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Assessment of blackflies (*Simulium damnosum* s.l.) nuisance on Guiembé hydro-agricultural development site in Poro region, Côte d'Ivoire

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Abstract

Backgroung : *Onchocerca volvulus,* the causal agent of river blindness, is transmitted by the blackfly *Simulium damnosum* s.l., which breeds in turbulent river waters. This study focused on the nuisance, biting rates and transmission potential of *S. damnosum* s.l. on the hydro-agricultural scheme site in the commune of Giembe in northern Côte d'Ivoire.

Methods : Blackflies were collected using the human landing catch collection method on two consecutive days of each month during the rainy season from September to October 2023. Samples from each site were dissected monthly for estimation of parity and *O. volvulus* infection rates.

Results : A total of 3714 blackflies of the species *S. damnosum* s.l. were captured during the study period, of which 1795 (48.3%) were caught at site A and 1919 (51.7%) at site B. These were species from the savannah group (3688; 99.3%) and species from the forest group (26; 0.7%). Of the 3703 flies dissected, 1029 (27.6%) were parous, while the remainder were nulliparous female flies 2674 (72.4%) and no black flies were found harbouring *O. volvulus* parasites. It composed The daily cycle of aggression showed two peaks : one between 7 and 9 a.m. and the other between 2 and 6 p.m. Monthly bite rates were very high, ranging from 13800 to 3975 bites/person/month and the average daily aggression rate in the study area was 168.8 bites/person/day.

Conclusion : This study revealed that almost all the blackflies responsible for the nuisance in Guiembé are savannah species, with high biting rates. In addition to the low parity rates recorded, no flies infected with *Onchocerca volvulus* were found during the study period.

Keywords : Blackflies, *Simulium damnosum*, onchocerciasis, *Onchocerca volvulus*, Giembé, Côte d'Ivoire

Introduction

Onchocerciasis, commonly known as "river blindness", is a neglected tropical disease caused by a parasitic worm, and is geographically widespread but mainly found in sub-Saharan Africa (WHO, 2019a). Blackflies of the genus *Simulium* transmit the filarial nematode *Onchocerca volvulus,* which can cause severe visual loss and severe itching accompanied by dermal changes (Boakye *et al*., 1998 [2]; Opara *et al*., 2008). 37 million people in 34 endemic countries are currently at risk of infection, and around 99% of the global disease burden currently occurs in Africa (Maegga *et al*., 2011 [4]; WHO, 2020).

In Africa, onchocerciasis transmission varies according to various factors, such as vector species, vector abundance, seasons and habitats (Cheke *et al*., 1992) [6]. *Simulium neavei* and species of the *S. damnosum* complex are the most important vectors, with a wide range throughout Africa (Amazigo *et al*., 2006) [7]. Thus, in ecological zones where vectors are cytogenetically different, the disease tends to vary in severity (Adeleke *et al*., 2011) [11], with the division of *S. damnosum* s.l. into the savannah fly responsible for *O. volvulus* which causes blindness, and the forest fly which causes a skin disease (Sam-Wobo *et al*, 2013). [9]

In West Africa, the geographical distribution of onchocerciasis follows that of its vector along of fast-flowing rivers and people at risk are fishermen, farmers and children who spend much of their daytime near *S. damnosum* s.l. infested rivers (Duke, 1968) [10]. The bites of these species are most often annoying for humans and sometimes harmful to their health due to their toxicity. As a result, populations generally abandon infested areas (Marchal, 1978 [11]., Yapi *et al*, 2014) [12].

In 2017, the global disease burden was estimated at 220 million people in need of at least chemoprevention, of whom,14.6 millions suffered dermal damage and 1.15 million vision loss (WHO, 2019b, Debela, *et al*., 2021) [14]. Aware of the epidemiological scale and socio-economic repercussions of this disease, World Health Organization (WHO) has mobilized human, material, technical and financial resources to combat onchocerciasis (Hougard *et al*., 2012 [15]; Yapi *et al*., 2014) [12]. Thus, Between 1974 and 1990, the WHO launched a vast onchocerciasis control programme called OCP (Onchocerciasis Control Program), covering the endemic countries of West Africa. This program eliminated the risk of disease transmission with a considerable drop in blackfly density, making it possible to resettle populations on river basin land that had previously been abandoned for fear of blindness and simulidian nuisance (Yapi *et al.*, 2014) [12].

In Côte d'Ivoire, after the program was halted in 2002 due to the polico military crisis, blackflies reappeared exponentially leding to a high nuisance, especially in rural communities where activities are essentially agricultural or fishing along fast-flowing rivers (Yapi *et al*., 2014 [12], Coulibaly *et al*., 2022) [16]. Since 2019, Guiembé municipality has benefited from a hydro-agricultural development project on the Bandama tributary, to enable the irrigation of several hundred hectares of agricultural crops. The impact of such a project can also be negative, with changes to the local ecosystem leading to an amplification of pathogens, particularly those resulting from the proliferation of disease vectors. The installation of the dam and the irrigation channels will also modify the water flow upstream and downstream of the dam, making it slower or faster in places, thus creating breeding grounds for simulium larvae. This modification of the ecosystem could therefore result in a major nuisance for the simulium vectors of onchocerciasis. The possibility and necessity for each country to carry out entomological activities in order to implement vector control is one of the reasons that prompted this study in the commune of Guiembé, whose populations are exposed to the effects of blackflies recrudescence due to the proximity of the Solomougou dam. As has been the case in other large regions, study was designed to assess the nuisance caused by blackflies at the Guiembé hydro-agricultural development site. Indeed, it has been showed that the prevalence of human onchocerciasis is directly linked to the presence and abundance of its vector ; consequently, a detailed understanding of parasite and vector population dynamics is crucial for vector control.

Methods

Ethical approval

All study participants were provided with full and detailed information on study procedures and objectives in their Local language. Each participant verbally agreed and signed an informed consent to participate as a study volunteer. All study volunteers, including blackfly catchers, received treatment with Ivermectin, as members of the local population. The study was approved by community leaders and District health officials.

Study area

The study was conducted in Guiembé municipality in Poro region of northern Côte d'Ivoire, 30 km from Korhogo and about 580 km from Abidjan. It is located between 9° 14′ N and 5° 43′ W and covers an area of 470 km² with an estimated population of 19872 (RGPH, 2021). The municipality benefits from a hydro-agricultural scheme on the tributary of the Bandama River called Solomougou. It is bordered to the north by the sub-prefectures of Tioroniaradougou, Dassougbo and Kombolokoura, to the west by Sirasso, to the south by those of Dikodougou and Kiémou and to the east by Napié (Figure 1). This region has a Sudanese climate with two main seasons. The dry season, which lasts from November to April, precedes the rainy season, which is characterised by two rainfall peaks, one in August and the other in September.

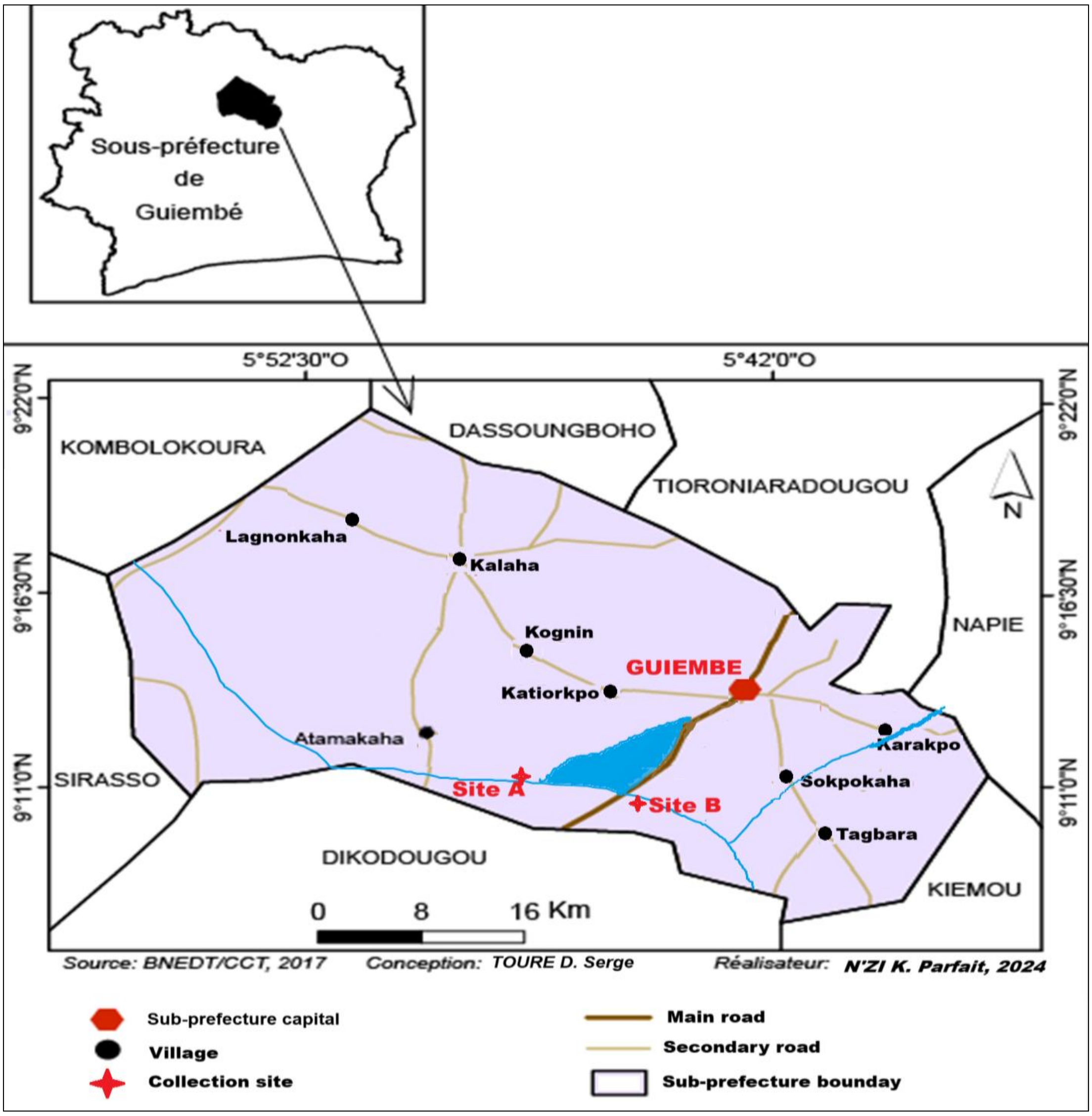

Fig 1: Map of Guiembé showing the study site

The daily and annual thermal amplitudes are relatively large and the humidity is lower than in the south of the country. The area is characterised by the intermittent presence of a cool, dry wind, the harmattan, between December and February. Annual rainfall varies between 1000 mm and 1400 mm.

Adult blackfly sample collection

At each site within the hydro-agricultural perimeter, adult blackfly collections were timed to coincide with periods of peak *O. volvulus* biting activity and transmission during the rainy season from July to November 2023. Four blackfly catchers, two per site, were trained in standard "human landing collection" methods for collecting adult blackflies (Walsh, 1983 [18] ; Higazi *et al*., 2013 ; Zarroug *et al*., 2014). Test samples were collected over five days, between 7 a.m. and 6 p.m., to identify sites with high blackfly activity and assess staff performance in capturing black flies. The actual collection was done two days each month from July to November 2023, simultaneously at both sites. The two staff capturing blackflies were at least 500 m apart and the flies were caught on an hourly rotation system, to minimize any potential bias relating to the efficiency of staff capturing blackflies. At the end of each hour, the date, time, name of the catching point and that of the catcher were specified. The catchers then recorded the number of females caught per hour on a capture sheet, as well as information on meteorological parameters such as ambient air temperature and relative humidity (recorded every hour).

Identification and dissection of adult blackflies

All the flies were identified morphologically into savanna and forest flies using the colours of the wing tufts, for ecoxa, abdominal tergite, scutellum and antennae (Quillivéré *et al*., 1977 ; Baker *et al*., 1985 ; Davies and Crosskey, 1991[30] ; Wilson *et al*., 1993). Each identified fly was dissected using dissecting pins and a microscope. The ovaries of the dissected fly were stretched and classified as parous or nulliparous after observing characters such as the transparency of the Malpighian tubules and the presence of fat bodies (Cupp and Collins, 1979 ; Finney, 1980). The flies were then dilacerated in a drop of 4.5% physiological water, and carefully examined for *O. volvulus* larvae, whose numbers and evolutionary stages make it possible to establish the infestation rates and parasite loads of *S. damnosum* complex females.

Entomological indicators of blackfly density

The density indicators taken into account in the Onchocerciasis Control Program (OCP) to estimate the number of Simulium damnosum s.l. coming to bite a man who is exposed to bites for almost every hour of the day every day were calculated as the daily bite rate and the monthly bite rate (Walsh *et al*., 1978) [27].

Daily biting rates (DBR)

Hourly-based black fly distribution was recorded and added to calculate flies biting-rate per man-day.

$$\text{DBR} = \frac{\text{Number of female blackflies collected}}{\text{Number of catching days x Number of cather}}$$

Monthly biting rates (MBR)

MBR is a global estimate of number of bites a person exposed to a population of Simulium vectors will receive over the course of a month. It is probably an overestimate, as not all flies that land on a person successfully obtain a blood meal. The MBR was calculated as follows (Davies and Crosskey, 1991 [30], WHO, 2023)

$$\text{MBR} = \frac{\text{Number of blackflies collected X Number of days in month}}{\text{Number of catching days}}$$

Results

Blackfly diversity and abundance

3714 flies were caught during the study period, of which 1795 (48.3%) were caught at point A and 1919 (51.7%) were caught at point B (Table 1). Morphological identification of blackflies revealed the presence of two groups of species. These are species in the savannah group 3688 (99.3%) and species in the forest group 26 (0.7%) (Fig 2, Fig 3 and Table 2).

Parity and infection rate

Of the 3703 flies dissected, 1029 (27.6%) were parous, while the rest were nulliparous female flies 2674 (72.4%). Proportion of nulliparous flies was higher than parous flies in savannah species (72.5%) while in forest species, parous flies were high than nulliparous flies (69.2%) (Table 2). Dissection revealed that no blackflies were found harbouring *O. volvulus* parasites. The potential for monthly transmission was therefore nil during the study period**.**

Table 1: Monthly relative abundance of blackflies

	July	August	September	October	November	Total
No. of blackfly caught (Point A)	420	350	400	504	121	1795
No. of blackfly caught (Point B)	464	430	423	438	164	1919
Total	884	780	823	942	285	3714

Table 2: Female blackfly species caught and Entomological indices in the study area

Blackfly species group	No. Of blackflies collected (%)	Number flies dissected	No. Of parous flies (%)	No. Of nulliparous flies (%)	Monthly transmission potential
Savanna species.	3688 (99.3%)	3677	1011 (27.5)	2666 (72.5)	0
Forest species	26 (0.7%)	26	18 (69.2)	8 (30.8)	0
Total	3714	3703	1029 (27.8)	2674 (72.2)	0

Daily and monthly blackfly nuisance

Overall, daily bite rates were almost similar over the study period. The highest daily bite rate was recorded in October (230 bites/person/day), while the lowest daily bite rate was recorded in November (66.3 bites/person/day). Monthly bite rates were very high, ranging from 13800 to 3975 bites/person/month (Table 3).

The mean daily aggressiveness rate in the study area was 168.8 b/p/d. The results revealed that blackflies were Abundant throughout the day on the agricultural perimeter, becoming more aggressive from the afternoon onwards (Fig 2). Hourly variations in simulidian nuisance show two vertices, one in the morning and the other in the early afternoon. The morning peak occurs between 7 and 9 a.m., with an average of 197.3 b/p/d. From 2 p.m., this aggressiveness increases exponentially and peaking between 3 and 4 p.m. (259.5 b/p/d).

Table 3: Daily and monthly simulidean nuisance

Months of collection	No. of capture days	No. of black flies collected	Aggressivity (bites/person/day)	Aggressivity (bites/person/month)
July	2	894	223.5	13410
August	2	780	195.0	11700
September	2	855	213.8	12825
October	2	920	230.0	13800
November	2	265	66.3	3975
TOTAL	10	3714	185.7	11142

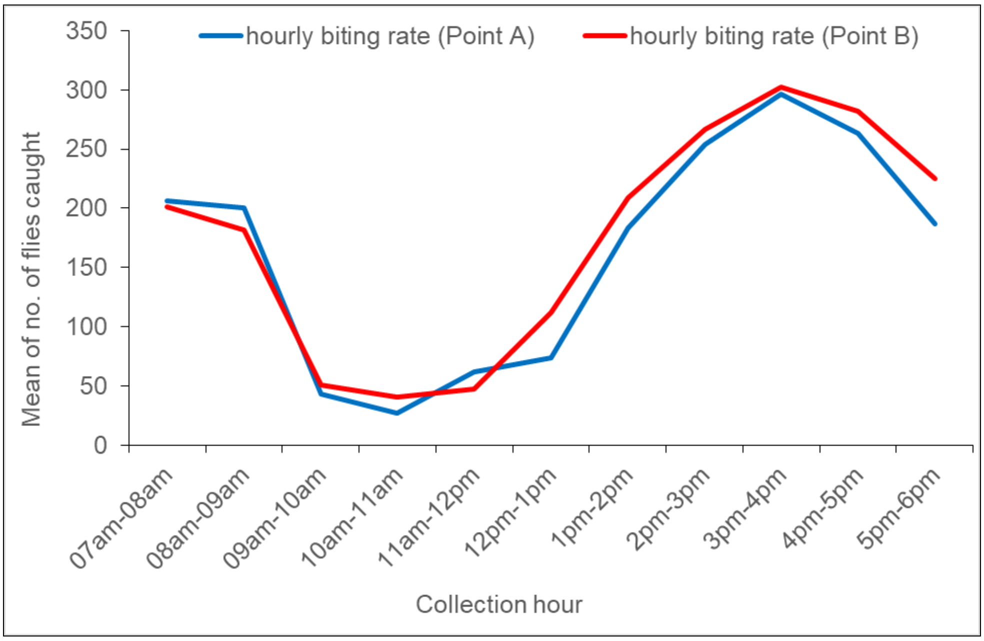

Fig 3: Hourly bitting pattern of Blackflies in Guiembé

Discussion

The data collected in this study showed that savannah species make up the majority of the blackfly fauna with very high biting rates. The high abundance of flies at the hydro-agricultural site may be linked to the hydrophysical characteristics of the Solomougou River, which irrigates the site. This observation is consistent with previous reports that fly abundance tends to coincide with the rainy season, accompanied by high river flows and rapid. According to the authors, these conditions usually favour the breeding of blackflies, leading to an increase in the adult fly population (Crosskey, 1990 [30]; Opara *et al*., 2008 ; Busari *et al*., 2021) [31]. Given that this study was carried out during the rainy season when populations are preoccupied with field work, predominance of savannah species of *S. damnosum* s.l. could however pose a problem due to the nuisance of black flies, which disturbed strongly agricultural and fishing activities, which could lead to a drop in annual productivity in this region.

The coexistence of forest and savannah black flies is thought to be due to the fact that the Guiembé hydro-agricultural site stems from a tributary of the Bandama river, which offers vegetation that as a whole is covered with fairly dense wooded savannah.

In fact, simulids, which travel several hundred kilometres in search of blood meals, can follow the forests along the river to reach the savannah zone to the north. Deforestation of forested areas by farmers and climatic variations could disrupt their living environment, leading them to conquer areas suitable for their development (Philippon, 1978) [32]. The phenomenon of migration could explain presence of forest blackflies in our study area, where we can also assume a simple phenomenon of occupation by extension of the area that has become favourable to forest species.

Dissection results showed that the majority of flies caught in the study area were nulliparous. This observation is consistent with the report of Opara *et al.* (2008), but contrary to those of Usip *et al.* (1999) [33] and Okolo *et al.* (2004) [34]. The high proportion of nulliparous flies could be due to their local production. In addition, the relatively high number of nulliparous flies could also be a contributing factor, since only older flies have the opportunity to transmit the parasite. It is also worth noting that the low parity rates recorded in this study indicate a young blackfly population and may reflect local production of black flies (Opara *et al*.,

2008) [3]. According to WHO, (1995), a high proportion of nulliparous flies would indicate incomplete vector control or high productivity of breeding sites. Conversely, a high proportion of parous females could indicate successful vector control or the presence of migratory flies. As no vector control programme has been implemented in the study area, the high proportion of nulliparous females could indicate high productivity of breeding sites.

The results also showed a significant variation in monthly fly biting activity in the study area. In addition, a bimodal peak in the daily rhythm of fly biting was recorded. The first peak occurred between 9am and 11am in all months, while the second peak occurred between 3pm and 5pm. Similar observations have been reported elsewhere in several sub-Saharan African countries (Adewale *et al*., 1999 [36] ; Okolo *et al*., 2004 [34]; Opara *et al*., 2008) [3]. Monthly variability in blackfly densities confirms the highly dynamic nature of the population and can be explained by the water flows in the river, which are strongly correlated with climatic conditions, mainly rainfall and temperature (Domche *et al*., 2022) [37]. At the end of the rainy season, the volume of water decreases and the egg-laying sites become more accessible to female blackflies, which can lay their eggs easily and safely. Similar trend observed in the monthly dynamics of blackfly densities could be explained by the fact that the two sampling sites are located in the same area and about 3 km apart. However, the evening peak was higher than the morning peak. This observation is consistent with previous reports, which generally indicate an increase in evening peaks. This observation could be related to the behaviour of the inhabitants of the region, most of whom are farmers who carry out rural activities at this time of year when fly densities are high. It could also be explained by environmental factors not considered in this study. The number of samples obtained was insufficient to meet the threshold set by the WHO. Furthermore, in the absence of data on blackfly infection rates in this study, it is difficult to rule out the possibility that cycles of endemic disease may persist even in the absence of Onchocerca-infected flies in the vector population, especially when biting rates are high (Tekle *et al*., 2012) [39]. The absence of infection in the flies dissected could indicate a low level of microfilarial load in the reservoirs of the human population of the commune of Giembé (Rodringuez-Peres *et al*., 1999) [40]. Although none of the flies were infected, the high rates of fly bites in the early and late morning represent a high risk to residents and farmers in the area. In this case, a migration of infected human hosts with a high load of microfilariae could, within a few years, change the transmission pattern of onchocerciasis in the region, given the presence of potential vectors. In view of the results of this study, the high rate of black fly bites could correspond to a strong nuisance for the human population rather than to transmission of the disease to Giembé, and could well favour the persistence of the disease in this region.

Conclusion

This entomological study found that the blackfly population in the study area was predominantly savannah species, with high biting rates in the early morning and afternoon. It provided basic data on the biting behavior and transmission of onchocerciasis of the S. damnosum s.l. complex in the Guiembe hydro-agricultural perimeter. Bionomic differences between sister species of the S. damnosum complex should be taken into account when designing entomological monitoring protocols for onchocerciasis control and elimination interventions.

In order to understand further the epidemiology and transmission of onchocerciasis in the study area, it is paramount that molecular studies targeting the aspect of vector competence and parasite vector complexes be conducted. In this case, a more in-depth study using more sensitive techniques to detect the parasite in blackflies is required to provide decision-makers with timely data for the implementation of integrated strategic control measures.

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