

## Original Research Article

### Presence and abundance of chiropteran ectoparasites of Zoonotic importance 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 from December 22, 2020, to January 28, 2021, in the Abidjan district, within the forest relics of the Abidjan National Zoo (ZNA) and the Adiopodoumé site of the Institut Pasteur de Côte d'Ivoire (IPCI)

**Methodology:** The first step was to capture the bats using mist nets and identify them with the help of research assistants. For each bat captured, 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 Insecta and Arachnida 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 belonging to the genera *Ornithodoros* and *Nycteribia*. These ectoparasites are vectors of zoonoses such as intermittent fever due to *Borellia* and vectors of animal diseases such as African swine fever. This research needs to be extended to the entire Côte d'Ivoire in order to identify ectoparasite vectors and gain a better understanding of local biodiversity and zoonotic risks (the transmission of diseases from animals to humans).

**Keywords:** Chiroptera - Ectoparasite - Zoonosis - Surveillance

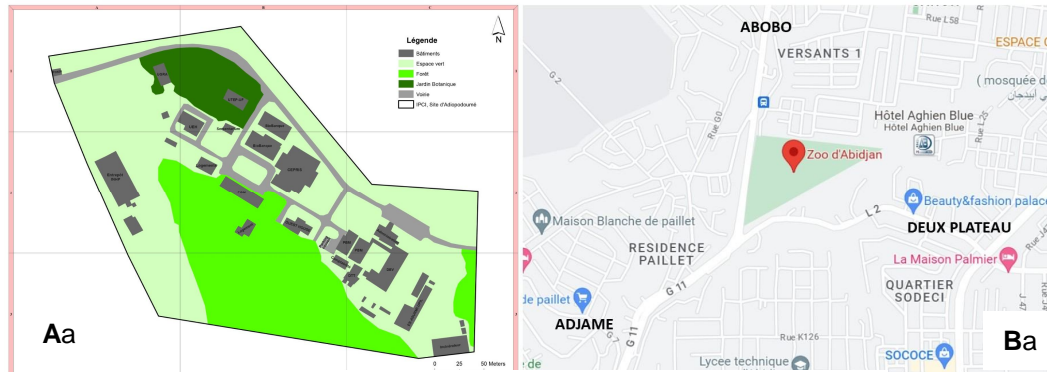
## 1. INTRODUCTION

In Côte d'Ivoire, studies on chiropterans have led to a better understanding of the diversity of this fauna. To date, there are 75 species belonging to 7 (seven) different families, including the Pteroptidae (*Hypsignathusmonstrosus*, *Eidolon helvum*, *Micropteropus* sp.), Amballonarudae, Nycteridae, Rhinilophidae, Hipposideridae, Vespertilionidae (*Pipistrellus* sp...) and Molossidae (13; 14). Studies have also been conducted in Côte d'Ivoire on ectoparasites and the relationship between ticks and the diseases they can transmit to both animals and humans. These studies revealed 6 (six) species, all belonging to the Arachnida class (*Amblyommavariegatum*, *Rhipicepalusmicroplus*, *Rhipicephalus senegalensis*, *Hyalomma truncatum*, *Hyalomma marginatum rufipes* and *Hyalomma impressum*). These studies have mainly focused on animals (ruminants, chicken, and swine) but very rarely on bats. The aim of our study is to produce information on the diversity and abundance of ectoparasites in bats and their related pathogens in the Abidjan district.

## 2. MATERIAL AND METHODS

### 2.1 Study area and period

This work was conducted in Côte d'Ivoire by setting up net traps at two sites, namely the wooded perimeter around the Institut Pasteur de Côte d'Ivoire (Adiopodoumé site) WGS84 (Latitude: 5° 19' 7.49" N; Longitude: -4° 8' 7.43" W) and the Abidjan National Zoo WGS84 (Latitude: 5° 22' 51.58" N; Longitude: 4° 0' 16.72" W) from December 22, 2020, to January 28, 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 meters), each 3 meters high and 12 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 (Fig. 1). 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 Webalaand al. (16).



Fig 1. Capture and identification of bats (Mist nets break; B. Capture of Bats; C and D. Identification of bats)

### 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 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

The ectoparasites were then identified in the laboratory of the Institut Pasteur's Entomology and Herpetology Unit (UEH) (Fig 2). They were identified using an binocular magnifier (OPTIKA) and dichotomous identification keys. The identification key of Walker et al. (18) was used for tick identification and that of Ramel for insect identification (19). The specimens were sorted according to their class, then identified by genus and species.



Fig 2. Parasite identification

### 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 Chi<sup>2</sup> test.

### 3. RESULTS AND DISCUSSION

#### 3.1. Results

##### 3.1.1. Identification of bats

Eighty-nine bats were collected at the two sites, i.e. the wooded area around the Institut Pasteur 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 1: DISTRIBUTION OF COLLECTIONS ACCORDING TO SPECIES (N=89)**

Species	absolute Frequencies (n)	relative Frequencies (%)
<i>Eidolon helvum</i>	36	40,4
<i>Hypsignathus monstrosus</i>	12	13,5
<i>Micropteropus</i> sp.	37	41,6
<i>Pipistrellus</i> 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 2).

**TABLE 2: breakdown of observations by sex (n=89)**

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

##### 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, 1804) (n=39), i.e. 69.7%, and ticks of the species *Ornithodoros erraticus* (n=17), representing 30.3% of ectoparasites (Table 3).

**TABLE 3: Diversity and abundance of ectoparasites according to the areas where bat species were captured.**

Group	Family	Genus	Taxons	Study area			relative Frequencies
				IPC forest	Zoo National	Cumulative number	
Insect Mite	<i>Nycteribiidae</i>	<i>Nycteribia</i>	<i>N. biarticulata</i>	0	39	39	69,7
	<i>Argasidae</i>	<i>Ornithodoros</i>	<i>O. erraticus</i>	0	17	17	30,3
<b>Total</b>			<b>2</b>	<b>0</b>	<b>56</b>	<b>56</b>	<b>100</b>

Bivariate analysis chi-square, showed that there was no statistically significant link ( $p= 0.074$ ) between the presence of ectoparasite species and the collection site (Table 4).

Table 4: Distribution of ectoparasites according to collection area (N=89)

		PRESENCE OF ECTOPARASITES		
		YES N (%)	NO N (%)	TOTAL
SITE	IPCI FOREST	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 5).

Table 5. Distribution of ectoparasites by species (N=89)

		PRESENCE OF ECTOPARASITES		
		YES N (%)	NO N (%)	TOTAL
GENUS AND SPECIES	<i>Eidolonhelvum</i>	17 (94,4)	19 (26,8)	36
	<i>Hypsignathusmonstrosus</i>	0 (0,0)	12 (16,9)	12
	<i>Micropteropus</i> sp	1 (5,6)	36 (50,7)	37
	<i>Pipistrellus</i> sp	0 (0,0)	4 (5,6)	4
TOTAL		18 (100,0)	71 (100,0)	89

### 3.1.2.3. Ectoparasite species present on bat species

Both types of ectoparasite (Arachnida and Insecta) were collected on the bat species *Eidolon helvum*, but only one insect was collected on the bat of the genus *Micropteropus* (Table 6).

Table 6. Matrix of bat species and their parasites

SPECES OF BATS	NUMBER	FREQUENCY (%)	PARASITES
<i>EIDOLONHEL VUM</i>	55	98,21	<i>Nycteribiabiarticulata</i>  <i>Ornithodoroserraticus</i>
<i>MICROPTEROPUSSP</i>	1	1,79	<i>Nycteribiabiarticulata</i>

### 3.2. DISCUSSION

In this study, four genera of Chiroptera were 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). The genera *Micropteropus* (n=37) and *Eidolon* (n=36) were the majority of bats found. The two species identified in this study, *Eidolon helvum* and *Hypsignathus monstrosus*, are megachiropters. A study carried out by Niamienet al. (21) on the population of hammerhead bats, *Hypsignathus monstrosus* 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 Rickettset al. (22), among mammals, chiropterans constitute a group of threatened species, some of which have seen their numbers decline at a particularly alarming rate

Several studies have shown that bats are infected by a wide variety of endoparasites and ectoparasites. Several studies have shown that bats are infected by a wide variety of endoparasites and ectoparasites, and to date many groups of ectoparasites, such as bat flies (Diptera), bugs (Hemiptera), fleas (Siphonoptera) and several arachnids such as fleas (Mesostigmata) and ticks including *Ornithodoros spp.* (24, 25, 26). In this study, two species of ectoparasites were identified, namely insects of the species *Nycteribiabiarticulata* (Speiser 1900) and ticks of the species *Ornithodoroserraticus* (Lucas 1849). 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  $p < 0, 001$ . According to Parola and Raoult (27), 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 Ticks of the genus *Ornithodoros* are known to be reservoirs of medically important pathogenic bacteria found throughout the world in birds, as well as the African swine fever virus. (28; 29). In Algeria, the species *Ornithodoroserraticus* has been found in rodent burrows in the east of the country (30).

In Côte d'Ivoire, knowledge of soft ticks is incomplete and less extensive than that of hard ticks (Ixodidae), given their veterinary and public health importance (31; 32). 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 different from the one discovered in this study (33). This could be explained by the fact that this ectoparasite species has a host diversity. Several species of *Nycteribia* in the Nycteribiidae family have been identified in bats in several studies (34; 35; 36).

These diseases are unique in that the diversity and complexity of the epidemiological cycles of the pathogens responsible complicate their study and monitoring. 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.

#### 4. CONCLUSION

In Côte d'Ivoire, studies have identified numerous bat species in different biotopes. 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 insecta and arachnida respectively *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 center of Côte d'Ivoire's economic capital. Many studies have shown the role of these two types of ectoparasite (Insecta and Arachnida) in the transmission of both human and animal pathogens such as *Bartonella*, *Borellia* and African swine fever virus.

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## REFERENCES

1. Don E. Wilson et DeeAnn M. Reeder. Mammal Species of the World: A Taxonomic and Geographic Reference. JHU Press. Vol 1. 2005
2. WILSON, D. E. Bats in question: the Smithsonian answer book. Smithsonian Institution, 2015. (ISBN 978-1-58834-511-0, lire en ligne [archive])
3. MORATELLI, R. and CALISHER, C. H. Bats and zoonotic viruses: can we confidently link bats with emerging deadly viruses?. *Memórias do Instituto Oswaldo Cruz*. 2015 ; vol. 110, p. 1-22. (ISSN 0074-0276, DOI 10.1590/0074-02760150048, lire en ligne [archive],
4. Raharimanga, V., Goodman, S. M., & Robert, V. (2003). Les ectoparasites des chauves-souris malgaches : Inventaire et implications écologiques. *Revue de Parasitologie*, 25(3), 125-138
5. Dick CW, Dittmar K. Parasitic bat flies (Diptera: Streblidae and Nycteribiidae): Host specificity and potential as vectors, in *Bats (Chiroptera) as Vectors of Diseases and Parasites*. Klimpel S, Mehlhorn H, Editors. Springer: Berlin. 2014 ; 187 p. (pp. 131–155).
6. Bendjeddou, M. L., Loumassine, H. A., Scheffler, I., Bouslama, Z., & Amr, Z. (2017). Bat ectoparasites (Nycteribiidae, Streblidae, Siphonaptera, Heteroptera, Mesostigmata, Argasidae, and Ixodidae) from Algeria. *Journal of Vector Ecology*, 42(1), 13-23. <https://doi.org/10.1111/jvec.12235>
7. Bendjoudi, D., Yedou, W., Beneldjouzi, A., et al. (2019). Ectoparasites des chauves-souris (Nycteribiidae, Streblidae, Siphonaptera, Mesostigmata et Ixodidae) du Parc National de Chréa (Monts de l'Atlas Central), Algérie. *Bulletin de la Société Zoologique de France*, 2, 67-76
8. Kkjh
9. Hopla, C. E., Durden, L. A. et Keirans, J. E. (1994). Ectoparasites et classification. *Revue scientifique et technique-Office international des épizooties*, 13(4), 985-1034.
10. Ritzi C. M., and Whitaker, J. O. Jr., 2003. Ectoparasites de petits mammifères du dépôt chimique de Newport, comté de Vermillion, Indiana. *Naturaliste du Nord-Est* ; Vol. 10, n° 2 , p. 149-158
11. Marquina, D., Buczek, M., Ronquist, F., & Łukasik, P., 2021. The effect of ethanol concentration on the morphological and molecular preservation of insects for biodiversity studies. *PeerJ*, 9, e10799. DOI 10.7717/peerj.10799
12. Kurta, A., Whitaker, J.O., Jr., Wrenn, W. & Soto-centeno, A., 2007. Ectoparasitic assemblages on mormoopid bats (*Chiroptera: Mormoopidae*) from Puerto Rico. *Journal of Medical Entomology*, 44, 953-958
13. Brosset, A., 1985. Chiroptères d'altitude du Mont Nimba (Guinée). Description d'une espèce nouvelle, *Hipposideros lamottei*. *Mammalia*; 48 (4): 545-555.
14. Fahr, J. and Kalko, E. K.V., 2010. Biome transitions as centres of diversity: habitats heterogeneity and diversity patterns of West African bat assemblages across spatial scales. *Ecography*; 33: 1-19.
15. Ehounoud, C. B., Yao, K. P., Dahmani, M., Achi, Y. L., Amanzougaghene, N., Kacou N'Douba, A., ... & Mediannikov, O. (2016). Multiple pathogens including potential new species in tick vectors in Côte d'Ivoire. *PLoS neglected tropical diseases*, 10(1), e0004367.
16. Webala, P. W., Oguge, N. O., & Bekele, A., 2004. Bat species diversity and distribution in three vegetation communities of Meru National Park, Kenya. *African Journal of Ecology*, 42(3), 171-179..

17. Oyetola W. D., 2015. Gestion technico-economique et sanitaire du zoo nationld'abidjan : etat des lieux, defis et perspectives. *These de medecineveterinaire*. EISMV dakar Senegal.
18. Walker, A. R., 2003. *Ticks of domestic animals in Africa: a guide to identification of species* (Vol. 74). Edinburgh: Bioscience Reports.
19. Ramel Alain 2021 Les Insectes, aramel.free.fr/INSECTES44.shtml, consulté en ligne le 30/06/2021
20. Denys, C., Monadjem, A., & Richards, L. 2016. An African Bat Hotspot: The Exceptional Importance of Mount Nimba for Bat Diversity. *ActaChiropterologica*, 18(2), 359-375. <https://doi.org/10.3161/15081109ACC2016.18.2.005>
21. Niamien, C. J. M., Kadjo, B., Kone, I., & N'goran, K. E., 2015. Données préliminaires sur la distribution spatio-temporelle des chauves-souris à tête de marteau, *Hypsignathusmonstrosus* H. Allen, 1861 dans la commune du Plateau (Abidjan, Côte d'Ivoire). *Afrique Science: Revue Internationale des Sciences et Technologie*, 11(1), 227-236.
22. Ricketts, T.H., Dinerstein, E., Boucher, and al., 2005. Pinpointing and preventing imminent extinctions. *Proc. Nat. Acad. Sci. USA* 102(51): 18497-18501
23. Paul Langevin and Robert M. R. Barclay., 1990. *Hypsignathusmonstrosus*. MAMMALIAN SPECIES No. 357, pp. 1-4, 4 figs
24. Léger, C., 2020. Bat parasites (Acari, Anoplura, Cestoda, Diptera, Hemiptera, Nematoda, Siphonaptera, Trematoda) in France (1762–2018): a literature review and contribution to a checklist. *Parasite*, 27.
25. Sonenshine DE, Anastos G. Observations on the life history of the bat tick *Ornithodoroskelleyi* (Acarina: Argasidae). *J Parasitol.* (1960) 46:449–54.
26. Szentiványi T, Estók P, Földvári M. Checklist of host associations of European bat flies (Diptera: Nycteribiidae, Streblidae). *Zootaxa.* (2016) 4205:101. doi: 10.11646/zootaxa.4205.2.1
27. Parola, P. Raoult, D., 2001. Ticks and tickborne bacterial diseases in humans: an emerging infectious threat. *Clin Infect Dis.*; 32, pp. 897-928
28. Wilkinson, D.A., Dietrich, M., Lebarbenchon, C., Jaeger, A., Le, R.C., Bastien, M. et al.; Massive infection of seabird ticks with bacterial species related to *Coxiellaburnetii*; *Appl Environ Microbiol.* 2014; 80, pp. 3327-3333
29. Boinas, F., Ribeiro, R., Madeira, S., Palma, M., de Carvalho, I. L., Nuncio, S., & Wilson, A. J., 2014. The medical and veterinary role of *Ornithodoroserraticus* complex ticks (Acari: Ixodida) on the Iberian Peninsula. *Journal of vector ecology*, 39(2), 238-248.
30. Lafri I., W., Benredjem, F., Neffah-Baaziz, R., Lalout, K., Abdelouahed, B., Gassen, S., and al., 2018. Inventaire et mise à jour sur les tiques argasidés et les agents pathogènes associés en Algérie ; Volume 23; 110-114
31. Madder, M., Thys, E., Geysen, D., Baudoux, C., & Horak, I., 2007. Tiques *Boophilusmicroplus* trouvées en Afrique de l'Ouest. *Acarologieexperimentaleetappliquée*, 43, 233-234.
32. Adjogoua, E. V., Diaha-Kouamé, C. A., Guindo, C. N., Diane, K. M., & Kouassi KARMC, D. M., 2021. Première détection du virus de la fièvre hémorragique Crimée Congo dans les tiques circulant à Bouaflé, région de la Marahoué, Côte d'Ivoire. *Revue Bio-Africa*, 25, 51-60.
33. Jaunbauere G., Salmane I., Spuis V. Occurrence of bat ectoparasites in latvia. – *Latvijasantomologs.* 2008 ; 45: 38-42.

34. Dick CW, Dittmar K. Parasitic bat flies (Diptera: Streblidae and Nycteribiidae): Host specificity and potential as vectors, in *Bats (Chiroptera) as Vectors of Diseases and Parasites*. Klimpel S, Mehlhorn H, Editors. Springer: Berlin. 2014 ; 187 p. (pp. 131–155).
35. Léger, C. Bat parasites (Acari, Anoplura, Cestoda, Diptera, Hemiptera, Nematoda, Siphonaptera, Trematoda) in France (1762–2018): a literature review and contribution to a checklist; *Parasite*. 2020 ; 27.
36. Diniz U.M., Gomez, L. F., Gnali, G. F., Raji, I. A. Drivers of Ectoparasite diversity and abundance in Paletropical bats. *First Bat Course on Ecology, Diversity, Conservation, And Ecosystem Services (Global South Bat)*. , 2020 ; Vol 1, N°1. Pp 82-88

#### **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

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