# BAT FORAGING RESPONSE TO INTRODUCED FISH IN THE SIERRA NEVADA

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*Abstract.*—Stocking of trout into naturally fishless water bodies in the mountains of western North America has reduced populations of many native species in those systems, with benthic aquatic invertebrates being particularly impacted. Although many bats consume emergent aquatic insects, no published studies have focused on how they could be affected by changes to prey populations at lakes subsequent to trout stocking. The aim of this study was to determine whether fishless lakes or lakes stocked with trout provide higher quality foraging habitat for bats. We recorded and analyzed bat echolocation calls to assess overall bat activity, foraging activity, and foraging rates at nine feature-matched pairs of stocked and unstocked high elevation lakes in the central Sierra Nevada of California. Bat species with echolocation calls classified as around 25 kHz and 50 kHz showed little to no behavioral change between stocked lakes and fishless lakes. In contrast, bats that echolocate around 40 kHz showed higher levels of overall activity, foraging habitat. Alternatively, these bats could be foraging on suboptimal prey, pursuing small insects such as mosquitoes, and this could represent a cost to these bats due to the lower energetic return of small prey. Because of the recent arrival in California of the potentially deadly fungus *Pseudogymnoascus destructans*, which causes White-nose Syndrome (WNS), introduced trout may constitute an additional challenge to populations of bats that are already threatened by WNS.

Key Words.---bats; introduced trout; conservation; emergent aquatic insects; echolocation; foraging habitat; restoration

#### INTRODUCTION

Introduction of trout into naturally fishless water bodies in the mountains of western North America has been shown to have profound negative impacts on native species diversity and trophic exchange in these systems (Bahls 1992; Knapp 1996; Finlay and Vredenburg 2007). Many organisms with aquatic life stages, such as amphibians and emergent aquatic insects, have experienced population declines as a result of trout predation (Knapp and Matthews 2000; Pilliod and Peterson 2001; Tyler et al. 1998; Pope et al. 2009). Terrestrial predators that rely on prey from aquatic ecosystems have likewise been affected, including spiders (Benjamin et al. 2011), garter snakes (Matthews et al. 2002; Lawler and Pope 2006), and the Graycrowned Rosy-finch (Leucosticte tephrocotis dawsoni), which nests at high elevations (Epanchin et al. 2010).

The presence of trout particularly impacts the abundance, diversity, and biomass of benthic aquatic invertebrates (Knapp et al. 2001; Finlay and Vredenburg 2007; Pope et al. 2009). In the Sierra Nevada of California, Knapp et al. (2001) found significantly lower numbers of most orders of invertebrates at lakes with fish compared to those without, including emergent aquatic insects such as mayflies (Ephemeroptera) and caddisflies (Trichoptera). Pope et al. (2009) showed that removing trout from lakes in the Trinity Alps Wilderness of California increased the abundances of emerging Ephemeroptera and Trichoptera, as well as overall insect biomass. In these studies, the only prevalent insects that

were found in high abundance at stocked lakes were small dipterans, particularly midges (Chironomidae) and mosquitos in the genus *Culex*; the aquatic larvae of these insects avoid being eaten by trout due to their small size, and they also experience lowered predation pressure as fish reduce populations of large predatory insects.

Although Kalcounis-Ruppell et al. (2007) reported the effects of a wastewater treatment plant on bat foraging on aquatic emergent insects, there are no published studies that focus on how bats are affected by changes in insect abundance, diversity, and biomass at mountain lakes subsequent to trout stocking. High-elevation ecosystems are resource limited, and seasonal pulses of emergent aquatic insects make up a large part of their overall ecosystem productivity (Finlay and Vredenburg, 2007; Epanchin et al. 2010). Subsequently, trout stocking in naturally fishless lakes could cause a loss of high-quality foraging habitat for some species of local bats such as has been reported for other vertebrates (Lawler and Pope 2006; Pope et al. 2009; Joseph et al. 2011).

For bats that breed in the high Sierra Nevada, where the maternity season is short (Elizabeth Pierson et al., unpubl. report), a loss of foraging habitat near a maternity roost could reduce reproductive success. Although bats can fly far in search of prey, during lactation bats decrease their average foraging range size so that they can return to a roost several times a night to nurse non-volant young (Henry et al. 2002). If abundance or biomass of nearby available prey is reduced, bats would suffer energetic costs by foraging for longer time periods or flying further distances to find suitable prey, which could result in poor body condition for mothers or offspring (Kurta et al. 1989).

Bats have been shown to respond to anthropogenic modifications of aquatic habitats, especially when modifications result in changes to insect emergence, but responses vary among species, and most likely depend on the prey base of a bat (Vaughan et al. 1996; Fukui et al. 2006; Kalcounis-Rueppel 2007; Abbott et al. 2009; Akasaka et al. 2009; Scott et al. 2010). For example, Abbott et al. (2009) found that the Common Pipistrelle (*Pipistrellus pipistrellus*) is more active downstream of sewage outflows, whereas the Daubenton's Bat (*Myotis daubentonii*) is more active upstream. In this case, *P. pipistrellus* may be foraging for small Chironomidae, which are abundant in eutrophic conditions; while *M. daubentonii* are likely concentrating on Trichoptera that do not occur in eutrophic conditions.

We hypothesize that foraging behavior of bats at lakes without trout would be different than their foraging behavior at lakes with introduced trout. We predict that bats that emit echolocation calls at 40 kHz and 50 kHz and which forage on aquatic emergent prey would forage more at lakes without introduced trout than at lakes with introduced trout. Both Yuma Myotis (Myotis yumanensis), a 50 kHz bat, and the Little Brown Bat (M. lucifugus), a 40 kHz bat, forage over still water (Herd and Fenton 1983; Buchler 1976). We also predict that bats that emit echolocation calls in the 25 kHz range would not exhibit a response to changes in the aquatic environment as these bats are open aerial foragers and prey on terrestrial insects (Barclay 1991; William Rainey et al., unpubl. report). To determine the differences in foraging behavior, we assessed the amount and timing of overall bat activity, foraging activity, and foraging effort at lakes with and without stocked trout using echolocation detectors. Assessing foraging effort might be a way to establish whether trout stocking is changing insect availability and causing bats to hunt for atypical prey. We also measured the timing of bat activity throughout the night to look for increases in foraging effort that may indicate energetic stress (Duverge et al. 2000).

### METHODS

*Study design.*—In July of 2012 and 2013, we recorded bat echolocation calls at 18 lakes (nine feature-matched pairs) in the central Sierra Nevada of California (i.e., we recorded at two lakes for each of nine non-consecutive nights totaling 18 survey nights). We paired lakes based on elevation, location (within 2 km of each other), area, and depth. We surveyed partner lakes simultaneously to reduce the impact of temporal variability.

*Study area.*—Our sites comprised naturally fishless lakes in the central Sierra Nevada. In the first season (July 2012), we used study lakes in the northwest corner

of Yosemite National Park. In the second season (July 2013), we used sites in the southwest corner of the Emigrant Wilderness in Stanislaus National Forest. Study lakes were small (0.2–1.8 ha) and located in granite basins surrounded by conifers, meadows, and rock features. The average distance between lake pairs was 499 m and sites were between 1,800 and 2,700 m above sea level (MASL), which places them between the lower montane and upper montane forest zones (Storer et al. 2004). We chose lake pairs based on accessibility and the availability of site information. Lake locations, fish status data, and some variables we assessed for covariance (lake depth, lake area, and lake perimeter) were provided by the Division of Resources Management and Science of Yosemite National Park, the North Central Region of the California Department of Fish and Wildlife, and the Mi-Wok Ranger District of Stanislaus National Forest. We computed other potential covariates (the distance from the lake to the nearest river, the distance from the lake to the nearest lake, and the distance from the lake to the nearest water feature) with images from Google Earth (Google Inc., Mountain View, California)

Field methods.—During each of nine survey nights, we recorded bat echolocation calls at both lakes in a given pair (one with fish, one without) using Anabat II<sup>TM</sup> detectors and Zero-crossings Analysis Interface Modules (Z-CAIM)<sup>™</sup> recorders (Titley Electronics, Ballina, New South Wales, Australia). To maximize coverage, we placed four bat detectors, one at each of the four cardinal directions, with microphones on 3-m-tall poles and facing the interior of the lake from the edge of the lake. The detectors recorded bat activity for roughly 8 h, from the evening of placement (usually an hour before sunset, but occasionally later) to an hour after sunrise, when we collected the units. We discarded data collected on nights with low temperatures, high wind speeds, and/ or heavy precipitation and the area we resurveyed as recommended by Hayes (1997) and Erickson and West (2002).

Anabat data analysis.—After we collected bat detectors, we extracted the data, identified echolocation call sequences, and sorted calls into groups for statistical analysis. Echolocation call sequences are defined as a series of two or more consecutive echolocation clicks produced by a bat as it flies within range of the detector (Fenton 1977; Hayes 1997; Johnston, D.S. 2002. Data collection protocol: Yuma bat (Myotis yumanensis). Wetlands Regional Monitoring Program Plan, Part 2: Data Collection Protocols:1-6. https://calisphere.org/ item/ark:/86086/n2fj2fq2/). We extracted call data with CF Read 4.4s<sup>TM</sup> and processed it using AnalookW 4.1b<sup>™</sup> (Titley Electronics, https://www.titley-scientific. com/us/downloads/firmware-software), which creates a file showing an acoustic frequency (kHz) by time display



FIGURE 1. A bat call sequence with two feeding buzzes (shown in boxes), which indicate foraging attempts.



FIGURE 2. An example of call sequences included in the 50 and 40 kHz echolocation categories. Bats in the 50 kHz category that echolocate at characteristic frequencies of 45-50 kHz include California Myotis (*Myotis californicus*) and Yuma Myotis (*Myotis yumanensis*). Bats in the 40 kHz category that echolocate at characteristic frequencies of 35–40 kHz include the Little Brown Bat (*Myotis lucifugus*), Long-legged Myotis (*Myotis Volans*), and Western Small-footed Myotis (*Myotis ciliolabrum*).

(O'Farrell et al. 1999). After visual assessment of each file, we labeled call sequences according to their phonic grouping and extracted those labels into a text file using the Anabat utility Dataget (after Miller 2001; from Titley Scientific, Brendale, Queensland 4500, Australia).

We quantified foraging activity by identifying foraging attempts known as feeding buzzes (Fig. 1), which are sequences in which echolocation clicks move closer together as a hunting bat closes in on its prey (Griffin et al., 1960, Johnston 2002, *op. cit.*). We determined foraging effort by computing the ratio of foraging activity to overall activity (feeding buzzes per unit of activity). We chose zero-crossing technology for ease of use in remote locations. We classified



FIGURE 3. An example of call sequences included in the 25 and 10 kHz echolocation categories. Bats in the 25 kHz category that echolocate at characteristic frequencies of 20–30 kHz include the Big Brown Bat (*Eptesicus fuscus*), Hoary Bat (*Lasiurus cinereus*), Silver-haired Bat (*Lasionycteris noctivagans*), Brazilian Free-tailed Bat (*Tadarida brasilienses*), and Long-eared Myotis (*Myotis evotis*). Bats in the 10 kHz category that echolocate at characteristic frequencies of 8–15 kHz include the Western Mastiff Bat (*Eumops perotis*) and Spotted Bat (*Euderma maculatum*).

echolocation call sequences of bats to phonic group, which is ecologically related to size of prey (Barclay and Brigham 1991), to reduce the number of calls that would need to be discarded when calls could not be classified to a specific species. Therefore, we organized all recorded call sequences into four phonic groups based on the characteristic call frequency (Keinath 2004; Joeseph Szewczak et al., unpubl. report; Elizabeth Pierson et al., unpubl. report; Figs. 2 and 3). Each of these four groups contains calls from one or more of the 11 species of bats that are commonly found in the Sierra Nevada above 2,000 MASL (Elizabeth Pierson et al., unpubl. report). Bats in the 50 kHz emitting group (M. californicus and M. yumanensis) and the 40 kHz group (M. lucifugus, Long-legged Myotis, M. volans, and Western Smallfooted Myotis, M. ciliolabrum) are relatively small and have diets restricted to smaller prey. Of these, M. lucifugus (40 kHz) and M. yumanensis (50 kHz) are most reliant on emergent aquatic insects. Bats in the 25 kHz (Big Brown Bat, Eptesicus fuscus, Hoary Bat, Lasiurus cinereus, Silver-haired Bat, Lasionycteris noctivagans, Brazilian Free-tailed Bat, Tadarida brasiliensis, and Long-eared Myotis, Myotis evotis) emitting group and the 10 kHz (Western Mastiff Bat, Eumops perotis, and Spotted Bat, Euderma maculatum) emitting group tend to be larger and can take a wider variety of prey items, including large prey such as moths (Waters et al. 1995).

We completed further categorization to improve analysis. To reduce the chance of over- or underestimating the activity of a given species, we used the acoustic activity index described in Miller (2001). In this index, a survey period is separated into 1-min blocks and then bat activity is defined as the number of minutes in a survey period that contain at least one echolocation call sequence. Also, to compare activity levels of each of the phonic groups at different times, we divided data from a night of recording into three time periods of bat activity: Early (1900–2300), Mid (2300–0300), and Late (0300– 0600).

Statistical analysis.—One complication in assessing differences in bat activity is that variables unrelated to prey abundance, such as weather, proximity to roosting habitat, and nearby vegetation and landscape features, can influence bat behavior in an area (Furlonger et al. 1987; Hayes 1997; Erickson and West 2002; Downs and Racey 2006; Hagen and Sabo 2011). To control for these variables, we used a block design with feature-matched (elevation, location, area, and lake depth) pairs of lakes (one with trout, one without) as blocks (Hayes 1997; Seidman and Zabel 2001) in the analyses. For each of the three dependent variables: overall bat activity, foraging activity, and foraging effort (each measured in minutes of echolocation call sequences), we used a separate Three-way Analysis of Covariance (ANCOVA; Zar 1999) to determine the effect of whether or not the lake was stocked, phonic group (25 kHz, 40 kHz, and 50 kHz), and time period of bat activity (Early: 1900-2300, Mid: 2300-0200, Late: 0200-0500) on the dependent variable. Lake pair was used as a block. We included lake depth, area, perimeter, and the distance of the lake to the nearest river as covariates in the analyses to filter out their effects on an analysis. If all of the covariates did not significantly impact an analysis, we reanalyzed the data with a Three-way Analysis of Variance (ANOVA; Zar 1999) to maximize the power of the analysis. We used planned (a priori) comparisons to test for differences among means in lieu of post-hoc tests because the power and assumptions of the planned comparisons were the same as the ANOVA and ANCOVA. To determine if the assumption of homogeneity of variance was met, we initially conducted Levene's tests for each ANOVA or ANCOVA; for all tests the *P*-value was > 0.05indicating that the dependent variables did not need to be transformed. We analyzed data with IBM SPSS (Statistical Package for the Social Sciences) statistical software (IBM Corporation, New York, New York).

*Invertebrate sampling.*—We also sampled emergent insect populations to evaluate the potential prey base for bats and to see whether invertebrate populations at our study lakes are consistent with past research. We placed insect traps at the lakes at the same times and roughly the same locations as the bat detectors and we collected

traps in the morning when we collected detectors. In 2012, we used floating emergence traps (Epanchin et al. 2010; William Rainey et al., unpubl. report), and in 2013 we switched to sticky traps (Collier and Smith 1994). Emergence traps were round, floating 0.25 m<sup>2</sup> traps constructed of wire, bicycle tubes, and 0.3 mm white mesh. We constructed sticky traps using one  $21.6 \times 28$  cm ( $8.5 \times 11$  in) transparency sheet bent into a cylindrical shape and covered with liquid Tangle Trap (Tanglefoot, Grand Rapids, Michigan). We collected the invertebrates in these traps, identified them to order (Bland and Jaques 1978), and preserved them in 70% ethanol.

To determine if the relative abundances of insect orders at the stocked and unstocked lakes reflected trends found in other studies, we assessed the differences in the ratio of small (< 3 mm) to large (> 3 mm) insects using an ANCOVA as done in Ober and Hayes 2008). The independent variable was trout stocking (Yes/No). We included lake depth, lake area, lake perimeter, the distance from the lake to the nearest river, the distance from the lake to the nearest river, the distance from the lake to the nearest lake, and the distance from the lake to the nearest state (whether a river or lake) as covariates.

Conditions associated with location (elevation, location, area, and depth) differed sufficiently such that significant differences might be obscured with a simple random design. We initially assessed the importance of lake depth, area, perimeter, and the distance of the lake to the nearest river as sources of variation that might obscure results using those parameters as covariates in a Three-way ANCOVA. Because none were significant (P > 0.050), we used the results of the Three-way ANOVA instead to obtain more power.

### RESULTS

For 18 survey nights (nine non-consecutive nights, two lakes per night), we recorded 7,889 min of total activity, which included 4,249 min of foraging activity, from bats in the 25 kHz, 40 kHz, and 50 kHz phonic groups. Only about 3.5% (276) of the calls recorded were from bats in the 10 kHz group. Because of the small numbers, we did not use the 10 kHz group in our analysis. We recorded calls in small numbers from this group at nearly every lake, however, and routinely heard audible echolocation calls (most likely from *Euderma maculatum*) over study areas.

**Overall activity**.—The significant Stocked  $\times$  Phonic Group interaction in the analysis (Table 1) shows that differences in overall bat activity with respect to stocked and unstocked lakes differed among Phonic Groups (Fig. 4). Bats in the 40 kHz phonic category exhibited much greater activity (approximately four times) at stocked lakes than unstocked lakes, the 50 kHz phonic group also showed greater activity (approximately one and a third times) at stocked lakes than unstocked lakes than unstocked lakes, whereas the



**FIGURE 4.** The number of minutes of activity (minutes with echolocation call sequences recorded) by stocked lake (yes or no) and phonic group (25 kHz, 40 kHz, and 50 kHz). The dependent variable of Lake pair was used as a block. Confidence bars are 95%.

25 kHz group showed virtually no difference (Fig. 4). Total bat activity varied between time periods (Table 1). The planned comparisons between Early, Mid, and Late periods showed that the combined mean of bat activity from all three phonic groups during the Early and Late periods was significantly greater than the mean activity in the Mid period and the mean activity level during the Early period was significantly higher than during the Late period (Fig. 5). Controlling for elevation, location, area, and lake depth with a paired lake design (block) was essential for determining the effects of the independent variables on overall bat activity (Table 1). This was indicated by the highly significant Lake Pair (block) effect.

*Foraging activity.*—The significant Stocked × Phonic Group interaction shows that differences in overall bat activity with respect to stocked and unstocked lakes differed among Phonic Groups (Table 2). Bats in the 40 kHz phonic category exhibited much greater activity (approximately four times) at stocked lakes than unstocked lakes, the 50 kHz phonic group also showed greater activity (approximately one and a third times) at stocked lakes than unstocked lakes, whereas the 25 kHz group showed virtually no difference (Fig. 6). There was a slight effect of lake area and distance to the nearest river on foraging activity (Table 2). The Lake Area covariate was significant and indicated a slight, positive correlation (r = 0.201, t = 0.54, df = 7, P = 0.011) between lake area and the amount of foraging activity and greater area would tend to inflate the estimate of activity. The significant Distance to Nearest River covariate indicated a slightly negative correlation (r = -0.083, t = 0.22, df = 7, P < 0.010) between foraging activity at a lake and the distance of that lake from the nearest river.

*Foraging effort.*—Differences in foraging effort (feeding buzzes per unit of activity) varied significantly with respect to whether or not the lakes were stocked among the three phonic groups (Stocked × Phonic Group interaction; Table 3). Bats in the 40 kHz phonic group had about one and a third times more foraging effort at lakes with stocked trout; 25 kHz bats showed about one and a quarter times more foraging effort at lakes that did not have stocked trout; and 50 kHz bats show almost no difference (Fig. 6). Overall, foraging effort did not vary significantly between time periods, and the power for this test is relatively low (Table 3).

The responses of the three phonic groups to stocking did not vary significantly among the three time periods, based on the Stocked  $\times$  Phonic Group  $\times$  Time Period

TABLE 1. Results of a Three-Way ANOVA for the total activity of bats at surveyed lakes. The dependent variable is the number of minutes of activity (minutes with echolocation call sequences recorded). The independent variables are stocked (yes or no), phonic group (25 kHz, 40 kHz, and 50 kHz), and time period of bat activity (Early: 1900–2300, Mid: 2300–0200, Late: 0200–0500). The two planned comparisons for Time Period are included: Early and Late versus Mid, and Early versus Late. Lake pair was used as a block. Lake depth, area, perimeter, and the distance of the lake to the nearest river were initially included as covariates but were not significant and were not included in final analysis. The abbreviation df = degrees of freedom.

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Source	df	Mean Square	F	P-value	Observed Power	
Stocked	1	34002.5	17.66	N/A	0.987	
Phonic Group	2	10807.3	5.612	N/A	0.852	
Time Period	2	18072.0	9.385	< 0.001	0.977	
Early and Late vs. Mid	1	12970.7	6.736	0.010	0.731	
Early vs. Late	1	23173.4	12.03	0.001	0.931	
Lake Pair	8	11370.2	5.905	< 0.001	1.000	
Stocked×Phonic Group	2	19487.1	10.12	< 0.001	0.984	
Phonic Group×Time Period	4	3153.2	1.637	0.168	0.493	
Stocked×Time Period	2	649.4	0.337	0.714	0.103	
Stocked×Phonic Group×Time Period	4	2148.5	1.116	0.352	0.344	
Error	136	1925.6				



**FIGURE 5.** The number of minutes of total activity (minutes with echolocation call sequences recorded) by time period of bat activity (Early: 1900–2300, Mid: 2300–0200, Late: 0200–0500). (Top) Comparison of activity for Early and Late activity versus Mid activity and (Bottom) Early versus Late. Lake pair was used as a block. Confidence bars are 95%.

interaction (Table 3). Based on the Stocked × Phonic Group interaction, stocked trout did not affect the level of foraging effort for bats as a whole over the course of the night (Table 3). The Phonic Group × Time Period interaction suggests the three phonic categories do not change their foraging effort during the three time periods (Table 3). Lake pair was found to have a block effect, indicating that the blocking parameters (elevation, location, area, and depth) could have significantly impacted results if they were not taken into account. Lake depth, area, perimeter, and the distance of the lake to the nearest river were assessed as covariates, but none was a significant correlate at the 0.05 level.

Invertebrate sampling.—Frequencies of insect captures at both emergence traps and sticky traps were very low and variable (stocked lakes: mean = 40.2, standard deviation [SD] = 21.0 from insects; unstocked lakes: mean = 32.0, SD = 19.1). Total emergent aquatic insect sampling comprised three orders: Diptera that were all < 3 mm (92 % of total; 220 from stocked lakes and 178 from unstocked lakes), Ephemeroptera (0.6% of total; one from stocked lakes and two from unstocked lakes, and Trichoptera (7.3% of total; 20 from stocked lakes and 12 from unstocked lakes). Stocking did not change the abundance of insect orders at the study lakes  $(F_{14} = 2.111, P = 0.220)$ . The extremely low power of this analysis (power = 0.203) suggests that a larger sample size and higher capture rates are needed to address this question.

#### DISCUSSION

**Response from different bat species.**—The results of our study are consistent with previous studies showing that bats that forage over aquatic ecosystems change

**TABLE 2.** Results of a Three-Way ANCOVA for the foraging activity time (min) of bats at surveyed lakes. The dependent variable is the number of minutes of foraging activity (minutes with feeding buzzes recorded). The independent variables are stocked (yes or no), phonic group (25 kHz, 40 kHz, and 50 kHz), and time period (Early: 1900 - 2300, Mid: 2300 - 0200, Late: 0200 - 0500). Lake pair was used as a block. Lake area and the distance of the lake to the nearest river were significant covariates. The abbreviation df = degrees of freedom.

Source	df	Mean Square	F	P-value	Observed Power
Stocked	1	55.8	17.66	N/A	0.987
Phonic Group	2	16451.7	5.612	N/A	0.852
Time Period	2	4808.0	9.385	< 0.001	0.977
Early and Late vs. Mid	8	4726.1	6.736	0.010	0.731
Early vs. Late	1	5492.5	12.03	0.001	0.931
Lake Pair	1	3259.7	5.905	< 0.001	1.000
Stocked×Phonic Group	2	10251.5	10.12	< 0.001	0.984
Phonic Group×Time Period	4	2379.2	1.637	0.168	0.493
Stocked×Time Period	2	888.7	0.337	0.714	0.103
Stocked×Phonic Group×Time Period	4	791.1	1.116	0.352	0.344
Error	136	818.8			



their behavior when modifications to these systems affect their prey base (Vaughan et al. 1996; Kalcounis-Rueppel 2007; Abbott et al. 2009; Scott et al. 2010). We found that the 40 kHz group (M. lucifugus, Myotis volans, and M. ciliolabrum) displayed greater overall activity, foraging activity, and foraging effort at lakes with stocked trout, suggesting that bats in this phonic group might be spending more time searching for insects at lakes with trout. Given the paucity of aquatic emergent insects caught in the emergent traps, the greater overall activity, foraging activity, and foraging effort of these bats at lakes with stocked trout are not likely because of higher quality foraging habitat. As predicted, we did not find a difference in activity levels from bats in the 25 kHz (Eptesicus fuscus, Lasiurus cinereus, Lasionycteris noctivagans, Tadarida brasilienses, and Myotis evotis). Although we also predicted that the 50 kHz bats (M. californicus and M. yumanensis) would forage more at lakes without introduced trout, this phonic group exhibited no difference between the overall activity, foraging activity, and foraging effort between lakes with and without introduced trout.

We believe that the majority of calls recorded in the 40 kHz category can be attributed to *M. lucifugus*, indicating that this species shows a strong response to the presence of stocked trout. Myotis lucifugus is very active over aquatic habitats, forages on aquatic insects, and is common above 2,000 feet in the Sierra Nevada (Ober and Hayes 2008; Clare et al. 2011; Elizabeth Pierson et al., unpubl. report). The other potentially occurring 40 kHz bats, M. ciliolabrum and M. volans, are not associated with aquatic habitats and are not common at the elevations of our study area (Elizabeth Pierson et al., unpubl. report). There are few diet records for M. ciliolabrum, but it is known to forage over rocky bluffs and cliffs, and the closely related Eastern Small-footed Myotis (M. leibii) consumes moths, beetles, and flies (Rodriguez and Ammerman 2004; Moosman et al. 2007; Johnson et al. 2012). Myotis volans forages mainly on moths (Johnson et al. 2007) primarily under the canopy of trees (Bell and Fenton 2011).

We expected to see a response from the 50 kHz group because this group included *M. yumanensis*, which is known to concentrate on emergent aquatic prey (Ober and Hayes 2008; Clare et al. 2011). Because we used zerocrossing technology, we were unable to reliably separate *M. yumanensis* calls from those of *M. californicus*, which does not typically forage over aquatic areas, and may have shown no response to the presence or absence of

**FIGURE 6.** (Top) The number of minutes of foraging activity (minutes with feeding buzzes recorded) for the interaction of stocked (yes or no) lakes and phonic group (25 kHz, 40 kHz, and 50 kHz) based on ANCOVA adjusted for area of the lake and distance of the lake from the nearest river. (Middle) The number of minutes of foraging activity (minutes with feeding buzzes recorded) for the interaction of phonic group (25 kHz, 40 kHz, and 50 kHz) and time period (Early: 1900–2300, Mid: 2300–0200, and Late: 0200–0500) based on ANCOVA adjusted for area of the lake and distance of the lake from the nearest river. (Bottom) The number of minutes of foraging effort (feeding buzzes per unit of activity) for the interaction of stocked (yes or no) and phonic group (25 kHz, 40 kHz, 40 kHz, and 50 kHz) based on ANCOVA adjusted for area of the lake and distance of the lake from the nearest river. (Bottom) The number of minutes of foraging effort (feeding buzzes per unit of activity) for the interaction of stocked (yes or no) and phonic group (25 kHz, 40 kHz, 40 kHz, and 50 kHz) based on ANCOVA adjusted for area of the lake and distance of the lake from the nearest river. For all, lake pair was used as a block and confidence bars are 95%.

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TABLE 3. Results of a Three-Way ANOVA for the foraging effort of bats at surveyed lakes. The dependent variable is the ratio of
the number of minutes of activity to the number of minutes of foraging activity. The independent variables are stocked (yes or no),
phonic group (25 kHz, 40 kHz, and 50 kHz), and time period (Early: 1900 - 2300, Mid: 2300 - 0200, Late: 0200 - 0500). Lake pair
was used as a block. Lake depth, area, perimeter, and the distance of the lake to the nearest river were initially included as covariates
but were not significant and were not included. The abbreviation $df = degrees$ of freedom.

Source	df	Mean Square	F	P-value	Observed Power
Stocked	1	0.08	2.078	N/A	0.299
Phonic Group	2	2.28	59.42	N/A	1.000
Time Period	2	0.05	1.249	< 0.001	0.268
Lake Pair	8	0.17	4.531	< 0.001	0.996
Stocked×Phonic Group	2	0.14	3.653	< 0.001	0.664
Phonic Group×Time Period	4	0.04	0.931	0.168	0.289
Stocked×Time Period	2	0.03	0.757	0.714	0.176
Stocked×Phonic Group×Time Period	4	0.03	0.831	0.352	0.260
Error	129	0.04			

trout (Ober and Hayes 2008; Harvey et al. 2011). Using a full-spectrum analysis to separate these species from one another might yield more definitive results for these species.

The species in the 25 kHz group do not specialize on aquatic insects, so as predicted, we did not observe a response to changes in the aquatic environment. Eptesicus. fuscus, L. cinereus, L. noctivagans, and M. evotis prefer prey of terrestrial origin such as lepidopterans and non-aquatic coleopterans (Barclay 1991; Agosta 2002; Ober and Hayes 2008). Tadarida brasiliensis, E. fuscus, L. cinereus, and L. noctivagans are open aerial foragers in the 25 kHz group that typically forage in wide open spaces, usually well above the landscape (Johnson et al. 2007). Tadarida brasiliensis is a generalist with a broad diet; although this bat will take advantage of large swarms of aquatic insects, it also appears to prefer large, terrestrial prey (Whitaker et al. 1996; McWilliams 2005). Relatively low levels of foraging activity coupled with consistent foraging effort indicated that bats in this group were probably pursuing prey that are occasionally present at lakes regardless of fish status. A diet analysis would confirm that trout stocking does not impact the prey base of these bats.

*Timing of activity.*—The timing of bat activity changed throughout the night in unexpected ways, but the stocked status of the lakes did not influence the temporal pattern. Bats that depend on crepuscular swarms of insects generally follow a bimodal activity pattern, with a peak at dusk and a second peak at dawn (Rydell et al. 1996; Hayes 1997); however, in our study, foraging activity for 40 kHz and 50 kHz bats stayed relatively high from sunset to 0200 and then dropped off in the early morning. These results were expected because all surveys took place at the height of maternity season when lactating females forage for longer periods of time and return to the roosts several times a night to nurse young (Henry et al. 2002). Activity levels for bats in the 25 kHz category remained constant, and relatively low, throughout the

night; bats that feed on insects that do not swarm at dusk or dawn do not tend to follow a bimodal pattern, and any peaks in activity may not have been recorded in this group because most of their foraging takes place away from the lakes (Jones and Rydell 1994).

Foraging effort and the response to trout stocking.— The high levels of foraging effort (minutes with feeding buzzes per minutes of overall activity) from 40 kHz bats (likely M. lucifugus) at lakes with trout, coupled with invertebrate data based on the literature, suggest that bats were foraging at these lakes for small, abundant insects. Previous studies in these systems found increased abundance of < 3 mm dipterans (mostly mosquitoes and midges), but substantially reduced populations of largerbodied Ephemeroptera and Trichoptera (Finlay and Vredenburg 2007; Knapp et al. 2007; Pope et al. 2009; Epanchin et al. 2010). Because a higher number of small prey compared to large prey are needed to satisfy energy requirements, a bat hunting relatively small insects will exhibit greater foraging effort than one eating highercalorie insects (Gonsalves 2013).

To help implement meaningful conservation strategies for bats in the Sierra Nevada, we suggest more studies to determine what bats and introduced trout are eating. A dietary study of both groups should reveal any overlap in diets and might explain why the 40 kHz group of bats has higher activity levels at lakes with trout. It is unknown whether abundant small dipterans represent a high-quality prey base for M. lucifugus, or if the scarcity of larger, preferred prey is driving compensatory feeding on smaller prey (Cruz-Rivera and Hay 2000). Optimal foraging theory predicts that marginally profitable prey items will be included in the diet of a predator only as more profitable prey becomes unavailable (Davies 1977), but some studies have found that the inclusion of small prey items is positively correlated with their abundance (Turner 1982). While the relative availability of small dipterans will decrease the amount of energy bats spend pursuing them, this may not offset their small size.

Turner et al. (1982) showed that the inclusion of many small items in the diet of foraging swallows resulted in a decrease in their energy gain rate.

The energetic costs associated with compensatory feeding would particularly impact bats during maternity season. While M. lucifugus does consume midges and mosquitoes, females have been shown to selectively forage on larger bodied insects such as ephemeropterans and trichopterans as those prey become available during the summer maternity season (Anthony and Kunz 1977; Ober and Hayes 2008; Clare et al. 2011). In fact, the increased energetic demands of lactation may cause bats to prey switch. Clare et al. (2011) found that small dipterans comprised 63% of the diet of pregnant M. lucifugus, but ephemeropterans made up 66% of their diet during lactation. If stocking of trout in lakes causes preferred high-calorie prey to be absent from the foraging habitat, local breeding populations of M. lucifugus could suffer reduced survivorship, growth, and fecundity (Cruz-Rivera and Hay 2000).

Removing introduced trout from lakes may be one straightforward way to restore foraging habitat for bats and add to their resilience in the face of other environmental threats. Populations of *M. lucifugus* and other bat species at winter hibernacula have plummeted by 75–99% because of White-nose Syndrome (Harvey et al. 2011). Now that White-nose Syndrome has arrived in northeastern California, the Sierra Nevada bat populations are facing additional challenges and the implementation of conservation strategies are becoming more important. A long-term comparison of bat foraging behavior, diets, and population dynamics before and after trout removal from a lake would clarify how bats are being impacted.

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