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Evaluation of *Onchocerca volvulus* Infectivity Status of Black Flies in Post-Ivermectin Regime in Kachia, Kaduna state, Nigeria

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Abstract

The infectivity status of Simulium species is an important factor in assessing the transmission of Onchocerca species. In this study, we determined the biting rate and presence of the larval stage of the parasite to determine the status of onchocerciasis in the post-ivermectin era in the study area. A total of 1.063 black flies were collected in September and October 2023. Dissection was carried out under a microscope to examine the internal organs for the presence of microfilaria and record their condition. The findings indicated that parous flies 311(29.3%) were markedly less prevalent compared to nulliparous flies 752(70.7%). Among the dissected flies, a total of 14 (1.3%) were found to be infected with Onchocerca species. The flies exhibited a bimodal biting pattern, with a peak in the morning occurring between 9 and 10 am and afternoon between 4:00 and 5:00 pm. The monthly biting rates of 5177.0 and 5807.3 in September and October respectively exceeded the World Health Organization's threshold of 1,000 bites per person per month. It is essential to investigate the potential resurgence of onchocerciasis in the study area, which may be attributed to the overlapping presence of cattle, Simulium vectors, and humans.

Keywords: *Simulium damnosum*, Ivermectin, infection, Onchocerca species, Kachia

INTRODUCTION

The black flies are predominantly known for their significant role in public health as vectors of parasitic nematodes responsible for the transmission of *Onchocerca volvulus* Leuckart (Spirurida: Onchocercidae), which is the etiological agent of onchocerciasis. The disease, which causes blindness and debilitating skin conditions, presently occurs in 30 African countries, Yemen, and two Latin American countries (Adler *et al.,* 2010, Rodriguez-Perez et al. 2015). Nigeria is the most affected country in the world, accounting for approximately one-quarter of all 37 million recent onchocerciasis cases (Ikani *et al.* 2022), In Nigeria, ivermectin

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treatment is the sole control method for onchocerciasis (Ojorongbe et al. 2015), elimination of the disease is thought to be feasible (Tekle et al. 2012).

Recent empirical findings indicate that interventions aimed at control have significantly reduced the populations of parasites; however, vector surveillance remains essential where the infection continues to be endemic. As initiatives aim to eliminate the disease in endemic communities, entomological evaluation is critically necessary to inform control strategies and avert the potential resurgence of infection.

This is all the more true in Africa where transmission cycles are still strong (Kamga et al. 2016, Komlan et al. 2018, Hendy et al. 2018). The recent WHO guidelines for verifying interruption of transmission stipulated that the upper bound of the 95% confidence interval of the estimated prevalence of flies carrying parasites in the infective stage (L3) in the head must be less than 0.1% (< 1/1000) in parous flies or less than 0.05% (WHO, 2016).

Kaduna State in Nigeria has long been reported to be endemic for onchocerciasis based on the results of rapid epidemiological mapping of onchocerciasis (REMO) conducted in 1995, 1996, and 2008 (APOC, 2008), and community-directed treatment with ivermectin (CDTI) has been ongoing in the state. (WHO, 2016) also reported a prevalence of 0% of all the 3,703 examined individuals using skin snip, after 15 to 17 years of treatment of ivermectin treatment with more than 75% reported coverage. (Tekle et al, 2012) This may not be sufficient to conclude.

The knowledge of entomological indices of riverine Simulium blackflies is crucial for assessing the transmission of onchocerciasis. Studies carried out in Cross River State, Cameroon, and the Kakoi-Koda focus have highlighted the importance of assessing biting rates, transmission potentials, and parity rates of Simulium flies (André *et al*. 2022, Friday *et al* 2023). These entomological parameters provide valuable insights into the prevalence of infection and the potential for disease transmission. Furthermore, empirical studies elucidate the factors influencing vector population dynamics and transmission rates[Dd1], thereby underscoring the necessity for integrated vector control strategies in conjunction with mass drug administration to achieve efficacious disease eradication (Opara et al. 2005).

In this study, we present an assessment of the transmission potential of *O. volvulus* in Kachia, an endemic Local Government Area located in Kaduna State, Nigeria by determination of the biting rate of blackflies and the transmission parameters of the vector flies collected in Kachia, an endemic Local Government Area, Kaduna State, using dissection technique.

MATERIALS AND METHODS

Area of study

The research was carried out in Kachia Local Government Area, Gurara community characterized by the endemic presence of onchocerciasis in Kaduna State, Nigeria. The geographical co-ordinates are 9°52′57.5″ N 7°58′04.1″E. The sampling location for the *S. damnosum* complex is the Gurara River which supplies water to nearby communities for domestic and agricultural purposes (Abaje and Oladipo, 2019) and are suitable habitat for *S. damnosum* complex breeding. Kaduna State generally has an annual rainfall ranging from 1323 -1525mm with its peak in August, with two seasons of six to seven months rainy season between (March/April to September/October) and a dry season (end of October and November to March of the following year) (Abaje and Oladipo, 2019) The people are

predominantly farmers and fishermen, occupations that come with high exposure to black fly bites.

Collection of samples

Prior to the beginning of the study, the black flies breeding sites were surveyed as an integral component of the community engagement protocol, aimed at establishing rapport with the leaders and senior figures of the village. The research period was in September and October, 2023, flies were collected for six days consecutively for each month. The research procedures were communicated to the community members. Individuals volunteering to participate were identified, and two individuals who provided informed consent received training in the methodologies pertinent to vector collection.

. One vector-catching site was selected where 2 catchers worked alternatively on hourly basis (7:00 am – 6:00 pm local time) and flies were collected using the Onchocerciasis Control Programme protocol for human landing catches technique with slight modifications (Walsh, et al, 1983; WHO, 1995). Each fly caught was preserved in individual plastic tubes containing moist cotton wool until dissection in a field laboratory. The fly collection was done on six consecutive days each month. The data acquired from adult black flies was used to determine the daily and monthly biting rates, parous daily and monthly rates.

Procedure

A total of 1063 black flies were dissection and assessed for parity and the presence of *O. volvulus* using a dissecting microscope (Micron OPTIK) at a magnification of 40×. Black flies were classified as parous if they had stretchy ovaries, reduced and clear malpighian tubules, while black flies with numerous fat bodies, non-stretchy ovaries, and dense malpighian tubules were classified as nulliparous (Monkey 1980). The hourly biting behaviours of the black flies were systematically observed, and the periods of biting intensity were determined.

Calculation of transmission indices

Transmission indices, such as the monthly population density (MPD) of the *S. damnosum* complex in the study site, the daily biting rate (DBR), the monthly biting rate (MBR), and the percentage of parous flies in the total dissected (proportion of parous flies), were calculated using standard methods (WHO 1995, 2001).

Data Analysis

Data on the Transmission indices were calculated by the formula described by Lamberton *et al*. (2014). The hourly biting rate was presented in a graph using Microsoft Excel.

RESULTS

The overall transmission indices resulting from the dissection of the 1,063 is shown in Table 1.

During this period, the total number of parous flies was 311 (29.3%), while nulliparous flies were 752 (70.7%). A total of 14 (1.3%) sampled dissected black flies were infected with the first and second stages of *Onchocerca* larva. In this study, the parous flies exhibited two modes of biting pattern: between 9 to 10 am and 4:00 to 5:00 pm (see Fig. 1). The lowest monthly Biting Rate (MBR) was recorded in September at 5177.0 flies per person per month, while the peak occurred in October at 5037.5 bites (Table 1). During the sampling, 311 out of 1060 (29.3%) S. damnosum s.l. caught were parous, while the rest were nulliparous female flies 752 (70.7%). The daily biting rate was highest in October (187.3 flies per person per day).

Table 1: Indices of Black fly transmission potentials in Kachia, Kaduna state

Transmission parameters	September	October	Total
Total flies dissected	501	562	1063
No (%) Parous flies	145(29.0%)	166(29.5)	311(29.3)
No (%) Nulliparous flies	356(71. %)	396(73.3)	752(70.7)
No (%) of flies infected with *Onchocerca* species	10(2.0)	4(1.0)	14(1.3)
Flies with L1 and L2 *Onchocerca* Spp	10(2.0)	4(1.0)	14(1.3)
Flies with L3 *Onchocerca* Spp	0	0	0
Biting density of flies	22.8	17.0	-
Daily Biting Rate (DBR)	167.0	187.3	-
Daily Parous Biting Rate (DPBR)	48.3	55.3	
Monthly Biting Rate(MBR)	5177.0	5807.3	-
Monthly Parous Biting Rate(MBR)	1450.0	1715.3	

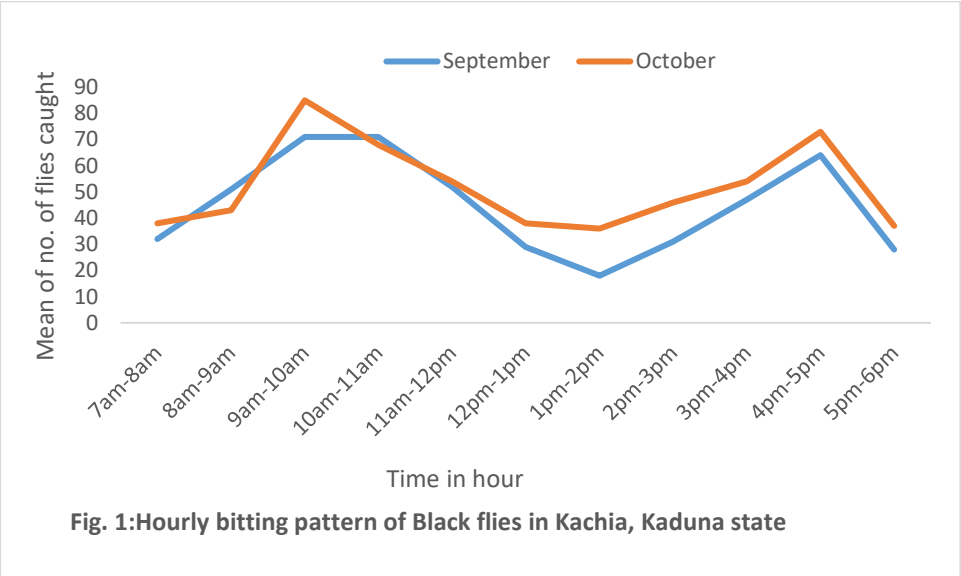

Fig. 1:Hourly bitting pattern of Black flies in Kachia, Kaduna state

DISCUSSION

Study on black flies abundance and biting rate in a post-Ivermectin-treated local Government Area in Kuduna state revealed evidence of the first stage of *Onchocerca* infection. The number of samples obtained was not enough for the threshold set by the WHO. Additionally, since there was no pre-control data on black fly infection rates, it's difficult to rule out the possibility that endemic disease cycles can persist even if only a small fraction of the vector population is infected with onchocerca, especially if biting rates are high, hence, we are careful to show as evidence of the continued efficacy of mass ivermectin treatment, and as a potential sign that *Onchocerca* transmission cycles in Kaduna State have been eliminated (Tekle *et al.* 2012). In this case, a further and more comprehensive survey is needed with more sensitive techniques in the detection of the parasite in the blackflies as to furnish the public health policy-makers with timely data for the implementation of integrated strategic control measures.

The biting time varies due to synchronized daytime hours with the occupational activities, such as farming and fishing in the study area. The corresponding activities between black flies biting pattern increase the risk of the disease among other factors.

It would be extremely difficult to change when and where people are active during the day, but such changes could contribute to the disruption of disease transmission cycles (Adeleke et al. 2010). However, in Kaduna State there is no established reason for low parity. However, Service (2004) suggested that it is likely a reflection of the age/phenological structure of local black fly populations.

According to the previous findings, this result contradicts earlier reports of the absence of Onchocerca species in many parts of southwest Nigeria in black flies as reported by Adeleke et al. (2010) and Sam-Wobo et al. (2013) but agrees with reports of continued endemic transmission in post-Ivermectin treatment in Northeast, Nigeria (Akafyi *et al*. 2021) and in Africa (Lamberton et al. 2015, Hendy et al. 2018). The infectivity of black flies could reflect the overlap of cattle, *Simulium* vectors, environment and humans in the study area. The community location has the potential of both the bovine and human *Onhocerca* species transmission (Akafyi *et al*. 2023 there exist additional transmission parameters that may significantly influence the detectability of recent infections; however, we have no direct evidence of this claim

In conclusion, it is essential to investigate the threat associated with the resurgence of onchocerciasis, which may be attributable to the interactions among cattle, Simulium vectors, and human populations in the study area

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