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Avian Individual and Population Health on Los Alamos National Laboratory 2005: Foraging Effects and Egg Contaminant Residues

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Table of Contents

1.0 Introduction to the Project 3

1.1 Methods 4

1.2 Avian Monitoring Network 2005 Summary 6

2.0 Foraging Effects on Contaminant Uptake in Cavity-Nesting Birds 9

2.1 Introduction 9

2.2 Methods 11

2.2.1 Productivity 11

2.2.2 Home Range Foraging 12

2.2.3 Prey Selection 13

2.3 Results 13

2.3.1 Productivity 13

2.3.2 Home Range Foraging 16

2.3.3 Prey Selection 17

3.0 Contaminant and Residue Exposure of Cavity-Nesting Birds 18

4.0 Acknowledgments 30

5.0 Literature Cited 30

1.0 Introduction to the Project

In 1997, an avian nestbox monitoring network was established on Los Alamos National Laboratory (LANL), Los Alamos County and U.S. Forest Service land in northern New Mexico to investigate the health and condition of cavity-nesting bird populations on the Pajarito Plateau. The purpose of this study is to evaluate the magnitude and sources of ecological risks from contaminants and other environmental stressors for cavity-nesting birds at LANL. The main objective is to evaluate the ecological and physiological costs of exposure to various contaminants at LANL and their potential impact on population processes. During the past two decades it has become increasingly important to be able to predict risks from potential adverse effects of exposure to chemical and physical hazards in the environment. This has resulted in the critical need for estimates of the relationship between exposure of organisms to contaminants and the response of the population.

This project was started in 1997 with 450 nest boxes placed in potentially contaminated and noncontaminated areas on LANL. Data on individual bird health and condition and population-level parameters have been collected for nine consecutive years through 2005. During the nine years the nest box monitoring project has seen the Cerro Grande fire that burned ¼ of the boxes and several areas of bluebird habitat, several years of severe drought, a large-scale tree thinning project, and now an 80% mortality of pine trees from bark beetles in several of the areas where nest boxes are placed. Many of the trees that boxes are placed on have died during the 2002-breeding season. Although each of these events will undoubtedly add variation in any study attempting to understand

the environment, it also gives us the opportunity to gain insight into the effects of environmental change and stress.

1.1 Methods

Avian Monitoring Network

To investigate health and condition of birds in areas of concern for contaminants, an avian monitoring network of nest boxes was initiated at LANL. During the winter of 1997, 438 nest boxes were placed on LANL in total of 18 both potentially contaminated and reference areas. Nest boxes were placed approximately two meters off the ground on trees and spaced approximately 50-75 meters apart. Boxes were placed in the open ponderosa pine forest of the canyons and piñon–juniper woodland on the plateau mesas. Boxes were placed in 18 locations or areas on LANL land with an average of 29 boxes per location.

The western bluebird (*Sialia mexicana*) (WEBL) is a widely distributed, sexually dichromatic, and monogamous species. The ash-throated flycatcher (*Myiarchus cinerascens*) (ATFL) is not as widely distributed or sexually dichromatic. Both species nest in secondary nest cavities, are insectivorous during the breeding season, and use small amounts of grit in their gizzards that are potentially important exposure pathways. These two species have similar life history traits, although the ATFL has a faster rate of development, fledging 4-5 days earlier than the bluebird, and has a significantly higher field metabolic rate during development (Mock et al. 1991). This difference in duration of development period could affect the relative exposure and risks to contaminants. If intake of contaminants in soil is proportional to dry matter intake as is assumed in ecological risk methodology, the higher metabolic rate for the ATFL compared to the WEBL may increase

their relative risk of toxic exposure. Sexual dichromatisim differs in the two species, with the WEBL being sexually dichromatic and the ATFL having no sexual differences. Although the specific migratory pattern of both species is unknown, WEBLs are more common in the winter months in the study area and it is thought that the ATFLs migrate farther south, even in mild winter years. Both bird species in this study readily utilized nest boxes and are common in northern New Mexico.

The main objective of the avian nest box-monitoring network is to investigate population level parameters such as survival, nest productivity, and return rates or recruitment into the population. All adults and nestlings western bluebirds are to be banded and return band numbers are recorded. This data will be used in a population viability analysis that can determine the status of the population. The LANL bluebird population will be compared with a western bluebird population in Oregon, California, and Arizona.

Data Analysis

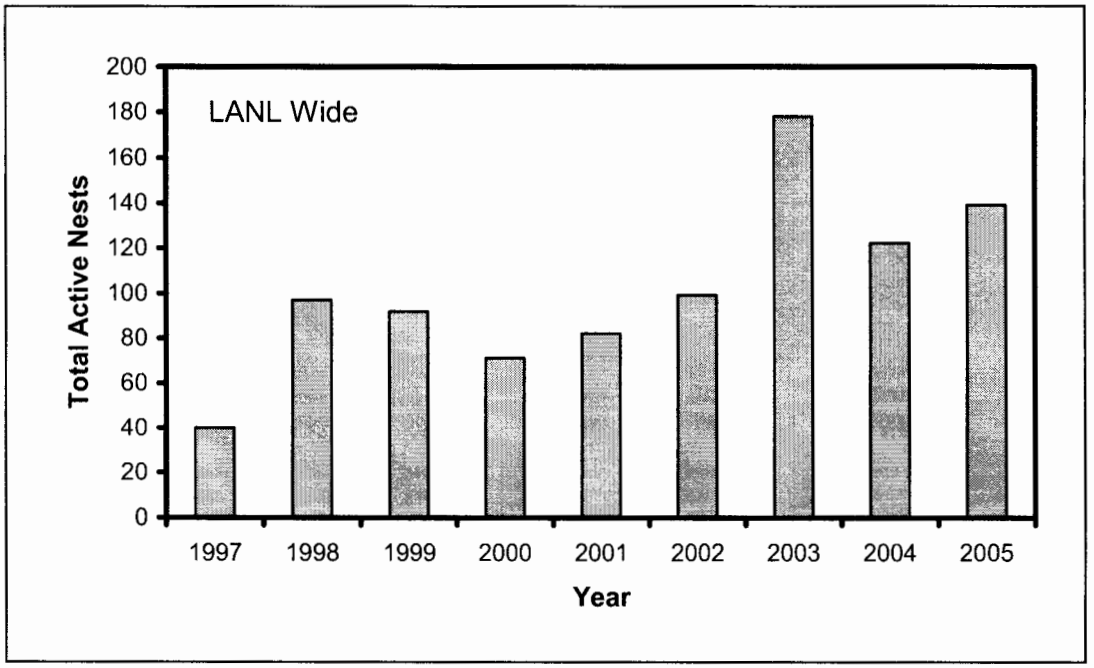
The Statistical Analysis System (SAS, Institute, Inc. 1987) was used for all statistical analyses, and assumptions for parametric statistics were examined. Growth and physiological parameters were compared using repeated measures Analysis of Variance models (ANOVA). Means for each treatment were compared with Duncan's Multiple Range Test. Data not normally distributed or having unequal variances were compared with Kruskal-Wallis nonparametric tests.

1.2 Avian Monitoring Network 2004 Summary

The number of active boxes in the project was 122 total (Figure 1). However, there were 33% more boxes placed in 2002 bringing the percentage of active boxes down to

15.5%. Population dynamics are dependent on two primary things: the number of offspring produced and the survival of individuals from each year. An accurate estimate of survival rates requires the banding and recapture of numerous individuals over multiple years. For the estimation of annual survival rates, individuals must be recaptured over time, which will give valuable information on dispersal distance as well. For example in the California central valley western bluebirds have been found to have a 33% recruitment of nestlings into the breeding population (C. Graham, personal communication). For the LANL bluebird population, the recruitment is a much lower 2-3%. Clearly, one of the future goals of the project should be to determine the reason for such a low recruit into the population on the Pajarito Plateau. It could be the due to the fire, drought, anthropogenic reasons, or other environmental variables such as heterogeneity of habitat.

Figure 1. Total active nests for 1997-2005 for the entire laboratory nest box network.



Hinds (1984) stated that a successful ecological monitoring program must be ecologically relevant, statistically credible, and cost-effective. The LANL nestbox network

has collected data over six breeding seasons. Thus, the nestbox network has sought to reach all parts of the tripartite requirements. The large sample size of birds utilizing the nestboxes allows for significant statistical power in the analyses. A total of 3,013 birds have been banded in the nine years of this project. The Golf Course had the highest occupancy in 2005 relative to the number of boxes (Table 1). Mortandad canyon had lower occupancy in 2005 compared to previous years.

Table 1. Active boxes for 2005.

Location	Original # of boxes	# Burned / Torn Down	Current # of boxes	2005 Active WEBL	2005 % Occupancy WEBL
Mortandad	111	51	60	18	30%
Canada del Buey	29	0	29	11	38%
Golf Course	12	0	12	7	58%
Cemetery	16	0	16	6	38%
Pueblo	61	0	61	11	18%
Bayo	82	0	82	5	6%
TA-35	28	3	25	3	12%
TA-51	5	0	5	2	40%
TA-48	45	0	45	1	2%
LA	112	0	112	5	4%
DP	12	0	12	2	17%
Ancho Canyon	18	0	18	1	6%
Ancho Mesa	12	0	12	1	8%
A/W Mesa	12	0	12		0%
Water	39	0	39	3	8%
Acid	31	5	26		0%
Guaje	11	11	0		n/a
Gate 9	6	0	6	1	17%
Gate 11	10	0	10	2	20%
DX	37	13	24		0%
TA-33	27	0	27		0%
Sandia	33	0	33		0%
LANL-Wide	749	83	666	79	11.9%

A large majority of the trees that boxes were placed on succumbed to the drought-induced bark beetle infestation. Tree mortalities continued throughout the breeding season through October, 2003.

Table 2. Total active boxes by species in 2005 (N = 139).

Species	Number of Active Boxes	Percent Active Boxes represented by species
Western Bluebird	79	56.8%
Violet-green Swallow	24	17.3%
Ash-throated Flycatcher	15	10.8%
House Finch	7	5.0%
Mountain Bluebird	9	6.5%
House Wren	4	2.9%
Juniper Titmouse	1	0.7%

2.0 Foraging Effects on Contaminant Uptake in Cavity-Nesting Birds

2.1 Introduction

The 2005 field season was the ninth year of the nestbox network-monitoring program. This field monitoring study was begun with the intention of comparing two similar cavity nesting bird species in a gradient of potential exposure to a mixture of contaminants at Los Alamos National Laboratory (LANL) in New Mexico. Although these species share similar life history traits, ATFLs and WEBLs differ in several respects that may affect contaminant uptake and subsequent effects.

The first major difference between the two species is the discrepancy in rate of development. The ATFL has a more rapid rate of development, and fledges about 4-5 days faster than the WEBL (Mock et al. 1991). Energy expenditure between the adults of the two species show that the ATFL daily energetic requirement is approximately 22% (between 16-32%) higher than the WEBL (Mock et al. 1991). Thus, the ATFL must acquire up to 20 g more food each day than WEBL adults with the same clutch size (Mock

et al. 1991). Therefore, the ATFL's greater demand for food resources relative to the WEBL may increase its likelihood of accumulating greater contaminant burden.

Secondly, although both target species are insectivorous, WEBLs and ATFLs have different diets, and therefore they may be exposed to correspondingly different contaminant loads. For example, ATFLs typically choose large grasshoppers as one of their main prey specimens (Fair, unpublished data). Grasshoppers are herbivorous, ephemeral arthropods living above ground where they may be exposed to fewer contaminants. WEBLs typically choose ants as one of their primary prey specimens (Fair, unpublished data), indicating selection of arthropods that are omnivorous, long-lived, and dwell underground. Thus, the WEBLs diet will likely reflect prey items that have a greater length of time to accumulate contaminant loads, resulting in prey items that carry greater contaminant burdens. ATFLs are expected to have lower contaminant loads because of their prey selection, but will likely have greater overall effects than WEBLs due to higher metabolic demands.

Differences in dietary composition may likely be a result of different foraging areas utilized by each species. WEBLs have been found to forage over short distances (50 to 100 m) from their nests during the breeding season (Fair, unpublished data). Since WEBLs forage within localized areas, resulting food chain effects may effectively monitor local contaminants (Wayland et al. 1998). Home range in this study refers to the area utilized by a bird in its day-to-day activities (i.e. foraging). Home ranges are restricted to finite areas around the nest due to energetic demands and the need to return to nestlings with prey (Linkhart et al. 1998). In a study investigating spatial variability and mobility of organisms on contaminant exposure, the probability that an individual is exposed to a high

level of contamination increased with increasing home range size (Marinussen and Zee 1996).

The main objectives were to examine how differences in foraging activity between two cavity-nesting passerines affect contaminant uptake, and how energetics may affect the relative impact of contaminant exposure. Even though contaminants are known to affect passerines, virtually no research has been conducted relating corresponding exposures to home range usage and extent.

2.2 Methods

2.2.1 Productivity

The 2005 monitoring has 749 nestboxes this year across the 112 km^2 LANL, Los Alamos County and U.S. Forest Service land (Table 3). One hundred twenty-two active nests in 18 locations were monitored throughout 2005, both in contaminated and reference areas.

Nests with warm eggs were considered to be active and were visited every two days until hatching (day=0). A minimum of ten active nests were chosen for monitoring based on close location within 100 m of a contaminated potential release site (PRS) boundary). A minimum of 3 controls were also chosen based on a distance greater than 100 m from a PRS. After the clutch was complete, one viable egg was collected randomly from each nest during incubation.

After day 0, the five pairs of nests were visited every two days to monitor nestling productivity. Productivity parameters measured included: fluctuating asymmetry, weight, growth rate, brood size, and fledging success. Each nestling was banded with a standard USFWS band. The sex of WEBL nestlings was determined by plumage color. The gender

of ATFL nestlings was determined by obtaining a single blood smear (<210 ul) from the brachial vein after day 12. All nests that had eggs removed were collected once nestlings fledged.

Eggshells were cleaned and measured for length, width, and volume. Eggshell thickness was measured on all collected eggs using a minimum of four points along the equator of each shell half (units of 0.01 mm). Egg yolks were frozen and stored for future contaminant residue analysis.

2.2.2 Home Range Foraging

Each female in the paired nests was fitted with a color band and tracked from early May through early August in 2005. To identify individuals visually while they foraged, one or two color bands were fitted onto the females' left/right tarsus. Each female was caught using mist nets placed directly in front of the nestbox opening, set immediately following hatching.

Individuals were tracked between 7 am to 10 am MT. Banded females were located visually using binoculars and/or spotting scope for a minimum duration of 30 minutes per visit. Observers were evenly distributed across opposite ends of the home range, and remained stationary at fixed points during the tracking period. A single GPS point was taken for each visually confirmed location. Two or more sessions were made on following days to collect a minimum of 30 GPS points for each individual, averaging 10 points per session. At each visual confirmation of a female, foraging activity was also noted if relevant (i.e. aerial hawking, perching, ground foraging).

2.2.3 Prey Selection

The color-marked females from the ten nests were monitored for prey selection. During the three visits at the nestbox for home range collection, a minimum of ten prey observations were made per visit. Each time the female of a nest returned with a prey item, it was visually identified to the Order and recorded. Nests were collected after nestlings fledged and stored in a freezer at –30°C until analysis. Prey in the mouth of an adult female was collected if caught in a mist net simultaneously. All prey was inventoried following each nest collection. Inventoried prey was compared with foraging observations and stored for future contaminant residue analysis.

2.3 Results

2.3.1 Productivity

Seven different species were found nesting in the boxes, with the majority being Western Bluebirds dominating over half the boxes (57%), Violet-green Swallows (17%), and Ash-throated Flycatchers (11%) (Table 2). LANL-wide occupancy was 20.9%, increased from 19.8% in 2004, and 18.3% immediately after the fire in 2000 (Table 3).

Table 3. LANL-wide occupancy for all species.

	1997	**1998**	**1999**	**2000**	**2001**	**2002**	**2003**	**2004**	**2005**
Percent Occupancy	9.2	22.3	21.2	18.3	16.1	18.9	27.7	19.8	20.9
Total Active Nests	40	97	92	71	81	99	172	122	139
Total Number of Boxes	435	435	435	388	502	523	636	616	666

Clutch size remained constant for WEBLs and increased for ATFLs (Table 4). The average clutch size for WEBLs and ATFLs was 5 and 4, respectively.

Table 4. Clutch size for western bluebirds and ash-throated flycatchers.

Year	WEBL	ATFL
1997	5	4
1998	4	4
1999	5	4
2000	4	3
2001	5	4
2002	4	3
2003	5	4
2004	5	3
2005	5	4

There was a slight increase in the percentage of eggs that successfully hatched in 2005 for WEBLs and ATFLs (Table 5).

Table 5. Percent of egg hatched for WEBLs and ATFLs.

Year	% WEBL Hatched	% ATFL Hatched
1997	78	75
1998	59	73
1999	82	79
2000	80	80
2001	72	63
2002	74	71
2003	66	61
2004	84	71
2005	85	77

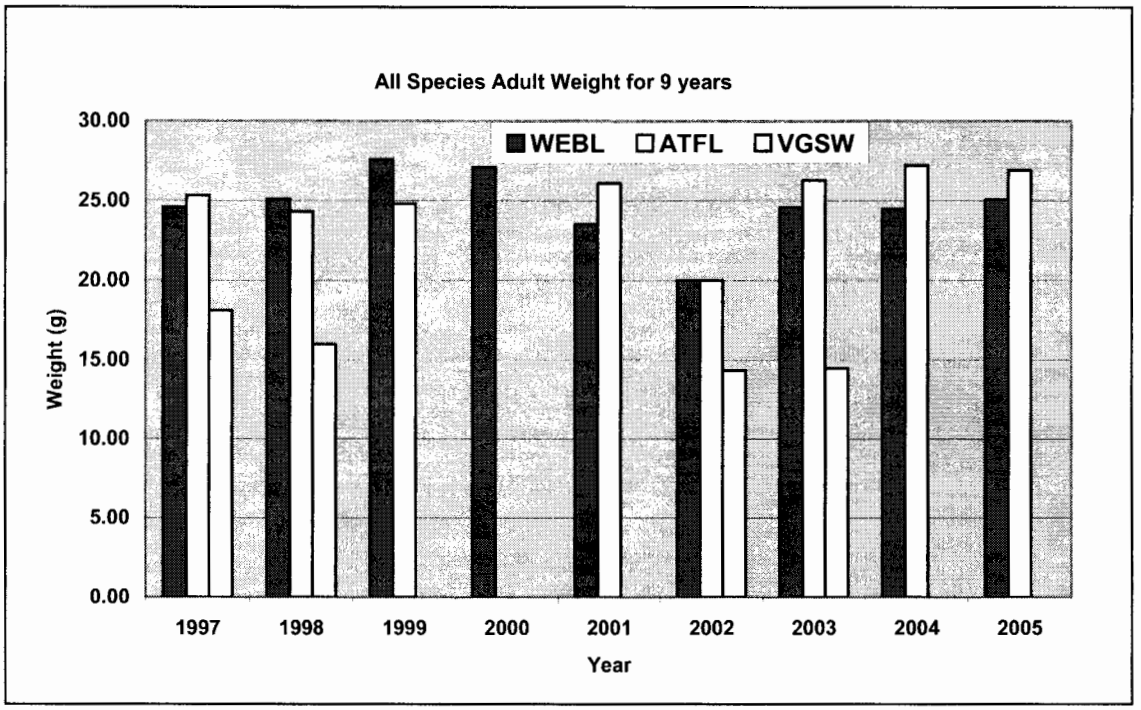
Average hatch date was approximately two weeks later in 2005 for WEBLs and ATFLs than in 2004, respectively (Table 6).

Table 6. Average Julian hatch date for WEBLs and ATFLs.

Year	WEBL Julian Date	ATFL Julian Date
1997	163	176
1998	167	168
1999	165	181
2000	170	171
2001	154	156
2002	166	175
2003	149	168
2004	152	148
2005	165	162

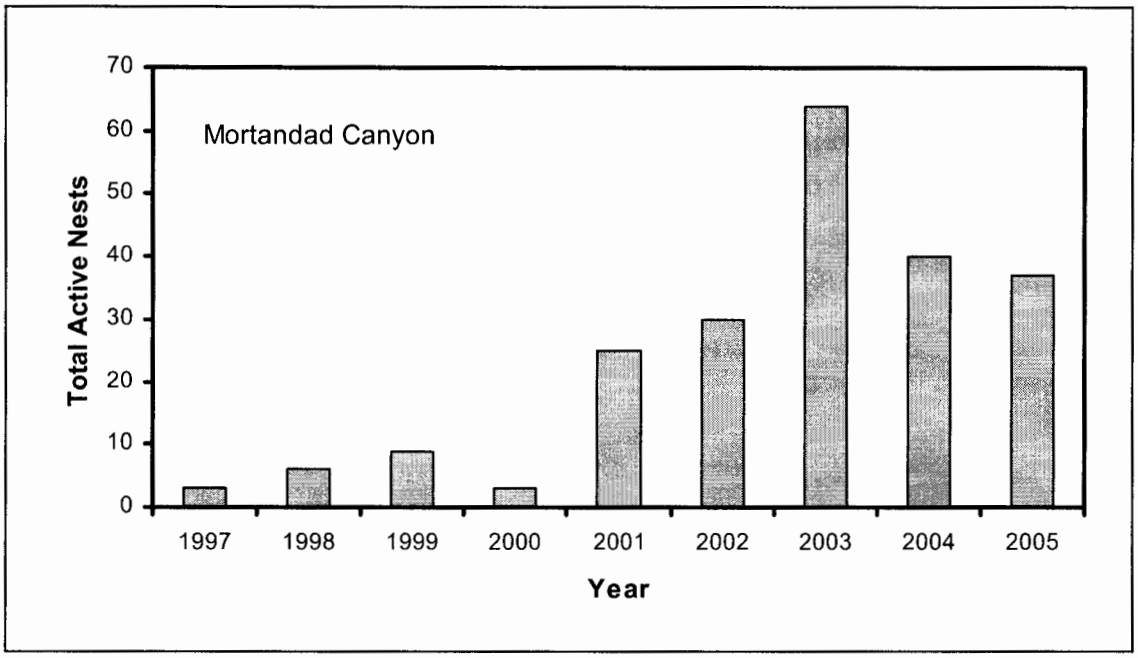
Adult condition can be used as an indicator of health across a population. Adult mass was measured for the three species and compared across nine years. Body weight lowered in 2001 and 2002 as a likely result of the Cerro Grande Fire, whereas it increased to pre-fire conditions in recent years (Figure 2). ATFL weight was considerably higher than WEBLs from 2003 through 2005.

Figure 2. Adult body weight (g) for 1997 to 2005.



Mortandad canyon continues to harbor one of the highest concentrations of potential release sites (PRS). In addition to which, the occupancy rate in Mortandad Canyon is the highest across the Lab (Figure 3). A combination of numerous PRS locations with high occupancy rates in Mortandad could result in subsequent effects on those species frequently foraging the canyon.

Figure 3. Total active nests (all species) for Mortandad Canyon from 1997-2005.



2.3.2 Home Range Foraging

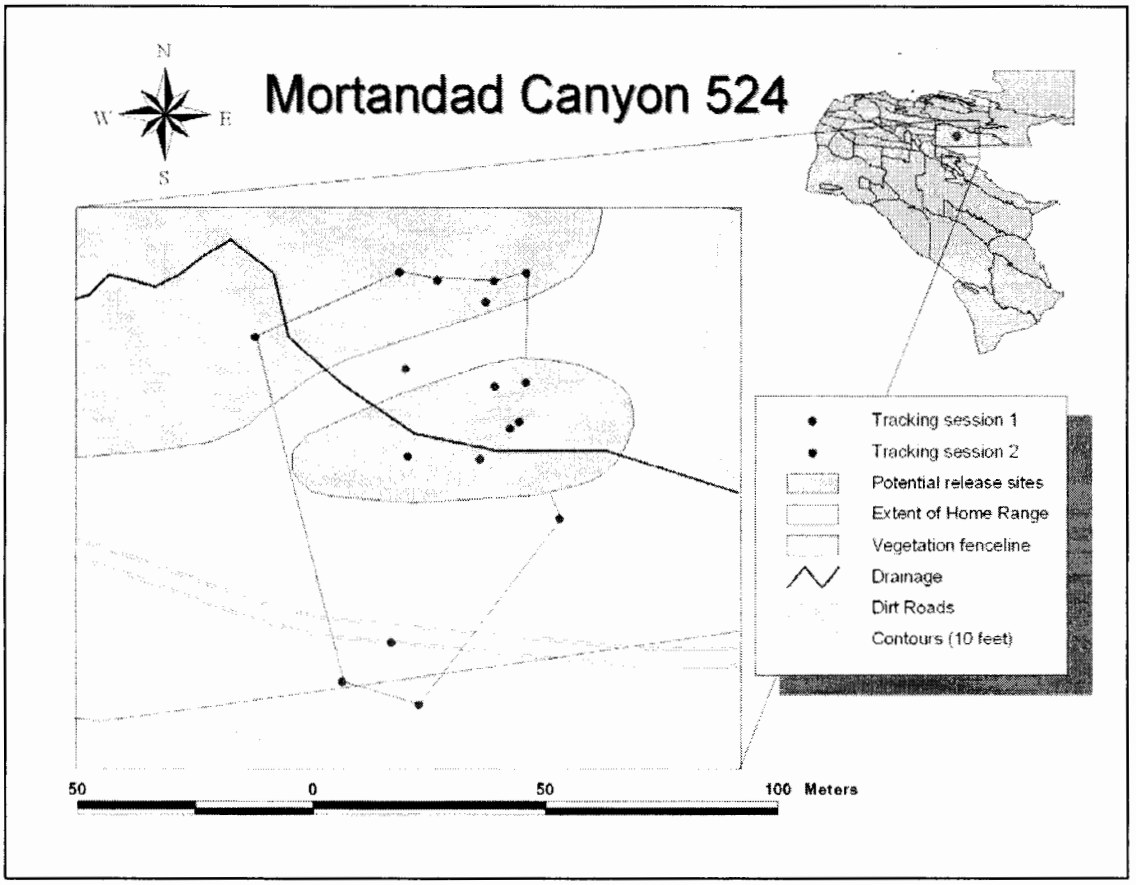
The average area for WEBLs home ranges was 5811 m^2 or 1.4 acres (Table 7). Analyses of the amount of PRS overlap are underway. WEBLs and ATFLs were found to forage between 50 to 100 m from the nestbox. Hot spots of contamination will be considered when examining the frequency of foraged locations within a home range. The amount of energy a bird utilized in any particular area may help determine contamination effects. This nestbox network project will be important in evaluating more accurately the connections between foraging dynamics and home range of passerines.

Table 7. Area of WEBL home ranges.

Location	HR #	Area (m^2)	Acres
Golf course	8A	4058	1.00
Golf course	8	7141	1.76
TA-35	742	1505	0.37
TA-48	736	1417	0.35
Golf course	6	5114	1.26
Golf course	5II	10246	2.53
Golf course	5AII	5756	1.42
Golf course	5A	11121	2.75
Canada del Buey	587	3166	0.78
Canada del Buey	582	2999	0.74
Canada del Buey	581	4302	1.06
Mortandad	551	2932	0.72
Mortandad	524	7839	1.94
Mortandad	522	15455	3.82
Mortandad	517	4387	1.08
Mortandad	513II	10585	2.62
Mortandad	513	10786	2.67
Mortandad	512	5393	1.33
Mortandad	511	4826	1.19
Golf course	5	8025	1.98
TA-35	30	6146	1.52
Cemetery	178	8640	2.14
Cemetery	175	4550	1.12
Cemetery	174	5562	1.37
Cemetery	172	2687	0.66
Cemetery	171	2658	0.66
Cemetery	168	4281	1.06
TA-51	326B	1123	0.28
Average		5811	1.44

Figure 4 illustrates an example 2004 WEBL home range in Mortandad canyon with PRS overlap of 46.1%.

Figure 4. Home range extent for nestbox 524 in Mortandad Canyon in 2004.



2.3.3 Prey Selection

Blowflies were the most abundant item found in WEBL nests, with the highest count in 2005 at an average of 42 per nest, compared to previous years (Table 8). The two most common insect Orders found in nests this year included *Hymenoptera* (ants, bees, etc.) and *Coleoptera* (beetles). We plan on obtaining contaminant residue analysis results this year on composite samples of prey by location. WEBL adult females were not

observed taking *Araneae*, *Diplopoda*, and *Diptera* to the nest, however they were found after the nestlings fledged and included in the final inventory.

Table 8. WEBL average number of prey and/or items found per nest.

Item	2003	2004	2005
Araneae	.	.	2
Blowflies	24	22	42
Catepillars/worms	1	2	2
Coleoptera	3	.	7
Diplopoda	1	10	2
Diptera	.	1	.
Hemiptera	1	2	1
Homoptera	1	.	1
Hymenoptera	1	14	13
Odonata	.	.	1
Orthoptera	1	1	2

3.0 Contaminant and Residue Exposure of Cavity-Nesting Birds

Each breeding season a percentage of eggs did not hatch or were abandoned. Table 9 shows the number of all unhatched eggs, as well as potentially viable eggs that were collected and refrigerated for contaminant residue analysis. Dead nestlings were also collected and frozen for residue analysis. There were 26 dead WEBL nestlings found (no other species) and 41 WEBL eggs collected in 2005 (Table 9).

Table 9. Collected eggs and dead nestlings for each species for 2005.

	WEBL	ATFL	VGSW	MOCH	MOBL	HOFI
Collected eggs	41	5	.	.	1	2
Dead nestlings	26	.	.	.	.	.

At total of 32 eggs and 14 composite insect samples from 2004 to 2005 were sent in for contaminant residue analysis (Table 10 and Table 11, respectively). All samples are currently being processed. Limits for eggs, in ppb, will be the detection limits divided by the mass of the eggs (g). Lower quantitation limit for most metal will be the 1-10 ppb

range. A variety of heavy metals will be analyzed from the eggs, including fifteen major elements (Table 12). Figure 5 illustrates all Mortandad sampling locations and corresponding reaches.

Table 10. Total WEBL egg samples for contaminant residue analysis from 2003-2005.

Date	Location	Box	Reach	<100 m from PRS	Analysis	Weight (g)
06/18/03	Mortandad	527	M-4W	X	Metal	.
06/27/03	Mortandad	548	M-4E		Metal	.
05/25/04	Cemetery	175	Cem	X	Metal	2.3
05/25/04	Golf Course	6	GC	X	Metal	2.5
06/02/04	Mortandad	520	M-5W		Metal	2.2
06/02/04	Mortandad	524	M-4	X	Metal	2.1
05/19/05	CDB	581	CDB		Metal	1.83
05/17/05	CDB	582	CDB		Metal	1.71
05/23/05	CDB	587	CDB		Metal	2.43
05/20/05	Cemetery	168	Cem	X	Metal	1.49
06/02/05	Cemetery	171	Cem	X	Metal	1.85
05/25/05	Cemetery	172	Cem	X	Metal	2.49
06/24/05	Cemetery	174	Cem	X	Metal	2.38
06/02/05	Cemetery	175	Cem	X	Metal	2.34
05/16/05	Cemetery	178	Cem	X	Metal	2.31
06/29/05	Golf Course	5	GC	X	Metal	1.91
05/17/05	Golf Course	6	GC	X	Metal	1.37
05/17/05	Golf Course	8	GC	X	Metal	1.1
05/31/05	Golf Course	5A	GC	X	Metal	2.55
06/29/05	Golf Course	5A	GC	X	Metal	2.4
06/29/05	Mortandad	510	M-5W		Metal	2.62
06/29/05	Mortandad	511	M-5W		Metal	2.48
05/17/05	Mortandad	512	M-5W		Metal	2.07
05/16/05	Mortandad	513	M-5W		Metal	1.99
06/29/05	Mortandad	513	M-5W		Metal	2.43
05/23/05	Mortandad	517	M-5W		Metal	1.53
06/28/05	Mortandad	522	M-4E		Metal	1.99
05/11/05	Mortandad	524	M-4	X	Metal	2.23
05/19/05	Mortandad	551	M-5W		Metal	2.24
05/26/05	TA-35	30	TS-2W	X	Metal	2.69
05/26/05	TA-48	736	E-1FW	X	Metal	2.87
05/24/05	TA-51	326B	CDB	X	Metal	2.1

	2003	2004	2005	
Potentially contaminated	1	3	15	
Controls	1	1	11	
Total 2003-2005	2	4	26	**32**

Table 11. Total composite insect samples for contaminant residue analysis from 2003-2005.

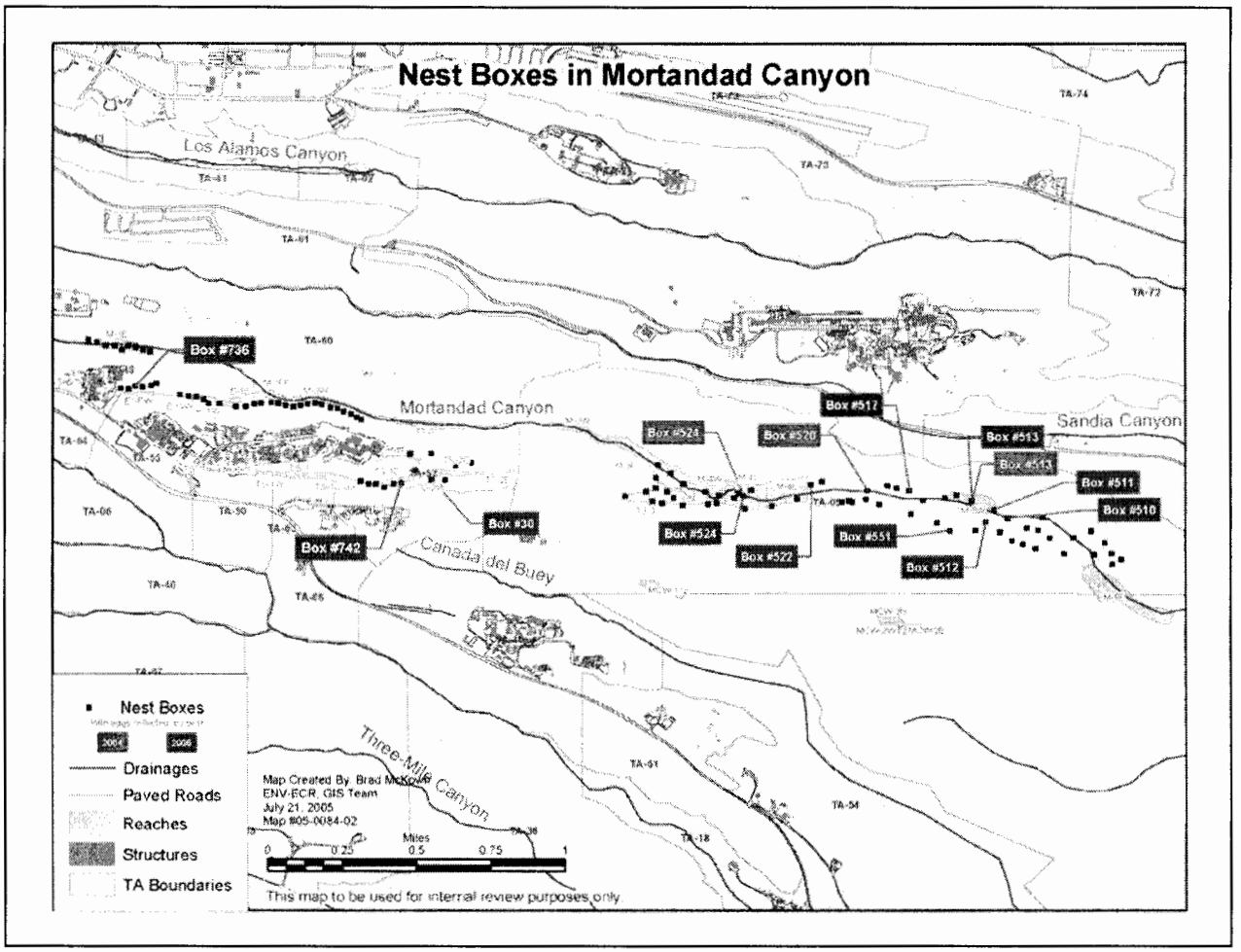
REACHES	Boxes	<100 m from PRS	Weight (g)
2003			
M-4W	527		0.35
M-4E	548		0.418
2004			
M-4	524	X	0.271
M-5W	513, 520		0.252
Cem	175	X	0.34
2005			
E-1FW	736	X	0.491
TS-1E	742	X	0.317
TS-2W	30	X	0.272
M-4	524	X	0.535
M-4E	522		0.115
M-5W	510, 511, 512, 513, 517, 551		2.701
GC	5, 5A, 6, 8A, 8	X	2.497
Cem	168, 171, 172, 174, 175, 178	X	1.253
CDB	581, 582, 587		1.565

	2003	2004	2005	
Potentially contaminated	0	2	6	
Controls	2	1	3	
Total 2003-2005	2	3	9	**14**

Table 12. Elements analyzed in eggs.

Ag
As
Ba
Be**
Cd
Co
Cr
Cu
Hg
Mn
Pb
Sb*
Se
Tl
Zn

Figure 5. Map of Mortandad canyon reaches and overlaying boxes with sampled eggs from 2003-2005.



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