Original Research Article

Diversity and abundance of chiropteran ectoparasites in Abidjan district, Côte d'Ivoire

.

ABSTRACT

Aims: The aim of this study is to produce information on the diversity and abundance of ectoparasites in bats in Côte d'Ivoire.

Place and Duration of Study: It was carried out in 2021 in the Abidjan district, in the forest relics of the Zoo National Abidjan (ZNA) and the Institut Pasteur de Côte d'Ivoire (IPCI) Adiopodoumé site.

Methodology: The first step was to capture and identify the bats, then for each bat collected, ectoparasites were collected and identified using the Walker and Ramel keys.

Results: In the course of this study, 89 bats were collected, most of them adult (91%) and female (50.6%). Most belonged to the genera *Micropteropus* (41.6%) and Eidolon (40.4%). Ectoparasites were collected at a rate of 20.22% (+/- 0.187), and identification revealed *Nycteribiabiarticulata* (69.7%) and *Ornithodoroserraticus* (30.3%), belonging to the insect and tick classes respectively. A causal link was established in the univariate analysis (p<5%) between the presence of the parasite and the bat species collected.

Conclusion: These results provide non-exhaustive information on the diversity and abundance of bat ectoparasites in the Abidjan district.

This research needs to be extended to the whole of Côte d'Ivoire, in order to identify the ectoparasites that are potential vectors of zoonoses transmitted by ectoparasites in a One Health concept.

Keywords: Chiroptera - External parasite - Zoonosis - Surveillance

1. INTRODUCTION

~~The order Chiroptera is the most diverse order of placental mammals after Rodentia, with nearly 1,400 species quarter (¼) of all mammal species known at the time of writing (1;2;3).~~

~~Many studies have highlighted them as the predominant host of both internal and external parasites (4; 5; 6; 7).~~

~~External parasites (ectoparasites) are small organisms that live and feed on the integument of various hosts, s which can colonise the host's body cavities (8). Some of these ectoparasites are potential vectors of disease in ma particularly chiropterans and rodents (9; 10). In a large part of the world, knowledge of bat ectoparasites is still limited. Most ectoparasites share co-evolutionary links with their hosts, as many of them remain on their hosts for most of their lives, while others are only present during the various reproductive stages of the host, such as gestation or lactation (11). The diversity of bat habitats makes them natural hosts for ectoparasites and therefore excellent models for studying host-parasite relationships, given their extremely diverse taxonomy and behaviour (12).~~ In Côte d'Ivoire, stud chiropterans have provided some insight into the diversity of this fauna, which currently accounts for 24% of the c known mammals (13; 14; 15; 16). However, in Côte d'Ivoire, very few documented studies have reported on th parasite relationship that exists between chiropterans and the parasites that live in their environments. The aim study, which is the first of this initiative, is to produce information on the diversity and abundance of ectoparasites in the Abidjan district. ~~For this study, we tested the following hypotheses: 1) bats in the Abidjan district~~

Comment [RBS1]: You are talking about only two species found, among more than 15 different families that can parasitize bats. The title should correspond to the results.

Comment [RBS2]: Missing italics

Comment [RBS3]: This is also not the class of insects. The correct name is Insecta

Comment [RBS4]: Tick is not a class, it is the common name for mites of the order Ixodida, the class is Arachnida. Serious errors of taxonomy should not be accepted.

Comment [RBS5]: What does this mean? You found only two new records. What is the total ectoparasitofauna associated with bats of Côte d'Ivoire?

Comment [RBS6]: All places we need this.

Comment [RBS7]: Use the correct term in Parasitology to name this group of parasites.

Comment [RBS8]: At no time was there any monitoring of pathogens or anything like that carried out. What do these two keywords have to do with the work?

Comment [RBS9]: Why does the text have short, loose sentences? The introduction needs to be rethought. Crucial information about the topic of the work is not being considered, such as:What is the current diversity of bat ectoparasites in Africa, or in the country under study? Have the species found already been recorded in the country?

Comment [RBS10]: Your introduction starts here. Avoid stating the obvious. This is a scientific article, not a book.

Comment [RBS11]: What does this sentence mean? 24% of the country's bat species were found parasitizing bats? That's it. It needs to be clear what the authors mean.

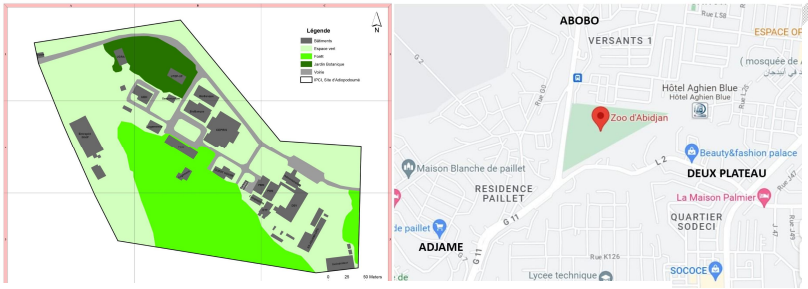
Comment [RBS12]: Two species only, not all.

~~ectoparasites; 2) the diversity and abundance of ectoparasites influenced by the bat species; 3) the diversity and abundance of ectoparasites influenced by the bat collection area.~~

2. MATERIAL AND METHODS

2.1 Study area and period

This work was carried out in Côte d'Ivoire by setting up net traps on two sites, namely the wooded perimeter around the site of the Institut Pasteur de Côte d'Ivoire (Adiopodoume site) WGS84 (Latitude: 5.31874808; Longitude: -4.13539601) and the Abidjan National Zoo WGS84 (Latitude: 5.380995; Longitude: -4.004645) from 22 December 2020 to 28 January 2021. Located in Adiopodoumé, the 17-hectare Adiopodoume site is made up of a degraded secondary forest that was used to build the institute's infrastructure facilities. The Abidjan National Zoo, currently estimated at around 18 hectares, is located in the commune of Cocody, precisely at the intersection of the roads linking the communes of Abobo and Adjamé to the Plateau-Dokui and Deux Plateau districts. It is made up of secondary forest that is in a very advanced state of degradation. It is home to many free-living animals (bats and some birds) and animals in captivity (17).



A) Institut Pasteur of Côte d'Ivoire, Adiopodoumé site **B) Abidjan National Zoo, limits and situation**

Figure 1: Administrative map of study area

2.2. Capturing and identifying bats

At the Institut Pasteur and Zoo National d'Abidjan, trapping carried out continuously throughout the study. Three mist nets at 4 levels and of different sizes (12, 9 and 6 ~~metres~~meters), each 3 ~~metres~~meters high and 12 ~~metres~~meters long, were set up in the undergrowth of each site using bamboo fixed in the ground and ropes to hold them in place. Set up from 6.30 in the evening and taken down before 6.30 in the morning, the traps visited every 30 minutes to remove the bats so that they do not injure themselves by becoming entangled in the mesh of the nets. They are placed in individual cotton bags and hung from a branch or rope provided for this purpose. The animals were then taken to the Animal Resource Management Unit (UGRA) of the Institut Pasteur de Côte d'Ivoire (IPCI) at the Adiopodoumé site for identification of the bat species and ectoparasites. To identify the bats, information such as weight, sex, age, physiological state and, using a ruler, the various measurements (forearms, fingers, head, body and tail) of each bat were determined according to a determination key described by Kadjo~~and Coll.~~et al. (16).

2.3. Collecting ectoparasites

After each capture, the bats were taken back to the Animal Resource Management Unit (UGRA) of the Institut Pasteur de Côte d'Ivoire (IPCI) at the Adiopodoumé site. Once at the Unit, some of the animals underwent flash ~~anaesthesia~~anesthesia with Isoflurane, while others were directly inspected in their entirety. Animal inspection and ectoparasite collection were carried out under SHPII in the UGRA laboratory, in compliance with biosafety measures. Ectoparasites were collected using blunt-tipped forceps, sometimes aided by a binocular magnifying glass. The parasites collected were then immersed in jars containing 70° alcohol (11). Each jar was then identified with the animal's ID number and stored at room temperature pending identification of the parasites collected. For each bat sampled, the examiner recorded the number of parasites collected. Once the ectoparasites had been collected, the bats were each placed in transport bags containing fruit and then transported to their capture areas to be released.

2.4 Identification of ectoparasites

Comment [RBS13]: No need.

Comment [RBS14]: Please provide geographic coordinates in degrees, common in scientific works.

Comment [RBS15]: Who is A, and who is B? There is no scale on the maps, much less any direction. The maps need to be self-explanatory. The way it was made is totally wrong. Which part of the Côte d'Ivoire is this?

Comment [RBS16]: How can a PhD thesis be used as a basis for host identification? Is there no work that has pictorial keys, identification keys for bats in the country or at least in Africa? PhD theses should never be used for such purposes. Nor should they be cited.

Comment [RBS17]: I do not think that ectoparasite collections began in 2011, so a reference to this date should not be used to explain how the ectoparasites were collected. Please review this.

The ectoparasites were then identified in the laboratory of the Institut Pasteur's Entomology and Herpetology Unit (UEH). They were identified using an OPTIKA binocular magnifier and dichotomous identification keys. ~~The insects were first separated from the ticks.~~ The identification key of Walker ~~and coll.~~ et al.(18) was used for tick identification and that of Ramel(19) for insect identification. Specimens were sorted according to their stage of development and then identified by genus and species.

2.5. Data analysis

After data management of the database using MS Excel. The actual analysis was carried out using R software version 4.2.1 (MacOS) on the Rstudio 2022.07.1 editor. It consisted firstly of a univariate analysis known as the descriptive method for calculating means, standard deviations and confidence intervals, and secondly of a multivariate analysis for calculating the p-value and the Chi2 test.

3. RESULTS AND DISCUSSION

3.1. Results

3.1.1. Identification of bats

~~A total of 89~~Eighty-nine bats were collected at the two sites, i.e. ~~The~~the wooded area around the InstitutPasteur site at Adiopodoumé and the forest at Zoo National Abidjan.

3.1.1.1. Distribution of bats according to species

Among the 89 observations, the majority were species belonging to the genus *Micropteropus* with 37 individuals, i.e. 41.6%, followed by the species *Eidolon helvum* with 36 individuals, i.e. 40.4%.

TABLE I: DISTRIBUTION OF COLLECTIONS ACCORDING TO SPECIES (N=89)

Species	absolute Fr~~é~~equencies (n)	relative Frequencies (%)
Eidolon helvum	36	40,4
Hypsignathus monstrosus	12	13,5
*Micropteropus*sp.	37	41,6
Pipistrellus sp.	4	4,5

3.1.1.2. Distribution of bats according to sex and age

Of the 89 bats collected, 45 (50.6%) were females and adults represented the majority with 81 observations (91%) (Table II).

TABLE II: breakdown of observations by sex (n=89)

Sex	absolute ~~Fréquencies~~Frequencies (n)	relative Frequencies (%)
~~Femal~~Female	45	50,6
Male	44	49,4

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Comment [RBS19]: What reference is this? Please fix it.

Comment [RBS20]: Which ones? Why is it not described anywhere what the stages of ectoparasites are?

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3.1.2. Diversity and abundance of ectoparasites in relation to collection areas and bat species.

3.1.2.1. Prevalence of ectoparasites as a function of bat capture areas

Among the 89 bats, 20.22% [20.13-20.30] carried ectoparasites for the entire study. No ectoparasites were found on the bats captured at the Adiopodoumé site.Two ectoparasites were identified, namely insects of the species *Nycteribiabiarticulata* (~~hermann~~Hermann, 1804) (n=39), i.e. 69.7%, and ticks of the species *Ornithodoroserraticus* (n=17), representing 30.3% of ectoparasites (Table III).

TABLE III: Diversity and abundance of ectoparasites according to the areas where bat species were captured.

Group	Family	Gender	Taxons	Study area			
				Forêt IPCI	Zoo National	Cumulative number	relativ
Insect	*Nycteribiidae*	*Nycteribia*	*N. biarticulata*	0	39	39	69,7
Mite	*Argasidae*	*Ornithodoros*	*O. erraticus*	0	17	17	30,3
Total			**2**	**0**	**56**	**56**	**100**

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Bivariate analysis showed that there was no statistically significant link (p= 0.074) between the presence of ectoparasite species and the collection site (Table IV).

Table IV: Distribution of ectoparasites according to collection area (N=89)

		PRESENCE OF ECTOPARASITES		
		YES **N *(%)***	**NO** **N *(%)***	**TOTAL**
SITE	**FORET IPCI**	**0 *(0,0)***	**15 *(21,1)***	**15**
	ZNA	**18 *(100,0)***	***56 (78,9)***	**74**
TOTAL		**18 *(100,0)***	**71 *(100,0)***	**89**

3.1.2.2. Prevalence of ectoparasites according to bat species

However, there was a statistically significant link (P< 0.001) between the presence of parasites and the bat species collected (Table V).

Table V. Distribution of ectoparasites by species (N=89)

		PRESENCE OF ECTOPARASITES		
		YES **N *(%)***	**NO** **N *(%)***	**TOTAL**
GENDER AND SPECIES	*Eidolon helvum*	***17 (94,4)***	**19 *(26,8)***	**36**
	Hypsignathus monstrosus	***0 (0,0)***	***12 (16,9)***	**12**
	Micropteropus	***1 (5,6)***	***36 (50,7)***	**37**
	Pipistrellus	***0 (0,0)***	***4 (5,6)***	**4**
TOTAL		**18 *(100,0)***	**71 *(100,0)***	**89**

3.1.2.3. Ectoparasitespecies present on bat species

Both types of ectoparasite (tick and insect) were collected on the bat species *Eidolon helvum*, but only one insect was collected on the bat of the genus *Micropteropus* (Table VI).

Table VI. Matrix of bat species and their parasites

SPECES OF BATS	NOMBER	FREQUENCE (%)	PARASITES
EIDOLON HELVUM	**55**	**98,21**	***Nycteribiabiarticulata***
			Ornithodoros. erraticus

Comment [RBS22]: The most crucial information was not included in the results: how were these ecoparasites identified? What characteristics separate them from the other species in each group? Beside that, No images of the ectoparasites were taken, in which collection were these ectoparasites deposited, so that if someone in the future wants to work with them, they can consult them?

MICROPTEROPUSSP	**1**	**1,79**	***Nycteribiabiarticulata***

3.2. DISCUSSION

In this study, four genera of Chiropterawere identified. These are the genera *Eidolon, Hypsignathus, Micropteropus* and *Pipistrellus*. Studies carried out in several regions of Côte d'Ivoire have identified species belonging to these genera and several other Chiroptera genera (14; 20; 16).The genera *Micropteropus* and *Eidolon* were the majority. The two species identified in this study, *Eidolon helvum* and *Hypsignathusmonstrosus*, are megachiropters. A study carried out by Niamien~~and Coll~~et al.(21)on the population of hammerhead bats, *Hypsignathusmonstrosus* in the plateau commune of Abidjan showed that this species has a marked preference for Terminalia catappa l. (Combretaceae) breeding sites and that numbers varied according to tree species and season.
Eidolon helvum was the majority species, yet this species has conservation status, as it is classified as vulnerable in certain regions. And according to Ricketts~~and Coll.~~et al. (22), among mammals, chiropterans constitute a group of threatened species, some of which have seen their numbers decline at a particularly alarming rate

~~In the second species identified in this study, *Hypsignathus monstrosus*, there is a very pronounced sexual dimorphism between males and females (23). In addition to being larger, these males have a monstrous face and a wingspan exceeding one metre.~~
Several studies have shown that bats are infected by a wide variety of endoparasites and ectoparasites, and up to 1999, around 756 taxa were associated with bats worldwide (24). In this study, two species of ectoparasites were identified, namely insects of the species *Nycteribiabiarticulata* and ticks of the species *Ornithodoroserraticus*. The data collected demonstrate the presence of ectoparasites in the district's bats, and analysis of the data shows that the diversity and abundance of ectoparasites are influenced by bat species p< 0, 001. ~~These results corroborate some of the hypotheses put forward prior to this study.~~
According to Parola and Raoult(25), ticks of the genus *Ornithodoros*are known to be vectors of relapsing fever, which is an infection caused by certain species of *Borrelia* in humans. And in birds, ticks of the genus *Ornithodoros* are known to be reservoirs of medically important pathogenic bacteria found worldwide(26). In Algeria, the species *Ornithodoroserraticus*has been found in rodent burrows in the east of the country (27).
In Côte d'Ivoire, knowledge of soft ticks (Ixodida: Argasidae) is incomplete. Further studies are needed to determine the specific composition of Argasidae and the diseases they could transmit. Precise knowledge of the distribution of these ticks and their monitoring are vital for defining areas at risk of tick-borne diseases and for establishing appropriate tick control and prevention measures. Against this backdrop, ongoing tick monitoring is an ongoing need in our regions.The insect species *Nycteribiabiarticulata* identified in this study was also collected from a bat species in ~~latvia~~Latvia(28). Several species of *Nycteribia* in the Nycteribiidae family have been identified in bats in several studies (29; 30; 31).
A number of emerging diseases are transmitted by haematophagousectoparasitic arthropods. These diseases are unique in that the diversity and complexity of the epidemiological cycles of the pathogens responsible complicate their study and monitoring. ~~In addition, current climatic changes and the transformation of landscapes caused by mankind are creating new environments and changing the existing dynamics within certain ecosystems, sometimes creating favourable conditions for the establishment of new outbreaks of disease, and therefore for their emergence.~~ It is therefore important to understand these ectoparasites in order to better combat the diseases they could transmit.

4. CONCLUSION

In Côte d'Ivoire, studies have identified numerous bat species in different biotopes, as well as ectoparasites in the environment and on animals. However, none of these studies had focused on bat ectoparasites. This study, which is one of the first of its kind, highlights the presence of insects and the ~~haematophagous~~hematophagous tick *Nycteribiabiarticulata*and *Ornithodoroserraticus* on bats at two sites: the wooded area around the Institut Pasteur site at Adiopodoumé and the forest at Zoo National Abidjan. The vast majority of these ectoparasites were identified on the *Eidolon helvum*, a species classified as vulnerable in other countries, which nests in the business ~~centre~~center of Côte d'Ivoire's economic capital. The *Ornithodoroserraticus* tick has also been identified as a vector of animal and human diseases. Côte d'Ivoire therefore has a strong interest in stepping up its research into the diversity of ectoparasites on wildlife in order to understand their diversity, but also to step up surveillance of the diseases that these ectoparasites could transmit.

Comment [RBS23]: Could this have been cross-contamination? Have you thought about that?

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Comment [RBS25]: This is introduction.

Comment [RBS26]: In this study or in literature¿

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Comment [RBS27]: Does this discuss anything about your results? Is the work about bats or about ectoparasites?

Comment [RBS28]: If the work is up to 2018, why are these numbers only up to 1999? Old and out of date.

Comment [RBS29]: Puta de correct names, diptera.

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Comment [RBS31]: Diversity of two species¿

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Comment [RBS32]: What is the meaning of these three separate sentences, with the results obtained in the present work?

Comment [RBS33]: According to whom¿

Comment [RBS34]: But what is this composition? Is there no soft tick described for the country?

Comment [RBS35]: How can you explain that a diptera that was identified in Latvia has now been detected in Côte d'Ivoire? Isn't this a very widespread distribution of an ectoparasite associated with bats? Were they detected in the same species of bat?

Comment [RBS36]: Again, a loose sentence, which means nothing, with no reference and no connection to the text.

Comment [RBS37]: The work was entirely focused on geographic distribution, this sentence has no meaning and much less importance for the result of the work.

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Comment [RBS39]: Every tick of this genus is hematophagous.

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Comment [RBS41]: This is not a conclusion drawn from the results obtained in the present work.

Comment [RBS42]: Thi is dicussion. Not your conclusion.

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DEFINITIONS, ACRONYMS, ABBREVIATIONS

IPCI: Institut Pasteur of Côte d'Ivoire
ZNA: Zoo National of Abidjan
UGRA: Unit of Animal Ressources Management
UEH: Unit of Enthomology and Herpetology