

## Phlebotomine sand flies of edible-nest swiftlet cave of Lang Ga Jiew Island, Chumphon province, Thailand

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**Abstract.** The present study reported for the first time phlebotomine sandfly species inhabiting edible-nest swiftlet cave of the isolated island, based on field collections made during June 2010-May 2011. The insect diversity was relatively lower to that of mainland caves. All species, *Phlebotomus stantoni*, *Sergentomyia anodontis*, *Sergentomyia bailyi*, *Sergentomyia gemmea*, *Sergentomyia hodgsoni* and *Sergentomyia punjabensis* were either endemic island species or native elsewhere in Thailand. *Sergentomyia hodgsoni* was the most prevalent species accounted for 94.7% and classified as a troglophile species. Seasonal pattern of the phlebotomine abundance and some aspects of their population characteristics were described and discussed. Two ectoparasites, *Ornithodoros* and *Paracimex* sp. were also incidentally collected from the swiftlet cave.

### INTRODUCTION

Since the first reported autochthonous case of visceral leishmaniasis from southern Thailand in 1996, leishmaniasis received considerable attention at national level (Thisyakorn *et al.*, 1999). This was followed by several other indigenous cases (Kongkaew *et al.*, 2007; Maharom *et al.*, 2008; Sukmee *et al.*, 2008). Recently, the first case of cutaneous leishmaniasis was reported in a three-year old girl (Kattipathanapong *et al.*, 2012) in Thailand. The transmission cycle, the pattern of transmission and vectors responsible are of significant importance. Investigations on the natural occurrence of sand flies in Thailand, begun in 1987 following the increasing reports of labourers working in leishmaniasis endemic areas returning home infected. (Apiwathnasorn *et al.*, 1989). These surveys have been conducted in a broad range of habitats to determine the sand fly fauna and its geographic distribution with particular attention to anthropophilic,

zoophilic and cave-dwelling species. *P. hoepflii* and *P. major major* and the only anthropophilic species (Apiwathnasorn *et al.*, 1989; 1993; 2011; Polseela *et al.*, 2011a; 2011b). Appearance of sand flies in terms of high species diversity and apparent abundance was reported from many caves with sparse to clumped distribution (Apiwathnasorn *et al.*, 1989). Recently of the 26 species reported more than 10 species were of inland cave-dwelling sand flies. The sand fly fauna of sea caves remains unknown. The inaccessibility of the swiftlet islands make sand fly surveys challenging. This study reports for the first time the isolated island fauna of sand fly in a swiftlet cave that focused on diversity, species composition and seasonality.

### Study area

Lang Ga Jiew is a small offshore island of Chumphon province covering an area of 0.063 square kilometers and located 8 km away from the shore. Apart from being a tourist

attraction, the island is tendered as a bird's nest concession. The edible-nest swiftlets (*Collocalia fuciphaga*) are cave dwellers and their nesting areas are usually inaccessible for human and are primarily deep in limestone caves high up on the wall. The nests are usually harvested three times a year. The first nest-harvesting is in late March; around 45 days after the nests are built. The cave entrance (5 m diameter) is located on the limestone sloping cliff. From the entrance, a steep passage leads to the floor covering with swiftlet guano 20-30 cm thick. The depth from the entrance is approximately 28 meters. The cave temperature is  $27\pm1^{\circ}\text{C}$  with relative humidity of  $85\pm2\%$ . The visitors do not allow entering the bird's nest cave to avoid disturbing the swiftlets, especially during the breeding season (February-April). The little huts located on beach and in front of the cave are guard shack to protect the nests from being stolen and to protect the birds from natural predators such as monitors, snakes, geckos, seagulls and hawks.

## MATERIALS AND METHODS

Regarding permission obtained from the concessionaire to access the swiftlet cave we were requested to minimize our human impact on the bird living habitats. The sand flies were, therefore, captured by 3 CDC light traps hung from bamboo poles about 30-50 cm above the cave floor. They were monthly

operated from 1700-0700 h for 2 nights during June 2010-May 2011. All sand flies captured were stored in 95% alcohol, counted, sorted and identified according to sex and species. The morphological identification was based mainly on the keys and species descriptions provided by Lewis (1978, 1982).

## RESULTS

A total of 1,702 sand flies belonging to the genus *Phlebotomus* and *Sergentomyia* were collected. Six species inhabiting the edible-nest swiftlet cave were *P. stantoni*, *S. anodontis*, *S. bailyi*, *S. gemmea*, *S. hodgsoni* and *S. punjabensis* of which *S. hodgsoni* was the most common accounted for 94.7%. The other species accounted less than 5% of the total. The sex ratio of sand flies population slightly changed over time. Sex allocation of captured phlebotomines was 1,130 females (66.4%) and 572 males (33.6%). Sand flies inhabiting swiftlet cave occurred throughout the year with the lowest density of 5.7 and 10.7 flies per trap-night in September and January, respectively (Figure 1). The seasonal abundance of sand fly population showed trimodal pattern with two small peaks in April (67.0 flies per trap-night) and October (74.7 flies per trap-night) and the highest peak in June (170.7 flies per trap night). In addition *S. hodgsoni* and *S. barraudi* also occurred in very small number outside the cave nearby the guard shacks.

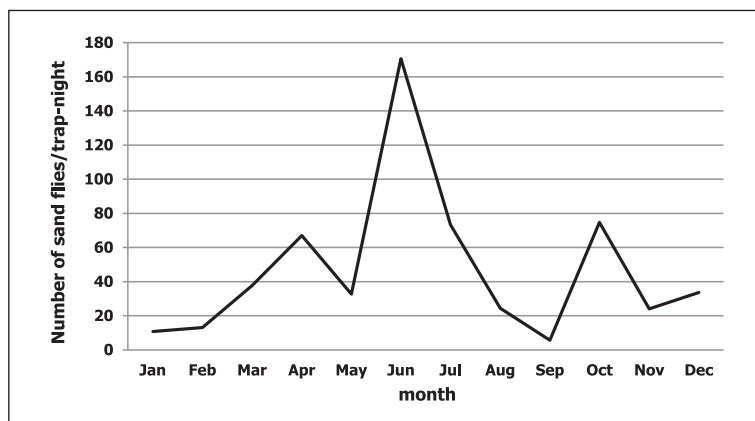


Figure 1. Seasonal variation of sand flies inhabiting swiftlet cave

There were frequent bug-bite complaints by bird's nest collector and guard while exposed to nest environment inside the cave. By investigating the complaint, attempts have been made to capture the responsible insects by direct search. Bat bugs of the genus *Paracimex* and bird-infesting soft ticks of the genus *Ornithodoros* were found from cracks and crevices in the cave walls and debris on the floor around bird nest areas.

## DISCUSSION

Ecosystems of the island cave are different from that of mainland communities owing to their relative simplicity of living communities and the isolation characteristics of the individual biota (Poulson & White, 1969). The fauna of sand flies in the present study had approximately two to three times lower in diversity on island than in the mainland habitats (Polseela *et al.*, 2007; Apiwathnasorn *et al.*, 2011; Polseela *et al.*, 2011b). The variation in species diversity probably is due to nature of living habitats on food availability, environmental conditions and selective pressures. Caves with higher resource availability had a higher biological complexity than those with less resource (Ferreira & Horta, 2001). Six species appeared to be endemic to the island swiftlet cave. They are also cavernicolous species in other inland limestone caves with varied density. Howarth (1987) revealed that the similarity of cave adaptations among various taxa in different caves is a general process and is the result of similar selection pressures. Number of sand flies in the cave varied throughout the year with a trimodal pattern in distribution as shown by the most frequently captured species *S. hodgsoni*. Therefore *S. hodgsoni* shows evidence of well adaptation to the cave environment. It was captured monthly and was also the most abundant. In addition *S. hodgsoni* also lived in subterranean environment and moist areas outside caves. This evidence demonstrates that it is a troglophile. The predominant species of sand flies were different and varied from cave to cave. In the present study *S. hodgsoni* was the most abundant species of

swiftlet island cave, whereas *Nemopalpus vietnamensis* occurred at highest densities in Naresuan Cave (Polseela *et al.*, 2011b). In addition *P. argentipes* have been reported as prevalent species in various limestone caves of Thailand together with common *Chinius barbazani*, *P. bargusae*, *P. statoni*, *S. anodontis* and *S. hodgsoni* (Apiwathnasorn *et al.*, 1989; 2011; Polseela *et al.*, 2011a). Individual species may respond and exploit differently to resource gradients of particular caves.

Males and females are produced in approximately equal numbers in most natural populations of animals with separate sexes (Omoloye, 2006). Female-biased sex allocation (1:2.0) observed in the present study was comparatively similar to those of inland caves in Phothisat Cave (1:2.5) and Naresuan Cave (1:1.3) reported by Polseela *et al.* (2011a; b). As mating takes place at or near hosts, the aggregation of males and females would depend on the distance from its hosts. Hence the female-biased sex ratio may be explained by this mating behaviour. Natural site-to-site variation in the sex ratio also depends on the number of hosts available. Placement variations which refer to the variability of light trap collection due to the location of the trap and/or trap bias possibly cause difference in sex ratio from places to places. Hamilton & Montgomerie (1989) found that technical errors in sampling method, time and location may lead to false estimation of sex ratio. It is also demonstrated by Stoks (2001) that operational sex ratios of the damselfly, *Lestes sponsa* were male-biased at the pond but became female-biased in a distance of 40 m from the shoreline. The main reason for this is that the female damselflies come to the pond for merely oviposition but stay away from the pond for several days to mature a new brood.

Caves have stable environments which enable the determination of the precise fauna, species composition and relative abundance of the various cavernicolous species as well as their interaction with other cave-adapted species and surrounding environments. Lewis (1971) stated that the cave-dwelling habitat was possibly a step in the development of anthropophily. Cimicidae, for

instance, probably evolved on bats and have subsequently spread onto birds and man, possibly because of early cave-dwelling habits (Busvine, 1976). Our anthropophilic species, *P. major*, were all collected outside the cave entrance. In conclusion, the present study revealed endemic species of phlebotomines that can be followed for further molecular, genetic diversity and evolution which may probably assist us in better understanding of vector potential and status of autochthonous leishmaniasis in Thailand.

Members of the genus *Paracimex* were primarily associated with the glossy swiftlet (*Collocalia esculenta* complex) with the distribution patterns rather more complex and parallel to the distribution of the *esculenta* complex (Ueshima, 1968). Only this species of the bat bug was found in the study island. There were reports of two species in Java, two on New Guinea and two in Bismark Archipelago. With regards to the soft ticks found during the study period, only *O. (Alectorobius) collocaliae* has been previously reported that is confined to the Oriental region belonging to hosts family *Apodidae* (Hoogstraal *et al.*, 1974). Approximately 12 argasid species including *Argas* and *Ornithodoros* are frequently reported attached to humans who intrude into tick-infested caves and burrows (Estrada-Peña & Jongejan, 1999). Little information is available on their medical significance in this region. Species identification is fundamentally required to contribute to the fauna of cave-swiftlet Argasid and to bring up awareness of risks associated with tick bites in cave environments. In addition, it should be interesting to know whether these ecto-parasites are pests in the bird-nest soup industry.

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