

## Habitat selections of Himalayan insectivorous bats in forest-dominated landscapes

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The alteration of landscape by anthropogenic activity has reduced the foraging habitats of insectivorous bats. Thus it is important to understand the habitat selections of insectivorous bats and patterns of prey availability, especially in human-altered environments. We investigated bat activity in five classified habitats, namely: old-growth forest, remnant forest, citrus orchard, cornfield and paddy field in the forest-dominated Himalayan country of Bhutan. We monitored bat activity acoustically across 120 sites from May to November 2018. Thirteen different taxa and one unknown QCF bat were identified from 2,558 bat passes. Overall bat activity was dominated by open-space foragers such as *Scotophilus kuhlii*, *Taphozous* sp., *Otomops wroughtoni* and *Nyctalus leisleri*, showing comparative variations in habitat selection. But, *Rhinolophus* specifically, *R. luctus* was rarely present in open habitats such as cornfield and paddy fields, and was found to be one of the most abundant species in cluttered habitats, e.g., old-growth forest and remnant forest. Bat activity was higher in old-growth forest compared to paddy and remnant forest, but was not significantly different from cornfield and citrus orchard. Insect biomass positively correlated with bat activity. Our findings suggest that the habitat selection of insectivorous bats is influenced by prey abundance and habitat types. Although remnant forest was poorly associated with insect prey, the species richness of bats found there was almost equal to that of old-growth forest. Thus, remnant forest was found to be an important habitat, apparently serving as a corridor for the narrow-space foragers in the study landscape in Bhutan.

**Key words:** acoustics, forest dominated, habitat selection, landscape, prey availability, *Otomops wroughtoni*

### INTRODUCTION

Anthropogenic activity has had at least two notable impacts on bats: the alteration of landscape and declines in foraging habitats (Mendes *et al.*, 2017). These, in turn, have negatively affected bat populations (Fenton *et al.*, 1992; Kunz and Lumsden, 2003). It is therefore important to understand the habitat types and features selected by bat species (Russo and Jones, 2003), especially in human-altered landscapes. Numerous studies have looked at the responses of bats to different landscape structures from forested habitats to agricultural fields in tropical and temperate regions (e.g., Duchamp and Swihart, 2008; Klingbeil and Willig, 2009; Fuentes-Montemayor *et al.*, 2013). The responses of bats vary among guilds, e.g., edge, narrow and open-space foragers, and the degree of species mobility depending on both local and landscape structures (e.g., landscape heterogeneity, patch density — Fuentes-Montemayor *et al.*, 2013; Mendes *et al.*, 2017).

Forest is one of the most important and ecologically diverse habitats for roosting and for foraging

habitat for bats (Lacki *et al.*, 2007; Frey-Ehrenbold *et al.*, 2013; Heim *et al.*, 2015). Forest types influence the abundance of bat activity (Kalda *et al.*, 2015) and the use of forested habitat by bats is species-specific (Fuentes-Montemayor *et al.*, 2013). Although a mosaic of habitats can support biodiversity (Robinson *et al.*, 2001; Antrop, 2005), expansion of agriculture during the last century has reduced natural diversity (Bianchi *et al.*, 2006; Liira *et al.*, 2008). In Europe, bat populations have experienced severe depletions during the 20th century (Hutson *et al.*, 2001), where the majority of land cover has been converted to farmland (Mendes *et al.*, 2017). In addition, agriculture intensification is identified as the main reason for the decline of bat populations in Britain where more than 76% of the land is used for agriculture (Robinson and Sutherland, 2002; Wickramasinghe *et al.*, 2003). However, in the Himalayan country of Bhutan, 70.4% of the total land area is dominated by broad-leaved forest and only 7% of the land is used for traditional agriculture (Neuhoff *et al.*, 2014). In this agrarian country, the conversion of natural forest into agriculture

has resulted in a mosaic of habitats with remnant forest mixed into an agricultural landscape. Bhutan is part of Eastern Himalayan biodiversity hotspot, where it houses almost 200 species of mammals of which 33% are bats (Wangchuk *et al.*, 2004). Of the 65 bat species, nine species are fruit eating and 56 species are insectivores (Wangchuk *et al.*, 2004). It is important to know whether bat activity is greatest in old-growth forest given that the habitat heterogeneity of farmland offers opportunities for bats to forage depending on their morphological characteristics and the flexibility in their foraging strategies (Mendes, *et al.*, 2014). Several studies have reported that undisturbed forests are biologically diverse systems that provide important habitats for different species of bats (Fenton *et al.*, 1992; Kunz and Lumsden, 2003; Lacki *et al.*, 2007; Heim *et al.*, 2015; Mendes *et al.*, 2017). However, the presence of remnant and regrowth forest in the agricultural landscape has gained attention in conservation biology as these habitat patches enhance the structural connectivity that maintains biodiversity in human-managed landscapes (Fuentes-Montemayor, 2013). Insect availability has been shown to affect bat distribution and foraging activity (Fukui *et al.*, 2006). Forest fragmentation was shown to reduce biodiversity mostly through the edge effects, however, some insects respond positively to edge. In addition, some matrix types such as regrowth can support forest species more than open areas like pastures (Laurence *et al.*, 2002).

This study investigated the habitat selection of insectivorous bats in five different habitats, including old-growth forest, remnant forest, citrus orchard, cornfield and paddy field. Old-growth forest was defined here as relatively large patches of well-structured forest with a thick forest floor due to fewer disturbances. Remnant forests are isolated patches of young regrowth vegetation or fragmented old-growth forest with some degree of disturbance within the agricultural landscape. We hypothesized that bat activity will be the highest in forested habitat because it is important foraging habitat for many species of bats (Fenton *et al.*, 1992; Kunz and Lumsden, 2003; Lacki *et al.*, 2007; Heim *et al.*, 2015; Mendes *et al.*, 2017).

## MATERIALS AND METHODS

### *Study Site*

This study was conducted in two contiguous districts in southern Bhutan, namely Dagana ( $27^{\circ}0'N$ ,  $89^{\circ}55'E$ ) with an area of  $1,722.8 \text{ km}^2$  and Tsirang ( $27^{\circ}01'N$ ,  $90^{\circ}06'E$ ) with an

area of  $637.8 \text{ km}^2$ . Temperature ranges from  $12.0\text{--}21.3^{\circ}\text{C}$  with an annual rainfall of 1,271 mm in these two regions (Weather and Climate Services Division, 2018). The study sites range from 400 m to 2,000 m above sea level. There are four distinct seasons: winter (December to February which are the driest months), spring (March to May), summer (June to August which has heavy rainfall) and autumn (September to November). The study area was classified into five main types of habitats: forest (88.05%), agricultural land consisting primarily of corn and paddy fields (8.27%), remnant forest (0.58%), and citrus orchards (0.48%). The area was dominated by broad-leaved forest interspersed with patches of agricultural land. Farmers in this area have been using traditional farming methods since 2003 because the government aimed to ensure that all agriculture is organic farming by 2020 (Neuhoff *et al.*, 2014). Corn and paddy rice were the main food crops grown in this region, while citrus orchards mainly of orange fruit trees (*Citrus sinensis*) and black cardamom (*Amomum subulatum*) provide cash crops. Corn is planted and harvested in early summer while paddy rice is grown in autumn and harvested in early winter. The resettlement scheme that began in 1997 (Ministry of Home and Cultural Affairs, 2011) has created areas of remnant forest, which resulted in a mosaic of habitats.

The study area was divided into 20 blocks, with each block containing all the habitats of interest. These blocks were separated by hills or mountain ridges or were at least 1,000 m apart from each other. In each block, the five habitats were classified as: i) old-growth forest (Forest), ii) remnant forest (Remnant), iii) citrus orchard (Citrus), iv) cornfield (Corn) and v) paddy field (Paddy). As farmland patches are very small, sampling sites were established in those with a minimum size of 1–4 ha for orchards, corn and paddy fields, 3–5 ha for remnant forest, and 5 ha for old-growth forest. The old-growth and remnant forests were differentiated on the basis of structure and location within the landscape. Old-growth forest was structurally dense with a thick forest floor due to fewer disturbances as it was farther from settlements compared to remnant forest. In this study, young successional vegetation and fragmented forest within the agricultural landscape were categorized as remnant forest.

### *Acoustic Monitoring*

A total of 20 acoustic sampling nights each were recorded in four different habitats (Forest, Remnant, Corn, Citrus — Appendix I) from sunset (based on Bhutan Time (BTT) recorded in the study, the sunset time during autumn season ranges from 06:43–06:54 pm, and in winter the sunset time ranges from 05:06–05:15 pm) until midnight. In addition, 20 sampling nights occurred from paddy rice and remnant habitats in the month of October to November 2018 (paddy rice is grown from September and November). Acoustic recordings from paddy fields in autumn were added to the samples taken from other habitats recorded in summer. For comparison, it is suggested that acoustic sampling from different habitats should be conducted in the same period (Walsh *et al.*, 2004). This requirement was missed as rice is grown in a different period from corn. However, because there was no significant seasonal variation in bat activity between summer and autumn in the remnant forest, it was assumed that the seasons may not have a significant impact on bat activity at least in the remnant forest. For remnant forest, only samples from summer were included in the analyses of habitat selection.

Echolocation calls of insectivorous bats were recorded using a D240x Ultrasound Detector (Pettersson Elektronik AB, Uppsala, Sweden) connected to a digital Zoom H2 recorder (ZOOM Corporation). The recorders were set to trigger automatically, and was recorded for 1.7 s, then played back for 17 s. The trigger level was set to 'low'. The digitized bat play back calls were stored in the Zoom H2 recorder. The bat detector was kept in a metallic box (25 × 20 × 10 cm) and mounted on a pole 1.5–2 m above ground level. The microphone was tilted to 45° facing away from obstacles that might distract its recordings. Each sampling station was a minimum distance of 500 m apart. The temperature and relative humidity were recorded every 30 mins by a data logger (TM-305U — Tenmars Electronics Co., Ltd, Taiwan). Sampling was not conducted on nights with unfavorable weather conditions or more than 75% illuminated moon.

### *Establishing Call Reference and Identifying Echolocation Calls*

Mist-nets (3 m x 14 m) and harp traps (1.4 m wide and 1.4 m high) were set across forest trails, remnant, rivers and in the paddy fields to capture bats, while in roosting places sweep nets were used. A total of 47 individuals were captured from three different sites (forest, remnant and river). We measured the morphological characteristics of every captured bat, such as mass (gram), forearms length (mm), ear length (mm), tail length (mm) and sex. Then we identified each one to species following Bates and Harrison (1997) "Bats of the Indian Subcontinent". These species included: *Hipposideros armiger*, *Rhinolophus affinis*, *R. lepidus*, *R. luctus*, *R. pearsonii*, *Tylonycteris fulvina*. For more complex species, bacula (*Pipistrellus javanicus*, *Kerivoula hardwickii*) and skull/dentition (*Hipposideros cf. larvatus*) were used to confirm the species identification with reference to specimens in the Princess Maha Chakri Sirindhorn Natural History Museum, Prince of Songkla University.

The echolocation calls of captured bats were recorded by using a D240x Ultrasound Detector. Bats with constant frequencies were recorded in-hand such as *Rhinolophus* spp. and *Hipposideros* spp. to reduce the frequency changes due to Doppler shift (Schnitzler and Denzinger, 2011). Those bats with frequency modulated calls were recorded when they were released in the open area, such as *P. javanicus*. Rare species were recorded by allowing them to fly around in the confined room (8 × 5 × 4 m). Echolocation calls were analyzed using Bat Sound software, version 4.2.1 (Pettersson Elektronik AB, Uppsala, Sweden) with a sampling rate of 44.1 kHz and FFT size to 1024 with a Hanning Window. Call parameters including call duration (d), frequency of maximum energy (FmaxE), frequency at half of call duration (Fmiddle), frequency at the beginning of call (Fstart), highest frequency (Fhighest), minimum/end frequency (Fmin) and call interval were measured following Pretoni *et al.* (2005). The frequency-based characters were measured from the dominant harmonic in kHz from the spectrogram. Call duration and call interval were measured in 'ms' from the oscillogram. Calls were identified to species or genus by comparing them to the call reference collection in this study (Appendix II). Those bat species that we were not able to capture from the study area and for which no local call reference was established, call identification was based on existing studies. We referred to Bumrungsrir and Parson (2005) for *Hipposideros cf. lekaguli*, *Scotophilus kuhlii* and *Taphozous* sp.; Wordley *et al.* (2014) for *Myotis horsfieldii*; Russo and Jones

(2002) for *Nyctalus leisleri*; Deshpande and Kelkar (2015) for *Otomops wroughtoni*. Apart from *O. wroughtoni*, these bats are already known to occur in Bhutan and were recorded in the guide book, "Mammals of Bhutan", by Wangchuk *et al.* (2004). One unknown taxa with quasi-constant frequency (QCF) calls was classified as an unknown QCF bat (Appendix II). For further authentication, the call types shown in the spectrogram of *O. wroughtoni* and an unknown QCF bat are provided in Appendix III (Appendix IIIA and IIIB, respectively). For *O. wroughtoni* we confirmed our recorded calls for this rare species by comparing them with those reported from the Western Ghats, India, by Deshpande and Kelkar (2015). In their study, they compared the echolocation calls of this species with three other molossid bats, namely *Tadarida teniotis*, *T. aegyptiaca* and *Chaerephon plicatus*. All the species were clearly distinguishable based on end-frequency and peak-frequency. Thus compared to those three species, *O. wroughtoni* was fairly well matched based on the two parameters mentioned above (Appendix II). Our recordings showed an FM-sweep call structure that ranges from 14.9–16.2 kHz with mean peak frequency of 15.4 (± 0.4) kHz (Appendix IIIA).

The echolocating bats in the present study were categorized into guilds following Schnitzler and Kalko (2001) and Denzinger and Schnitzler (2013). For example, open-space bats (insectivorous bats that forage in open spaces) include bats recorded from the families Molossidae, Emballonuridae, and Vespertilionidae. Rhinolophid bats were categorized as narrow-space bats (bat species that forage close to surfaces or in cluttered habitats). For hipposiderid bats found in this study (*H. armiger*, *H. cf. larvatus* and also *H. lekaguli* (similar size to *H. cf. larvatus*) were designated as edge-space bats following Furey and Racey (2016).

### *Insect Samplings*

The relative abundance of insects was sampled simultaneously from 55 sites (11 sites in each habitat) using a custom-made, light-suction trapping method. White LED lights, model no. LE-5050 and CPU cooling case suction fans with 4Pins, DC, 12 V, 120 mm were powered by 9 V rechargeable batteries. The traps were suspended 1–1.5 m above the ground at least 30 to 40 m away from the acoustic station on the same sampling night. Trapped insects were identified to order based on Triplehorn and Johnson (2005). Insect biomass was estimated with

$$W = (0.0305) L^{2.62},$$

where W denotes dry mass (mg) and L = body length (mm) following Rogers *et al.* (1976).

### *Data Analysis*

Measures of bat activity were calculated as the number of bat passes per hour. A bat pass was defined as a sequence of search-phase calls recorded within one second (Kalda *et al.*, 2015). Feeding buzzes which are feeding attempts were also measured. Feeding buzzes were differentiated from search-phase calls by a rapid fall of pulses into shorter inter-pulse call sequences (Griffin *et al.*, 1960).

Bat passes and feeding buzzes per hour and insect biomass were compared between habitats. A Shapiro-Wilk test showed that our data were not normally distributed, therefore we used a non-parametric statistical analysis. We found two outliers in our data after completing a box-and-whisker plot (Quinn and

Keough, 2002). These sites (one in remnant forest and one in a paddy field) were adjacent to continuous forest and may not be representative of regional variability in the categorized habitats, so we removed these outliers from the analysis. To analyze bat activity patterns, we used Kruskal-Wallis test, because the data on bat activity is represented by continuous data (bat passes/h). Dunn's post-hoc test was applied for pairwise comparisons unless a Bonferroni correction was needed. Kendall-Theil regression with the Siegel method as the default was used to test the correlation between bat passes, insect biomass, and other environmental factors (temperature and relative humidity). The richness was represented by count data (number species in the habitat), therefore general linear model (GLM) with Poisson distribution was applied to evaluate species richness between habitat types. All statistical analyses were conducted in R version 3.4.1 and RStudio version 1.0.153 (RStudio, Inc.). The significance level was set to 0.05 for all tests.

## RESULTS

A total of 2,558 bat passes with 137 feeding buzzes were recorded in 535 hours of recording time period. Fourteen species were identified, including one unidentified species, representing eight genera in the families Emballonuridae, Hipposideridae, Molossidae, Rhinolophidae and Vespertilionidae. Twelve species were recorded acoustically from the forest and 10 species in remnant, nine in citrus orchard, eight in cornfield and eight in paddy field. General linear model revealed no significant variation in species richness between habitats (deviance = 155.5, d.f. = 99,  $P > 0.05$ ). The five most dominant species recorded were *S. kuhlii* (30.2% of total bat passes) followed by *Taphozous* sp. (19.6%), *O. wroughtoni* (16.8%), *N. leisleri* (13.1%) and *R. luctus* (8.2%). These species were found in all habitats, except *R. luctus* which was recorded in forest, remnant and citrus (Table 1).

Bat activity was significantly different between habitats (Kruskal-Wallis test,  $\chi^2 = 19.76$ , d.f. = 4,  $P < 0.01$  — Fig. 1). Bat activity in forest (median = 4.2 passes/hr,  $Q_1-Q_3 = 2.1-8.1$ ) was significantly higher ( $P < 0.05$ ) than in paddy (median = 1.0 passes/hr,  $Q_1-Q_3 = 0.5-1.8$ ) and in remnant (median = 0.8 passes/hr,  $Q_1-Q_3 = 0.1-2.8$ ). But bat activity in forest was not significantly different from that in corn (median = 3.1 passes/hr,  $Q_1-Q_3 = 1.7-8.8$ ) and citrus (median = 4.6 passes/hr,  $Q_1-Q_3 = 1.4-11.9$ ) with  $P > 0.05$ . Feeding buzzes were also significantly different between habitats (Kruskal-Wallis test,  $\chi^2 = 13.03$ , d.f. = 4,  $P < 0.05$  — Fig. 2). The feeding buzzes in the cornfield were significantly different from paddy and remnant ( $P < 0.05$ ), but no significant differences were observed among citrus, forest, paddy rice and remnant habitats ( $P > 0.05$ ).

## Guilds and Their Activity

When comparing bat activity among guilds, there was a significant difference (Kruskal-Wallis test,  $\chi^2 = 8.6332$ , d.f. = 2,  $P < 0.05$ ). Activity of open-space foragers was significantly higher than edge and narrow-space foragers where  $P < 0.05$  and  $P < 0.01$  respectively, but there was no significantly different between edge and narrow-space forager ( $P$ -value = 0.4). Further analysis of open-space foragers revealed that the activity of *S. kuhlii* was significantly different between habitats (Kruskal-Wallis test,  $\chi^2 = 20.9$  d.f. = 4,  $P < 0.001$ ). In corn and citrus, the activity of *S. kuhlii* was significantly higher than in remnant forest ( $P < 0.05$ ) (Fig. 3A). The activity of *Taphozous* sp. and *N. leisleri* was also significantly different between habitats (Kruskal-Wallis test,  $\chi^2 = 15.53$ , d.f. = 4,  $P < 0.05$ ; and  $\chi^2 = 16.51$ , d.f. = 4,  $P < 0.01$ , respectively — Fig. 3B and 3C). The activity of *Taphozous* sp. and *N. leisleri* in the corn and citrus habitats was significantly higher than in paddy ( $P < 0.05$ ). Although the activity of *O. wroughtoni* was statistically different among habitats (Kruskal-Wallis test,  $\chi^2 = 11.23$ , d.f. = 4,  $P < 0.02$ ), post-hoc tests using the Bonferroni adjustment revealed no significant differences.

## Insect Biomass

A total of 10,223 mg of insect biomass from eight different orders was sampled. The insect biomass in the forest, corn and citrus habitats was significantly higher than in paddy ( $P < 0.05$ ). There was no significant difference among the corn, citrus

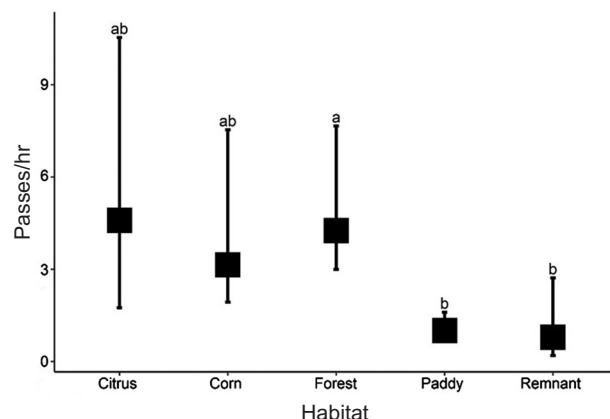


FIG. 1. Bat activity in passes per hour recorded from different habitats. The boxplot shows the medians with quantiles of bat activity (passes/hr) for each habitat along with 95% confidence intervals, indicated by error bars. Groups sharing the same letters are not significantly different ( $\alpha = 0.05$ )

TABLE 1. Bat pass per hour ( $\bar{x} \pm \text{SE}$ ) recorded in five habitats. Bat calls are divided into guild from different habitats. Paddy fields were surveyed in the autumn (October–November 2018) while the other habitats were surveyed in summer (May–July 2018)

Family/Species	Forest	Remnant	Citrus	Corn	Paddy	Guilds
Emballonuridae						
<i>Taphozous</i> sp.	$5.55 \pm 1.24$	$1.84 \pm 0.41$	$6.25 \pm 1.40$	$10.85 \pm 2.43$	$0.68 \pm 0.15$	open-space forager
Hipposideridae						
<i>Hipposideros armiger</i>	$0.05 \pm 0.01$	0.0	0.0	0.0	0.0	edge-space forager
<i>H. cf. larvatus</i>	$0.10 \pm 0.02$	$0.16 \pm 0.04$	0.0	0.0	$0.21 \pm 0.05$	edge-space forager
<i>H. lekaguli</i>	0.0	0.0	$0.15 \pm 0.03$	0.0	$0.68 \pm 0.15$	edge-space forager
Molossidae						
<i>Otomops wroughtoni</i>	$4.75 \pm 1.06$	$4.00 \pm 0.89$	$4.00 \pm 0.89$	$3.05 \pm 0.68$	$6.16 \pm 1.38$	open-space forager
Rhinolophidae						
<i>Rhinolophus affinis</i>	0.0	$0.26 \pm 0.06$	0.0	0.0	0.0	narrow-space forager
<i>R. lepidus</i>	$0.55 \pm 0.12$	$0.21 \pm 0.05$	$0.05 \pm 0.01$	$0.05 \pm 0.01$	$0.32 \pm 0.07$	narrow-space forager
<i>R. luctus</i>	$9.45 \pm 2.11$	$0.78 \pm 0.19$	$0.20 \pm 0.04$	0.0	0.0	narrow-space forager
<i>R. pearsonii</i>	$1.60 \pm 0.36$	$0.45 \pm 0.12$	$0.05 \pm 0.01$	$0.10 \pm 0.02$	0.0	narrow-space forager
Vespertilionidae						
<i>Myotis horsfieldii</i>	$2.05 \pm 0.46$	0.0	0.0	0.0	0.0	edge-space forager
<i>Pipistrellus javanicus</i>	$0.05 \pm 0.11$	$0.89 \pm 0.20$	$1.75 \pm 0.39$	$1.05 \pm 0.23$	0.0	edge-space forager
<i>Scotophilus kuhlii</i>	$3.70 \pm 0.83$	$3.63 \pm 0.81$	$15.55 \pm 3.48$	$13.50 \pm 3.02$	$2.53 \pm 0.56$	open-space forager
<i>Nyctalus leisleri</i>	$4.75 \pm 1.06$	$2.11 \pm 0.47$	$6.10 \pm 1.36$	$3.65 \pm 0.82$	$0.26 \pm 0.06$	open-space forager
Unknown QCF sp.	$0.10 \pm 0.02$	$2.15 \pm 1.74$	$0.60 \pm 0.13$	$0.37 \pm 0.08$	0.0	edge-space forager
Mean per habitat	$35.0 \pm 7.84$	$14.6 \pm 3.27$	$34.9 \pm 7.80$	$32.8 \pm 7.35$	$11.7 \pm 2.64$	
Species richness	12	10	9	8	8	

and remnant habitats ( $P = 0.93$ ), but forest was significantly higher than remnant ( $P < 0.05$ ). The majority of insect biomass was from four orders: Hymenoptera (median = 12 mg,  $Q_1-Q_3 = 0.0\text{--}53.5$ ), Lepidoptera (median = 21.7 mg,  $Q_1-Q_3 = 4.6\text{--}44.5$ ), Coleoptera (median = 8.1 mg,  $Q_1-Q_3 = 1.8\text{--}67.8$ ) and Diptera (median = 3.4 mg,  $Q_1-Q_3 = 1.2\text{--}9.0$ ). The proportion of insect orders varied between habitats and it was significantly different within the various habitats, (Kruskal-Wallis test,  $\chi^2 = 22.53$ ,  $d.f. = 4$ ,  $P < 0.001$ ). Furthermore, our results showed a significant positive correlation between bat activity and insect biomass ( $\tau = 0.42$ ,  $P < 0.001$ ), but there was no significant correlation between bat passes and other environmental factors such as temperature, humidity, or elevation. Feeding buzzes also correlated with insect biomass ( $\tau = 0.25$ ,  $P = 0.01$ ).

## DISCUSSION

Our results revealed that the use of different habitats was associated with insect availability. In this forest-dominated landscape, rice paddy and remnant were the least ideal foraging habitats, especially for *Rhinolophus*. However, citrus orchard and forest have provided major foraging habitats for insectivorous bats in this region.

The present study is also the first record of *O. wroughtoni* in Bhutan.

In addition to having the highest insect biomass, old-growth forest also provides shelter from predators (Walsh and Harris, 1996; Lima and O'Keefe, 2013) and an ambient environment that reduces thermoregulatory costs (Vonhof, 1995). It should be

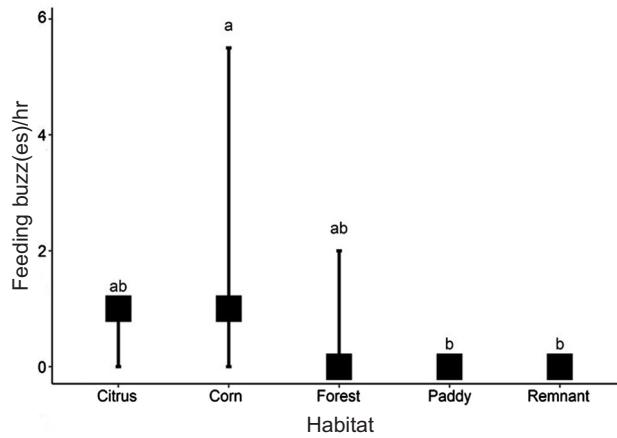


FIG. 2. Feeding buzzes per hour recorded from different habitats. The boxplot shows the medians with quantiles of feeding buzzes for each habitat along with the 95% confidence intervals using the percentile method. Error bars indicate the 95% confidence intervals for the median with the percentile method. Groups sharing the same letters are not significantly different ( $\alpha = 0.05$ )

noted, however, that the use of old-growth forest by bats in this study was under-represented, as dense forest obscure the foraging activity captured by bat detectors (Yates and Muzika, 2006). Cornfield and citrus orchards were the primary feeding grounds of two dominant open-space foragers, *Taphozous* sp. and *S. kuhlii*. A fecal analysis of *S. kuhlii* (Srinivasulu *et al.*, 2010; Chen *et al.*, 2016) and *Taphozous* sp. (Wei *et al.*, 2008; Weterings *et al.*, 2015) reported that Coleoptera is their main prey and these insects had the largest biomass in cornfields and citrus orchards. Traditional organic farming practices in this area may also enrich food resources for bats. Organic farming supports a high diversity and high biomass of insects (DeWalt *et al.*, 2003), which make these sites favored by insectivorous bats (Wickramasinghe *et al.*, 2003).

The results of our study show that old-growth forest is a better foraging habitat than remnant forest especially for narrow-space bats such as *Rhinolophus*. In old-growth forest, the activity of *R. luctus* was almost 10 times higher than in remnant forest.

This species usually roosts in hollow, fallen logs, which are the most common in old-growth forest (Duckworth *et al.*, 1999). By contrast, the remnant forest was partially disturbed due to human activities such as fodder and firewood collection, gathering shed leaves for composting, and continuous grazing by livestock, all of which are common practices in the study area (T. Dendup, personal observation). However, the number of the species recorded acoustically in the remnant forest was almost equal with that of old-growth forest. This indicates that the remnant forest could provide night roosting sites for these insectivorous bats (Fuentes-Montemayor *et al.*, 2013), especially hipposiderid and rhinolophid bats which forage in mixed-remnant forest-farmland landscapes. In addition, remnant forests can serve as corridors between forest patches within farmland landscapes (Bowen *et al.*, 2007). Therefore, it is good to maintain fragmented forest in an agricultural landscape to support bat diversity, which in turn may help to suppress to abundance of pests (Russo *et al.*, 2018).

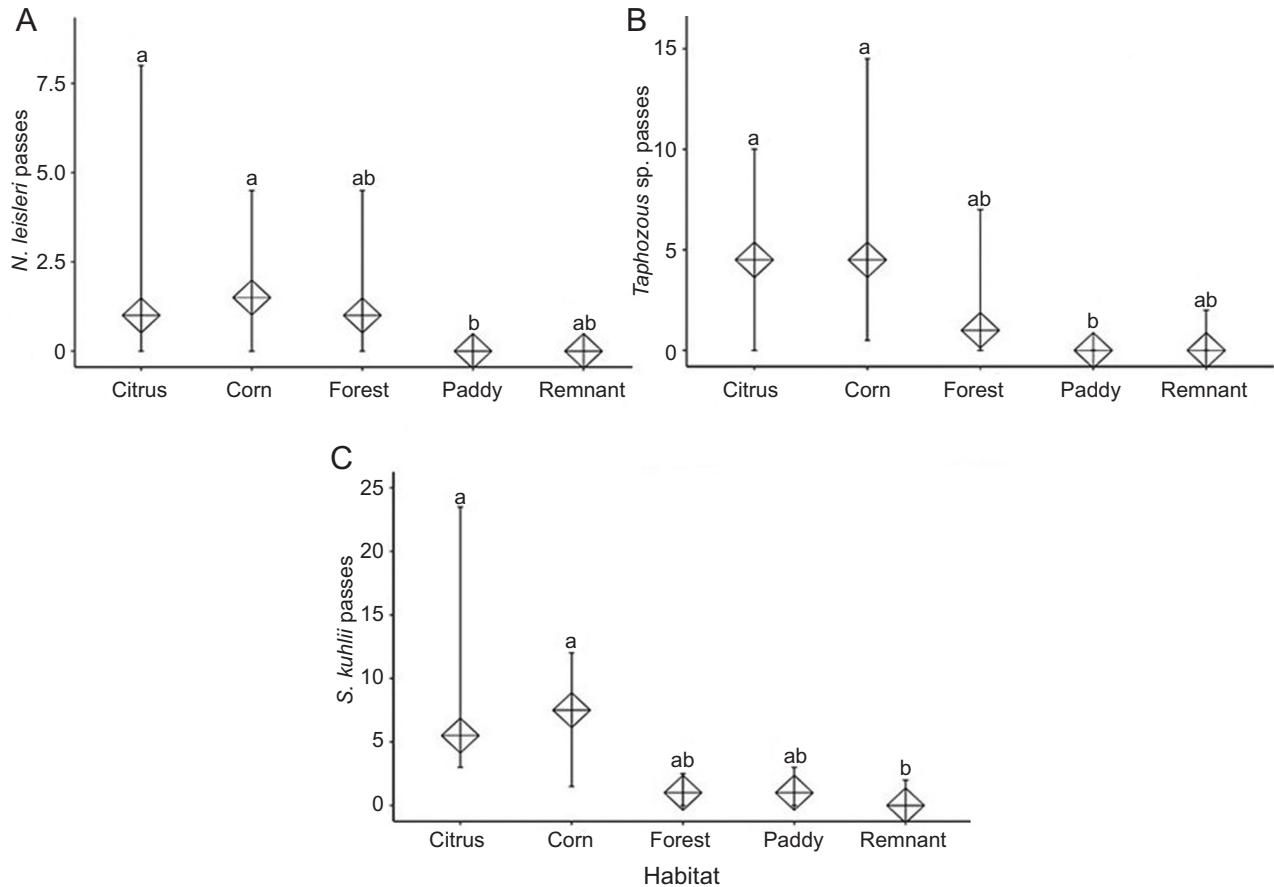


FIG. 3. The median of passes by each bat species in different habitats: (A) *N. leisleri*, (B) *Taphozous* sp. and (C) *S. kuhlii*. Plot of medians and confidence intervals for each habitat with 95% confidence intervals, indicated by error bars, using the percentile method. Groups sharing the same letters are not significantly different ( $\alpha = 0.05$ )

Overall, open-space foragers dominated all the habitats including old-growth forest. For example, *S. kuhlii*, *Taphozous* sp. and *N. leisleri* were most abundant in open space habitats like cornfield and citrus orchard. Their morphology with long and narrow wings are well suited to forage in open habitats (Altringham, 1996). In old-growth forest, these bats could forage opportunistically over the canopy for insects such as Diptera, which generally swarm above the canopy at dusk (Kunz and Fenton, 2003). Contrarily, *R. luctus*, *R. affinis*, *R. pearsonii* and *R. lepidus* were rarely recorded in open habitats. These bats have short, broad wings with low aspect ratios, low wing loading (Norberg and Rayner, 1987, also see Furey and Racey, 2016) and echolocation calls that are designed for foraging in cluttered habitats (Schnitzler and Kalko, 2001; Denzinger and Schnitzler, 2013). Consequently, if anthropogenic activities further fragment or degrade old-growth forest, narrow-space foragers such as small rhinolophid and hipposiderid bats, both known as forest specialists (Kingston, 2010), may be negatively affected because they are restricted to forested areas and avoid commuting through open space (Furey and Racey, 2016; Huang *et al.*, 2019). As Huang *et al.* (2019) demonstrated that bats with small body size and short and high-frequency echolocation calls were extremely sensitive to vegetation simplification, therefore foragers in cluttered habitats are more vulnerable to forest fragmentation than generalist aerial foragers.

In this study, *O. wroughtoni*, which was thought to be rare (Ruedi *et al.*, 2014), was commonly recorded in every habitat, especially in paddy fields. A several studies have demonstrated that molossid bats are pest suppressors in open farmland, including rice fields (Brown *et al.*, 2015; Srilopan *et al.*, 2018). *Otomops wroughtoni* has been found in India's Western Ghats and the Jaintia Hills District of Meghalaya, (Ruedi *et al.*, 2014), about 250 km from the present study, and in Cambodia (Walston and Bates, 2001). It was reported to form small colonies in limestone crevices within large openings of collapsed dolines (Ruedi *et al.*, 2014). Such crevices, although not common, can be found in the study area, especially in the steep slopes of rocky outcrops and in crevices along the river sides, which might provide suitable roosting sites for this species. They may forage at high altitude to follow their prey as reported for another molossid bat; *C. plicatus* (Nguyen *et al.*, 2019). This study is the first to reveal the foraging habitat of this little-known bat as the acoustic surveys are highly effective for rare bats

that have loud, clear acoustic signatures (Deshpande and Kelkar, 2015). Acoustic monitoring has proved to be an appropriate technique for bat surveys, however, it has its own intrinsic technical constraint that would miss low intensity calls e.g. whispering bat species (Hayes, 2000; Zukal and Řehák, 2006), which might influence current study results. Another caution in this study is acoustic monitoring was conducted mainly in summer except in paddy field. Thus, the finding revealed the summer pattern of habitat selection only. Lower temperature of other seasons could reduce both insect and bat activity (Lacki, 1984) and bat selection patterns may be different from the current study.

In conclusion, although anthropogenic activity has had a notable negative impact on bats (Fenton *et al.*, 1992; Kunz and Lumsden, 2003; Mendes *et al.*, 2017), the presence of patchily distributed agriculture in the forest-dominated landscapes resulted to a mosaic of foraging habitats for insectivorous bats. Furthermore, remnant and regrowth forest in the agricultural landscape has enhanced the structural connectivity (Fuentes-Montemayor, 2013) and apparently served as a corridor for the narrow-space foragers in the study landscape in Bhutan.

### *Conservation Implication*

As far as we know, this is the first study of the foraging activity of insectivorous bats in this region and may therefore provide a baseline for acoustic studies of echolocating bats. Further studies on the ecology of, and threats to, *O. wroughtoni* are needed to bridge the gap in the data deficiencies (Prabhukanolkar, 2016) of this species. As open space bats (*S. kuhlii*, *Taphozous* sp., *O. wroughtoni* and *N. leisleri*) were commonly recorded in agricultural landscapes in our study, additional studies on these common bats are needed, as they probably play an important role in insect pest control in farmland (Russo, *et al.*, 2018). To enhance the connectivity of landscapes and to enrich bat diversity, future conservation plans should include protecting and maintaining remnant forests in agricultural landscapes and encouraging traditional organic farming systems.

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Declaration of interest. The authors declare that there is no conflict of interest.

## LITERATURE CITED

- ALTRINGHAM, J. D. 1996. Bats, biology and behaviour. Oxford University Press, Inc., New York, 272 pp.
- ANTROP, M. 2005. Why landscapes of the past are important for the future. *Landscape and Urban Planning*, 70: 21–34.
- BATES, P. J. J., and D. L. HARRISON. 1997. Bats of the Indian Subcontinent. Harrison Zoological Museum Publications, Sevenoaks, England, 258 pp.
- BIANCHI, F. J. J. A., C. J. H. BOOIJ, and T. TSCHARNTKE. 2006. Sustainable pest regulation in agricultural landscapes: a review on landscape composition, biodiversity and natural pest control. *Proceedings of the Royal Society*, 273B: 1715–1727.
- BOWEN, M. E., C. A. MCALPINE, A. P. HOUSE, and G. C. SMITH. 2007. Regrowth forests on abandoned agricultural land: a review of their habitat values for recovering forest fauna. *Biological Conservation*, 140: 273–296.
- BROWN, V. A., E. B. DE TORREZ, and G. F. McCACKEN. 2015. Crop pests eaten by bats in organic pecan orchards. *Crop Protection*, 67: 66–71.
- BUMRUNGSRI, S., and D. PARSON. 2005. Acoustic identification of insectivorous bats in southern Thailand. Bat Research Unit, Prince of Songkhla University, Thailand, 55 pp.
- CHEN, S. F., S. S. HUANG, D. J. LU, and T. J. SHEN. 2016. Post-natal growth and age estimation in *Scotophilus kuhlii*. *Zoo Biology*, 35: 35–41.
- DENZINGER, A., and H.-U. SCHNITZLER. 2013. Bat guilds, a concept to classify the high diverse foraging and echolocation behaviors of microchiropteran bats. *Frontiers in Physiology*, 4: 164.
- DESHPANDE, K., and N. KELKAR. 2015. Acoustic identification of *Otomops wroughtoni* and other free-tailed bat species (Chiroptera: Molossidae) from India. *Acta Chiropterologica*, 17: 419–428.
- DEWALT, S. J., S. K. MALIAKAL, and J. S. DENSLAW. 2003. Changes in vegetation structure and composition along a tropical forest chronosequence: implications for wildlife. *Forest Ecology and Management*, 182: 139–151.
- DUCHAMP, J. E., and R. K. SWIHART. 2008. Shifts in bat community structure related to evolved traits and features of human-altered landscapes. *Landscape Ecology*, 23: 849–860.
- DUCKWORTH, J. W., R. E. SALTER, and K. KHOUNBOLINE (comp.). 1999. Wildlife in Lao PDR: 1999 status report. IUCN, Vientiane, Lao PDR, xiv + 275 pp.
- FENTON, M. B., L. ACHARYA, D. AUDET, M. B. C. HICKEY, C. MERRIMAN, M. K. OBRIST, and D. M. SYME. 1992. Phyllostomid bats (Chiroptera: Phyllostomidae) as indicators of habitat disruption in the Neotropics. *Biotropica*, 24: 440–446.
- FREY-EHRENBOLD, A., F. BONTADINA, R. ARLETTAZ, and M. K. OBRIST. 2013. Landscape connectivity, habitat structure and activity of bat guilds in farmland-dominated matrices. *Journal of Applied Ecology*, 50: 252–261.
- FUENTES-MONTEMAYOR, E., D. GOULSON, L. CAVIN, J. M. WALLACE, and K. J. PARK. 2013. Fragmented woodland character and landscape context on bats and their insect prey. *Agricultural, Ecosystem and Environment*, 172: 6–15.
- FUKUI, D., M. MURAKAMI, S. NAGANO, and T. AOI. 2006. Effect of emerging aquatic insects on bat foraging in a riparian forest. *Journal of Animal Ecology*, 75: 1252–1258.
- FUREY, N., and P. A. RACEY. 2016. Can wing morphology inform conservation priorities in Southeast Asian cave bats? *Biotropica*, 48: 545–556.
- GRIFFIN, D. R., F. A. WEBSTER, and C. R. MICHAEL. 1960. The echolocation of flying insects by bats. *Animal Behaviour*, 8: 141–154.
- HAYES, J. P. 2000. Assumptions and practical considerations in the design and interpretation of echolocation-monitoring studies. *Acta Chiropterologica*, 2: 225–236.
- HEIM, D., M. TSCHAPKA, KNORNSCHILD, and K. JUNG. 2015. The importance of landscape elements for bat activity and species richness in agricultural areas. *PLoS ONE*, 10: e0134443.
- HUANG, J. C.-C., E. L. RUSTIATI, M. NUSALAWO, and T. KINGTON. 2019. Echolocation and roosting ecology determine sensitivity of forest-dependence bats to coffee agriculture. *Biotropica*, 51: 757–768.
- HUTSON, A. M., S. P. MICKLEBURGH, and P. A. RACEY. 2001. Microchiropteran bats: global status survey and conservation action plan. IUCN/SSC Chiroptera Specialist Group, Gland, Switzerland, x + 258 pp.
- KALDA, O., R. KALDA, and J. LIIRA. 2015. Multi-scale ecology of insectivorous bats in agricultural landscapes. *Agriculture, Ecosystems and Environment*, 199: 105–113.
- KINGSTON, T. 2010. Research priorities for bat conservation in Southeast Asia: a consensus approach. *Biodiversity and Conservation*, 19: 471–484.
- KLINGBEIL, B. T., and M. R. WILLIG. 2009. Guild-specific responses of bats to landscape composition and configuration in fragmented Amazonian rainforest. *Journal of Applied Ecology*, 46: 203–213.
- KUNZ, T. H., and M. B. FENTON (eds.). 2003. Bat ecology. University of Chicago Press, Chicago, IL, 779 pp.
- KUNZ, T. H., and L. F. LUMSDEN. 2003. Ecology of cavity and foliage roosting bats. Pp. 3–89 in Bat ecology (T. H. KUNZ and M. B. FENTON, eds.). University of Chicago Press, Chicago, 779 pp.
- LACKI, M. J. 1984. Temperature and humidity-induced shifts in the flight activity of little brown bats. *Ohio Journal of Science*, 84: 264–266.
- LACKI, M. J., S. K. AMELON, and M. D. BAKER. 2007. Foraging ecology of bats in forests. Pp. 83–127 in *Bats in forests: conservation and management* (M. J. LACKI, J. P. HAYES, and A. KURTA, eds.). Johns Hopkins University Press, Baltimore, MD, xvi + 329 pp.
- LAURENCE, W. L., T. E. LOVEJOY, H. L. VASCONCELOS, E. M. BRUNA, R. K. DIDHAM, and P. C. STOUFFER, C. GASCON, R. O. BIERREGAARD, S. G. LAURANCE, and E. SAMPAIO. 2002. Ecosystem decay of Amazonian forest fragment: a 22-year investigation. *Conservation Biology*, 16: 605–618.

- LIIRA, J., T. AAVIK, O. PARREST, and M. ZOBEL. 2008. Agricultural sector: rural environment and biodiversity in the central and eastern European EU member states. *Acta Geographica Debrecina: Landscape & Environment*, 2: 46–64.
- LIMA, S. L., and J. M. O'KEEFE. 2013. Do predators influence the behaviour of bats? *Biological Reviews*, 88: 626–644.
- MENDES, E. S., M. J. R. PEREIRA, S. F. MARQUES, and C. FONSECA. 2014. A mosaic of opportunities? Spatio-temporal patterns of bat diversity and activity in a strongly humanized Mediterranean wetland. *European Journal of Wildlife Research*, 60: 651–664.
- MENDES, E. S., C. FONSECA, S. F. MARQUES, D. MAIA, and M. J. RAMOS PEREIRA. 2017. Bat richness and activity in heterogeneous landscapes: guild-specific and scale-dependent? *Landscape Ecology*, 32: 295–311.
- MINISTRY OF HOME AND CULTURAL AFFAIRS. 2011. National Rehabilitation Program. Available at [https://www.nics.gov.bt/?page\\_id=50](https://www.nics.gov.bt/?page_id=50). Accessed 24 June 2021.
- NEUHOFF, D., S. TASHI, G. RAHMANN, and M. DENICH. 2014. Organic agriculture in Bhutan: potential and challenges. *Organic Agriculture*, 4: 209–201.
- NGUYEN, T. N., A. RUANGWISET, and S. BUMRUNGSRI. 2019. Vertical stratification in foraging activity of *Chaerephon plicatus* (Molossidae, Chiroptera) in Central Thailand. *Mammalian Biology*, 96: 1–6.
- NORBERG, U. M., and J. M. V. RAYNER. 1987. Ecological morphology and flight in bats (Mammalia; Chiroptera): wing adaptations, flight performance, foraging strategy and echolocation. *Philosophical Transactions of the Royal Society of London*, 316B: 335–427.
- PRABHKHANOLKAR, R. 2016. *Otomops wroughtoni*. The IUCN Red List of Threatened Species 2016: e.T15646A22112971. Available at <https://dx.doi.org/10.2305/IUCN.UK.2016-2.RLTS.T15646A22112971.en>.
- PREATONI, D. G., M. NODARI, R. CHIRICHELLA, G. TOSI, L. A. WAUTERS, and A. MARTINOLI. 2005. Identifying bats from time-expanded recordings of search calls: comparing classification methods. *Journal of Wildlife Management*, 69: 1601–1614.
- QUINN, G. P., and M. J. KEOUGH. 2002. Experimental design and data analysis for biologists. Cambridge University Press, Cambridge, 537 pp.
- ROBINSON, R. A., and W. J. SUTHERLAND. 2002. Post-war changes in arable farming and biodiversity in Great Britain. *Journal of Applied Ecology*, 39: 157–176.
- ROBINSON, R. A., J. D. WILSON, and H. Q. P. CRICK. 2001. The importance of arable habitat for farmland birds in grassland landscapes. *Journal of Applied Ecology*, 38: 1059–1069.
- ROGERS, L. E., W. T. HINDS, and R. L. BUSCHBOM. 1976. A general weight vs. length relationship for insects. *Annals of the Entomological Society of America*, 69: 387–389.
- RUEDI, M., K. B. MUKHIM, O. M. CHACHULA, T. ARBENZ, and A. THABAH. 2014. Discovery of new colonies of the rare Wroughton's free-tailed bat *Otomops wroughtoni* (Mammalia: Chiroptera: Molossidae) in Meghalaya, northeastern India. *Journal of Threatened Taxa*, 6: 6677–6682.
- RUSSO, D., and G. JONES. 2002. Identification of twenty-two bat species (Mammalia: Chiroptera) from Italy by analysis of time-expanded recordings of echolocation calls. *Journal of Zoology (London)*, 258: 91–103.
- RUSSO, D., and G. JONES. 2003. Use of foraging habitats by bats in a Mediterranean area determined by acoustic surveys: conservation implications. *Ecography*, 26: 197–209.
- RUSSO, D., L. BOSSO, and L. ANCILLOTTO. 2018. Novel perspectives on bat insectivory highlight the value of this ecosystem service in farmland: research frontiers and management implications. *Agriculture, Ecosystem and Environment*, 266: 31–38.
- SCHNITZLER, H. U., and A. DENZINGER. 2011. Auditory forevea and Doppler shift compensation: adaptation for flutter detection in echolocating bats using CF-FM signals. *Journal of Comparative Physiology*, 197A: 541–559.
- SCHNITZLER, H.-U., and E. K. V. KALKO. 2001. Echolocation by insect-eating bats. *Bioscience*, 51: 557–569.
- SRILOPAN, S., S. JANTARIT, and S. BUMRUNGSRI. 2018. The wrinkle-lipped free-tailed bat (*Chaerephon plicatus* Buchanan, 1800) feeds mainly on brown planthoppers in rice fields of central Thailand. *Acta Chiropterologica*, 20: 207–219.
- SRINIVASULU, B., C. SRINIVASULU, and P. VENKATESHWARLU. 2010. First record of lesser yellow house bat *Scotophilus kuhlii* Leach, 1821 from Secunderabad, Andhra Pradesh, India with a note on its diet. *Journal of Threatened Taxa*, 2: 1234–1236.
- TRIPPLEHORN, C. A., and N. F. JOHNSON. 2005. Borror and DeLong's introduction to the study of insects. Thomson Brooks/Cole, Belmont, CA, 888 pp.
- VONHOF, M. J. 1995. Roost-site preferences of big brown bats (*Eptesicus fuscus*) and silver-haired bats (*Lasionycteris noctivagans*) in the Pend d'Oreille Valley in Southern British Columbia. Pp. 62–79, in *Bats and Forests Symposium*, October 19–21, 1995, Victoria, British Columbia, Canada (R. M. R. BARCLAY and R. M. BRIGHAM, eds.). Research Branch, B.C. Ministry of Forests, Victoria, British Columbia, Working Paper 23/1996, ix + 80 pp.
- WALSH, A. L., and S. HARRIS. 1996. Foraging habitat preferences of vespertilionid bats in Britain. *Journal of Applied Ecology*, 33: 508–518.
- WALSH, A. L., R. M. R. BARCLAY, and G. F. McCACKEN. 2004. Designing bat activity surveys for inventory and monitoring studies at local and regional scales. Pp. 157–165, in *Bat echolocation research, tool, techniques and analyses* (R. M. BRIGHAM, E. K. V. KALKO, G. JONES, S. PARSON, and J. G. A. LIMPENS, eds.). Bat Conservation International, Austin, TX, 167 pp.
- WALSTON, J., and P. BATES. 2001. The discovery of Wroughton's free-tailed bat *Otomops wroughtoni* (Chiroptera: Molossidae) in Cambodia. *Acta Chiropterologica*, 3: 249–252.
- WANGCHUK, T., P. THINLEY, K. TSHERING, C. TSHERING, and D. YONTEN. 2004. A field guide to the mammals of Bhutan. Bhutan Trust Found for Environmental Conservation, Thimphu, Bhutan, 179 pp.
- WEATHER AND CLIMATE SERVICES DIVISION. 2018. Climate data book of Bhutan. National Center for Hydrology and Meteorology, United Printing Press, Thimphu, 261 pp.
- WEI, L., N. HAN, L. ZHANG, K. M. HELGEN, S. PARSONS, S. ZHOU, and S. ZHANG. 2008. Wing morphology, echolocation calls, diet and emergence time of black-bearded tomb bats (*Taphozous melanopogon*, Emballonuridae) from southwest China. *Acta Chiropterologica*, 10: 51–59.
- WETERINGS, R., J. WARDENAAR, S. DUNN, and C. UMPONSTIRA. 2015. Dietary analysis of five insectivorous bat species from Kamphaeng Phet, Thailand. *Raffles Bulletin of Zoology*, 63: 91–96.
- WICKRAMASINGHE, L. P., S. HARRIS, G. JONES, and N. V. JENNINGS. 2003. Abundance and species richness of nocturnal

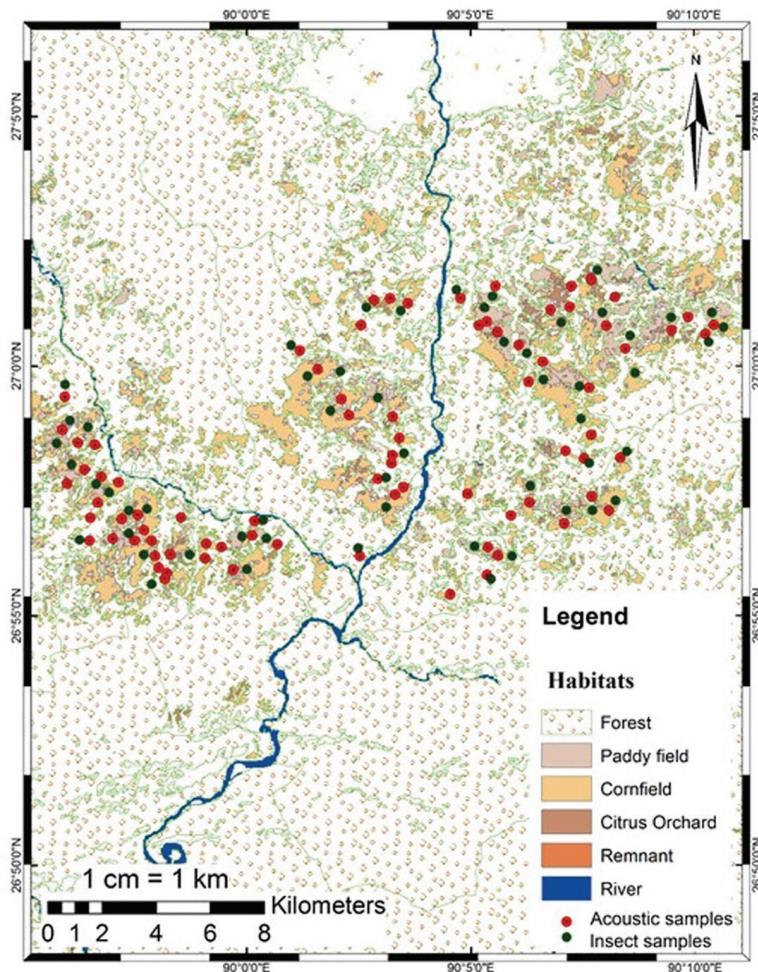
- insects on organic and conventional farms: effects of agricultural intensification on bat foraging. *Conservation Biology*, 18: 1283–1292.
- WORDLEY, C. F. R., E. K. FOUI, D. MUDAPPA, M. SANKARAN, and J. D. ALTRINGHAM. 2014. Acoustic identification of bats in the southern Western Ghats, India. *Acta Chiropterologica*, 16: 213–222.
- YATES, M. D., and R. M. MUZIKA. 2006. Effect of forest structure and fragmentation on site occupancy of bat species in Missouri Ozark forests. *Journal of Wildlife Management*, 70: 1238–1248.
- ZUKAL, J., and Z. ŘEHÁK. 2006. Flight activity and habitat preference of bats in a karstic area, as revealed by bat detectors. *Folia Zoologica*, 55: 273–281.

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## APPENDIX I

Distribution of acoustic sampling points ( $n = 80$ ) and insect samples ( $n = 55$ ) from different habitats. Sampling points in paddy fields and remnant forest ( $n = 20$  each) in the autumn season (September–November 2018) are not shown on the map because the samplings from paddy fields are the same as those from cornfields. These crops are grown in the same place in different seasons. Remnant forest samplings are taken from the same sites in summer (June–August 2018) and autumn



## APPENDIX II

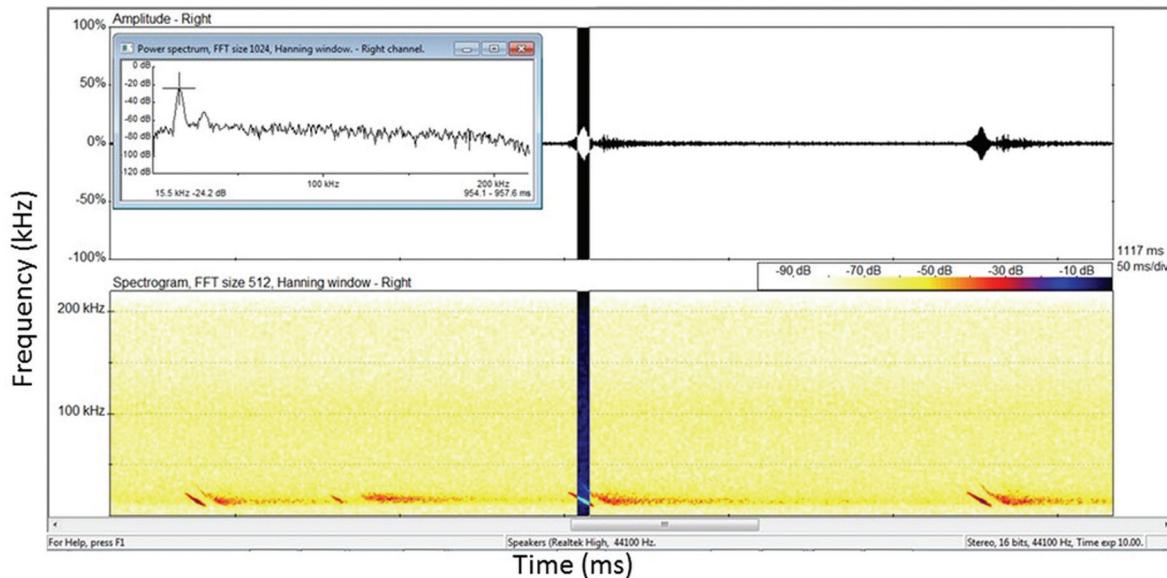
Mean ( $\bar{x}$ ), standard deviation (SD), and range of at least seven call characteristics: frequency of maximum energy (FmaxE), frequency at half of the call duration (Fmiddle), frequency at the beginning of call (Fstart), highest frequency (Fhighest), and minimum frequency (Fmin), call duration (d), and call interval. The total number of individuals recorded is denoted by letter ‘n’

Species	n	d	FmaxE	Fmiddle	Fstart	Fhighest	Fmin	Call interval	Dominant harmonic	References
<i>Taphozous</i> sp.	5	$\bar{x}$	10.1	30.4	30.1	40.7	43.5	24.6	140.5	2nd
		SD	0.9	1.4	1.7	3.2	3.6	1.9	41.6	Bumrungsri and Parson (2005)
		max.	11.5	31.8	32.0	43.4	48.0	27.0	212.5	
		min.	9.2	28.1	28.0	36.5	39.6	22.0	112.2	
<i>Hipposideros armiger</i>	3	$\bar{x}$	8.2	69.0	67.9	67.3	70.7	58.5	40.5	2nd
		SD	0.9	1.0	1.1	1.3	1.1	0.2	8.4	This study
		max.	9.1	69.6	69.0	68.3	71.7	58.3	50.1	
		min.	7.3	67.8	66.8	65.8	69.5	58.3	34.6	
<i>Hipposideros</i> cf. <i>larvatus</i>	5	$\bar{x}$	8.3	82.7	81.8	80.4	84.1	69.1	54.6	2nd
		SD	0.6	0.9	0.7	1.5	0.7	2.9	19.2	This study
		max.	9.2	83.7	82.8	82.0	85.0	72.0	87.2	
		min.	7.6	81.6	81.0	78.8	83.3	64.3	39.8	
<i>Hipposideros lekaguli</i>	4	$\bar{x}$	4.9	44.5	42.7	44.1	45.8	29.9	97.3	2nd
		SD	0.4	3.0	3.4	4.0	3.8	4.9	12.2	Bumrungsri and Parson (2005)
		max.	5.3	48.0	46.0	48.0	49.0	35.0	110.4	
		min.	4.4	41.8	39.0	40.0	42.0	24.6	86.3	
<i>Rhinolophus affinis</i>	2	$\bar{x}$	40.6	78.4	76.8	73.7	78.5	65.2	99.1	2nd
		SD	5.3	0.4	0.3	5.2	0.7	4.5	8.8	This study
		max.	44.3	78.6	77.0	77.3	79	68.3	105.3	
		min.	36.8	78.1	76.6	70.0	78.0	62.0	92.8	
<i>Rhinolophus lepidus</i>	7	$\bar{x}$	41.7	102.3	100.3	93.4	104.7	85.7	95.5	2nd
		SD	8.7	2.6	3.0	2.8	3.4	2.9	13.1	This study
		max.	55.0	105.2	104.5	97.5	108.8	89.5	113.2	
		min.	33.0	99.5	96.7	89.0	100.5	81.0	72.6	
<i>Rhinolophus luctus</i>	4	$\bar{x}$	40.2	31.0	30.3	27.0	32.8	23.8	87.2	2nd
		SD	4.6	0.1	0.4	0.2	0.7	1.4	11.9	This study
		max.	43.8	31.0	30.7	27.3	33.7	25.3	101.4	
		min.	33.4	30.9	29.8	26.8	32.0	22.0	76.4	
<i>Rhinolophus pearsonii</i>	6	$\bar{x}$	43.3	63.4	61.8	56.9	64.8	43.2	115.6	2nd
		SD	11.1	0.8	1.1	2.5	1.4	3.1	55.1	This study
		max.	58.3	64.9	64.0	59.8	67.7	47.8	178.0	
		min.	26.6	62.7	61.0	53.0	64.0	40.5	33.8	
<i>Myotis horsfieldii</i>	5	$\bar{x}$	4.9	48.5	49.7	65.4	68.0	45.4	78.7	1st
		SD	1.1	1.1	2.4	5.3	5.3	1.3	6.4	Wordley <i>et al.</i> , 2014
		max.	5.9	50.0	53.0	71.0	73.7	46.5	87.2	
		min.	3.6	47.2	47.3	59.0	61.8	43.3	69.4	
<i>Nyctalus leisleri</i>	6	$\bar{x}$	11.4	31.2	31.3	37.3	39.1	27.8	175.8	Russo and Jones (2002)
		SD	1.8	1.4	2.3	3.6	3.7	1.3	47.3	
		max.	13.9	33.3	35.5	41.0	43.5	30.3	250.0	
		min.	8.4	29.7	29.0	32.5	34.5	27.0	110.0	
<i>Scotophilus kuhlii</i>	6	$\bar{x}$	7.1	39.6	39.5	52.7	54.2	35.8	93.0	1st
		SD	0.9	1.7	1.2	5.9	6.4	2.3	15.9	Bumrungsri and Parson (2005)
		max.	8.4	42.4	41.5	64.3	66.5	40.0	113.2	
		min.	6.1	38.0	38.3	48.5	49.8	33.0	69.6	
<i>Pipistrellus javanicus</i>	7	$\bar{x}$	2.7	50.6	54.8	90.0	95.0	41.7	84.8	1st
		SD	0.7	1.4	8.8	12.8	13.0	1.0	61.4	This study
		max.	3.5	52.6	71.7	107.0	112.5	43.0	219.1	
		min.	1.9	48.4	48.0	72.0	75.7	40.3	36.9	
<i>Otomops wrouhgtoni</i>	7	$\bar{x}$	11.2	15.4	16.2	22.8	23.8	11.3	283.5	1st
		SD	4.9	0.4	0.8	1.0	0.9	1.3	218.4	Despande and Kelkar (2015)
		max.	19.0	16.2	17.5	24.0	25.0	13.0	754.0	
		min.	5.7	14.9	15.3	21.0	22.5	9.3	138.6	
Unknown QCF bat	4	$\bar{x}$	8.6	45.8	44.1	47.9	49.3	42.4	113.0	1st
		SD	1.2	4.1	4.2	4.4	4.2	3.7	5.5	This study
		max.	10.9	50.7	49.0	55.0	56.6	47.3	125.0	
		min.	7.6	40.7	38.5	43.0	45.2	38.0	108.4	

## APPENDIX III

Echolocation calls of bats recorded in the study area: A) the call of *O. wroughtoni* showing the FM-sweep call structure of free-flying bats, which ranges from 9.3–13.0 kHz with an average minimum frequency of 11.3 ( $\pm 1.3$ ) kHz; B) calls of an unknown QCF bat showing the QCF call type of free-flying bats. The call ranges from 38.0–47.33 kHz of the minimum frequency with an average of 42.43 ( $\pm 3.75$ ) kHz

A



B

