# Diet preferences of insectivorous bats (Mammalia: Chiroptera) in Chambok, Kampong Speu Province, Cambodia

SIN Sopha<sup>1,\*</sup>, CHHORN Soksan<sup>1</sup>, DOEURK Bros<sup>1</sup>, HAK Kosal<sup>2</sup>, ITH Saveng<sup>2</sup>, PHAUK Sophany<sup>1</sup> & SOR Ratha<sup>2,3,4</sup>

- <sup>1</sup> Cambodian Entomology Initiatives, Department of Biology, Faculty of Science, Royal University of Phnom Penh, Confederation of Russia Boulelvard, Phnom Penh, 12156, Cambodia.
- <sup>2</sup> Centre for Biodiversity Conservation, Royal University of Phnom Penh, Confederation of Russia Boulevard, Phnom Penh, 12156, Cambodia.
- <sup>3</sup> International Relations Office, Chea Sim University of Kamchaymear, No. 157, Preah Norodom Boulevard, Khan Chamkarmon, Phnom Penh, 12300, Cambodia.
- <sup>4</sup> Wonders of the Mekong Project, c/o Inland Fisheries Research and Development Institute, No. 186, Preah Norodom Boulevard, Khan Chamkarmon, P.O. Box 582, Phnom Penh, 12300, Cambodia.
- \* Corresponding author. Email sopha.sin702@gmail.com

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# មូលន័យសង្ខេប

សត្វប្រចៀវដើរតូនាទីយ៉ាងសំខាន់ក្នុងការកំចាត់សត្វល្អិតចង្រៃ។ ដោយហេតុថា ការស្រាវជ្រាវពីបំណែកសត្វល្អិតក្នុងលាមកសត្វ ប្រចៀវនៅមានកម្រិតនៅឡើយ យើងបានប្រមូលសំណាកលាមកប្រចៀវពីតំបន់ទេសចរណ៍ធម្មជាតិចំបក់ ក្នុងខេត្តកំពង់ស្ពឺ ដើម្បីធ្វើ ការសិក្សា និងប្រៀធៀបពីប្រភេទអាហារដែលវាចូលចិត្ត។ សំណាកលាមកចំនួន២៩៤គ្រាប់ ត្រូវបានប្រមូលពីសត្វប្រចៀវ ៥ប្រភេទ (Rhinolophus shameli, R. pusillus, Hipposideros pomona, H. cf. larvatus និង Megaderma spasma) ដែលក្នុងចំនោម នោះ ១២០គ្រាប់ត្រវបានយកទៅពិនិត្យជាមួយមីក្រទស្សន៍។ សំណាកលាមករបស់ប្រចៀវទាំងអស់មានបំណែករបស់សត្វល្អិតចំនួន ៩លំដាប់ ហើយបំណែកមួយចំនួនទៀតជារបស់ពពួកចៃ និងកណ្តុរ។ លំដាប់សត្វល្អិតដែលសត្វប្រចៀវចូលចិត្តស៊ីច្រើនបំផុតគឺ Lepidoptera និង Coleoptera និងបន្ទាប់មកគឺ Heteroptera, Hymenoptera, Homoptera និង Diptera។ ចំណែកឯ លំដាប់សត្វល្អិតដែលសត្វប្រចៀវមិនសូវចូលចិត្តស៊ីមាន Isoptera, Trichoptera និង Orthoptera និង Diptera។ ចំណែកឯ លំដាប់សត្វល្អិតដែលសត្វប្រចៀវមិនសូវចូលចិត្តស៊ីមាន Isoptera, Trichoptera និង Orthoptera និងពពួកចៃ និងកណ្តុរ។ លទ្ធផលបានបង្ហាញទៀតថា សត្វប្រចៀវ rhinolophid និង hipposiderid ចូលចិត្តស៊ីសត្វល្អិតក្នុងបរិមាណខុសៗគ្នាក្នុងកម្រិត ជឿជាក់ រឹងប្រចៀវប្រភេទ *M. spasma* មានចំណូលចិត្តស៊ីសត្វឆ្អឹងកងតូចៗ។ ការសិក្សាស្រាវជ្រាវបន្ថែមពិតជាចាំបាច់ដើម្បីបង្កើន ចំណេះដឹងអំពីចំណូលភាពនៃចំណីអាហាររបស់សត្វប្រចៀវទាំងអស់ដែលមានវត្តមាននៅកម្ពុជា ពីព្រោះសត្វប្រចៀវចូលរួមយ៉ាង សំខាន់ក្នុងការផ្តល់សេវាកម្មប្រព័ន្ធអកូឡូស្តីដែលទាក់ទងនឹងសេដ្ឋកិច្ច។

# Abstract

Bats provide economically significant ecosystem services in insect pest suppression. As little is known regarding the diets of insectivorous bats in Cambodia, we sampled these in the Chambok area of Kampong Speu Province in August 2018 so as to describe and compare their specific diet preferences. Our field survey yielded 294 faecal pellets produced by five insectivorous species (*Rhinolophus shameli, R. pusillus, Hipposideros pomona, H. cf. larvatus* and *Mega-derma spasma*), 120 of which were examined by microscopy. These samples contained fragments representing nine insect

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orders, and included fragments of mites (Acari) and Rodentia. The most common orders in terms of their percentage frequency of occurrence were Lepidoptera and Coleoptera, followed by Heteroptera, Hymenoptera, Homoptera and Diptera, whereas the least common were Rodentia, Acari, Isoptera, Trichoptera and Orthoptera. Comparisons based on the percentage volume of the most common orders indicated significant differences in the amounts consumed by rhinolophid and hipposiderid bats, whereas *M. spasma* showed a preference for small vertebrates. Further studies are warranted to develop understanding of the diet preferences of Cambodian bats, not least because these likely give rise to economically significant ecosystem services.

**Keywords** Ecosystem services, frequency percentage, frequency volume, *Hipposideros, Megaderma, Rhinolophus*.

# Introduction

Insectivorous bats provide economically significant ecosystem services in insect pest consumption (Boyles *et al.*, 2011). For example, the value of these services to agriculture in the continental USA has been estimated at approximately US\$ 22.9 billion /year, including the reduced costs of pesticides that are not needed to suppress the insects consumed by bats (Boyles *et al.*, 2011). In Thailand, populations of the wrinkle-lipped free-tailed bat *Mops plicatus* (Molossidae) are regarded as important pest control agents because they may consume as many as 55 tons of insects per night, a significant portion of which comprises major agricultural pests (Leelapaibul *et al.*, 2005).

Analysis of bat faeces can provide valuable information on the insect taxa consumed by bat species. These have shown that insect orders commonly consumed by insectivorous bats include Homoptera, Coleoptera, Heteroptera, Lepidoptera, Diptera, Hymenoptera, Neuroptera, Trichoptera, Odonata and Araneae (Leelapaibul *et al.*, 2005; Oliveira *et al.*, 2015). Some studies have shown that certain bat species mostly consume soft-bodied insects such as moths, whereas other species are less specific in their preferences (Bogdanowicz *et al.*, 1999; Ghazali & Dzeverin, 2013; Weterings & Umponstira, 2014).

At least 80 bat species including 68 animalivorous taxa are presently known to occur in Cambodia (Kingsada *et al.*, 2011; Neil Furey, unpublished data), although this figure is relatively low compared to neighbouring countries such as Lao PDR with  $\approx$ 100 bat species, Thailand with  $\approx$ 146 species and Vietnam with  $\approx$ 125 species (Soisook 2011; Kruskop 2013; Thomas *et al.*, 2013; Karapan *et al.*, 2016; Neil Furey, unpublished data). Aside from a single study on the cave nectar bat *Eonycteris spelaea* (Pteropodidae) (Hoem *et al.* 2017), nothing has been published regarding the diet preferences of bats inhabiting the country to date. To address this gap, we sampled insectivorous bats in the Chambok area of southwest Cambodia in August 2018 with the aim of documenting and comparing their diet preferences.

# Methods

#### Study area

Our study was undertaken in the Chambok Community-Based Ecotourism Site (hereafter 'Chambok') in Kampong Speu Province, southwest Cambodia. Chambok occupies a total area of 8,257 ha and borders Kirirom National Park (Lonn, 2013). The site includes three areas under different forms of community-based management: 1) community forestry areas established by the Forest Administration in 2005, which cover 286 ha and border the national park; 2) community protected areas created by the Ministry of Environment in 2002, which occupy 758 ha within the national park; and 3) community-based ecotourism areas created by the Ministry of Environment in 2003, which extend over 161 ha inside the national park (FAO, 2012). Four villages with a total population of 3,670 people are located within Chambok (Chambok Dangkum, Beng, Krangchek and Thmei). The natural vegetation of the Chambok landscape includes bamboo, degraded semievergreen forests and grasslands with deciduous forest.

#### Field sampling

We employed two four-bank harp traps and one mist net to sample seven sites in Chambok in August 2018. Six of these sites (S01-S06) were located within the interior of semi-evergreen forests at the site, whereas one site (S07) was situated in bamboo forest adjacent to agricultural land (Table 1). Our harp traps and mist net were employed from 18:00 to 21:00 hrs on a single occasion at each site. All insectivorous bats captured in the harp traps and net were gently removed by hand and retained in individual cloth bags until the following morning when their faeces were collected. Before release, each bat was identified to species based on its external morphology and standard measurements for the group using Francis (2008). The faecal pellets produced by each bat overnight were transferred from their individual cloth bags with soft forceps into appropriately labelled vials containing

70% ethanol and the bags used to retain individual bats were thoroughly cleaned before each reuse.

#### Faecal analysis

Insectivorous bats chew their prey into small fragments, the less digestible portions of which are passed into their faeces. The faecal samples obtained from bats in Chambok were processed by randomly selecting and placing one pellet at a time on a petri dish containing a few drops of glycerol and teasing this apart with a pair of needles under an Olympus SZ51 microscope. All insect fragments present in each faecal pellet (e.g., legs, wings, antennae) were then mounted onto slides for observation under an Olympus CX23 microscope and photographed using an OPTIKA Microscope P6 Pro Camera. These were sorted according to size and identified to order level using McAney *et al.* (1997), Pokhrel & Budha (2014) and Ponmalar & Vanitharani (2014).

#### Data analysis

Following identification, we visually estimated the percentage volume of each insect order in each faecal pellet by placing the relevant fragments on paper with six 1 x 1 cm grid squares (Fig. 1). In each instance, the number of squares employed for this purpose depended on the quantity of material for a given order in each faecal pellet. We then calculated the percentage frequency (PF) and percentage volume (PV) of each insect order using the following formulae (Kunz & Parsons, 2009):

$$PF = \frac{\text{number of fecal pellets in which food items present}}{\text{total number of fecal pellets}} \times 100$$
$$PV = \frac{\text{sum of the individual volume}}{\text{total volumes of samples}} \times 100$$



**Fig. 1** Grid scheme used to visually estimate the precentage volume of a given animal order in faecal samples.

As such, PF represents the average occurrence of each food type (insect order) consumed, whereas PV represents the average percentage volume of each food type. We employed PV values to classify each food type into one of four categories following Shetty & Sreepada (2013): 1) PV >20% representing 'preferred food'; 2) PV 5–20% representing 'frequent food'; 3) PV 1–5% representing 'supplementary food'; 4) PV <1% representing 'incidental food'. Where more than 10 faecal pellets were obtained for a bat species, Kruskal Wallis tests were used to test for significant differences in the PV values of each food type between bat species. Probability values <0.05 were regarded as significant and all statistical tests were undertaken using *IBM SPSS Statistics for Windows* vers. 21.0 (IBM, New York, USA).

Table 1 Sampling sites in Chambok, Kampong Speu Province, southwest Cambodia.

Site	Latitude, Longitude	Elevation (m)	Sampling Date	Habitat
S01	11°21'29.40"N, 104°06'09.0"E	407	13 August 2018	Semi-evergreen forest, ≈60 m from a stream
S02	11°21'39.6"N, 104°06'07.2"E	371	13 August 2018	Semi-evergreen forest along a stream
S03	11°21'45.5"N, 104°06'07.9"E	374	14 August 2018	Semi-evergreen forest with bamboo
S04	11°21'59.9"N, 104°06'13.7"E	266	15 August 2018	Mixed bamboo and semi-evergreen forest, ≈400m from a cave bat colony
S05	11°22'06.9"N, 104°06'25.0"E	203	15 August 2018	Mixed bamboo and semi-evergreen forest
S06	11°22'10.2"N, 104°06'33.1"E	177	16 August 2018	Mixed bamboo and semi-evergreen forest
S07	11°22'22.1"N, 104°06'41.4"E	123	16 August 2018	Bamboo forest and agriculture land use

# Results

We captured 58 individuals of five bat species belonging to three families: *Rhinolophus shameli*, *R. pusillus* (Rhinolophidae), *Hipposideros pomona*, *H.* cf. *larvatus* (Hipposideridae) and *Megaderma spasma* (Megadermatidae) (Fig. 2; Table 2). These provided a total of 294 faecal pellets, 120 of which were randomly selected for analysis (Tables 2 & 3).

The faecal samples included a total of nine insect orders (Coleoptera, Lepidoptera, Heteroptera, Homoptera, Diptera, Orthoptera, Hymenoptera, Trichoptera and Isoptera), as well as mites (Acari) and rodent fragments (Rodentia) (Fig. 3; Table 3). Insect fragments identified in faecal pellets produced by *R. pusillus* included eight insect orders, whereas those produced by *R. shameli*, *H. pomona* and *H.* cf. *larvatus* comprised six insect orders and one mite order. Food types observed in faeces produced by *M. spasma* differed greatly in including just two insect orders and fragments of a rodent (Table 3).

The most commonly eaten insect orders in terms of percentage frequency were Lepidoptera and Coleoptera, followed by Heteroptera, Hymenoptera, Homoptera and Diptera, whereas the least common were Rodentia,

**Table 2** Sample sizes of bat species and faecal pellets obtained in Chambok, August 2018.

Species	n	Male	No. ♂ pellets	Female	No. ♀ pellets	
Rhinolophus shameli	25	4	41	21	77	
R. pusillus	19	10	67	9	38	
Hipposideros pomona	5	-	-	5	58	
H. cf. larvatus	8	4	-	4	11	
Megaderma spasma	1	-	-	1	2	
Total	58	18	108	40	186	

Acari, Isoptera, Trichoptera and Orthoptera (Table 3). Based on percentage volume, *R. shameli* apparently preferred three insect orders (Lepidoptera, Coleoptera and Hymenoptera, with PV values of 33.3%, 31% and 20%, respectively), whereas *R. pusillus* preferred a single insect order (Lepidoptera, PV=38%) (Table 4, Fig. 4). In



**Fig. 2** Bat species captured in August 2018 in Chambok: A) *Rhinolophus shameli*, B) *R. pusillus*, C) *Hipposideros pomona*, D) *H. cf. larvatus*, E) *Megaderma spasma* (© Phauk S.).

contrast, *H. pomona* preferred Heteroptera (55.7%), while *H.* cf. *larvatus* preferred Heteroptera (54.4%) and Coleoptera (35.6%). Food types consumed on a supplementary or incidental basis included the orders Orthoptera, Trichoptera, Isoptera and Acari. Not surprisingly, the percentage volume of the most common six insect orders differed significantly between four bat species (all except *M. spasma* which was excluded from comparisons due to low sample sizes) (Fig. 4; Table 4).

# Discussion

Our study represents the first published information on the diets of insectivorous bats in Cambodia. All of the bat species we sampled are currently considered Least Concern (IUCN, 2020) and typically regarded as caveroosting taxa, although some also roost in anthropogenic sites (e.g., temples, mines or houses) or hollow trees (Francis, 2008).

We found the most common insect orders consumed by bats in Chambok in terms of percentage frequency were Lepidoptera and Coleoptera, followed by Heteroptera, Hymenoptera, Homoptera and Diptera, whereas the least common were Rodentia, Acari, Isoptera, Trichoptera and Orthoptera. Comparisons based on percentage volume indicated significant differences in the relative amounts of six insect orders consumed by rhinolophid and hipposiderid bats, whereas *M. spasma* showed a pref-

**Table 3** Percentage frequency of food types consumed by bat species in Chambok. Abbreviations: Col=Coleoptera,Lep=Lepidoptera, Het=Heteroptera, Hym=Hymenoptera, Hom=Homoptera, Dip=Diptera, Ort=Orthoptera, Tri=Trichoptera,Iso=Isoptera, Aca=Acari, Rod=Rodentia.

Species	No. of pellets	Col	Lep	Het	Hym	Hom	Dip	Ort	Tri	Iso	Aca	Rod
Rhinolophus shameli	40	78	90	53	68	20	30	-	-	-	5	-
R. pusillus	47	77	94	43	49	55	77	2	6	-	-	-
Hipposideros pomona	20	25	100	95	60	85	-	-	-	45	5	-
H. cf. larvatus	11	73	45	91	18	9	18	-	-	-	18	-
Megaderma spasma	2	100	100	-	-	-	-	-	-	-	-	100

**Table 4** Percentage volume of food types consumed by bat species in Chambok. Abbreviations for each type are provided in Table 3 above. Asterisks indicate significant differences in values for specific insect orders between bat species (\*\* P<0.01, \*\*\* P<0.001).

Species	No. of pellets	Col ***	Lep ***	Het ***	Hym **	Hom ***	Dip ***	Ort	Tri	Iso	Aca	Rod
Rhinolophus shameli	40	31.0 ±5.3	33.3 ±4.7	11.4 ±2.5	20.0 ±3.7	2.3 ±1.1	1.7 ±0.5	-	-	-	0.3 ±0.2	-
R. pusillus	47	14.2 ±2.4	38.0 ±3.9	11.0 ±3.4	10.3 ±1.8	17.2 ±2.8	8.9 ±1.8	0.2 ±0.2	0.3 ±0.2	-	-	-
Hipposideros pomona	20	4.9 ±2.6	12.2 ±2.1	55.7 ±4.4	6.3 ±1.9	18.6 ±2.7	-	-	-	2.3 ±0.8	0.1 ±0.1	-
H. cf. larvatus	11	35.6 ±11	3.6 ±1.6	54.4 ±12.8	2.1 ±1.4	2.5 ±2.5	1.2 ±0.9	-	-	-	$\begin{array}{c} 0.7 \\ \pm 0.5 \end{array}$	-
Megaderma spasma	2	4.0 ±1	7.5 ±0.5	-	-	-	-	-	-	-	-	88.5 ±1.5



**Fig. 3** Examples of insect fragments commonly found in bat faecal samples from Chambok: A) Coleoptera—anntennae, B) Lepidoptera—wing scales, C) Heteroptera—forewing, D) Hymenoptera—head, E) Homoptera—hind limbs, F) Diptera—abdomen.



Fig. 4 Percentage volume (mean ± standard error) of six insect orders in faecal samples of four bat species in Chambok.

erence for small vertebrates (Rodentia). In general terms, these differences could be due to variations in the life history and ecology (e.g., body size and foraging behaviour) of each species (Bogdanowicz *et al.*, 1999; Weterings & Umponstira, 2014).

Our data suggest that populations of *R. shameli* in Chambok may prefer lepidopteran, coleopteran and hymenopteran insects, whereas *R. pusillus* may prefer lepidopterans (all PV values  $\geq$ 20%). Rhinolophid bats are generally small-bodied animals which predisposes them to prey upon smaller, softer-bodied insects (Ponmalar & Vanitharani, 2014). Studies elsewhere have found that

members of the genus exhibit a preference for Lepidoptera, small Coleoptera and Diptera (Bogdanowicz *et al.*, 1999; Fukui *et al.*, 2009; Weterings & Umponstira, 2014).

The two hipposiderids we captured at Chambok (*H. pomona* and *H.* cf. *larvatus*) showed a marked preference for heteropterans (PV values >50%) and *H.* cf. *larvatus* also frequently consumed coleopterans (PV=35.6%). Although the two species differ in body size (*H. cf. larvatus* being larger than *H. pomona*), both taxa produce high-frequency echolocation signals whose call durations are shorter than those emitted by *R. shameli* and *R. pusillus* (Phauk *et al.*, 2013). Previous studies have

found that larger hipposiderids such as *H. armiger* and *H. diadema* also consume heteropterans and coleopterans (Pavey & Burwell, 1997; Weterings *et al.*, 2015).

Somewhat interestingly, fragments of mites (Acari) occurred in faecal samples produced by three of our bat species (*R. shameli, H. pomona* and *H. cf. larvatus*). Mites are parasitic invertebrates which occur on bats (Whitaker *et al.*, 1983; Baker & Craven, 2003) and have been found in faecal samples produced by *Hypsugo cadornae* (Vespertilionidae) in Thailand (Weterings *et al.*, 2015). They also parasitize coleopterans (Abbot & Dill, 2001; Almane & Elnov, 2009). Additionally, in containing fragments of Rodentia, our data for *M. spasma* is consistent with previous studies suggesting the taxon consumes small vertebrates (Balete, 2010; Vanitharani *et al.*, 2015). More broadly, the species is thought to forage on large and hard-bodied insects, lizards and other small vertebrates (Francis, 2008).

Our study provides a useful baseline for future research on the diets of insectivorous bat species in the Chambok area. They also demonstrate that these frequently feature insect orders which include important pest species for agriculture (e.g., Lepidoptera, Coleoptera and Heteroptera; Sallam, 2001; Thongdara *et al.*, 2009; Rabitsch, 2010) and to a lesser extent, disease vectors for humans (Diptera). As such, we recommend further studies to develop understanding of the diet preferences of Cambodian bats, not least because these likely give rise to economically significant ecosystem services.

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