# BAT SPECIES ABUNDANCE AND DISTRIBUTION, THE EFFECTS OF FOREST THINNING AND BURNING ON BAT FORAGING ACTIVITY, INCIDENCE OF WEST NILE VIRIS IN BATS, AND CALCIUM WATER HOLE EXPERIMENTS AT HEIL VALLEY RANCH, 2006



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### SUMMARY POINTS FOR 2004 FIELD SEASON

- ➤ 142 bats were captured and released
- Most captures during study period were *Myotis evotis*, approx. 39%
- > 704 unknown sonar calls was recorded
- ▶ More than 100 hand-release calls were recorded for the Colorado bat sonar library
- Most unknowns were recorded in Thinned forest plots
- Most biomass of insects was highest in Thinned plots
- > Diptera and Lepidoptera made up the majority of insects captured in light traps
- Blood was drawn from 8 bats for WNV testing, with a single female *M. evotis* testing positive for WNV antibodies
- ▶ 41 bats were PIT-tagged, of these 16 (38%) were reacquired at the artificial water hole
- > Of 24 female *M. thysanodes* PIT-tagged, 14 were reacquired (54%)
- ▶ Of 93 drinking passes, 90 (97%) were at the higher calcium water hole

<u>Abstract</u>: In 2006 we conducted research at Heil Valley Ranch (HVR) to continue 1) measuring bat species abundance and distribution that was begun in 2002, 2) measuring the effects of forest thinning on bat foraging patterns, 3) measuring blood antibody levels to West Nile virus (WNV) infections and 4) integrating a novel PIT-tag system reader that allowed us to follow visitation patterns of individuals at water holes. In addition, we ran an experiment on calcium water hole preferences of lactating female bats.

Netting began on 8 June and continued until 3 October. A total of 142 bats were captured (Table 1). Of these, 55 were *M. evotis*, 43 were *M. lucifugus*, 36 *M. thysanodes*, 4 *M. ciliolabrum*, 1 *M. volans*, and 2 *C. townsendii*.

Blood was drawn from 8 individuals for testing for WNV antibodies in 2006.

Sonar data collected from meadow, thinned, unthinned and burned sites at HVR continue to indicate that thinned and meadow areas support the highest bat species richness and species evenness at HVR. Insect biomass and diversity data gathered from sonar plot sites showed that Thinned areas contained highest total insect biomass. Kruskal-Wallis one-way ANOVA on ranks showed a significant difference among plots in insect biomass collected (H = 8.77, p = 0.033). Insect traps in Meadows collected the least amount of insect biomass. Biomass across plots showed that Diptera were the most insects captured in Meadow, Thinned and Forest plots, with Thinned areas having a much higher proportion of Diptera than the others. Lepidoptera came in second in biomass across plots, with the exception of Burn plots which where Lepidoptera dominated. Coleoptera were only collected from the Burn and Forest plots, but they were in relatively low numbers. Meadows showed the greatest balance of insect diversity across orders.

Using the new PIT-tag system the choice test for calcium loaded water hole was run from 8/10 till 9/13. Over this time period, 90 visits, all females, were recorded for the artificial calcium water hole versus 3 passes by a single male bat at the natural water hole.

## **INTRODUCTION**

Three aspects of the study of bats at Heil Valley Ranch (HVR) have evolved over the past four years including Species Abundance and Distribution, Forest Thinning and Bat Foraging Patterns, and Incidence of West Nile Virus in bats at HVR. In 2006 a new and innovative PIT-tagging system for understanding water use patterns by bats at water holes was initiated.

**Part I:** *Tracking Species Abundance and Distribution:* Because bats are difficult to catch and they change their foraging patterns and areas seasonally, long-term studies of bat populations are required to ascertain presence and abundance of bat populations. In 2004, a previously undocumented eastern bat species (*Pipistrellus subflavus*) was found on OSMP property in north Boulder (Armstrong et al., 2005) In addition, because bats are susceptible to human disturbance, infectious diseases, and are responsive to climate variation, year-to-year patterns may shift and thus require long-term efforts to understand regional ecology, population dynamics and stability. In this third year of capture and release data collection, we are beginning to better understand ecological patterns of bats at HVR in Boulder County.

In 2002, we began mist netting bats at ephemeral and permanent water holes throughout HVR. In addition, radio tagging of some lactating females allowed us to locate and map maternity roost sites as well as conduct outflight counts and document emergence times of various colonies and species. These data are paramount in the management and conservation of bat species in the West. In 2006, we continued with our mist netting efforts that contributed new information on species abundance and species presence at HVR.

*Part 2: Forest Thinning Practices*: Protecting critical foraging habitats for bats is of paramount importance. Loss of critical foraging habitat can affect the stability and survivorship of bat

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populations. Several critical factors need be in balance. For insectivorous bats, foraging in less cluttered habitats is most energy efficient because obstacle avoidance is limited as they hunt. However, foraging in open areas has its own risks, such as predation from owls at night, or other raptors before darkness (Erickson and West, 2002). *Myotis* bats also show choice in selecting habitat types along a riparian corridor in Utah, whereas *Eptesicus fuscus* showed no significant differences among habitats (Rogers, 2006).

Human impacts to foraging habitats usually come in the form of forest cutting and various other degradations. Clear-cutting practices have likely caused the loss of some bat populations, however, the overall effects will never truly be known due to lack of precutting sampling for bats. Studies in the West indicate bat activity is low where clear-cutting has occurred. Conversely, the less-severe practice of forest thinning may enhance bat foraging areas (Parker et al., 1996; Perdue and Steventon, 1996; Humes et al., 1999; Patriquin and Barclay, 2003). However, Tibbels and Kurta (2003) found that thinned areas of red-pine did not enhance foraging areas for bats which instead used intra-forest clearing more predominately. In 2003, we began a study to understand the effects of forest thinning practices currently underway at HVR using set 0.25 hectare plots in four habitat types. This study continued in 2006, but monitoring for insects occurred in each plot beginning in 2006.

*Part 3: Incidence of West Nile Virus in HVR Bat Populations*: Insectivorous bats are the predominant foragers of night-flying insects, including adult mosquitoes (Gould 1955; Griffin et al. 1960; Findley, 1993; Altringham, 1996). In Colorado, myotis species consume mosquitoes in variable amounts (Adams, 2003). In Moffat County, *M. lucifugus* consumed 21% of their diet in flies and mosquitoes (Diptera), with other myotis species consuming about 10% of their diet in flies (Freeman, 1984). Diets of bats, however, differ regionally. Adams (1993, 1997) found

seasonal differences in consumption of species of Diptera by *M. lucifugus*, from 28% in spring to 38% in fall. The relationship between mosquito consumption and incidence of WNV in bats remains unknown. However, some bat species have tested positive for WNV infection in New York (Marra et al., 2003; Davis et al., 2005). Because Boulder, Larimer, and Weld counties were the "hot-zone" for human cases of WNV infections in 2003, we initiated a study to document the incidence of WNV infection in *Myotis* species at HVR in 2004 and continued this monitoring in 2005. We did not intend to check for WNV in bats in 2006, but we did gather blood samples in September because there was a sudden rise in incidence of WNV in Boulder County.

*Part 4.—Monitoring activity at water holes and testing the calcium hypothesis*: Adams (2003) showed a significant correlation between the amount of dissolved calcium in the water and visitation by reproductive female bats and their young. In 2006, Adams received support from Boulder County Parks and Open Space, The Lois Webster Foundation, Boulder County Nature Association, and the University of Colorado to purchase a BioMark, Inc., passive integrative transponder (PIT) tagging system with waterproof plate antennae and reader. This system gave previously unknown insights into water use by bats at HVR and in the future will continue to give such insights.

### **METHODS**

*Capture and Release*: We continued mist netting bats in Geer Canyon and at Ingersol Quarry. All captures were made in mist nets, and all individuals were released within one hour of capture.

*Marking & PIT-tag data acquisition*.—Bats captured in Geer Canyon were PIT-tagged with BioMark 12mm tags under the dorsal skin between the shoulder blades. Individuals were kept

for 20 minutes to ensure closure of the entry wound, and then released. A plate antennae and recorder were positioned in an artificial water holes west of a maternity colony of *Myotis thysanodes*.



**Figure 1**. (left) plate antennae placed 1 cm below water surface in artificial water hole, (middle) water proof data recording station with solar panel, data reader, antennae tuner, and car battery, and (right) PIT-tagging station with ring antennae reader.

We tested two *Null Hypotheses*:  $HO_1$ : There will be no significant differences in drinking patterns between reproductive and none reproductive female *M. thysanodes*.  $HO_2$ : There will be no significant difference in number of drinking passes recorded for female reproductive *M. thysanodes* between the artificial calcium water hole and the natural water hole.

*Bat foraging pattern surveys*: Pettersson 240x time-expansion, sonar detectors interfaced with Sony tape recorders were position in 0.25 hectare fixed plots in unthinned forest, recently thinned forest, open meadows, and a burned site in Geer Canyon. *Null hypotheses:*  $H_0$ : There are no significant differences in bat foraging activity as measured by sonar pass recordings between unthinned, human-thinned, montane meadow, and natural burned habitats (i.e. treatment plots).  $H_1$ : Bat species composition will not be significantly different between treatment plots.

*Sonar Library Hand-Release Recordings*: We recorded sonar calls of hand-released known individuals for comparison to unknown calls collected in the sonar plots.

Insect Diversity and Sonar Plots: In 2006 we hung insect light traps in each plot on night when

sonar recordings were in progress and at times when sonar plots were not in progress. The lights were hung at sunset and allowed to run for three hours after sunset to sample for insects during the time interval that bat sonar grids were run.

*Blood Sampling for West Nile Virus*: Bats were captured in mist nets and anaesthetized using Isoflurane. Approximately 30  $\mu$ L of blood was drawn from an artery in the interfemoral membrane by puncturing with a 25 gauge needle. Samples were collected in heparinized, glass capillary tubes (Lollar and Schmidt-French 2002; Kunz and Nagy 1988). Pressure was applied to the wound with the researcher's index finger, until blood-loss ceased. Individuals were returned to capture sacs for 20 minutes to ensure that bleeding did not reoccur. Individuals were then released. Blood samples were put on ice and later spun down using an Autocrit Ultra 2 micro-centrifuge and stabilized using Ambion, Ribopure blood kits. Samples were analyzed using a 1-step RT-PCR kit (Ambion) called Retroscript (Kauffman, et al., 2003).

*Null hypothesis*: H<sub>0</sub>: There will be no antibodies present for WNV in bats at HVR.

#### RESULTS

*Capture Data:* Netting began on 8 June and continued until 3 October. A total of 142 bats was captured (Table 1). Of these, 55 were *M. evotis*, 43 were *M. lucifugus*, 36 *M. thysanodes*, 4 *M. ciliolabrum*, 1 *M. volans*, and 2 *C. townsendii*.

DATE	SITE	SPP	SEX	AGE	WGT	REPRO	Bled
6/8/2006	Ingersol	M. evotis	М	А	5.6	NS	
6/8/2006	Ingersol	M. evotis	F	А	8.4	NLNP	
6/8/2006	Ingersol	M. evotis	М	А	5.6	NS	
6/8/2006	Ingersol	M. evotis	М	А	4.9	NS	
6/24/2006	Ingersol	M. evotis	М	А	4.9		
6/24/2006	Ingersol	M. evotis	F	А	8.4	NLNP	
6/24/2006	Ingersol	M. evotis	F	А	7.9	Р	
6/24/2006	Ingersol	M. evotis	F	А	8.2	L	

**Table 1.** Capture and release data from 2006, organized by date and capture site.

C/24/2000		M avatia		٨	6.0	П	
6/24/2006	Ingersol	M. evotis	F	A	6.8	P	
6/24/2006	Ingersol	M. evotis	M	A	7.1	NS	
6/24/2006	Ingersol	M. luci	M	A	6.5	NS	
6/24/2006	Ingersol	M. luci	M	A	8	NS	
6/24/2006	Ingersol	M. luci	M	A	No Wgt	NS	
6/24/2006	Ingersol	M. luci	M	A	7.3	NS	
6/24/2006	Ingersol	M. luci	М	A	6.9	NS	
6/24/2006	Ingersol	M. luci	M	A	No Wgt	NS	
6/24/2006	Ingersol	M. luci	М	Α	6.9	NS	
6/24/2006	Ingersol	M. luci	М	Α	8.3	NS	
6/24/2006	Ingersol	M. luci	M	А	No wgt	NS	
6/24/2006	Ingersol	M. thys	M	A	7.9	NS	
6/24/2006	Ingersol	M. evotis	F	А	7.9	Р	
6/24/2006	Ingersol	M. evotis	F	А	6.8	Р	
6/24/2006	Ingersol	M. evotis	М	А	7.1	NS	
6/24/2006	Ingersol	M. evotis	F	А	8.2	L	
6/24/2006	Ingersol	M. evotis	escaped		no wgt		
6/24/2006	Ingersol	M. evotis	escaped		no wgt		
6/24/2006	Ingersol	M. luci	М	А	7.2	NS	
6/24/2006	Ingersol	M. luci	М	А	6.5	NS	
6/24/2006	Ingersol	M. luci	М	А	8	NS	
6/24/2006	Ingersol	M. luci	М	А	no wgt	NS	
6/24/2006	Ingersol	M. luci	М	А	7.3	NS	
6/24/2006	Ingersol	M. luci	М	А	no wgt	NS	
6/24/2006	Ingersol	M. thys	М	А	7.9	NS	
7/13/2006	Lower Geer	СОТО	М	А	No wgt	NS	
7/13/2006	Lower Geer	СОТО	М	А	No wgt	NS	
7/13/2006	Lower Geer	M. evotis	F	А	No wgt	L	
7/13/2006	Lower Geer	M. evotis	F	А	No wgt	NLNP	
7/13/2006	Lower Geer	M. luci	М	А	No wgt	NS	
7/13/2006	Lower Geer	M. luci	М	А	No wgt	NS	
7/13/2006	Lower Geer	M. luci	М	А	No wgt	NS	
7/13/2006	Lower Geer	M. luci	М	А	No wgt	NS	
7/13/2006	Lower Geer	M. thys	F	А	No wgt	L	
7/13/2006	Lower Geer	M. thys	F	А	No wgt	L	
7/13/2006	Lower Geer	M. thys	F	А	No wgt	L	
7/13/2006	Lower Geer	M. thys	F	А	No wgt	L	
7/13/2006	Lower Geer	M. thys	F	A	No wgt	L	
7/13/2006	Lower Geer	M. thys	F	A	No wgt	L	
7/13/2006	Lower Geer	M. thys	F	A	No wgt	 L	
7/13/2006	Lower Geer	M. thys	M	A	No wgt	NS	
7/13/2006	Lower Geer	M. thys	F	A	No wgt	L	
7/13/2006	Lower Geer	M. thys	F	A	No wgt	 L	
7/13/2006	Lower Geer	M. thys	F	A	No wgt	 L	
7/15/2006	Lower Geer	M. thys	F	A	No wgt	 L	
7/15/2006	Lower Geer	M. thys	F	A	No wgt	<b>–</b>	
1/10/2000		111. 11193	I	7	No wgi		1

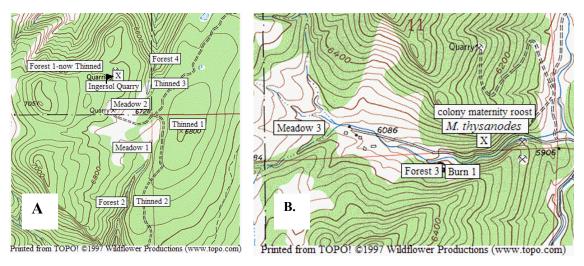
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7/15/2006	Lower Geer	M. thys	M	A	No wgt	NS	
7/15/2006	Lower Geer	M. thys	F	A	No wgt	L	
7/15/2006	Lower Geer	M. thys	F	A	No wgt		
7/15/2006	Lower Geer	M. thys	F	A	No wgt	NLNP	
7/15/2006	Lower Geer	M. thys	F	A	No wgt	L	
7/15/2006	Lower Geer	M. thys	F	A	No wgt	L	
7/15/2006	Lower Geer	M. thys	F	A	No wgt	NLNP	
7/18/2006	Lower Geer	M. thys	F	Α	No wgt	L	
7/18/2006	Lower Geer	M. thys	М	Α	No wgt	NS	
7/18/2006	Lower Geer	M. thys	F	A	No wgt	NLNP	
7/19/2006	Ingersol	M. cilio	М	A	9.2	NS	
7/19/2006	Ingersol	M. evotis	М	A	5.8	NS	
7/19/2006	Ingersol	M. evotis	М	A	6.3	NS	
7/19/2006	Ingersol	M. evotis	М	A	6.2	NS	
7/19/2006	Ingersol	M. evotis	М	A	6.8	NS	
7/19/2006	Ingersol	M. evotis	F	А	7.1	L	
7/19/2006	Ingersol	M. evotis	F	Α	6.9	NLNP	
7/19/2006	Ingersol	M. evotis	F	Α	6.3	NLNP	
7/19/2006	Ingersol	M. evotis	М	А	6.8	NS	
7/19/2006	Ingersol	M. evotis	М	Α	7.4	NS	
7/19/2006	Ingersol	M. evotis	М	Α	6.5	NS	
7/19/2006	Ingersol	M. evotis	М	А	6.1	NS	
7/19/2006	Ingersol	M. evotis	М	А	6.4	NS	
7/19/2006	Ingersol	M. evotis	М	А	8	NS	
7/19/2006	Ingersol	M. luci	М	А	6.4	NS	
7/19/2006	Ingersol	M. luci	F	А	6.1	L	
7/19/2006	Ingersol	M. luci	F	Α	7.5	L	
7/19/2006	Ingersol	M. luci	М	А	7	NS	
7/19/2006	Ingersol	M. luci	М	А	6.6	NS	
7/19/2006	Ingersol	M. luci	F	А	7.4	NLNP	
7/19/2006	Ingersol	M. luci	F	А	7.1	NLNP	
7/19/2006	Ingersol	M. luci	F	SA	7.4	NLNP	
7/19/2006	Ingersol	M. luci	F	SA	7.7	NLNP	
7/19/2006	Ingersol	M. luci	М	J	6.7	NS	
7/19/2006	Ingersol	M. luci	М	А	7.1	NS	
7/19/2006	Ingersol	M. luci	М	Α	7.4	NS	
7/19/2006	Ingersol	M. luci	М	Α	6.6	NS	
7/19/2006	Ingersol	M. luci	М	Α	6.3	S	
7/19/2006	Ingersol	M. thys	М	Α	7	NS	
7/19/2006	Ingersol	M. thys	F	Α	8.6	L	
7/19/2006	Ingersol	M. thys	М	Α	8.2	NS	
7/19/2006	Ingersol	M. thys	M	A	7.2	NS	
7/24/2006	Lower Geer	M. cilio	M	A	5	NS	
7/24/2006	Lower Geer	M. evotis	M	A	No wgt	NS	
7/24/2006	Lower Geer	M. thys	F	A	No wgt	L	
7/24/2006	Lower Geer	M. thys	F	A	No wgt	L	
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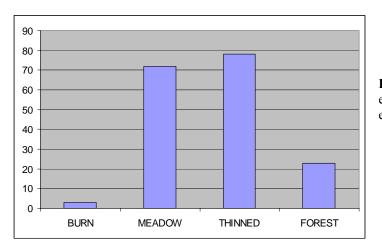
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7/24/2006	Lower Geer	M. thys	F	A	No wgt	L	
7/24/2006	Lower Geer	M. thys	F	А	No wgt	NLNP	
7/24/2006	Lower Geer	M. thys	F	А	No wgt	L	
7/26/2006	Lower Geer	M. evotis	М	А	No wgt	NS	
7/26/2006	Lower Geer	M. evotis	М	А	No wgt	NS	
7/27/2006	Upper Geer	M. cilio	escaped	А	No wgt	NS	
7/27/2006	Upper Geer	M. evotis	F	А	No wgt	NLNP	
7/27/2006	Upper Geer	M. evotis	F	А	No wgt	L	
8/3/2006	Ingersol	M. evotis	М	А	7.1	NS	
8/3/2006	Ingersol	M. evotis	F	А	6.5	PL	
8/3/2006	Ingersol	M. luci	М	А	7.1	NS	
8/3/2006	Ingersol	M. luci	М	А	6.3	NS	
8/3/2006	Ingersol	M. luci	М	А	6.4	NS	
8/3/2006	Ingersol	M. luci	М	А	7.8	NS	
8/3/2006	Ingersol	M. evotis	М	А	7.2	NS	
8/3/2006	Ingersol	M. evotis	М	А	7.1	NS	
8/3/2006	Ingersol	M. evotis	F	А	6.5	L	
8/3/2006	Ingersol	M. luci	М	А	7.1	NS	
8/3/2006	Ingersol	M. luci	М	А	6.3	NS	
8/3/2006	Ingersol	M. luci	М	А	6.4	NS	
8/3/2006	Ingersol	M. luci	М	А	7.8	NS	
8/8/2006	Upper Geer	M. thys	М	А	No wgt	NS	
8/8/2006	Upper Geer	M. thys	М	J	No wgt	NS	
8/22/2006	Ingersol	M. evotis	F	А	No wgt	NLNP	Bled
8/22/2006	Ingersol	M. evotis	М	А	No wgt	NS	Bled
8/22/2006	Ingersol	M. evotis	М	А	No wgt	NS	Bled
8/22/2006	Ingersol	M. evotis	М	А	No wgt	NS	Bled
8/22/2006	Ingersol	M. luci	М	А	No wgt	NS	Bled
8/22/2006	Ingersol	M. luci	М	А	No wgt	NS	Bled
8/22/2006	Ingersol	M. volans	М	А	No wgt	NS	Bled
8/22/2006	Ingersol	M. cilio	М	А	3.9	S	
8/22/2006	Ingersol	M. evotis	М	А	6	NS	
8/22/2006	Ingersol	M. evotis	F	А	5.7	NLNP	
8/22/2006	Ingersol	M. evotis	F	А	5.9	NLNP	
8/22/2006	Ingersol	M. evotis	М	А	5.8		
8/22/2006	Ingersol	M. evotis	М	А	5.9	NS	
8/22/2006	Ingersol	M. evotis	М	А	6.5	NS	
8/22/2006	Ingersol	M. evotis	М	SA	7.1	NS	
8/22/2006	Ingersol	M. evotis	М	А	5.6	NS	
8/22/2006	Ingersol	M. evotis	М	А	6.4	NS	
9/3/2006	Ingersol	M. evotis	М	А	6.2	NS	

*Sonar Plot Analyses*: Forest, Burn, Meadow, and Thinned plots were sampled 3 times each in 2006. Fifty four detector nights resulting in 704 bat sonar passes. Of these 544 (77.3%) were

*Myotis* species, 116 (15.5%) we *Eptesicus fuscus*, and 44 (6.2%) were either *Lasiurus cinereus* or *Lasionycteris noctivagans*. Highest numbers of passes were recorded in Thinned, Meadow, Forest, and Burn plots respectively (Fig. 3).

**Fig. 2.** Locations of <sup>1</sup>/<sub>4</sub> hectare sonar plots in Meadow, Thinned, Forest, and Burn habitats **A**) near Ingersol Quarry and **B**) in Geer Canyon.





**Fig. 3.** Total numbers of passes recorded for each sonar test plot. All plots were sampled equally.

Analysis of sonar calls by general showed that Meadows in 2006 had the most equal balance of species use. Thinned areas were used predominately by *Myotis* species, although *Eptesicus* and *Lasiurus* were recorded there on occasion (Fig. 4).

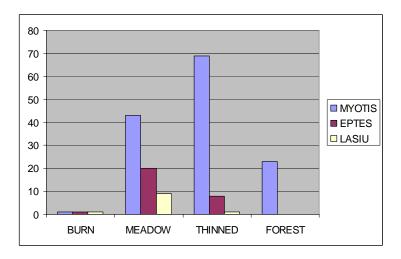


Fig. 4. Number of sonar passes by genus per sonar plot.

*Insect Diversity by Sonar Plot*: Fifteen sampling nights gave a total Biomass per plot (Fig 5) shows that Thinned areas contained highest total insect biomass. Kruskal-Wallis one-way ANOVA on ranks showed that a significant difference occurred in insect biomass collected (H = 8.77, p = 0.033). Insect traps in Meadows collected the least amount of insect biomass. It should be noted, however, that the type of insect traps used might be biased towards smaller body-sized insects.

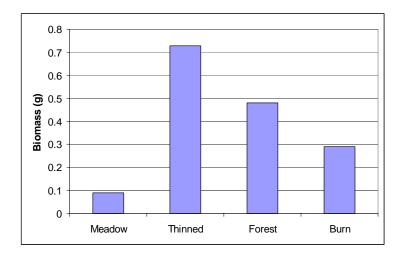
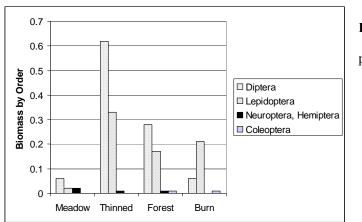


Fig. 5. Biomass of insects per plot type in grams.

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Biomass across plots (Fig 6) shows that dipterids were the most insects captured in Meadow, Thinned in Forest plots, with Thinned areas having a much higher proportion of Diptera. Lepidopterids come in second in biomass across plots, with the exception of Burn plots where lepidopterids dominated. Coleopterids were only collected from the Burn and Forest plots, but they were in relatively low numbers. Meadows showed greatest balance of insect diversity across orders.



**Fig. 6**. Biomass of insects by order in each sonar plot in grams.

*PIT-Tag Reacquisition*: Of 29 *M. thysanodes* individuals PIT-tagged, 14 were reacquired (48%). Of the 24 female *M. thysanodes*, 13 were reacquired (54%). Of seven *M. evotis* PIT-tagged, two were reacquired. Three *M. lucifugus* were PIT-tagged, but none were reacquired. Two male *C. townsendii* were PIT-tagged and none were reacquired (Table 2).

M. thysanodes	M. evotis	M. lucifugus	

Table 2. Numbers of bats PIT-tagged by species and sex

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	M. thysanodes	M. evotis	M. lucifugus	C. townsendii
Males	0	3	3	2
Females	29	4	0	0

Reacquisition data gathered at the artificial water hole gave precise visitation times of PIT-

tagged individuals to the second (Table 3).

Night 1	Night 2	Night 3	Night 4
20:52:23	20:56:51	20:38:23	20:51:09
20:52:29	20:56:52	20:38:29	5:00:41
20:52:35	20:56:58	20:38:39	5:00:48
20:52:51	20:57:05	20:45:34	
20:52:52	20:57:12	20:45:40	
20:59:24	4:49:46	20:45:53	
0:14:53	4:49:51	23:38:31	
0:14:54			

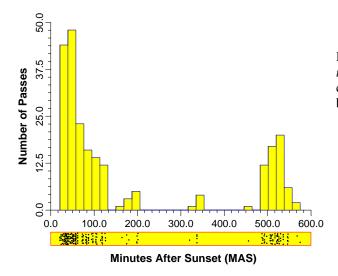
**Table 3.** Example of PIT-tag reacquisition data for a female, lactatingM. thysanodes, PIT-tag number 1BF249802D.

Data sorted between lactating and nonreproductive females showed that lactating females drank significantly more than did nonreproductive females. Table 4 shows that lactating females averaged 21 passes per night (SD 8.59), whereas nonreproductive females averaged 2.5 (SD 1.37) passes per night. The difference was significant (p = 0.001). Range for number of drinking passes by individual lactating females was 3 to 16 drinking passes per night.

**Table 4.** Numbers of lactating versus nonlactating M. thysanodes, females visiting the artificial water hole by date. There was significant difference between groups in number of passes. Kruskal-Wallis, p = 0.001

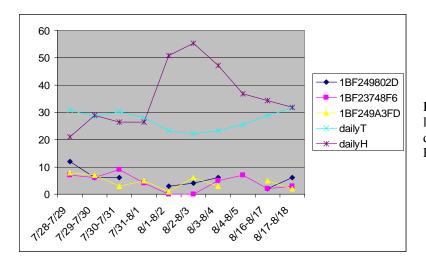
Date	Lactating (10)	<u>NonRepro (5)</u>
7/28-7/29	42	4
7/29-7/30	32	1
7/30-7/31	18	
7/31-8/1	13	1
8/1-8/2	24	
8/2-8/3	18	
8/3-8/4	14	
8/4-8/5	18	
8/16-8/17	22	3
8/17-8/18	16	4
8/22-8/23	19	2
SUM	236*	15
MEAN	21.45	2.5
SD	8.595	1.378

Analysis of visitation patterns of pooled lactating female data showed a bimodal drinking pattern with the majority of passes occurring directly after evening emergence. There was also significant drinking at dawn, before re-entering the dayroost (Fig 7).



**Figure 7**. Histogram of utilization times for lactating *M*. *thysanodes* at the artificial water hole. Most visitations were clustered directly after evening emergence from the roost and before re-entering the roost at dawn.

Data from a temperature-humidity data logger (Endurance, Inc.) placed at the water hole, shows fluctuation in these variables and some fluctuation in the visiting patterns of three lactating females that visited the water hole.



**Figure 8**. Line graph of three female lactating *M. thysanodes* plotted against daily Temperature (T) and daily Humidity (H).

Deriving an index of temperature and humidity (temperature/humidity) and testing the visitation patterns of reproductive and nonreproductive female *M. thysanodes*, Pearson-rank correlation showed no significant correlation between T/H Index and lactating females (r = 0.51, p = 0.13); whereas there was a significant correlation with drinking patterns of nonreproductive females (r = 0.63, p = 0.05). Lactating females showed high numbers of drinking passes regardless of temperature and humidity (Table 5).

TH Index	Lactating	NonRepro	Date
1.477327	42	4	7/28-7/29
0.9827049	32	1	7/29-7/30
1.1467577	18		7/30-7/31
1.0669456	13	1	7/31-8/1
0.4549744	24		8/1-8/2
0.4011218	18		8/2-8/3
0.4926922	14		8/3-8/4
0.6964091	18		8/4-8/5
0.8456592	22	3	8/16-8/17
0.9943271	16	4	8/17-8/18

**Table 5**. Temperature/Humidity indices as compared to visitation patterns of lactating and nonlactating M. thysanodes by date.

*Calcium Hypothesis Test*: The choice test for calcium loaded water hole was run from 8/10 till 9/13. Because I had only one plate antennae, it had to be moved back and forth between the artificial calcium site and the natural water hole. Over this time period, 90 visits, all females, were recorded for the artificial calcium water hole versus 3 passes by a single male bat at the natural water hole (Table 6)

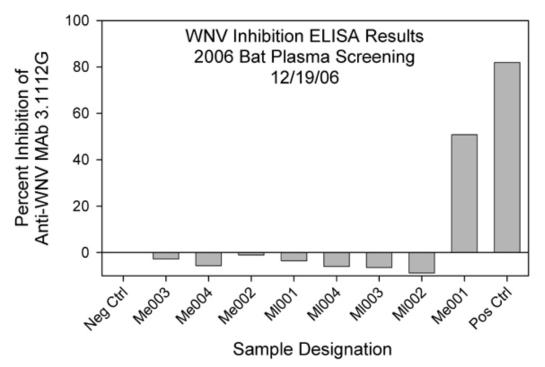
Date	Artificial	Natural
8/10-8/11		0
8/11-8/12		0
8/12-8/13		0
8/13-8/14		0
8/14-8/15		0
8/15-8/16		0
8/16-8/17	25 ♀, 0 ♂	
8/17-8/18	21 ♀, 0 ♂	
8/22-8/23	26 ♀, 0 ♂	

**Table 6**. Number of passes by sex at artificial versus natural water holes between the dates of 8/10 and 9/13.

8/28-8/29		0
8/29-8/30		0
8/30-9/1		3 ♂, 0 ♀
9/3-9/4	0	
9/4-9/5	9♀,0♂	
9/12-9/13	9♀,0♂	
TOTAL	<b>90</b> ♀, <b>0</b> ♂	3 ♂, 0 ♀

*West Nile Virus Data:* Eight individuals were bled to check for WNV antibody presence. A single adult female, nonreproductive *M. evotis* tested positive for antibodies (Fig. 9) indicating that it was infected with West Nile virus and survived.

**Figure 9.** Results from ELISA test on blood samples for incidence of West Nile Virus inhibin response among. Me =  $Myotis \ evotis$ , Ml = M. *lucifugus*. Me001 showed positive results for WNV antibodies.



#### **DISCUSSION**

Capture data over the last five years shows that the majority of captures at HVR are males (365 of 550 captures, 64%); whereas females compose about 33% of all captures (185 of 550

captures). In 2006, approximately 65% of captures were males, whereas 35% were females

(Table 7).

**Table 7**. Comparative capture data by male/female across four years at HVR for individuals of known sex.
 BNN =

 bats per net per night.
 BNN =

Species	2002	2003	2004	2005	2006	Total
MYCI	1 (♀)	7 (4♀, 3♂)	7 (1♀, 6♂)	5 (1♀, 4♂)	3 (ථ)	23 (7 ♀, 16 ♂)
MYEV	21 (60, 113)	15 (9♀, 6♂)	34 (9♀, 25♂)	61 (19♀, 42♂)	53 (19♀, 34♂)	184 (62 ♀, 118 ♂)
MYLU	23 (120, 113)	14 (7우, 7중)	9 (1♀, 8♂)	<b>50</b> (7♀, 43♂)	44 (6 ♀, 38 ♂)	139 (32 ♀, 10 ♂)
MYTH	17 (130,43)	22 (90,113)	14 (11♀, 3♂)	16 (11♀, 5♂)	36 (26 ♀, 10 ♂)	103 (70 <sup>0</sup> , 33 ♂)
MYVO	0	1 (0)	4 (°+)	14 (6우, 8소)	1 (රී)	20 (8 ♀, 12 ♂)
EPFU	7 (ථ)	38 (්)	18 (1♀, 17♂)	9 (බ)	0	72 (2 ♀, 62 ♂)
LACI	1 (්)	0	2 (්)	0	0	3 (0 ♀, 3 ♂)
LANO	1 (රි)	1 (්)	2 (්)	0	0	4 (0 ♀, 4 ♂)
СОТО	1 (රි)	0	2 (♀)	3 (2♀, 1♂)	2 (්)	8 (4 ♀, 4 ♂)
Total	32 ♀, 36 ♂	30 ♀, 66 ♂	25 ♀, 63 ♂	46 ♀, 112 ♂	<b>47</b> ♀ <b>, 88</b> ♂	<b>556 (185</b> ♀, <b>356</b> ♂)
BNN	6.8	5.6	7.3	15.8	11.25	8.17

The age distribution at HVR continues to favor adults. Of all captures in 2006, only 3.6% were juvenile bats (Table 4). This is surprising especially for *M. thysanodes* where mist netting occurred in proximity to a maternity site. Over the 5-year study, only 10.9% of captures were juveniles. This pattern remains inexplicable.

Species	2002	2003	2004	2005	2006
M. ciliolabrum	A(1) J(0)	A(3) J(2)	A(6) J(1)	A(3) J(2)	A (4) J (0)
M. evotis	A(8) J(9)	A(8) J(7)	A(32) J(2)	A(55) J(6)	A (53) J (1)
M. lucifugus	A(19) J(4)	A(14) J(0)	A(9) J(0)	A(49) J(1)	A (37) J (3)
M. thysanodes	A(13) J(4)	A (12) J(10)	A(13) J(1)	A(13) J(3)	A (38) J (1)
M. volans	A(1) J(0)	A(1) J(0)	A(4) J(0)	A(14) J(0)	A (1) J (0)
E. fuscus	A(5) J(2)	A(38) J(0)	A(18) J(0)	A(9) J(0)	A (0) J (0)
Total	A(47) J(19)	A(76) J(19)	A(82) J(4)	A(143) J(12)	A (133) J (5)

**Table 8**. Comparative age distributions per species across years at HVR.

Sonar data from 2006 showed the continued importance of meadows and thinned plots for foraging by bats. DesPITe the thinning efforts, *M. evotis*, a clutter specialist, continues to occur in high numbers at HVR. Highest insect biomass was observed in thinned plots. However, Meadow plots, where bat activity is also high, showed very little insect biomass. This is likely a sampling bias in that larger insects, especially beetles, were not readily captured in our light traps, and meadows may be more prone to having larger-bodied insects using them. This is likely why larger-bodied bats such as the big brown bat (*Eptesicus fuscus*) and the hoary bat (*Lasiurus cinereus*), forage predominately in meadow habitats. We intend to continue this part of the survey and perhaps invest in other methods of insect capture if funding is available.

The calcium experiments conducted in 2006 using an artificial water hole with artificially increased calcium load and a plate antennae for scanning PIT-tagged individuals gave tremendous insight into water use patterns of lactating versus nonreproductive female sand indicated that: 1) water resources are extremely important to the health of bats, in particular, those that are lactating and that 2) reproductive females greatly preferred the artificial calcium water hole over the natural water hole. Because bats lose large amounts of body water during diurnal roosting, they drink most prolifically just after evening emergence and then again just before dawn when re-entering the day roost.

West Nile virus continues to show up in HVR bats, but apparently in relatively low frequencies. In 2006, a single *M. evotis* tested positive, but this species appears to be doing well in terms of populations numbers in Boulder County.

#### **FUTURE NEEDS**

- Continue analysis of water use patterns
- Radio-tag several female C. townsendii in order to locate maternity roost site
- Continue sonar data collection from forest thinning operation with insect collection
- Replication of calcium water hole experiments

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