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Malaria transmission in the coastal zone and in the Centre of Côte d'Ivoire during the dry season

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Abstract

Background Malaria remains a threat in sub-Saharan Africa, particularly in Côte d'Ivoire, where it is endemic and represents the leading cause of hospital consultations, morbidity and mortality. The strong climatic variations that exist between coastal and savannah areas of Côte d'Ivoire suggest that vector control interventions should be scheduled according to the eco-epidemiological diversity. This study evaluates bioecological parameters of vectors and malaria transmission in two health districts, one coastal and one central of Côte d'Ivoire.

Methods The study was conducted in the coastal (Jacqueville) and savannah (Béoumi) areas of Côte d'Ivoire from November 2018 to March 2019. Human Landing Catches (HLC) were conducted monthly at the study sites to determine *Anopheles* vector species composition, biting behaviour as well as entomological parameters of malaria transmission. Mosquitoes were collected over 12 h, from 6:00 pm to 6:00 am during 2 days per month. Mosquitoes infectivity was revealed by enzyme-linked immunosorbent assay (ELISA) for *Plasmodium falciparum* circumsporozoite protein. A random sample of 100 *Anopheles gambiae* sensu lato (s.l.) including all CSP-positive females, were further classified by polymerase chain reaction (PCR) at the species and molecular form levels.

Results In Jacqueville, 853 (99.7%) *An. gambiae* s.l., and 3 (0.35%) *Anopheles pharoensis* were collected. In Béoumi, 811 (96.3%) *An. gambiae* s.l., 23 (2.73%) *Anopheles funestus* and 8 (0.95%) *An. pharoensis* have been found. *Anopheles coluzzii* represented the only species of the *An. gambiae* complex in Jacqueville. Among the *An. gambiae* s.l. samples tested in Béoumi, 29 (58%) were *An. coluzzii* and the rest 21 (42%) was *An. gambiae* sensu stricto. The human biting rate (HBR) in Jacqueville increased from 5.7 (b/p/n) in November to 17.3 (b/p/n) in March. Conversely in Béoumi the HBR decreased from 16.4 (b/p/n) in November to 0.69 (b/p/n) in March. In Jacqueville, the entomological inoculation rate (EIR) varies from 0.21 to 0.56 (ib/p/n) with the pic of 0.56 (ib/p/n) in February. In Béoumi no infection was detected in the parous *An. gambiae* s.l. samples tested during the study period.

Conclusions This study evaluates bioecological parameters of vectors and malaria transmission in two health districts, one coastal and one central of Côte d'Ivoire.

Keywords Malaria, Mosquitoes, Coastal area, Savannahs, *Anopheles gambiae*, Côte d'Ivoire

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Background

Malaria is a major global public health problem and a leading cause of morbidity and mortality in many countries [1]. Globally, the number of malaria cases has been estimated at 247 million in 2021 in 84 malaria-endemic countries, up from 245 million in 2020. Most of the additional cases are estimated to be in the World Health Organization (WHO) African Region [2]. In 2020, malaria deaths increased by 10% compared with 2019, to an estimated 625 000. Estimated deaths declined slightly in 2021 to 619 000. The percentage of total malaria deaths in children aged under 5 years reduced from 87% in 2000 to 76% in 2015. Since then there has been no change [2]. Malaria remains a threat in sub-Saharan Africa, particularly in Côte d'Ivoire where it is endemic and is the leading cause of hospital consultations, morbidity and mortality [3]. It is endemic and its transmission is stable throughout the year with peaks during the rainy season. In Côte d'Ivoire, while the incidence rate of malaria in the general population was 115 cases per 1,000 and 389 % in children under 5 years age, it increased between 2016 and 2018, from 155 to 189.9 % in the general population and from 287 to 492.9 % in children under age 5. The low-incidence health was Abidjan1-Grands Ponts (Jacqueville) (170.1%) and the health region with the highest incidences are Indenié-Djuablin (859.7%), Gbeke (Beoumi) (754.6%) and Hambol (712.4%) in 2018 [4].

The risk of malaria transmission is heterogeneous and varies over time depending on the season but also on the year depending on climatic events [5]. Vector control remains the commonly prioritized measure for malaria control as it allows to reduce and interrupt malaria transmission [6, 7]. The main control method used to reduce the burden of malaria in Côte d'Ivoire is the distribution and use of long-lasting insecticidal nets (LLINs). Recently, the National Malaria Control Programme (NMCP) adopted indoor residual spraying as supplementary strategy to reduce malaria morbidity.

The climatic diversity in the country, predisposes to a great phyto-geographical diversity, which could support the variation of epidemiological situations [8]. In the coastal areas of the Ivory Coast, little is known about the entomological parameters of malaria transmission in the dry season compared to the savannah areas. Comprehensive monitoring of vector bionomics at these sites will provide information on vector distribution and infectivity and help determine the best time for vector control interventions.

Malaria vector bionomics was monitored in Jacqueville and Beoumi, from 2018 to 2019, over a period of 5 months. The objective of this study was to evaluate entomology parameters of vectors and malaria transmission in two health districts of Côte d'Ivoire.

Methods

Study area

The study was conducted in Jacqueville city ($5^{\circ} 12' 21.532''$ N $4^{\circ} 25' 24.071''$ W) and in Beoumi ($7^{\circ} 40' 23.131''$ N $5^{\circ} 34' 20.028''$ W). Jacqueville is located in the South of Côte d'Ivoire in the coastal zone, characterized by four seasons: a main rainy season (March–July), a minor rainy season (September–November), a main dry season (December–February), and a minor dry season (August), with a mean annual temperature of 28 °C and more than 1,500 mm of rainfall per year.

Beoumi is located in the savannah area characterized by two seasons: one long rainy season from March to November followed by a short dry season from December to February. The mean annual temperature is 26 °C with a mean annual rainfall of 801 mm. In each district, one urban and one rural site were selected (Fig. 1).

Adult mosquito collection

The adult mosquito collections were carried out in Jacqueville and Béoumi, from November 2018 to March 2019s over two different sub-seasons: From November to December at end of the rainy season and from January to March, in the dry season. Human Landing Catches (HLC) were conducted monthly to determine the *Anopheles* vector species composition, the human biting density, the biting cycle and the biting location (indoor/outdoor) as well as entomological parameters of malaria transmission. Volunteers were used as human bait to collect landing mosquitoes for 12 h, from 6:00 pm to 6:00 am using haemolysis tubes. Two houses were selected per site and collection were performed for two consecutive nights. As each pair of houses generates 1 measurement, two pairs of houses have been required per site. To avoid for attractivity biases collectors switched indoors and outdoors during the collection period. All mosquitoes collected through each method were morphologically identified to genus using identification keys. The parity of female anopheline mosquito was assessed by dissecting and observing ovaries [9]. All collected *Anopheles* females were preserved individually on desiccant silica gel in Eppendorf tubes, until laboratory processing to determine sibling species, *Plasmodium* infection using polymerase chain reaction (PCR) and enzyme-linked immunosorbent assay (ELISA), at the Centre Suisse de Recherche Scientifique (CSRS).

Ethical considerations

The study protocol received the approval of Côte d'Ivoire's national ethics committee and by local health and administrative authorities before commencing

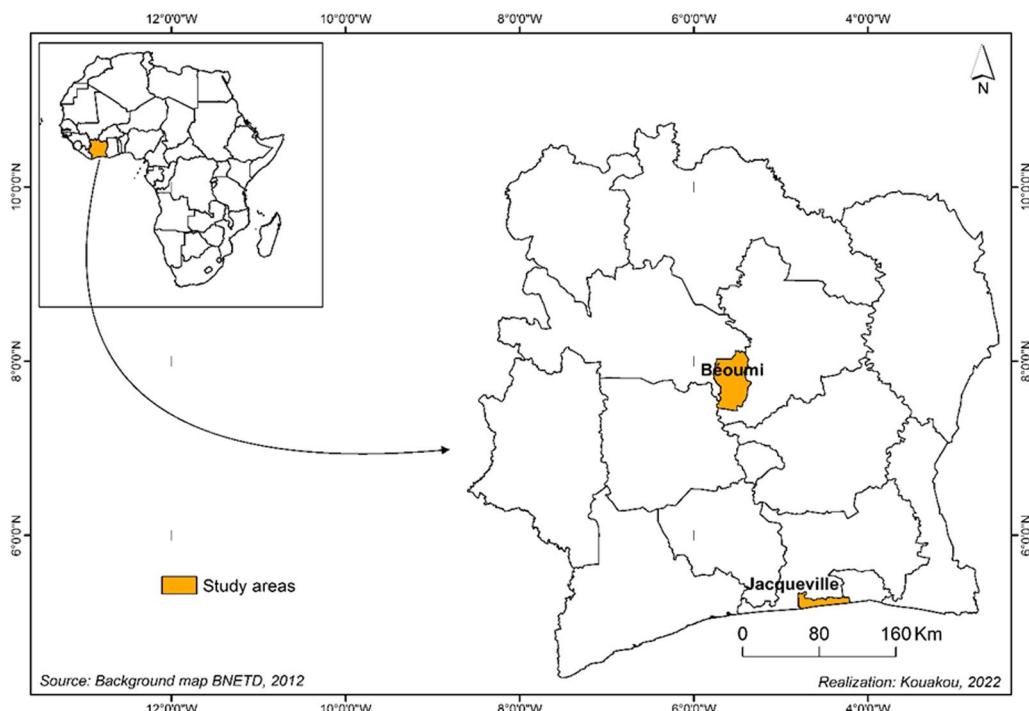


Fig. 1 Map of the study areas

activities under the number: N/Ref: 168-18/MSHP/. Volunteers vaccinated against yellow fever and placed on anti-malarial prophylaxis were used as human bait.

Laboratory mosquito processing

The presence of *Plasmodium falciparum* circumsporozoite protein (CSP) inside infected mosquitoes were revealed by the enzyme-linked immunosorbent assay (ELISA) [10–12]. Genomic DNA was extracted from the legs and abdomens of a random sample of 100 *An. gambiae* s.l. females using the method of Collins (2007) [13]. *Anopheles gambiae* complex species were identified as *Anopheles gambiae*, *Anopheles coluzzii*, or hybrids of the two species, following the Short-Interspersed Element protocol described by Santolamaza (2008) [14].

Entomological parameters

Entomological parameters of transmission were calculated in the MACDO-NAL method after dissection of *An. gambiae* females. The human biting rate (HBR) is expressed as the number of female anopheline bites per person per night (b/p/n); The parity is revealed by dissecting the ovaries and by inspecting of the tracheoles according to the DETINOVA method [15]. in order to calculate the parity rate which is the ratio of the number of parous females to the total of dissected

mosquitoes; the infection rate (IR) corresponds to the proportion of females infected by *P. falciparum*; the entomological inoculation rate (EIR), expressed as the number of infective anopheline bites per person per year, is calculated as the product of the HBR and the IR of mosquitoes collected on humans.

Statistical analysis

Data were first entered in Excel and analysed the statistical software R logiciel version R4.1.2.

The following entomological parameters were determined are the human biting rate (HBR), expressed as the number of female anopheline bites per person per night (b/p/n); the parity rate (PR) was calculated as the proportion of parous females; the infection rate (IR) corresponding to the proportion of females infected by *P. falciparum*; and the EIR, expressed as the number of infective anopheline bites per person per year, was calculated as the product of the HBR and the IR of mosquitoes collected on humans. Chi-square tests were used to compare different proportions, and Kruskal-Wallis test was used to compare HBR between the two and equality of population rank test was used to compare density of species between the two sites. All differences were considered significant at $P < 0.05$.

Table 1 Species composition of mosquitoes collected in Jacqueville and Béoumi

Locality	Species	November 2018	December 2018	January 2019	February 2019	March 2019	Total
Jacqueville	<i>An. gambiae s.l.</i>	91 (10.6)	138 (16.1)	112 (13.1)	235 (27.5)	277 (32.4)	853 (99.)
	<i>An. pharoensis</i>	0 (0)	0 (0)	1 (0.1)	0 (0)	2 (0.2)	3 (0.3)
Béoumi	<i>An. gambiae s.l.</i>	535 (63.5)	235 (27.9)	23 (2.7)	7 (0.8)	11 (1.30)	811 (96.3)
	<i>An. funestus s.l.</i>	0 (0)	6 (0.7)	9 (1.1)	8 (0.9)	0 (0)	23 (2.7)
	<i>An. pharoensis</i>	0 (0)	0 (0)	8 (0.9)	0 (0)	0 (0)	8 (0.9)

Table 2 Species identification of *An. gambiae* s.l. in the Jacqueville and Beoumi

Locality	Total tested	<i>An. coluzzii</i>	<i>An. gambiae</i>
Jacqueville	50	50 (100%)	0 (0%)
Béoumi	50	29 (58%)	21(42%)

Results

Adult mosquito species composition

A total of 5,167 female adult mosquitoes were collected by HLC in the two study districts with 2,998 in Jacqueville and 2,169 in Béoumi. In Jacqueville, anopheline mosquitoes represented 28.55% (856) of all mosquitoes collected while culicinae represented 71.45% ($n = 2142$).

Among anopheline mosquitoes, *An. gambiae* sensu lato (*s.l.*) was the predominant species 99.6% ($n=853$), followed by *Anopheles pharoensis* 0.4% ($n=3$) (Table 1). *Anopheles coluzzii* was the only species of the *An. gambiae* *s.l* molecularly identified (Table 2).

In Beoumi, anopheline mosquitoes represented 38.82% (842) of all mosquitoes collected while culicinae represented 61.18% ($n=1327$). Among anopheline mosquitoes, *An. gambiae s.l.* was the predominant species 96.3% ($n=811$) followed by *An. pharoensis* 1% ($n=8$) and *Anopheles funestus* 2.7% ($n=23$) (Table 1). *Anopheles coluzzii* represented 58% ($n=29$) and the

rest represented 42% ($n=21$) of *An. gambiae* sensu stricto (s.s.) (Table 2).

Biting behaviours and biting rates

Out of the 853 *An. gambiae* s.l. collected in Jacqueville, 52.9% were found indoors. In Béoumi, out of the 811 *An. gambiae* s.l. collected, 58.7% were collected outdoors. *Anopheles funestus* collected in Beoumi in January and February was predominantly found indoors (70.6%; n=24). There were different HBR trends in the two sites over the collection period (Table 3). The human bite rate (HBR) in Jacqueville rose from 5.7 b/p/n in November to 17.3 b/p/n in March. The highest densities were observed during the dry season from February to March (14.7–17.3 b/p/n) in Jacqueville.

The average human bite rate of *An. gambiae* s.l. in Jacqueville was 10.66 b/p/n. The human bite rate (HBR) in Jacqueville increased from 5.7 b/p/n in November to 17.3 b/p/n in March. The highest bite rates were observed during the dry season from February to March (14.7 to 17.3 b/p/n), and the lowest bite rate of *An. gambiae* s.l. was 5.7 b/p/n (Table 4).

In Beoumi the average human bite rate was 11.08 b/p/n. The highest bite rate ranged from (14.7 to 33.5 b/p/n) and the lowest from (1.38 to 4 b/p/n). The human bite rate fell from 16.4 b/p/n in November to 0.7 b/p/n in March. The highest densities were observed at the end of the rainy season from November to December (33.5 to 14.7 b/p/n).

Table 3 Biting behaviour of *An. gambiae* and *An. funestus* per site

Locality	Species		November	December	January	February	March	Total
JACQUEVILLE	<i>An. gambiae</i>	In N (%End)	44 (48.4)	79 (57.2)	64 (57.1)	122 (51.9)	142 (51.3)	451 (52.9)
		Out N (%Exo)	47 (51.6)	59 (42.8)	48 (42.9)	113 (48.1)	135 (48.7)	402 (47.1)
		Total	91	138	112	235	277	853
BEOUMI	<i>An. gambiae</i>	In N (%Endo)	236 (44.1)	78 (32.4)	22 (47.8)	4 (28.6)	14 (63.6)	354 (41.3)
		Out N (%Exo)	299 (55.9)	163 (67.6)	24 (52.2)	10 (71.4)	8 (36.4)	504 (58.7)
		Total	535	241	46	14	22	858
	<i>An. funestus</i>	In N (%Endo)	0	0	0	10 (55.5)	14 (87.5)	24 (70.6)
		Out N (%Exo)	0	0	0	8 (44.4)	2 (12.5)	10 (29.4)
		Total	0	0	0	18	16	34

Table 4 Monthly biting rate results using HLC

Locality	Months	IBR <i>An. gambiae</i>	OBR <i>An. gambiae</i>	IBR <i>An. funestus</i>	OBR <i>An. funestus</i>	Total biting rate (bpm)
JACQUEVILLE	November	2.8	2.9	–	–	5.7
	December	4.9	3.7	–	–	8.6
	January	4	3	–	–	7
	February	7.6	7.1	–	–	14.7
	March	8.9	8.4	–	–	17.3
BEOUMI	November	14.8	18.7	–	–	33.5
	December	4.6	10.1	–	–	14.7
	January	1.4	1.5	0.6	0.5	4
	February	0.25	0.6	0.9	0.1	1.85
	March	0.88	0.5	0	0	1.38

IBR Indoor biting rate, OBR Outdoor biting rate

Table 5 Monthly parity rates of dissected mosquitoes from indoor and outdoor collections per sites

Locality	Month	Dissected	Parous	Parity rate
Jacqueville	November	88	88	100
	December	132	46	34.85
	January	110	104	94.55
	February	231	166	71.86
	March	274	264	96.35
Beoumi	November	407	383	94.10
	December	237	215	90.72
	January	21	8	38.09
	February	5	3	60
	March	11	3	27.27

(Table 4). The average bite rate of *An. gambiae* collected was 10.66 b/p/n and that of *An. funestus* was 0.42 b/p/n. There were different trends in the human bite rate at the two sites over the collection period (Table 4).

Parity rate

The parity rates varied between 34.9% in December and 100% in November in Jacqueville. In Beoumi, the parity rates varied between 27.3% and in March and 94.1% in November. The mean parity in Jacqueville (74.4%) was lower than the mean parity rate observed in Beoumi (81.6%). However, there was no significant difference between the parity rates of the two sites ($P=0.241$) (Table 5).

Infection and entomological inoculation rate

200 samples were tested in Jacqueville and 301 in Beoumi. Samples were selected on the basis of the mosquitoes had already taken a blood meal after dissection. In Jacqueville, all *An. gambiae* s.l. samples examined

contained at least one infection. The infection rate of *An. gambiae* s.l. collected was 0.075%. In Jacqueville, the EIR varied from 0 in November to 0.56 (ib/p/n) in February. They varied from 0.013% to 0.024% per month.

In Beoumi, in contrary no infection was found in the examined samples. As no infection was observed in Beoumi, the infection rate of *all species* collected was 0%. The EIR was 0 ib/p/n (Table 6). Table shows the monthly entomological inoculation rates (EIRs) for *An. gambiae* s.l. in the two areas.

Discussion

The present study shows that malaria transmission is essentially ensured by *An. gambiae* s.l. and that is the main malaria vector in Jacqueville (99.7%) and Beoumi (96.3%). Previous work has also shown that the major vector for malaria transmission is *An. gambiae* s.l. [16, 17]. In Jacqueville, it was observed that the vector biting densities of mosquitoes seemed to be more evenly distributed over the entire period of the study. Indeed, the distribution of rainfall throughout the year would have favoured this state of fact because practically every two months it rains in this part of the country. The permanent presence of water in the south of the country could have favoured this distribution of mosquitoes obtained in high proportion each month during the study period. However, in Béoumi, the distribution of identified malaria vectors appears largely at the end of the rainy season from November to December and in very small quantities between January and March. In fact, at the end of the rainy season, there is still the presence of water and, therefore, favourable to mosquito breeding sites. On the other hand, the months of January to March correspond to the dry season in the centre of the country where the breeding sites are absent and, therefore, less conducive to

Table 6 Monthly sporozoite and EIRs of the two districts of bionomic survey

Locality	Month	Total <i>An. gambiae</i> s.l.	Number of circum-sporozoite positive	Sporozoite rate (%)	HBR (b/p/n)	EIR (ib/p/n)
Jacqueville	November	0	0	0	5.7	0
	December	42	1	0.024	8.6	0.206
	January	0	0	0	7.0	0
	February	78	3	0.038	14.7	0.5559
	March	80	1	0.013	17.3	0.225
	Total					
Beoumi	November	134	0	0.0	16.4	0.000
	December	138	0	0.0	7.4	0.000
	January	15	0	0	1.45	0
	February	6	0	0	0.425	0.000
	March	8	0	0	0.69	0.000

the spread of malaria vectors [18]. The diversity and variation in *Anopheles* mosquito species composition may be related to human activities (incl. rice farming, vegetable crops) and with vector control interventions, as well [19]. *Anopheles coluzzii* was the only species of the *An. gambiae* complex in Jacqueville. A study conducted in coastal cities such as San Pedro, Abidjan and Aboisso showed that the *An. gambiae* complex was dominated by the molecular form M (*An. coluzzii*) [17, 20]. The abundance of *An. coluzzii* in the samples from forest areas could be related to the type of breeding sites and the climatic conditions in these study sites [21]. Several studies carried out in Côte d'Ivoire have shown a predominance of *An. coluzzii* in forest area especially in the western [20] and south-eastern [22] parts. The results obtained corroborate with those of the forest zone and the intermediate zone.

In Beoumi, the species were present in relatively equal proportions. The presence of these two species in this locality would be due to the difference of the climatic conditions between these two localities. A study conducted in Côte d'Ivoire shows that there are three distribution areas where the species *An. gambiae* s.s. is dominant (sites located in savanna) an area with predominance of *An. Coluzzii* (in the southern forested area) and an intermediate area where the two species were in a same proportion (pre-forested site) [23]. The presence of both species in almost equal proportions in Beoumi could be explained by the fact that it is located in an intermediate area (in a pre-forest zone). The relative dominance of these two species may be associated with specific and characteristic breeding sites. The presence of *An. gambiae* s.s. and *An. coluzzii* has been previously reported in Côte d'Ivoire [23, 24]. In Jacqueville malaria vectors are both endophagic and exophagic unlike in the town of Beoumi, where the vectors had an exophagic

behaviour. The observation of these results shows that the biting behaviour different from one area to another within the same country. This could be explained by the fact that climatic factors differ from one area to another. In Jacqueville the biting rate increased from 5.7 (b/p/n) to 17.3 (b/p/n) while it has evolved in the opposite way to that in Beoumi 16.4 (b/p/n) to 0.69 (b/p/n). This suggests that, vector control measures must be scheduled according to the existing eco-epidemiological diversity, which has to be well understood before embarking upon any extensive action [25].

Parity rate was high in both sites. Similar findings have previously been reported in Côte d'Ivoire [26, 27]. This shows that the mosquito vector population is old. Given the dry season and the scarcity of puddles, female mosquitoes will be looking to reproduce at any moment to ensure the survival of their species. Bed nets were distributed in May 2017, one month and a half later the study was conducted. It may be the nets are not normally used by the household occupants. Also the resistance status of malaria vectors to pyrethroids is increasingly high in Côte d'Ivoire [28, 29]. As a result, even if vectors come into contact with insecticide-impregnated nets they do not necessarily die and can continue to bite. There is also the fact that, the only means of protection is impregnated mosquito nets which are only used at night. This situation leads to increased human-vector contact. Although treated nets are distributed, the rate of use by the population is low [30]. The fact that the vectors are both endophagous and exophagous could explain the high rate of parasitic females. Indeed, the populations have no means of protection when they are out of the bedrooms, so they are continuously exposed to mosquito bites [31]. The entomological inoculation rate (EIR) varied from 0 to 0.56 (ib/p/n) with a high EIR rate in February 0.56 (ib/p/n) in Jacqueville. Lowest malaria prevalence in

Jacqueville could be due to the low vector density and the relatively low EIR in this locality. Moreover, annual EIR of over 1 ib/p/year may be sufficient to maintain *Plasmodium* transmission [32]. Contrary to Beoumi where no *An. gambiae* species was infected with *Plasmodium*. The study of infectivity to *P. falciparum* shows that no infected individual was found in the population of *An. gambiae s.l.*, in Beoumi. This could be explained by the fact that the mosquito population collected in Jacqueville only took their blood meal from individuals not infected with *Plasmodium*. The fact of having a large number of mosquitoes tested, a high biting rate also is not synonymous with the presence of infection.

Conclusion

This study shows us how much the climatic differences at the local scale influence the various eco-epidemiological and bioecological parameters of the coastal zones and the savannahs.

Mosquitoes were endophagous in Jacqueville and exophagous in Beoumi. Entomological parameters of malaria transmission of these two districts are very different. It also teaches us that the level of biting rate does not necessarily influence the risk of transmission. This is what the results obtained at Béoumi show us; where despite the high number of mosquitoes captured, there were no cases of infections. This result suggests that vector control measures must be selected according to the existing eco-epidemiological. The main malaria vector control method that can be used in the two health districts is the distribution and use of ITNs.

Abbreviations

ITNs	Insecticidetreated nets
WHO	World Health Organization
EIR	Entomological inoculation rate
HBR	Human biting rate
IR	Infection rate
PCR	Polymerase chain reaction
ELISA	Enzyme-linked immunosorbent assay
CSRS	Centre Suisse de Recherche Scientifique
LLINs	Long-lasting insecticidal nets
HLC	Human landing catches
CSP	<i>Plasmodium falciparum</i> Circumsporozoite protein
NMCP	National Malaria Control Programme

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Author contributions

GJR was a major contributor in data collection, writing original draft, analyzing and interpreting the manuscript. CAVE was involved in the project administration, supervision, study design and review. EK, CGNG and JZBZ supervised and

approved the manuscript. GBK has designed the study, supervised, read and approved this document and was involved in funding acquisition.

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Availability of data and materials

No datasets were generated or analysed during the current study.

Declarations

The findings and conclusions expressed herein are those of the author(s) and do not necessarily represent the official position of neither USAID nor PMI, nor those of Centers for Disease Control and Prevention (CDC). Ethics approval and consent to participate

The study protocol has been approved by the Côte d'Ivoire national ethics committee prior to the start of the experiments. The study protocol has been approved by the local health and administrative authorities. Before starting the study, the village chiefs gave their oral informed consent. In addition, all entomological surveys and sample collections carried out on private land or private residential areas were carried out with the authorization and informed written consent of the residents. Volunteers participating in the HLC have given written informed consent for their participation.

Consent for publication

Not applicable.

Competing interests

The authors declare no competing interests.

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