Mosquito (Diptera: *Culicidae*) Larval Ecology in Rubber Plantations and Rural Villages in Dabou (Côte d'Ivoire)

ABSTRACT:

In Côte d'Ivoire, rubber cultivation has more than doubled since 2010. These mass agricultural areas require a large workforce with little information on how this environment might impact risk of mosquito-borne diseases. The objective of this study **was to assess the larval ecology of mosquitoes in rubber areas of Dabou, Côte d'Ivoire**.

From January to June 2017, an entomological survey was conducted of mature (MP) and immature (IP) rubber plantations, as well as in villages surrounded by rubber plantations (SV) and remote from rubber plantations (RV). The number and type of potential and positive breeding sites were recorded, and mosquito larval densities and diversity were estimated.

Seven genera divided into 31 species including major vector such as *Anopheles gambiae* s.l. and *Aedes aegypti*, were identified. A total of 1,660 waterbodies were identified with a larvae positivity rate of 63.1 %. A majority of waterbodies were identified in SV (N=875, 53.4 % positivity rate), followed by MP (N=422, 81.8 % positivity rate), IP (N=194, 72.2 % positivity rate) and least in RV (N=169, 57.4 % positivity rate). The most important breeding sites for disease vectors were leaf axils in IP **(N=108, 77.1%)**, latex collection cups in MP **(N=332, 96.2%)** and the containers abandoned in the SV **(N=242, 51.8%)** as well as in the RV **(N=59, 60.8%).**

All these results allow us to affirm that the cultivation of rubber trees has an impact on the larval ecology by increasing the number of available sites and favoring a high larval density and diversity.

Introduction

Among hematophagous arthropods, *Culicidae* are the most formidable in terms of numbers, nuisance and diseases that they transmit (Halbach et al. 2017)**.** Many mosquito species are vectors of pathogens that cause millions of deaths per year, adding to the global economic and health care burden (World Health Organization 2014). The genera *Anopheles*, *Aedes* and *Culex* are the most important disease vectors (Manguin 2013). *Aedes* species are vectors of arboviral diseases, such as yellow fever, dengue, chikungunya, Rift Valley fever, and Zika that are of considerable public health relevance (Liang et al. 2015)**.** A recent study by Weetman et al. 2018 estimated that 831 million people, 70% of them in Africa, would be exposed to arboviruses transmitted by mosquitoes of the genus *Aedes*. The occurrence of an epidemic is conditioned by the pre-existence or introduction of the pathogen or vector in an area, the presence of susceptible (host) populations and climatic and environmental conditions conducive to vector outbreaks (Mariner 2019). Recent studies in Africa have reported the expansion of *Aedes aegypti* in Ghana, Mozambique, and Namibia, and *Ae. albopictus* in Mali, Morocco, Mozambique, São Tomé and Príncipe (Weetman et al. 2018), and Côte d'Ivoire (Konan et al. 2013). Recent reported arbovirus epidemics have included Rift Valley Fever in Niger Senegal and Mali, yellow fever and dengue fever in 2018 in Burkina Faso, Côte d'Ivoire and Senegal (Sow 2018). Moreover, despite the many efforts made by the international community to fight malaria, African countries remain endemic to this parasitosis transmitted by *Anopheles* mosquitoes. The number of malaria cases worldwide is estimated at 228 million with 405,000 deaths in 2018. The WHO African Region continues to bear a disproportionate share of the global malaria burden with 93% (213 million) of cases and 94% (381,000) of deaths due to malaria (WHO, 2019). Also, 39 African countries in the WHO region totalling 405 million habitats are endemic to lymphatic filariasis transmitted by species of the genus *Culex* (WHO 2009) and *Anopheles* (De Souza et al. 2012).

The resurgence of these mosquito-borne diseases and their geographic expansion has long been associated with human-induced landcover change (Leisnham and Juliano 2012). Land use changes for human benefit can have a large impact on mosquito species diversity and human dynamics (Lee et al. 2020). In many tropical regions, mosquitoes are particularly problematic and widespread in agricultural landscapes (Tangena et al. 2017 ; Zahouli et al. 2017). Disease transmission can be exacerbated by land use changes due to alteration to the biophysical environment, with implications for the dispersal of mosquitoes between landscapes, hence modifying vector ecology patterns and disease transmission dynamics (Patz et al. 2004). In recent decades, the forest areas of Côte d'Ivoire have undergone major changes in favour of the expansion of rubber cultivation. Planted areas have increased from 318,000 hectares in 2010 to 650,000 hectares in 2018 (APROMAC, 2019), an increase of more than 40,000 hectares per year. All these transformations that modify the natural habitat are likely to change the behavior of the vectors and promote the emergence or re-emergence of vector-borne diseases in these rubber-growing areas, as has been the case in Southeast Asia. Indeed, in Southeast Asia, where rubber tree cultivation is the most widespread, vectors of malaria (*An. Dirus* s.l. and *An. Minimus* s.l.) (Yasuoka and Levins 2007) and arbovirus (*Ae. Albopictus*) (Paily et al. 2013) have adapted to rubber tree landscapes, leading to outbreaks of malaria (Guyant et al. 2015), dengue and Chikungunya (Sensing and Control 2014).

Côte d'Ivoire is experiencing waves of arbovirus epidemics threatening the health security of the population. This is due to the abundance of vectors, presence of arboviruses and non-immune populations. In 2008, a double epidemic of dengue virus type 3 and yellow fever with the vector *Aedes aegypti* was declared in Côte d'Ivoire (WHO 2008). Since then, these arboviruses have continued to appear in the Ivorian population. The latest to date are those of 2019, when 89 cases and 1 death due to yellow fever and 300 cases and 2 deaths due to dengue fever were reported (ECOWAS 2019 ; Fofana et al. 2019). As for malaria, it is the first reason for consultations in health facilities and is the cause of hospitalization in 32% of cases in pregnant women and 62% of cases in children under 5 years old (WHO 2013). Transmission is ensured by three main vectors: *An. gambiae* s.s., *An. funestus* s.s., and *An. nili* s.s., (Koffi et al. 2009 ; Adja et al. 2011).

In the absence of specific treatment and vaccination against most of the infectious agents transmitted by Culicidae, vector control remains the most important means of control and prevention (**Diallo** 2018). The main control method used to reduce the burden of malaria in Côte d'Ivoire is the use of long-lasting insecticidal nets (LLINs), whose effectiveness depends on biting behavior and vector susceptibility to insecticide treatment (WHO 2018) . In addition, strategies to control and prevent arbovirus outbreaks in Côte d'Ivoire are based on vector surveillance and vector control interventions, including elimination of mosquito breeding sites through source reduction measures (destruction, modification, removal, covering or recycling of non-essential containers that provide habitat for larvae) and fumigation to eliminate adult mosquitoes (WHO 2010 ; WHO 2017). Even though the population in rural areas are at equally high risk of mosquito-borne diseases, vector control campaigns remain concentrated in urban areas, particularly in Abidjan. For vector control in rural areas to be successful, it must be adapted to the local environment, i.e. prior knowledge of the ecology of these vectors is essential (WHO 2014). Little is known about the larval ecology of mosquitoes in these expanding rubber-growing areas in Côte d'Ivoire. However, in addition to potential natural breeding sites, the presence of latex collection cups, the microclimate due to the canopy and the vegetation could favor a high density and diversity of mosquito larvae in rubber growing areas. Knowledge of the larval ecology of mosquitoes in these rubber-growing areas could help improve the control of mosquito-borne diseases in these localities. Thus, the objective of this study was to assess the impact of rubber plantations on mosquito larval density and diversity.

Materials and methods

Study area

The work was carried out in the department of Dabou, which is located about 40 km from the city of Abidjan, the Ivorian economic capital. The population of Dabou is estimated at more than 884,300 (I.N.S. 2015). About twenty years ago, the region still possessed several thousand hectares of dense rainforest. However, due to agricultural development and urbanization, these forests have practically disappeared. Landcover is now dominated by rural villages and industrial rubber plantations. A few old oil palm and cocoa plantations are also present in this rubber-dominated landscape. The climate of the Dabou region is sub-equatorial, hot and humid with four seasons: a long rainy season from April to July and a short rainy season from mid-September to November. The two rainy seasons are separated by a large dry season from December to March and a small dry season from August to mid-September (Kouadio et al. 2003). The average annual rainfall is around 1,400 mm, with average temperatures varying between 25 and 26 °C (Kouadio et al. 2003).

Samples were collected in four types of habitats in Dabou department, including: (i) immature rubber plantations (IP: young rubber plants not yet tapped for latex, less than 5 years old), (ii) mature rubber plantations (MP: rubber plants tapped for more than 10 years), (iii) villages surrounded by rubber plantations (SV), and (iv) villages remote from plantations (RV). For each of these four habitat types, three replicates of seven hectares each were selected in different localities (Table I). These locations were at least 5 km apart (Figure 1). The selection of rubber plantations was guided by size (at least seven hectares), age (3-5 years for immature plantations and more than 10 years for mature plantations) and, for mature rubber plantations, regularity of human activities (still actively tapped for latex every week). For villages, they were selected based on the distance from the rubber plantations. Surrounding villages should be in close proximity to the rubber plantations with at least three within 300 m of them. Remote villages should be at least 2 km from the rubber plantations. The remote villages were the control habitat for the study and were in the same locality of Lopou. These controls were used to compare mosquito diversity in rural villages surrounded by rubber plantations.

Larval survey

Surveys of breeding sites were carried out in the previously described four habitat types, each consisting of three replicates (N = 12 survey sites). All survey sites were entirely surveyed for waterbodies once every month from January to March 2017 (dry season) and from May to June 2017 (rainy season).

In each of the 12 survey sites, all waterbodies were identified and recorded. Waterbodies were classified into one of the following waterbody types: 1 : cut bamboo (In cultivation or used to make chairs or sheds), 2 : leaf axil (banana : *Musa paradisiaca*, pineapple : *Ananas comosus*, taro : *Colocasia esculenta*), 3 : abandoned containers (cut cans, used tires, coconuts, plastic and glass bottles, broken seals, canaries), 4 : water container (canaries, basins, drums, plastic and iron barrels, cisterns), 5 : puddle/tyre track, 6 : latex collection cup (with latex, without latex), 7 : tree trunks (tree growing or cut down), 8 : ditch (gutter and pit left after soil is removed for house construction), 9 : leaf puddle, 10 : roof tarpaulins (bag used to cover the roof of the houses), 11 : snail shell and 12 : water meter cover (Figure 2).

The presence or absence of immature mosquitoes was determined, and immature mosquitoes were collected using standard ladles (250 ml), sieves and pipettes. Ladles were used in larger bodies of water. In these bodies of water, sampling consisted of about ten dipping's at several locations in the waterbody with 30 seconds in between dips in order to maximize captures (Sy et al. 2016). Small, shallow impoundments were sampled using sieves and/or pipettes.

The immature mosquitoes were transported to the field laboratory where they were counted and sorted by genera. The immature stages were reared until the emergence of adult mosquitoes. The adult mosquitoes were identified morphologically under a binocular magnifying glass using the Ethiopian mosquito identification keys (Edwards 1941 ;.Gillies and Coetzee 1987; Huang 2004).

Data analysis

The ecological compositional indices represented by relative abundance, frequency and larval density were calculated. Relative abundance was both the number of larvae in a habitat or site type and the number of sites in a habitat type. Frequency was equal to the percentage of the number of positive sites over the number of sites in water. Larval density was equal to the number of larvae over the number of water sites. Generalized Linear Models (GLM) were used to estimate differences in larval densities across habitats, waterbody types and seasons using a negative binomial model with a Log linkage function (IBM SPSS statistics, version 20).

Results

Diversity

The identification of adult mosquitoes after their emergence revealed the presence of seven genera (*Aedes, Culex, Anopheles, Eretmapodites, Ficalbia, Toxorhynchites* and *Uranotaenia*) divided into 31 species including the important malaria and filariasis vector (*Anopheles gambia*e s.l.), two arbovirus vectors (*Aedes aegypti* and *Aedes africanus*) and a urbain filariasis vector (*Culex quinquefasciatus*) (Table II). The distribution of species richness in the different habitats was as follows: 20 species in immature rubber plantations, 25 species in both mature rubber plantations and surrounding villages, and 16 species in remote villages.

Larvae of the potential malaria and filariasis vector (*Anopheles gambiae* s.l.) were collected only in the rural villages, both surrounded by and away from rubber plantations. In the surrounded villages, they were collected from gutters, water storage drums, and abandoned containers, while in the remote village they were found only in abandoned containers (Figure 3). Immature stages of the potential urban filariasis vector (*Culex quinquefasciatus*) were collected from all habitats in the study except immature rubber tree plantations. Larval densities of *Culex quinquefasciatus* were statistically different from one habitat to another (Table III). In the mature plantations, *Culex quinquefasciatus* were mostly collected from latex collection cups, while abandoned containers and water storage drums were the most important waterbodies in the surrounded villages. In the remote village, they predominated in the abandoned containers.

Larvae of the potential arbovirus vector *Aedes aegypti*, were collected in all four habitats, with higher larval densities in the surrounded villages (16 larvae/positive waterbody) and mature plantations (9 larvae / positive waterbody) compared to remote villages (GLM, P < 0,001). In mature plantations, this vector reproduces mainly in latex collection cups, whereas in immature plantations it reproduces in leaf axils. In surrounded villages, *Aedes aegypti* larvae were collected in leaf axils, snail shells, cut bamboo, water storage drums and predominantly in abandoned containers. In the remote village, larvae of *Aedes aegypti* were predominantly collected from abandoned containers.

Abundance and typology of waterbodies

A total of 1,660 potential mosquito breeding sites were identified in this study, of which 63.2% (1049/1660) contained at least one mosquito larva or nymph (Table IV). Waterbodies positive for mosquito larvae were mostly found in the villages surrounded by rubber plantations, with a positivity rate of 53.4% (467/875). The second highest number of larval breeding sites were found in the mature plantations with 81.7% (345/422) positive sites, followed by the immature plantations with 72.2% (140/194) positive sites, and those in the village away from plantations with 57.4% (97/169) positive sites. The bodies of water were grouped into 12 types of waterbody, including five natural (bamboo hollows, leaf axils, waterlogged leaf, tree trunk and snail shell) and seven artificial (abandoned container, storage drum, water puddle/tire track, latex collection cup, gutter/pit, roof tarp and water meter cover; Figure 2). Overall, there were more artificial waterbodies positive for mosquitoes, with a positivity rate of 60.0% (806/1344) than natural waterbody with a positivity rate of 42.1 % (243/577). Specifically, the most abundant artificial waterbody types were latex collection cups with a positivity rate of 85.8% (347/404) and abandoned containers with a 58.7% positivity rate (313/536).

The most common types of waterbodies per habitat were leaf axils in immature plantations (74.5% positive deposits (108/145)), latex collection cups in mature plantations (85.6 positive waterbodies (332/388)), abandoned containers (61.6% positive waterbodies (242/396)) and water storage drums

(115/320)) in the villages with rubber plantations and abandoned containers in the village away from rubber plantations (52.2% positive waterbodies (59/113).

Larval productivity

Mature plantations and the surrounding villages were the most productive habitats, with an average of 36 larvae per positive waterbody (Table V). For the cumulative potential vectors (*Aedes aegypti*, *Culex quinquefasciatus* and *Anopheles gambiae* s.l), the highest larval productivity was observed in mature plantations **(14.2(11.9-17.0) larvae)** (Table VI). It was followed by those in surrounded villages **(9.5 (8.6-10.5) larvae)**, remote villages (**8.7 (7.4-10.2) larvae)** and immature plantations **(4.6(2.9-7.1) larvae)**. The difference between the larval densities of the potential vectors collected in remote and surrounded villages was not significant (GLM, P = 0,342).

In immature plantations, the most productive waterbody types were tree trunks (69.6 (33.0-146.7) larvae/waterbody) (Table VII). Larval productivity was 1,6 times higher for tree trunks compared to abandoned containers and water storage barrels but is not significant (GLM, P = 0,062). In mature rubber plantations latex collection cups were the most productive type of waterbody (37.4 (33.8-41.4) larvae/waterbody) with a larval density three times higher than that of all other types of waterbodies (GLM, P = 0,001). The most productive waterbodies in surrounded villages were roofing tarpaulins (2372.0 (321.0-28071.3) larvae/waterbody), followed by latex collection cups (133.8 (66.8-267.9) larvae/waterbody). Water storage drums (42.0 (27.0-65.4) larvae/waterbody), water meter lids (30 (4.1-220.0) larvae/waterbody), and abandoned containers (22.4(18.6-27.1) larvae/waterbody) have been the most productive waterbodies in remote villages.

Seasonality

Larval densities were higher in the dry season than in the wet season for all habitats except immature rubber plantations (Table VIII). In the dry season, the larval densities per waterbody observed in the surrounded villages (50.8 (44.8-57.5) larvae/waterbody), remote villages (38.6 (23.5-53.3) larvae/positive waterbody) and mature plantations (47.8 (42.3-54.0) larvae /waterbody) were between 0,8 and 1,1 times higher than those in the immature plantations (9.2 (7.0-12.1) larvae/waterbody). Similarly, during the rainy season the larval density per positive waterbody in immature plantations (11.4 (9.2-14.1) larvae/waterbody) was lower than in the other three habitats. The densities observed in both village types were similar for both seasons (GLM, P > 0,260).

As for the disease vectors *Aedes aegypti* and *Culex quinquefasciatus,* their larval densities were higher in the dry season, while *Anopheles gambiae* s.l. densities were higher in the rainy season (Table IX).

Discussion

This study assessed the impact of rubber plantations on the species richness and larval abundance of mosquitoes in rubber plantation areas of Dabou. It revealed the presence of an important culicidal fauna composed of seven genera divided into 31 species, including two major vectors of infectious agents (*Anopheles gambiae* s.l. and *Aedes aegypti*). The 31 species identified in this study are representative of the habitats studied. A previous study conducted in two rural villages of Dabou (Fofana et al. 2010) identified a lower species richness of 16 species belonging to five genera using the larval survey method. This high diversity of species collected in the present study could be due to the favorable ecological conditions offered by the environment of the rubber plantations studied to the immature stages of mosquitoes. This hypothesis is supported by the difference in the number of species collected in the two types of villages. In villages surrounded by rubber plantations, 25 species were collected compared to 16 species in villages away from rubber plantations. Indeed, the combination of different ecological and climatic factors that shaped the rubber plantation areas provided specific sites for the development of various pre-imaginal populations of each species: storage containers, abandoned containers, tree trunks, rubber tapping cups, and leaf axils. Similarly, larval densities were higher in villages surrounded than in villages distant from rubber plantations. The relatively higher species richness and larval density observed in villages surrounded by rubber plantations was due to the proximity of these villages to the rubber plantations. This assertion is in agreement with previous studies (Mayana et al. 2014; Golding et al. 2015) that stated that the specific composition of mosquito populations is influenced by the immediate environment.

Anopheles gambiae s.l., a major vector of malaria in Côte d'Ivoire (Zoh et al. 2020 ; Assouho et al. 2020) and rural filariasis in West Africa (De Souza et al. 2012) was only weakly collected in both types of villages with relative dominance in villages surrounded by rubber plantations. The low density of this species during the present study could be due to the scarcity of favorable developmental sites for immature stages. As for Aedes aegypti, the main arbovirus vector responsible for the recent dengue and yellow fever epidemics in Côte d'Ivoire (Fofana et al. 2019), it was identified both in rubber plantations and in villages. This presence of Aedes aegypti in all four habitat types is due to the fact that its larvae adapt to both natural roosts, such as tree holes, and artificial roosts, such as latex collection cups (Xia et al. 2021). Overall, immature stages of Aedes aegypti colonized peri-domestic roosts, specifically abandoned containers in villages and latex collection cups in rubber plantations, and domestic roosts, including water storage containers. These observations are consistent with previous results reported in Cameroon (Tedjou et al. 2020) and Asia (Hammond et al. 2007). Also, larval densities of Aedes aegypti were higher in habitats in the rubber-growing area than in villages far from the rubber plantations. This is simply because latex collection cups, abundantly present in rubber-growing areas, retain water as well as leaves and become favorable sites for the development of immature stages of this vector. These observations raise the question of the management of latex collection cups in rubber plantations. These cups become water bodies rich in organic matter. Yet, the presence of organic matter in Aedes aegypti larval habitats could serve as a food resource (Barrera et al. 2006 ; Kamgang et al. 2013) or as a micro-habitat to hide and avoid predators (Cox et al. 2007). These observations concur with those of Joly et al. (2017) who note that the vegetative cycle and functioning of rubber trees have aspects favorable to mosquito development. A simple intervention would be to keep latex collection cups and store them in waterproof shelters when not in use for extended periods. Rain covers for these cups, which would prevent water from seeping into the latex collection cups, could also be used to control larvae.

In addition, the study found 12 types of mosquito breeding sites throughout the study locations. The most numerous and most colonized breeding sites for immature stages of mosquitoes were leaf axils in immature rubber plantations, latex collection cuts in mature rubber plantations, and storage containers and abandoned containers in villages. The abundance of storage containers and abandoned drums in villages is consistent with findings in many rural and urban locations in Africa. Poorly controlled urbanization, lack of appropriate hygiene measures, and lack of sanitation around huts in most villages are responsible for the high incidence of mosquito breeding sites (Rodhain et al. 1985 : (Carnevale and Robert 2009 ; Bouabida et al. 2012). Overall, mosquito larvae production was greater in the dry season than in the wet season. This decrease in larval density in the wet season could be explained by the fact that during the wet season, the heavy rains prevalent in the study area cause water bodies to overflow. This observation contrasts with Koumba et al. (2018) who observed greater abundance during the rainy season in agricultural areas in the Mouila region of Gabon. Vector control programs should take this observation into account so that they no longer focus solely on the rainy seasons for control. All these results allow us to affirm that the cultivation of rubber trees has an impact on the larval ecology by increasing the number of available sites and favoring a high larval density and diversity.

This information is of significant importance for the implementation of a vector control strategy according to the specificities of each habitat.

Conclusion

The large variations in breeding sites for mosquitoes in the rubber-growing areas have been identified in this study. Especially latex collection cups and artificial containers (garbage) are important breeding sites for mosquitoes, including disease vector species. Certain practices that accompany the cultivation of rubber trees in their immature and mature state contribute to a consequent increase in the presence of mosquito vector breeding sites in these landscapes. The abundance of collection cups, their high larval density and species richness could increase Culicidian nuisances and the risk of arbovirus epidemics in this rubber tree farming area, especially during the dry season. However, further studies on the epidemiology of the mosquito-borne diseases, the physico-chemical composition of the contents of latex collection cups, adult mosquitoes and other rubber tree growing areas of Côte d'Ivoire should be carried out in order to provide additional information necessary for the improvement of available vector control methods.

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Table 1: Location of collection sites

Localities	Habitats	GPS coordinates	
		Latitudes	**Longitudes**
Kotokodji	immature rubber plantations	05°26’354	004°36’437
	mature rubber plantations	05°26’193	004°36’370
	villages surrounded	05°26'354	004°36'437
Adangba-eby	immature rubber plantations	05°26’787	004°35’686
	mature rubber plantations	05°26’889	004°35’561
	villages surrounded	05°26'976	004°35'388
Agnimangbo	immature rubber plantations	05°28’220	00 4°34’590
	mature rubber plantations	05°28’300	00 4°35’600
	villages surrounded	05°27'879	004°34'749
Lopou	villages remote 1	05°25'130	004°28'105
	villages remote 2	05°25'321	004°28'194
	villages remote 3	05°25'300	004°28'117

Table II: Specific richness of mosquitoes in rural areas of Dabou, Côte d'Ivoire

Species	Immature Plantation (N)	Mature Plantation (N)	Surrounded village (N)	Remote Village (N)	Total (N)
Aedes aegypti	2,4 (110/4468)	29,9 (1335/4468)	61,6 (2751/4468)	6,1 (272/4468)	**100 (4468/4468)**
Aedes africanus	13,4 (13/97)	66,0 (64/97)	20,6 (20/97)	0 (0/97)	**97**
Aedes apicoargenteus	0 (0/5)	100 (5/5)	0 (0/5)	0 (0/5)	**100 (5/5)**
Aedes cumminsii	0 (0/3)	100 (3/3)	0 (0/3)	0 (0/3)	**100 (3/3)**
Aedes denderensis	1,0 (4/404)	83,2 (336/404)	15,6 (63/404)	0,2 (1/404)	**100 (404/404)**
Aedes lilii	91,0 (10/11)	0 (0/11)	9,1 (1/11)	0 (0/11)	**100 (11/11)**
Aedes metallicus	44,6 (108/242)	9,1 (22/242)	40,9 (99/242)	5,4 (13/242)	**100 (242/242)**
Aedes simpsoni	35,1 (65/185)	9,7 (18/185)	44,9 (83/185)	10,3 (19/185)	**185**
Aedes subargenteus	44,1 (15/34)	20,6 (7/34)	35,3 (12/34)	0 (0/34)	**100 (34/34**
Anopheles gambiae s.l.	0 (0/93)	0 (0/93)	98,9 (92/93)	1,1 (1/93)	**100 (93/93)**
Culex albiventris	0 (0/8)	62,5 (5/8)	37,5 (3/8)	0 (0/8)	**100 (8/8)**
Cx bitaeniorhynchus	0,5 (2/365)	10,1 (37/365)	70,1 (256365)	19,2 (70/365)	**100 (365/365)**
Culex cinereus	0,6 (7/1152)	53,2 (613/1152)	26,4 (304/1152)	19,8 (228/1152)	**100 (1152/1152)**
Culex decens	0 (0/17)	82,3 (14/17)	11,8 (2/17)	5,9 (1/17)	**100 (17/17)**
Culex ethiopiensis	0 (0/37)	0 (0/37)	100 (37/37)	0 (0/37)	**100 (37/37)**
Culex horridus	0,6 (7/1134)	26,8 (304/1134)	15,3 (173/1134)	57,3 (650/1134)	**100 (1134/1134)**
Culex lacticinctus	0 (0/7)	14,3 (1/7)	71,4 (5/7)	14,3 (1/7)	**100 (7/7)**
Culex nebulosus	0,7 (18/2623)	33,5 (879/2623)	33,2 (871/2623)	32,5 (855/2623)	**100 (2623/2623)**
Culex quinquefasciatus	0 (0/3048)	15,9 (484/3048)	45,1 (1376/3048)	39,0 (1188/3048)	**100 (3048/3048)**
Culex rubinotus	8,3 (36/430)	83,3 (358/430)	5,6 (24/430)	2,8 (12/430)	**100 (430/430)**
Culex tigripes	0,0 (1/256)	12,5 (32/256)	73,8 (189/256)	13,3 (34/256)	**100 (256/256)**
Culex univittatus	3,6 (2/55)	0 (0/55)	61,8 (34/55)	34,5 (19/55)	**100 (55/55)**
Eretmapodites chrysogaster	6,5 (73/1120)	83,6 (936/1120)	9,8 (110/1120)	0,1 (1/1120)	**100 (1120/1120)**
Eretmapodites inornatus	50,0 (2/4)	50,0 (2/4)	0 (0/4)	0 (0/4)	**100(4/4)**
Eretmapodites quinquevittatus	13,0 (3/23)	78,3 (18/23)	8,7 (2/23)	0 (0/23)	**23**
Ficalbia sp	100 (23/23)	0 (0/23)	0 (0/23)	0 (0/23)	**100 (23/23)**
Toxorhynchites brevipalpis	8,3 (1/12)	75,0 (9/12)	16,7 (2/12)	0 (0/12)	**100 (12/12)**
Toxorhynchites conradi	0 (0/3)	100 (3/3)	0 (0/3)	0 (0/3)	**100 (3/3)**
toxorhynchites evansea	0 (0/11)	90,9 (10/11)	9,1 (1/11)	0 (0/11)	**100 (11/11)**
Toxorhynchites sp	0 (0/45)	80,0 (36/45)	20,0 (9/45)	0 (0/45)	**45**
Uranotaenia sp	100 (6/6)	0 (0/6)	0 (0/6)	0 (0/6)	**100 (6/6)**
Number of species	**20**	**25**	**25**	**16**	**31**

Table III: Larval density of the main potential vectors in four rubber-bearing facies of the Dabou region (Côte d'Ivoire)

Vectors putatives	Habitats	N	Average number of larvae/Positive waterbody (CI 95%)	OR	P
Aedes aegypti	Immature Plantation	110	4,6 (2,9 - 7,1)	1,8	0,027
	Mature plantation	1335	16,1 (12,9 - 20,1)	6,1	<0,001*
	Villages Surrounded	2751	8,7 (7,7 - 9,7)	3,3	<0,001*
	Villages remote	272	2,6 (2,0 - 3,3)	1	
Culex quinquefasciatus	Mature plantation	484	10,8 (7,9 - 14,6)	0,6	0,003*
	Villages Surrounded	1376	12,0 (9,9 - 14,5)	0,6	0,004*
	Villages remote	1188	18,9 (14,6 - 24,3)	1	
Anopheles gambiae s.l.	Villages Surrounded	92	7,8 (4,3 - 14,3)	7,8	0,155
	Villages remote	1	1,0 (0,1 - 15,9)	1	

N: number of larvae; **CI**: 95% confidence interval; **P**: P value; **OR**: order ratio;* results are significant

Table IV: Immature mosquito positivity rate for the different waterbody types found in the four different habitats.

	Immature Plantation		Mature Plantation		Village Surrounded		Village Remote		Total	
waterbody type	**positivity rate (N)**	**overall positivity rate (N)**	**positivity rate (N)**	**overall positivity rate (N)**	**positivity rate (N)**	**overall positivity rate (N)**	**positivity rate (N)**	**overall positivity rate (N)**	**positivity rate (N)**	**overall positivity rate (N)**
Cut bamboo*	0 (0/0)	0 (0/140)	0 (0/2)	0 (0/345)	88,9 (8/9)	1,7 (8/467)	0 (0/0)	0 (0/97)	72,7 (8/11)	0,8 (8/1049)
leaf axils*	74,5 (108/145)	77,1 (108/140)	80 (4/5)	1,2 (4/345)	79,4 (81/102)	17,3 (81/467)	91,3 (21/23)	21,6 (21/97)	77,8 (214/275)	20,4 (214/1049)
Tree tunks*	100 (7/7)	5,0 (7/140)	0 (0/0)	0 (0/345)	0 (0/3)	0 (0/467)	0 (0/0)	0 (0/97)	70 (7/10)	0,7 (7/1049)
Dead leaves*	78,6 (11/14)	7,9 (11/140)	0 (0/0)	0 (0/345)	33,3 (1/3)	0,2 (1/467)	100 (1/1)	1,0 (1/97)	72,2 (13/18)	1,2 (13/1049)
Snail shell*	0 (0/0)	0 (0/140)	0 (0/0)	0 (0/345)	100 (1/1)	0,2 (1/467)	0 (0/1)	0 (0/97)	50 (1/2)	0,1 (1/1049)
Abandoned containers	55,6 (5/9)	3,6 (5/140)	38,9 (7/18)	2,0 (7/345)	61,1 (242/396)	51,8 (242/467)	52,2 (59/113)	60,8 (59/97)	58,4 (313/536)	29,8 (313/1049)
Water storage drum	25,0 (3/12)	2,1 (3/140)	50 (1/2)	0,3 (1/345)	35,9 (115/320)	24,6 (115/467)	60 (12/20)	12,4 (12/97)	36,9 (131/355)	12,5 (131/1049)
Water puddle	50 (1/2)	0,7 (1/140)	25,0 (1/4)	0,3 (1/345)	0 (0/16)	0 (0/467)	0 (0/1)	0 (0/97)	8,7 (2/23)	0,2 (2/1049)
Latex collection cup	100 (3/3)	2,1 (3/140)	85,6 (332/388)	96,2 (332/345)	100 (10/10)	2,1 (10/467)	66,7 (2/3)	2,1 (2/97)	85,8 (347/404)	33,1 (349/1049)
Gutter and ditch	0 (0/0)	0 (0/140)	0 (0/2)	0 (0/345)	100 (3/3)	0,6 (3/467)	50 (1/2)	1,0 (1/97)	57,1 (4/7)	0,4 (4/1049)
Roof tarpaulins	100 (2/2)	1,4 (2/140)	0 (0/0)	0 (0/345)	50 (5/10)	1,1 (5/467)	0 (0/4)	0 (0/97)	43,8 (7/16)	0,7 (7/1049)
Water meter cover	0 (0/0)	0 (0/140)	0 (0/0)	0 (0/345)	50 (1/2)	0,2 (1/467)	100 (1/1)	1,0 (1/97)	66,7 (2/3)	0,2 (2/1049)
Total	**72,2 (140/194)**	**100 (140/140)**	**81,7 (345/422)**	**100 (345/345)**	**53,4 (467/875)**	**100 (467/467)**	**57,4 (97/169)**	**100 (97/97)**	**63,2 (1049/1660)**	**100 (1049/1049)**

*: waterbody natural

Table V: Cumulative larval density of waterbodies in four rubber-bearing facies of the Dabou region (Côte d'Ivoire)

Habitats	**N**	**Average number of larvae/ Positive waterbody (CI 95%)**	**OR**	***P***
Immature Plantation	1553	11,2 (9,4 - 13,3)	0,4	0,001*
Mature Plantation	14302	36,3 (33,0 - 40,2)	1,4	0,002*
Village Surrounded	19577	36,1 (33,1 - 39,5)	1,4	0,002*
Village Remote	2765	25,8 (21,3 - 31,4)	1	

N: number of larvae; **CI**: 95% confidence interval; **P**: P value; **OR**: odds ratio; * results are significant

Table VI: Cumulative putative vector larval density of waterbodies in four rubber-bearing facies of the Dabou region (Côte d'Ivoire)

Habitats	**N**	**Average number of larvae/ Positive waterbody (CI 95%)**	**OR**	***P***
Immature Plantation	110	4,6 (2,9 - 7,1)	0,5	0,008*
Mature Plantation	1819	14,2 (11,9 - 17,0)	1,6	0,001*
Village Surrounded	4219	9,5 (8,6 - 10,5)	1,1	0,342
Village Remote	1461	8,7 (7,4 - 10,2)	1	

N: number of larvae; **CI**: 95% confidence interval; **P**: P value; **OR**: odds ratio; * results are significant

Table VII: Larval density of larval waterbodies types in rubber plantation habitats in the sub-prefecture of Lopou (Dabou, Côte d'Ivoire)

Habitats	**Type of waterbody**	**N**	**Average number of larvae/waterbody (CI 95%)**	**OR**	**P**
Immature Plantation	Leaf axil	642	4,5 (3,7-5,3)	0,2	0,001*
	Water storage drum	403	28,78 (16,9 - 49,0)	1,1	0,875
	Latex collection cup	501	4,7 (1,3 - 16,2)	0,2	0,015*
	Tree trunk	487	69,6 (33,0 - 146,7)	2,6	0,062
	dead leaf in water	259	18,5 (10,8 - 31,7)	0,7	0,392
	Roof tarpaulin	3	1,5 (0,3 - 8,9)	0,1	0,003*
	Abandoned container	242	26,9 (13,8 - 52,3)	1	
Mature Plantation	Leaf axil	21	4,2 (1,6 - 11,1)	0,3	0,050
	Water storage drum	12	2.0 (0,8 - 5,3)	0,2	0,001*
	Latex collection cup	14515	37,4 (33,8 - 41,4)	3,0	0,001*
	Abandoned container	224	12,4 (7,7 - 20,1)	1	
Village Surrounded	Cut bamboo	465	51,7 (26,7 - 99,9)	1,7	0,106
	Leaf axil	741	7,3 (5,9 – 9,0)	0,2	0,001*
	snail shell	178	178.0 (24,9 -1270,6)	6,0	0,075
	Water storage drum	3942	12,3 (11,0 - 13,8)	0,4	0,001*
	Puddle/tire track	28	2,9 (0,9 - 8,8)	0,1	0,001*
	Latex collection cup	1071	133,8 (66,8 - 267,92)	4,5	0,001*
	Gutter/Ditch	135	45,7 (14,4 - 144,8)	1,5	0,469
	Roof tarpaulin	2858	2372.0 (321,0 - 28071,3)	79,5	0,001*
	Water meter cover	34	17,3 (4,1 - 72,8)	0,6	0,459
	Abandoned container	11786	29,8 (27.0 – 33.0)	1	
Village Remote	Leaf axil	104	4,5 (2,9 - 7,1)	0,2	0,001*
	Water storage drum	840	42.0 (27,0 - 65,4)	1.9	0,011
	Latex collection cup	2	0,7 (0,1 – 4,0)	0,0	0,001*
	Dead leaf in water	14	14.0 (1,8 - 106,5)	0.6	0,651
	Water meter cover	30	30.0 (4,1 - 220,0)	1.3	0,775
	Abandoned container	2533	22,4 (18,6 - 27,1)	1	

N: number of larvae; **CI**: 95% confidence interval; **P**: P value; **OR**: odds ratio; * results are significant

Table VIII: Seasonal productivity of breeding sites in the rubber tree habitats of the sub-prefecture of Lopou (Dabou, Côte d'Ivoire)

Habitats	**Dry Season**				**Rainy season**			
	N	**average number of larvae per site (IC 95%)**	**OR**	**P**	**N**	**average number of larvae per site (IC 95%)**	**OR**	**P**
Immature Plantation	516	9,2 (7,0 - 12,1)	0,2	< 0,001*	1049	11,4 (9,2 - 14,1)	0,5	<0,001*
Mature plantation	12375	47,8 (42,3 - 54,0)	1,2	0,411	1927	20,1 (16,4 - 24,6)	0,9	0,276
Villages Surrounded	12941	50,8 (44,8 - 57,5)	1,3	0,293	6926	26,4 (23,4 - 29,9)	1,1	0,268
Villages remote	617	38,6 (23,5 - 53,3)	1		2874	23,4 (19,5 - 28,1)	1	

N: number of larvae; **CI**: 95% confidence interval; **P**: P value; **OR**: order ratio; ***** results are significant

Table IX: Seasonal productivity of *Aedes aegypti*, *Anopheles gambiae* s.l. and *Culex quinquefasciatus*

Putatives Vectors	Seasons	N	Average number of larvae/waterbody (CI 95%)	OR	P
Aedes aegypti	Dry	2637	10,4 (9,1 - 11,8)	1,6	< 0,001*
	Rainy	1831	6,7 (5,9 - 7,6)	1	
Culex quinquefasciaus	Dry	1845	17,6 (9,1 - 11,8)	1,7	< 0,001*
	Rainy	1203	10,2 (8,4 - 12,3)	1	
Anopheles gambiae s.l.	Dry	7	3,5 (0,7 - 16,8)	0,4	0,301
	Rainy	86	8,6 (4,5 - 16,5)	1	

N: number of larvae; **CI**: 95% confidence interval; **P**: P value; **OR**: order ratio; * results are significant

LISTE DES FIGURES