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Species diversity and activity of insectivorous bats in three habitats in La Virgen de Sarapiquí, Costa Rica

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Abstract: Pineapple farms make up 45 000ha of Costa Rican landscape and are the second most exported crop. This is economically beneficial for the Costa Ricans, but greatly affects the natural flora and fauna because it is such a low growing crop. This study examined the differences in insectivorous bat species diversity and activity in the habitat gradient between the forest in Tirimbina Biological Reserve in La Virgen de Sarapiquí, Heredia, Costa Rica and the nearby pineapple farm called Finca Corsicana. Over a four week period in March and April 2013, ultrasonic recorders were placed at different sites to pick up the bats’ calls. Then the recordings were analyzed to identify the species. There were four families present and 19 different species. There was a significant decrease in the number of bat passes (the number of times a bat passes the recorder) in the pineapple farm ($\bar{x}=22.6$), in comparison to the border ($\bar{x}=39.9$), and the forest ($\bar{x}=44.2$) ($p=0.0028$). Agricultural environments affected and lowered bat presence. Also, a greater mean number of bats recorded between 1900-1930hrs compared to 1730-1800hrs, coincided with the setting of the sun and beginning of bat activity. More research is need throughout the night and the year to establish clearer patterns of bat use and activity in different habitats. Rev. Biol. Trop. 62 (3): 939-946. Epub 2014 September 01.

Key words: bat activity, Costa Rica, habitat gradient, insectivorous bats, tropical rain forest, ultrasonic recorders.

Conservation efforts are being made all over the world to help protect the environment from human destruction. In order for these conservation efforts to be implemented, there must be an understanding of: a) how changes to the landscape affect the flora and the fauna, and b) how to correct the problem or at least create a balance between the needs of humans and the natural life. This study focuses on how the gradation in diversity between a biological reserve (protected forest), the border of the forest, and a pineapple farm affect bat species diversity and activity.

Pineapple is the second most exported crop in Costa Rica. Of the pineapples consumed in the US and Europe, 85% and 71% are from Costa Rica, respectively (Ingwersen, 2012). This makes pineapple economically important to Costa Rica. As of 2011, there were 45 000ha of land cultivated in pineapple in Costa Rica. However, because it is a low-growing plant it drastically changes the landscape and thus the habitat of many species. Can bat species adapt to these new environments?

Miller, Arnett, and Lacki (2003) examined 56 bat studies on habitat management of forest-roosting bats and concluded that without roosting areas, bats will leave. Thus conservation efforts should be made to maintain roosting areas. It is highly beneficial to farmers to keep bats in an area because insectivorous bats greatly decrease pests and frugivorous and nectivorous bats help with pollination. It has been estimated that a colony of big brown bats (Eptesicus fuscus) can eat 1.3 million insects a year, or each bat can eat 4 to 8g per night. In monetary terms this is around $74/acre that farmers in south-central Texas, USA save in pesticides (Boyles, Cryan, McCracken, & Kunz, 2011).
Of 1,116 species of the bats in the world, 113 are found in Costa Rica and about 60 are known in the rural town of La Virgen de Sarapiquí, where this study was conducted (Laval & Rodríguez-Herrera, 2002). Around 75% of all bat species are insectivores, the rest eat fruit, nectar, fish, birds, or blood (Bat Conservation International, Inc., 2011). Even though insectivorous bats make up the majority of bat species, there are fewer studies on insectivorous bats in comparison to other types because insectivorous bats usually fly higher than mist nets, which are typically used in bat studies. The solution to this problem is recording devices.

Within the past 15 years there has been an increase in sonic recorder use to identify bat species difficult to catch in nets. Initially, research groups worked to identify all sounds made by each bat species by capturing them and recording calls of each species in open rooms. With species recordings, they were able to identify bats in the field with recording equipment (O’Farrell & Miller, 1999; Rydell, Arita, Santos, & Granados, 2002; Jung, Kalko, & von Helversen, 2007). Evidence of the usefulness of recording calls was shown in a study (Sampaio, Kalko, Bernard, Rodríguez-Herrera, & Handley, 2003) that focused on bat species adaptability to new surroundings in the Amazon Basin in Brazil with mist net captures at ground and canopy levels. However, sonic recordings detected 15 bat species not caught during the 2.5 yrs of the study. MacSwiney, Clarke, and Racey (2008) specifically tested how effective the recorders are in the identification of bat species and found that mist nets missed 30% of bat fauna and aerial insectivores (insectivorous bats that do not land to catch insects) were not caught at all; they were only identified by recorders.

The use of recording devices is possible because the majority of bat species use echolocation to navigate and find food during the night (Rodríguez-Herrera & Montero, 2009). The bats send out the call and use the echo that bounces back to form an image of their surroundings, called an eco-image. This eco-image includes information about the form and the texture of an object. The frequency of these calls is higher than the human range, which stops at 20kHz. One bat can call as many as 400,000 times per night. Bats use two types of calls, the frequency-modulated (fm), which is in a wide frequency band for a short period of time, and the quasi-constant frequency (qcf), which is in a narrow frequency band for a longer period of time (Schnitzler & Kalko, 1998). Each species’ fm and qcf calls are different, which is how recording devices can be used to identify different bat species.

The main objectives of this study were to determine species richness in a gradient of disturbance in La Virgen de Sarapiquí. Through the use of sonic recorders, this study aimed to identify bat species in order to determine their activity and diversity within: a) the forest at Tirimbina Biological Reserve, b) the edge of this forest, and c) the edge of the pineapple farm, Finca Corsicana.

This will help build a better understanding of the interaction between bats and humans based on the effects of agriculture and create a better understanding of how best to protect bat habitats.

**MATERIALS AND METHODS**

**Study area:** This study took place in La Virgen de Sarapiquí, Heredia, in northcentral Costa Rica (10°24′41″ - 10°27′05″ N, and 84°06′52″ - 84°07′43″ W) during March and April 2013. La Virgen is a small town that runs along the Sarapiquí River. About 7km to the north of the center of La Virgen is Tirimbina Biological Reserve and about 2km further North, along Route 126, is the Finca Corsicana, a pineapple farm. The majority of the study was done in Tirimbina Biological Reserve, which is 345ha of protected forest (Tirimbina Biological Reserve, 2010) (Table 1).

**Habitat sites:** There were three main habitat sites: 1) the forest, 2) forest border, and 3) pineapple farm. The forest was part of Tirimbina Biological Reserve, which is 98.55%
forest, 1.15% open area; mostly comprised of walking trails, and 0.30% wetlands. The forested area is made up of 85% primary forest and 15% secondary forest and these include tropical pre-montane forest that transitions into basal and tropical forest. The west border of the reserve is made up of the Sarapiquí River and Route 126, so the recorder-collection sites along the border have forest on one side and open space on the other. Finally the 1200ha pineapple farm is less than 2km NW of Tirim-bina and is found on both sides of Route 126.

**Recorder-collection sites:** At each of the three habitat sites there were four recorder-collection sites (one used per week). During week A (March 11, 2013 to March 15, 2013) one recorder was placed on the tower of the second bridge within the forest (A1), the second within an open building at the edge of the forest (A2), and the third at house 1 along the edge of the pineapple farm (A3). During week B (March 18, 2013 to March 22, 2013) one recorder was placed just off the Corteza trail, which is past the bridge (B1), the second in the cacao plantation, which is along the river (B2), and the third on the side of a building part of Escuela Las Palmitas next to the pineapple farm (B3). During week C (April 8, 2013 to April 12, 2013) one recorder was placed on the Hunter trail (C1), the second was placed on the road to Tirimbina’s field station (C2), and the third at La Quinta hotel (C3). During week D (April 15, 2013 to April 19, 2013) the first recorder was placed within the forest off the Botarrama trail (D1), the second on an area known as the island, which divides the river in two (D2), and the third at house 2 near the pineapple farm (D3).

**Bat call collection:** Each week one recorder (Song Meter SM2BAT+, Wildlife Acoustics, Concord, Massachusetts, USA) was placed in each of the three habitat sites and this process was repeated for 4wks. They were placed out on Monday or Tuesday and collected Saturday or the following Monday to ensure five whole nights of data collection. Table 2 illustrates the nights data was collected. Recorders ran from 1730hrs to 0530hrs. Each week, after retrieving the recorders, the data chips from the recorders were inserted into my computer (MacBook Pro) and the calls were transferred to an external hard drive. For each night there were eight files in wav format, each 1.5hrs in length. Then before moving the recorders to new recorder-collection sites the batteries were changed (only alkaline batteries will last the full length of time) and the data was deleted off the data chips in the recorders from the previous week to make space for new data.

**Bat call identification:** During daylight hours I analyzed the recordings using Raven Pro 1.5, (The Cornell Lab of Ornithology, Ithaca, NY, USA), a computer program designed to analyze sounds. I opened each file within this program, so that I was able to see 3min at a time. Passes were defined as the number of times a bat flew next to the recorder. Based on the definition by Estrada-Villegas, Meyer, & Kalko (2010), a pass ends when the time between two calls is three times longer than the time between the first two calls. I recorded the number of passes/species/ min in an excel spreadsheet.

To identify bat species, I compared the peak frequency, the length of the call, and the shape of the call to known calls (O’Farrell &

### TABLE 1
The 12 recorder-collection sites and their GPS coordinates

<table>
<thead>
<tr>
<th>Site</th>
<th>GPS Coordinates</th>
</tr>
</thead>
<tbody>
<tr>
<td>A1 Second Bridge</td>
<td>10°24’51” N, 84°07’07” W</td>
</tr>
<tr>
<td>A2 Open Building</td>
<td>10°24’58” N, 84°07’23” W</td>
</tr>
<tr>
<td>A3 House 1</td>
<td>10°25’37” N, 84°07’43” W</td>
</tr>
<tr>
<td>B1 Corteza Trail</td>
<td>10°24’44” N, 84°07’03” W</td>
</tr>
<tr>
<td>B2 Cacao Farm</td>
<td>10°24’41” N, 84°07’18” W</td>
</tr>
<tr>
<td>B3 School</td>
<td>10°25’37” N, 84°07’43” W</td>
</tr>
<tr>
<td>C1 Hunter Trail</td>
<td>10°24’34” N, 84°06’52” W</td>
</tr>
<tr>
<td>C2 Road to Field Station</td>
<td>10°25’12” N, 84°07’17” W</td>
</tr>
<tr>
<td>C3 La Quinta</td>
<td>10°27’05” N, 84°07’17” W</td>
</tr>
<tr>
<td>D1 Botarrama Trail</td>
<td>10°25’04” N, 84°06’52” W</td>
</tr>
<tr>
<td>D2 Island</td>
<td>10°25’08” N, 84°07’07” W</td>
</tr>
<tr>
<td>D3 House 2</td>
<td>10°26’52” N, 84°07’38” W</td>
</tr>
</tbody>
</table>
Miller, 1997; Fenton, Rydell, Vonhof, Eklof, & Lancaster, 1999; Koss, Mora, Coro, & Vater, 1999; O’Farrell & Miller, 1999; O’Farrell, Miller, & Gannon, 1999; Siemers, Kalko, & Schnitzler, 2001; Mora, Macías, Vater, Coro, & Koss, 2004; Jung et al., 2007; MacSwiney et al., 2008; Pio, Clarke, MacKie, & Racey, 2010; Jung & Kalko, 2011). Peak frequency is the frequency at the maximum power of the call. These researchers have captured different species and recorded their calls in order to accurately identify the species. Many of the articles mentioned above contained pictures of the sounds that I compared to the images in Raven Pro 1.5.

Time constraints hindered the analysis of all 12h from each night. Instead I analyzed the calls only from 1730-1800hrs and 1900-1930hrs for each night.

Differences between means of the variable, “number of passes”, were estimated between: a) the three habitats and two time periods; b) bat families and two time periods; c) bat families and three habitats; and d) four weeks and three habitats, with parametric two-way ANOVA. Differences between means of the variable, “number of species”, were estimated between: a) three habitats and two time periods with parametric two-way ANOVA. Assumptions were tested prior to execution of parametric ANOVAS (Sokal & Rohlf, 1995). Tests were executed with Statgraphics statistical software (www.statgraphics.com).

RESULTS

In 94 30-min segments over 4wks, several trends appeared. There were fewer bat passes for the pineapple farm (\(\bar{x}=22.6\)) than for the forest (\(\bar{x}=44.2\)) or border (\(\bar{x}=39.9\)) (F=5.69, d.f.=2, 312, p=0.0028, Fig. 1). There was a significant increase in the mean number of bat passes between 1730-1800hrs (\(\bar{x}=25.4\)) and 1900-1930hrs (\(\bar{x}=44.3\)) (F=11.71, d.f.=1, 313, p=0.0001). This trend was consistent for all habitats (F=0.65, d.f.=2, 309, p=0.5241, Fig. 1).

**TABLE 2**

Data collection dates. Data not collected during the first two weeks for the full five nights at each site was because batteries did not last long enough.

<table>
<thead>
<tr>
<th>Site</th>
<th>Week A</th>
<th>Week B</th>
<th>Week C</th>
<th>Week D</th>
</tr>
</thead>
<tbody>
<tr>
<td>A1 Forest</td>
<td>X</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>A2 Edge</td>
<td>X</td>
<td>X</td>
<td></td>
<td></td>
</tr>
<tr>
<td>A3 Pineapple</td>
<td>X</td>
<td>X</td>
<td></td>
<td></td>
</tr>
<tr>
<td>B1 Forest</td>
<td>X</td>
<td>X</td>
<td></td>
<td></td>
</tr>
<tr>
<td>B2 Edge</td>
<td>X</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>B3 Pineapple</td>
<td>X</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>8-Apr</td>
<td>9-Apr</td>
<td>10-Apr</td>
<td>11-Apr</td>
</tr>
<tr>
<td>C1 Forest</td>
<td></td>
<td>X</td>
<td>X</td>
<td>X</td>
</tr>
<tr>
<td>C2 Edge</td>
<td>X</td>
<td>X</td>
<td>X</td>
<td>X</td>
</tr>
<tr>
<td>C3 Pineapple</td>
<td>X</td>
<td>X</td>
<td>X</td>
<td>X</td>
</tr>
<tr>
<td></td>
<td>15-Apr</td>
<td>16-Apr</td>
<td>17-Apr</td>
<td>18-Apr</td>
</tr>
<tr>
<td>D1 Forest</td>
<td>X</td>
<td>X</td>
<td>X</td>
<td>X</td>
</tr>
<tr>
<td>D2 Edge</td>
<td></td>
<td></td>
<td></td>
<td>X</td>
</tr>
<tr>
<td>D3 Pineapple</td>
<td>X</td>
<td>X</td>
<td></td>
<td>X</td>
</tr>
</tbody>
</table>
There was also a significant difference in the mean number of species per habitat and 30-min period (F=6.98, d.f.=2, 88, p=0.0015, Fig. 2). At 1730-1800hrs, there were more species in the forest (x̄=3.69) and border (x̄=3.59) than during 1900-1930hrs (forest x̄=3.0, border x̄=3.47). The opposite occurred at the pineapple site where there were fewer species during 1730-1800hrs (x̄=2.18) than during 1900-1930hrs (x̄=4.06).

Four families of bats were found: Emballonuridae, Molossidae, Vespertilionidae, and only two passes of Moormopidae (removed for statistical testing). Of the four families, 19 different species were identified (Cormura brevirostris, Cynomops greenhalli, Cyttarops alecto, Diclidurus albus, Eptesicus brasilienensis, Eptesicus furinalis, Euderma maculatum, Eumops spp, Lasiusurus ega, Molossus currentium, Molossus molossus, Myotis albeckens, Myotis nigricans, Peropteryx kapleri, Peropteryx macrotis, Pteronotus pardus, Rhogeessa io, Saccopteryx bilineata, Saccopteryx leptura). There was a significant difference in mean number of bat passes per family with Emballonuridae having the least (x̄=24.0), then Vespertilionidae (x̄=34.3), and Molossidae with the greatest mean number of passes (x̄=45.9) (F=4.32, d.f.=2, 308, p=0.0141). When families were compared to the three habitats there was a significant difference (F=3.33, d.f.=4, 302, p=0.0109). The mean number of bat passes for Emballonuridae was greatest at the border sites (x̄=40.5) and the smallest in the forest (x̄=11.5, Fig. 3). Mean number of passes of Molossidae was greatest at the border as well (x̄=54.4) and smallest in the pineapple farm (x̄=16.4, Fig. 3). In contrast, the greatest mean number of passes of Vespertilionidae was in the forest (x̄=53.7) and the least in the border habitat (x̄=28.3).

There was no significant difference in mean number of passes at each habitat between weeks (F=1.45, d.f.=6, 303, p=0.939, Fig. 4). For all four weeks the mean number of passes
per habitat increased, decreased and, except for the pineapple farm, increased again in week D. Between the two 30-min periods was an hour of time. This hour created a significant difference in the activity of the different families (F=6.61, d.f.=2, 305, p=0.0015, Fig. 5). There were fewer passes for the Emballonuridae (\(\bar{x}=11.57\)) and the Molossidae (\(\bar{x}=23.06\)) during 1730-1800hrs than between 1900-1930hrs (Emballonuridae \(\bar{x}=35.53\), Molossidae \(\bar{x}=64.40\)). Mean number of passes of Vespertilionidae changed little and unlike the other two families, more passes occurred during 1730-1800hrs (\(\bar{x}=36.14\)) than during 1900-1930hrs (\(\bar{x}=32.64\)).

**DISCUSSION**

The main objective of this study was to examine the effect of habitat gradation on bats and results showed a difference in bat activity with the greatest mean number of passes in the forest and decreasing to the smallest mean number of passes in the pineapple farm. No significant difference existed in mean number of species between habitats, but a significant difference existed in mean number of passes per family in each habitat.

Based on different calls, different families seemed better suited to different habitats. For example, Vespertilionidae use fm calls with a long frequency and short duration, making them suited for forest habitats, where the greatest number of passes occurred (Schnitzler & Kalko, 1998). Voigt and Holderied (2012) found Molossidae better suited for open spaces partly because of their physical conformation (body size and wing length) and their calls are qcf, almost cf, which means they have a small change in frequency and a longer length. In their study of *M. currentium* and *M. sinaloe* in open and closed spaces, the *M. currentium* performed better in the dense forest-like areas than the *M. sinaloe*. In my study there were a number of *M. currentium* and no *M. sinaloe*, which may explain why there was a greater number of Molossidae found in the forest and the border than the pineapple farm. Alpizar, Viquez, Hong, Rodriguez-Herrera, and Gonzalez-Mayo (2012) in Tirimbina, with bat species in dense and open spaces within the forest, found greater numbers of Emballonuridae in the forest than the open spaces, but overall there was more species richness in open spaces (Jaccard=0.308). I found more Emballonuridae in the border than the other two habitats. Emballonuridae use gcf calls, which have both a large change in frequency and a long length of time making them ideally suited for border spaces (Schnitzler & Kalko, 1998).
While there was no significant difference in number of passes per habitat between weeks, there were some interesting trends that can be explained by the individual recorder-collection sites. In week B, the mean number of passes in the forest and the border were almost the same (forest $\bar{x}=48.07$, border $\bar{x}=48.14$). During week B the forest recorder was placed low to the ground, which could have prevented the microphone from picking up all of the calls and the border site was in the cocoa plantation, which is along the Sarapiquí River, but the recorder was more in the plantation than on the edge of the forest. In week C the forest recorder was placed in a tree on the side of a slope that was above a small river, thus creating more of an open space than forest space, which could have lead to a decrease in passes ($\bar{x}=27.0$). The forest recorder in week D was the furthest from the trail than in the other weeks creating a denser environment which may be why there is the highest mean number of passes at this site ($\bar{x}=65.60$).

The last major trend was the increase of mean number of passes between 1900-1930hrs when compared to 1730-1800hrs ($p=0.0001$). Bats are nocturnal species and the sun sets around 1730hrs, so by 1900hrs there has been more time for the bats to wake up and head out to feed. Jung & Kalko (2010) noticed a decrease in bat activity after 2030hrs in their human populated site, but no change throughout the night in their forest site. More analysis would have to be done of the bat calls from my study in order to clearly notice a trend in bat activity throughout the night.

ACKNOWLEDGMENTS

I would like to thank Bernal Rodríguez-Herrera for helping to guide me through this project with his vast knowledge of bats. Also, I would like to thank Christopher Vaughan and Michael McCoy for their help in editing and advising during the study. Judy Magnan arranged my home stay for 2mo and Alejandra Loiaza was instrumental in helping to collect all of my equipment. Additionally, Priscilla Alpízar helped in teaching me how to use the computer programs and how to identify the species based on the images. Finally, I would like to thank all of Tirimbina’s staff for their encouragement throughout the data collection process, especially Emmanuel Pequeño for helping me put out and take down the recorders every week.

REFERENCES


