Annals of Parasitology

eISSN 2300-6706 ISSN 2299-0631 (formerly *Wiadomości Parazytologiczne*)

edited by the Polish Parasitological Society



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Tit	tle of paper:		A Report of Ectoparasites Bat (<i>Eonycteris spelaea</i>) Am Dwelling in Lombok Islan Tenggara	ong Cave-
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Keywords:	
Keyword 1:	Eonycteris spelaea
Keyword 2:	Ectoparasites
Keyword 3:	Cave
Keyword 4:	Lombok Island
Keyword 5:	West Nusatenggara

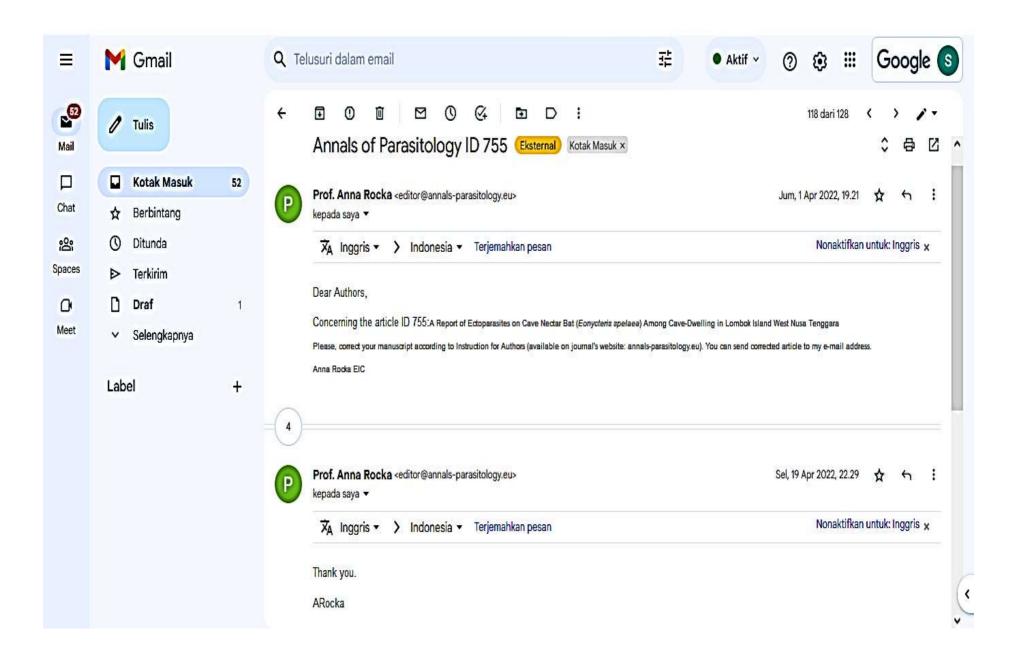


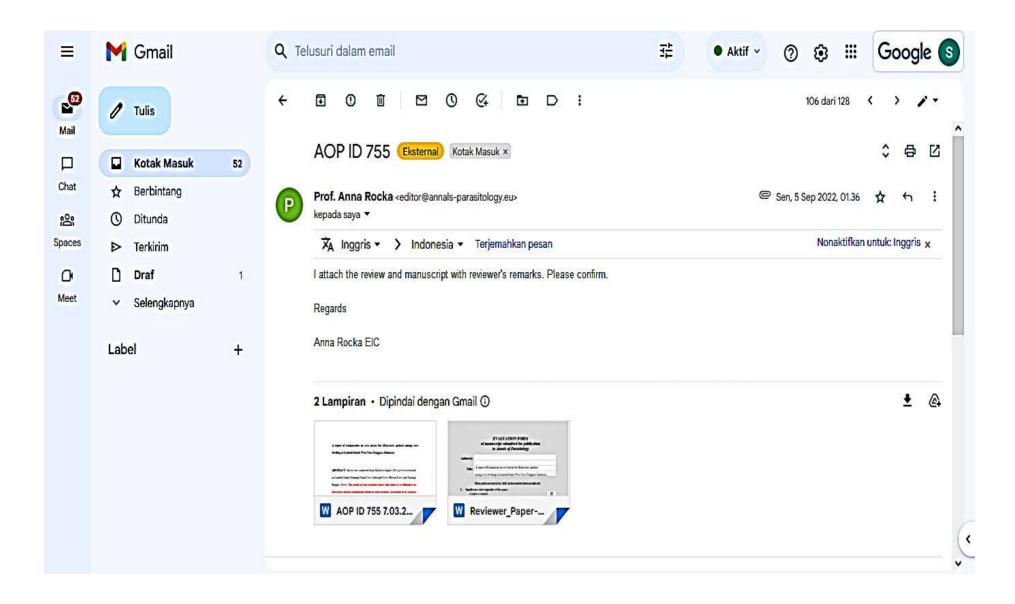
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A Report Of Ectoparasites On Cave Nectar Bat (Eonycteris Spelaea) Among Cave-

Dwelling In Lombok Island, West Nusa Tenggara, Indonesia

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ABSTRACT. Survey was conducted from March to August 2021 in 4 caves located in Lombok Island (Gunung Tunak Cave, Gale-gale Cave, Buwun Cave, and Tanjung Ringgit Cave). The result of this research shows that there is a difference in *Eonycteris spelaea* morphology found in each location. According to *E. spelaea* morphology analysis and identification found in some caves, it is known that from 42 species caught and morphologically analyzed, *E. spelaea* species found in Gale gale and Buwun Caves have similarity. It was seen from the similar size of forearm of the wings 78.8 mm and 74.9 mm. Furthermore, the sizes of tibia of the species were also similar with the value of 16.1 mm and 16.7 mm. Whereas, *E. spelaea* found in the other two caves (Tanjung Ringgit Cave and Gunung Tunak Cave), also showed similar size morphologically, such as the size of forearm wings with the values of 65.9 mm and 67.5 mm Regarding ectoparasite observation found in *Eonycteris spelaea*, there were 7 species of ectoparasites consisting of 4 species from the class Insecta in which 2 species from the order Diptera (*Eucampsipoda sundaica* and *Cyclopodia horsfieldii*), 2 species from the order Siphonaptera (*Ischnopsyllus*

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hexactenus and *Thaumapsylla breviceps*), and 3 species from the class Arachnida (*Amblyomma* sp., *Ancystropus* sp, and *Meristaspis* sp.).

Keywords: Eonycteris spelaea, ectoparasites, cave, Lombok Island, West Nusatenggara

Introduction

Family Pteropodidae consists of 42 genera and 182 species worldwide [1]. In Indonesia, this family has 21 genera and 72 species [2]. Members of this family are known as seed dispersers, flower pollinators, and guano producers.

Eonycteris spelaea is one of the bats that plays a role in pollinating flowers commonly found in caves. This is why the species is often called cave fruit bat or cave nectar bat. The distribution of *Eonycteris spelaea* is very wide ranging from Malay-Indonesia, the southern Chinese islands, and the Indian continent [1].

In addition, it is widely distributed from southwest and north of India, Southern China, Andaman, Myanmar (Burma), Malaya, Philippines, Sumatra, Java, Kalimantan, Sulawesi, Bali, Lombok, Sumba, Timor and Halmahera [3]; [4]; [5]; [6]

Morphologically, *E. spelaea* has medium size with weight ranging from 53-84 grams, long snout, second finger of the wings having no claws, kidney shaped glands on the left and right of the anus, and 2 pairs of small incisors [7]. The habitat of this species can be found in agricultural area, primary forest, secondary forest, and coastal mangrove [7];[8]. This species forages at a distance of more than 38 km from their nesting sites at night [9], while on a trip, *Eonycteris spelaea* has been

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reported to cover a distance of up to 17.9 km to 38 km between the roost and foraging areas in Thailand and Malaysia [9] and Khteas Cave in southern Cambodia [10].

In general, bats play an important role in ecosystems, including seed dispersal, pollination, and insect predation. Meanwhile, *Eonycteris spelaea* bat plays a special role as plant pollinator. Various types of plants have been reported to have pollination assisted by the bat species. These plants include durian (*Durio zibethinus*), a leguminous canopy tree *Parkia* spp, and petai (*P. speciosa*) and the Indian trumpet flower (*Oroxylum indicum*) in managed agricultural habitats in southern Thailand [11]; [12]. Thavry et al [10] Mentioned that, *Eonycteris spelaea E. spelaea* is an economically important pollinator for plants in Cambodia. The colony of *Eonycteris spelaea* is able to provide protection to mangrove plants and is also able to provide benefits to durian farmers.

The threats to bats population of *Eonycteris spelaea* are habitat loss, hunting [13], hunting for humans consumption [14] wildlife trade and death due to the presence of ectoparasites and bacteria in their bodies. Fruit bats are more likely to have ectoparasites than insectivorous bats [14]. The presence of ectoparasites can affect bats physical condition and will have an impact on long-term survival. Several studies have shown that ectoparasites tend to decrease the fecundity of bats. Ticks, mites, chiggers, bugs, fleas, and flies are some of the types of ectoparasites found in bats [15], [16]. Some ectoparasites are associated with pathogens that can cause disease in humans or animals, both wild and domestic [17]. Fajri et al [18] mentioned that there were 14 types of ectoparasites found in 9 species of bats that perched in caves on Lombok Island-Indonesia. The ectoparasites consist of Diptera, Siphonaptera, Ixodida, and

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Mesostigmata groups. The bat fly, *Cyclopodia horsfieldii*, one of the ectoparasites, dominated the ectoparasites found from 9 species of bats examined. In Batuputih Nature Tourism Park, Sulawesi-Indonesia, A total of 479 ectoparasites representing three families, namely Nycteribiidae, Streblidae (bat flies), and Spinturnicidae (mites) were collected. Majority of these ectoparasites (n = 475) belonged to the family Nycteribiidae genus Leptocyclopodia [14]. Sauqi et al [19] mention that identification of ectoparasites on fruit bats (*Cynopterus brachyotis*) was infested with *Leptocyclopodia ferrarii* (8%). Further, in Goa Jepang Bukit Plawangan, Sleman Yogyakarta Indonesia, showed that ectoparasite species were found in the bats of the Microchiroptera Suborder from the Subclass Acarina and the Insect Class. The species of Subclass Acarina found were *Periglischrus sp., Spinturnix plecotinus, Blattisocius sp.,* and *Glycyphagus sp.* Species of the Insect Class found include *Megastrebla sp., Stylidia caudata, Basilia sp.,* and *Brachytarsina sp.,* [20].

Thus, this study aims to analyze the morphology of bat species of *Eonycteris spelaea* in 4 caves in Lombok Island and to identify the ectoparasites present in *Eonycteris spelaea*. *Eonycteris spelaea* it is important to study ectoparasites in bats to determine their potential role as vectors of zoonotic pathogens [21]. New report from Singapore, Lim et al [22] mention that there are 261 species of *Eucampsipoda sundaica* found in *Eonycteris spelaea*. This research is also very important because the distribution of *Eonycteris spelaea* in the caves of Lombok Island is quite high.

Materials and Methods

Study area

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The study was conducted in Lombok Island in the four location (Figure 1), which included Central Lombok Regency: Bangkang Gale-gale Cave (S 08°53.188' and E 116°15.254' Elev 111m) and Buwun Cave (S 08°53.375' and E 116°15.160' Elev. 92m) (Prabu Village, Pujut District), Gunung Tunak Cave, and Mertak Village (S 08°56'6,61164" and E 116°23'45,23964" Elev 116m); and East Lombok Regency: Raksasa Cave in Tanjung Ringgit Village, Jerowaru District (S08°42.76' and E 116°39.56' Elev. 92m).

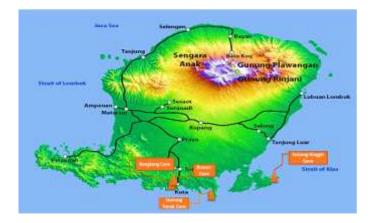


Figure 1. Map showing the sampling locality (Modification of Fajri et al, 2021)

Bats sampling

Bat species were collected using the trapping trap method with a mist net installed at the mouth of the four cave. The sampling was carried out when the bats came out of the cave at 18.30 – 19.00 WITA (Central Indonesian Time). Captured bats were removed from the net with a gloved hand) and transferred into a cotton cloth bag for transport to the nearby processing station for morphometric measurements and identification using morphological criteria by Suyanto et al (2002) [23] and Kitchener et al (2002) [24].

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Ectoparasites collecting techniques

Eonycteris spelaea species successfully collected were then examined, and samples of ectoparasites were collected manually from the bat's body was carried out sequentially starting from the head, ears, neck, wing bases, base of the feet, and toes using tweezers. In addition, the bats were carefully combed to collect additional samples of ectoparasites remaining in their fur. The ectoparasites collected were then placed in collecting tubes containing 95% alcohol. Before release, bats were marked on the head with a permanent marker to avoid resampling. All collected ectoparasites were subsequently sorted, identified, counted, mounted on microscope slide, and deposited for further study in the Laboratory Biology Center , National Research and Innovation Agency. Specimens were taxonomically identified using the taxonomic keys by Baker and Delfinado (1964) and Maa (1975).

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Results and Discussion

Eonycteris spelaea morphology

Eonycteris spelaea species were found in all caves chosen as research sites, namely Tanjung Ringgit Cave, Buwun Cave, Gale-gale Cave, and Gunung Tunak Cave. Based on the result of morphological measurements (Table 1), it is known that each individual bat found in each habitat has a unique characteristic regarding its size. The result of morphology analysis of 42 species captured shows that there are 2 groups that have similar measurement results. The first group consists of bats found in Gale-gale Cave and Buwun Cave, whereas the second group consists of bats found in Tanjung Ringgit Cave and Gunung Tunak Cave. **Commented [W112]:** Procedure identify the ectoparasite

 Table 1. Morphological measurements (mean and range) of adult and subadult

 Eonycteris spelaea captured in Lombok Island Caves in this study (research sites) (n =

 42) compared to other references.

Measurements	This Stud	ly				
Source	Tanjung Ringgit Cave	Gale-gale Cave	Buwun Cave	Gunung Tunak Cave	Suyanto	Kitchener
N	12	9	11	10	-	-
WT	67	78	65	66.9	-	66.6
HB	105	91.7	97.8	109.9	÷	110.5
Т	19.9	18.4	25.1	20.4	÷	16
Ε	20.5	20.5	20.6	19.8	16-22	17.9
ТВ	32.4	16.1	16.7	32.7	25-37	34
FA	65.9	78.8	74.9	67.5	60-81	72.4
HF	19.8	35.9	33.9	20.3	17-21	ł.

Description: n= Total Individu, WT=Weight (mm), HB=Head Body (mm), T= Tail, (mm) E= Ear (mm), TB= Tibia (mm), FA=Forearm (mm), HF= Hind Foot (mm)

The bats found in Gale-gale Cave and Buwun Cave have similar sizes of forearm which are 78.8 mm and 74.9 mm, respectively. Moreover, the sizes of their tibias are also similar, with the value of 16.1 mm and 16.7 mm. Meanwhile, the bats in Tanjung Ringgit Cave and Gunung Tunak Cave also show similar morphological sizes of the forearm (FA) which are 65.9 mm and 67.5 mm, and similar calf sizes (TB) with a length of 32.4 and 32.7 mm, respectively. Long and strong arms are very beneficial to support flying ability of this species. Long hind legs really help them to grip when nesting (Kamilah et al., 2021).

Ectoparasites Eonycteris spelaea

According to the result of the identification of ectoparasites collected from 42 species of *Eonycteris spelaea* in 4 caves in Lombok Island (Table 2), it is found that there are 7 types of ectoparasites consisting of 4 species from the class Insecta (2 species from the order Diptera, namely *Eucampsipoda sundaica* and *Cyclopodia horsfieldii* and 2 species from the order Siphonaptera namely *Ischnopsyllus hexactenus* and *Thaumapsylla breviceps*) and 3 species from the class Arachnida (*Amblyomma* sp., *Ancystropus* sp., and *Meristaspis* sp.).

Table 2. Ectoparasites of Eonycteris spelaea

	Number of Pa	rasites (Indiv	idual)	
Ectoparasites	Tanjung	Gale-gale	Buwun	Gunung
	Ringgit Cave	Cave	Cave	Tunak Cave
Insecta				
Diptera				
Nyteribiidae				
Eucampsipoda sundaica	1	1	-	2
Cyclopodia horsfieldii	2	3	2	3
Siphonaptera				
Ischnopsyllidae				

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Thaumapsylla breviceps	-	2	-	1	
Ischnopsyllus hexactenus	3	2	2	4	
Arachnida					
Ixodida					
Ixodidae					
Amblyomma sp.	12	9	17	19	Formatted: Font: Not Italic
Mesostigmata					
Spinturnicidae					
Ancystropus sp.	5	5	3	9	Formatted: Font: Not Italic
Meristaspis <u>sp.</u>	5	4	3	7	Formatted: Font: Not Italic

Eucampsipoda sundaica

Morphology

The result of observation shows that *Eucampsipoda sundaica* has a brown dorsal body, a flat body shape dorsoventrally, no wings, abdomen and legs covered with setae hair, claws on the tips of the feet, a head that can be folded towards the thorax, and femur and tibia nearly equal in length The eye has a single elliptical lens. Small head, pentagonshaped chest. Clasper short and tapered and hair on the lateral. The femur has a white ring and the tibia has two white rings [26].

Host

E. sundaica was found in bat species *E. spelaea* and *R. amplexicaudatus* [27][28]. In addition, *E. sundaica* has also been found infecting *Rousettus leschenaultia* and *Cherephon aplicata* [29]; [30]; [19]

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Distribution

E. sundaica has been reported infecting *Eonycteris spelaea* bats found in two places on the Lombok Island, namely in Tanjung Ringgit Cave and Gale-gale Cave. E. sundaica has also been found in E. spelaea bats in Gunung Reng, Jeli, Kelantan, Tioman Island Wildlife Reserve, Pahang, Wang Kelian State Park, Perlis [30], Yunnan Province of China at the Sino-Burmese border [29].

Cyclopodia horsfieldii

Morphology

Cyclopodia horsfieldii is frequently found in the genera *Cynopterus* and *Rousettus. C. horsfieldii* has a non-angled sterna plate with a curved tip. On the ventral side, white sutures diagonally across the abdomen are clearly visible. Thorn-like structures called ctenidia are clearly visible on thorax and abdomen. The head folds ventrally and can rotate 180°. White segmentation on the abdomen is clearly visible, and abdomen is also covered with setae [31].

Host

Cyclopodia horsfieldii is commonly found in bat from genus *Cynopterus*. However, its presence has also been reported in *Pteropus*, *Acerodon* and *Rousettus* Gray bats, new host records including *Cynopterus brachyotis* and Ptenochirusjari [32]. In addition, research conducted by Fajri et al [33] stated that *Cyclopodia horsfieldii* was found in bat species *Eonycteris spelaea*, *Rosettus amplexicaudatus* and *Miniopterus pusillus*.

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Distribution

Cyclopodia horsfieldii has been found in four locations on the Lombok island, namely the Gale-gale cave, Tanjung Ringgit cave, Buwun cave and Gunung Tunak cave. While in other places, Philippines including Luzon, Mindoro, Busuanga, Culion, Palawan, Balabac, Leyte, Iloilo, Panay, Guima-ras, Camiguin, Mindanao, Negros and Jolo [34][35][28]. In addition, this species has also been found around the mountains in Indonesia, Cambodia, Malaysia, Thailand and Timor Leste [28].

Thaumapsylla breviceps

Morphology

Thaumapsylla breviceps is a flea from the order Siphonaptera belonging to Ischnopsyllidae family. Ischnopsyllidae family is a family with the highest number of species infecting bats [36]. *T. breviceps* is classified into subfamily Thaumapsyllinae, the newest subfamily known as fleas that infect bats. *T. breviceps* has shorter head compared to all identified fleas and has a very short pronotum (separated from the ctenidium) [37].

Host

According to several studies of ectoparasites, it is known that *T. breviceps* has a narrow ectoparasite-host relationship. *T. breviceps* is only found in fruit-eater bats such as *Rousettus amplexicaudatus* [35] and *Eonycteris spelaea* [33].

Distribution

Three *Thaumapsylla breviceps* were found in this study. This ectoparasite is found on the body of *E. spelaea* in the Gale-gale cave and Gunung Tunak cave. Populations of *Thaumapsylla breviceps* have been found in southern Africa [38], southern China [39].

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In addition, its distribution is also widely found in Indonesia [33], Ethiopian region and Philippines-Mindoro [38]; [35].

Ischnopsyllus hexactenus

Morphology

Although bat lice belong to different families, they are generally similar to fleas that infect cats, dogs and other mammals. They are often seen as small oval shapes, move quickly through bat fur and are difficult to catch without a quick reaction. Like all fleas, only adults that become the hosts: egg and larval stages are found in the detritus on which bats roost.

Host

Ischnopsyllus hexactenus is one of the lice specifically found in bats, and is not found in other mammal species. Bats that have served as hosts for *I. hexactenus* are *Myotis* frater, Murina hilgendorfi, Pipistrellus pipistrellus and R. microphyllum [33]. *I. hexactenus* is an ectoparasite that can be a vector of several zoonotic pathogenic bacteria such as Trypanosomes. The abundance of *I. hexactenus* in bats is quite high, and *I. hexactenus* is even being able to make bats as the sole host.

Distribution

Eleven *Ischnopsyllus hexactenus* has been found in four locations on the Lombok island: the Gale-gale cave, Tanjung Ringgit cave, Buwun cave and Gunung Tunak cave. The distribution of *I. hexactenus* infesting bats has been reported in Indonesia, Malaysia, Latvia, and Mongolia [40], Siberian province, European province; Great Britain, Belgium, Netherlands, Germany, Svizzera Confederazione, Hungary, Romania and Czechia [41].

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Amblyomma sp.

Morphology

Ectoparasites from the class Arachnida consist of two orders, namely Ixodida and Mesostigmata. *Amblyomma* sp. belongs to the Ixodidae family. The Ixodidae family is also known as the hard tick because it has a clear capitulum characteristic. There is also scutum (chitinous dorsal board). According to the result of *Amblyomma* sp. observation, this ectoparasite has a black body and is still in the larval stage as proven by its only 3 pairs of legs. *Amblyoma* sp. has some special characteristics including mouth apparatus (palpus and chelicera) much longer than the base of the capituli, the palps longer than the chelicerae and the length of the palps twice the width [42]. *Amblyoma* sp. is a vector of Francisellatularensis, the agent of tularemia and *Ehrlichiach affeensis* causing monocyticehrlichiosis in humans. This tick also transmits *Rickettsia amblyommii*, Borrelialonestari, and Heartland viruses that cause disease in humans [43]. The order Mesostigmata has the largest number of parasitic species whose members are mostly blood suckers at the adult stage [42].

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Host

Amblyomma sp. is found in several bat species including Rousettus aegyptiacus [35], Eonycteris spelaea, Chaerephon plicata, and Taphozous melanopagon [33].

Distribution

Fifty-seven *Amblyomma* sp. has been found in four locations on the Lombok island: the Gale-gale cave, Tanjung Ringgit cave, Buwun cave and Gunung Tunak cave. Distribution of this species broad enough, among others in Caribbean and Africa [43], Brazil [44], Southwest Georgia and Northwest Florida, USA [45].

Ancystropus sp.

Morphology

Ancystropus sp. is a mite from the family Spinturnicidae which has very enlarged legs I and claws I. It usually has 2-3 pairs of setae surrounding the anterior border of the dorsal shield, slender distal setae on tarsus I, three pairs of setae surrounding the anterior border of the dorsal shield, epigynial shield located at the level of coxa IV, no lateral hooks on tarsus I and coxa I [46].

Host

The existence of *Ancystropus* sp. has been reported infesting several bats including *Rousettus leschenaulti* (Pteropodidae), *Rhinolophus rouxi* and *Hipposideros sporis* (Rhinolophidae). In addition, it has also been found in the species of *Eonycteris spelaea*.

Distribution

Most of *Ancystropus* sp_ is found on the wing membranes of bats. In this study, twenty two *Ancystropus* sp_ were found in the four observation sites on Lombok Island. The distribution of *Ancystropus* sp. infesting several bats has been found in some caves in Sri Lanka, Indonesia, Philippines, and Thailand.

Meristaspis sp.

Morphology

Meristaspis sp. is a species from the genus Meristaspis characterized by 4 pairs of setae surrounding the anterior border of the dorsal shield. Tarsus I has a pair of large distal

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setae, flat distal setae, no enlarged claws I; large tritosternum usually dilated, epiginal shield at coxa III-IV, anal shield which has no postanal seta, enlarged leg I, but no enlarged claw [46].

Host

Hosts from this species of ectoparasite are generally found in bats belonging to Megachiroptera including *Eonycteris spelaea*, *Macroglossus minimum*, *Rosettus amplxicaudatus* [33], and *Rousettus aegyptiacus* [47].

Distribution

Nineteen *Meristaspis* sp. has been found in four locations on the Lombok island: the Gale-gale cave, Tanjung Ringgit cave, Buwun cave and Gunung Tunak cave. This ectoparasite species distributes almost throughout Southeast Asia regions and some parts of American continent such as Mexico [47].

Commented [R20]: Add the measurement of the body of
this ectoparasite

Infected Number of **Number Bats** Prevalance **Ectoparasites species** (Per Bat's Intensity Examined Parasite (%) Tail) Insecta Diptera Nyteribiidae 10 40 1.00 Eucampsipoda sundaica 4 4 10 10 Cyclopodia horsfieldii 10 100 1.00 Siphonaptera Ischnopsyllidae Thaumapsylla breviceps 10 5 3 50 0.60 2 11 20 Ischnopsyllus hexactenus 10 5.50 Arachnida Ixodida Ixodidae Amblyomma sp 10 10 57 100 5.70 Mesostigmata Spinturnicidae 100 Ancystropus sp 10 10 22 2.20

Commented [R21]: It would be more complete if author add some informations such as photos of ectoparasites, prevalence data, and intensity

	• . •	
Me	eristaspis	SD

10

19

10

1.90

100

Conclusion

The result of this study shows that the *Eonycteris spelaea* species are distributed in all caves in Lombok Island. Based on the result of morphological measurements, there are differences in morphology of *E. spelaea* species found in each location. *E. spelaea* are infested by seven ectoparasites consisting of 4 species from the class Insecta which are 2 species from the order Diptera (*Eucampsipoda sundaica* and *Cyclopodia horsfieldii*) and 2 species from the order Siphonaptera (*Ischnopsyllus hexactenus* and Thaumapsylla); and 3 species from the class Arachnida (*Amblyomma* sp., *Ancystropus* sp., and *Meristaspis* sp.). It is also addressed that there are several ectoparasites that are quite dangerous for humans and other animals found infecting *E. spelaea* These ectoparasites can cause several types of diseases. Thus, it is necessary to carry out special care and to give attention to this species of bat.

Acknowledgements

We would like to thank the Ministry of Education, Culture, Research and Technology, the Directorate General of Higher Education, Research and Technology for fully funding this research. We also thank the foundation, rector and LPPM of Mandalika University of Education for facilitating us so that we could carry out and complete this research.

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EVALUATION FORM of manuscript submitted for publication to Annals of Parasitology

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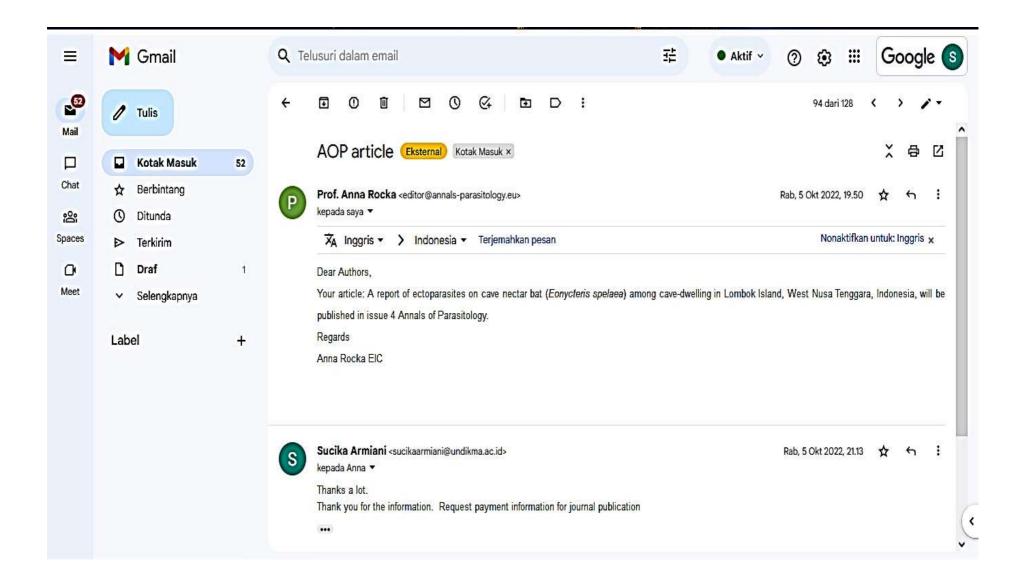
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ACCEPTANCE LETTER

I have a pleasure to inform that the article: "A report of ectoparasites on cave nectar bat (*Eonycteris spelaea*) among cave-dwelling in Lombok Island, West Nusa Tenggara, Indonesia"; authors: Siti Rabiatul FAJRI, Sucika ARMIANI, Akhmad SUKRI, Ibnu MARYANTO, will be published in journal *Annals of Parasitology* number 4 vol. 68/2022.

With kind regards,

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