atom % curium-244). Intravenous doses, approximately 5 ml in volume and buffered to a pH of 6 with citrate (0.15 M NaCl--0.05 M sodium citrate--0.0025 M citric acid), were administered by jugular venipuncture. The oral doses (in 100 μl of 0.6 M HCl) were placed in gelatin capsules containing cellulose fiber and administered using a balling gun.

Table 1. Summary Information on the Six Goats Used to Investigate the Biological Transport of Curium-243

Goat Number	^{243}Cm Dose	Animal Age (y)	Pre-treatment Weight (kg)	Sacrifice Weight (kg)	Average Daily Output During Study (kg)		
					Milk	Urine	Feces
	<u>Oral Dose</u>						
1	200 μCi	2	43.5	37.5	1.3	1.0	0.8
2	200 μCi	1	38.5	35.5	1.2	0.9	0.7
3	200 μCi	3	46.5	49.5	1.5	1.3	0.8
	<u>I.V. Dose</u>						
4	20.8 μCi	2	53.5	52.0	1.5	1.4	0.9
5	20.8 μCi	3	44.5	39.0	0.9	0.8	0.3
6	20.8 μCi	1	34.5	33.0	1.1	1.0	0.6

Since routine radiochemical assay procedures for curium are still somewhat in the development stage, gamma counting was considered the most feasible analytical technique for this experiment. Curium concentrations in the sample material were, therefore, determined by counting the 228 and 278 keV gamma rays of curium-243 using a NaI(Tl) detector and pulse height analyzer.

Overall measurement error was assessed by considering potential uncertainties in each step of the sampling and analytical scheme. The NaI(Tl) detector was energy-calibrated with appropriate gamma-ray emitting nuclides. Checks were also made for gain shifts and changes in efficiency after every 10 samples with an aliquot of the dosing solution. Backgrounds were taken before, during, and after a series of counts to confirm that contamination of the counting chamber had not occurred. To establish differences in counting variability between sample types (milk, urine, feces, etc.) and the administered dose, "spiked" samples of each type were prepared in triplicate and assayed. These "spiked" samples were recounted after a period of time to determine any time-related variations due to the settling out of sample material. Throughout the experiment, multiple samples were collected for analysis. Tissue samples were not homogenized but are being counted twice (top-bottom rotation). The variability between these two tissue counts will be used in the overall assessment of those uncertainties not associated with analytical hardware and counting duration.

RESULTS AND DISCUSSION

All samples collected during this study have been analyzed at least once, but because of a few very low curium concentrations, a portion of the samples are being recounted (1,000-minute counts). Furthermore, to determine the feasibility of radiochemical assays, selected animal tissues and some metabolic (urine, milk, etc.) samples will be prepared for analysis by an outside laboratory. As a result, data reported below are not totally comprehensive, but they do provide significant information on the caprine absorption of orally administered curium. Considerably more information is given on the biological transport of intravenously administered curium.

Experimental data were initially calculated as the curium concentration per gram of sample. For milk, urine, and fecal samples, the concentration per gram of sample was multiplied by the respective total daily output (weight) of these substances. Results for the intravenously dosed goats are presented as a percentage of administered dose per total collection (Table 2) and as a percentage of dose per kg of material (Table 3).

In the case of the orally dosed goats, approximately 99 percent of the dose was recovered in the feces; the majority of milk, urine, and blood samples had no detectable curium concentrations. Curium concentrations in the feces from these orally dosed goats were relatively high from 24 to 96 hours after dosing. The greatest curium output occurred in the feces at the 48-hour collection period.

Table 2. Percentage of Curium-243 Dose Transferred to Milk, Urine, and Feces of Three Goats for 144 Hours After Each Animal Had Received a Single 20.8 μ Ci Intravenous Injection of Citrate-Buffered Curium-243 Chloride

Time post-injection (hrs)	(Transfer to MILK)		
	Goat #4	Goat #5	Goat #6
8	0.894	0.713	0.814
24	0.387	0.850	0.291
48	0.211	0.504	0.272
72	0.112	0.303	0.146
96	0.0803	0.180	0.0880
120	0.0731	0.125	0.0716
144	0.0572	0.0856	0.0495
Total	1.82	2.76	1.73
Time post-injection (hrs)	(Transfer to URINE)		
	Goat #4	Goat #5	Goat #6
8	2.57	1.90	1.77
24	1.11	0.442	1.43
48	0.583	0.353	0.247
72	0.349	0.220	0.316
96	0.372	0.177	0.326
120	0.459	0.153	0.264
144	0.287	0.146	0.188
Total	5.73	3.39	4.54
Time post-injection (hrs)	(Transfer to FECES)		
	Goat #4	Goat #5	Goat #6
8	0.00510	0.00269	0.00284
24	0.0207	0.0856	0.102
48	0.834	0.594	0.654
72	0.739	0.483	0.440
96	1.48	0.724	0.580
120	1.88	0.656	0.867
144	1.46	1.31	0.640
Total	6.42	3.86	3.29

72	1.51	2.28	0.885
96	1.91	1.79	0.914
120	1.47	2.56	1.12
144	1.15	3.23	0.721

Goats that received curium intravenously retained approximately 64 percent of the injected dose at time of sacrifice (Table 4). As noted in the table, nearly half of this retained curium was found in the liver. On both a percentage of dose per organ and a percentage of dose per kilogram basis, liver, bone, and kidney had consistently high curium values. The thyroid gland also had readily detectable curium-243 concentrations. Individual curium concentrations for bile are probably of doubtful significance since several variables affect the volume of bile produced. However, a large part of the fecal curium (for the I.V. dosed goats) was probably transported to the gastrointestinal tract by the bile. The total curium output in the feces of the three intravenously injected goats averaged 4.5 percent of the administered dose.

Curium concentrations in the tissues taken from the orally dosed animals were of course much lower than the concentrations noted in the intravenously exposed goats, but the basic distribution pattern was similar. Liver, bone, and kidney retained the highest concentrations of curium on a percentage of dose per kilogram basis. However, the above-mentioned organs plus cardiac muscle were the only tissues from the orally dosed group to consistently have detectable amounts of curium. Furthermore, the total amount of curium-243 retained in all tissues collected from any individual goat did not exceed 0.01 percent of the original oral dose.

Previously acquired information on the biological transport of plutonium following a 50 μ Ci acute intravenous injection of citrate-buffered plutonium-238 nitrate to one lactating goat is presented in Table 5 (Stanley and Mullen, 1971). The two groups of data, expressed as a percentage of administered dose per kilogram of milk, urine, or feces, are not strictly comparable. Plutonium data are based on a single goat, while the curium results shown represent an average of the three goats used in this study. There were also slight differences in dosing and collection schedules, and no comparison has been included for the detection efficiency of the analytical techniques. Nonetheless, this information does provide an opportunity for a gross comparison between the physiological transport of plutonium and curium in the same ruminant species under somewhat similar conditions.

Curium was excreted more rapidly than plutonium in the first week post-injection. Shortly after injection, the percentage of administered dose per kilogram of milk and urine was noticeably higher for curium (Table 5). On this percentage of dose per kilogram basis, curium values remained consistently greater than plutonium values in most milk, urine, and fecal collections. This relationship between curium and plutonium was also evident on a total excretion basis. The total amount of curium transported to the milk and urine by 144 hours post-injection was 2.2 and 4.6 percent of dose, respectively. Total plutonium transport to milk and urine over this 144-hour period was 0.8 and 1.8 percent of the administered dose.

Table 4. Curium Retention Pattern in Three Dairy Goats 162 Hours After Each Animal Had Received a Single Intravenous Dose (20.8 μ Ci/animal) of Citrate-Buffered Curium-243 Chloride

Tissue/Organ	Percentage of Injected Curium-243 Retained		
	Goat #4	Goat #5	Goat #6
Liver	37.7	40.3	33.6
Bone	17.3	13.5	21.9
Muscle	3.86	7.68	3.03
Kidney	1.30	1.89	1.53
Lung	0.607	0.832	0.753
Heart	0.260	0.487	0.192
Spleen	0.0133	0.133	0.119
Bile	0.0134	0.0260	0.0591
Thyroid	0.00712	0.00871	0.00529
Gonads	0.00401	0.00564	0.00505
Tissue/Organ	Percentage of Injected Curium-243 Retained per kg		
	Goat #4	Goat #5	Goat #6
Liver	30.3	46.0	50.6
Kidney	7.30	11.4	13.7
Bone	5.28	5.46	10.5
Bile	2.68	2.36	1.25
Thyroid	1.18	2.18	1.70
Heart	1.04	2.59	1.24
Gonads	0.803	1.88	1.68
Lung	0.914	1.49	1.49
Spleen	0.0721	0.899	1.89
Muscle	0.164	0.433	0.202

Table 5. Percentage of Dose per kg of Milk, Urine, and Feces From Dairy Goats Following an Intravenous Dose of Either Curium-243 or Plutonium-238

Average of 3 goats (20.8 μ Ci of citrate-buffered curium-243 chloride per animal):			
Time post-injection (hrs)	Milk	Urine	Feces
8	1.59	6.86	0.0417
24	0.612	1.32	0.301
48	0.288	0.500	1.60
72	0.181	0.356	1.56
96	0.109	0.284	1.54
120	0.0866	0.229	1.72
144	0.0561	0.147	1.70
One goat (50 μ Ci of citrate-buffered plutonium-238 nitrate):			
Time post-injection (hrs)	Milk*	Urine*	Feces
6	0.117	2.54	0.0280
23	0.210	0.256	0.692
47	0.0458	0.0250	0.420
71	0.0188	0.0170	0.418
95	0.0180	0.0166	0.480
119	0.0142	0.0212	0.406
143	0.00840	0.0104	0.318

*Based on volume, not weight (%/l)

FUTURE PLANS

While caprine studies are definitely a logical step toward understanding the transport of curium to milk, definitive studies must ultimately be conducted using the major milk producing animal, the cow. The second curium study in this series will use two lactating dairy cows. One animal will receive an acute oral dose, and the second cow will be given a single curium dose via intravenous injection. Initial selection of an appropriate oral dose usually represents an estimate based on several considerations. Dosing considerations in this case include the observed percentage transfer of intravenously administered curium-243 to goat's milk, a best estimate gut reduction factor, the approximate detection limits for curium-243 analyses on milk, and the probable bovine milk production. Collection procedures will be similar to those just described in the caprine experiment. Milk, urine, feces, and blood samples will be collected for approximately 144 hours after dosing. Both animals will then be sacrificed and tissue samples analyzed for curium content.

Metabolism studies will also be conducted this fiscal year to determine the biological transport of neptunium in dairy animals. These experiments are still in the planning stage, but will be basically similar to those described for curium. Dairy goats will be used in the first project where individual animals will receive either oral or intravenous neptunium doses. Subsequent neptunium experiments will then be conducted in the fall of 1976 using dairy cows.

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BOVINE TRANSPORT AND RETENTION
OF PLUTONIUM-238 WITH SPECIAL EMPHASIS
ON THE GASTROINTESTINAL UPTAKE OF
IN VIVO LABELED MILK

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ABSTRACT

A two-phase experiment was conducted to determine whether *in vivo* plutonium-labeled milk presents the nuclide in a more biologically available form than the *in vitro* plutonium preparations typically administered for intestinal uptake studies. Dairy calves were fed either *in vivo* plutonium-labeled milk or an *in vitro* plutonium-labeled milk prepared through the addition of plutonium citrate to uncontaminated milk. Plutonium retained in the tissues, collected at the time of calf sacrifice, will be used to compare the relative biological availability of plutonium in the two treatment groups. Analytical results are not currently available on these calf tissues but will be presented later.

An additional objective was to examine the excretion patterns and tissue retention of plutonium in the dairy cows used to produce the *in vivo* plutonium-labeled milk. A total of six cows received various acute intravenous doses of citrate-buffered plutonium-238 nitrate, and the subsequent transport to milk, urine, and feces is reported. Tissue samples were taken from four of the adult cows, but the plutonium concentration in these samples, along with the above-mentioned calf tissue samples, will be presented later in the comprehensive report.

INTRODUCTION

The long physical and biological half-life of plutonium has dictated that considerable effort be devoted to quantifying plutonium transport through the various trophic levels. Results on the absorption, distribution, and excretion

of plutonium by dairy cows have been reported at previous Nevada Applied Ecology Group meetings. These studies were undertaken by (1) establishing what portion of plutonium was retained in the tissues following initial absorption; (2) determining the amount of activity transported to milk, urine, and feces; and (3) observing these phenomena after various chemical forms of the nuclide were administered orally. Due to the large reduction factor occurring with gastrointestinal absorption, the transport and retention characteristics were based on relatively low levels of systemic accumulation. Comparisons should be made on the percentage of plutonium transported to milk and edible bovine tissues following a larger systemic dose.

Transport coefficients are frequently valid only under experimentally defined conditions and some reports have addressed factors that can cause variations in plutonium uptake. Ragan (1975) noted an approximately fourfold increase for the gastrointestinal absorption of plutonium-239 citrate in iron-deficient mice. There was also a more rapid translocation of plutonium from soft tissues to bone in these iron-deficient mice. Another problem of potential concern is whether *in vivo* plutonium-labeled milk represents a more biologically available nuclide form than the various *in vitro* plutonium preparations typically administered to establish gastrointestinal transport coefficients. This question is particularly relevant in reference to juvenile animals, not only because human infants consume relatively large quantities of milk, but also because of the increased juvenile gut absorption of plutonium in some species (Ballou, 1958).

To obtain information on the biological availability of *in vivo* plutonium-labeled milk, two closely related studies were performed and these experiments have been outlined in this paper. A basic objective was to compare the gastrointestinal uptake of *in vivo* and *in vitro* plutonium-labeled milk by dairy calves. The definitive phase of these experiments has just been completed and the samples of primary concern, i.e., calf tissues, have not been analyzed. This paper should therefore be considered as a progress report.

METHODS AND MATERIALS

The experiments were conducted in two phases. A feasibility study (Phase I) used a total of two lactating cows and four calves, while the more elaborate definitive study (Phase II) used four cows and twelve calves.

Phase I

Phase I was directed primarily toward confirming the approximate quantity of plutonium for two different doses, an oral dose for calves and an intravenous dose for the adult cows. Selection of an appropriate dose for the calves concerned plutonium concentrations needed for the *in vivo* labeled milk. This *in vivo* plutonium-labeled milk would have to contain a sufficiently high nuclide concentration to allow for the subsequent detection of plutonium in selected calf tissues. The second dose requiring confirmation was directly

related to the oral dose for calves. It concerned the original intravenous dose to adult cows that would eventually result in the appropriate plutonium concentration per gram of milk. It should be noted that there are normal variations in daily milk production which can apparently alter either the plutonium concentration per gram of milk or the total amount of plutonium transferred to milk per collection. Furthermore, while the daily milk production of adult Holstein cows often approximates 20 liters, an individual 10-day-old calf will ingest only 2 to 4 liters of this milk per day.

Table 1 presents some background information on the animals and the plutonium doses administered during the feasibility study. As stated above, all intravenous doses to the lactating cows were given as citrate-buffered plutonium-238 nitrate injections. The first adult cow received a single 83 mCi injection. Milk, urine, and fecal samples were taken for 72 hours after injection, but no tissue samples were collected at time of sacrifice 90 hours post-dosing. The second adult cow received an initial injection of 9.95 mCi following which milk, urine, and fecal samples were collected. As noted in Table 1, three additional plutonium injections were sequentially administered to this animal (one per day for three consecutive days) beginning 144 hours after the initial dose.

The *in vivo* plutonium-labeled milk collected following these sequential injections was fed to two calves. Background information on these calves is also presented in Table 1. The second adult cow was sacrificed approximately 13 days after the initial dose, but no tissues were collected.

The calves ingested plutonium-labeled milk for 6 days. For 2 days after the last treatment feeding, the calves were given uncontaminated milk from the dairy herd. All calves were then sacrificed, including two control calves that had received uncontaminated milk throughout the study. Calf tissues were taken at sacrifice and included samples of liver, lung, bone (femur-diaphysis and epiphysis, sternum and marrow), gall bladder and bile, gonads, duodenum (mucosa and serosa), spleen, muscle (skeletal and cardiac), lymph nodes, thymus, adrenals, thyroid, abomasum, omasum, reticulum, rumen, and blood.

Phase II

Information gained during the feasibility study was applied to the design of a definitive effort (Phase II). This Phase II program has recently been conducted and will thus be briefly summarized. Tables 2 and 3 provide an outline for this portion of the project. Four adult cows were given a single intravenous injection of citrate-buffered plutonium-238 nitrate (approximately 16.6 mCi per animal). Plutonium-labeled milk from these cows, collected during the period of peak concentration, was fed to four calves. Four additional calves received *in vitro* plutonium-labeled milk prepared with approximately the same plutonium concentration as in the *in vivo* labeled milk. Each group of calves contained two control animals which were sacrificed along with the experimental calves. Tissues were collected from 11 of the 12 calves for plutonium analysis. Furthermore, for the Phase II effort, tissues were taken from the adult cows and analyzed for plutonium content. Additional details of this second phase experiment will be presented, along with the analytical results, in a subsequent comprehensive report.

Table 1. Adult and Juvenile Animals Used in the Phase I Feasibility Study

Cow #1	I.V. ^{238}Pu Dose 83 mCi	Weight (kg) 641	Age (Years) 4	Sacrifice Time After Dosing 90 hours			
Cow #2	I.V. ^{238}Pu Dose 9.95 mCi 10.6 mCi 10.6 mCi 10.7 mCi	Time After First Dose (hours) - 144 168 192	Weight (kg) 614	Age (Years) 3	Sacrifice Time Post Initial Dosing (days) 13		
Calf Number	Dose	Location of Calf During Study	Sex	Age at Beginning of Study (days)	Pre-Study Weight (kg)	Sacrifice Weight (kg)	Age at Sacrifice (days)
#2637	174 μCi in <i>vivo</i> plu- tonium-labeled milk	Metabolism Room	Male	6	45.0	44.6	17
#2638	153 μCi in <i>vivo</i> plu- tonium-labeled milk	Metabolism Room	Male	6	35.9	36.8	17
#2641	Control	Metabolism Room	Male	6	43.2	40.0	17
#2634	Control	Outside Pen	Male	6	35.5	48.2	17

Table 2. Summary Information on the Four Lactating Dairy Cows Used in the Phase II Definitive Experiment

Cow Number	^{238}Pu I.V. Dose (mCi)	Pre-Study Weight (kg)	Dose (μCi) per kg	Animal Age (Years)	Approximate Sacrifice Time Post-Dosing
119	16.0	843	19.0	8	135 hours
179	16.3	764	21.3	6	140 hours
123	17.5	863	20.3	7	13 days
128	16.6	736	22.6	7	13 days

Table 3. Summary Information on the Twelve Calves Used (in Phase II) to Investigate the Relative Biological Availability of *In Vivo* and *In Vitro* Plutonium-238 Labeled Milk

Calves Receiving <i>In Vivo</i> Plutonium-238 Labeled Milk					
Calf Number	Pre-Study Weight (kg)	Sex	Pre-Study Age (days)	Sacrifice Age (days)	Sacrifice Weight (kg)
92*	38.6	Male	7	16	30.9
96	35.9	Male	7	16	32.7
7278	27.7	Female	7	16	30.0
101	34.1	Male	8	17	34.1
7284	30.9	Female	7	16	30.0
93**	35.0	Male	7	16	31.8
Calves Receiving <i>In Vitro</i> Plutonium-238 Labeled Milk					
Calf Number	Pre-Study Weight (kg)	Sex	Pre-Study Age (days)	Sacrifice Age (days)	Sacrifice Weight (kg)
7359	33.8	Female	4	13	31.4
170	41.3	Male	4	8***	-
7349*	35.8	Female	6	15	33.2
163	42.3	Male	5	14	40.0
NT	29.3	Female	7	16	28.2
171**	42.8	Male	4	13	40.0

*Control animal maintained with experimental calves

**Control animal maintained in outside pen

***Age at time of death, no tissue samples collected

Both phases of the experiment were conducted at the Nevada Test Site (Area 15) Farm and made use of the recently installed waste disposal system. Adult cows were confined to metal metabolism stalls designed for the total collection of urine and feces. The cows were catheterized with an indwelling inflatable urinary catheter. Urine was collected in 20-liter plastic bottles placed at the rear of the stall and connected to the catheter by polyethylene tubing, while a grid-covered pan lined with polyethylene sheeting was used to collect the feces. A Hobart mixer was used to mix the fecal collections prior to sampling. Milk was collected with individual bucket milkers.

Blood samples, collected by jugular venipuncture, were taken periodically from the adult animals. Blood collections were centrifuged and the plasma and cells separated using a disposable pipette. Packed cells were washed with physiological saline solution during the preparation process.

Calves were kept in small metabolism stalls located in the same room as the adult cows. Urine and feces were collected to prevent floor contamination but, in the case of the calves, were not analyzed for plutonium content. Milk collected from the plutonium-treated cows was placed in suitable buckets and fed to the calves. During Phase II, milk collected during the period of peak plutonium concentration was refrigerated and saved for subsequent calf feedings. The refrigerated milk was warmed in a modified water bath prior to feeding. In both phases, each calf had an individual plastic feeding bucket to reduce the possibility of cross contamination.

DOSE ADMINISTRATION AND SAMPLE ANALYSIS

Plutonium-238 was obtained as a dioxide from the Oak Ridge National Laboratory. It was dissolved in concentrated HNO_3 with a trace of HF before being converted to the citrate form. The doses were calibrated by liquid scintillation counting of the plutonium-238 alpha particles.

Radionuclide solutions, approximately 5 ml by volume and 5 to 6 in pH, were administered to the adult cows by jugular venipuncture. The *in vitro* plutonium-labeled milk, given to the Phase II calves, was prepared by the addition of approximately 5 ml of a plutonium citrate solution per gallon of uncontaminated milk. This *in vitro* labeled milk was thoroughly shaken and samples were removed for direct counting to ensure homogeneity and known dosing concentrations.

Samples collected throughout the study were analyzed for plutonium-238, based on the 17 keV X ray from the plutonium isotope. All samples were counted in 200-ml aluminum cans using a phoswich detector, containing a thin NaI scintillator, backed by a thick CsI scintillator. Overall measurement error was assessed by considering potential uncertainties in each step of the sampling and analytical scheme. The phoswich detector was energy-calibrated with appropriate plutonium-238 solutions. Checks were also made for gain shifts and changes in efficiency after every 10 samples with an aliquot of the dosing solution. Backgrounds were taken before, during, and after a series of counts to confirm that contamination of the counting chamber had not occurred. To establish differences in counting variability between sample types (milk,

urine, feces, etc.) and the administered dose, "spiked" samples of each type were prepared in triplicate and assayed. These "spiked" samples were also recounted after a period of time to determine any time-related variations due to the settling out of sample material. In many cases, multiple samples were collected for analysis. Tissue samples were not homogenized, but selected samples were counted twice (top-bottom rotation). The variability between these two tissue counts will be used in the overall assessment of those uncertainties not associated with counting hardware and counting duration.

Selected samples (primarily of low plutonium concentrations) are also being prepared for analyses by another laboratory which will employ more sensitive assay techniques.

RESULTS AND DISCUSSION

In this report, Phases I and II have been discussed separately in order to emphasize the experimental approach and to sequentially present some of the resulting data.

Phase I

The total plutonium transported to milk during the first 72 hours following intravenous plutonium injections was essentially one percent of the administered dose (Tables 4 and 5). At 72 hours post-injection, 1.3 and 0.81 percent of the dose had been secreted in milk for the first and second cows, respectively. While the percentage per kg of milk was definitely higher for the first cow (83 mCi dose) at hour 72, the basic secretion pattern was, as expected, quite similar between animals. The percentage of plutonium transported to urine was also similar, but the fecal excretion of plutonium did reveal differences between the two treatments. It should be noted, however, that the first cow developed a problem, apparently digestive in nature, and was not consuming a normal amount of feed. Consequently, the gross amount of fecal material had decreased sharply by the third collection day (0.8 kg for Day 3).

Following multiple plutonium injections to the second animal, milk collections were fed to two calves. The dosing sequence and the respective amounts of plutonium ingested by each calf are shown in Table 6. Oral doses of 174 μ Ci and 153 μ Ci, ingested over the six-day dosing period, resulted in detectable plutonium concentrations for some calf tissues (Table 7). Several tissues collected at the calf sacrifice have been omitted from this table due to their low plutonium concentrations. Samples of muscle, gonads, thyroid, thymus, and blood were of limited value having very low nuclide concentrations. Two additional calves were used as control animals during the study to evaluate potential cross contamination. One was located inside the metabolism room, while a second control calf was housed in an outdoor pen completely removed from the experimental operation. The calf inside the metabolism room was, of course, the primary control animal and received identical handling as the experimental calves, i.e., it received milk at the same time as did the experimental animals and it was maintained in a metabolism stall between the two

Table 4. Percentage of Plutonium-238 Transported to Milk, Urine, and Feces of One Cow Over a 72-Hour Period Following a Single 83 mCi Intravenous Injection of Citrate-Buffered Plutonium Nitrate

Post-Dosing Time (h)	Percentage of Dose per Total Day's Collection			Percentage of Dose per kg of Material		
	Milk	Urine	Feces	Milk	Urine	Feces
24	0.55	1.4	0.35	0.027	0.10	0.028
48	0.52	0.31	0.42	0.033	0.034	0.12
72	0.25	0.12	0.15	0.025	0.028	0.19
TOTAL	1.3	1.8	0.92			

Table 5. Percentage of Plutonium-238 Transported to Milk, Urine, and Feces of One Cow Over a 72-Hour Period Following a Single 9.95 mCi Intravenous Injection of Citrate-Buffered Plutonium Nitrate

Post-Dosing Time (h)	Percentage of Dose per Total Day's Collection			Percentage of Dose per kg of Material		
	Milk	Urine	Feces	Milk	Urine	Feces
24	0.39	1.4	0.82	0.023	0.084	0.038
48	0.28	0.35	1.0	0.015	0.019	0.053
72	0.14	0.23	0.61	0.0078	0.0094	0.024
96	0.10	0.15	0.37	0.0055	0.0073	0.018
120	0.060	0.11	0.31	0.0033	0.0054	0.016
TOTAL	0.97	2.2	3.1			

Table 6. Dosing Sequence for the Two Phase I Calves That Ingested *In Vivo* Plutonium-238 Labeled Milk

Dosing Day	Calf No. 2637			Calf No. 2638		
	Pu Concentration (nCi/g)	Total Milk Ingested (kg)	Total Pu Ingested (nCi)	Pu Concentration (nCi/g)	Total Milk Ingested (kg)	Total Pu Ingested (nCi)
Pre-Dosing Day 1 (PM)	BKG	2.1	BKG	BKG	2.1	BKG
Pre-Dosing Day 2 (AM)	BKG	2.1	BKG	BKG	2.1	BKG
(PM)	BKG	0.6	BKG	BKG	0.6	BKG
Dosing Day 1 (AM)	6.0	2.1	12,600	6.0	2.1	12,600
(PM)	6.0	2.1	12,600	6.0	2.1	12,600
Dosing Day 2 (AM)	7.5	2.1	15,750	7.5	2.1	15,750
(PM)	11.3	2.1	23,730	11.3	2.1	23,730
Dosing Day 3 (AM)	11.3	2.1	23,730	11.3	2.1	23,730
(PM)	11.3	2.1	23,730	11.3	2.1	23,730
Dosing Day 4 (AM)	7.5	2.1	15,750	7.5	2.1	15,750
(PM)	7.5	2.1	15,750	7.5	1.2	9,000
Dosing Day 5 (AM)	4.2	2.1	8,820	4.2	1.0	4,200
(PM)	4.2	1.6	6,720	4.2	1.0	4,200
Dosing Day 6 (AM)	3.5	2.1	7,350	3.5	1.0	3,500
(PM)	3.5	2.1	7,350	3.5	1.1	3,850
Post-Dosing Day 1 (AM)	BKG	2.1	BKG	BKG	1.0	BKG
(PM)	BKG	2.1	BKG	BKG	2.1	BKG
Post-Dosing Day 2 (AM)	BKG	2.1	BKG	BKG	2.1	BKG
(PM)	BKG	2.1	BKG	BKG	2.1	BKG
Total Pu Ingested			173,880			152,640

Table 7. Percentage of Plutonium Dose Per Kg of Tissue Retained by the Phase I Calves Following Ingestion of *In Vivo* Plutonium-Labeled Milk

Calf Number	2637	2638
Breed	Holstein	Holstein
Average Daily Milk Intake (kg)	4.0	3.0
Total Pu Dose (μCi)	174.0	153.0
Percentage of Dose Per kg of Tissue/Organ:		
Bone	4.9×10^{-2}	9.2×10^{-2}
Liver	4.0×10^{-2}	3.8×10^{-2}
Lymph Nodes	2.6×10^{-2}	1.3×10^{-2}
Spleen	-	9.2×10^{-3}
Adrenals	9.2×10^{-3}	9.8×10^{-3}
Duodenum mucosa	7.5×10^{-3}	3.3×10^{-2}
Abomasum	1.7×10^{-2}	-
Omasum	3.5×10^{-2}	7.8×10^{-3}
Reticulum	7.5×10^{-3}	-
Rumen	6.3×10^{-3}	-

experimental calves. Based on the very low, essentially background, plutonium concentrations in the control animal tissues, any unintentional plutonium exposure to the calves was shown to be minimal.

Results from the Phase I calf tissue, along with results from past plutonium investigations, suggested that the variety of calf tissues collected during the definitive study (Phase II) could be reduced without compromising the validity of comparisons between the *in vivo* and *in vitro* plutonium-labeled treatments. Extrapolations to a percentage of dose retained per kg of organ are presented in Table 7. Extrapolated calculations, especially from very low concentrations, can sometimes obscure variability and, in other instances, result in significant retention figures which were unfortunately based on concentrations approaching the lower limit of detectability for direct counting. Final results from the two phases will be based on radiochemical analysis and will be significantly more valid.

Phase II

The transport of plutonium to milk, urine, and feces of the four adult cows is shown in Table 8. An average of 1.5, 2.4, and 2.2 percent of the intravenous dose was transported to milk, urine, and feces, respectively, during the first 120 hours after injection. These transport values are basically similar to those observed in Phase I.

Many samples, including all calf tissues, quality control assays on both the *in vivo* and *in vitro* plutonium-labeled milk, and some blood samples, still require final analysis. Most of these remaining samples have been screened for plutonium activity with the phoswich system, but many will also be analyzed using radiochemical techniques. A subsequent report will include the significant findings from both Phase I and Phase II.

SMALL VERTEBRATES

Table 8. Percentage of Plutonium-238 Transported to Milk, Urine, and Feces in Four Dairy Cows Following Acute Intravenous Doses of Citrate-Buffered Plutonium-238 Nitrate (Approximately 16.0 mCi per Animal)

Time (h) After Injection	Percentage of Dose Transported to MILK			
	Cow #119	Cow #179	Cow #123	Cow #128
24	0.507	0.549	0.331	0.413
48	0.610	0.516	0.268	0.341
72	0.346	0.320	0.233	0.193
96	0.235	0.228	0.141	0.141
120	0.171	0.155	0.118	0.123
Total	1.87	1.77	1.09	1.21
Time (h) After Injection	Percentage of Dose Transported to URINE			
	Cow #119	Cow #179	Cow #123	Cow #128
24	0.975	1.27	0.897	0.820
48	0.397	0.507	0.430	0.570
72	0.573	0.259	0.340	0.401
96	0.374	0.209	0.259	0.271
120	0.374	0.139	0.213	0.247
Total	2.69	2.38	2.14	2.31
Time (h) After Injection	Percentage of Dose Transported in FECES			
	Cow #119	Cow #179	Cow #123	Cow #128
24	0.134	0.467	0.156	0.330
48	0.179	1.01	0.613	0.617
72	0.458	0.870	0.371	0.357
96	0.332	0.637	0.306	0.417
120	0.307	0.455	0.309	0.494
TOTAL	1.41	3.44	1.76	2.22

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239Pu and 241Am CONTAMINATION
OF SMALL VERTEBRATES IN NAEG
STUDY AREAS OF NTS AND TTR

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ABSTRACT

Nevada Applied Ecology Group ecological studies of small vertebrates in three plutonium (Pu) contaminated study areas of Nevada Test Site began in Spring, 1972, and were expanded to include four areas of Tonopah Test Range in Fall, 1973. Much of the basic inventory and ecological data on small vertebrates have been previously reported. This progress report consists primarily of presentation and analysis of radioanalytical data on rodents and lizards from Area 11-C, Nevada Test Site. In addition, methodology and preliminary results of initial hematologic studies are presented.

Dipodomys microps is a dominant rodent species in all study areas. Concentrations of ²³⁹Pu and ²⁴¹Am in pelt, GI tract, and carcass of 74 resident *D. microps* from five study areas were determined. The only consistent trend evident was that carcass burdens were lower than pelt or GI tract burdens by a factor of 10². Mean ratios of Pu/Am in tissue aliquots were variable, and many were significantly different than ratios in soil or vegetation samples.

Rodents which were resident near GZ, Area 11-C, in high activity strata (>25,000 CPM ²⁴¹Am) had higher Pu concentrations in the three tissue components examined than rodents which resided in lower activity strata (<25,000 CPM Am) at greater distances from GZ. Whereas Pu concentrations in small vertebrates were variable and did not differ significantly between trophic categories, Pu/Am ratios in carcasses of insectivore-omnivores were significantly higher (P<.01) than mean ratios reported in soil and vegetation samples from Area 11-C. Studies on food sources and microhabitats frequented by rodents are needed before Pu and Am uptake can be properly evaluated.

Trapping data indicate that resident rodent species and most rodent populations were fewer in number in the higher Pu activity strata in Area 11-C. Therefore, collections of adequate samples of resident rodents from the vicinity of GZ will require extended collecting effort encompassing several reproductive seasons.

Preliminary results of hematologic studies of rodents in Area 11-C indicate that there is a correlation between Pu body burdens and depressions in some leukocyte counts. Regression analysis revealed a significant ($P < .01$) reduction in relative number of lymphocytes, with an increase in Pu carcass burdens of *Dipodomys microps* and *Perognathus longimembris*. Leukocyte counts were significantly lower ($P < .01$) with an increased Pu carcass burden in *D. merriami*. More data from animals with high Pu tissue burdens are needed before effects of Pu on the hemopoietic system of rodents can be properly evaluated.

INTRODUCTION

Plutonium (Pu) is an important environmental factor in certain areas of Nevada Test Site (NTS) and Tonopah Test Range (TTR). The Nevada Applied Ecology Group (NAEG) has been engaged in studies related to distribution of Pu in these areas in order to evaluate effects of Pu on native plants, animals, and ecosystems and their possible relationships to man.

Ecological studies of vertebrates in three Pu-contaminated areas of NTS began in Spring, 1972, and were expanded to include four areas of TTR in Fall, 1973. During the initial phases of these studies, emphasis was on the modification of standardized procedures for qualitative and quantitative inventories of small vertebrate populations. Additional techniques for the collection and preparation of vertebrate samples for radioanalysis were developed and improved. These methods and much of the basic inventory and ecological data for small vertebrates have been previously reported (Moor and Bradley, 1974; Bradley and Moor, 1975).

Emphasis has changed since the last formal report (Bradley and Moor, 1975) with the completion of initial species inventories of NTS and TTR. Dominant vertebrate species of various trophic levels have now been identified in NAEG Study Areas, and much is known about their numbers and seasonal population dynamics. Efforts are now being directed at evaluating uptake of Pu and Am in different trophic levels in relation to radioisotope concentrations within the general activity range of individual animals. Initial attempts to evaluate some possible irradiation effects have recently been undertaken with initiation of hematological studies of lizards and rodents resident in NAEG Intensive Study Areas.

This report consists primarily of presentation and analysis of radioanalytical data on rodents and lizards of some NAEG Study Areas of NTS and TTR. In addition, methodology and some preliminary results of initial hematological studies of rodents are presented.

MATERIALS AND METHODS

Census and Collection

Various standard census methods were employed to obtain qualitative and quantitative inventories of lizard and rodent faunas in each NAEG Study Area. These methods varied between vertebrate taxa because of differences in ease of capture and observation and have been reported previously (Moor and Bradley, 1974; Moor *et al.*, 1976a).

In general, permanent grid systems developed by NAEG in the study sites were used as locations for census and collection of small vertebrates. These grid systems identify the coordinates of Pu and Am activity strata developed from FIDLER surveys and Pu soil analysis (Gilbert *et al.*, 1975).

Utilizing capture-recapture techniques and a system of toe-clipping to enable recognition of individual animals, data on population densities, biomass, and seasonal activities were gathered. Rodents were collected in Sherman live traps and lizards were noosed. Data recorded at time of capture included the location of the capture in the grid, species identification, toe clip, sex, relative age, and reproductive condition.

The density of small mammals in the trapping grid was estimated using mark-recapture methods described by Hayne (1949). The trapping grid was also used to gather data on animal movements in order to establish home ranges based upon recapture locations in the grid. A center of activity was determined for individual animals using individual recapture locations and the relative frequency with which an animal was found at various locations in the grid (Hayne, 1949; Calhoun and Casby, 1958). Centers of activity were then examined in relation to Pu activity strata and distance from GZ. Recapture radii were averaged by sex for each species and were used to estimate home range size and effective trapping area of the grid. When sufficient data to estimate recapture radii for a particular species were not available, data from Jorgensen and Hayward (1965) collected from NTS were utilized from similar areas to estimate home-range diameters.

Density estimates were also utilized to compute a species diversity index. The Shannon formula (Shannon, 1948; Pielou, 1966; Lloyd *et al.*, 1968) was used as a general index of species diversity in each Study Area:

$$\frac{C}{N} (N \log_{10} N - \sum n_i \log_{10} n_i)$$

where C converts logarithms from base 10 to an arbitrary base, N is the total number of individuals of all species, and n_i is the number of the i th species.

In addition to estimates of home range, movements, population densities, and trophic components found in the Study Areas, data gathered were utilized for

other purposes. Aboveground activity of each species in each area was based on the relative number of individuals captured seasonally; reproductive status and recruitment were estimated by the percentage of the populations which were described as either sexually active adults, sexually inactive adults, young adults, or immatures.

Radioanalysis

Individual animals known to be residents in the Study Areas for at least three months were captured, sacrificed, and taken to the CETO building in Mercury, where they were autopsied. Animals were also collected off-site for analysis following the same procedures used in NAEG Study Areas. Tissue aliquots for radioassay included pelt or skin, GI tract, and carcass. Laboratory procedures for preparation of tissue samples have been reported (Moor and Bradley, 1974; Moor *et al.*, 1976b) and included dipping animals in hot paraffin wax to minimize cross-contamination between respective aliquots. Levels of ^{239}Pu and ^{241}Am in the tissue samples were determined by LFE Environmental Analysis Laboratories.

Hematological Studies

The following procedures were used to obtain baseline hematological data from animals collected off-site and in NAEG Intensive Study Areas.

Animals were captured alive utilizing Sherman live traps or nooses and returned to the laboratory, either CETO or UNLV, and examined within 24 hours of capture. Blood was obtained from lizards by decapitation without anesthetic and was obtained from rodents by cardiac puncture using ethyl ether as an anesthetic. In all instances, blood was collected in heparinized (ammonium sulfate) syringes. All analyses were done in duplicate, and any test exceeding a 1% difference was repeated. All analyses were done following standard methods (Hepler, 1966).

Differential Stain--Blood smears were prepared on clean slides and allowed to air dry. Slides were then fixed with Wright's stain. One hundred cells were counted and identified using the 4-field meander method, beginning at the margin of the smear and counting toward the center and then to the margin alternately.

Hemoglobin--The cyanmethemoglobin method was used for hemoglobin determination. To 5.0 ml Drabkins solution, 0.02 ml of whole blood was added. Hemoglobin is converted to cyanmethemoglobin by the Drabkins solution which contains ferricyanide and cyanide. The resultant color is indicative of hemoglobin concentration which was read on a Fisher Flo-thru Hemophotometer in gm/100 ml or Bausch and Lomb Spectronic 20.

Packed Cell Volume--The packed cell volume (PCV) is a measurement of percentage of red blood corpuscles in whole blood. Precalibrated 75 mm heparinized micro-hematocrit tubes were centrifuged in an Adam's Readacrit Centrifuge at 8500 rpm for five minutes and PCV measured on the built-in reader (1-100%).

Erythrocyte Count--Blood was drawn into an RBC pipette ($\pm 1\%$) and diluted with Haymen's dilution fluid (1:200). A Specto-Bright-line hemocytometer was used for counting erythrocytes. Numbers were estimated in millions/cm³.

Leukocyte Counts--Blood was drawn into a WBC diluting pipette ($\pm 1\%$) and diluted (1:20) with 1% glacial acetic acid. A Specto-Bright-line hemocytometer was used for counting leukocytes. Numbers were estimated in thousands/cm³.

Plasma Protein--Plasma from the packed cell volume test was utilized to estimate concentration of plasma proteins using an American Optical total solids meter No. 10400 Refractometer. Total plasma proteins were read directly on the meter (g/100 ml).

Albumin concentration was determined colormetrically using an American Monitor buffered albumin dye No. 1007. The plasma albumin binds quantitatively to the day 3, 3', 5, 5'-tetrabromo-m-cresolsulfonphthalein. The albumin dye combination produces an intense blue chromophore which was measured at 600nm on a Bausch and Lomb Spectronic 20. A standard curve was developed for each sample series, and the albumin concentration calculated in gm/100 ml.

Glucose--Glucose concentration was determined using Harleco reagent and standard set No. 64147. To 10.0 ml reagent (ortho-toluidine 6% in glacial acetic acid), 0.1 ml of plasma was added. After boiling, the resultant green color was measured photometrically on a Bausch and Lomb Spectronic 20 at 630 nm, and the concentration (mg/100 ml $\pm$ 2 mg/100 ml) was determined from a standard curve.

Plasma Cholesterol--Plasma cholesterol was determined colormetrically using Harleco standard 7653B and reagent 7653A (acetic acid, acetic anhydride, and sulfuric acid). Plasma (0.1 ml) was added to 5.0 ml of reagent and incubated for 10 minutes at 37^o C. The resultant color is proportional to the cholesterol concentration and read on a Bausch and Lomb Spectronic 20 at 625 nm.

RESULTS AND DISCUSSION

Concentrations of ²³⁹Pu and ²⁴¹Am in Small Vertebrates of NAEG Intensive Study Areas

Small vertebrates found in six NAEG Intensive Study Areas of NTS and TTR have been previously reported (Moor and Bradley, 1974; Bradley and Moor, 1975) and, in general, species composition agrees with earlier investigations of vertebrates from comparable habitats of NTS outside of the Study Areas (Hayward *et al.*, 1963; Tanner and Jorgensen, 1963; Jorgensen and Hayward, 1965).

Dipodomys microps is a dominant rodent species in the Study Areas. Table 1 presents Pu and Am concentrations in tissues of *D. microps* from five Study Areas. In Area 5, NTS, where *D. microps* and *D. merriami* are co-dominant, these two similar granivore species are pooled.

In general, *Dipodomys* had relatively low Pu tissue burdens. In addition, tissue burdens and their Pu/Am ratios exhibited high variability, especially in carcasses, relative to values reported in both soil and vegetation samples. In each Study Area, the mean of at least one *Dipodomys* tissue ratio, pelt, GI tract, or carcass, was significantly different ($P < .01$) from one or both mean soil or vegetation ratios. These differences are less pronounced in Area 11-C.

In respect to Pu and Am tissue burdens, the only consistent trend evident was that carcass burdens were much lower than pelt or GI tract burdens; otherwise, either pelt or GI tract had the highest concentrations depending on the Study Area examined. The highest tissue burdens for *D. microps* were from Clean Slate 2, TTR. We believe this is due to biased representation of samples from near GZ. Data on the relationships between centers of animal activity, Pu activity strata, and Pu tissue burdens are discussed later in this report.

Differences between levels of Pu and Am and their ratios in tissues is expected between samples from distinct event sites, principally because of dissimilar concentrations of original source materials used in the respective events. The high variability in *Dipodomys* tissue samples and the differences between Pu/Am ratios in animal, vegetation, and soil samples from the same Study Area are more difficult to explain but may be attributable to several factors. It has been well established that Pu and Am are not distributed homogeneously in animal tissues upon uptake by inhalation or ingestion (Wick, 1967; and others). The amount of these transuranic elements taken up by a particular organ varies due to interaction of chemical and physiological processes and upon the chemical form of the radioisotope. Thus, rates of uptake and elimination of Pu and Am may be related to the species of animal investigated, its age, general physiological condition, and the mode of radioisotope uptake (Wick, 1967; Jee, 1972; Lindenbaum and Rosenthal, 1972). In addition, it has been suggested that plants may preferentially uptake Am over Pu (Romney *et al.*, 1975).

Hence, variations in Pu and Am concentrations and their ratios in animal tissues from the same event site may be due to physiological differences between species or even individuals, age during radioisotope uptake, mode and chemical form of radioisotope uptake, and differences in food items ingested. In addition, animals are mobile to varying degrees; hence, they are exposed to a more varied transuranic environment than individual plants.

Additional sources of variability may be related to laboratory techniques. Methods of preparation of samples for radioanalysis vary between soil, vegetation, and animals. Counting errors and tracer yields are less consistent in tissue samples than soil samples (Major, Pers. comm.). This source of variability may be due to interfering reactions or to the process of homogenizing specimens and obtaining subsamples of homogenate. Difficulties in obtaining subsamples of homogenates from animal samples, where elements may be concentrated in particular tissues, have been reported in the literature (Sturges *et al.*, 1974).

Table 1.

^{239}Pu and ^{241}Am in Dipodomys microps from Five NAEG
Study Areas of NTS and TTR (values in nCi/g ash)

NAEG Study Area	N	^{239}Pu $\bar{X} \pm \text{SE}$	N	^{241}Am $\bar{X} \pm \text{SE}$	N	Pu/Am $\bar{X} \pm \text{SE}$	Soil Pu/Am* $\bar{X} \pm \text{SE}$	Veg. Pu/Am* $\bar{X} \pm \text{SE}$
AREA 5**							10.2 $\pm$ 0.24	12.5 $\pm$ 0.25
Pelt	16	0.096 $\pm$ 0.040	16	0.015 $\pm$ 0.009	3	5.29 $\pm$ 0.57		
GI Tract	16	0.029 $\pm$ 0.012	16	0.004 $\pm$ 0.002	9	6.16 $\pm$ 0.64		
Carcass	16	0.0003 $\pm$ 0.0001	16	0.0001 $\pm$ 0.0001	10	6.39 $\pm$ 2.03		
AREA 11-C							6.0 $\pm$ 0.08	5.2 $\pm$ 0.10
Pelt	23	1.930 $\pm$ 0.850	23	0.546 $\pm$ 0.320	21	5.95 $\pm$ 0.56		
GI Tract	23	0.742 $\pm$ 0.380	23	0.117 $\pm$ 0.060	23	6.45 $\pm$ 0.40		
Carcass	23	0.0040 $\pm$ 0.0010	21	0.0010 $\pm$ 0.0002	19	6.14 $\pm$ 0.79		
AREA 13							9.4 $\pm$ 0.15	7.9 $\pm$ 0.20
Pelt	13	0.772 $\pm$ 0.220	13	0.077 $\pm$ 0.030	10	8.99 $\pm$ 1.23		
GI Tract	13	0.498 $\pm$ 0.360	13	0.065 $\pm$ 0.040	12	5.87 $\pm$ 0.48		
Carcass	12	0.0040 $\pm$ 0.0020	13	0.0002 $\pm$ 0.0003	1	6.70		
CLEAN SLATE 2							22.2 $\pm$ 0.41	11.6 $\pm$ 0.64
Pelt	15	0.236 $\pm$ 0.042	15	0.008 $\pm$ 0.002	11	27.17 $\pm$ 1.23		
GI Tract	15	6.976 $\pm$ 3.601	15	0.325 $\pm$ 0.174	14	21.50 $\pm$ 1.30		
Carcass	15	0.0116 $\pm$ 0.0039	15	0.0004 $\pm$ 0.0002	13	26.85 $\pm$ 4.33		
DOUBLE TRACK							23.5 $\pm$ 0.73	15.8 $\pm$ 1.40
Pelt	7	0.146 $\pm$ 0.110	7	0.014 $\pm$ 0.104	5	13.41 $\pm$ 4.79		
GI Tract	7	0.274 $\pm$ 0.160	7	0.0290 $\pm$ 0.023	4	13.92 $\pm$ 4.32		
Carcass	7	0.0013 $\pm$ 0.0008	7	0.0002 $\pm$ 0.0001	5	7.29 $\pm$ 3.31		

*Soil and Vegetation Data from Romney et al., 1975.

**Means represent pooled samples of D. merriami and D. microps.

Table 2 presents data on Pu and Am in small vertebrates from one Study Area, Area 11-C, NTS. Considerable variation is evident in Pu and Am levels in the three tissue components of each taxon. *Perognathus longimembris*, a granivore, and lizards, which are generally insectivorous, had relatively high mean Pu tissue burdens of 0.37 and 0.17 nCi/g ash, respectively, in the carcasses. These values are two orders of magnitude higher than the mean values in the granivore, *D. microps*, and the insectivore, *O. torridus*. Mean concentrations in the carcass of *A. leucurus*, an omnivore, are intermediate between the examples of high and low values given above. Highest Pu tissue burdens for any individual rodent (*P. longimembris*) were 132, 28, and 2.1 nCi/g ash for pelt, GI tract, and carcass, respectively.

Considerable variation is also evident in Pu/Am ratios of two tissue components, pelt and carcass (Table 2). The GI tract is the only tissue component which had mean ratios which varied little between species representing several trophic levels and which had comparable ratios with those reported in soil and vegetation samples (Table 1).

A more consistent pattern of ratios was expected at least between pelt and GI tract of the same species from the same event site. Both tissues are exposed to resuspendable contamination with Pu/Am ratios which should be consistent with those reported for soil and vegetation samples. The rodents examined all engage in dust bathing and utilize underground burrow systems for shelter. In addition, much locomotor activity of lizards exposes the ventrum to surface soil contamination, and both nocturnal and seasonal inactivity (hibernation) usually involves withdrawal to underground burrow systems. These burrow systems may provide significant soil contamination for both rodents, lizards, and major food items. Ingestion of foods provides additional sources of contamination from both vegetation and soil. Ratios of Pu/Am in food items such as plant parts and arthropods are unknown. Contamination, however, from soil is important for both food items, particularly insects and seeds, and in the form of soil particles which are commonly ingested with food by lizards.

The variability in pelt or skin ratios of rodents and lizards is difficult to explain. For example, the mean Pu/Am ratio of the pelt of *P. longimembris* was significantly different ($P < .01$) than mean ratios found in other rodent pelts, soil, or vegetation (Tables 1 and 2). Differences in tissue ratios may indicate that some vertebrate species and individuals of the same species are exposed to varied physical-chemical forms of the radioisotopes and that these forms have not been characterized as to their concentrations or distributions within microhabitats frequented by smaller vertebrates. In addition, some variation in Pu/Am ratios found in small vertebrates may be due to morphological, physiological, and behavioral differences between species. For example, there is a wide range of body size in rodent species, from *P. longimembris* which weighs about 7 g to *A. leucurus* which weighs approximately 100 g. Body size in rodents is related to size of home range (McNab, 1963) and probably the range of microhabitats utilized by species.

Table 3 is an initial attempt to group species in Area 11-C by similar ecological and spatial patterns. Animals are grouped by general trophic categories and activity strata from which they were collected. The activity strata of Area 11-C, as reported by Gilbert *et al.* (1975), were grouped into Low Activity Stratum ($< 25,000$ CPM ^{241}Am) and High Activity Stratum ($> 25,000$ CPM ^{241}Am).

Table 2.

^{239}Pu and ^{241}Am in Small Vertebrates from Area 11-C, NTS (values in nCi/g ash)

TAXA	N	^{239}Pu $\bar{X} \pm \text{SE}$	^{241}Am $\bar{X} \pm \text{SE}$	Pu/Am $\bar{X} \pm \text{SE}$
<u>Dipodomys merriami</u>	7			
Pelt		10.91 $\pm$ 8.36	1.71 $\pm$ 1.55	10.3 $\pm$ 4.3
GI Tract		3.41 $\pm$ 1.66	0.48 $\pm$ 0.22	6.5 $\pm$ 0.3
Carcass		0.013 $\pm$ 0.005	0.0031 $\pm$ 0.0018	8.2 $\pm$ 2.0
<u>Dipodomys microps</u>	23			
Pelt		1.93 $\pm$ 0.85	0.55 $\pm$ 0.32	6.0 $\pm$ 0.6
GI Tract		0.74 $\pm$ 0.38	0.12 $\pm$ 0.60	6.5 $\pm$ 0.4
Carcass		0.004 $\pm$ 0.001	0.0010 $\pm$ 0.0002	6.1 $\pm$ 0.8
<u>Perognathus longimembris</u>	11			
Pelt		23.31 $\pm$ 13.14	9.08 $\pm$ 5.24	2.8 $\pm$ 0.2
GI Tract		1.51 $\pm$ 0.90	0.23 $\pm$ 0.14	5.7 $\pm$ 0.4
Carcass		0.367 $\pm$ 0.193	0.0560 $\pm$ 0.0290	5.3 $\pm$ 0.5
<u>Ammospermophilus leucurus</u>	6			
Pelt		5.15 $\pm$ 3.24	1.32 $\pm$ 0.86	7.1 $\pm$ 3.0
GI Tract		5.54 $\pm$ 4.47	0.16 $\pm$ 0.10	7.3 $\pm$ 0.4
Carcass		0.040 $\pm$ 0.030	0.0050 $\pm$ 0.0050	7.5 $\pm$ 0.9
<u>Onychomys torridus</u>	6			
Pelt		0.95 $\pm$ 0.39	0.18 $\pm$ 0.11	5.9 $\pm$ 0.5
GI Tract		0.31 $\pm$ 0.24	0.06 $\pm$ 0.05	6.2 $\pm$ 1.0
Carcass		0.006 $\pm$ 0.002	0.0006 $\pm$ 0.0003	16.6 $\pm$ 7.1
Lizards	9			
Skin		7.41 $\pm$ 3.34	0.91 $\pm$ 0.48	8.9 $\pm$ 2.0
GI Tract		7.79 $\pm$ 4.49	1.11 $\pm$ 0.64	6.8 $\pm$ 0.2
Carcass		0.170 $\pm$ 0.050	0.0224 $\pm$ 0.0075	11.4 $\pm$ 3.6

Table 3.

^{239}Pu and Pu/Am Ratios in Granivores and Insectivore-omnivores
from Area 11-C, NTS (values in nCi/g ash)

Trophic Category	N	Low Activity Stratum*			N	High Activity Stratum**		
		^{239}Pu $\bar{X} \pm \text{SE}$	N	Pu/Am $\bar{X} \pm \text{SE}$		^{239}Pu $\bar{X} \pm \text{SE}$	N	Pu/Am $\bar{X} \pm \text{SE}$
GRANIVORE								
Pelt	26	0.714 ± 0.267	20	5.90 ± 0.59	16	22.792 ± 9.320	16	5.92 ± 1.78
GI Tract	26	0.155 ± 0.039	26	6.07 ± 0.39	16	3.344 ± 0.950	16	6.59 ± 0.15
Carcass	26	0.008 ± 0.005	20	5.58 ± 0.84	16	0.263 ± 0.137	16	7.17 ± 0.67
INSECTIVORE-OMNIVORE								
Pelt/Skin	11	1.316 ± 0.499	8	7.18 ± 1.75	11	8.911 ± 2.948	10	7.82 ± 1.93
GI Tract	12	1.159 ± 0.530	8	6.75 ± 0.34	11	9.891 ± 4.217	9	6.67 ± 0.26
Carcass	12	0.022 ± 0.012	9	10.84 ± 3.12	11	0.148 ± 0.302	11	12.17 ± 3.67
COMBINED VERTEBRATES								
Pelt/Skin	37	0.893 ± 0.240	28	6.27 ± 0.64	27	17.136 ± 5.732	26	6.66 ± 1.31
GI Tract	38	0.472 ± 0.182	34	6.23 ± 0.31	27	6.011 ± 1.869	25	6.59 ± 0.13
Carcass	38	0.013 ± 0.005	29	7.21 ± 1.18	21	0.216 ± 0.083	21	9.20 ± 1.58
SOIL***			23	6.2 ± 0.39			24	6.0 ± 0.30
VEGETATION***	26	2.0 ± 0.40	26	5.3 ± 0.20	22	8.3 ± 0.30	22	5.0 ± 0.25

* $<25,000$ CPM ^{241}Am based on Fidler surveys by Reeco personnel.

** $>25,000$ CPM ^{241}Am based on Fidler surveys by Reeco personnel.

***Data from Romney et al., 1975.

In Area 11-C, granivores (*D. merriami*, *D. microps*, and *P. longimembris*) did not differ significantly ($P > .05$) from insectivore-omnivores (*A. leucurus*, *O. torridus*, and lizards) in mean Pu tissue burdens from either Activity Stratum. Mean Pu tissue burdens of both trophic categories, however, were significantly greater ($P < .01$) from the High Activity Stratum where Pu carcass burdens were approximately 2×10^{-1} nCi/g ash.

Mean Pu/Am ratios of both pelt/skin and carcasses were more variable in insectivore-omnivores than in granivores from both strata. As shown in Table 3, however, ratios in GI tracts of both trophic categories were consistent with soil and vegetation ratios and exhibited relatively low variability. In contrast, mean ratios in carcasses of insectivore-omnivores were not only more variable, they were significantly higher ($P < .01$) than either means of the GI tract samples or soil and vegetation ratios. This phenomenon is difficult to explain in light of the consistent ratios found in GI tracts of rodents. In addition, Romney *et al.* (1975) present data which indicate relatively consistent values for Pu/Am in soil and vegetation samples and suggest consistent Pu/Am ratios in resuspendable contamination in Area 11-C. The high ratios present in carcass samples of insectivore-omnivores may indicate that these rodents are exposed to different environmental conditions than plants which have been sampled and occupy microhabitats which have not been sampled by current soil sampling design. In addition, insectivore-omnivores have larger home ranges than granivores (McNab, 1963; our studies) and, hence, may be exposed to a wider variety of microhabitats. A supplemental hypothesis may be that some food sources of insectivore-omnivores, such as arthropods and possibly small vertebrates, contain Pu and Am which is more or less soluble, hence, more or less available for uptake, than that of resuspended contaminants.

It is well known that plants and animals play a significant role in development and dynamics of soil ecosystems. Plants aid in chemical and mechanical weathering of rock and soil. Plant roots help break up large soil aggregates and rocks and provide avenues of access of air and water to subsurface soil. Fungi and bacteria excrete metabolic acids which can dissolve rock aggregates, and possibly can solubilize essentially insoluble soil components, such as plutonium. When plants die, various soil invertebrates, fungi, and bacteria convert plant tissue to available food for microinvertebrates, and the resulting processes add nutrients to the soil. Whereas desert soils are relatively sterile, the available nutrients are not uniformly distributed. We believe that nutrient enrichment associated with burrow systems may be a major link in nutrient cycling of desert ecosystems.

Animal activity, particularly burrowing activity, has been shown to be a principal force affecting soil chemistry and structure. Taylor (1935) has reported that ground squirrels move 30 to 40 tons of soil per acre yearly in arid midwest areas. Thorp (1949) estimated that burrow mounds consisted of 7-9 Kg of subsoil for each square meter of surface in semiarid portions of the United States. Burrowing activity has also been shown to be extensive as far as depth is concerned. For example, some ants at Rock Valley, NTS, have been found at depths of 10 meters. Kangaroo rats (*Dipodomys*), the most abundant rodents of NAEG Study Areas, have been shown to significantly alter soil chemistry in their burrow systems. Green and Reynard (1932) reported that kangaroo rats defecated extensively throughout their burrow systems. They

reported mean soluble nitrate content in regions of two burrows at 221 to 570 ppm; whereas, a maximum of 15 ppm was found in surrounding desert soils. It is also well established that burrows are much higher in oxygen and water content than typical desert soils and temperatures in burrows are more stable and more favorable for living systems than those found in open desert (Bradley and Yousef, 1972).

The animal burrow, then, is a different environment than that found in the surrounding desert. Soluble nutrients, gases, humidity, and temperatures are at levels which are more conducive for maintenance and growth of organisms. Burrowing activity may significantly influence distribution and resuspension of soil components, including probably Pu and Am.

It is important to realize that time spent in burrows by small vertebrates and many invertebrates is significant and, in many cases, is greater than that spent in aboveground activity. Nonhibernating rodents spend approximately 85% of their active lifetime in burrows. For hibernating species (*P. longimembris*, for example), burrow occupancy increases to approximately 90% of an average year (Bradley, unpublished data). We estimate that lizards may utilize hibernals for approximately 30-40% of an average year and inhabit nocturnal retreats throughout their activity season. Food stored in animal burrows provides an ideal substrate for the growth of fungi and bacteria and also a convenient food source for many invertebrates. Au and Beckert (1975) have reported that *Aspergillus niger*, an ubiquitous soil fungus, can take up several chemical forms of Pu, including plutonium dioxide, from a cultural medium and have reported on microbial numbers and relative abundance of fungi in surface and subsurface soils of NTS. From analyses of stomach samples, Thomas (1975) reported that darkling beetles (Tenebrionidae) consume large quantities of fungi, particularly *Aspergillus*. In a preliminary effort to estimate tissue burdens in some possible food sources of small vertebrates, darkling beetles were collected from GZ of Area 11-C, washed with alcohol to attempt to remove surface contamination, and gamma scanned by REECO personnel. Plutonium tissue burdens were estimated at 0.5 nCi/g ash.

Burrow microenvironments may differ radiochemically from the soil surface and may provide significantly different sources of contamination by inhalation and ingestion. Therefore, Pu and Am concentrations in burrows and hibernals may show higher correlations with tissue concentrations than activity strata developed from surface soil samples. Laboratory error may account for some of the variability in samples, particularly the homogenate problem already discussed. It is obvious, however, that before more conclusive statements can be made, more data on radiochemical and biological inventories of microhabitats, such as burrows, and food sources, of some vertebrates have to be available for evaluation. These data may be critical in order to properly evaluate Pu and Am uptake by inhalation and ingestion. Radioisotopes in these sources of contamination may differ in relative concentrations, physical state, and solubility from those of soil samples analyzed to date.

Resident Rodent Populations of Area 11-C

One particularly difficult problem in evaluating tissue burdens of small vertebrates occupying different trophic levels and inhabiting defined activity

strata is the apparent reduced resident rodent populations in the vicinity of GZ. Figure 1 presents a diagram of the inner trapping grid in Area 11-C in which isopleths mark the approximate boundaries of activity strata defined by Gilbert *et al.* (1975). Trapping stations are 50 feet apart. The inner grid with a width of 350 feet and a length of 1200 feet was trapped most frequently. The area encompassed by the High Activity Stratum ($>25,000$ CPM ^{241}Am) represents approximately 24% of the inner grid and is indicated by a solid-lined isopleth.

Table 4 presents data on total number of captures and number of resident rodents encountered in the High and Low Activity strata of the inner grid during 18 months of study in Area 11-C. If we assume random distribution of animals in the grid, an assumption which is probably met only under ideal conditions, an estimate of the number of animals captured in the High Activity Stratum can be made by multiplying the total number of captures (447) by the proportion of the area encompassed by the High Activity Stratum (23.5%). One hundred and five captures are expected using these calculations, which is not significantly different than the number actually captured in this stratum (103). All species shown in Table 4 were captured in both High and Low Activity strata. The number of resident animals expected in the High Activity Stratum can be estimated in the same manner. There were 82 resident animals in the grid, of which 19 were expected from the High Activity Stratum. Only nine resident animals, however, were recorded from this stratum, a significantly ($P<.01$) lower number than expected. Of the six resident species, only four were found in the High Activity Stratum. In addition, of these resident animals, only one species (*D. merriami*) was represented by more than two individuals. These data show a reduced species richness, species diversity (H'), and population density of resident rodents in the High Activity Stratum near GZ and illustrate the problems of collecting adequate samples of most species of rodents.

We do not now have an explanation for the reduced number of residents in the High Activity Stratum. Beatley (1976) reported that *D. merriami* replaced *D. microps* as a dominant rodent in some disturbed areas of NTS. Whereas the low number of *D. microps* may be explained by habitat alteration around GZ, it does not explain the absence of resident *A. leucurus* or *O. torridus*. Whereas a reduced number of some species of rodents may be expected when native habitat is altered, experience by the investigators in the Mohave desert suggests that many rodents, particularly *A. leucurus*, are likely to be more numerous in disturbed habitats with sufficient vegetative cover. We do not believe, therefore, that the depauperate rodent fauna of Area 11-C, GZ, can be explained satisfactorily strictly on the basis of mechanical disturbance, as the area is not denuded of vegetation. We are not suggesting at this time that this phenomenon is related directly to Pu or Am levels; however, it is a hypothesis worthy of further investigation.

Hematologic Studies

Tables 5 and 6 present comparative hematologic values of three species of heteromyid rodents from Area 11-C, NTS, and a control site in the Charleston Mountains of southern Nevada. Whereas substantial variation is seen in Table 5 for most of the hematologic characteristics due partially to small sample size, the two populations, in general, compare favorably with three notable

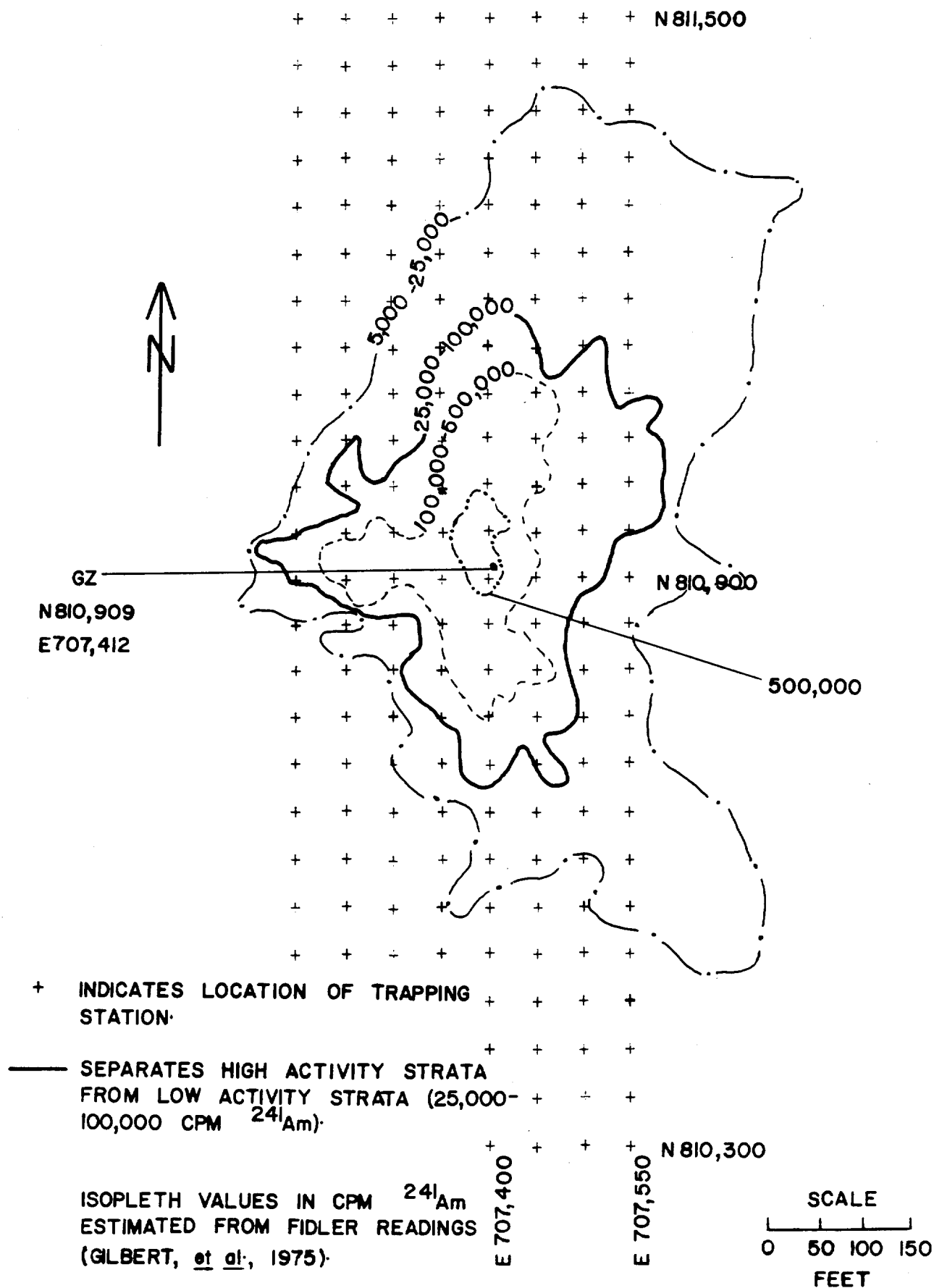


Figure 1. Location of small vertebrate trapping grid in relation to Pu activity strata in Area 11-C, NTS.

Table 4.
A Comparison of Trapping Success and Residency of Rodents
in Relation to Activity Strata of Area 11-C, NTS

Species	Number of Captures			Number of Residents		
	Low Activity Stratum	High Activity* Stratum		Low Activity Stratum	High Activity* Stratum	
		Expected	Observed		Expected	Observed
<u>Ammospermophilus leucurus</u>	51	14.1	9	14	3.3	0
<u>Dipodomys merriami</u>	60	21.2	30	6	2.6	5
<u>Dipodomys microps</u>	107	29.1	17	25	6.3	2
<u>Perognathus longimembris</u>	60	18.8	20	12	3.1	1
<u>Neotoma lepida</u>	2	0.9	2	0	0	0
<u>Onychomys torridus</u>	34	10.1	9	12	2.8	0
<u>Peromyscus</u> spp.	30	10.8	16	4	1.2	1
Totals	344	105	103	73	19.3	9**
Number of Species	7	7	7	6	6	4
Species Diversity (H')				2.37	2.43	1.66

*Represents 23.5% of trapping grid (>25,000 CPM ^{241}Am).

**Significantly different than expected ($P < .01$); chi square.

Table 5.

Mean Hematologic Values of Three Species of Heteromyid Rodents from Area 11-C, NTS and an Offsite (Charleston Mountains) Control Area (Mean $\pm$ 1 Standard Error and Range)

Species	Site	N	Body Weight (g)	PCV (%)	Hb (g/100ml)	Total Protein (g/100ml)	Glucose (mg/100ml)	CHOL (mg/100ml)	Albumin (g/100ml)	RBC ($10^6/\text{cm}^3$)	WBC ($10^3/\text{cm}^3$)
<u>D. mer.</u>	Control	21	37.0 $\pm$ 0.7 28-42	49.6 $\pm$ 0.9 45-55	15.9 $\pm$ 0.6 13.9-17.3	6.1 $\pm$ 0.3 4.4- 6.9	177.8 $\pm$ 4.3 146-204	191.2 $\pm$ 2.4 178-200	1.8 $\pm$ 0.1 1.4-2.6	10.3 $\pm$ 1.1 7.4-14.4	7.4 $\pm$ 0.8 4.9-10.4
	NTS	7	36.6 $\pm$ 0.9 35-41	43.5 $\pm$ 0.4* 40-51	15.1 $\pm$ 0.9 11.8-18.0	5.4 $\pm$ 0.3 4.6- 6.2	174.6 $\pm$ 2.1 144-207	184.4 $\pm$ 1.3 166-215	1.9 $\pm$ 0.2 1.4-2.4	7.2 $\pm$ 0.7* 4.6- 9.2	6.5 $\pm$ 1.3 2.9-11.2
<u>D. mic.</u>	Control	20	59.1 $\pm$ 1.9 43-73	46.7 $\pm$ 2.2 40-55	15.6 $\pm$ 1.4 12.0-17.2	6.2 $\pm$ 0.9 4.5- 7.4	181.3 $\pm$ 3.0 160-193	185.1 $\pm$ 2.8 165-200	1.9 $\pm$ 0.4 1.1-3.0	8.2 $\pm$ 0.8 6.7-10.6	9.2 $\pm$ 1.1 5.0-16.0
	NTS	10	60.4 $\pm$ 3.4 43-71	42.9 $\pm$ 1.7 36-53	14.8 $\pm$ 0.7 12.4-17.8	5.4 $\pm$ 0.4 3.9- 7.8	184.4 $\pm$ 1.4 165-200	176.2 $\pm$ 1.6 140-197	2.1 $\pm$ 0.3 1.2-2.5	5.8 $\pm$ 0.8** 3.8-11.4	6.5 $\pm$ 0.6** 3.7- 9.6
<u>P. lon.</u>	Control	6	7.8 $\pm$ 0.4 6- 9	48.3 $\pm$ 1.5 44-54	16.4 $\pm$ 0.6 14.4-18.2	5.4 $\pm$ 0.4 4.9-10.4	I	I	I	10.3 $\pm$ 1.1 7.4-14.4	7.4 $\pm$ 0.8 4.9-10.4
	NTS	12	7.7 $\pm$ 0.3 6- 9	45.8 $\pm$ 1.6 33-52	15.9 $\pm$ 0.7 11.2-18.1	5.1 $\pm$ 0.2 4.4- 6.1	142.1 P	166.0 P	2.2 P	9.2 $\pm$ 0.8 6.4-12.4	7.1 $\pm$ 0.7 3.5-10.4

D. mer. = Dipodomys merriami; D. mic. = Dipodomys microps; P. lon. = Perognathus longimembris.

PCV = Packed cell volume; Hb = hemoglobin; CHOL = cholesterol; RBC = erthrocyte; WBC = leucocyte.

P = Represents one pooled sample.

* = Significantly different from control animals, $P < .01$ (Student's T-test).

** = Significantly different from control animals, $P < .05$ (Student's T-test).

I = Results incomplete.

Table 6.

Relative Leukocyte Differential Counts of Three Species of Heteromyid
Rodents from Area 11-C, NTS and an Offsite (Charleston Mountains)
Control Area (Mean $\pm$ 1 Standard Error and Range)

Species	Site	N	Neutrophils (%)	Eosinophils (%)	Basophils (%)	Lymphocytes (%)	Monocytes (%)
<u>Dipodomys</u> <u>merriami</u>	Control	14	26.6 $\pm$ 0.6 10.0 - 35.0	2.3 $\pm$ 0.4 0 - 5.0	0.4 $\pm$ 0.2 0 - 3.0	69.9 $\pm$ 1.9 44.0 - 83.0	3.1 $\pm$ 0.3 0 - 15.0
	NTS	7	25.3 $\pm$ 2.9 14.0 - 31.0	3.0 $\pm$ 0.9 0 - 7.0	0.7 $\pm$ 0.3 0 - 2.0	69.4 $\pm$ 3.9 59.0 - 79.0	3.0 $\pm$ 0.6 1.0 - 6.0
<u>Dipodomys</u> <u>microps</u>	Control	12	29.3 $\pm$ 0.9 12.0 - 44.0	2.6 $\pm$ 0.4 0 - 7.0	0.5 $\pm$ 0.4 0 - 3.0	71.1 $\pm$ 1.2 58.0 - 86.0	3.8 $\pm$ 0.5 0 - 8.0
	NTS	10	25.5 $\pm$ 1.5* 20.0 - 35.0	2.4 $\pm$ 0.6 0 - 7.0	1.0 $\pm$ 0.3 0 - 3.0	67.5 $\pm$ 1.9 54.0 - 74.0	3.5 $\pm$ 0.6 0 - 3.0
<u>Perognathus</u> <u>longimembris</u>	Control	6	32.3 $\pm$ 3.4 22.0 - 44.0	2.3 $\pm$ 0.5 1.0 - 4.0	0.7 $\pm$ 0.3 0 - 5.0	61.3 $\pm$ 3.5 49.0 - 71.0	3.3 $\pm$ 0.9 1.0 - 6.0
	NTS	12	31.0 $\pm$ 2.9 22.0 - 50.0	2.3 $\pm$ 0.5 0 - 4.0	1.1 $\pm$ 0.6 0 - 3.0	61.4 $\pm$ 3.5 42.0 - 76.0	4.1 $\pm$ 0.6 0 - 7.0

*Significantly different from control animals, $P < .05$ (Student's T-test).

exceptions. Mean leukocyte and erythrocyte counts of *D. microps* from NTS are significantly ($P < .05$) depressed when compared to control animals. In addition, packed cell volumes were significantly ($P < .01$) reduced in *D. merriami* from NTS. Mean relative leukocyte differential counts (Table 6) are comparable for the two populations with one exception. Neutrophils are significantly depressed ($P < .01$) in *D. microps* from NTS.

There have been many laboratory studies on the effects of Pu on the hemopoietic system of mammals. These studies, summarized in Wick (1967), have pointed out that Pu causes destruction of bone marrow, anemia, and leukopenia. Depressed levels of lymphocytes were reported by Paglia (1968) in a natural population of *D. microps* from a Pu-contaminated area of NTS.

To determine if Pu body burdens were related to blood cell reductions in rodents of Area 11, NTS, correlation coefficients were determined for blood cell counts and Pu body burdens. Significant depressions in some leukocyte counts were correlated with elevated Pu carcass burdens.

In Fig. 2, relative lymphocyte counts were plotted against Pu carcass burdens of *D. microps* and a least squares fit for the power curve plotted. Relative lymphocyte counts were negatively correlated ($P < .01$) to Pu carcass burdens. In Fig. 3, in a similar manner, lymphocytes were negatively correlated ($P < .01$) with Pu carcass burdens in *P. longimembris*. In Fig. 4, leukocyte counts were negatively correlated ($P < .01$) with Pu carcass burdens in *D. merriami*.

Although these results are preliminary, and based on small sample sizes, levels of Pu in carcasses of some rodent species in Area 11, NTS, appear to be related to depressed leukocyte counts on a statistical basis. There is no evidence at this time that these depressions are of a deleterious nature. Additional data from examination of animals with high Pu body burdens is needed. We plan to evaluate blood cell formation in the bone marrow of these animals. By comparing blood cell counts in bone marrow with peripheral blood cell counts, one can determine if depressed peripheral counts are caused by a block in the synthesis of certain cells or a reduction in the number of circulating cells.

FUTURE PLANS

In addition to proposed initial investigation of nuclear event sites and surveillance of safety shot areas, we recommend continued and concentrated study in one safety shot area (Area 11) with the following specific objectives:

1. Census, collection, and radioanalysis of small vertebrates with emphasis on the higher activity strata.
2. Study and analysis of trophic relationships including analysis of Pu and Am in major food sources of trophic categories.

Figure 2. Relative lymphocyte counts in relation to ^{239}Pu carcass burdens in *D. microps* from Area 11-C, NTS.

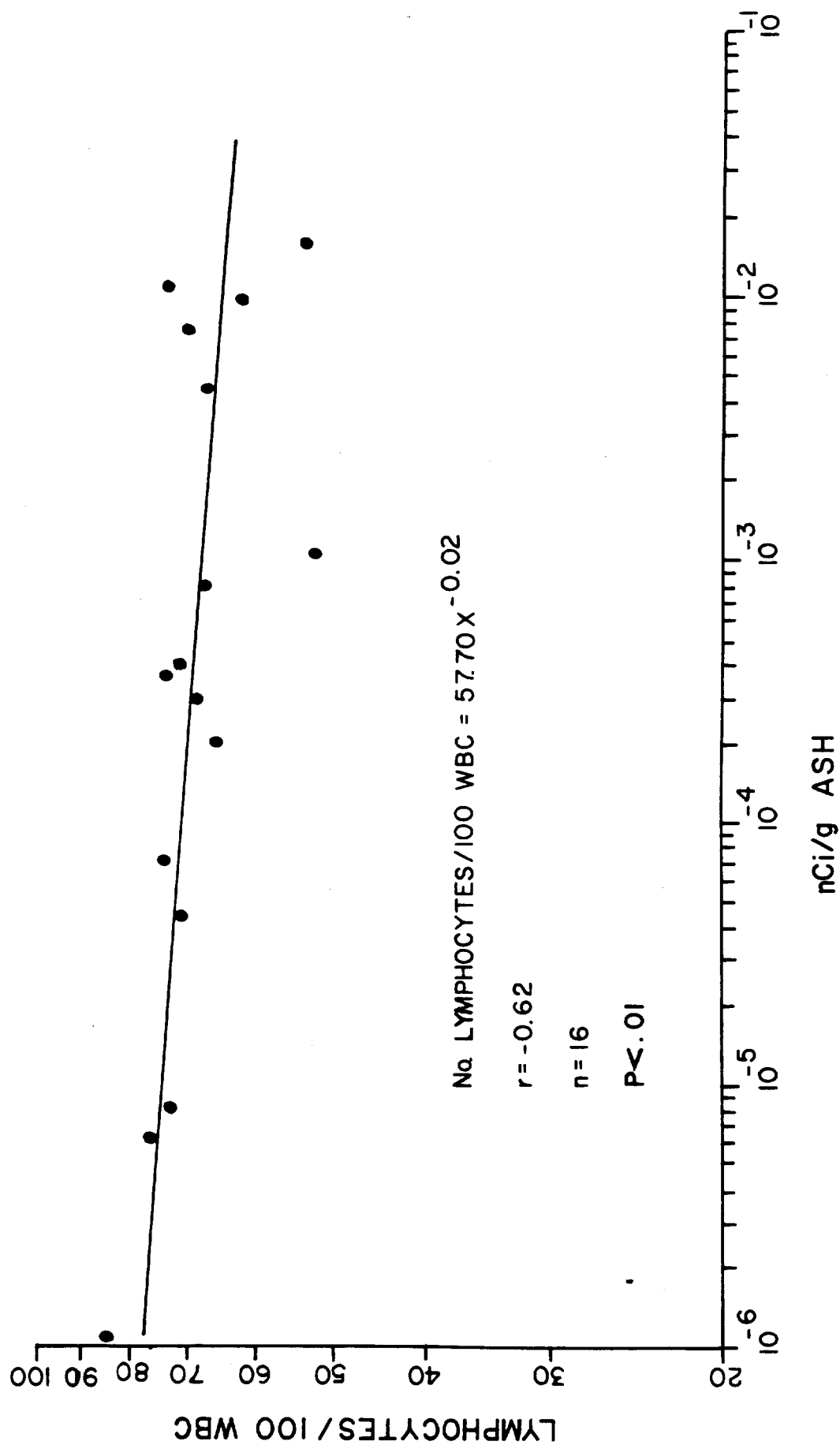


Figure 3. Relative lymphocyte counts in relation to ^{239}Pu carcass burdens in *P. longimembris* from Area 11-C, NTS.

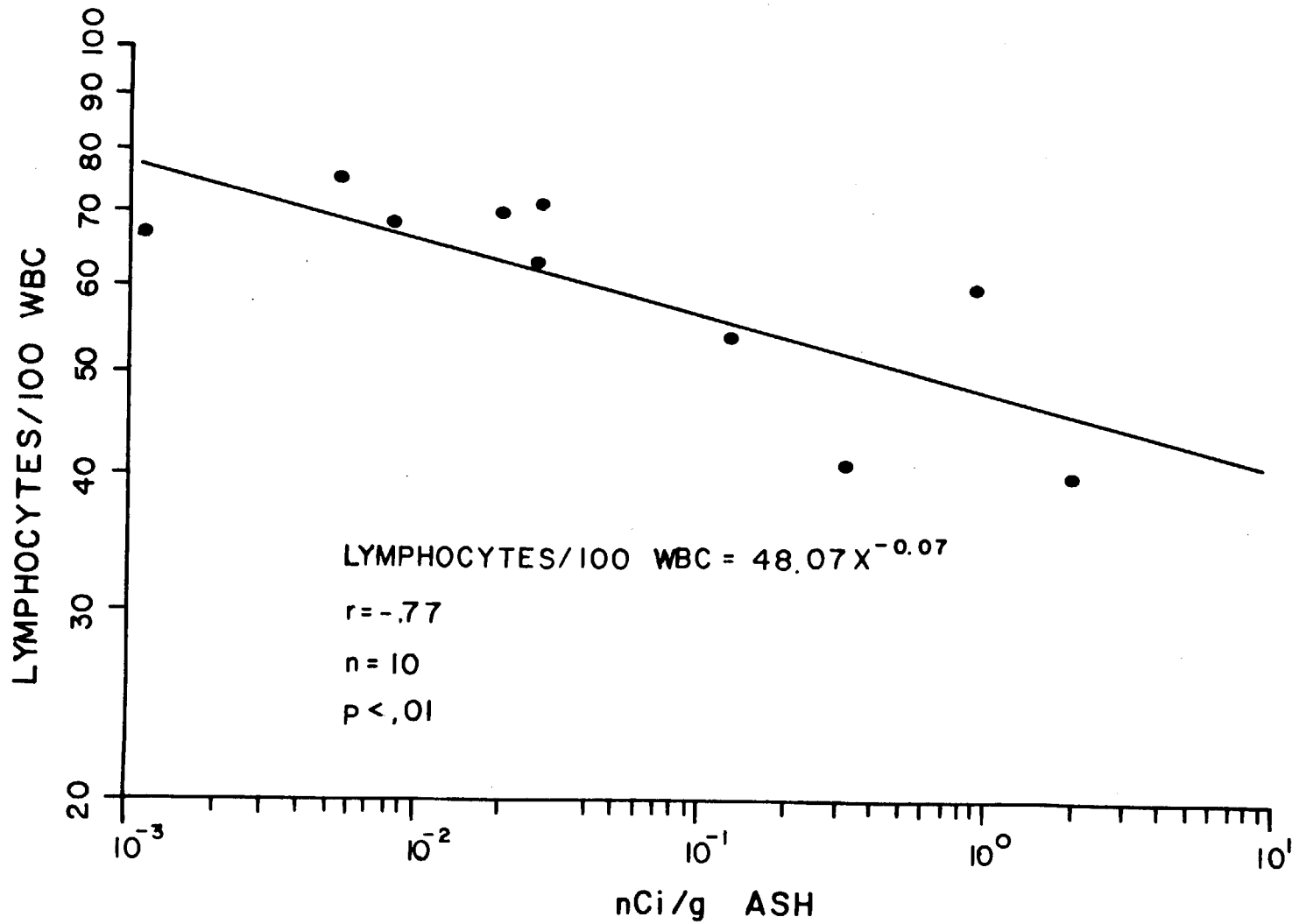
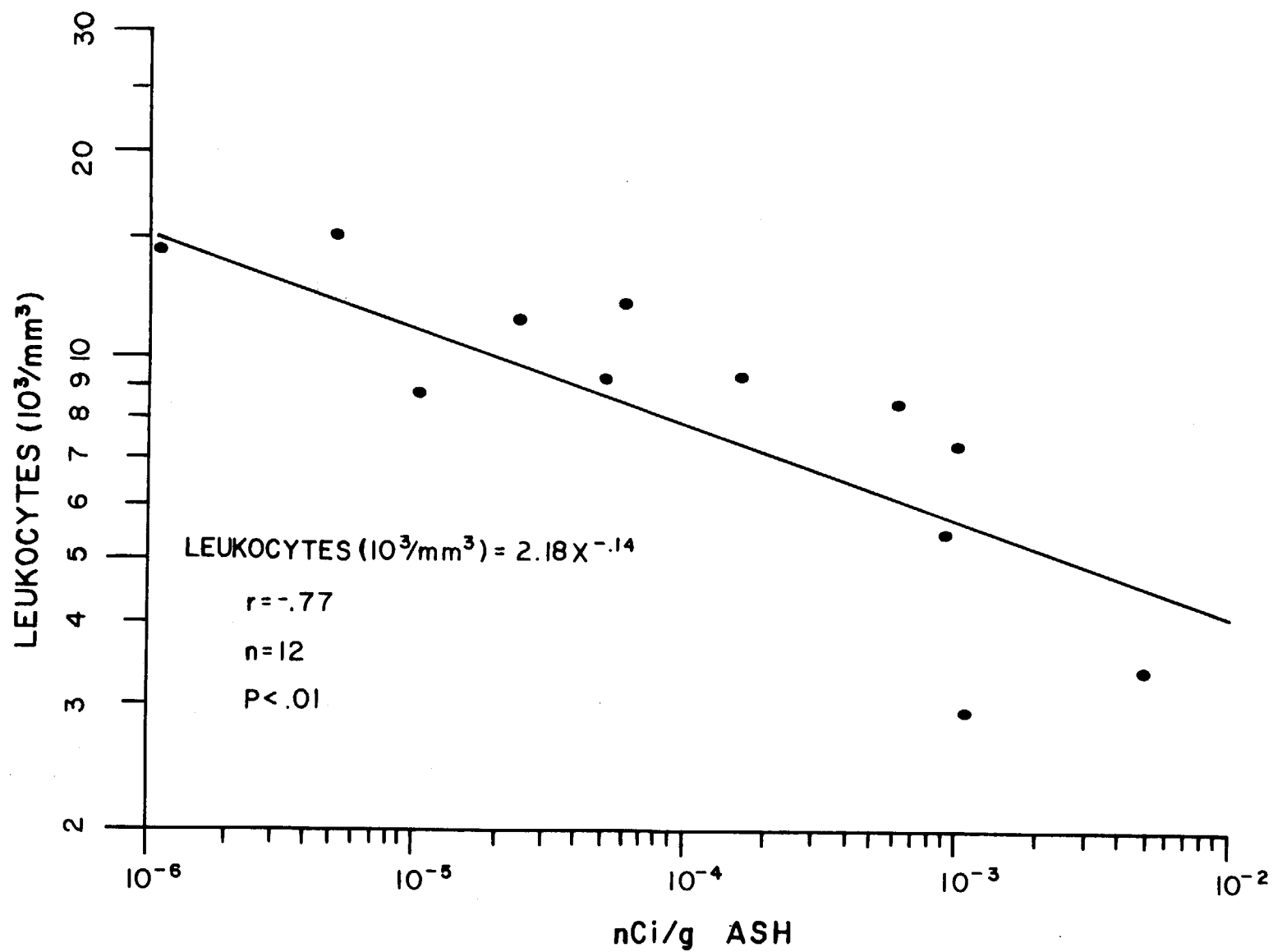


Figure 4. Total leucocyte counts in relation to ^{239}Pu carcass burdens in *D. merriami* from Area 11-C, NTS.



3. Development of procedures for sampling rodent burrows and lizard hibernals, including associated soil, fungi, invertebrates, food caches, and waste products.
4. Continued examination for histopathological effects, particularly the hemopoietic system of small vertebrates with high Pu carcass burdens. Techniques are being developed to examine rodent bone marrow to analyze blood cell formation and chromosome morphology as they relate to Pu body burdens.

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MICROORGANISMS

INFLUENCE OF MICROBIAL ACTIVITIES ON AVAILABILITY AND BIOTRANSPORT OF PLUTONIUM

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ABSTRACT

In this report, the data and conclusions from previous studies are summarized and discussed in context. Special attention is directed toward the effects of native vegetation and agricultural practices on soil microbial populations and the probable effects of changes in these microbial populations on the bioavailability and transport of plutonium to plants. In addition, the biological movement of transuranics in soil systems is addressed. A special technique developed for these studies to collect small particles is briefly outlined. The use of this technique, which eliminated cross-contamination problems, made possible uptake studies with various plutonium compounds using the soil fungus *Aspergillus niger*. The same technique is applicable to various phases of ongoing studies.

Experiments currently in progress are designed to elucidate the roles of various common soil microorganisms on the movement of transuranics in soil and on the bioavailability of transuranics to plants and animals. The results of these studies are expected to bring us one step closer toward a broader understanding of the importance of soil microbial interaction with transuranics in the environment.

INTRODUCTION

Microorganisms in plutonium-contaminated soils may play important roles as solubilizers and translocators of deposited plutonium. Solubilization processes will probably result in an increased transfer of plutonium from soil and plants to livestock, such as cattle, since solubilized plutonium is more readily absorbed by plants and animals. Translocation processes will contribute to plutonium migration within the soil profile.

In this report, the results of our laboratory and field experiments are summarized and discussed to show the influence of soil microbial activities on the bioavailability and biotransport of transuranics in desert environments.

DISCUSSION

We reported previously that *Aspergillus niger*, a commonly occurring soil fungus, took up plutonium from culture media and transported it to the fungal spores (Au, 1974). The chemical form of the plutonium added to the culture media, and the pH of the growth media, had a marked influence on plutonium uptake and translocation (Au and Beckert, 1975a). To better perceive the concept of plutonium transport, we used the term transport factor (TF) which is applicable for culture media where the distribution of nutrients and pollutants is uniform (Au *et al.*, 1976a). The concentration-independent transport factor is defined as the fraction of the total plutonium that is transported from the medium to the tissue divided by the fraction of the total dry mass transported from the medium to the tissue, or

$$TF = \frac{Pu_T/Pu_M}{M_T/M_M}$$

where, Pu_T = total plutonium content of tissue (e.g., mycelium, spores)

Pu_M = total plutonium originally present in the parent medium.

M_T = dry mass of tissue

M_M = dry mass originally present in the parent medium.

The TF shows immediately if accumulation of or discrimination against the pollutant has occurred: $TF > 1$ indicates accumulation, and $TF < 1$ defines discrimination against the pollutant.

In general, it was found that for *Aspergillus niger* all transport factors defining the movement of plutonium from culture media via the mycelium to the spores were smaller than 1, indicating discrimination against the transport of plutonium from the agar media to the spores. This was true for all chemical forms of plutonium tested at pH 2.5 and 5.5. As required by the definition, the transport factors derived from experiments using plutonium dioxide spheres were fairly concentration-independent, although the plutonium concentrations applied to the culture media varied by a factor of 100. The TF values derived from experiments using plutonium nitrate and plutonium citrate, respectively, were reasonably close at pH 2.5, but showed a greater variation at pH 5.5 (Beckert and Au, 1975).

The major problem encountered in these types of quantitative studies was the prevention of cross-contamination of collected material with the contaminant contained in the culture medium and in the parent mycelium. Cross-contamination was eliminated by the use of a sample collection technique that permitted the selective massing of aerial fungal spores, as well as a variety of other small particles, on a filter (Au and Beckert, 1975b). This new and simple collection technique was extensively evaluated at this Laboratory using *Aspergillus niger* grown on a buffered agar medium and on soil, both of which contained transuranic elements. Microscopic examinations of the filter contents showed the absence of mycelial and conidiophore (stalk) fragments. Because this technique prevented any direct contact between the spores and the medium containing the transuranic element, reliable and accurate transfer coefficients from medium to spores could be established for the first time.

The soil fungus *Aspergillus* was chosen as our test organism because it is ubiquitous and is morphologically well described (Raper and Fennell, 1965). More importantly for our experiments, it produces aerial spores atop lengthy stalks. This feature prompted the development of the collection technique mentioned above. *Aspergillus* is also present in the Nevada Test Site (NTS) soils; however, it is certainly not the only important soil microorganism in these soils. In fact, the soil fungal and bacterial populations of desert soils can be surprisingly high and diversified even in locations without plants. As plant growth increases, the number of the soil microorganisms increases, and their relative abundance may vary. Soil microbial surveys conducted in Area 13 of the NTS showed that in the 0-5-cm soil segment the fungal and bacterial populations were generally 2 to 4 times as high in the hummocks as in the soils without plant growth (Table 1). When the desert soil was cultivated and planted to produce crops under greenhouse coverings to simulate more typical agricultural conditions, the total fungal and bacterial numbers in the 0-6-cm soil segment increased during the growing season by factors of 3 and 16, respectively (Au *et al.*, 1976b). The averaged results are listed in Table 1, together with the relative abundance (in percent) of the major fungal genera as determined in these soils. Also included in Table 1 are the fungal and bacterial biomasses expressed in kilogram per hectare (kg/ha) and calculated very conservatively according to the method used by Alexander (1961). The biomass figure might provide a better appreciation of the potential of the microbial activities in these soils.

How can soil microorganisms influence the transfer of transuranics from soil to man? It has been well established that soil microbes can attack various minerals which are normally biologically unavailable to plants and can change them by a number of processes into forms available to plants (Waksman, 1927; Bollen, 1959; Alexander, 1961). A major role in these solubilization processes seems to be attributable to complexing cell exudates such as certain organic acids. It is known that many fungi, notably the black-spored *Aspergilli* (*Aspergillus niger*, *A. carbonarius*, *A. japonicus*, and *A. phoenicis*), and certain species of *Penicillium* produce and exude relatively large amounts of nonvolatile organic acids, such as citric, oxalic, and gallic, into the culture media (Hawker, 1950; Raper and Fennell, 1965; Chmiel, 1975). Smaller amounts are produced by some species of the *Mucoraceae*. In addition, *Aspergillus* spp.

Table 1. Microbial Populations of Soil Without Plant Growth, Hummock Soil, and Greenhouse (GH) Soil of Area 13

	Soil Depth Segment (cm)	FUNGI ¹						BACTERIA ¹	
		Total Numbers per Gram of Oven-Dry Soil (Thousands)	Biomass (kg/ha)	% <i>Mucors</i>	% <i>Aspergilli</i>	% <i>Penicillia</i>	% <i>Dematiaceae</i>	Total Numbers per Gram of Oven-Dry Soil (Millions)	Biomass (kg/ha)
Soil Without Plant Growth	0 -2.5	5.0 ± 1.2	11.4 ± 2.7	4 ± 3	73 ± 12	0 ± 0	2 ± 2	2.9 ± 0.5	1.2 ± 0.2
	2.5-5.0	1.5 ± 0.6	3.4 ± 1.4	6 ± 7	51 ± 20	6 ± 8	0 ± 0	2.4 ± 0.3	1.0 ± 0.1
	0 -5.0	3.2 ± 0.9	14.8 ± 4.1	5 ± 5	62 ± 16	3 ± 4	1 ± 1	2.6 ± 0.4	2.2 ± 0.3
Hummock Soil	0 -2.5	10.6 ± 1.3	24.2 ± 3.0	8 ± 4	0 ± 0	16 ± 5	16 ± 1	5.1 ± 0.5	2.2 ± 0.2
	2.5-5.0	16.3 ± 2.7	37.3 ± 6.2	4 ± 4	1 ± 1	45 ± 5	4 ± 1	7.4 ± 2.1	3.1 ± 0.9
	0 -5.0	13.4 ± 2.0	61.5 ± 9.2	6 ± 4	1 ± 1	30 ± 5	10 ± 1	6.2 ± 1.3	5.3 ± 1.1
GH Soil Before Planting	0-3	4.2 ± 1.6	11.5 ± 4.4	22 ± 6	7 ± 4	11 ± 4	22 ± 4	3.9 ± 0.5	2.0 ± 0.3
	3-6	9.6 ± 3.2	26.3 ± 8.8	10 ± 2	25 ± 8	36 ± 10	2 ± 1	4.7 ± 0.6	2.4 ± 0.3
	0-6	6.9 ± 2.4	37.8 ± 13.1	16 ± 4	16 ± 6	23 ± 7	12 ± 2	4.3 ± 0.6	4.4 ± 0.6
GH Soil at Harvest	0-3	27.9 ± 8.1	76.5 ± 22.2	48 ± 7	14 ± 4	22 ± 10	3 ± 1	101.3 ± 30.2	51.4 ± 15.3
	3-6	12.6 ± 3.1	34.6 ± 8.5	28 ± 5	10 ± 4	7 ± 2	25 ± 9	37.4 ± 13.4	19.0 ± 6.8
	0-6	20.2 ± 5.6	111.1 ± 30.7	38 ± 6	13 ± 4	15 ± 6	14 ± 5	69.4 ± 21.8	70.4 ± 22.1

¹All values are expressed as the average ± 1 standard error and are based on the same sample size.

and some other microorganisms are known to produce acids of the humic acids type which also have complexing properties. These complexing agents then attack minerals, transforming the mineral constituents into forms which can more easily be taken up by microorganisms and plants. We can assume that deposited, relatively insoluble plutonium (or other transuranics) is altered by similar processes, namely by the action of complexing organic substances. This view is supported by the results of extraction studies of NTS soil from Area 13 which demonstrated that citric acid solutions can remove in excess of 5% of the deposited plutonium (Beckert and Au, 1975). Similar results showing the efficiency of certain naturally occurring chelating agents for solubilizing plutonium were reported by others (Bondietti *et al.*, 1975; Tamura, 1975). Obviously, the production of complexing agents is not limited to soil microorganisms; compounds with complexing properties are also contained in and exuded by plant roots and other organisms.

Once the soil-deposited, relatively insoluble plutonium has been transformed into a more soluble form, a larger fraction will probably be taken up by microorganisms and by plant roots. Also, more soluble forms of plutonium may more easily be translocated vertically and horizontally by rain, or transferred to soil-ingesting animals ranging from earthworms to cattle, or become airborne with dust particles. Microbial cells can be transported or dispersed laterally or vertically in the soil by water or by predatory actions (Griffin, 1972; Ireland, 1975a, 1975b). This adds another dimension to the translocation and bioavailability of plutonium with time because predators, such as protozoa, nematodes, and arthropods, have a broader range of movement than soil fungi or bacteria.

It has been emphasized that the importance attached to the incorporated and soluble over the nonincorporated, insoluble forms of plutonium in soils lies in the probable differences in bioavailability and in biotransport to other trophic levels (Dunaway, 1976). We do not know in what complexed form or forms plutonium is stored in microbial cells; but whatever form it may be, it is very probable that the complexed plutonium is more soluble upon release from cells than the relatively insoluble plutonium as deposited in soil. However, definite proof is lacking as to whether successive microbial generations enhance the bioavailability of plutonium, and thus, with time, increase its availability to other trophic levels and its biotransport in the soil system. Experiments have therefore been initiated to determine if under laboratory conditions the availability and transfer of plutonium are increased during successive generations of microbial growth. Several fungal genera including *Aspergillus* will be cultured and incubated on malt agar plates which contain plutonium-238. Following incubation, part of the fungal materials will be collected and analyzed for plutonium; the remaining fungal growth will be terminated by ethylene oxide in part of the experiment and by heat treatment in another part. The plates will then be inoculated with spores of *Aspergillus niger*. After two or three weeks of incubation, aerial spores of *A. niger* will be collected with the collection system described earlier (Au and Beckert, 1975b) and analyzed for plutonium. As many collections as feasible will be made to measure the transport of plutonium to successive generations.

Our earlier experiments have shown that fungi can take up plutonium and translocate it within their domain. The present study should elucidate whether or not plutonium, once incorporated into fungal tissue, will be passed on to different fungal genera, as well as to successive generations of the same fungal species.

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DISTRIBUTION AND INVENTORY

PLUTONIUM DISTRIBUTION IN THE ENVIRONS OF THE
NEVADA TEST SITE--STATUS REPORT

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INTRODUCTION

Subsequent to the October 1974 Information Meeting of the Nevada Applied Ecology Group (NAEG), EPA efforts for the NAEG distribution and inventory studies have been directed toward analyzing the backlog of soil and air samples collected under the program and collecting and analyzing special-study soil samples.

DISCUSSION AND CONCLUSIONS

Plutonium in Soil

Analytical results from the soil sample backlog have not shown any significant differences from data previously generated. Many of those results are undergoing in-house review and will be reported later in a more thorough fashion.

The data generated from soil sampling to define the plutonium distribution around the NTS are again being reviewed to evaluate the associated errors. Analytical results from samples collected in an area northeast of the NTS, where concentrations up to 1.5 pCi/gm (91 nCi/m²) have been reported, have been evaluated. Twenty adjacent surface soil samples were collected from one location. Each sample was split and each split received two plutonium analyses. The procedures and results have been recorded, but not published (Church, 1973). The range of plutonium concentrations in these 80 analyses was 0.15 to 18 pCi/gm. These 80 results have been treated in numerous fashions; however, the logarithm of the data best fits a normal distribution. The computed log mean was 1.05 pCi/gm with upper and lower 95% confidence limits of 14.5 and 0.077 pCi/gm, respectively.

Numerous attempts have been made to develop a set of equations which could be used to define the distribution of Pu in soil in the off-NTS area. No attempt tested thus far has resulted in a correlation coefficient larger than 0.34.

Concern for the assumptions made in defining soil bulk density when converting concentration values to deposition values prompted the EPA to perform a field study in which two sets of samples were collected as reported by Church *et al.*, 1974, and another set collected where the actual volume was measured. Some soil collectors presume a sample volume by using a collection tool of known size and use an average soil bulk density to define the sample weight. This procedure is subject to errors in collection technique as well as the variability in soil bulk density. In this study, the third collection included refilling each sampled hole with a standard sand. The measured weight of sand was used to define the sample volume and bulk density. Each of the first two sample collectors sampled four profiles to 20 cm deep in 5-cm increments, i.e., collected 16 samples, while the third collected four samples. The average densities computed for the first two collectors were 1.70 and 1.62 g/cm³ with standard deviations of 0.08 and 0.05. By the actual volume measurement used in the third collection set, the computed density was 1.53 ± 0.05. It was noted that the former two collectors had errors in collection depths of +2.8 and +1.9 cm. Correcting for this excess volume revises their bulk density measurements to 1.49 and 1.48, both within the one-sigma error of the method in which actual sampling volume was measured. It may be concluded, then, that careful sampling with a known volume tool is sufficiently accurate to calculate bulk density for the objectives of the NAEG survey.

Plutonium in Air

Results from the analysis of air filters from the eight western U.S. locations previously reported (Bliss and Jakubowski, 1975) show ambient levels of ²³⁹Pu, ²⁴⁰Pu to be remaining within the range of data collected for the period 1966 through 1973.

An ongoing study which should be concluded by early 1977 will allow reporting the uncertainty of the air data. Laboratory analytical and counting errors can be determined but sampling errors have not been fully evaluated. Duplicate sampling has begun for the purpose of estimating the total error associated with the sampling and analyzing of air samples.

Three sampling locations near NTS have been selected for routine plutonium analysis in addition to the eight above. These locations are Las Vegas, Lathrop Wells, and Diablo. No analyses have been completed for these locations.

All locations routinely analyzed for plutonium are now using filters of polystyrene fiber mat which are easily dissolved in organic solvents.

Related Projects

1. Basin Studies--Sampling has been completed in three drainage basins to assess the concentrating effects of hydraulic movement. These areas are Fortymile Canyon, a basin southeast of Frenchman Flat, and a basin 160 km

north-northeast of the NTS. Analysis of samples is complete for Fortymile Canyon and the Frenchman Flat area. Preliminary review of the data does not show definable movement or concentration of plutonium. Uncertainties in the results have not been fully assessed; therefore, this lack of trend is not conclusive.

2. Vegetable and Fruit Sampling Study--Vegetable and fruit samples were collected from 26 locations around the NTS. Analysis for plutonium in the vegetable material collected near the NTS is not complete; however, a ^{239}Pu concentration of 24 pCi/kg wet weight was detected in a sample purchased from a Las Vegas commercial distributor. A second aliquot of ash from that sample yielded 5.9 pCi/kg of ^{239}Pu . Additional samples have been purchased and are undergoing analysis.

FUTURE PLANS

Analysis of backlogged samples will continue for soil and air. Few samples will be collected to further define the areal distribution of plutonium in soil; however, more special interest sampling is planned. Plutonium analysis of filters from the 11 air sampling locations reported above will be performed on a routine basis indefinitely.

A field study is planned for spring 1976 to sample hummocks created around shrubs by wind erosion. This study will be patterned after the recent on-NTS sampling performed by Los Alamos Scientific Laboratory (LASL) and Reynolds Electrical and Engineering Co., Inc. (REECo).

Two high-volume air samplers and one high-volume cascade impactor sampler will be fielded this spring to augment the routine air sampling network. The data will be used to evaluate the potential for resuspension in the off-site area.

Further studies are planned to evaluate the sampling errors involved with soil collection. Additional sampling will be accomplished if necessary. Evaluation of data already generated will continue. Plans include defining what differences may occur between current sample locations. These will be included in the total error. These adjustments will be used in analyzing all the data for trends such as the effect of distance from the NTS and of changing elevation in a drainage basin.

A study is under way to evaluate the statistical significance of analyzing 10 grams of soil as compared to analyzing 1 gram of soil. Plans include analyzing 20-gram aliquots to determine if this will significantly decrease the variability of replicate analyses.

A report is planned for the joint Environmental Protection Agency/Health and Safety Laboratory soil sampling effort conducted in 1974. Both members have analyzed their samples and a joint review of the data is planned.

Data preparation is under way for defining the ^{137}Cs distribution around the NTS in a fashion similar to ^{239}Pu .

In conclusion, primary emphasis of the plutonium survey is being diverted from sampling and data generation to data evaluation and reporting. A major portion of the effort in 1976 will be directed toward finishing the above plans and developing a comprehensive report of the plutonium distribution in the environs of the Nevada Test Site.

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CALENDAR YEAR 1975 STATUS:

NEVADA APPLIED ECOLOGY GROUP DISTRIBUTION AND
INVENTORY SAMPLING DATA

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U.S. Energy Research and Development Administration

ABSTRACT

To date, under this program over 6,000 soil samples have been taken from Areas 1, 4, and 5 of the Nevada Test Site. All of these samples were Ge(Li) scanned for Am-241 and other gamma-emitting radionuclides by Reynolds Electrical & Engineering Co., Inc. (REECo), and the results indicate only about 5% contained Am-241 in quantities above the detection limit (1 pCi/g of Am-241). In addition, approximately 10% of the samples were subjected to wet chemistry analysis for Pu-239 by REECo. The results indicate all contained Pu-239 in quantities above the detection limit (0.01 pCi/g of Pu-239). The largest concentrations of Am-241 and Pu-239 appear in the Hamilton and Small Boy sites of Area 5. The Hamilton soil data may be sufficient to estimate both an inventory and a distribution for plutonium over most of the site, but the data from the majority of the other sites are sufficient only to set upper limits of plutonium inventories. Extensive additional soil sampling and/or analysis will likely be required before estimates of plutonium inventories and distribution can be made for these sites.

INTRODUCTION

The Nevada Applied Ecology Group (NAEG) Distribution and Inventory Program's soil sampling program intensified in late 1974, and has continued to date. Over 6,000 soil samples from Areas 1, 4, and 5 have been analyzed. Wet chemistry analysis for plutonium has been used on some of these samples, while Ge(Li) scanning to infer plutonium concentration has been used for all. The inferred method is several orders of magnitude less sensitive than the wet chemistry analysis, and as a result, the inferred method for most samples can only be used to determine an upper limit of plutonium concentration.

METHODS

Soil samples were taken at predetermined locations for each event site. In general, the locations were the intercept points of a 200-foot by 200-foot grid surveyed from the ground zero point of each event site. In the immediate vicinity of ground zero for the Hamilton, Small Boy, and BFa sites of Area 5, and for the T-1 site of Area 1, additional locations were chosen using a 100-foot grid. In addition to these locations, samples were also taken at two random points within each 200-foot by 200-foot grid square of the Hamilton site.

The majority of sample locations were sampled at five different depths: the surface, 30, 60, 90, and 120 cm. Each location was at least sampled at the surface. Sampling was accomplished by first digging a 24-inch wide by 4-foot deep by 6-foot long trench using a backhoe. Then a 10 cm long by 10 cm wide by 5 cm deep sampling tool was used to remove the soil samples of interest.

A selected number of these samples were dried, weighed, ball-milled, and a 10 g aliquot used for wet chemistry analysis for plutonium. All of the soil samples were Ge(Li) scanned for 100 minutes.

A more detailed description of the sampling and analysis methods used in this program is given in the Nevada Applied Ecology Group Progress Report NVO-153, June, 1975.

DATA DISCUSSION

Table 1 lists the sampling grid size, the number of sample locations, and the number of soil samples taken from each site in the areas sampled to date. It also lists for each site the number of plutonium concentrations determined by wet chemistry, as well as the number of plutonium concentrations which can be inferred from Ge(Li) counting Am-241 and the number of concentrations of the three most significant gamma-emitting radionuclides.

Plutonium concentrations were determined for each sample taken from the Hamilton site of Area 5. Since two different sample location systems (grid system and random system) were used for the Hamilton site, there are two sets of plutonium concentration data for this site. This should allow both estimates of inventory and distribution, and estimates of their respective errors to be made.

The data from the Airdrop site and Able site of Area 5 contain so few plutonium concentration numbers that no plutonium distribution can possibly be estimated. However, an upper limit to the plutonium inventory for these sites can be estimated, if it becomes necessary to use these limited data.

Table 1. NAEG Distribution and Inventory Program Sampling Data Summarized by Site

Area	Site Name	Grid Bounds (x 100 ft.) ^[8]				Grid Spacing (in ft.)		No. of Sampling Locations	No. of Samples	No. of Samples With Usable Values					
		N.	S.	E.	W.	GZ	Other			Pu	Am ^[7]	Pu/Am	¹³⁷ Cs ^[7]	⁶⁰ Co ^[7]	¹⁵² Eu ^[7]
5	Hamilton ^[5]	6	10	4	8	100	200	140	745	743	57	55	104	349	363
	Smallboy	52	6	52	4	100	200	863	2,041	535	429	125	1,072	840	709
	Able ^[1]	4	6	6	4	200	200	32	157	40	8	2	22	102	116
	Airdrops ^[2]	6	6	6	6	200	200	47	234	40	0	0	18	8	2
	8Fa ^[3]	12	10	12	12	100	200	184	889	230	12	2	182	599	619
1	T-1 ^[4]	26	30	16	18	100	200	653	1,685	212	73	8	-	-	-
4 ^[6]	-	-	-	-	-	-	-	-	305	68	23	-	-	-	-

[1] Tumbler-Snapper series

[2] Ranger series: Baker I & II, Easy, Fox, Able

[3] Upshot-Knothole: Encore & Grable; Hardtack II: Wrangell & Sandford; Teapot: Met; Plumbbob: Priscilla; Coulomb "A"

[4] Tumbler-Snapper: Easy; Upshot-Knothole: Simon; Plumbbob: Galileo; Teapot: Apple II

[5] Includes data on a random pattern within each 200-ft. grid square

[6] Sampling discontinued pending evaluation of the sampling approach

[7] Most are from surface samples

[8] Measured from ground zero

The data from the Balloon facility of Area 5 and the T-1 site of Area 1 contain many plutonium concentration numbers. However, these represent such a small fraction of the total area of these sites that again, no plutonium distribution can possibly be estimated. However, an upper limit to the plutonium inventory can be estimated.

The data from the Small Boy site of Area 5 include a good fraction with plutonium concentrations. However, these concentrations vary considerably between adjacent sample locations, and it is not clear if these data can be used for more than determining an upper limit of the inventory for this site.

Sampling in Area 4 had been terminated until evaluations of the usefulness of this sampling approach can be made. Area 4 data have not as yet been examined to determine usefulness.

Current efforts of the PIDP studies are being directed at the evaluation of the program's sampling and analysis procedures, as well as quantification of the plutonium in the sites sampled to date.

CONCLUSIONS

Although thousands of soil samples have been taken, and sampling has proceeded from site to site and area to area, the current data are not sufficient to estimate distribution of plutonium for more than one or two of these sites. A limit for maximum plutonium inventory for each site can be made, but this would probably not be much more accurate than a mass balance on the original device. In order to estimate both plutonium distribution and inventory for all sites sampled to date, most of the soil samples will need to be analyzed for plutonium by wet chemistry, or by Ge(Li) scanning with more sensitive instruments (Pu inferred from Am-241). The only other alternatives are to resample by some more sensitive procedure, or to infer plutonium concentration from the fission product data. In the latter case, plutonium inference from Cs-137 does show promise.

STATISTICS

AN INITIAL SYNTHESIS OF AREA 13 ^{239}Pu DATA
AND OTHER STATISTICAL ANALYSES

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ABSTRACT

An initial effort is made here to synthesize the Nevada Applied Ecology Group $^{239,240}\text{Pu}$ data currently available from Area 13 (Project 57) on the Nevada Test Site. Plutonium concentrations for soil, vegetation, small vertebrates, and various tissues in beef cattle (grazed on the Pu-contaminated vegetation) are plotted on a single graph for visual comparison. Hypothetical $^{239,240}\text{Pu}$ concentrations for lung, skeletal bone, and kidney of a Standard Man assumed to live in and obtain most of his food from the area are also plotted. These hypothetical values were obtained using results from the plutonium transport and dose estimation model of Martin and Bloom (1976).

We also discuss here some methods for analyzing and reporting the underlying structure in environmental radionuclide data that are characterized by skewed (asymmetrical) distributions. The inadequacy, in many cases, of reporting only the arithmetic mean and standard error for a data set is emphasized. In particular, we illustrate the construction of a stem-and-leaf display using Pu data from Area 13 for graphically conveying the information content of a data set. A comparison is made between several estimates of central tendency, and the suggestion made that more than one such estimate should be routinely given.

Some recent efforts at experimenting with estimating Pu concentration contours with various computer algorithms are also displayed for Area 5 (GMX Site). Contours estimated on log-transformed data appear to have less bias than those obtained on untransformed data. The nearest-neighbor data search routines

QUAD and NEAR appear to result in contours with less bias than obtained using a polynomial fitting routine called TREND. It is suggested that Universal Kriging should be evaluated for its applicability for estimating plutonium contours and, possibly, inventory.

INTRODUCTION

Considerable effort has been expended in sampling soil, vegetation, small vertebrates, large vertebrates, and air for plutonium, americium, uranium, and other radionuclides in connection with the NAEG environmental sampling program on the Nevada Test Site (NTS) and the Tonopah Test Range (TTR). The results of this work to date have been presented primarily in the NAEG progress reports edited by Dunaway and White (1974), and White and Dunaway (1975), and the recent IAEA/ERDA Symposium on Transuranium Nuclides in the Environment.¹ It would appear useful, however, if these data could be pulled together in a concise manner so that the reader could obtain a better grasp of the total data picture to see how the soil results relate to those for vegetation, small mammals, etc. In this paper, we attempt the first step in such a synthesis by displaying many of the data collected at Area 13 (Project 57) on a single graph.

This area was chosen for our initial effort primarily because of the cattle grazing study currently under way in Area 13 (Smith, 1974, 1975, 1976; Smith *et al.*, 1976a). A limited number of data on plutonium concentrations in cattle tissue are available for comparison with the pelt, gastrointestinal (GI) tract, and carcass of small vertebrates as well as with soil and vegetation concentrations. While we are concerned entirely with Area 13 in this paper, there is an obvious need to graphically pull together the data from the other nine safety-shot sites being studied.²

The information plotted for Area 13 consists of simple arithmetic means (AM), standard errors (SE) and ranges (maximum minus minimum observation) of the data. We suggest, however, that these summary statistics often do not adequately describe the underlying structure in the data (they may even hide it). This is due in part to the skewed (lognormal-type) distributions often observed in data sets. Consequently, we discuss some additional ways that data sets can be quickly summarized and displayed graphically that convey additional information. Different methods of estimating the "center" of a skewed distribution are considered relative to how they "weight" the largest observations in a data set.

¹Held in San Francisco, November 17-21, 1975.

²Clean Slates 1, 2, and 3 on the TTR; Double Tracks on the Nellis Air Force Bombing and Gunnery Range; Area 5 (GMX Site) and Area 11 (Sites A, B, C, and D) on NTS.

We also take this opportunity to review the field sampling design used at the safety-shot sites to estimate the inventory of plutonium in soil and vegetation. Our experience analyzing these data obtained using stratified random sampling (sampling at random locations within strata) suggests that the use of a systematic (grid) pattern in conjunction with random sampling may result in better estimates of the geographical distribution of plutonium than we have obtained thus far, while still providing data suitable for estimating inventory. We also display some recent results from our continuing effort to experiment with estimating plutonium concentration contours using SURFACE II, a computer contouring package under continuing development by the Kansas Geological Survey (Sampson, 1973). These results were presented orally at the above-mentioned IAEA/ERDA Symposium, but most could not be published in the proceedings due to space limitations.

SUMMARIZATION OF $^{239,240}\text{Pu}$ DATA FOR AREA 13

Background Information

Figures 1 and 2 show the soil and vegetation sample locations for the Area 13 (Project 57) study site. These were chosen at random within each stratum discussed by Gilbert and Eberhardt (1974). Figure 3 gives the AM, SE,³ and range for soil, vegetation, small vertebrates, and beef cattle. The soil and vegetation data are in units of nCi/g dry weight for comparison purposes. These data were taken from Gilbert *et al.* (1975, Table 14) and Romney *et al.* (1975, Table 2). The vegetation data are also given in units of nCi/g ash (from Romney *et al.*, 1975, Table 3) for comparison with the small vertebrate and cattle tissue results which are in units of nCi/g ash. The soil and vegetation data are given separately for each of the six activity strata. The results for pelt, GI tract, and carcass for the 11 *Dipodomys microps* taken from strata 1 through 4 within the inner fence (inner compound) were pooled together since the data presently available do not indicate consistent differences in Pu concentrations with increasing soil activity strata. The center of activity of each small vertebrate collected thus far in Area 13 (of which the 11 here are a subset) is given in Figure 4. Most of the rodent data used here were reported by Moor and Bradley (1974, Table 9). Bradley and Moor (1975) emphasize that about half of the radioanalysis data on rodents were not available at that time.

The data given in Figure 3 for beef cattle have been discussed in part by Smith *et al.* (1976a). The design of this cattle study is also discussed in Smith (1974, 1975). Note that the number of samples varies from 1 to 3 so that estimates of precision are either impossible to obtain or are extremely unreliable. More data will soon be available. The cattle data discussed here may be divided into three groups based on length of grazing: {2, 3, 8}, {1, 4, 6}, {5}, where the numbers in brackets are identifying cattle numbers.

³SE = $s/\sqrt{n}$ = (standard deviation)/ $\sqrt{n}$.

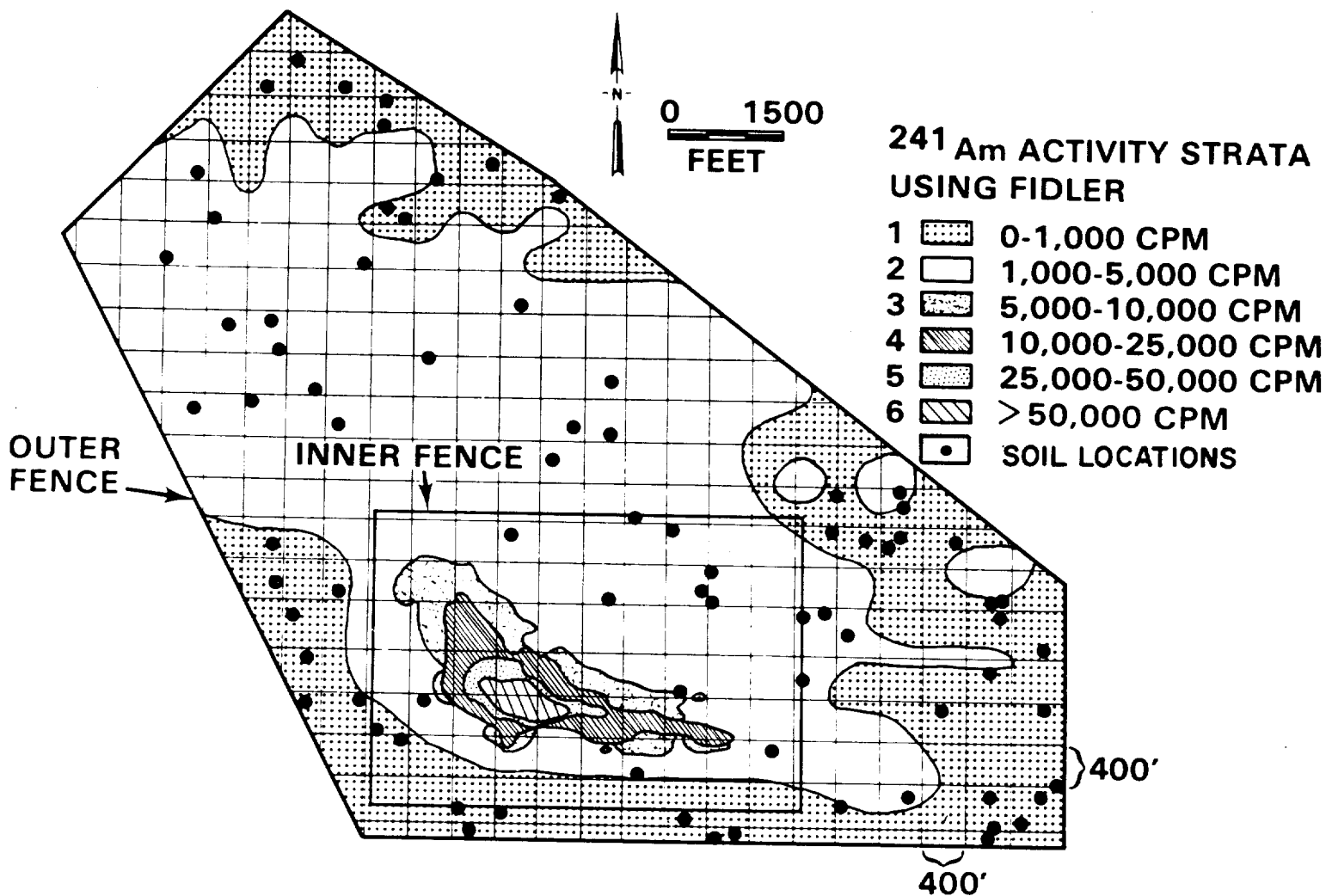


FIGURE 1. Area 13--Random Soil Locations Within Strata 1 and 2.

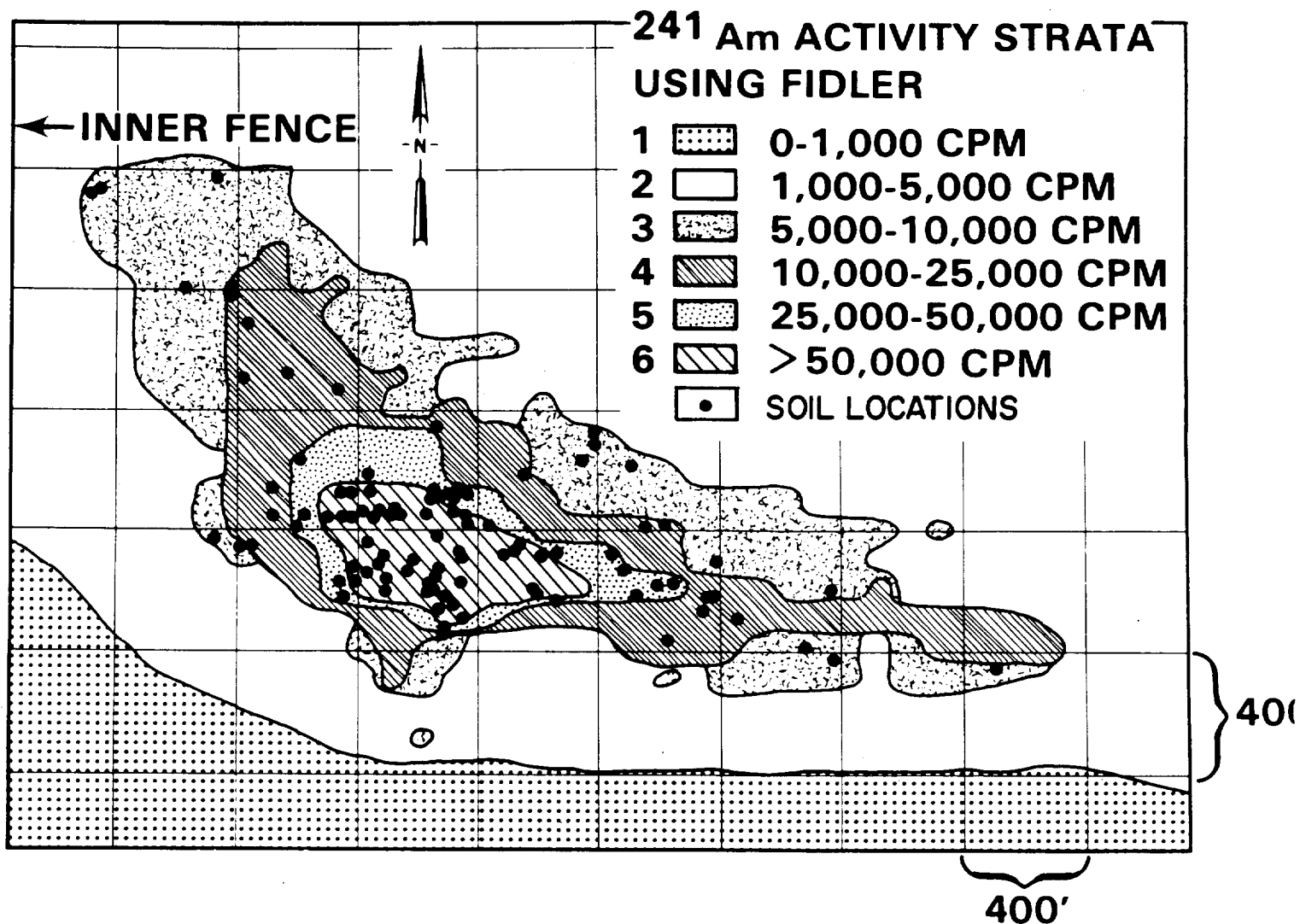


FIGURE 2. Area 13--Random Soil Locations Within Strata 3, 4, 5, and 6.

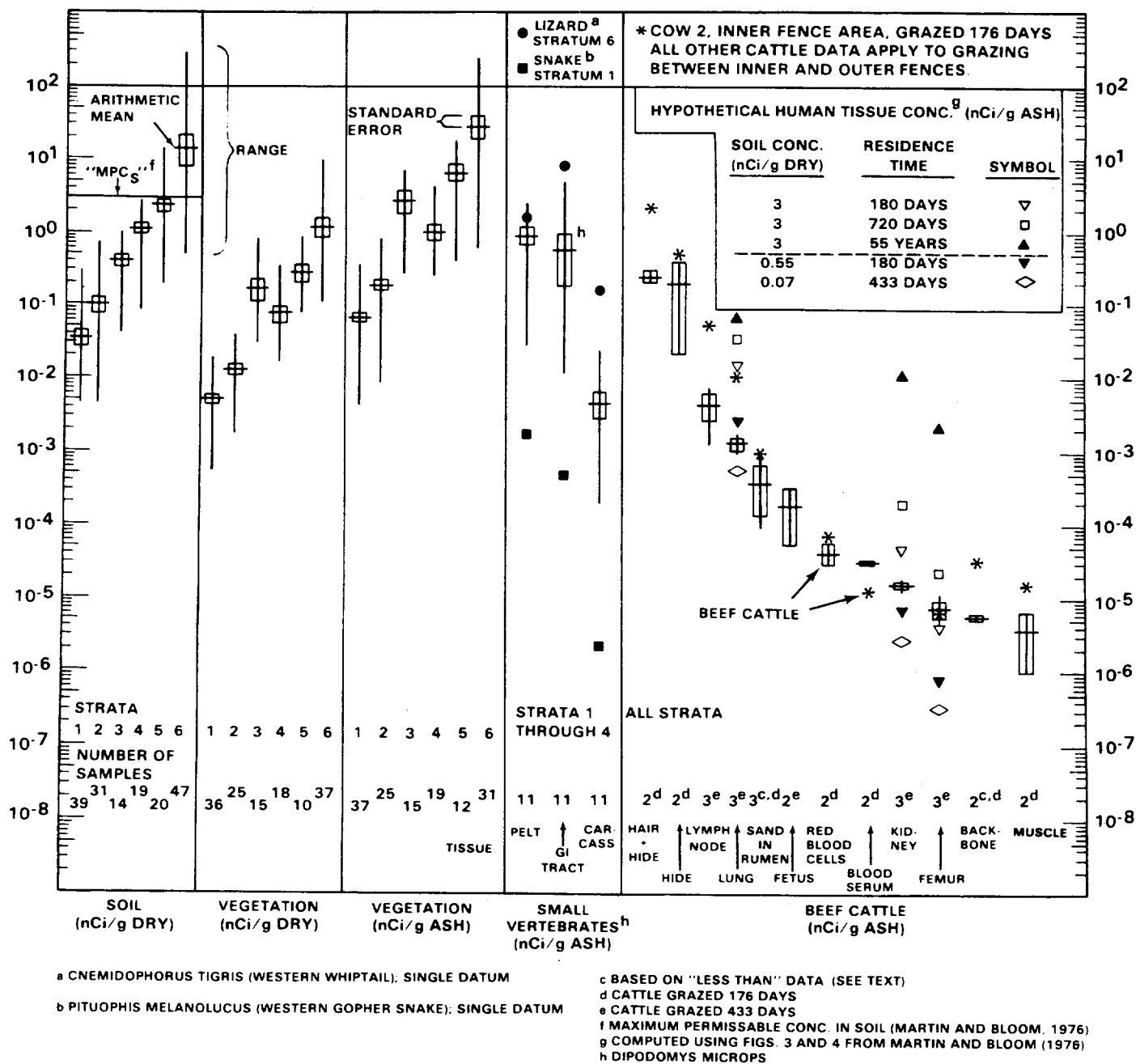


FIGURE 3. Concentration of $^{239,240}\text{Pu}$ in Soil, Vegetation, Small Vertebrates, Beef Cattle, and Hypothetical "Standard Man" in Area 13 (Project 57), NTS.

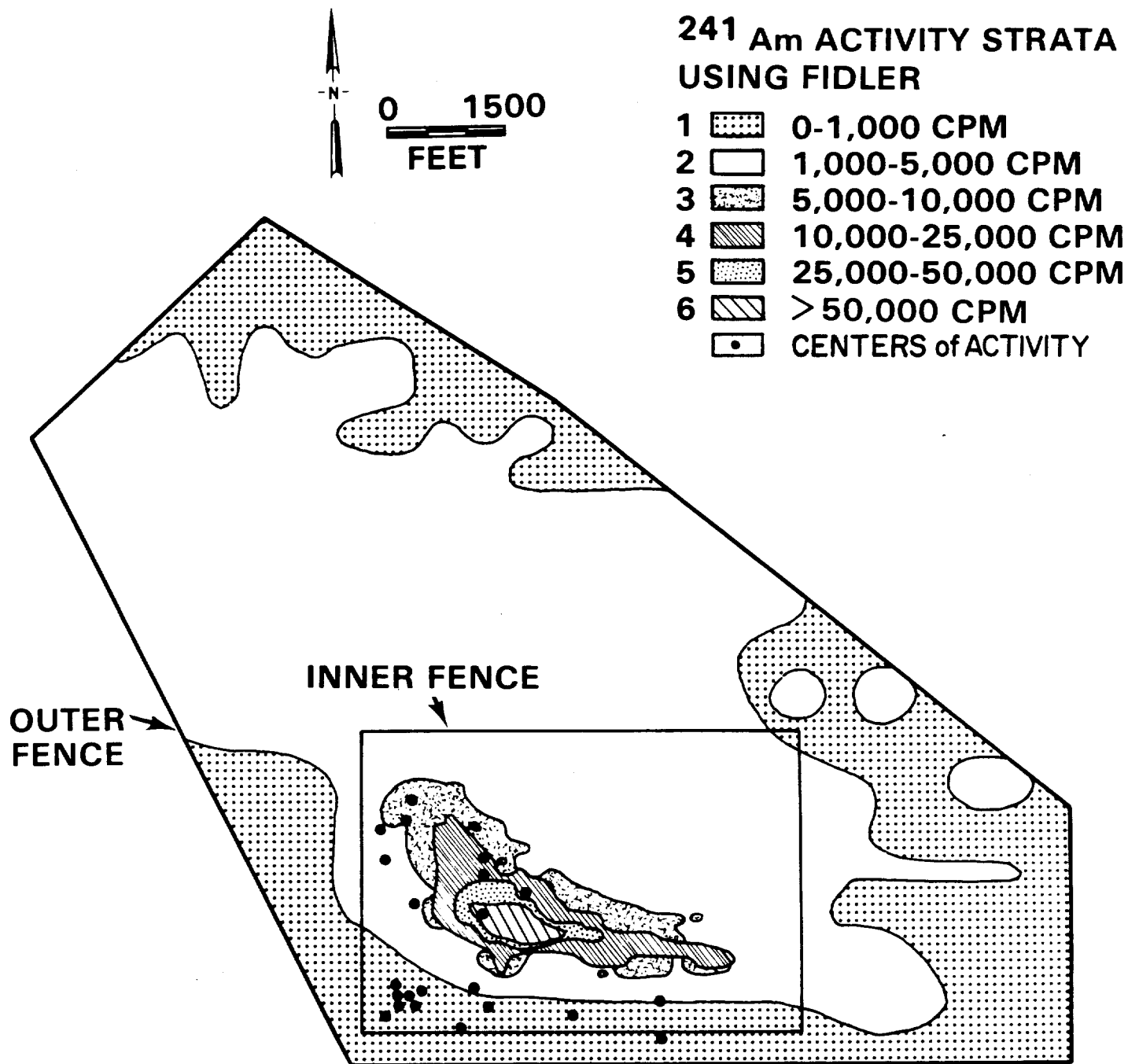


FIGURE 4. Area 13--Centers of Activity for Sacrificed Small Vertebrates.

These three groups were grazed 176, 433, and 637 days, respectively. Cow number 2 was grazed in the inner compound within the inner fence enclosing the area around ground zero (Figure 4). The remaining six cattle were grazed in the outer compound between the inner and outer fences, except for one week in May, 1974, when they inadvertently grazed the inner compound area. The data for cow 2 are plotted separately from the other cattle data to compare tissue concentrations resulting from grazing the two compounds. The weighted⁴ average ^{239}Pu concentration in surface soil in the inner and outer compounds is roughly 0.55 and 0.07 nCi/g dry, respectively. The weighted averages for the vegetation are roughly 0.06 and 0.009 nCi/g dry, respectively. The data identified by footnote symbols d and e in Figure 3 refer to groups {3, 8} and symbol e refers to group {1, 4, 6}. Data for group {3, 8} were used when data for {1, 4, 6} were not available. Concentrations for cow 5 (grazed 637 days) are not shown in Figure 3 but are larger than for group {1, 4, 6} (grazed 433 days) by a factor of 3 for kidney, 1.1 for muscle, 1.1 for lymph node, and 1.3 for femur.

Comparisons Between Soil, Vegetation, Small Vertebrates, and Beef Cattle

It is clear from Figure 3 that average soil and vegetation ^{239}Pu concentrations decrease with distance from ground zero (GZ) (this is shown more forcefully, perhaps, by the 3-dimensional plot of the soil data in Figure 15 of Gilbert *et al.*, 1975). The vegetation samples were taken in close proximity of the soil samples (usually within 5 to 10 feet). The relationship between Pu concentrations in surface (0-5 cm) soil and vegetation is summarized and discussed in Romney *et al.* (1975, 1976) by the computation of average vegetation, $\bar{x}$, to soil, $\bar{y}$, concentration ratios computed as $\bar{x}/\bar{y}$. They note that these average ratios tend to decrease with increasing soil concentrations in surface soil, due presumably to a differential particle size distribution occurring within the fallout pattern of Area 13. Gilbert *et al.* (1975, Figures 29-32) have plotted the vegetation-soil data pairs for selected strata to illustrate the poor correlations often observed between vegetation and soil even when collected at adjacent locations.

Concerning the relationship between concentrations in soil and small vertebrates, Bradley and Moor (1975) point out the lack of an obvious correlation (based on limited data) between ^{239}Pu concentrations in pelt, GI tract, and carcass with increasing concentrations in soil. They suggest that the mobility of rodents results in exposure to a wide range of Pu concentrations in soil due to normal traversing of several activity strata. Indeed, from the amount of variability encountered in soil concentrations even within each stratum (indicated by the ranges for soil plotted in Figure 3), it is not surprising to see large variation in rodent pelt and tissue concentrations. Also, the rodent data given in Figure 3 apply to rodents whose centers of activity fall in strata 1 through 4 (Figure 4). The analysis of tissues from animals whose centers of activity are nearer ground zero (strata 5 and 6) would be helpful in understanding the relation between soil and rodent concentrations.

⁴Each stratum average concentration was weighted by the approximate proportional area of the stratum within the compound.

grazing period in the inner compound area.⁵ These estimates agree fairly well with the 620 ± 130 nCi/day and $1.1 \times 10^5 \pm 2.4 \times 10^4$ nCi over 176 days that we estimated would be consumed by a cow grazing the inner fence area. This latter estimate is based on average ^{239}Pu concentrations for shrubs *Eurotia lanata* and *Atriplex canescens* as reported in Romney *et al.* (1975, Table 1) for Area 13, assuming the cow's diet consisted entirely of these two shrubs; 36% of the former, 64% of the latter. These proportions are approximately those reported by Smith (1975) as obtained from vegetation analysis of the rumen contents of fistulated steers for grazing date September 5, 1973.⁶ The latter estimate (620 ± 130 nCi/day) does not consider the ^{239}Pu that is ingested with soil. Presumably, the estimate of 560 nCi/day obtained by Smith *et al.* (1976a) is influenced to some extent by plutonium ingested with soil since the estimate is based on concentrations of vegetation and liquid taken from the rumen.

Smith's estimate is based on Pu concentrations found in rumen contents ingested over a 24-hour grazing period for months July through November, 1973 (cows 2, 3, and 9 were grazed in May through October, 1973). These concentrations remained relatively stable until August, 1974, when they increased by an order of magnitude (Smith *et al.* (1976a), Figure 3). Smith noted the increase might be due to a change in vegetation composition in the diet or to the ingestion of greater amounts of soil. Another possibility is that the cattle concentrated their grazing in the GZ area on that 24-hour period. Whatever the cause, this variability illustrates that the ingestion estimates above may not be generally applicable for all field situations.

The estimated ingestion rates above may be compared with the estimate of 500 nCi/day obtained using the model of Martin and Bloom (1976, eq. 7.1) (see below) if the amount of Pu ingested with soil is ignored, and using the weighted average soil concentration within the inner compound (0.55 nCi/g dry). An estimate of 770 nCi/day is obtained using their eq. 7.1 if the Pu in ingested soil is included (assumed to be 500 g/day; see footnote 7).

None of these estimates for ingested Pu consider the rate at which Pu is inhaled. The only estimate of this rate for beef cattle available to us comes from Martin and Bloom (1976). Their model specifies this rate to be $0.0110 C_s$ (pCi/day), where C_s is the Pu concentration (pCi/g) in soil. Using $C_s = 550$ pCi/g as we did above for estimating the ingestion rate, this equation gives

⁵Using the same data (supplied by D. D. Smith) but substituting the arithmetic mean for the geometric mean, we obtained 700 ± 130 nCi/day (mean $\pm$ standard error) or $1.2 \times 10^5 \pm 2.3 \times 10^4$ nCi over the 176 day period.

⁶There is considerable variability, however, in the botanical composition in the diet of these fistulated steers over time. Grasses and forbs were often the major items in the diet rather than shrubs. The average intake per day was based only on *Eurotia lanata* and *Atriplex canescens* since samples of forbs and grasses were not collected for plutonium analyses.

⁷Eq. (7.1) assumes cattle ingest 2000 g of soil/day. Smith (1976) found that less than 500 g/day was more accurate and is the figure used in calculations here.

about 0.006 nCi/day. We note for consideration in the next section that their model specifies the Pu inhalation rate for man to be 5.5 times less than that for beef cattle, or in this case about 0.001 nCi/day.

The model of Martin and Bloom allows us to calculate hypothetical ^{239}Pu concentrations in muscle of grazing beef cattle resulting from inhalation and ingestion over time period t . It is of interest to compare these model estimates with those reported in Figure 3 for cattle that grazed Area 13. Using data in Martin and Bloom's Figure 2 and their equations 7.1,⁷ 9, and 10 we obtained hypothetical ^{239}Pu concentrations in muscle of 2.4×10^{-6} and 3.1×10^{-7} nCi/g (wet weight) for soil concentrations (C_s) of 0.55 and 0.07 nCi/g, respectively, for grazing time $t = 176$ days.⁸ On an ash weight basis, these muscle concentrations become approximately 6.5×10^{-5} and 8.2×10^{-6} nCi/g ash. The conversion from wet to ash weight was made by multiplying by the average ratio of received weight to ash weight (see footnotes 9 and 10) of calves and cows grazed in Area 13. This average $\pm$ approximate SE was 27 ± 6 . From Figure 3, we see that these estimates of Pu concentrations are factors of about four and two larger than actually found in cattle muscle tissue in the inner and outer compounds, respectively. Of course, these computations do not constitute an adequate evaluation of Martin and Bloom's model since so few data are available. Also, the parameter estimate used in the model are not known with any accuracy. The equations of the model may also require modification.

In Figure 3, the Pu concentrations for tissue samples are given on an ash weight basis. Due to the difficulty of obtaining a reproducible ash weight on tissue samples in the laboratory, it may be preferable to report results on a dry weight basis. Smith (1976) prefers wet weight to ash weight for cattle tissue samples due to high variability in percent ash reported by laboratories for similar tissues, but he does not consider the dry-weight comparison. A problem with reporting on a wet weight basis is that samples tend to dry out, and different laboratories may use different handling procedures that affect the recorded wet weight. Lee *et al.* (1976) report on a rapid dissolution method for tissue sample analyses for reporting on a dry weight basis. Their procedure involves cutting the wet tissue (up to 250 grams) into small cubes followed by successive six-hour drying periods until a constant weight is obtained. This technique may allow the determination of reliable dry weights so that soil, vegetation, small mammals, and cattle tissue samples can be reported and compared on a dry weight basis. We hope to be able to statistically examine this question of reporting on a wet, dry, or ash basis in the near future.

Hypothetical ^{239}Pu Concentrations in Human Tissue

An important goal of the NAEG program is to assess the potential health hazard to man from the plutonium present in the environs of NTS. Martin and Bloom (1976) have devised a plutonium transport and dose estimation model based on NTS studies that estimates inhalation and ingestion rates, organ burdens, accumulated doses, and dose commitments for a Standard Man assumed to live in and obtain most of his food from a Pu-contaminated area such as Area 13 at NTS. Using this model based on parameter values found in ICRP (International Committee on Radiological Protection) publications, they estimated that the

average concentration of ^{239}Pu in the soils of contaminated areas at NTS which would result in a predicted dose rate of 1.5 rems/year to the lung is about 3 nCi/g dry. They use this rate as a "maximum permissible concentration" of ^{239}Pu in soil (MPC; plotted with the soil concentrations in Figure 3). The rate of 1.5 rems/year is the dose rate for individual members of the public that the ICRP (1966) has recommended should not be exceeded.

Our interest here is to use this model for estimating hypothetical ^{239}Pu concentrations in lung, skeletal bone, and kidney of Standard Man for soil concentrations at the MPC level (3 nCi/g dry) and for the average soil concentrations existing within the outer and inner compounds of Area 13 (0.07 and 0.55 nCi/g dry, respectively). These hypothetical human tissue concentrations are obtained for one or more of the following exposure (residence) times of Standard Man: 180 days, 433 days, 720 days, and 55 years. The results of the model for residence time 180 days and soil concentration 0.55 nCi/g can presumably be compared with tissue burdens from cow 2 that grazed the inner fence area for 176 days. Similarly, it is of interest to compare the model results for 0.07 nCi/g and 433 days with the observed average tissue concentrations for cows 3 and 8 since they grazed the outer compound for 433 days.

These results for Standard Man were obtained by first dividing total organ burdens⁸ (in units of curies) as estimated by the model, by the mass in grams of each organ (500, 7000, and 300 grams for lung, bone, and kidney, respectively; from Martin and Bloom, Figure 3). These concentrations were converted to an ash weight basis by multiplying by the average ratio of received⁹ weight to ash weight obtained for the lung, bone, and kidney tissues of cows and calves grazed in Area 13; the assumption being made that these ratios also apply to man. These average¹⁰ ratios were estimated to be (average $\pm$ approximate SE) 36 ± 2.5 , 4.6 ± 0.7 , and 37 ± 3 for lung, bone, and kidney. Multiplying the ^{239}Pu concentrations (nCi/g ash tissue per 1 pCi/g dry of soil) by the concentration in soil (pCi/g dry) gives the estimates plotted for hypothetical Standard Man in Figure 3.

Before discussing these results, we quote from Martin and Bloom:

"The present model is but one of several that have been investigated in the course of the NAEG plutonium study. It presents our best effort to judge and interpret the information currently available. The design of the model as well as the assumptions and parameter values selected for its implementation comprise what we believe to be a reasonable working hypothesis which is subject to continuing reappraisal as new information comes to light. While it is not the last word on the subject, it does provide a provisional method for evaluating potential health hazards associated with Pu-contaminated areas at NTS."

⁸Obtained from W. E. Martin. Also available from Figure 4 in Martin and Bloom (1976).

⁹Tissue samples were frozen until analyzed.

¹⁰Average ratio computed as $\bar{x}/\bar{y}$, where $\bar{x}$ = average received weight, $\bar{y}$ = average ash weight; n was 8, 11, and 7 for lung, bone (femur, vertebra), and kidney. SE approximated using formula given in footnote to Table 17 in Gilbert *et al.* (1975).

The reader is referred to their paper for details concerning the assumptions and parameter values used in the model.

Turning to Figure 1, we see that the hypothetical ^{239}Pu concentrations in lung, bone, and kidney of Standard Man for 433 days residence in an area with soil concentrations of 0.07 nCi/g are less by factors of (roughly) 2, 22, and 6 than the mean ^{239}Pu concentrations in these same tissues for the beef cattle that grazed 433 days in the outer compound. The hypothetical values for human lung and bone for 180 residence days at 0.55 nCi/g of soil are factors of 5 and 8 less than obtained for cow 2, which grazed 176 days in the inner compound (no data are available for the kidney of cow 2).

Factors of 2 and 5 are perhaps not unexpected for lung since in the model the inhalation rate of plutonium (pCi/day) for beef cattle is taken to be (based on NTS studies) 5.5 times greater than that of man (see eq. 1 and the discussion on cattle inhalation rates in Martin and Bloom). Hypothetical human concentrations in lung and bone at the MPC level (3 nCi/g of dry soil) for 180 residence days are about equal to concentrations found in these two tissues of cow 2. According to the model, human tissue concentrations in lung, bone, and kidney increase by factors of 3, 6, and 6 in the time span from 180 to 720 residence days when soil is at the MPC level. The ^{239}Pu concentrations in these human organs increase by factors of roughly 2, 80, and 70, respectively (according to the model), between residence times 720 days and 55 years for soil at MPC. The greater increases for bone and kidney between 720 days and 55 years occur because the model assumes no losses of plutonium from these organs (see Figures 3 and 4, Martin and Bloom).

Discussion

We don't have now, nor are we likely to have in the near future, estimates of the accuracy (bias) of these hypothetical human tissue concentrations since no humans live in these contaminated areas. We also do not have any precise knowledge of the precision (variability) of these model estimates. We presume this variability is largely due to the high variability of most estimates of parameters used in the model. As new information becomes available from the continuing NAEG studies, and we learn how to design better field studies and extract more information from the data collected (see next section), the model will undoubtedly be modified and improved. As Martin and Bloom stress, the present model is not the last word on the subject.

More data are clearly needed to obtain better estimates of plutonium burdens in tissues of small vertebrates and grazing cattle, particularly for GZ areas. Considerably more data will soon be available, at which time Figure 3 should be revised. Information is also available on concentrations of ^{238}Pu , ^{238}U , and ^{241}Am that needs to be related to concentrations for soil, vegetation, and small vertebrates.

SUMMARIZATION OF DATA SETS

Measures of Central Tendency

Several thousand radiological analyses have been completed for the NAEG program over the past several years. These data contain a great deal of information relative to NAEG objectives, but much of it may not be apparent without appropriate statistical treatment of the data. Up to this point in time, we have used simple (arithmetic) means and standard errors to describe data sets such as the "average" ^{239}Pu soil or vegetation concentrations in various soil activity strata of safety-shot sites (Table 14, Gilbert *et al.*, 1975). In this section, we would like to briefly discuss some alternative ways of summarizing a data set that give additional information to the reader. A common characteristic of radionuclide data collected from environmental studies is the occurrence of one or more observations that are much larger in magnitude than the bulk of the data. This is illustrated in Figure 5 by comparing the symmetric normal distribution with the asymmetric lognormal distribution (Aitchison and Brown, 1969, page 9) for parameters $\mu = 0$ and $\sigma^2 = 0.50$.

Our primary concern here is with how one should summarize the information contained in a data set from an underlying distribution of the asymmetric type. We will be particularly interested in choosing the most appropriate estimator of the "center" of the distribution. If the underlying distribution (from which the actual data values are drawn) is symmetric, then the mode, median, and arithmetic mean (AM) fall at the same place on the distribution. The mode is the value that occurs most frequently; the median is that value above which and below which half the values occur. For an asymmetric distribution, the mode, median, and AM do not coincide. For the lognormal distribution (Figure 5), the mean is always larger than the median which in turn is larger than the mode. The extent of the difference between the mean, median, and mode for the lognormal can be determined for any μ and σ^2 by using the formulae in Figure 5.

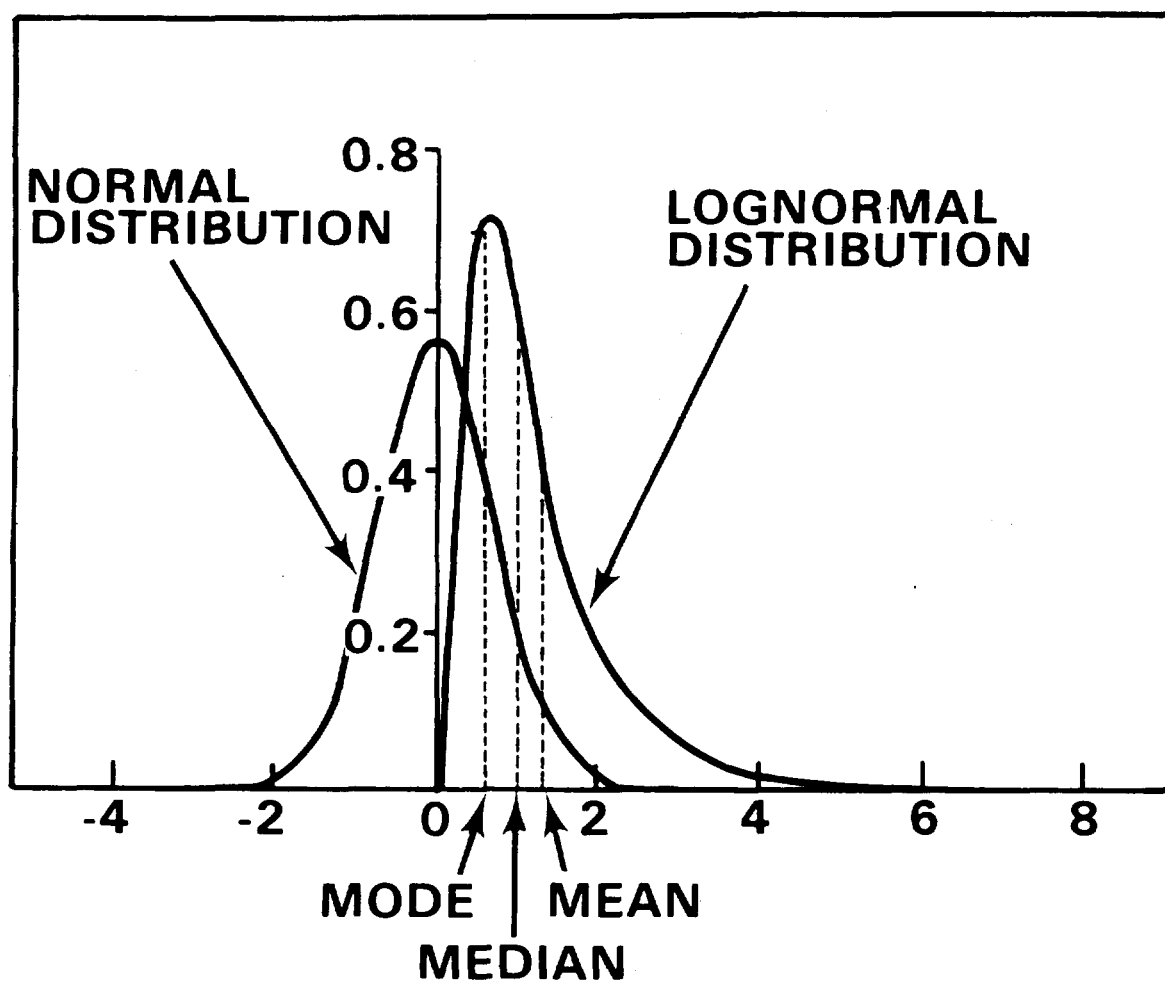
Another measure of the "center" of any distribution is the geometric mean (GM) computed as

$$GM = \left(\prod_{i=1}^n X_i \right)^{\frac{1}{n}}$$

or equivalently

$$GM = \exp \left(\frac{1}{n} \sum_{i=1}^n \log_e X_i \right) .$$

This mean gives less weight to the larger data values than does the arithmetic mean. In fact, the GM will always be less than the AM unless all data points are equal, in which case the AM and GM will be numerically equal. The GM is sometimes referred to as the "lognormal mean." However, the GM is an estimate of the median, not the mean, of the lognormal distribution. Actually, it is a biased estimate of the median for this distribution (an unbiased estimator is given by Zellner, 1971), but this may be of little practical importance since the true distribution is usually unknown and may not be lognormal.



	<u>NORMAL</u>	<u>LOGNORMAL</u>
MODE:	μ	$\exp (\mu - \sigma^2)$
MEDIAN:	μ	$\exp (\mu)$
MEAN:	μ	$\exp (\mu + \frac{1}{2} \sigma^2)$

FIGURE 5. Frequency Curves of Normal and Lognormal Distributions for $\mu = 0$ and $\sigma^2 = 0.5$.

of ^{239}Pu inventory for stratum 6 soil would have been about 8 rather than 19 curies. This in turn would have reduced the total estimated inventory for all six strata from 44 to 36 curies.¹¹ The apparent precision of the estimate would also have improved due to the elimination of this single datum.

¹¹The estimated total inventory for ^{239}Pu in Area 13 (Project 57) is roughly 24 Ci if the GM is used in place of the AM and the datum 305 nCi/g is not deleted.

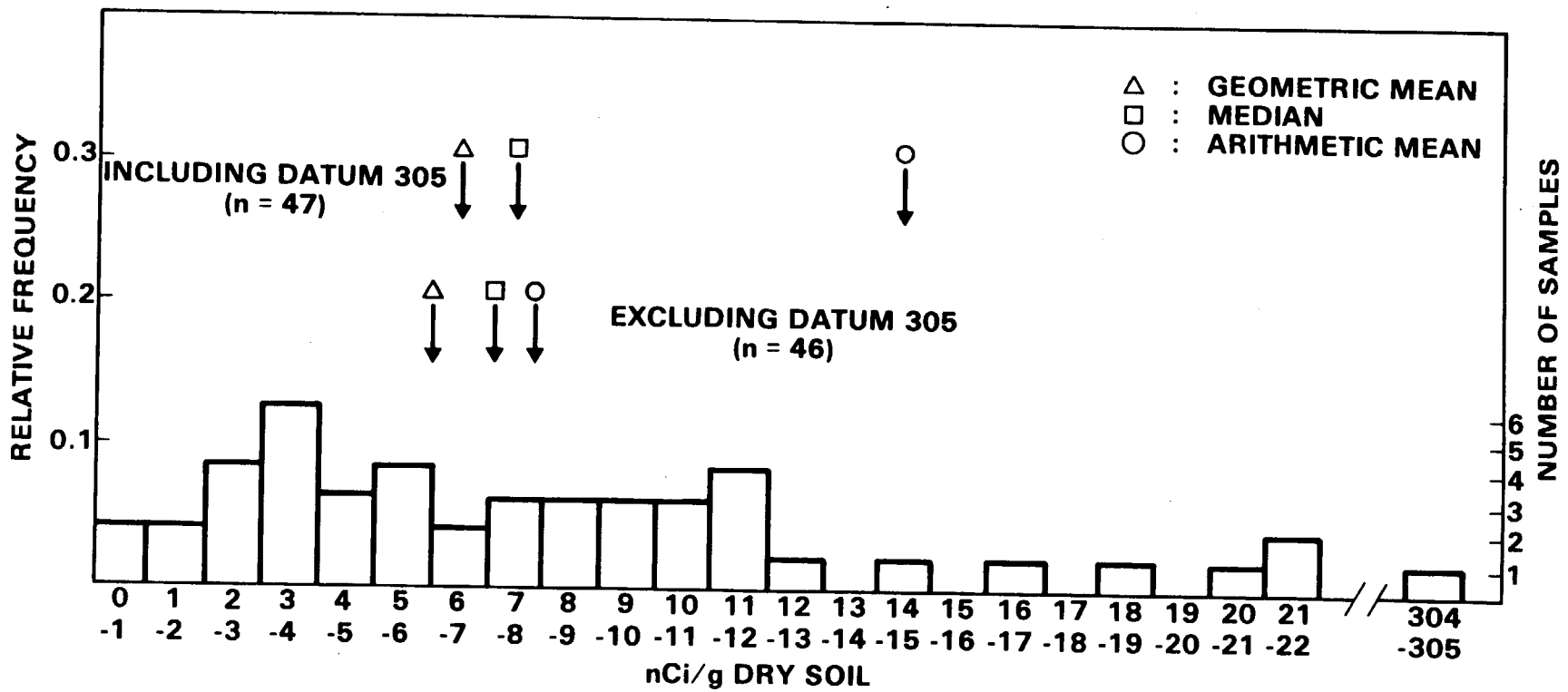


FIGURE 6. ^{239}Pu Concentrations in Soil From Stratum 6 of Area 13 (Project 57).

We are faced then with deciding which estimator to use for asymmetric distributions. Our choice should depend on the objective of the study and the use to be made of the estimator. Does one want to estimate where most of the data in a data set lie, or is it important to give extra weight to the extreme values for the purpose at hand? In Figure 6, the AM clearly overestimates where the bulk of the data lies. One could argue, however, that when working with a potentially harmful substance such as ^{239}Pu in the environment, it may be preferable to be conservative in the sense that we tend to overestimate rather than underestimate average soil concentrations.

Stem-and-Leaf Displays

Probably the best approach when working with asymmetric distributions is to compute more than one estimate of the "average" of the data set. Even this, however, will not convey much information about the scatter or variability present in the data. One method for obtaining such information is to plot the data in histogram form as was done in Figure 6. A preferred method, however, is called a "stem-and-leaf" display (Tukey, 1972). This gives all the information of a histogram in addition to retaining the actual numerical values which makes it a simple matter to find the median. The construction of a stem-and-leaf display is illustrated using the following ^{239}Pu concentrations in soil that are displayed in histogram form in Figure 6:

8.2	0.8	2.4	18.2	1.9
9.4	9.0	3.6	3.0	14.3
12.8	3.5	16.4	3.4	7.9
1.7	0.5	4.8	5.6	9.2
7.9	10.7	5.6	11.2	5.9
8.9	2.0	4.4	11.0	2.6
6.7	7.6	5.8	2.5	10.2
10.3	4.4	305.0	21.0	
21.3	3.1	6.8	3.6	
11.3	11.3	8.7	20.0	

The first step is to select a "stem" which corresponds to the intervals of a histogram. For the above data set, units of 10s appear to be a reasonable choice. The stem appears as in column (a) in Table 2. The "leaf" of the display is the next digit of the number, illustrated in column (b) of Table 2 for the first 5 numbers in column 1 of the above data set. Doing this for all 47 numbers gives column (c) in the table. Note that two data with the same stem value appear in the same row, *e.g.*, 21.3 and 21.0. By reordering the leaf values from smallest to largest for each stem, and by adding a depth column, we obtain the final stem-and-leaf display given in column (d). Note that the "leaf" part is just a histogram, but each "bar" of the histogram now contains the actual numerical values of the data. The "depth" column is constructed by counting the number of observations starting at both ends. Thus, the entry at position 7 on the stem contains the median (central observation).

260

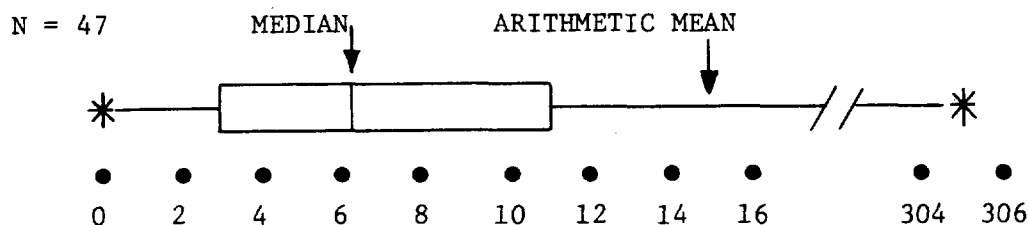
Table 2. Construction of a Stem-and-Leaf Display Using ^{239}Pu Concentrations (nCi/g Dry) in Surface Soil From Stratum 6 of Area 13 (Project 57).

(a)		(b)	(c)		(d)		
Stem	Stem	Leaf	Stem	Leaf	Completed Stem-and-Leaf Display		
					Depth	Stem	Leaf
0	0		0	5 8	2	0	5 8
1	1	7	1	7 9	4	1	7 9
2	2		2	0 4 5 6	8	2	0 4 5 6
3	3		3	5 1 6 0 4 6	14	3	0 1 4 5 6 6
4	4		4	4 8 4	17	4	4 4 8
5	5		5	6 8 6 9	21	5	6 6 8 9
6	6		6	7 8	23	6	7 8
7	7	9	7	9 6 9	(3)	7	6 9 9
8	8	2	8	2 9 7	21	8	2 7 9
9	9	4	9	4 0 2	18	9	0 2 4
10	10		10	3 7 2	15	10	2 3 7
11	11		11	3 3 2 0	12	11	0 2 3 3
12	12	8	12	8	8	12	8
13	13		13			13	
14	14		14	3	7	14	3
15	15		15			15	
16	16		16	4	6	16	4
17	17		17			17	
18	18		18	2	5	18	2
19	19		19			19	
20	20		20	0	4	20	0
21	21		21	3 0	3	21	0 3
304			304			304	
305			305		1	305	
306			306			306	

This data set is clearly asymmetric with a very long tail as a result of the extreme datum 305. This stem-and-leaf display gives the same information as the histogram in Figure 6, but in addition retains the actual numerical values of the data for use in latter calculations. The depth column, which counts the number of data points from each end, is useful for computing summary statistics such as the minimum, 25th percentile, median (50th percentile), 75th percentile, and the maximum. For this data set we find

minimum	=	0.5
25th percentile	=	3.5
median	=	7.6
75th percentile	=	11.0
maximum	=	305

[see Conover (1971) for discussions of the median and other percentiles]. These statistics can be displayed as a "box" plot as follows (the AM is also shown for comparison):



The box area contains the middle 50% of the data (called the interquartile range) and tells us where the bulk of the data lies. Taken as a whole, the plot above gives us information on the center of the distribution, its variability, symmetry (or asymmetry), and the occurrence of outlying values. The stem-and-leaf display in conjunction with this summary plot conveys additional information over that provided by the AM and SE. Additional information such as the GM and other percentiles could also be displayed on the above plot if desired.

To further illustrate the usefulness of the above approach, we display box plots in Figure 7 for the rodent (*Dipodomys microps*) data that were summarized in Figure 3. The AM, GM, SE, and range are displayed in column [A] and the box plots in column [B]. The GMs are seen to be considerably smaller than the AMs, and slightly less than the medians. This was also true for the soil data in Figure 6. The tendency of the GM to be smaller than the median for these data illustrates our feeling that the GM may not give sufficient weight to the larger observations, at least if we take a conservative approach of not wanting to underestimate "average" concentrations. The interquartile ranges in Figure 7 give us additional information on the possible downward shift in Pu concentrations in the GI tract relative to those in pelt. Since the number of samples ($n = 11$) is so small for these three data sets, a stem-and-leaf display does not give as much information as we would like and the estimates of the interquartile range and other parameters are not precise. It is still useful, however, to go through the procedure if for no other reason than it requires a look at each individual datum to see how it relates to the other data in the set.

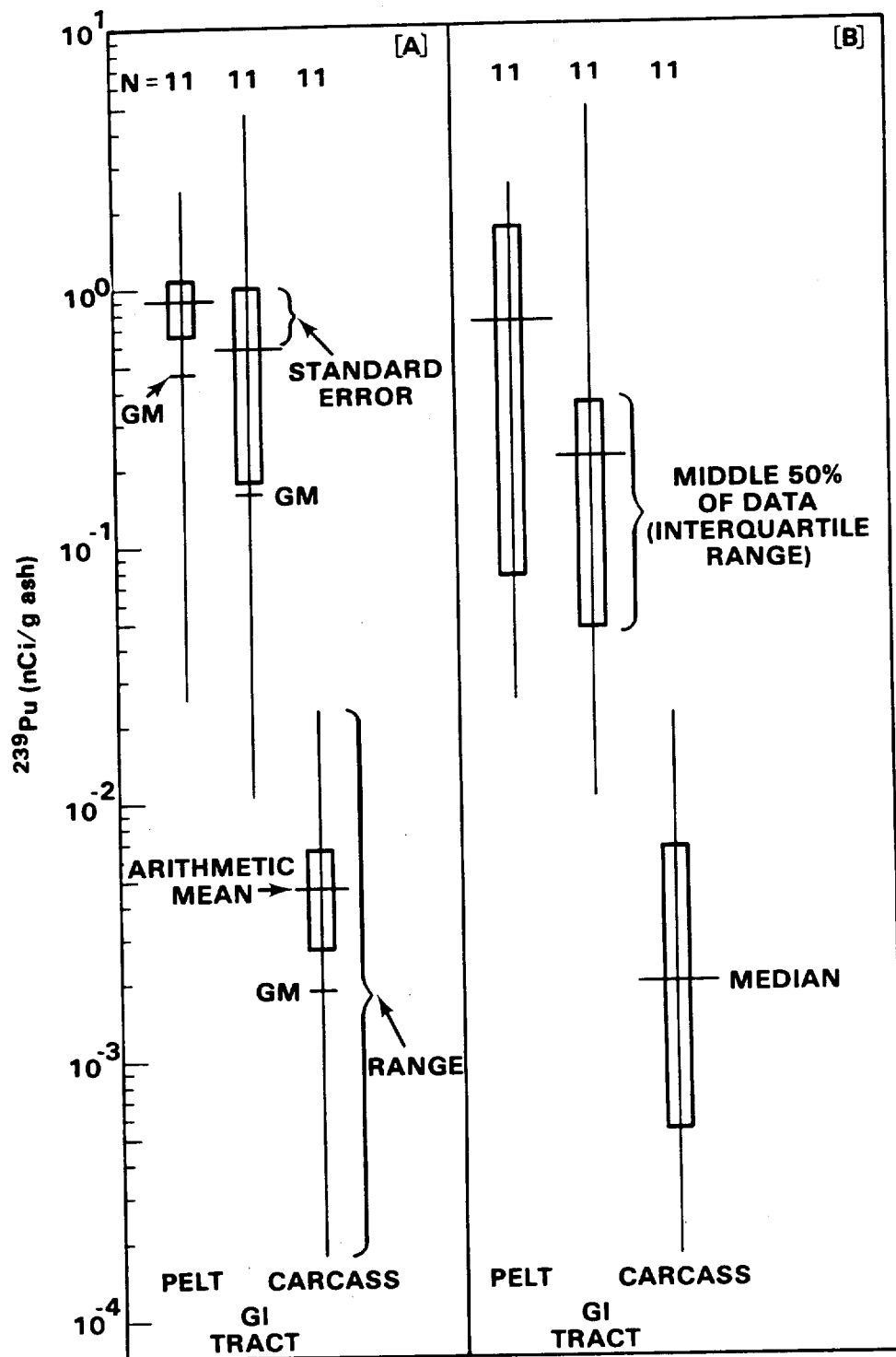


FIGURE 7. Three Sets of Rodent Data (*Dipodomys microps*) Summarized Two Different Ways: [A] Using Geometric Mean (GM), Arithmetic Mean, and Standard Error; [B] Using the Median and the Interquartile Range.

Designing Field Sampling to Estimate Spatial Pattern

Stratified random sampling, where soil samples are collected at random locations within strata (subregions based on ^{241}Am activity), has been used to estimate ^{239}Pu inventory in soil at the safety-shot sites (Gilbert and Eberhardt, 1974; Gilbert *et al.*, 1975, 1976). This design appears to have given more precise estimates of ^{239}Pu inventory than would have resulted if stratification had not been used and soil had been collected at random over the entire study site. However, these data have not proved ideal for estimating the ^{239}Pu concentration "surface" (depicted by contour lines) using computer algorithms since portions of the study area where concentration levels change rapidly were sometimes left unsampled. In these areas, the estimated contours are sometimes biased. For example, the soil Pu contours for GMX site in Area 5 obtained using the computer algorithm NEAR¹² on the computer package SURFACE II (Sampson, 1973) are erroneous west of GZ due to a lack of data immediately west of the GZ bunker (Gilbert *et al.*, 1975, Figures 18, 24, and 25). Gilbert *et al.* (1976, Figure 3; also see Figure 8 in this report) showed that the magnitude of this problem was reduced if the contours were computed on logarithms of the data.

Our experience suggests that we might consider the efficient estimation of the geographical distribution of Pu the primary objective of future sampling efforts at safety-shot or nuclear event sites on NTS. We can think of the concentrations in soil as a continuous three-dimensional surface, the height of which at a particular location gives the Pu concentration in soil at that point (examples are Figures 20, 21, 24, and 25 in Gilbert *et al.*, 1975). It is possible to estimate this continuous surface in units of nCi/m^2 at grid points over the study site using SURFACE II. The inventory could then be estimated by simply summing the grid point concentration estimates and multiplying by a suitable constant to convert nCi/m^2 to nCi . The variance of an estimate of inventory obtained in this manner requires the variance of the estimated concentration at each grid node. It appears that it might be possible to obtain these estimates of variance if the grid node concentrations are estimated using a method called Universal Kriging (Davis, 1973; Delfiner and Delhomme, 1975). One version of this method should be available on SURFACE II (Sampson, 1975) by the fall of 1976. It is not yet apparent whether this approach would yield more or less precise estimates of inventory than stratified random sampling, or indeed, whether Universal Kriging is really suitable for estimating spatial pattern of Pu.

An important aspect of the problem is to identify or develop field sampling designs that are optimum for estimating the spatial pattern or "location" of environmental contaminants such as plutonium. Sampling locations can be chosen many different ways (completely at random, random within strata, on a systematic (grid) pattern, combination of systematic and random, etc.). Gilbert *et al.* (1975, page 419) discuss some of the issues involved in choosing a particular design. Some aspects of the design problem for estimation of

¹²NEAR, as used here, finds the 8 nearest data points (regardless of their orientation or distance) from the point at which the height of the surface is to be estimated.

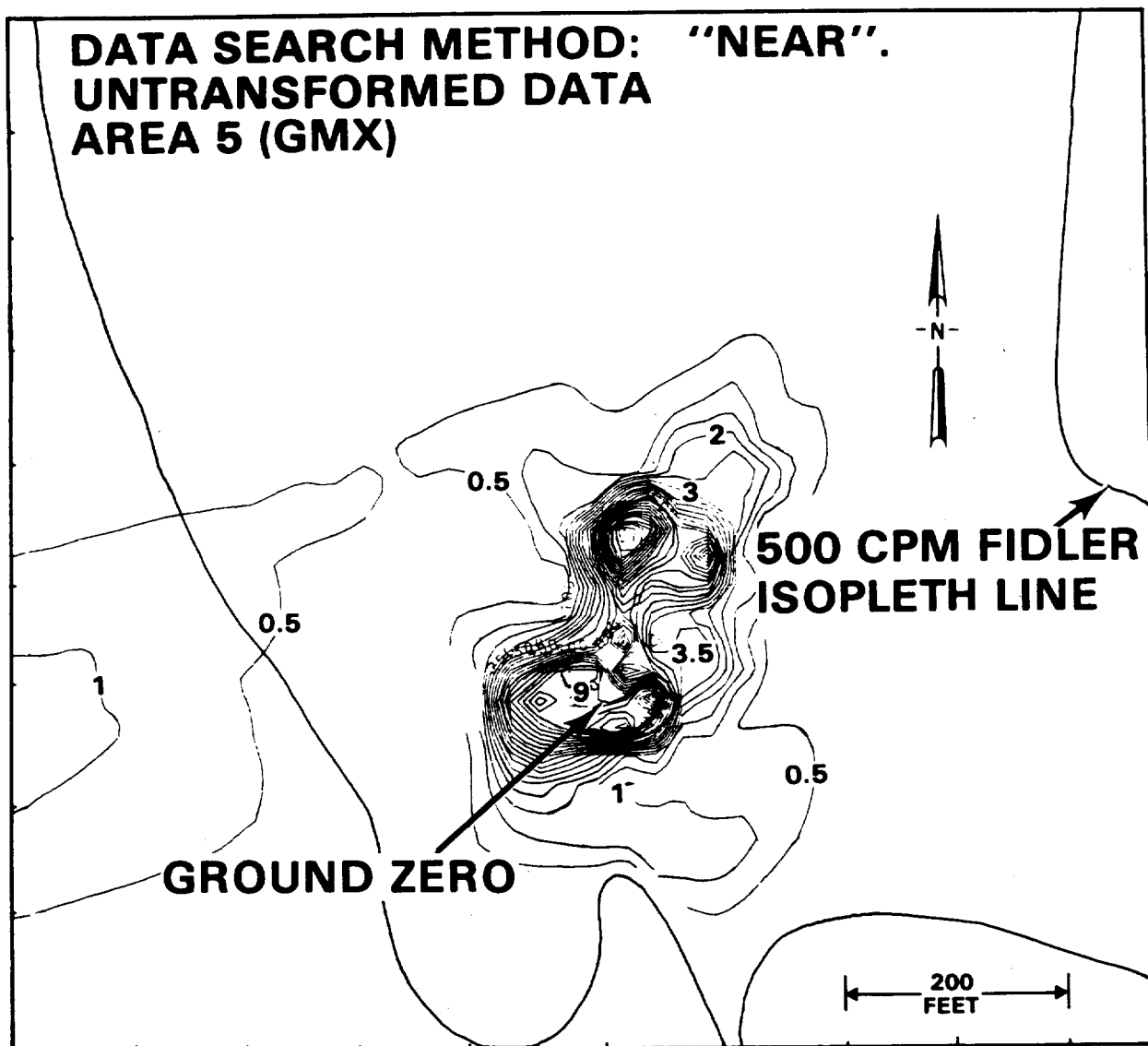


FIGURE 8. Estimated Pu Concentration Contours for Soil, Data Search: "NEAR," Untransformed Data, Area 5 (GMX).

spatial pattern as contrasted with estimating a total were discussed by Eberhardt and Gilbert (1976) at the 1st ERDA Statistical Symposium held at Los Alamos, November 2-5, 1975. The topic was presented as a "problem" for discussion, and many valuable suggestions were offered by the statisticians in attendance, particularly Dr. John W. Tukey of Princeton University and Bell Laboratories. We hope to continue working on this design problem in conjunction with continuing experimentation with computer contouring algorithms.

In preparation for the IAEA/ERDA and the ERDA symposia, we estimated ^{239}Pu concentration contours in surface soil of the GMX site using algorithms QUAD and TREND available on SURFACE II. Since most of these could not be published in the Proceedings of the IAEA/ERDA symposium due to space limitations, we include them here as Figures 9 through 14. All of the contours shown here are in units of nCi (of ^{239}Pu) per gram (dry) of surface soil. Figure 8 shows questionable contours west of GZ obtained using NEAR as mentioned above. Figure 9 was obtained using NEAR on the logarithms of the data. As mentioned above, the use of logarithms eliminated some of the doubtful contours obtained on the untransformed data (Figure 8). More samples must be collected just west of GZ, however, to obtain accurate contours for that region. The algorithm QUAD, which searches for a minimum number of data points in each quadrate within a specified distance¹³ of the point to be estimated, was also applied to the logarithms of the data (Figure 10). Note that contours are not estimated west of ground zero. This occurred because of too few data points in this area. Hence, QUAD can indicate when insufficient data are a problem. Figures 11 and 12 show contours resulting from fitting 4th and 6th degree polynomials to the logarithms of these GMX soil data using the algorithm TREND. These contours show considerably less detail than those obtained above using NEAR or QUAD. The 6th degree fit to the data is somewhat better than that provided by the 4th degree equation. However, the black line along the left margin of the 6th degree fit (Figure 12) indicates severe edge effects. That is, the regression equation is giving grossly inaccurate estimates in this area where no data were collected. Figure 13 shows contours obtained by using TREND to fit a 6th degree polynomial to the untransformed data. Note that the area included within the 0.5 nCi/g contour is much larger for the original scale (Figure 13) than for the log-transformed scale (Figure 12). Serious edge effects are also present in Figure 13. These are plotted in Figure 14 to illustrate the wild Pu estimates obtainable using polynomial fits in regions of the study site where few data have been collected.

Of the contour methods applied thus far to the soil Pu data collected via simple random sampling within strata, it appears that QUAD or NEAR applied to the logarithms of the data are the most promising (see Gilbert *et al.*, 1976, Table III for a statistical evaluation). Improved estimates appear to result if an iterative fitting procedure is applied to the residuals of previous (log) fits. This iterative approach was suggested by Tukey (see discussion after paper by Eberhardt and Gilbert (1976)) and has been investigated in part by Gilbert (1976) using NEAR on SURFACE II.

¹³For Figure 8, we specified (a) maximum distance to nearest data point must be ≤ 100 feet, (b) maximum search radius = 200 feet.

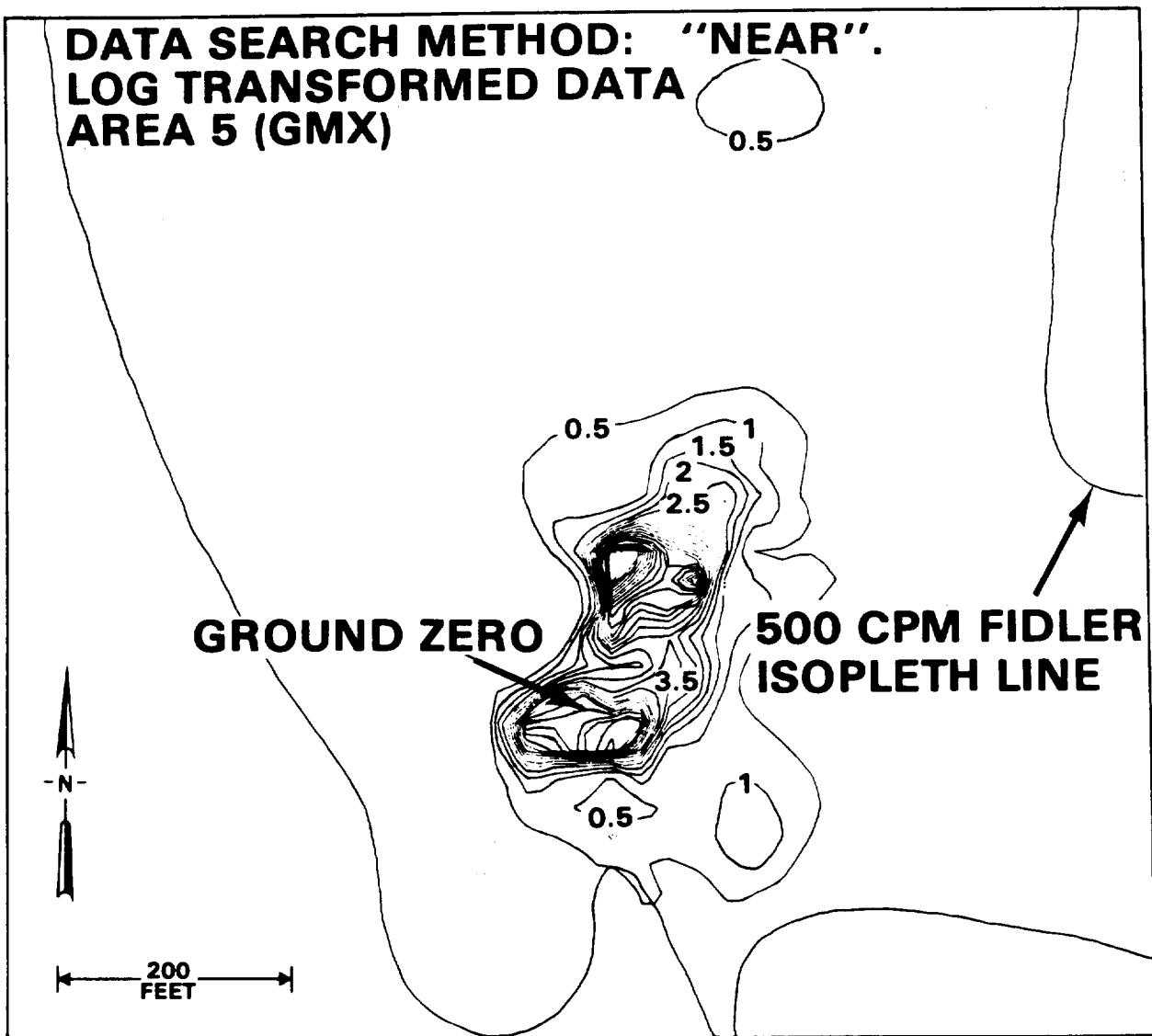


FIGURE 9. Estimated Pu Concentration Contours in Soil, Data Search Method: "NEAR," Log-Transformed Data, Area 5 (GMX).

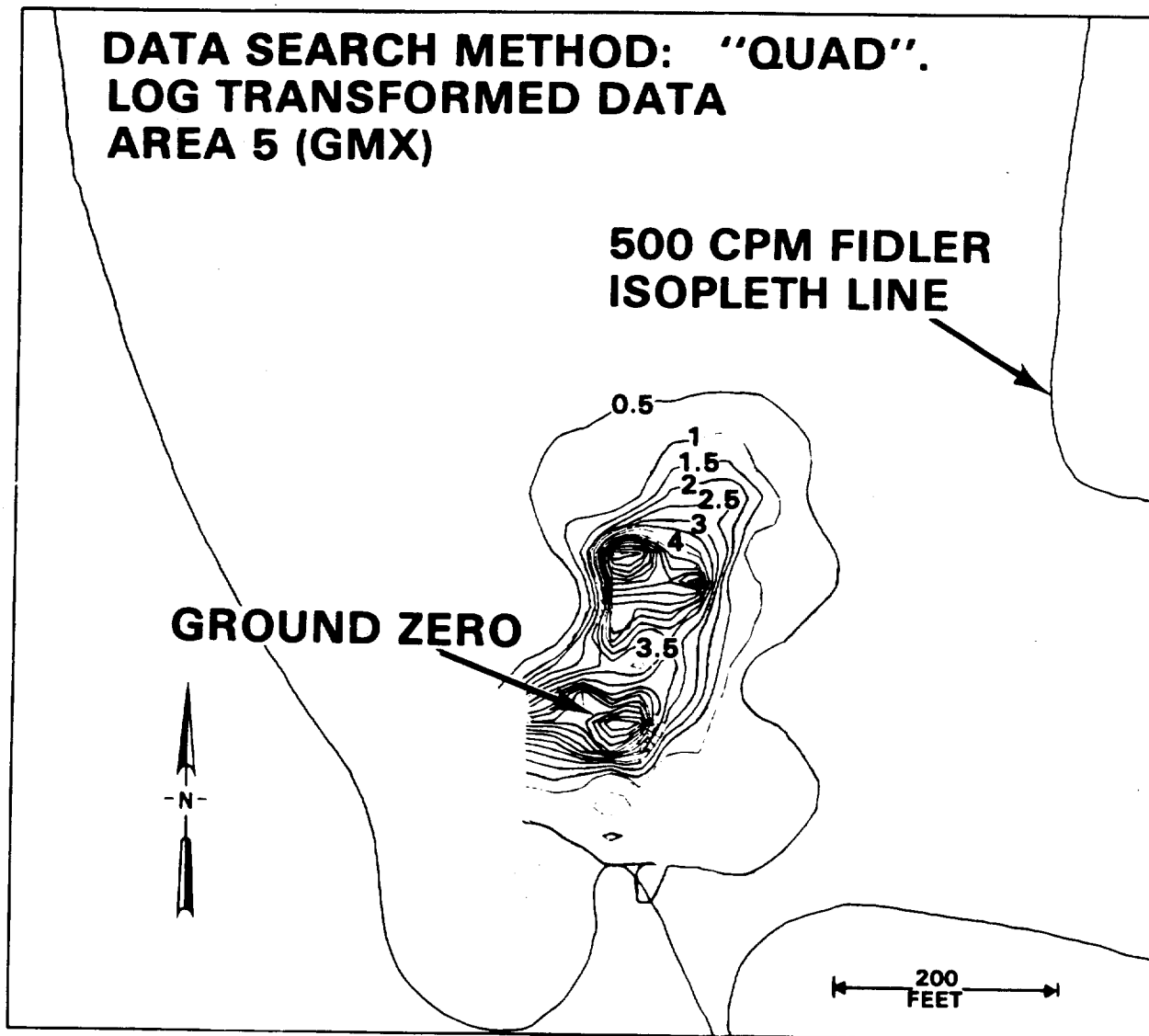


FIGURE 10. Estimated Pu Concentration Contours in Soil, Data Search Method: "QUAD," Log-Transformed Data, Area 5 (GMX).

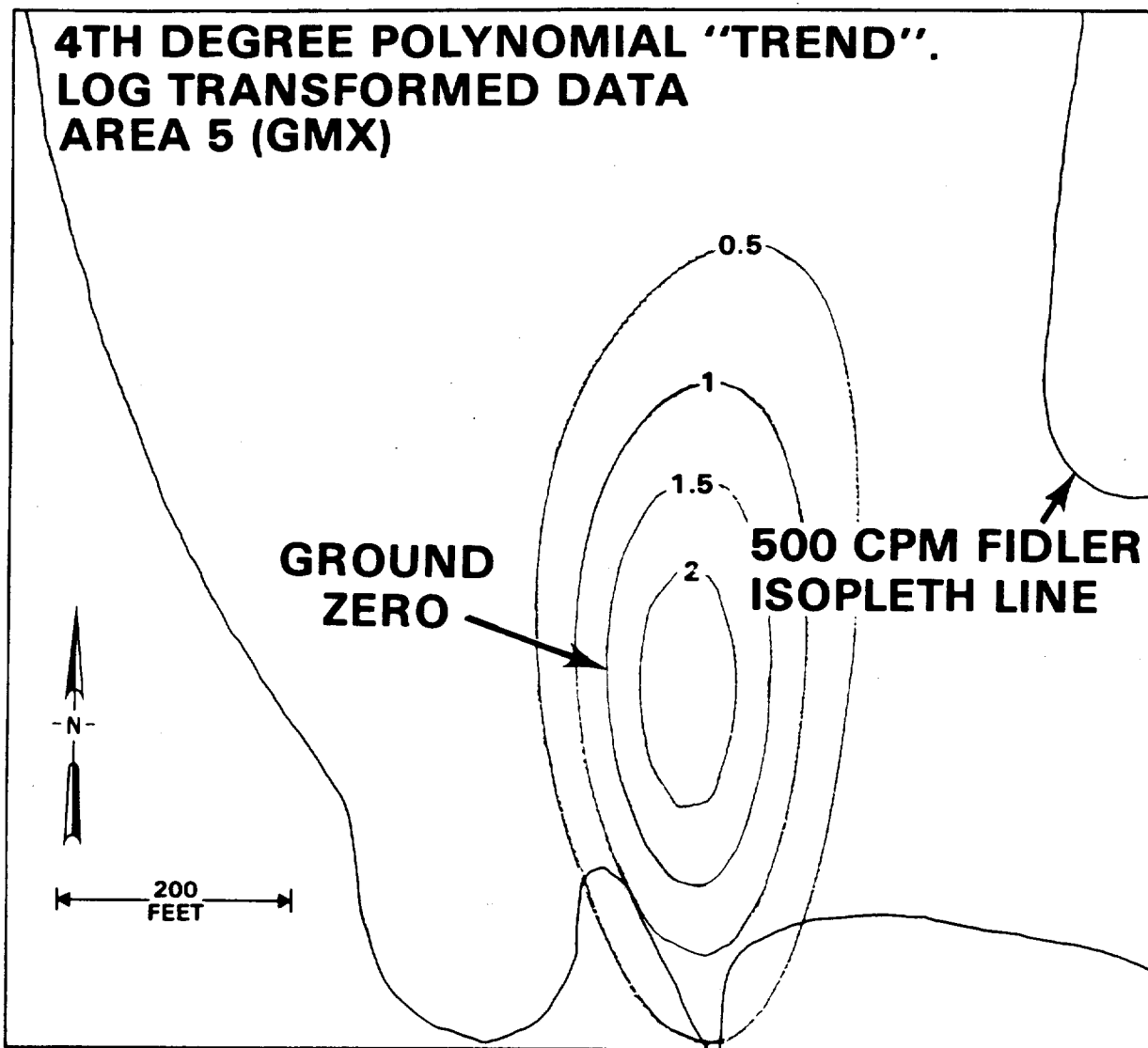


FIGURE 11. Estimated Concentration Contours for Soil, 4th Degree Polynomial "TREND," Log-Transformed Data, Area 5 (GMX).

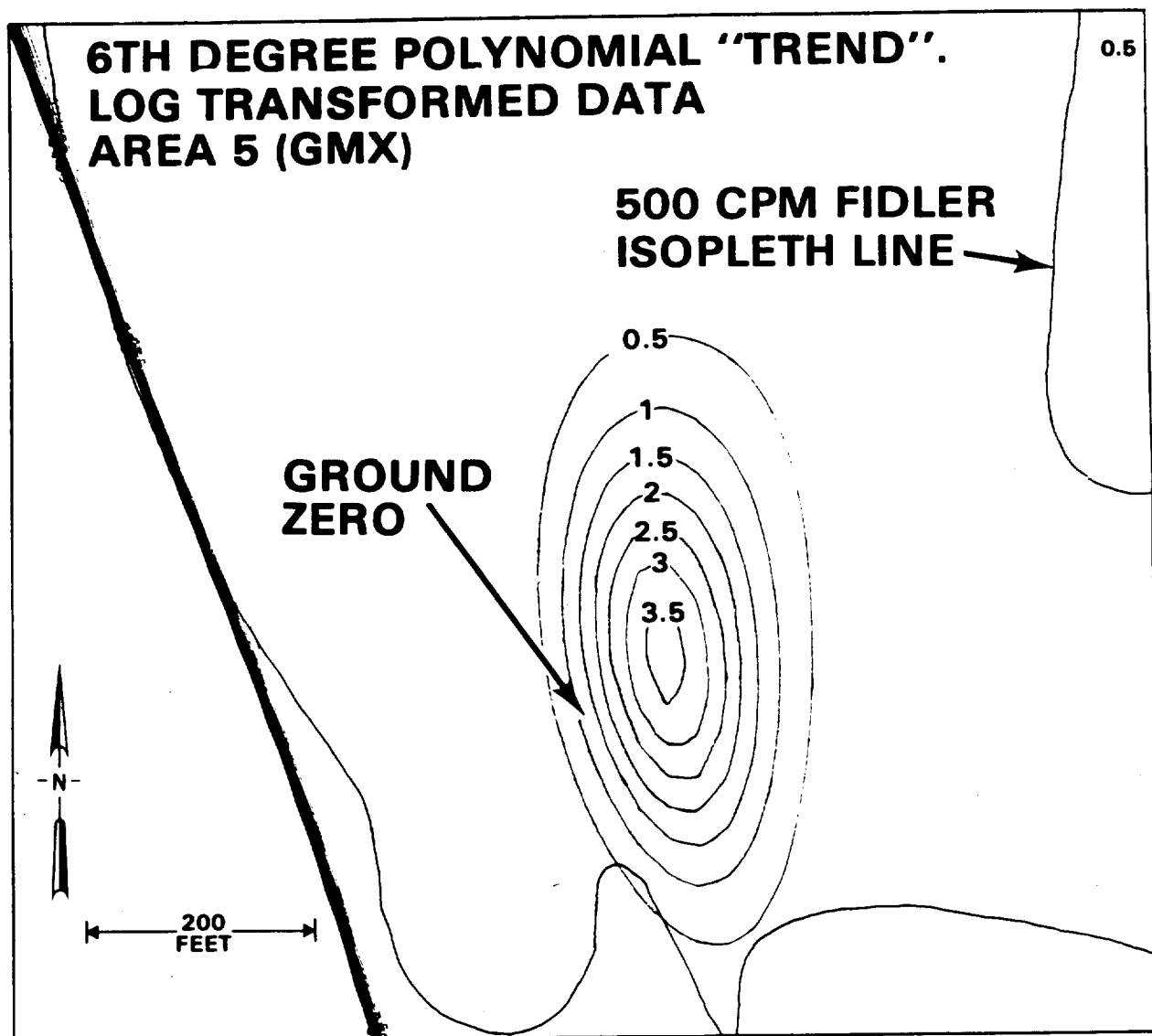


FIGURE 12. Estimated Pu Concentration Contours for Soil, 6th Degree Polynomial "TREND," Log-Transformed Data, Area 5 (GMX).

2.10

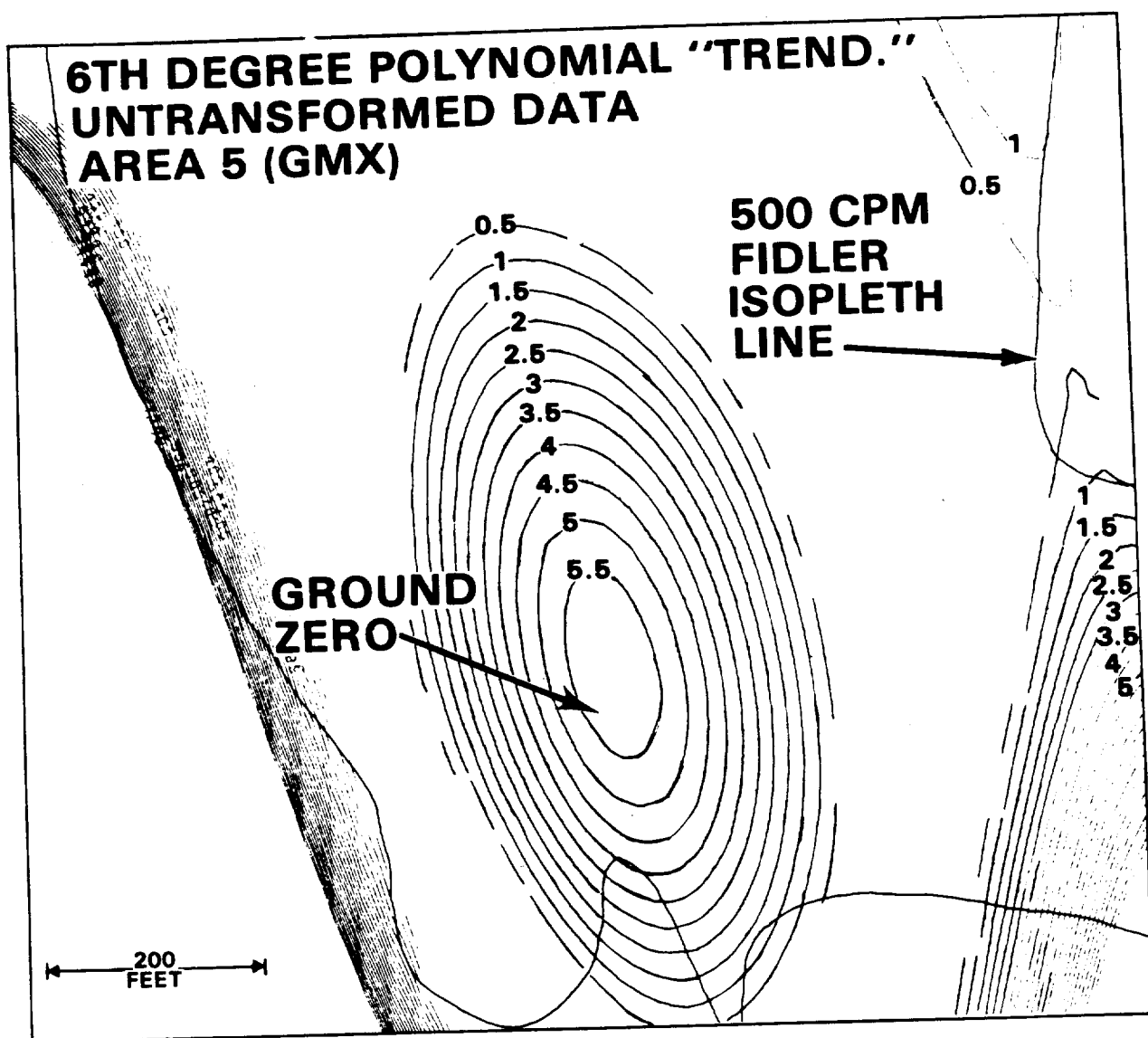
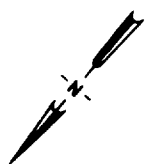


FIGURE 13. Estimated Pu Concentration Contours for Soil, 6th Degree Polynomial "TREND," Untransformed Data, Area 5 (GMX).

AREA 5 (GMX)



500 FEET

FIGURE 14. Estimated Pu Concentration Surface for Soil, 6th Degree Polynomial "TREND," Untransformed Data, Area 5 (GMX).

It appears at this point, however, that Universal Kriging may be the preferred method since in theory it should yield more accurate estimates of contours than any other approach, at least when underlying assumptions are satisfied. Also, as mentioned above, it provides estimates of variability not available using any other method.

SUMMARY

In the first part of this paper, we have attempted an initial synthesis of the $^{239,240}\text{Pu}$ concentration data currently available from Area 13 (Project 57) on the Nevada Test Site for soil, vegetation, small mammals, and grazing cattle. For the most part, this synthesis has consisted of simple visual comparisons between different trophic levels by plotting all the data on a single graph.

The soil and vegetation data are seen to be highly variable within strata, but with strata means increasing with increasing proximity to GZ. This large variability also exists in small mammal tissue concentrations. There is a lack of correlation between rodent concentrations and concentrations in the soil where animals live, due perhaps in part to the mobility of rodents. $^{239,240}\text{Pu}$ concentrations in cattle tissue are also plotted and compared with other trophic levels. These comparisons are hampered by the small number of cattle tissue samples available for statistical analysis. The lowest and highest concentrations were found in muscle and hide and hair, respectively, with lung concentrations falling between these extremes. The single cow grazed in the inner compound (enclosing the GZ area) had concentrations higher than the average for the three cattle grazed in the outer compound for hair and hide, tracheo-bronchial lymph nodes, lung, muscle, and backbone. Equal or lower concentrations were indicated for hide, rumen sediment, red blood cells, femur, and blood serum. Again, we stress the need for more data, particularly for the inner compound, to increase our confidence in these initial comparisons. The concentrations in lungs of the Area 13 cattle are 2 to 3 orders of magnitude greater than for cattle grazed in a control area (Searchlight, Nevada). As for muscle, there are no apparent differences between the outer-compound cattle in Area 13 and the control herd. The muscle data from the one cow grazed in the inner compound suggest a possible elevation of about one order of magnitude above the control herd for that tissue.

Observed muscle $^{239,240}\text{Pu}$ concentrations in Area 13 cattle were compared with hypothetical concentrations obtained using Martin and Bloom's plutonium transport and dose estimation model. Their model gave estimates from 2 to 4 times larger than actually observed, suggesting, perhaps, that their model as presently constituted may yield conservative estimates for cattle. Hypothetical human concentrations were also obtained in Area 13 for residence times equal to cattle grazing times before sacrifice. The hypothetical values for human lung and bone were factors of 5 and 8 less than obtained for the single cow in the inner compound. Hypothetical concentrations are also obtained for residence times of 180 days, 720 days, and 55 years for an estimated "maximum permissible concentration" of 3 nCi/g dry of $^{239,240}\text{Pu}$ in soil.

An estimate of the average intake of $^{239,240}\text{Pu}$ per day by a cow grazing the inner compound is computed from average $^{239,240}\text{Pu}$ concentrations for shrubs *Eurotia lanata* and *Atriplex canescens*. This result (620 ± 130 nCi/day) agrees quite well with the 560 nCi/day estimated by Smith *et al.* (1976a) using average $^{239,240}\text{Pu}$ activities in the vegetation and liquid components of rumen contents of fistulated steers grazing the inner compound of Area 13. An estimate of 770 nCi/day was obtained using Martin and Bloom's model assuming 500 grams of soil is ingested per day by grazing cattle.

The second part of this paper discusses some aspects of displaying and analyzing skewed data sets. Some relationships between the arithmetic mean, geometric mean, median, and mode are discussed. It is shown that estimates of inventory can be greatly influenced by the particular measure of central tendency (average) used to characterize a skewed data set. The point is made that the choice of a measure of central tendency depends on the objective of the study and the use to be made of the estimate. Stem-and-leaf displays for displaying the information content of a data set are discussed and illustrated using soil and rodent plutonium concentrations.

The third and final portion of this paper reviews some of the bias problems we have encountered in attempting to estimate plutonium concentration contours in Area 13 (Project 57) and Area 5 (GMX site). The possibility of estimating inventory by first estimating the spatial pattern of plutonium concentrations in surface soil is suggested. An evaluation of Universal Kriging for this purpose is encouraged. Finally, some estimated contours for Area 5 (GMX) are displayed to illustrate what appears to be improved estimates using log-transformed data. Contours obtained using polynomial fits are shown to be susceptible to large biases in areas of the study site where few data were collected.

FUTURE PLANS

We expect to:

1. Continue to provide statistical help in the synthesis of safety-shot data for ^{239}Pu and for other radionuclides (^{238}Pu , ^{241}Am , ^{238}U).
2. Continue our assistance to program investigations in the analysis of their data.
3. Help design environmental transuranic studies at nuclear sites on NTS.
4. Identify or develop efficient field sampling designs for estimating spatial patterns (geographical distribution).
5. Continue to experiment with estimating Pu concentration contours using computer algorithms.

6. Develop or identify the most appropriate ways of analyzing, summarizing, and reporting data for maximum communication of content.

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SUPPORT ACTIVITIES

INTERIM REPORT OF LFE ANALYTICAL SERVICES FOR NAEG

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Since the October 1974 meeting of the Nevada Applied Ecology Group, LFE has analyzed several hundred samples for various radioactive elements in a wide range of matrices. Radiochemical and instrumental analysis were performed on such samples as field and greenhouse vegetation, soils, glass fiber and micro-sorban filters, rodents, bovine and caprine tissues and bone, and desert mammals. Radioisotopes determinations included ^{238}Pu , $^{239,240}\text{Pu}$, ^{241}Am , ^{210}Pb , ^{55}Fe , ^{0}Fe , ^{90}Sr , $^{230,232}\text{Th}$, total and isotopic U, and ^{226}Ra . The data generated was reported routinely to the NAEG for evaluation.

A special computer program was developed for data reduction and reporting of bovine and caprine numbers. The program allowed for computing and reporting the sample radioactivity on a wet, dry, and ash weight basis. A complete animal description, weights, ratios, and pertinent radiochemical data were included in the printout. A similar computer program was developed for soils, vegetation, and rodent animals.

A special test was performed at the Nevada Applied Ecology Group laboratory at Jackass Flats, to determine the transuranium content of large-size, high-level bovine and caprine tissue samples. The lab had been reconstructed by NAEG and REECO personnel to accommodate analysis of these samples and to confine any contamination to a remote area. In order to avoid spread of contamination and speed up dissolution of the samples, a wet ashing method was developed and employed on spiked samples of large tissues. The method consisted of spiking kilogram samples of liver, muscle, spleen, and bone with ^{60}Co , analyzing them using the special wet ashing techniques and classical radiochemical methods. Details of a similar procedure are attached.

A RAPID DISSOLUTION TECHNIQUE FOR TISSUE SAMPLE
ANALYSIS OF TRANSURANIUMS

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ABSTRACT

A rapid dissolution technique for the analysis of transuraniums in soft animal tissue samples has been developed. The sample is dissolved, tracer-free, in HNO_3 in the presence of HF while being warmed on a boiling water bath in a disposable plastic bottle. After the tissue is solubilized, boric acid is added to boil off fluoride ion. Fat separates on cooling and is filtered off, ashed, dissolved, and combined with the original sample.

The solubilized tissue sample may then be aliquoted for transuranium analysis. The aliquot is equilibrated with tracer by boiling in HNO_3 with added H_2O_2 . If Pu is being analyzed, the sample is cooled and NaNO_2 added so that the final solution is 6-8N HNO_3 and ready for an anion column separation of Pu and Np from Am and Cm. The analyses are then completed by the appropriate method.

The method is most suitable for samples in the 250-g size category. Larger samples can be done in increments. The dissolution is accomplished in about one hour.

The method is particularly suited to the analysis of samples from injection or uptake by animals. The dissolution puts the sample into a soluble form which is most useful for obtaining a uniform counting geometry for instrumental techniques or taking a small aliquot for subsequent isotope dilution analysis.

INTRODUCTION

In recent years, a number of methods have been devised for ashing and dissolving large tissue samples for analysis of Pu, Am, and other nuclides. Two obvious methods are furnace ashing plus acid treatment of the ash and direct wet ashing of the sample. Presently, many tissue samples at our laboratory are processed by a previously reported procedure (Major *et al.*, 1975) in which the

sample is ashed and then treated with HF, HNO₃, and H₃BO₃ so as to completely dissolve the transuranium elements.

Over the years, various methods have been developed for the wet ashing of large tissue samples. A combination of HNO₃, F-HNO₃, and HClO₄ has been used successfully but is time consuming and requires almost constant attention (NCRH, 1967). Also, insoluble (NH₄)₂ ClO₄ salts interfere with the dissolution. Another method (Major and Wessman, 1964) utilizing a reflux action with H₂SO₄ and an Hg catalyst was developed but was still time consuming. B. Sansoni and Kracke (1971) reported a very adequate method of using H₂O₂ in the presence of Fe⁺⁺ ion.

The method reported here is simple and economical, particularly for large high activity samples. The method offers a self-contained easy contamination control as shown in the procedure schematic Figure 1.

PROCEDURE

Cut the wet soft tissue sample (up to 250 g) into small cubes for dissolution. If a dry weight is required, then dry the sample in an oven at 105° C for 24 hours. Weigh the dried meat and repeat the drying and weighing at 6 hour intervals until a constant weight is obtained. In some cases, the ash content of the sample is required. It is suggested that a representative aliquot of the wet sample be taken for ash determination and that the ash be dissolved and recombined with the original sample.

Transfer the sample to a 2-liter wide-mouth poly bottle. The reacting solution may foam over if too much meat is used. Add an equal weight of HNO₃ and 10 ml of HF to the bottle. CAUTION: Cap the bottle but be sure it is loose so that the gases may escape during the reaction. Place the bottle in a hot water bath and heat on a hot plate until all the foaming reaction stops. Add 10 ml saturated H₃BO₃, and continue heating for 20 minutes. Transfer the solution into a glass beaker and wash the bottle with hot 6N HNO₃. Any fat will float to the top upon standing at this time and the aqueous phase should be clear. If a smaller volume is required, evaporate the solution to a smaller volume. Do not evaporate to dryness since some organics are present and the solution will turn into a black tar. Cool the sample to solidify the fat and filter it through a No. 541 Whatman filter paper. In some cases, the fat may be discarded at this point. Otherwise, ash the filtered fat in a 425° C furnace and dissolve the ash with HNO₃, HF, and H₃BO₃. Combine the resulting solution with the filtrate. Dilute the sample to 1,000 gram with 6N HNO₃.

The solubilized tissue sample is aliquoted for transuranium analysis. The aliquot is equilibrated with tracer by boiling in HNO₃ with added H₂O₂. If Pu is being analyzed, the sample is cooled and NaNO₂ added so that the final solution is 6-8N HNO₃ and ready for an anion column separation of Pu and Np from Am and Cm. The analyses are then completed by the appropriate method.

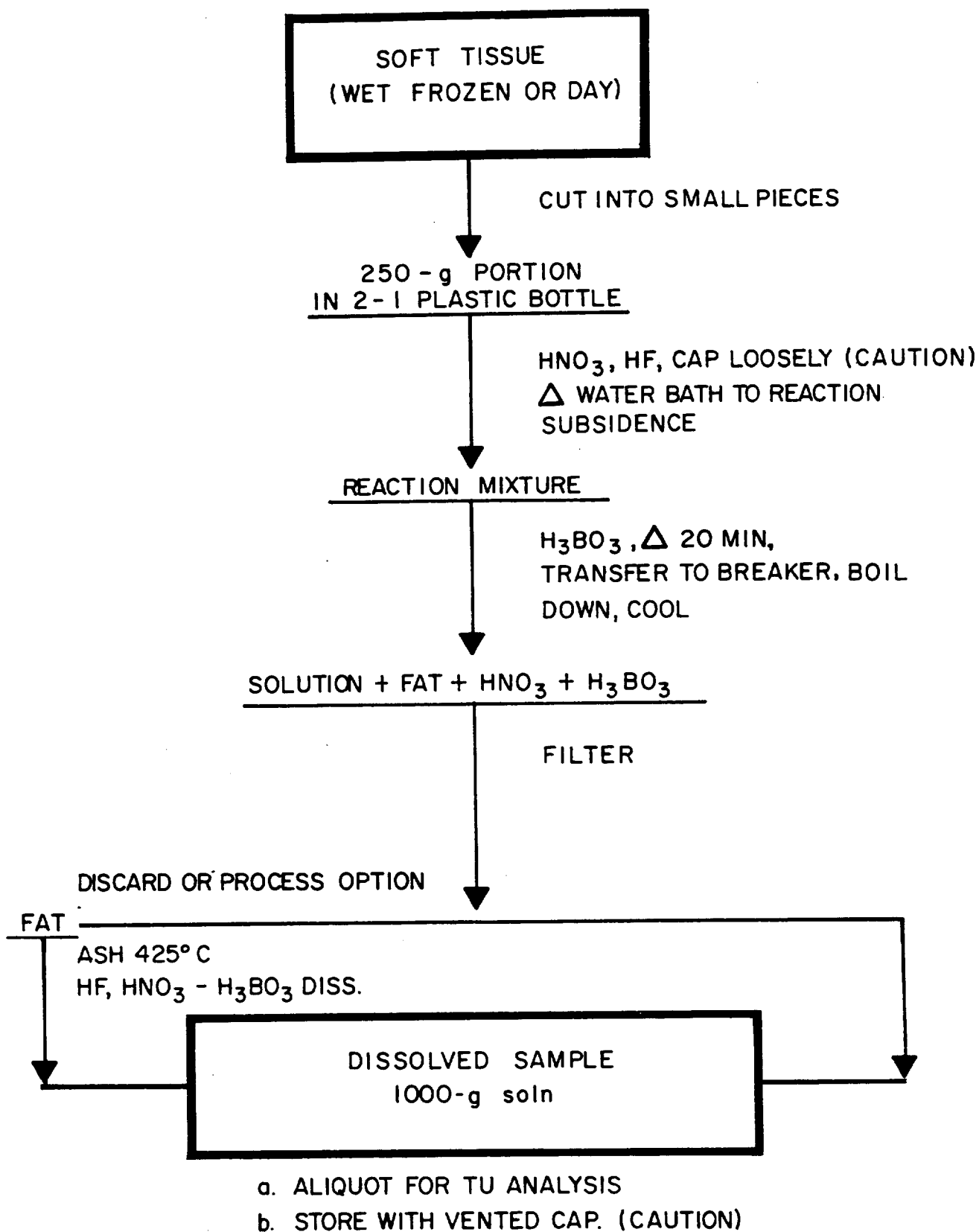


FIGURE 1.
WET DISSOLUTION OF SOFT TISSUE

DISCUSSION

An experiment was performed with a spike solution of ^{237}Pu , ^{241}Am , and ^{237}Np to determine the activity distribution in the fat and filtrate portions. The results show less than 1% of the activity in the fat after the cold 6N HNO_3 wash. These results might be expected since the spike solution has not been through metabolism mechanics. Four samples from an inhalation metabolism experiment for ^{241}Am in tissues were analyzed to determine the ^{241}Am content of the fat portion. The sample was dissolved, filtered, and ashed by the method described. The filtrate and fat portion were counted separately. The results are given in Table 1. In the four tests, the amount of ^{241}Am remaining in the fat portion ranged from 0.1 to 0.4% of the sample activity. For most applications, the small amount of ^{241}Am retained by the fat may be considered negligible and the fat could be discarded. To date, in our laboratory, we have adopted a conservative approach and processed all fat portions and combined them with the sample.

To determine the ability to analyze the dissolved sample and recover the ^{241}Am activity, seven metabolism tissue samples containing ^{241}Am were dissolved. The dissolved samples were assayed for ^{241}Am in a standardized jar counting geometry using a thin layer NaI (Tl) crystal as described by Major *et al.*, (1974). Aliquots, ranging in size from 0.01% to 5.0% of the dissolved sample, were analyzed for ^{241}Am by the isotope dilution procedure previously reported (Major *et al.*, 1974). Americium-243 tracer and alpha spectrometry are used. The results (Table 2) show satisfactory recovery of ^{241}Am from solution of various tissue types and activity levels, and there is no apparent bias within the limits of the procedures used.

In summary, the described dissolution procedure for tissue samples using HF and HNO_3 appears to be satisfactory for its intended purpose. Using this procedure, samples of 250 grams can be completely solubilized in less than an hour. The advantages of this procedure are:

- a. Equipment requirements such as charring and ashing furnaces and exhaust facilities are reduced.
- b. Ashing odors are reduced.
- c. Labor to tend ashing samples is reduced.
- d. Cost savings on laboratory ware and reagents is achieved.

The procedure is expected to be adaptable to the analysis of large, low-level samples, such as autopsy samples.

Table 1. Distribution of Americium-241 Between Filtered Fat and Filtrate of Dissolved Metabolism Tissue

Tissue	Sample FRACTION	^{241}Am in SAMPLE FRAC. (dpm)	$\frac{^{241}\text{Am (Fat)}}{^{241}\text{Am (Soln)}}$
Liver	Fat Filtrate	8.0×10^2 2.0×10^5	0.0040
Liver	Fat Filtrate	1.5×10^1 1.3×10^4	0.0012
Liver	Fat Filtrate	3.7×10^2 2.2×10^5	0.0017
Liver	Fat Filtrate	4.6×10^2 2.0×10^5	0.0023
		MEAN	0.002

Table 2. Recovery of Americium-241 From Dissolved Tissue

Tissue	Sample Wt. Range (g)	Added (a) (dpm)	Recovered (b) (dpm)	Deviation %
Liver	100-500	300 ± 30	341 ± 17	(-) 12.0
Nose	<50	484 ± 29	456 ± 41	8.5
Paw + Tail ^(c)	100-500	1,290 ± 52	1,260 ± 38	2.4
Liver	100-500	1,690 ± 17	1,690 ± 101	-0-
Lung	<50	2,600 ± 52	2,320 ± 46	(-) 11.0
Skel. Rem.	100-500	3,570 ± 71	3,740 ± 75	4.8
Paw + Tail ^(c)	100-500	10,900 ± 109	11,700	11.6
-----		Mean Deviation, Sign Neglected		7.2
		Mean Deviation, Sign Included		0.6

(a) Added--from gamma spec assay of total.

(b) Recovered--isotope dilution analysis from 0.01 to 5% aliquot of solution.

(c) Samples with bony material are dry ashed and dissolved.

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INTERACTIONS OF THE NAEG INFORMATION CENTER
WITH OTHER PROJECTS

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ABSTRACT

In the past year, the Information Support Project to the Nevada Applied Ecology Group (NAEIC) has interacted with many other research projects on the transuranics and other radionuclides. Group interactions through symposiums, workshops, and responding to search requests have proven to be mutually beneficial. The NAEIC will draw on the information resources of the Oak Ridge National Laboratory to produce a bibliography of the radionuclides (other than the transuranics) of interest to the Nevada Test Site.

PROGRESS

The major objective of the Information Support Project to the Nevada Applied Ecology Group (NAEIC) is to inform NAEG contractors of all the developments and techniques in their areas of interest. In the past year, the NAEIC has, while pursuing this objective, interacted with many other research projects on the transuranics and other radionuclides. Mutual benefits arising from these interactions have accrued to both groups, especially in the case of conference and workshop interactions. The staff of the NAEIC has relayed to the appropriate NAEG contractor, as often as possible, the information gained at conferences. We inform the NAEG contractor when we hear about a presentation at a meeting or a study being done that seems to coincide with the contractor's interests. We either send the contractor a preprint of the presentation or we offer to send the pertinent information so that the NAEG contractor can contact other researchers in his field. In the process of providing the most up-to-date information, we have found all NAEG researchers did not receive announcements

*Operated by Union Carbide Corporation for the Energy Research and Development Administration.

of meetings with presentations related to their fields because they did not belong to the society sponsoring the meeting. The computerized data base of the Information Support Project has provided a mechanism for generating distribution lists for conferences on transuranium elements since it can be machine searched to generate a directory of current researchers. The names of the NAEG personnel and contractors will always be on Information Support Project's generated distribution lists.

During July and October of last year, the NAEIC support capability was demonstrated at two workshops with a computer terminal connected via dial-up telephone lines to the Oak Ridge National Laboratory IBM 360/75. The terminal utilized an on-line search and retrieval program to access the data base on the Environmental Aspects of the Transuranics. These two workshops were: (1) the National Commission on Radiation Protection (NCRP) Committee #31 (Plutonium) Workshop held in Seattle, Washington, and (2) the Workshop on the Biological Effects and Toxicity of Plutonium 239 and Radium 226 held in Sun Valley, Idaho. These demonstrations gave non-NAEG researchers and decision makers the benefit of an information resource that had been directed and supported by the NAEG for the preceding four years. The quick response of the computer terminal and breadth and depth of the data base on the Environmental Aspects of the Transuranics were impressive to those new to automated, specialized data bases. This was the first time an on-line information resource had been used as a time-saving device at a committee meeting at which standards were being formulated.

The NAEIC has been informing NAEG researchers of requests in their areas of interest. This is a means of keeping the NAEG investigators informed of new projects being proposed and new research being initiated as well as giving the NAEG researchers an opportunity to help the Information Center to answer the requests with more current and detailed data. Responding to search requests is another way information support groups interact.

The NAEIC draws on the information resources of the Information Division and the Information Center Complex of Oak Ridge National Laboratory (ORNL) for selection of documents for inclusion in the data base on the Environmental Aspects of the Transuranics. Table 1 gives the data bases available for searching at ORNL. We select new additions of interest to the NAEIC by use of a profile or search procedure stored in the computer to search new magnetic tapes of these data bases available at ORNL. Use of these large-scale, automated data bases is only one of the ways the data base selects new documents. Being on the distribution lists for new documents is a much quicker way because the documents are available to be entered into the data base much sooner.

Several nontransuranium radionuclides on the Nevada Test Site will be considered by the NAEG in the near future in relation to decontamination efforts. The radionuclides so far identified for which information is desired are: cobalt 60; ruthenium 102; silver 108; antimony 125; cesium 137; europium 152, 154, and 155; zinc 65. A draft bibliography of these nuclides in relation to soil movement, plant uptake, and resuspension has been compiled by the NAEIC using a variety of computerized data bases. Table 2 gives a list of the computerized data bases built by the Ecological Sciences Information Center from which this bibliography was drawn. The large-scale, automated data bases available at

Table 1. Data bases available for searching at ORNL

<u>Non-ORNL data bases</u>
Nuclear Science Abstracts
National Agricultural Library Data Base
Government Research and Development Reports (NTIS)
Biological Abstracts
Chemical Abstracts
Bio-Research Index
Searchable Physics Information Notices
<u>Information Center Complex data bases</u>
Toxic Materials Information Center Data Base
Environmental Mutagenesis Data Base
Energy Abstracts for Policy Analysis
Energy R and D Inventory Data Base

Table 2. Data bases built by the Ecological Sciences Information Center in the subject area of radioecology

Date base	Years of literature covered	Abstracted	Indexed
Environmental Aspects of the Transuranics	1948 - 1976	yes	yes
Radionuclide Movement in Soils and Uptake by Plants	1948 - 1975	yes	yes
Klement & Schultz Radioecology Bibliographies	1962 - 1974	no	yes
Personal Literature Collections of ORNL Environmental Scientists & Publications by ESD Researchers	1956 - 1974	yes	yes
Behavior of U and Th in Th Fuel Cycle	1941 - 1975	yes	yes

ORNL were searched as well. This bibliography and the machine-searchable data base compiled as a basis for the bibliography are a logical foundation for a more extensive effort and may provide the literature background for making research decisions.

Our selection of material for the data base depends on research plans for the coming year as well as present interests. Please let the NAEIC at ORNL know when your research direction changes or when you anticipate new information needs, in order that the NAEIC can be ready with complete answers when you need them. The information from documents is abstracted with the needs of the NAEIC contractors in mind. Each year more detailed data is abstracted for you as we become more aware of your anticipated information needs.

PLANNED ACTIVITIES

The new data base on the radionuclides other than transuranics on the Nevada Test Site will be expanded to meet the needs of the NAEIC, and input of information to the data base on the Environmental Aspects of the Transuranics will continue. The fifth and sixth bibliographies of the data base on the Environmental Aspects of the Transuranics were published in 1975 and the seventh is in press. Two additional bibliographies are planned for this fiscal year. Special bibliographies on resuspension phenomena, decontamination methods and effects, and/or transport of the transuranics in the terrestrial environment are possible if the need is shown. The Nevada Applied Ecology Information Center will continue to keep the interests of the NAEIC as a first priority, but this service is contingent on our awareness of current interests.

REECO ACTIVITIES AND SAMPLE LOGISTICS
IN SUPPORT OF THE NEVADA APPLIED ECOLOGY GROUP

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ABSTRACT

The logistics in support of the collection, preparation, and shipment activities by REEC Co related to Nevada Applied Ecology Group (NAEG) soil, vegetation, and animal samples are discussed. These samples were taken from the NAEG plutonium intensive-study areas at the Nevada Test Site and the Tonopah Test Range from March 1, 1975, through December 31, 1975.

REEC Co activities in support of the collection, preparation, and analysis related to the Plutonium Inventory and Distribution Element (PIDP) are discussed. The PIDP soil samples were collected from Areas 5, 1, and 4 of the NTS during the period from January 1, 1975, through December 31, 1975.

DATA SUMMARY

During the period March 1, 1975, through December 31, 1975, the following NAEG sample logistics activities took place (see Table 1):

Table 1. NAEG Sample Logistics Activities

Sample Type	Number of Samples Collected (1975)	Number of Aliquots		
		Prepared		Shipped For Analysis (1975)
		1975	Total to Date	
Soil	479	653	3,315	191
Vegetation	97	251	1,415	267
Animal	---	553	1,217	847

Four hundred seventy-nine (479) soil samples were collected, 653 aliquots were prepared, and 191 aliquots were shipped to the four laboratories participating in NAEG analyses.

Ninety-seven (97) vegetation samples were collected, 251 aliquots were prepared, and 267 aliquots shipped to the analysis laboratories.

Five hundred fifty-three (553) animal aliquots were prepared, and 847 aliquots were shipped to the analysis laboratories.

The total number of NAEG intensive-study area soil aliquots which have been prepared for radiological analysis as of December 31, 1975, is 3,315; vegetation aliquots, 1,415; and animal tissues, 1,217.

The average number of NAEG soil, vegetation, and small animal aliquots collected, prepared, and shipped per month from January, 1972, to January, 1976, are shown in Figure 1.

During the period January 1, 1975, through December 31, 1975, a total of 4,898 PIDP soil samples were collected from NTS Areas 5, 1, and 4. A total of 5,363 soil samples were prepared in the Soils Laboratory. Analyses of these samples were performed in REEC's Environmental Sciences Department Chemistry and Counting Laboratories. A total of 6,023 soil samples were counted using Ge(Li) techniques for ^{241}Am and 1,390 samples were analyzed for $^{239-240}\text{Pu}$ by wet chemistry techniques.

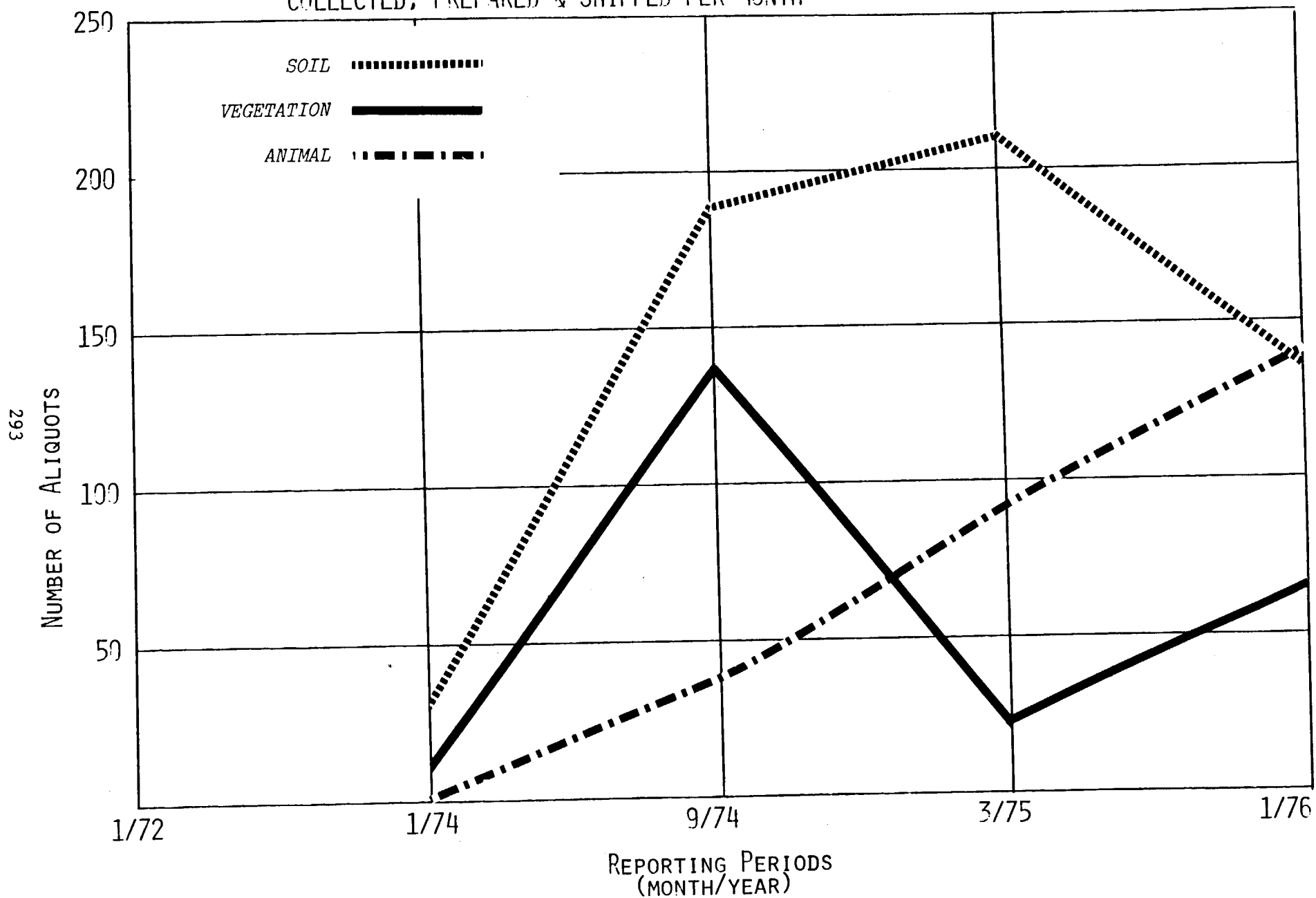
The expected increase in sample load in the Soils Laboratory in FY 76 resulted in modification of this facility to increase the volume of samples that can be handled. An additional drying oven was required to increase our sample drying capacity by 100 samples per loading.

Soil mound sampling studies were initiated during 1975 at the Tonopah Test Range (TTR). During the study, eight plots were sampled containing 79 soil mounds. Soil Mound Test Study No. 2 was also initiated in Area 13 consisting of 12 study plots. As part of this study, 40 soil mound test samples were collected and analyzed in the REEC laboratories. The results comprising ^{241}Am Ge(Li) measurements were forwarded to the investigators.

A study was conducted in the Soils Laboratory to determine the feasibility of altering the standard NAEG procedure to permit use of smaller sized soil containers, reduced number of steel balls, and shorter ball-milling time. The test results and recommendations were forwarded to the appropriate NAEG investigation for study.

A total of six grids were designated, staked, and sampled in Area 5 as part of the Plutonium Inventory and Distribution Program Element (PIDP). The largest grid, SMALL BOY, had to be increased several times to a maximum size of 4,800 feet by 6,400 feet. Soil samples were collected at 30 cm increments to a depth of 120 cm in those areas of these grids where preliminary sampling indicated contamination to these depths. The heavily disturbed nature of Area 5 has necessitated deep profile sampling in most cases.

FIGURE 1. AVERAGE NUMBER OF NAEG SOIL, VEGETATION & ANIMAL ALIQUOTS
COLLECTED, PREPARED & SHIPPED PER MONTH



Additionally, single, large-scale grids based upon 100-foot and 200-foot centers were surveyed, staked, and sampled in Areas 1 and 4. Deep profile sampling to a depth of 120 cm was required in the areas also due to a large amount of mechanical disturbance over the years.

The HAMILTON event in Area 5 was extensively sampled using both grid and random sampling techniques to permit evaluation of the relative merits of both systems. This evaluation is still under review by the PIDP.

Field tests were conducted to evaluate the possible usefulness of two new portable instrument systems in the PIDP program. An improved FIDLER system designed by Eberline Instrument Corporation and an LLL 256-channel portable analyzer developed as part of the VEST program was tested in Area 5, SMALL BOY.

PIDP soil sample results for SMALL BOY and HAMILTON in Area 5 were plotted and preliminary isopleths have been generated.

The REECO technical support staff assigned to assist in NAEG programs increased during 1975 to four with the addition of two staff positions.

FUTURE PLANS

Anticipated REECO support activities through September, 1976, include the following:

Combining NAEG field trips expected to the Tonopah Test Range (TTR) NAEG intensive study areas and Nevada Test Site (NTS) NAEG study areas, the projected total is 15.

Additional field trips are expected for the purpose of topographic and radiation surveys of nuclear detonation sites to select future NAEG nuclear study sites. Also, several aerial surveys are expected for the same purpose.

REECO NTS support of NAEG activities is planned to include:

- a. The collection, preparation, and shipment for analysis of approximately 250 soil samples for the ongoing NAEG Soils Element Soil Mound Study No. 2;
- b. The collection, preparation, and shipment for analysis of approximately 150 vegetation samples for the ongoing NAEG Vegetation Element Soil Mound Study No. 2;
- c. The collection, preparation, and shipment for analysis of approximately 170 soils and 170 vegetation samples to supplement present plutonium concentration contour data;

- d. The analysis of 18 soil samples, presently being stored in the NAEG Sample Storage Library, from NTS Area 5, stratum 2;
- e. The ongoing collection, preparation, and shipment of soil samples for the purpose of a continuing intercalibration of laboratories which are doing work for the NAEG;
- f. Shipment for analysis of approximately 50 vegetation samples for the NAEG Vegetation Element plant uptake study of ^{239}Pu and ^{241}Am from aged plutonium fallout areas;
- g. Support the collection, preparation by UNLV, and shipment for analysis of approximately 200 rodent samples for the NAEG Small Vertebrate Element intensive area studies;
- h. Radiological support of Resuspension Element studies and analysis of resuspension study samples; and
- i. Radiological support of the NAEG Plutonium Distribution and Inventory Element.

The need for considerable support for REECO Engineering is expected as plans become more firm for future cleanup, treatment, and grazing study sites. Topographic surveying and mapping will be needed to establish these sites. Surfacing, scraping, and other equipment will also be needed.

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Also, we wish to thank the NAEG investigators who have been so cooperative as well as informative during their field trips to the Nevada Test Site.

NAEG/REECo COORDINATION ACTIVITIES

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ABSTRACT

Coordination activities performed by the Reynolds Electrical & Engineering Co., Inc. (REECo), coordination staff, for the Nevada Applied Ecology Group (NAEG), are presented in this report.

Since this is the first NAEG report on NAEG/REECo coordination activities, a brief history is included to give the reader an orientation on the chronological development of NAEG/REECo coordination functions.

A description of NAEG/REECo coordination activities currently in progress is also included.

INTRODUCTION

NAEG/REECo coordination basically consists of bringing NAEG problems together with the people, equipment, and money needed to solve them.

The need for a Nevada Applied Ecology Group (NAEG) Coordinator to act as liaison between the NAEG and Reynolds Electrical & Engineering Co., Inc. (REECo), was identified late in 1971, at which time the first Radiological Services Coordinator was appointed.

The initial problems encountered by the coordinator were to determine the scope and support needs of the NAEG as defined by the NAEG Scientific Program Manager's "Coordination Plan for NAEG Studies," 1971.

This plan outlined formats for study proposals by NAEG investigators, established the general functions of a biostatistician and mathematical modeler, defined the need for effective coordination to ensure that conflicts did not occur with ongoing and planned NAEG activities, and identified the need for logistical requirements.

The scheduling of experiments, personnel, and logistical support, within the confines of a small research budget, soon followed.

By early 1972, Area 13 (Project 57), Area 5 (GMX), and Clean Slates I, II, and III and Double Tracks at the Tonopah Test Range had been identified as NAEG study areas and contracts had been let for the construction of 92,000 linear feet of fence to enclose contaminated areas at the Tonopah Test Range alone.

In the spring of 1972, four small experimental areas were laid out in Area 13 (Project 57). These small areas later became known as "microplots." Only one of these, a few hundred feet west of ground zero, was selected for sampling trials. In February, 1972, soil profile and vegetation samples were taken from the microplot (13-1) and prepared for analysis. These samples were collected by NAEG investigators under the watchful eye of the NAEG statistician.

By late 1972, the foundation for the coordinator's activities had been established in earnest. The activities of many different personalities, of as many different scientific disciplines, were being coordinated in an effort to study the radioecological aspects of the Nevada Test Site and the Tonopah Test Range.

In May of 1972, the groundwork had been laid for issuing grazing permits for the Tonopah Test Range. Subsequently, the coordination problem of keeping cattle out of radiation-contaminated areas where they were not supposed to be and in radiation areas where they were supposed to be soon developed.

Following the identification and fencing of the NAEG intensive study areas, as they came to be known, coordination efforts were concentrated on bringing scientists, engineers, radiological monitors, logistics, and money together to determine where the radioactivity was; what soil, vegetation, animal, resuspension, and other conditions existed; and how to go about sampling these conditions in a realistic way.

Soil was the first, most basic, ecological compartment, therefore, the first to be considered from a sampling and analysis point of view.

A team of experienced REECo radiological monitors was formed to perform radiation surveys of the intensive study areas and record radiation instrument measurements with respect to ground location.

These measurements were then compiled by REECo personnel and transferred to the NAEG statistician.

Liaison was established between REECo data evaluation personnel and the statistician, to coordinate field support with stake and sample location selection information.

By February, 1973, Field Instrument for the Detection of Low Energy Radiation (FIDLER) measurement surveys in Area 13 had been completed in coordination with the statistician, and hundreds of soil and vegetation samples had been collected by random location selection techniques. The earlier work with the Area 13 microplot provided sufficient information to establish standard methods for the sampling of soil and vegetation.

During March and April of 1973, FIDLER surveys, location staking, and the collection of hundreds of samples in Area 5 (GMX) was accomplished.

This surge of activity caused new areas of liaison to be required. Coordination was needed between the personnel who were developing new techniques for preparing soil and vegetation samples for analysis and the NAEG investigators responsible for the ongoing studies. Methods for instrumental and chemical analysis of the new sample matrix had to be developed. This expanded the scope of the coordination effort. In addition to interrelating the ongoing work of the statistician, engineers, radiological monitor teams, sample preparation personnel, and evaluation personnel, the radiochemists were introduced into the ecological study effort and liaison was again expanded to include their activities.

By the end of 1973, hundreds of Tonopah Test Range samples had been collected, prepared for analysis, and shipped to radiochemical laboratories. There were, by that time, thousands of samples from Area 5 and the Tonopah Test Range NAEG intensive study areas, in various stages of collection, preparation, and radiochemical analysis. Also, cattle grazing and soil resuspension studies were under way.

The advent of these activities produced an avalanche of sample information and analytical data. The manual methods being used to handle this increased data volume were inadequate. Either the data control and evaluation staff would have to be increased to the size of a small army with green eyeshades and quill pens, or the manual NAEG data base would need to be converted to a computerized data base. Before data could be handled by a computer system, it had to be organized. Therefore, meetings were held to discuss the problems involved. It was discovered that eight different methods had been used for selecting sample locations in the field. Different analytical methods were being used by the analytical laboratories. Arguments had long since developed concerning the most accurate formulas for calculating error terms for analytical result units. No agreement had been reached on which result units should be used throughout the NAEG system. As a result, in February of 1974, a meeting of all NAEG persons involved with these problems was held in Los Alamos, New Mexico, to discuss procedures being used and attempt to determine standard methods, especially concerning radiochemical techniques and resulting data.

The need for a central, computerized data bank became obvious. By June of 1974, initial groundwork had been laid for establishing a computerized NAEG/REECo data base. This resulted in the need for additional coordination efforts. REECo volunteered to take on the challenge of developing a computerized system capable of satisfying the myriad of ideas and requests inherent in the research effort. Coordination was employed in bringing together the investigators and their problems with the computer and data evaluation personnel, who hopefully had some answers. The data handling philosophy, at that time, was to try to anticipate the needs of each investigative study. As a result, meetings were held with investigators and data processing personnel to attempt to define the categories of data expected to result from each study. This approach did not bear much fruit; however, it did produce valuable information which was used in developing general data processing plans. A new philosophy for processing NAEG data developed. The new philosophy required that input data to the NAEG

data base be produced for each new study, in cooperation with NAEG investigators and NAEG management, prior to attempting to develop input formats. Also, it was recognized that "progressive programming" would be necessary rather than the traditional "program once and be through with it" approach.

In addition to the aforementioned coordination functions, the act of juggling support funds also became an area of coordination responsibility. An increased emphasis in one research area resulted in a corresponding decrease in effort that could be applied in another.

CURRENT ACTIVITIES

In order to keep abreast of and maintain a more active liaison with current NAEG activities, an NTS on-site coordinator has recently been added to the coordination effort. The addition of the on-site coordinator has increased the efficiency and thereby reduced the time required to comply with NAEG requests for REEC Co services.

During the period November, 1975, through January, 1976, sixty-five (65) requests to REEC Co to perform services for NAEG were received, processed, and complied with. Of these requests, two (2) concerned support budgets, eleven (11) field support, seventeen (17) sample preparation, twelve (12) sample analysis by REEC Co, and twenty-three (23) data processing.

The support budget requests involved changes in support work orders and the generation of a budget summary report.

Field support requests involved the collection of samples for inventory and distribution, the collection of transect samples from nuclear sites which may be selected for future NAEG studies, radiological personnel support to NAEG investigators visiting nuclear site locations which may be selected for future NAEG areas, the support of soil collection and sieving operations in Area 13, the collection of vegetation samples at the soil mound study No. 1 site in Area 11-C, radiological personnel support to the mobile GE Li system operating in Area 5, the collection of laboratory background control samples from Area 5, and a request for a report of field operation in Area 13.

Requests for sample preparation services included preparation of inventory and distribution samples from Area 5, preparation of nuclear site transect soil samples from proposed NAEG nuclear sites, performance of soil sample preparation tests to determine preparation parameters involved with preparing composite soil samples from soil mound study No. 2, assisting with coding of input shipping data, the preparation for shipment and shipment of small animal samples, performance of an experiment to determine ball milling duration for substandard containers, completion and transfer to the evaluation staff of soil mound study No. 2 field data, having genus and species identification performed on soil mound No. 2 vegetation samples, performance of a special mixing procedure on inventory and distribution surface samples from Area 5,

providing sample information on special profile samples for a REECO report, and assisting with providing information concerning quality control used during preparation operations.

Requests for REECO analytical services included analysis of soil resamples for inventory and distribution from Area 5 for 239-240 Pu and 241 Am (by Ge Li), analysis of soil samples from an Area 5 original grid location for 239-240 Pu and 241 Am (by Ge Li), analysis of soil transect samples from nuclear sites selected as possible future NAEG intensive study areas, the analysis of soil test samples from Area 13 for 241 Am (by Ge Li), the analysis of vegetation samples from soil mound study No. 1 in Area 11-C for 241 Am, the analysis of surface swipe samples taken following the NAEG sample preparation dry run at the NAEG high level laboratory at NRDS for alpha, beta, and gamma activity, the analysis of soil surface samples from Area 5 for 241 Am (by Ge Li), generation of a report defining quality control procedures used in radiochemical analysis, and analysis of special soil samples from Area 5 for 241 Am (by Ge Li).

Requests for data processing involved the generation of a computer printout for Area 5 Hamilton grid sample data, report of transect sample analysis results for Area 18 Danny Boy and Little Fellow II transect soil samples, generation of a sample analysis results report for Area 20 Palanquin soil transect data, generation of a computer printout of all NAEG soil profile data in the NAEG data base from the beginning of the NAEG Program, generation of a computer printout of Area 5 Small Boy data, generation of additional computer printout of Area 5 Hamilton data, generation of computer printout of Area 5 Hamilton data taken at random surface locations, generation of data report of soil mound study No. 2 soil preparation test, generation of a data report containing analysis results, weights, and parameters for vegetation samples from soil mound study No. 1 in Area 11-C, distribution of the soil mound study No. 2 field sheets, distribution of a genus and species report for soil mound study No. 2 TTR vegetation samples, generation of a data report for analytical data from Area 5 Hamilton surface samples, transfer of data tape containing all inventory and distribution analytical data from Area 5, distribution of a report on field data associated with a special NAEG soil profile study, generation of computer printout of analytical data for an inventory and distribution study of Area 1, and generation of a quality control information report.

At present, the NAEG system is "stretched out" with respect to sample and data flow through the system. The basic reason for this is that the rate of expansion of the NAEG Program is greater than the funding supplied to operate with. An area in which this is most noticeable is in the numbers and types of samples projected by investigators, many of which, if analyzed soon, would supply data which, in a year or so, could be used to fill in the gaps, so to speak, and complete the picture for the initial study sites for soil, vegetation, and small animal studies.

Plans are currently in progress for beginning work in NAEG nuclear study sites. This new effort will undoubtedly create new coordination problems throughout the NAEG System. We in NAEG/REECO coordination are looking forward to the challenge.

ACKNOWLEDGMENTS

We wish to express our appreciation to Russell D. Lease, the first Radiological Services Coordinator, for his efforts in initiating NAEG/REECo coordination. We wish to thank Henry J. Kayuha, the second Radiological Services Coordinator, for his diligence and energy in sustaining the coordination momentum. We also wish to thank Jared J. Davis, Paul B. Dunaway, Mary G. White, and Arden E. Bicker for their tolerance and understanding during times of hectic coordination activity.

NAEG FIELD PROBLEMS IN SAMPLING SOIL MOUNDS

AND

SOIL MOUND SAMPLE MIXING PROCEDURE

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ABSTRACT

This report discusses the problems, procedures, and solutions incurred during the Nevada Applied Ecology Group (NAEG) soil mound sampling study at the Tonopah Test Range, located approximately 45 miles southeast of Tonopah, Nevada.

Included is the means devised in the field for mixing and aliquoting soil samples of large volume, a method proven to be reliable.

INTRODUCTION

A workshop was conducted in September, 1974, to discuss soil sampling of mounds for defining plutonium distribution. A protocol was designed for evaluation of plutonium inventory in and around these mounds. The first study (feasibility, or pilot study) of mound sampling was at the Nevada Test Site, Area 11 in "C" site; the second mound sampling study was at the Tonopah Test Range in study site Clean Slate 3; and the third mound study occurred at the Nevada Test Site in the NAEG study site, Area 13.

PROBLEMS AND SOLUTIONS

Wind movement of particles from surface areas and dust control are very important factors when soil sampling, due to the possibility of alpha contaminated particles being transferred by disturbances such as wind onto the sample being taken.

The best method for controlling this problem was found to be through the use of a pressurized garden-type sprayer to wet the surface around and at the sampling location with water. By spraying a fine mist and then allowing time for the soil to absorb the moisture, and repeating this several times, control over particles being moved was believed to be quite satisfactory.

Perhaps in soil that does not accept moisture readily this procedure could be improved by adding a wetting agent to the water; however, this was not necessary in the areas in which the mound sampling studies were conducted.

Cross contamination from sampling equipment was prevented by decontaminating the items used after each sample was taken. This was done by using alcohol and rinsing with water. It was found that alcohol was needed to remove a slight film that was left on sampling equipment from the soil having been moist when sampled. The items were then dried and a smear-type swipe was taken and counted on a scintillation alpha particle counter. It was felt that this type of field decontamination would suffice and prevent a sample from being cross contaminated due to sampling tools.

It was imperative that the surface soil in the study area and sampling plots not be disturbed any more than was absolutely necessary; therefore, vehicle traffic was limited to existing roads only. This meant that due to the few roads in the study areas, most of the equipment used had to be carried a good distance to and from the sampling locations by hand, thus entailing numerous trips from the roadway where the vehicles were parked.

Boards 1" x 12" x 10' were used inside the sampling plots to walk on while taking inventory, measuring, and sampling. Foot traffic to and from the sampling plots was rerouted frequently to prevent the making of permanent trails.

When surveying the plots for sampling, care was taken not to disturb the soil or vegetation in these areas.

To determine the height of a mound or the desert pavement in relation to it, the best method without having a crew of surveyors was to use a surveyor's level (tripod-mounted type) and a grade stick. This enabled the highest, lowest, and midpoint heights to be easily taken.

Due to the large volume of soil in a mound caused by large animals such as coyotes or badgers, a problem arose: how to homogeneously mix and aliquot such a large sample in the field. After considering many ideas, the problem was quite readily solved by using a cement mixer (approximately 1/4 cubic yard in size). It was determined that by mixing the soil for 15 minutes, a homogeneous mixture could be obtained. From this an aliquot of approximately 5,000 grams was removed, sealed in a one-gallon can, and the remainder of the sample was returned to the location from which it had been taken. Some of these samples were in excess of 200 pounds. A medical scale with a capacity of 350 pounds was used for weighing the samples of large volume.

In order to prevent contaminating the mixer and to alleviate the possibility of cross contaminating a sample, the mixer was lined with five 36" x 54" plastic

bags. The sample was then placed inside the mixer, double-bagged, and sealed by taping, with a plastic cover taped in place over the opening. Plastic liners and cover were removed and replaced, after each mixing operation.

To help determine the size of a mound, a string or small rope was laid around the contours at the base to determine the edges or borders. Measurements were then made for the length, width, and any narrow or wide areas.

Due to the expertise needed in mound soil sampling, it is apparent that the sampling crew should be well trained in the methods, protocol, and radiological practices if the credibility of sampling is to meet the high standards expected for this type of study. When a crew is well qualified, the need to keep the same nucleus crew is quite apparent. With the large amount of paperwork involved in collecting the field data, the same person should handle this duty throughout the operation, from beginning to end, since this is the information to be programmed into the computer data base, and mistakes are more common when several persons are involved in recording complex data. It is felt that if the same people do the same phase of sampling each time a sample is taken, the probability of human error can be kept to a minimum.

Sampling at the Tonopah Test Range can cause logistic problems due to the remoteness and distance from warehousing and supplies at the Nevada Test Site. This problem was kept to a minimum with planning and foresight to anticipate what the logistic needs would be to complete the mound soil sampling. With the terrain of the type in the areas to be studied, a good practice is to have at least one four-wheel drive vehicle assigned for the sampling crew. Equipment and vehicles must be maintained to ensure they are in excellent operating condition.

ACKNOWLEDGMENTS

Thanks are to be given to the sampling crew made up of E. Milton, E. Hensley, J. Hardy, and C. Nash, for the professional and excellent manner in which they accomplished the sampling of the soil mounds.

NAEG COMPUTER PROCESSING ACTIVITIES

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ABSTRACT

The Information Systems Department provides services in systems design, programming, computer data processing, and administrative support to all organizational units within REECO. Additionally, these same services are provided to ERDA/NV and prime contractors such as Fenix & Scisson, Inc., and Wackenhut Services, Inc.

In November, 1974, the Environmental Sciences Department within REECO requested our assistance in the design of a system for the Nevada Applied Ecology Group (NAEG). We have summarized the efforts expended by both the REECO departments through December, 1975. This summary is organized according to past, present, and future activities.

Systems Effort in the PIDP and NAEG Data Base

In March, 1974, the Environmental Sciences Department requested our services to design a system for the Plutonium Inventory and Distribution program (PIDP). Data gathering and analysis for the PIDP was almost completed; therefore, early in our design effort, we established procedures to encode the data for ADP processing. Having the data in machine-readable form allowed the creation of the data base to parallel the system design for the PIDP.

Later, ERDA/NV suggested the data base be expanded to accept other NAEG data studies. A brief review indicated there were many facets of data which could be included in this data base. Recognizing the extent of the data available, ERDA/NV formally requested a feasibility study which was to include all NAEG data: Distribution and Inventory, Resuspension, Large Animal, REECO FIDLER, Small Animal, and Vegetation data. The study was completed in December, 1974; however, it was agreed not to include the LLL Resuspension, EPA Large Animal, or REECO FIDLER data at this time.

Along with the feasibility study, we continued to separately add data to the preliminary NAEG and PIDP data bases. Several months later, an addendum feasibility study was requested and received by ERDA to provide cost estimates to include REECO FIDLER and Large Animal data into the NAEG common data base.

Our fundamental problem, how to design a system which would encompass future studies for which the data criteria were unknown, kept surfacing. Meetings with both ERDA/NV and REECO Environmental Sciences Department personnel were held to resolve this problem.

After much deliberation, the efforts to create a common data base were abandoned in favor of a compatible data base. This approach was suggested by D. L. Wireman, coordinated with M. G. White of NAEG management, and it defined the data base as a series of elements with the capability for numerous studies within each element. The elements included were soil, vegetation, small animal, large animal, microorganism, and air data. This approach was defined in more detail and coordinated with the appropriate personnel.

Programming Effort in the PIDP and NAEG Data Base

The initial programming effort was strictly in support of the PIDP element. Programs were written to create a data base from manually prepared input. Programs were later written to interface the counting laboratory, computer-generated (DEC PDP11) analytical results with the PIDP data base. This eliminated the manual effort of entering that data. However, the header and aliquot information is still manually coded and keypunched for entry into the data base. Programs have been written to prepare the Distribution and Inventory report (see Attachment 1) and to create card or magnetic tape data files when requested by NAEG management.

Since this early PIDP support, the programming effort has been directed toward creating the NAEG data base. This effort went into developing conversion programs to input vegetation, small animal, and other soil data. Programming also has been initiated to accomplish data verification. However, the responsibility for the review and validation of the computer data remained within the Environmental Sciences Department, and personnel of that department performed the majority of the effort manually verifying the data contained within the data base.

Current Status of the NAEG System

Presently, the NAEG data base contains data from the Soil, Vegetation, and Small Animal studies. The data elements and samples contained therein are as follows:

<u>Element</u>	<u>Samples</u>
Soil	7,675
Vegetation	1,439
Small Animal	506

These data are stored on magnetic tape in coded form, and totals approximately 92,000 card image records. The NAEG Sample Results Report (See Attachment 2) documents each isotope measurement, as well as relevant background information for the reported sample.

In addition to report generation, data can be selected for punching into card formats or card image records on tape for subsequent use at other ADP installations. NAEG data can be processed with the Surface Approximation and Contour Mapping (SACM) program to create contour plots. It also can be statistically analyzed utilizing either the UCLA Bio-Med or the International Mathematical & Statistical Library programs. The NAEG reports can be prepared on microforms such as 105 mm microfiche with either 42X or 48X reduction, 35 mm roll microfilm, or 16 mm roll microfilm.

The start-up problems have been resolved; the data which reside in the compatible data base have been verified; and the capability to produce output from the data base has been proven.

Future Plans for the NAEG System

The Information Systems Department, in conjunction with the Environmental Sciences Department, has established the following goals to be accomplished during Calendar Year 1976:

1. Employ a qualified Scientific Programmer for one year under the Title X Program to create jobs for the unemployed.
2. Conversion of previous PIDP and NAEG data to the new formats.
3. Creation of procedures for setting up coding sheets for all new studies sponsored by NAEG.
4. Addition of the Mound Study (I and II data) to the NAEG Soil data element.
5. Addition of the REECO FIDLER data to the NAEG Soil data element.
6. Conversion and implementation of the studies within the vegetation, small animal, large animal, microorganism, and air data elements.

SUMMARY

The Information Systems Department personnel have provided a substantial amount of manpower to assist the Environmental Sciences Department personnel in designing a data base which will encompass data from all selected NAEG studies. Furthermore, inherent in the system design is the capacity and the flexibility for growth as future studies are conducted and added to this base of information.

ACKNOWLEDGMENTS

Our thanks go out to D. N. Brady, D. E. Engstrom, and L. M. Rakow for their efforts in bridging the gap between our ADP background and their scientific environment. In addition, we wish to thank D. L. Wireman for his invaluable assistance in developing and designing the framework for the NAEG data storage and retrieval system.

Our appreciation is also extended to M. C. Thompson and the Word Processing Branch for their editing and typing assistance.

AREA 05	SAMPLE TYPE 011	SOIL-GROSS	LOC. DESCRIPTION 03	DIRECTION AND DISTANCE	ISOTOPE	RESULT	PCT-ERR				
LIBRARY NUMBER	EVENT	LOCATION	DEPTH R-S	LAB TO	DESCRIPTION	ANALYSIS DATE	ALIQUOT NUMBER	TENS OF FEET	N-CI/GM		
						05-20-75	38340	K 40	1.79E	-2	15.4
						05-20-75	38340	AM241	2.46E	-3	36.1
						05-20-75	38340	AU198	2.43E	-4	39.9
						05-20-75	38340	CO 60	3.63E	-4	41.2
						05-20-75	38340	CS137	4.34E	-4	35.1
						05-20-75	38340	EU152	2.14E	-3	11.6
						05-20-75	38340	EU155	4.65E	-4	93.5
						05-20-75	38340	RA226	7.45E	-4	34.3
						05-20-75	38340	TH232	1.40E	-3	15.9
						05-20-75	38340	TH232	1.40E	-3	33.9
						06-11-75	38380	PU239	1.21E	-2	1.6
06023	100	S02CE010	000	S	RE	PU-GAMMA	COLLECTION DATE	05-13-75			
						05-22-75	38330	K 40	1.33E	-2	20.8
						05-22-75	38330	AM241	8.60E	-3	13.3
						05-22-75	38330	AU198	3.63E	-4	73.3
						05-22-75	38330	CO 60	2.47E	-4	17.2
						05-22-75	38330	CS137	1.43E	-3	21.5
						05-22-75	38330	EU152	7.45E	-4	13.3
						05-22-75	38330	TH232	1.15E	-3	60.9
						05-22-75	38370	AM241	1.15E	-3	95.5
						06-19-75	38370	PU239	1.41E	-1	.9
06018	100	S020E020	000	S	RE	PU-GAMMA	COLLECTION DATE	05-12-75			
						05-21-75	38325	K 40	1.16E	-2	26.2
						05-21-75	38325	CO 60	2.18E	-3	17.4
						05-21-75	38325	CS137	2.67E	-4	81.2
						05-21-75	38325	EU152	8.71E	-4	14.2
						05-21-75	38325	EU155	4.07E	-4	83.9
						05-21-75	38325	TH232	1.30E	-3	66.9
05361	100	S020E040	000	S	RE	PU-GAMMA	COLLECTION DATE	03-19-75			
						05-07-75	37443	PU239	3.25E	-3	3.0
06008	100	S020W000	000	S	RE	PU-GAMMA	COLLECTION DATE	05-13-75			
						05-21-75	38315	K 40	1.22E	-2	26.9
						05-21-75	38315	AM241	3.67E	-3	50.8
						05-21-75	38315	AU198	3.33E	-4	61.3
						05-21-75	38315	CO 60	1.76E	-4	19.4
						05-21-75	38315	CS137	4.57E	-4	41.4
						05-21-75	38315	EU152	8.05E	-4	10.8
						05-21-75	38315	RA226	4.22E	-4	58.0
						05-21-75	38315	TH232	1.22E	-3	56.1
						06-13-75	38355	PU239	2.98E	-2	1.1
06013	100	S020W010	000	S	RE	PU-GAMMA	COLLECTION DATE	05-13-75			
						05-21-75	38320	CO 60	2.90E	-4	39.2
						05-21-75	38320	EU152	1.08E	-3	24.4
						05-21-75	38320	RA226	7.33E	-4	30.1
						05-21-75	38320	TH232	8.13E	-4	55.0
						06-13-75	38360	PU239	2.32E	-3	4.1
06003	100	S020W020	000	S	RE	PU-GAMMA	COLLECTION DATE	05-13-75			
						05-20-75	38310	K 40	1.25E	-2	21.8
						05-20-75	38310	AM241	2.02E	-3	32.0
						05-20-75	38310	AU198	1.66E	-4	85.1
						05-20-75	38310	CO 60	1.22E	-3	24.1
						05-20-75	38310	CS137	3.99E	-4	61.3
						05-20-75	38310	EU152	6.88E	-4	11.5
						05-20-75	38310	RA226	6.52E	-4	56.4
						05-20-75	38310	TH232	1.02E	-3	71.8
						06-13-75	38350	PU239	2.39E	-2	2.0
05356	100	S020W040	000	S	RE	PU-GAMMA	COLLECTION DATE	03-19-75			
						05-07-75	37438	PU239	7.39E	-3	1.9
04462	100	S020W060	000	S	RE	PU-GAMMA	COLLECTION DATE	03-12-75			
						03-21-75	37179	K 40	1.04E	-2	26.2
						03-21-75	37179	CO 60	9.21E	-4	28.5

EVENT SITE 9		SAMPLE TYPE 040 VEGETATION-GROSS				ISOPLETH 2		LOC. DESCRIPTION 05 SMPL LOC/NEV GRID			
LIBRARY NUMBER	LOCATION/DESCRIPTION	STAKE NO	FT FROM SOIL/C	AZI- MUTH	DEPTH R OR S	GENUS	SPECIES	COLLECTION DATE	SPECIFIC INFORMATION		AREA
*****	LAB-ID LFE	ALIQ-NO		DESC-CD	N/A			ISOTOPE-ID 144-CE N-CI/GM	0.00E+00	PCT-ERR	.0
*****	LAB-ID LFE	ALIQ-NO		DESC-CD	N/A			ISOTOPE-ID 241-AM N-CI/GM	4.47E-02	PCT-ERR	1.0
*****	LAB-ID LFE	ALIQ-NO		DESC-CD	N/A			ISOTOPE-ID 239-PU N-CI/GM	3.28E-01	PCT-ERR	3.0
12749	NA12315F706791		00000	000	N/A	CHRYSO.	VISIDIFLORUS	04-29-74	N/A		11
*****	LAB-ID LFE	ALIQ-NO		DESC-CD	N/A			ISOTOPE-ID 137-CS N-CI/GM	9.00E-04	PCT-ERR	19.0
*****	LAB-ID LFE	ALIQ-NO		DESC-CD	N/A			ISOTOPE-ID 241-AM N-CI/GM	6.26E-02	PCT-ERR	1.0
*****	LAB-ID LFE	ALIQ-NO		DESC-CD	N/A			ISOTOPE-ID 239-PU N-CI/GM	4.77E-01	PCT-ERR	7.0
*****	LAB-ID LFE	ALIQ-NO		DESC-CD	N/A			ISOTOPE-ID 144-CE N-CI/GM	0.00E+00	PCT-ERR	.0
12749	NA12753E706046		00000	000	N/A	LYCIUM	ANDERSONII	04-29-74	N/A		11
*****	LAB-ID LFE	ALIQ-NO		DESC-CD	N/A			ISOTOPE-ID 239-PU N-CI/GM	3.46E-01	PCT-ERR	5.0
*****	LAB-ID LFE	ALIQ-NO		DESC-CD	N/A			ISOTOPE-ID 137-CS N-CI/GM	0.00E+00	PCT-ERR	.0
*****	LAB-ID LFE	ALIQ-NO		DESC-CD	N/A			ISOTOPE-ID 144-CE N-CI/GM	0.00E+00	PCT-ERR	.0
*****	LAB-ID LFE	ALIQ-NO		DESC-CD	N/A			ISOTOPE-ID 241-AM N-CI/GM	4.50E-02	PCT-ERR	3.0
12750	NA12675E706804		00000	000	N/A	ATRIPLEX	CONFERTIFOLIA	04-29-74	N/A		11
*****	LAB-ID LFE	ALIQ-NO		DESC-CD	N/A			ISOTOPE-ID 137-CS N-CI/GM	1.13E-03	PCT-ERR	47.0
*****	LAB-ID LFE	ALIQ-NO		DESC-CD	N/A			ISOTOPE-ID 239-PU N-CI/GM	1.61E+00	PCT-ERR	8.0
*****	LAB-ID LFE	ALIQ-NO		DESC-CD	N/A			ISOTOPE-ID 144-CE N-CI/GM	0.00E+00	PCT-ERR	.0
*****	LAB-ID LFE	ALIQ-NO		DESC-CD	N/A			ISOTOPE-ID 241-AM N-CI/GM	1.57E-01	PCT-ERR	1.0
*****	LAB-ID LFE	ALIQ-NO		DESC-CD	N/A			ISOTOPE-ID 239-PU N-CI/GM	9.39E-01	PCT-ERR	4.0
12751	NA12780F706526		00000	000	N/A	LYCIUM	ANDERSONII	04-29-74	N/A		11
*****	LAB-ID LFE	ALIQ-NO		DESC-CD	N/A			ISOTOPE-ID 144-CE N-CI/GM	0.00E+00	PCT-ERR	.0
*****	LAB-ID LFE	ALIQ-NO		DESC-CD	N/A			ISOTOPE-ID 241-AM N-CI/GM	8.83E-03	PCT-ERR	12.0
*****	LAB-ID LFE	ALIQ-NO		DESC-CD	N/A			ISOTOPE-ID 137-CS N-CI/GM	0.00E+00	PCT-ERR	.0
*****	LAB-ID LFE	ALIQ-NO		DESC-CD	N/A			ISOTOPE-ID 239-PU N-CI/GM	1.19E-01	PCT-ERR	5.0
12752	NA12302E706909		00000	000	N/A	ATRIPLEX	CONFERTIFOLIA	04-29-74	N/A		11
*****	LAB-ID LABX	ALIQ-NO		DESC-CD	N/A			ISOTOPE-ID 239-PU N-CI/GM	1.71E+00	PCT-ERR	7.3
*****	LAB-ID LABX	ALIQ-NO		DESC-CD	N/A			ISOTOPE-ID 238-PU N-CI/GM	3.30E-02	PCT-ERR	7.8
*****	LAB-ID LABX	ALIQ-NO		DESC-CD	N/A			ISOTOPE-ID 241-AM N-CI/GM	1.59E+00	PCT-ERR	7.5

U INDICATES RING OR SCOOP UNKNOWN - SAMPLER-CD 06 OR 07

* INDICATES NO ALIQUOT SUBMITTED FOR RESULT CARD

SAMPLE PREPARATION DRY RUN AT THE
NEVADA APPLIED ECOLOGY GROUP (NAEG)
HIGH RADIOACTIVITY LEVEL FACILITY

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ABSTRACT

An activity was conducted in a laboratory facility located at Area 400 of the Nevada Test Site to simulate and assess the feasibility of performing transuranic radiochemical analysis on large animal tissue samples that could contain millicurie quantities of radioisotopes.

INTRODUCTION

One of the objectives of the NAEG is to assure that contractor laboratories have developed analytical techniques for analysis. One of these techniques is the handling and analysis of relatively large tissue sample sizes and high radioactivities of the transuranics that are anticipated in the large animal studies program.

The LFE Environmental Analysis Laboratories, a contractor to ERDA, is a laboratory that has qualified under the NAEG's qualification procedure for the analysis of large animal samples for transuranics. This laboratory was required to perform a simulated exercise to assess the techniques, conditions, problems, and precautions associated with the chemical analysis of Class 1 (very high radiotoxicity) materials in tissue samples. This dry run was conducted in March, 1975, at the NAEG high radioactivity level facility at Test Cell A in Area 400 of NRDS.

The NAEG high radioactivity level facility required routine maintenance and power to be put into use. The laboratory equipment, including hoods, glove boxes, and air filtration systems, required refurbishing prior to use in order to meet the health and safety requirements.

The dry run was conducted in this rehabilitated laboratory facility since none of the presently qualified NAEG laboratories met the ANSI laboratory standards for handling transuranics in the millicurie and larger activity ranges.

The dry run was conducted after the REECO and LFE protocols were approved by ERDA/NV. LFE coordinated with REECO's Environmental Sciences Department for the initial preparation and stocking of the facility and the overall monitoring and health physics support during the exercise.

PROCEDURE

Approximately two pounds of various types of beef (muscle, bone, liver, etc.) were spiked with a radioactive source that was used to simulate the transuranics. The isotope chosen for use was cobalt-60. This isotope was chosen for its relative ease of detection and tracking through the sample preparation process.

The spiked samples were placed in a large aluminum pan and dried overnight in a large vented drying oven. After drying, the samples were cut into smaller pieces inside the designated hot laboratory area, this area being a controlled access area. All utensils and the work areas were monitored continuously and swipes were taken for documentation. In addition to lapel (breathing zone) air samples, general laboratory work area air samples were taken and analyzed. The results of these swipes and air samples indicated that all radioactive materials were being contained in the dissolving samples. All nuclear counting was performed by REECO in an area of the facility designated as the counting room. This room was equipped to immediately evaluate contamination problems without undue delay in an emergency.

CONCLUSION

The comprehensive program of taking air samples and swipes indicated that the work areas were free of radioactive contamination; the techniques of dry, wet, and dry ashing; and the sawing of bone under rather rigorously controlled conditions was indeed a feasible operation. It was also concluded that the NAEG facility itself could be made operational and functional to perform this type of sample preparation.

Current budget limitations have prevented the permanent upgrading of the facility to a Class B or Class A laboratory, but the facility in its present condition can be used as an adequate Class C radiological laboratory.

ACKNOWLEDGMENTS

We wish to express our appreciation to William J. Major and Kim D. Lee of the LFE Environmental Analysis Laboratories staff, who participated in the dry run exercise, for their close cooperation with the REECo staff.

SUMMARIZATION

SUMMARIZATION

M. G. White and P. B. Dunaway

The annual information conference of the Nevada Applied Ecology Group (NAEG) held in Las Vegas, Nevada, in February, 1976, provided the environmental research documents in this report. As an ERDA environmental integrated research studies group, the NAEG has varied disciplines represented in its meetings. One of the advantages of holding an annual meeting of this type is the interface of scientists and technicians interested in the different aspects of environmental investigations where a more thorough examination of desert environmental problems is permitted. Some answers, status of ongoing studies, and some new problems were addressed during the session.

The research efforts of the NAEG vegetation investigators resulted in five reports covering plant uptake of plutonium and americium through roots, estimation of vegetation plutonium, americium, and uranium radionuclide inventory, stabilization of soils in cleanup efforts by revegetation, a special report on possible cytological effects on shrubs from low-level radiation at the Nevada Test Site, and report of vegetation sample results from Area 11 (Plutonium Valley).

An investigation to observe the effects of soil amendments (nitrogen fertilizer and organic matter, with and without DTPA) on plant uptake of plutonium and americium was conducted by Romney *et al.* using soil from various NAEG NTS intensive study safety-shot sites. Alfalfa, barley, and soybean plants were grown in pot culture experiments, in order to prevent foliage contamination from resuspended soil particulate material as occurs under field conditions. No alteration of the root uptake of $^{239,240}\text{Pu}$ was evident with additions of soil amendments such as nitrogen, fertilizer, and organic matter. However, when sulfur was added with DTPA, root uptake significantly increased. The DTPA amendment apparently was the agent for this increase, as uptake in soybeans also was significantly increased with the addition of the chelate amendment only. The greater uptake of ^{241}Am through plant roots in proportion to $^{239,240}\text{Pu}$ was demonstrated by the amendment experiments, lending support to the belief that potential problems from americium in the environment should be addressed by research investigators as well as those problems from environmental plutonium.

Romney and co-investigators found that comparisons of soil and vegetation inventory estimates at NAEG study sites on NTS indicated that plutonium in the standing vegetation constitutes an insignificant portion (less than one-thousandth) of the total $^{239,240}\text{Pu}$ inventory in the study areas. Resuspendable materials on vegetation are the probable source of most of the $^{239,240}\text{Pu}$ present in vegetation samples from the safety-shot areas of NTS. The vegetation in desert environments is important in

prevention of wind erosion and acts as an intermediary trap for blowing particulate material.

Discussion of desert environment problems and associated recommendations are presented by Wallace and Romney for measures to be considered in the proposed cleanup and restoration of areas at the Tonopah Test Range and Area 13, NTS. Through examination of past decontamination and soil disturbance experiences in the Great Basin Desert, Wallace and Romney found indications of tendencies toward slow natural recovery in disturbed areas. Revegetation considerations of disturbed areas are addressed in detail, with excellent bibliographic material included.

The inventory of plutonium, americium, and uranium in/on vegetation samples from Plutonium Valley (Area 11) intensive study sites was determined from analyses of samples at McClellan Central Laboratory. (Mount's excellent report of analyses of ^{238}Pu , ^{241}Am , and ^{235}U in vegetative material is included in this document.) The soils under desert plants in mounds were investigated by Tamura, ORNL, to determine whether the character and behavior of the plutonium in the mounds were similar to that in desert pavement soils. Analysis of samples reflected some of the problems of sampling mounds. The distribution of radioactivity observed demonstrated the importance of sampling techniques. Preliminary data on the activity in particle size fractions, in terms of activity per unit mass, indicates that the coarse silt (50-20 μm) contains the highest activity in the desert pavement samples. The medium silt (20-5 μm) contains the highest activity in the desert mound samples. As considerable mechanical mixing had occurred in the mound sampled, it was difficult to interpret measurements of depth distribution. In making particle density measurements, Tamura found an indication that the desert mound particles in the 20-5 and 5-2 μm size classes are lower in density than the corresponding size classes in the desert pavement. In the smaller size classes in both types of samples, the data suggest that plutonium is distributed in the lighter fractions as well as the heaviest fraction.

Study of the implications for uptake by man and grazing animals through ingestion or inhalation of particles from desert mound samples characterized by Tamura awaits more definitive data. NAEG plans include investigation of various natural mechanisms (organic and inorganic soil complexing) by which biological uptake may be enhanced.

Fowler and Essington emphasized the fact that current soil profile data from samples taken down to 25 cm indicate that the maximum depth of penetration of plutonium in soils at NTS is still unknown. (Greater in-depth sampling, of course, depends on availability of funds.) Another problem indicated is the disparity in the plutonium-americium ratios among study sites. Difficult analysis of americium is suggested as a possible cause of the disparities. Also, other factors impinge on the data available, e.g., mechanical disturbance at some sites, differences in times and types of plutonium release, variance in climatic environments at NTS, etc.

NAEG studies to gain information concerning the influence of soil microbial activities on the bioavailability and biotransport of transuranics in desert environments continue. Au and Beckert suggest that the biomass figure for fungal and bacterial biomasses should provide some understanding of potential microbial activities in Nevada Test Site soils. Of course, many variables enter into such determinations. In fact, the soil fungal and bacterial populations of desert soils have been observed to be very high and diversified, even in locations without plants. Experiments using *Aspergillus niger* are under way to determine whether the availability and transfer of plutonium are increased during successive generations of soil microbial growth.

G.I. tract studies with fistulated steers grazing in Area 13 continue. The relative solubility and concentrations of plutonium and americium in rumen samples collected under field conditions in Area 13 vary, depending on several factors discussed by Barth: season, predominant species ingested, vegetative stage of growth, presence of fruiting involucres on *Eurotia lanata*, grazing intensity, and distance of grazing area from ground zero. Future plans include combined studies with microorganisms and artificial rumen systems. Investigation of differences between biological availability of biologically incorporated plutonium and nonbiologically available plutonium, and study of whether biological organic binding protects plutonium from being removed from solution by competing chemical reactions are among studies planned.

A comparison was made of data from cattle grazing in Area 13, NTS (a plutonium-contaminated safety-shot area), with data from cattle grazed in the area of the Rocky Flats Plant in Jefferson County, Colorado. Smith reported on Area 13 cattle, reflecting gonad concentrations equal to that of femurs and significantly higher than muscle. Similar relative tissue concentrations were found in the Rocky Flats cattle data, although generally the concentrations were less than those reported for Area 13 cattle. Smith also reported that concentrations of plutonium in the femurs of young animals were slightly higher than concentrations in adult animals.

Holstein dairy cows were used to measure rate of passage of sand particles tagged with ^{141}Ce , ^{85}Sr , ^{54}Mn , or ^{46}Sc , through the G.I. tract. Patzer, Sutton, and Potter performed the study to determine whether passage rate of particles through the bovine G.I. tract is related to size. Results indicated significant variance with particle size, with measurements of up to 12 days for passage of 90% of the particles in feces.

Comparisons between the biological transport of plutonium and curium in dairy goats were studied by Sutton *et al.* Intravenous or oral doses of curium-243 chloride were administered to 6 lactating goats, followed by milk, urine, fecal, and blood sampling for 6 days. Approximately 2% of the IV dose was transferred to milk during the 6-day period. Urine and feces averaged more than 4% of the administered IV dose. The oral dose results indicated excretion in feces of all but less than 1% of the administered dose, and nondetectable quantities in milk, urine, and blood. Compared with plutonium previously administered (IV) to a single

goat by Stanley and Mullen, the curium was excreted more rapidly. Shortly after injection, the percentage of the administered curium dose in the biological samples was higher than the plutonium values, based on percent per kilogram basis.

Sutton *et al.* also reported on an investigation of transport and retention of ^{238}Pu in calves fed *in vivo* labeled milk vs prepared *in vitro* labeled milk. This experiment will provide information on the relative biological availability of plutonium in the two experimental groups of animals. The cows which received the citrate-buffered plutonium-238 nitrate were also sampled for uptake. Results of these experiments will be provided in a future NAEG document as data were not available at the time of the reporting session.

Small vertebrate investigations for NAEG are performed by University of Nevada, Las Vegas (UNLV), at NTS and the Tonopah Test Range. Data on rodents and lizards from NAEG safety-shot sites, NTS, are presented in this report. Moor and others at UNLV reported lower carcass burdens of ^{239}Pu and ^{241}Am than pelt or G.I. tract burdens by a factor of 10^2 . Hematologic studies of rodents from the NTS and TTR areas are currently under way.

Methodology and preliminary analyses are discussed. Moore *et al.* report that some of the changes in NTS soil chemistry environments may be due to animal burrow environments. They suggest that soluble nutrients, gases, humidity, and temperatures are at levels which are more conducive for maintenance and growth of microorganisms than surface soil areas. Animal burrows, of course, are difficult areas to study in that many factors are involved. Mechanical stabilization of burrow areas for easier sampling (e.g., freezing tunnels for easier and more discrete soil samples), sampling that is meaningful, individual species of small vertebrate habits study, biological inventories of microhabitats, and food sources are a few such factors, not to mention radiochemical variance in differing environments.

Preliminary synthesis of some of the data generated in the various safety-shot areas studied by NAEG has been begun by several investigators. Until recently, field measurements of the concentration of Pu in various ecosystem components were the primary objective of most of the NAEG investigations as a beginning step toward providing information for an ecosystem model for the transport or movement of Pu in the environment.

Gilbert, NAEG statistician, presents efforts by several of the NAEG program investigators to synthesize the $^{239,240}\text{Pu}$ data currently available from Area 13, NTS (an NAEG safety-shot study site). Data on soil, vegetation, small vertebrates, large vertebrates, and air concentrations previously reported are combined within single graphs in order to evaluate trends or information not obvious by other means of presentation. The limitation of few numbers of animal data from the Area 13 grazing study to date causes restricted interpretation. However, it is evident that the lowest concentration in cow tissues is in muscle, and the highest in hide and hair. Statistical analysis indicates that lung concentrations

are higher by 2 to 3 orders of magnitude in Area 13 cattle than for cattle grazed in a control area. Based on limited data, muscle values from the cattle grazing the highest concentration area were possibly elevated about one order of magnitude above the control cattle for that tissue. As more data become available, these values will become more definite. And, as more data are available, it is clear that adjustments in Martin and Bloom's model are becoming essential to reflect actual ecosystem component measurements *vs* theoretical values. Gilbert and Eberhardt, using data provided by Smith, Bradley, Fowler, and Romney, discuss some aspects of meaningful interpretations of actual field data, including skewed data sets and bias problems.

Bliss and Jakobowski, in reporting further data on distribution of plutonium around the NTS, indicate that there has been no recent significant change in plutonium in soil data. Duplicate sampling for air concentrations of plutonium has begun as a study to estimate the total error associated with the sampling and analysis of air samples. Three drainage basins near the Nevada Test Site are being monitored for signs of any definable movement or concentration of plutonium. Preliminary observations are negative. Analysis is also under way of vegetables and fruits collected from areas surrounding NTS. Future plans include measurements of off-site plutonium resuspension, mounds under shrubs, study of soil collection errors (pertaining to sample locations, size, etc.), and definition of ^{137}Cs distribution off-NTS.

The support services section of this publication includes reports from those contractor personnel responsible for making it possible for the field-conducted experiments to take place. Not only does NAEG depend heavily on these people to surmount huge problems in logistics and data base operations (Reynolds Electrical & Engineering Co., Inc.), literature search (Oak Ridge National Laboratory), and analysis, but also for development of techniques, methods, and future guidance on potential field or laboratory problems, advance literature needs, and availability of new radiochemical and statistical analysis procedures or tools. The reports include special reports by Lee, Major, and Wessman; by Major and Leventhal, LFE Environmental Analysis Laboratories, on radiochemical services to NAEG, and tissue analysis for transuranic elements.

A report by Pfuderer of the Nevada Applied Ecology Information Center at ORNL on activities of that group reflects only a small portion of the in-depth background of this important service to the Nevada Applied Ecology Group. Recent innovations have included a special abstract report service to NAEG personnel on the most recent publications of interest, as they appear in the ORNL Environmental Sciences Information System. The massive data bases available at the ORNL Ecological Sciences Information Center in the area of radioecology are listed in Table 2 of Pfuderer's paper.

Reynolds Electrical & Engineering Co., Inc., provides NAEG scheduling and coordination, logistical support, rad-health services, data base processing, programming, and computer services, sample collection, preparation, handling, and laboratory analysis. Reports on the varied

REECo activities in support of NAEG field and laboratory needs are included, prepared by Brady, Wireman, Rakow, Rosenberry, Auer, Zellers, Straight, and Smith.

Many others, not mentioned in this summarization of current NAEG Nevada Test Site field and laboratory activities, have made contributions toward the objectives of the Nevada Applied Ecology Group. Some results and some future indications have been reported here, together with preliminary attempts to evaluate previous data. Hopefully, the readers will forgive certain compromises in order to effect timely presentation of the information included herein.

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