

## Diversity and dynamics of malaria-carrying mosquitoes in three villages in Aboisso, south-east Côte D'Ivoire

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### Abstract

Mosquitoes have an epidemiological role as a vector of many parasites which can affect populations. So, an inventory of mosquitoes during the period from 2012 to 2013 was conducted in three villages' names N'zikro, Ehania-V1 and Ayebo in the Aboisso health district located in the southeastern of Côte d'Ivoire. The aim was to identify mosquito breeding sites and the main vectors. Mosquito larvae were collected using the dipping method, and their breeding sites were characterised. Adults were captured using three methods: human landing catches (HLC), pyrethrum spray catches (PSC), and window trap (WT). Following these collections, mosquito species were morphologically identified. *Plasmodium falciparum* infections were detected by ELISA method. A total of 17 types of larval breeding sites were inventoried, including 4 natural and 13 anthropogenic. A total of 217 larvae were collected from the three villages. The proportions of *Culex*, *Anopheles*, and *Aedes* were 64.98%, 60.37%, and 12.90%, respectively. The three capture methods yielded 3,954 adult mosquitoes. Four genera were identified: *Anopheles* (9.43%), *Culex* (87.78%), *Mansonia* (0.53%), and *Aedes* (0.25%), with *An. gambiae s.l.* being the most prevalent. The average infestation rates recorded in the three villages were comparable. The annual inoculation rate in the village N'zikro was 0.0308 infectious bite/man/night or 11 infectious bites/man/year. The entomological inoculation rate in the other two villages was zero. The study shows that these agroecosystems offer various habitats favourable to vector proliferation and also influence mosquito dynamics.

**Keywords:** Malaria, agroecosystems, anopheles, aboisso, Côte D'Ivoire

### Introduction

In both rural and urban areas, the transformation of the environment for agricultural purposes is a daily occurrence (Afrane *et al.* 2004; Yadouléton *et al.* 2010) <sup>[1, 2]</sup>. In Côte d'Ivoire, 51.2% of the population lives in urban areas, which poses a recurring problem of food and economic security (Lovell 2010) <sup>[3]</sup>. To address this, urban and peri-urban agriculture appears as a very promising alternative to bridge this food and economic gap (Lovell 2010) <sup>[3]</sup>. These intensive agricultural practises promote environmental transformation, resulting in the creation of numerous Culicidae larval habitats (Afrane *et al.* 2004; Adja *et al.* 2021) <sup>[1, 4]</sup>, hence the risk of vector-borne diseases including malaria, dengue fever, chikungunya, and yellow fever (Bigoga *et al.* 2012; Afrane *et al.* 2012) <sup>[5, 6]</sup>. Indeed, intensive agricultural practises can promote the proliferation of mosquitoes (Culicidae) by creating numerous breeding sites, particularly areas of stagnant water (puddles, ponds) and disturbed habitats that are conducive to larval development, thus impacting public health through the transmission of vector-borne diseases. In the Aboisso region, vast areas of oil palm and rubber plantations have been established to contribute significantly to the country's agricultural and economic development. These industrial plantations are heavy water consumers, resulting in the proliferation of breeding sites and therefore the multiplication of disease-carrying mosquitoes. Several

studies conducted in tropical countries have shown a link between agricultural practises and the proliferation of disease-carrying mosquitoes (Afrane *et al.* 2004; Akono *et al.* 2015) <sup>[1, 7]</sup>. To conduct a sustainable and effective mosquito control campaign, it is essential to study species composition, population dynamics, and periods of abundance. Mosquitoes pose a significant health risk to humans and often to animals due to their ability to transmit pathogens and the nuisance they cause through their bites. To contribute to the preservation of the health of populations living near agricultural units, a study was conducted in three agroecosystems in the Aboisso region, located in the southeast of the country: an area with intensive oil palm and rubber plantations, and a control area of food crops. The objective of this work is to identify mosquito breeding sites and the main vectors in these areas. This is crucial for understanding how agriculture influences these vectors and public health.

### Material and methods

#### Study sites

The study was conducted in three agroecosystems (N'zikro, Ehania-V1 and Ayebo) within the Aboisso health district (5° 28' 06" N, 3° 12' 25" W), located in southeastern Côte d'Ivoire. Aboisso has a hot and humid climate with two rainy seasons (a major rainy season from May to July and a minor rainy season from September to November) and two

dry seasons (a minor rainy season in August and a major rainy season from December to April), characterised by a subequatorial climate with average temperatures around 27°C year-round. The wettest periods are June and October-November, while January is the driest month. Three villages were selected in this region based on the crops grown there: N'zikro, Ehania-V1, and Ayebo (Fig. 1). These three villages have been previously described elsewhere (Sadia-Kacou *et al.* 2021) <sup>[8]</sup>. The village and the plantation were grouped under the term "agroecosystem." Within each agroecosystem, one village was selected for the study. Thus, the N'zikro agroecosystem (7°32'30.0" N and 5°4'12" W), a crossroads village, had a population of 9,415 in 2013. Its population consists of indigenous Agni people and immigrants from Burkina Faso, Mali, Ghana, and Guinea. This village is a rubber-growing area with 255.5 hectares of plantations, some located less than 50 meters from dwellings. To the southwest of this village lie more than 3,600 hectares of oil palm plantations belonging to PALMCI (Palm Côte d'Ivoire, an agro-industrial company specialising in oil

palm plantations). The socio-economic infrastructure available to this village includes a health center and primary schools. It also has electricity and running water. The Ehania-V1 agroecosystem (5°18' N and 3°04' W) is the largest of the seven integrated agricultural units of PALMCI. It houses a palm kernel processing unit. It had 2,169 inhabitants in 2013. The population consists of indigenous Agni people and immigrants from Burkina Faso, Mali, and Ghana. This area is surrounded by over 5,157.15 hectares of industrial oil palm plantations and 100 hectares of preserved primary forest. The rainforest provides lush vegetation with dense cover.

The socio-economic infrastructure available to this village includes a modern health center well-stocked with medicines provided by PALMCI, primary schools, and a bank. Electricity and running water are also available. The Ayebo agroecosystem (5°26'27''N and 3°15'52''W) had 4812 inhabitants in 2013. The population consists of indigenous Agni people and migrants from Burkina Faso, Mali, Ghana, and Guinea. The main activity is the cultivation of tubers (yams and cassava).

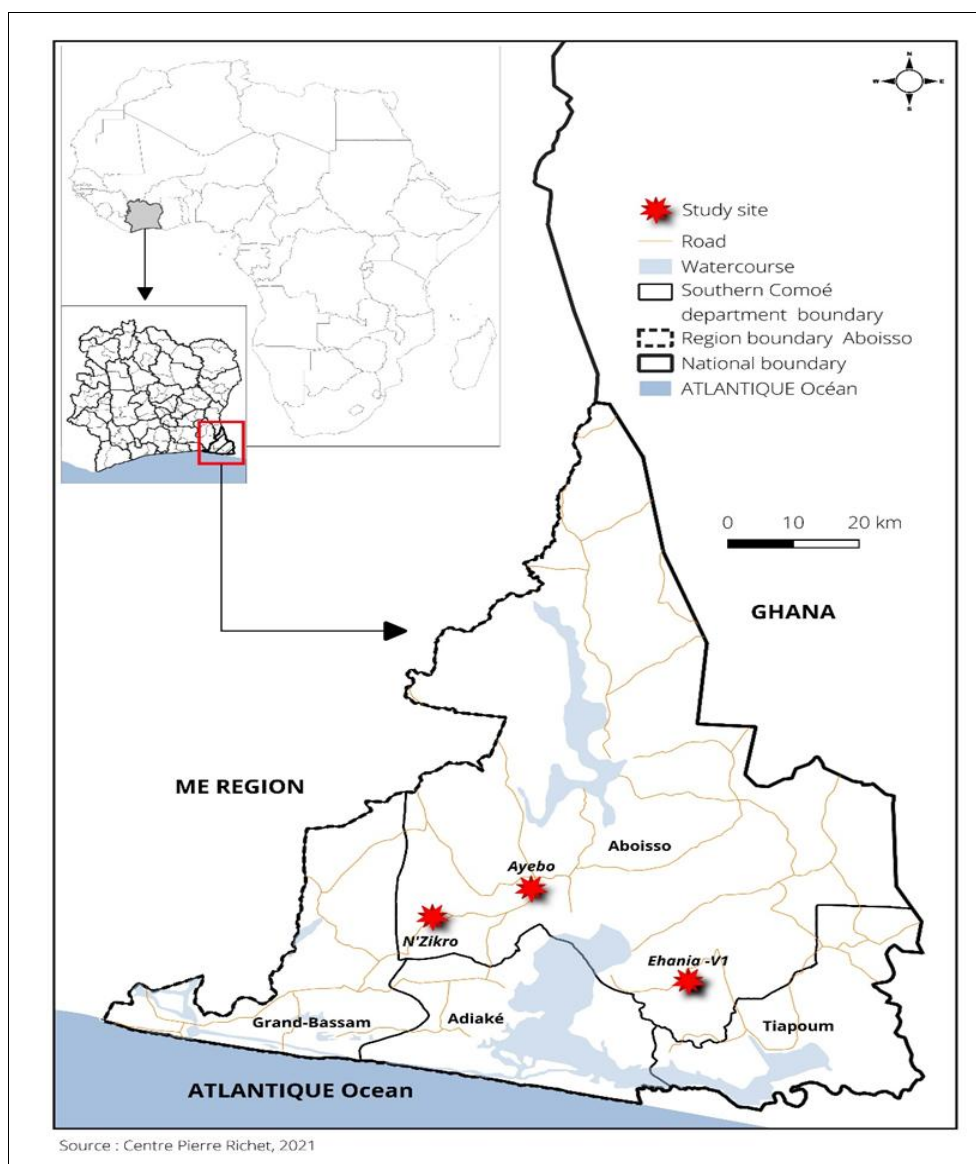


Fig 1: Studies sites

In addition, food crops, cereals, and fruit trees are grown. Agricultural activities in this village take place more than

two kilometers from the village center and are not extensive. This village serves as the pilot site. The socio-economic

infrastructure available in this village includes a health center and primary schools. It also has electricity and running water.

### **Mosquito breeding sites characterization and larval survey**

In each site, the habitat types containing larvae were identified as key variable, namely the nature of the breeding sites (anthropogenic or natural), was recorded. The census and characterization of larval breeding sites were carried out from February to July in the three agro-ecosystems in 2013. Pre-imaginal stages (larvae, pupae) were collected using the dipping method with 90 mls. A total of 10 dips were made in each container and then transported to insectaries at the Pierre Richet Institute in Bouaké. The larvae were then monitored until emergence and subsequently identified.

### **Adult mosquito sampling**

Three methods were used to maximise the capture of *Anopheles* mosquitoes: human landing catches (HLC), pyrethrum spray catches (PSC), and window trap (WT). Adult mosquitoes were captured at four capture sites located exclusively inside houses in each agroecosystem for two consecutive nights. Captures were carried out over a total of four missions (during August-September 2012, November-December 2012, February 2013, and June-July 2013) by two teams of four people. The first team captured mosquitoes from 6:00 PM to midnight, and the second team from midnight to 6:00 AM the following morning. A total of 32 capture devices were used at each site throughout the study. The mosquitoes were collected, and carefully stored in closed hemolysis tubes with cotton and kept in plastic bags by time slot and by catching point. The human biting rate (HBR) of *An. gambiae* was calculated as the average number of mosquitoes collected per person per night (bites/human/night, or year). Pyrethrum spray catches (PSC) involved spraying a 1% pyrethrin aerosol solution on the interior surfaces, walls, and roofs of 60 bedrooms during six surveys conducted between November 2012 and October 2014 to collect resting mosquitoes. This was carried out early in the morning, between 6:00 and 8:00 AM. The day before, the heads of households in the houses to be used for PS were informed and their consent obtained. They were asked to keep the bedrooms closed until the team arrived. Before spraying, all bedroom entrances were carefully sealed; a white sheet was spread in the room, and then the insecticide was sprayed. After a 10-minute wait outside the room, the sheet is removed, and all stunned mosquitoes are collected and placed in petri dishes. The name of the village, the house number, the type of building material, and the number of people who slept in the room the previous night are recorded in a specially designed form. Window traps are mounted on bedroom windows to capture mosquitoes that attempt to leave the rooms after feeding. Four traps were used per survey. Mosquitoes are collected via suction tubes inside the window traps. Collection takes place very early in the morning (6:00 AM). The mosquitoes are placed in petri dishes with the site name, the capture point number, the type of building material, and the number of people who slept in the room. Sampling was carried out over two days during each of the four (4) surveys, from March 2013 to November 2014. In total, 32 trap windows were used during the study period at each site.

**Identification and treatment of mosquitoes** Following collection, the mosquitoes were brought to the laboratory and then sorted according to sex, genus, and species based on morphological criteria. Two identification keys were used: Mattingly's (1971) for the identification of Culicidae and Gillies and Meillon's (1968) for the identification of *Anopheles* species. Females were counted and preserved for later analysis. *Plasmodium falciparum* infection was detected by the CSP ELISA technique at the Pierre Richet Research Center laboratory in Bouaké.

### **Statistical Analysis**

Data were entered into Microsoft Excel and analysed using STATA version 17.0 (Stata Corporation, College Station, USA). For each capture method, the number of species, the human biting rate (HBR), corresponding to the average number of mosquito bites received per person per night (b/p/n), and the infestation rate was estimated. The entomological inoculation rate (EIR), expressed as the number of infected bites per day and per night, was defined as the number of *Anopheles* mosquito bites received per person per night multiplied by the infestation rate. The Kruskal-Wallis (KW) test was used to compare the different variables between the three villages.

### **Ethical Considerations**

This study received approval from the National Malaria Control Program (PNLP) prior to its implementation. Health authorities in the Aboisso district and the farming villages were informed of the study's objectives, procedures, and benefits. Verbal agreement from members of the various communities and written consent from district health officials were obtained before data collection began. Volunteer mosquito trappers were treated before the mosquito-trapping period and again as soon as they developed symptoms suggestive of malaria. Their free and informed consent was obtained.

## **Results**

### **Characteristics and types of larval breeding sites**

Across all three agroecosystems, 17 types of larval breeding sites were visited, including 4 natural and 13 anthropogenic (Table 1). The highest number of breeding site types was observed in Ayebo (8) and Ehania-V1 (7), followed by N'zikro (6). Based on mosquito genera, three groups of breeding sites were identified: sites specific to *Anopheles* or *Culex*, mixed *Anopheles-Culex*, or *Culex-Aedes*. *Culex*-specific breeding sites were found primarily in the village of N'zikro in rainwater collection areas. *Anopheles*-specific breeding sites included cemented wells, unfinished houses, hoof prints, construction holes in the village of Ehania-V1, and holes in uprooted palm trees found in the village of Ayebo. The various roosts, such as the holes of uprooted palm trees, construction holes, and unfinished houses, can be considered atypical. All other roosts are mixed.

### **Pre-imaginal fauna following larval surveys**

The pre-imaginal fauna collected following surveys of larvae in the three villages includes *Anopheles*, *Culex*, and *Aedes* genera. A total of 217 larvae were collected from the three villages. The proportions of the genera *Culex*, *Anopheles*, and *Aedes* were 64.98%, 60.37%, and 12.90%,

respectively, of all larvae collected from the villages (Fig. 2). The proportions of mosquito genera varied among the villages.

In N'zikro village, 24 larvae were collected. Within this village, the genus *Culex* was dominant, representing 71% of the larvae collected. In Ehania-V1 village, 111 larvae were collected. The genus *Anopheles* was dominant, representing 56%, followed by the genus *Culex*, which represented 37%. In Ayebo, 82 larvae were collected. The genus *Anopheles* was dominant, representing 54% of the larvae collected. In general, in all villages, the *Aedes* genus was poorly represented with respective proportions of 8%, 7% and 13% in N'zikro, Ehania-V1 and Ayebo.

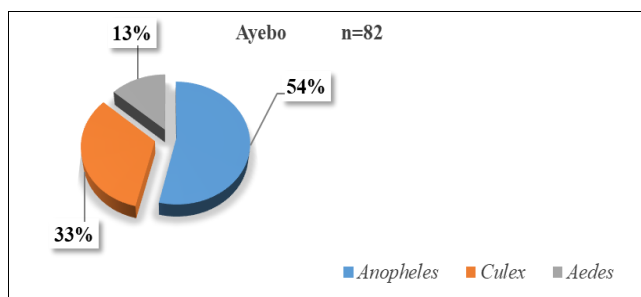
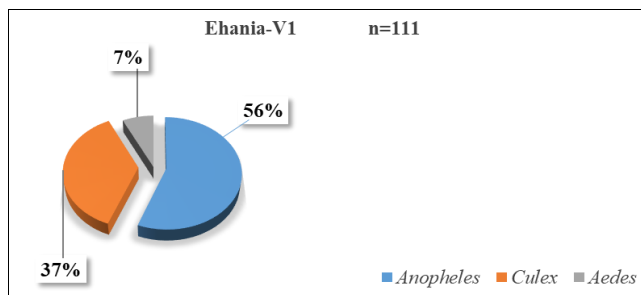
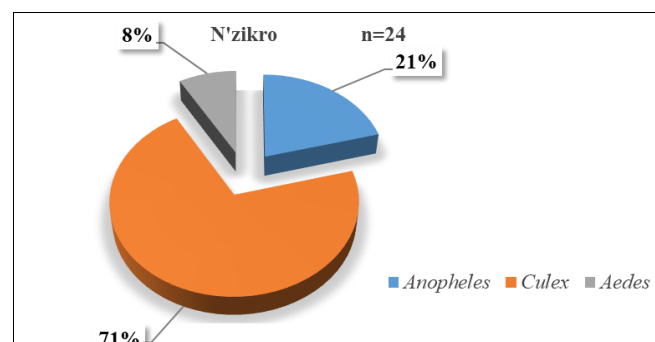
### Adult mosquito fauna using the three capture methods (HLC, WT, and PSC)

Regarding the adult mosquito fauna, the three capture methods allowed for the collection of a total of 3,954 mosquitoes across the three study sites. Four genera were identified following morphological identification of the mosquitoes: 9.43% *Anopheles*, 89.78% *Culex spp.*, 0.53% *Mansonia*, and 0.25% *Aedes*.

**Table 1:** Types of larval habitats inventoried in the villages of N'zikro, Ehania-V1 and Ayebo

	N'zikro	Ehania-V1	Ayebo
	Gender		
Anthropic			
Duck watering			Cx; Ae
Water bucket			An
Tyre tracks	An-Cx		
Gardering hole		An-Cx	
Water reservoir hole			An-Cx
Empty tomato can	An		
Cement drum	Cx; Ae		An-Cx
Cemented well		An	
Unfinished house***		An	
Animal footprints		An	
Construction hole***		An	An
Palm tree hole***			An
Bleeding cap	Cx-Ae		
Naturals			
Swamp			An-Cx
Rainwater collection	Cx	An-Cx	Cx-An
Expanse water		Cx-Ae	
Puddle	An		
Number of breeding sites	6	7	8

An: Anopheles; Cx: Culex; Ae: Aedes; \*\*\*: atypical breeding sites



**Fig 2:** Proportions of Culicidae genera collected following larval surveys in the villages of N'zikro, Ehania-V1 and Ayebo

At the species level, only one species of the genus *Anopheles* was identified: 9.43% *An. gambiae s.l.*; 82.95% *Culex quinquefasciatus*; 1.74% *Culex poicilipex*; 0.63% *Culex fraseri*; and 4.45% *Culex descens*. 0.53% *Mansonia uniformis* and 0.25% *Aedes aegypti*. *An. gambiae s.l.* was the only predominant vector species in all sampled villages (Table 2).

In N'zikro, a total of 1015 mosquitoes belonging to three genera (*Culex* and *Anopheles*, *Aedes*) were collected using the three sampling methods. The PSC method yielded the largest number of *Anopheles* mosquitoes (67 specimens).

In Ehania-V1, a total of 1614 mosquitoes belonging to two genera (*Culex* and *Anopheles*) were collected using the three methods. As in N'zikro, the PSC method yielded the largest number of *Anopheles* mosquitoes (16 specimens).

In Ayebo, a total of 727 mosquitoes belonging to four genera (*Culex*, *Anopheles*, *Mansonia*, and *Aedes*) were identified. Similar to the other agro-ecosystems, the PSC method yielded the largest number of *Anopheles* mosquitoes (218 specimens).

### Resting behaviour of *An. gambiae* by PSC

In N'zikro, 67 female *An. gambiae* were collected while resting in bedrooms. The mean resting density, estimated from the mean number of females per room per day (FRD), was 1.11 FRD [95% CI (0.97–1.26)] for this species.

In Ehania-V1 village of, 16 female *An. gambiae*, representing a resting density of 0.28 FRD [95% CI (0.11–0.44)], were collected in rooms.

In total, 218 female *An. gambiae* were collected while resting in rooms in Ayebo. The mean resting density was estimated at 3.63 FRD [95% CI (2.84–4.42)] for this species. The resting density of *An. gambiae* recorded at N'zikro is statistically lower than the resting density in Ayebo ( $p < 0.05$ ). However, this density is statistically higher than that observed in Ehania-V1 ( $p < 0.05$ ). Furthermore, the resting density of *An. gambiae* at Ehania-V1 is statistically lower than that in Ayebo ( $p < 0.05$ ).

**Average density of *Anopheles gambiae* by WT**

The mean resting density was estimated from the mean number of females per trap per day (FTD). In N’zikro, 11 female *An. gambiae*, representing a mean density of 0.34

FTD [95% CI (0.17–0.52)], were collected per trap window. In Ehania-V1, 4 females of *An. gambiae*, representing a mean density of 0.13 FTD [95% CI (0.04–0.25)], were collected per trap window

**Table 2:** Adultes mosquitos’ fauna collected using the three capture methods in the villages of N’zikro, Ehania-V1, and Ayebo

Species	Nzikro			Ehania-V1			Ayebo			Total
	HLC	WT	PSC	HLC	WT	PSC	HLC	WT	PSC	
<i>Anopheles Gambiae s.l</i>	13	11	67	5	4	16	21	18	218	373
<i>Culex Quinquefasciatus</i>	844	212	265	397	185	978	61	117	221	3280
<i>Poicilipex</i>	0	26	6	0	0	3	1	25	8	69
<i>Fraseri</i>	14	0	3	0		0	3		5	25
<i>Descens</i>	144	0	5	5		16	6		0	176
<i>Mansonia Uniformis</i>	0	0	0	5		0	15		1	21
<i>Aedes Aegypti</i>	0	0	3	0	0	0	0	2	5	10
Total	1015	249	349	412	189	1013	107	162	458	3954

HLC: human landing catches; WT: window trap; PSC: pyrethrum spray catches.

A total of 18 female *An. gambiae* were collected at rest by window traps in Ayebo, representing a mean density of 0.56 FTD [95% CI (0.30–0.82)]. The resting density of *An. gambiae* collected in N’zikro is comparable to that obtained in Ayebo (p = 0.1567). The average density N’zikro is statistically higher than that obtained in Ehania-V1 (p=0.0325). The resting density observed in Ayebo is statistically higher than that obtained in Ehania-V1 (p=0.0042).

**Culicidae nuisance by HLC**

The HLC method allowed for the collection of 1015 mosquitoes of all species in N’zikro, representing a culicid nuisance of 32 bites per person per night (b/p/n) [95% CI (20-43)]. In Ehania-V1, 412 mosquitoes were collected, representing a nuisance of 13 b/ p/n [95% CI (8.7-17)]. In Ayebo, 107 mosquitoes were collected, representing a culicid nuisance of 3.3 b/p/n. The culicid nuisance recorded in N’zikro was significantly higher than those in Ayebo and Ehania-V1 (p=0.0239). The nuisance in Ehania-V1 was significantly higher than that obtained in Ayebo (p=0.006).

**Human biting rate of *Anopheles gambiae*, a major malaria vector**

The captures made in N’zikro yielded 13 specimens of *Anopheles gambiae s.l.*, a major malaria vector. This represents an estimated average HBR of 0.41 b/p/n [95% CI (0.23-0.59)], or 149 b/p/yr. In Ehania-V1, 5 specimens of *An. gambiae* were collected. This represents a HBR of 0.2

b/p/n [95% CI (0.002-0.29)], or 58 b/p/yr. Overall, 21 specimens of *An. gambiae* were collected in Ayebo. The mean HBR is estimated at 0.7 b/p/n [95% CI (0.31-1.0)] for this species, or 241 b/p/yr. Statistical analyses revealed that the HBR in N’zikro was comparable to Ayebo’s (p=0.5259) and higher than that obtained in Ehania-V1 (p=0.0279). Furthermore, the HBR of *An. gambiae* in Ehania-V1 was statistically lower than Ayebo’s (p=0.0100).

**Infestation and entomological inoculation rates of *Anopheles gambiae s.l.***

A total of 280 female mosquitoes belonging to the species *An. gambiae*, collected using three different methods, were tested by the ELISA method. Across the three villages of Aboisso, 12 of the 280 collected female *Anopheles* mosquitoes were infected, representing a mean *P. falciparum* infestation rate of 4.3% [95% CI (1.9–6.7)]. The mean infestation rates of *Plasmodium falciparum* in the localities of N’zikro, Ehania-V1, and Ayebo were 4% [95% CI (4.3–0.12)], 3.3% [95% CI (3.5–0.10)] and 4.4% [95% CI (1.7–7.2)], respectively (Table 3). The mean infestation rates recorded in the three villages were comparable (P = 0.9585). Taking into account the collection methods, using the HLC, one mosquito out of 37 captured was found to be carrying *Plasmodium*, representing a IR of 2.7% [95% CI (2.8–8.2)]. IRs in the villages of N’zikro, Ehania-V1, and Ayebo were 7.70% [95% CI (1-24)] and 0% in the other two villages, respectively (Table 3).

**Table 3:** Infestation and human biting rates of *An. gambiae s.l.* of the three capture methods

Villages	Collection methos									Total	
	Human landing catches			Pyrethrum spray catches			Window trap			N (infested)	S (%)
	N (infested)	S (%)	Ma (b/p/n)	N (infested)	S (%)	Density (FRD)	N (infested)	S (%)	Density (FTD)		
N’zikro	13 (1)	7.7	0.4	10 (0)	0	1.1	2 (0)	0	0.3	25 (1)	4
Ehania-V1	3 (0)	0	0.2	22 (1)	4.5	0.3	5 (0)	0	0.1	30 (1)	3.3
Ayebo	21 (0)	0	0.7	193 (9)	4.7	3.6	11 (1)	9.1	0.6	225 (10)	4.4
Total	37 (1)	2.7	1.6	225 (10)	4.4		18 (1)	5.6		280 (12)	4.3

n: number of tested *Anopheles*  
s: sporozoitic index  
(): number of infested *Anopheles*  
FRD: number of *Anopheles per room per day*  
FTD: number of *Anopheles per trap per day*  
Ma : agressive density

Using the PSC method, 10 *Anopheles gambiae* specimens were found to be infested out of 225 tested, representing an IR of 4.4% [95% CI (1.7-7.2)]. IRs in the villages of N’zikro, Ehania-V1, and Ayebo were 0%, 4.66% [95% CI (1.66-7.66)], and 4.54% [95% CI

(4.91-14)], respectively. The IRs in the two villages were comparable (p=0.9842). Using the window trap method, only one mosquito out of the 18 tested was found to be infested with *Plasmodium*, representing a IR of 5.6% [95% CI (6.2-17)]. In

Ehania-V1, 10% [95% CI (8.66-32)] with an inoculation rate of 0.0308 ib/p/n or 11 ib/p/yr, while no infection was obtained in the other two villages.

## Discussion

Larval surveys have shown that agroecosystems offer a diversity of larval habitats. Regarding habitat types, an abundance of anthropogenic habitats was observed. These results are consistent with those obtained by Etang *et al.*, (2016)<sup>[9]</sup> et Koumba *et al.*, (2018)<sup>[10]</sup> in Gabon and Cameroon, respectively, who also reported that the majority of their identified habitats were artificial, created by human economic and industrial activities. This could be explained by human activities that generate water "containers" which constitute larval habitats for mosquitoes. Several atypical *Anopheles gambiae* habitats were identified in agroecosystems, including unfinished houses, construction pits of abandoned houses, and pits left by oil palm harvesting for palm wine extraction. This demonstrates that human activities can increase the proliferation of larval breeding sites in the environment.

A small number of breeding sites, typically associated with crops in the immediate vicinity of villages, were identified. These include tapping pits (N'zikro) and holes created by uprooting palm trees (Ayebo). With the arrival of the rains, these areas become breeding grounds for *Anopheles* mosquitoes. The impact of human activity on vector proliferation was highlighted in a rubber plantation in Cameroon by Assako Assako *et al.*, (2005)<sup>[11]</sup>.

The inventory of pre-imaginal and adult mosquitoes in the different study villages revealed the composition of the mosquito fauna inhabiting these agricultural villages. This investigation showed that *Anopheles gambiae s.l.* was the only malaria vector identified in the three villages. It also appears that these environments constitute an important reservoir of *Culex* species. Indeed, the abundance of *Culex quinquefasciatus* in the three agricultural villages could be explained by the presence of runoff wastewater. The high presence of this species in these agroecosystems highlights urbanisation leading to surface water pollution from industrial waste, which is conducive to the proliferation of *Culex* (Hougard *et al.* 1993)<sup>[12]</sup>.

The three methods used to capture adults allowed for the collection of the same *Culicidae* species, with a few exceptions, in the study sites. Throughout our investigations in the villages, and despite the use of multiple methods, only a small proportion of *Anopheles* mosquitoes were captured. These results do not align with observations made in rice-growing areas where irrigation systems and rice paddies create environments highly conducive to the proliferation of *Anopheles* mosquitoes, leading to an abundance of these vectors, as was the case in Côte d'Ivoire (Doannio *et al.* 2002; Koudou *et al.* 2007; Betsi *et al.* 2012; Diakité *et al.* 2015)<sup>[13-16]</sup>; and Cameroon (Tanga *et al.* 2011; Bigoga *et al.* 2012)<sup>[5,17]</sup>. In Papua New Guinea, Pluess *et al.*, (2009)<sup>[18]</sup> also demonstrated the influence of palm and rubber plantations on the proliferation of malaria vectors. Within the framework of this study, the low number of *Anopheles gambiae s.l.* species collected inside dwellings could be explained in villages like N'zikro and Ayebo by the scarcity of natural and permanent breeding sites. The majority of these sites are artificial and therefore dependent on human activities and rainfall. They cease to exist when conditions are no longer favourable, consequently reducing the *Anopheles* population during the dry season. Furthermore, the widespread use of insecticides and repellents by the population to combat the increased nuisance of *Culex* mosquitoes, which can be observed by their large numbers in agricultural systems, could explain this low presence. It was noted that *An. gambiae* was found infested with *P. falciparum* and is responsible for malaria transmission in each of the study sites. This study confirms the role of *An. gambiae* in malaria transmission. *An. gambiae* as the main vector of malaria in agricultural areas of Côte d'Ivoire. This role of *An. gambiae* in agricultural areas has already been mentioned by several other authors in other regions of Côte d'Ivoire (Dossou-yovo *et al.* 1995; Doannio *et al.* 2002; Ayala *et al.* 2009; Atangana *et al.* 2012)<sup>[13,19-21]</sup>.

## Conclusion

The nature of the identified larval habitats is not directly linked to crops but rather to human activities carried out on the sites. All of these villages harbour a mosquito fauna dominated by the genus *Culex*. The low presence of the genus *Anopheles* could be due to the scarcity of larval habitats in the villages of N'zikro and Ehania-V1. Nevertheless, this study shows that mosquito infestation is very significant in N'zikro and Ehania-V1, but that the HBR of *Anopheles gambiae s.l.* is low across all sites. However, a high level of *Plasmodium falciparum* vector infestation was observed at each of the sites. The low presence of larval habitats and the scarcity of *Anopheles* species in the study area indicate that malaria could easily be eliminated through effective public awareness campaigns on the need to destroy these habitats.

## References

1. Afrane Y, Klinkenberg E, Drechsel P, Owusu-Daaku K, Garms R, Kruppa T. Does irrigated urban agriculture influence the transmission of malaria in the city of Kumasi, Ghana? *Acta Tropica*,2004;89:125–134. <https://doi.org/10.1016/j.actatropica.2003.06.001>
2. Yadouléon A, N'Guessan R, Allagbé H, Asidi A, Boko M, Osse R, *et al.* The impact of the expansion of urban vegetable farming on malaria transmission in major cities of Benin. *Parasites and Vectors*,2010;3:118. <https://doi.org/10.1186/1756-3305-3-118>
3. Lovell ST. Multifunctional Urban Agriculture for Sustainable Land Use Planning in the United States. *Sustainability*,2010;2:2499–2522. <https://doi.org/10.3390/su2082499>
4. Adja AM, Zoh DD, Sagna AB, Kpan DMS, Guindo-Coulibaly N, Yapi A, *et al.* Diversity of *Anopheles gambiae s.l.*, Giles (Diptera: Culicidae) larval habitats in urban areas and malaria transmission in Bouaké, Côte d'Ivoire. *Vector-Borne and Zoonotic Diseases*,2021;21:593–601. <https://doi.org/10.1089/vbz.2020.2728>
5. Bigoga JD, Nanfack FM, Awono-Ambene PH, Patchoké S, Atangana J, Otia VS, *et al.* Seasonal prevalence of malaria vectors and entomological inoculation rates in the rubber cultivated area of Niéte, South Region of Cameroon. *Parasites and Vectors*,2012;5:197. <https://doi.org/10.1186/1756-3305-5-197>
6. Afrane YA, Lawson BW, Brenya R, Kruppa T, Yan G. The ecology of mosquitoes in an irrigated vegetable farm in Kumasi, Ghana: abundance, productivity and survivorship. *Parasites and Vectors*,2012;5:233. <https://doi.org/10.1186/1756-3305-5-233>
7. Akono PN, Mbida JAM, Tonga C, Belong P, Ngo Hondt OE, Magne GT, *et al.* Impact of vegetable crop agriculture on anopheline aggressivity and malaria transmission in urban and less urbanized settings of the South region of Cameroon. *Parasites and Vectors*,2015;8:293. <https://doi.org/10.1186/s13071-015-0906-2>
8. Sadiakacou CAM, Adja MA, Assi S-B, Poinsignon A, Coulibaly JT, Ouattara AF, *et al.* Seasonal prevalence of *Plasmodium falciparum* infection and use of insecticide-treated nets among children in three agroecosystems in Aboisso, Côte d'Ivoire. *Parasitology Research*,2021;120:3663–3671. <https://doi.org/10.1007/s00436-021-07326-1>
9. Etang J, Pennetier C, Piameu M, Bouraima A, Chandre F, Awono-Ambene P, *et al.* When intensity of deltamethrin resistance in *Anopheles gambiae s.l.* leads to loss of long-lasting insecticidal nets bio-efficacy: a case study in north Cameroon. *Parasites and Vectors*,2016;9:132. <https://doi.org/10.1186/s13071-016-1420-x>
10. Koumba AA, Koumba CRZ, Nguema RM, Djogbenou LS, Ondo PO, Ketoh GK, *et al.* Distribution spatiale et saisonnière des gîtes larvaires des moustiques dans les espaces agricoles de la zone de Mouila, Gabon. *International*

- Journal of Biological and Chemical Sciences,2018;12:1754–1769. <https://doi.org/10.4314/ijbcs.v12i4.19>
11. Assako Assako RJ, Bley D, Simard F. Apports des sciences sociales et de l'entomologie dans l'analyse de l'endémicité du paludisme à HEVECAM, une agro-industrie du Sud-Cameroun. 2005.
  12. Hougard J-M, Mbentengam R, Lochouarn L, Escaffre H, Darriet F, Barbazan P, *et al.* Lutte contre *Culex quinquefasciatus* par *Bacillus sphaericus*: résultats d'une campagne pilote dans une grande agglomération urbaine d'Afrique équatoriale. Bulletin of the World Health Organization,1993;71:367.
  13. Doannio JMC, Dossou-Yovo J, Diarrassouba S, Rakotondraibé ME, Chauvancy G, Chandre F, *et al.* Dynamics of malaria transmission in Kafiné, a rice growing village in a humid savannah area of Côte d'Ivoire. Bulletin de la Société de Pathologie Exotique,2002;95:11–16.
  14. Koudou BG, Adja AM, Matthys B, Doumbia M, Cissé G, Koné M, *et al.* Agricultural activities and malaria transmission in two eco-epidemiological settings in central Côte d'Ivoire. Bulletin de la Société de Pathologie Exotique,2007;100:124–126.
  15. Betsi AN, Tchicaya ES, Koudou BG. High proliferation of *Anopheles gambiae* and *Anopheles funestus* larvae in irrigated and non-irrigated rice fields in the Western forest region of Côte d'Ivoire. Bulletin de la Société de Pathologie Exotique,2012;105:220–229. <https://doi.org/10.1007/s13149-012-0219-z>
  16. Diakité NR, Guindo-Coulibaly N, Adja AM, Ouattara M, Coulibaly JT, Utzinger J, *et al.* Spatial and temporal variation of malaria entomological parameters at the onset of a hydro-agricultural development in central Côte d'Ivoire. Malaria Journal,2015;14:340. <https://doi.org/10.1186/s12936-015-0871-4>
  17. Tanga MC, Ngundu WI, Tchouassi PD. Daily survival and human blood index of major malaria vectors associated with oil palm cultivation in Cameroon and their role in malaria transmission. Tropical Medicine and International Health,2011;16:447–457. <https://doi.org/10.1111/j.1365-3156.2011.02726.x>
  18. Pluess B, Mueller I, Levi D, King G, Smith TA, Lengeler C. Malaria a major health problem within an oil palm plantation around Popondetta, Papua New Guinea. Malaria Journal,2009;8:56. <https://doi.org/10.1186/1475-2875-8-56>
  19. Dossou-Yovo J, Doannio JM, Rivière F, Chauvancy G. Malaria in Côte d'Ivoire wet savannah region: the entomological input. Tropical Medicine and Parasitology,1995;46:263–269.
  20. Ayala D, Costantini C, Ose K, Kamdem GC, Antonio-Nkondjio C, Agbor J-P, *et al.* Habitat suitability and ecological niche profile of major malaria vectors in Cameroon. Malaria Journal,2009;8:307. <https://doi.org/10.1186/1475-2875-8-307>
  21. Atangana J, Fomena A, Tamesse JL, Fondjo E. Pratiques agricoles et épidémiologie du paludisme en zone soudano-sahélienne du Cameroun. Bulletin de la Société de Pathologie Exotique,2012;105:23–29. <https://doi.org/10.1007/s13149-011-0202-4>