

Mvolo County, an Onchocerciasis Endemic Area in Western Equatoria State, South Sudan: An Entomological Study to Prepare for a “Slash and Clear” Community-Based Vector Control Intervention

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Background: Mvolo in Western Equatoria of South Sudan has been a hotspot for *Onchocerca volvulus* transmission since the 1940s. In Mvolo onchocerciasis is a disease of public health importance, associated with onchocerciasis-associated epilepsy including nodding syndrome.

Methods: We conducted an entomological study to map the breeding sites of blackflies (*Simulium damnosum*, *sensu lato*) on the river Naam, to allow the removal of vegetation from vector breeding sites, the “slash and clear”. Three blackfly catching sites were established along the river. Focus group discussions were also conducted to assess the willingness of the communities to support the “slash and clear” intervention and the semi-annual distribution of ivermectin.

Results: A total of 2466 female *S. damnosum* s.l. were caught in 14×11h (06.00–15.00) catches. The highest biting density of 4210.25 flies/month/h and monthly biting rate (MBR) of 11,482.25 bites/man/month were observed in November 2023. Biting density and MBR reduced to zero in the intervention site by April 2024. While the mean parity rate was 31% (CI: 0.2976±0.9176). Two diurnal biting peaks were observed, one from 9:00–10:00 (at the bridge site) and a prominent one from 14:00–15:00 in the two catching sites in Mvolo. Along the river Naam, only one site was found productive for *S. damnosum* s.l.; and the larvae and adults were morphologically associated with the anthropophilic *S. damnosum*. The “slash and clear” intervention was implemented at Dogoyabolu along the river Naam. Communities expressed willingness to support a “slash and clear” intervention and the semi-annual distribution of ivermectin.

Conclusion: *S. damnosum* active breeding was identified along the river Naam in a stretch of 3–5 km close to human settlements. Highest blackfly biting density was 4210.25 flies/month/h, and two fly biting peaks were observed. A community “slash and clear” vector control was implemented, and will be prospectively monitored.

Keywords: onchocerciasis, South Sudan, entomology, slash and clear, ivermectin

Introduction

Onchocerciasis is a neglected tropical disease (NTD) transmitted by female blackflies (Diptera: Simuliidae) breeding in fast flowing rivers and streams. In the sub-Saharan Africa, *Simulium damnosum* s.l. is the principal vector of the filarial parasite, *Onchocerca volvulus* the causative agent for onchocerciasis.¹ The female blackfly plays an important role in the transmission of onchocerciasis. While taking its blood meal from a human host the parasites enter the human skin and migrate to subcutaneous tissue. They develop into adult worms which reside in worm bundles located in palpable nodules or in deeper body tissues, and

produce microfilariae (mf) that migrate throughout the body, mainly to the skin and eyes.² Adult female worms live for 10 years on average and produce hundreds to thousands of mf daily.³ When the blackfly takes another blood meal from a human host, it ingests the microfilaria which penetrate the midgut and migrate to the thoracic muscle. The microfilariae later develop into an infective stage (L3) and migrate to the head and proboscis of the blackfly completing the cycle. Understanding the life cycle of the parasite allows us to appreciate the current interventions being deployed to control or eliminate onchocerciasis.

Onchocerciasis remains a public health problem at global level with 99% of the cases registered in sub-Saharan Africa.¹ The clinical symptoms are caused by the parasite's (*O. volvulus*) developmental phases and the host's inflammatory reactions to the parasite.¹ However, there is also strong epidemiological evidence that onchocerciasis is associated with epilepsy, particularly in areas with suboptimal onchocerciasis elimination programs.^{4,5} The type of epilepsy associated with onchocerciasis is currently called onchocerciasis-associated epilepsy (OAE) which includes the nodding syndrome.⁶⁻⁸

All NTDs except Chagas disease are present in South Sudan. Onchocerciasis is prevalent in approximately half the country.^{9,10} One of the states historically known for the severity of the disease since the 1940s has been Western Equatoria with a hotspot in Mvolo County.¹¹ Control of the disease has been through annual mass treatment with ivermectin, which has been recently upgraded to biannual treatment¹² to accelerate onchocerciasis elimination.

In 2017, a high prevalence of OAE and high ongoing *O. volvulus* transmission was documented in Maridi^{13,14} another onchocerciasis endemic area in Western Equatoria State. This high prevalence was found to be related to the high ongoing *O. volvulus* transmission at the Maridi Dam, the only blackfly breeding site in the area.¹⁵ Therefore, in 2018, in an effort to supplement ivermectin treatment, a low-cost community-based vector control method, "Slash and Clear" (S&C), was implemented at the Maridi dam, the local blackfly breeding site.¹⁶ It took only four volunteers and four working days to remove the vegetation at the Maridi dam and reduce the blackfly biting rate by more than 50%.¹⁶ Based on these promising results, the NTD program in collaboration with AMREF Health Africa, South Sudan thought of scaling up this strategy in Mvolo and Mundri East and West, counties with a high OAE prevalence.¹⁷ Prior to the implementation of this intervention in Mvolo an entomological and qualitative study was conducted to identify blackfly breeding sites, explore its feasibility, effectiveness and acceptability in the communities along the river Naam.

Materials and Methods

Description of Study Areas

Mvolo County is generally a wooded grassland with the Naam being the main river; and it meanders along its course with some section splitting into two or three sub-tributaries. However, within the 5 km stretch alongside Mvolo village it is characterized by huge rock outcrops that are mostly bare, and were earlier mentioned by Schweinfurth and Lewis¹¹ during their earlier exploratory visits to this area. In terms of size, the Naam is a medium-size river especially the stretch close to Mvolo village where some sections of the river measure less than five meters wide. These characteristics made prospection for the early stages of onchocerciasis vector feasible because one can enter the water and pick the trailing vegetation where the young stages attach themselves. In this village, most settlements are clustered and are within a distance of 1–2.5 km from the river. The communities were initially pastoralists, but after several armed conflicts, they are adjusting to subsistence farming being supplemented by fishing, particularly during the dry season. Fishing attracts most of the community members from young children to adults who are in the water most of the time setting their nets, while others swimming. These activities provide a lot of exposure to blackfly bites.

Mvolo is a hyperendemic area for onchocerciasis, and annual and biannual treatment has been undertaken in an effort to reduce the burden of the disease. By the time of study, the population had more than ten annual and one biannual rounds of ivermectin.

Study Design

This was a cross-sectional population based study. A river was prospected and villages were purposively selected for focus group discussions (FGD). The plan was to select two sites for the S&C intervention sites and two control sites without a vector control intervention and to monitor blackfly biting rates.

Study Procedures

Entomological Study

Training of Field Teams

Thirteen community members were selected with guidance of local authorities and Mvolo health department. They were provided a half-day theoretical training on onchocerciasis, its vectors, transmission, signs and symptoms, control, types of breeding sites, biting behavior, human landing catches, catching forms, and recording including the preservation of flies. Information on slash and clear vector control including supervision was also provided to trainees. Practical session was conducted the following day along the river Naam.

Larval Collection and Mapping Breeding Sites

A map of Mvolo County mainly used by United Nations High Commission for Refugees (UNHCR) was downloaded, this had some names of villages but lacked physical features vital for planning blackfly prospections. To help the team move forward, the supervisor was requested to list all the sites along the river Naam where there was fast running water. This list later guided the field team that moved upstream checking all these sites. At each site, standard prospection procedures were conducted,¹⁸ at least 30–45 min were spent checking trailing vegetation, sticks, hanging substrates in water, rocks, etc. Productive sites with young stages, these were collected in polythene bags for further identification and hatching based on procedures earlier described.¹⁸

On completion of mapping breeding sites on the river Naam, three catching sites were established. The choice of catching sites was made to ensure it was located in the shade, sheltered from the wind and usually accessible all year round.^{18,19}

Biting Activity of *S. Damnosum* s.l. Along the River Naam in Mvolo County

Diurnal Biting Activity

In the three established catching sites along the river Naam, two collectors of flies serving as volunteers were situated 250 m apart. The fly collectors were either seated under the shadow of a tree or near a huge rock gorge that equally provided shade to catch wild host-seeking females of *S. damnosum*. The fly collectors were not smokers and did not wear perfume or any lotion that could influence the host-seeking behavior of blackflies. The fly collectors mainly wore short pants and collected blackflies on exposed lower part of the skin. Host seeking females were captured while they were landing or attempting to feed on the human collector. Catches were conducted from 6:00 am to 5:00 pm following the earlier described procedures.²⁰ Due to changes made in the South Sudan time, hourly catching time had to be adjusted and volunteers were advised to start catches at 6:00 am and stop at 5:00 pm. The HLC method typically gives information on the relative density of the populations of biting females of *Simulium* spp. vectors per person in a given place and period.^{21,22} To mitigate the risks posed by HLC to vector collectors, they were treated with ivermectin prior engaging in this activity. They will get additional ivermectin doses from the routine program treatment that takes place twice a year.

Dissection of Flies for Parity (Age Grading)

Flies caught from the three catching sites along the river Naam were kept alive by wrapping them in a moist piece of cloth soaked in water. The field team confirmed the species of blackfly before the process of opening the abdomen began. Dissection of female black flies was done with a stereomicroscope field microscope. Before the dissection, the female flies were anesthetized with chloroform and then deposited one at a time in a drop of normal saline on the slide. One examined with the stereomicroscope, using the adjusted lighting system above, the morphological characters that identify the fly to species level. Next, dissecting needles were used to draw on the end of the abdomen to withdraw its contents. Dissections for parity were conducted following standard routine procedures¹⁹ and specimens were recorded as nulliparous or parous. For each site, 100 flies or more were dissected for each catching day.

Calculation of Entomological Indices

Entomological indices such as monthly population density (MPD) of *Simulium damnosum* complex in the study sites, the daily biting rate (DBR), monthly biting rate (MBR) and percentage of parous flies (PPF) were calculated according to previously published methods^{18,19,21,22}.

(a) Monthly Population Density: The monthly population density also called the biting density of flies was calculated through the fly per man hour (FMH) using the formula:

$$\text{MPD} = \frac{\text{Number of flies caught in a month}}{\text{Number of catching days} \times \text{time in hours}}$$

(b) Daily Biting rate (DBR)

The Daily Biting Rate (DBR) was calculated by using the formula:

$$\text{DBR} = \frac{\text{Total number of flies caught in a month}}{\text{Number of catching days}}$$

(c) Monthly Biting Rate (MBR)

The monthly biting rate (MBR) was computed by using the formula:

$$\text{MBR} = \frac{\text{Number of flies caught in a month} \times \text{number of days in the month}}{\text{Number of catching days}}$$

(d) Proportion of Parous flies

This represents the percentage of parous flies (PPF) in the total dissected. It is calculated by:

$$\text{PPF} = \frac{\text{Number of parous flies} \times 100}{\text{Total number of flies dissected}}$$

Qualitative Study

Focus Group Discussions (FGD)

FGDs were conducted with the communities in Gira, Dukaboro. Minimbere and Bogori 2 in Mvolo County to assess their knowledge and attitudes on onchocerciasis including the slash and clear intervention to be introduced in the area. Each FGD comprised 8–12 people, males and females. A checklist guiding discussions was used, and there was a moderator and a note taker for every discussion. All FGDs participants consented prior to engaging in the discussions. Topics discussed included: nuisance by blackflies, consequences of blackfly biting, time and places where people are bitten, coping mechanisms to blackfly bites, willingness to participate in S&C activities, challenges faced by volunteers during ivermectin distribution and attitudes toward ivermectin intake.

Slash and Clear Implementation

The breeding site identified for the S&C intervention was localized within a very small stretch along the river Naam. This site had stretches of fast running water measuring about 700 m upstream. The field team cut using sharpened machetes and removed trailing vegetation hanging on rocky outcrops and threw them along the riverbank. Slashing was 700 m upstream and 200 m downstream and all the vegetation cut was thrown onto the bank of the river. Since there was only one S&C feasible site, the original study design could not be adopted and the team only cleared all the trailing vegetation in this available site adhering to procedures earlier described by Jacob et al.²³

Data Handling and Analysis

Data on Human Landing catches were recorded in fly collection forms. These forms were checked every day by field supervisor for consistency and accuracy. Number of flies caught on monthly basis was summarized in an Excel database. Relevant entomological indices were calculated based on methods earlier described.^{18,21,22} Percentage of parous flies (PPF) from the three sites were also calculated indicating the estimate between the upper and lower bounds of the confidence interval.

Transcripts of FGDs were read and reviewed by a sociologist to come up with a list of codes which were grouped into themes. A thematic approach was used to analyze the qualitative data.²⁴ Some important phrases were extracted verbatim and used in presentation of the results.

Results

Training of Field Teams

Thirteen field teams were trained; six vector collectors, six slash and clear teams and one field supervisor. Those trained were earlier involved in other health-care delivery activities and had some background on onchocerciasis. Most of the trainees were males with only one female who is a fly catcher.

Larval Collection and Mapping Breeding Sites

Six sites were visited along the river Naam based on the guidance provided by the community (Figure 1). Most of the sites were characterized by huge bare rock outcrops either in the middle of the river or at the banks (Supplementary Table 1). Water gushing through rocks could be seen at some of these sites, and most of the rocks were covered by water due to the high-water level during the time of the visit. At one site (Dogoyabolu) where larvae and pupae were found, rock outcrops in the middle of the water were covered by vegetation, mainly plant climbers. Even island rock outcrops in

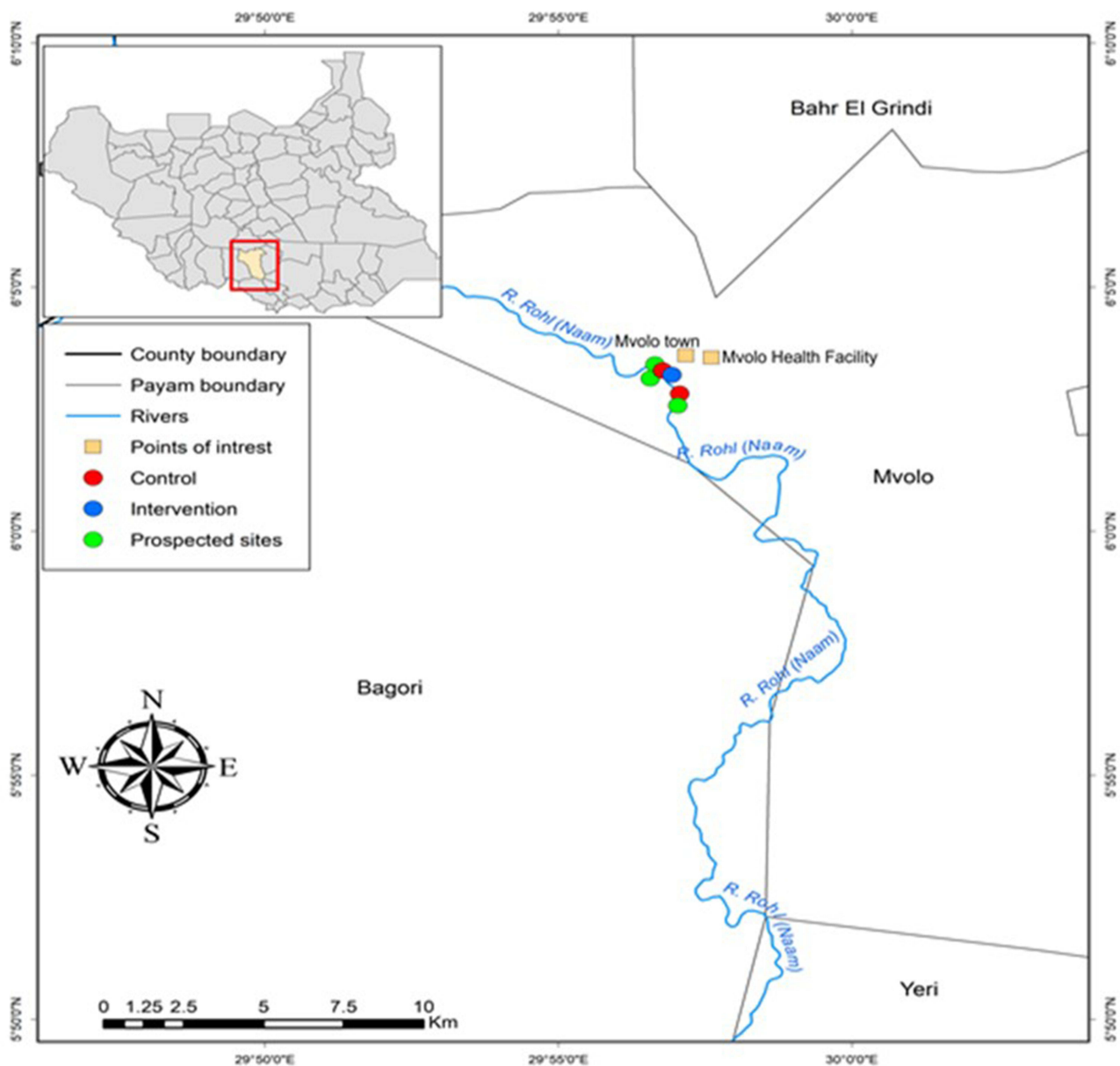


Figure 1 Prospected sites along the river Naam in Mvolo County, Western Equatoria, South Sudan.

the middle of the river were covered with vegetation. Numerous larvae and pupae were collected from this site, and majority identified were *S. damnosum*, the larvae characterized by their prominent dorsal tubercles.¹⁸

Biting Activity of *S. Damnosum* s.l. Along the River Naam in Mvolo County

A total of 2466 female *S. damnosum* s.l. were caught in 14×11 h (06.00–15.00) catches during the four months of catches in the study area. The daily biting rate (DBR) varied from 382.75 flies per man per day in November to 19 flies per man per day in January. The monthly biting rate (MBR) was highest in November with 11,482.5 flies per man per month while the lowest MBR was recorded in January with 589 bites per man per month. The monthly biting density (MBD) ranged from 4210.25 in November to 290 in January. A gradual reduction in biting density and MBR was observed in intervention site compared to the control site. In January 2024, MBR for control was 589 flies/man/month compared to 286.75 flies/man/month in intervention (Table 1).

Diurnal Biting Activity

According to baseline catches conducted from the three sites from 6 to 10 October 2023, it was observed that diurnal biting activity presented two peaks, one at 9:00–10:00 am (at bridge site) and another one at 14:00–15:00 pm (Figure 2).

Dissection of Flies for Parity (Age Grading)

A total of 760 flies were dissected from the three catching sites, and the overall percentage parity rate was 31%. There was not much intersite variation in parity rate (Table 2).

Monthly Biting Rates Pre- and Post-“Slash and Clear” Intervention

The highest MBR pre-slash and clear was 11,482.5 bites/man/month and this reduced to 0 bites/man/month in April 2024, five months after slash and clear (Table 1).

Qualitative Study

Focus Group Discussions

Blackfly Biting

Communities living along the river Naam complained about the nuisance caused by blackfly bites, itching, and wounds caused as a result of scratching. A 37-year-old female participant from Bogori 2 said: “It bites and brings itching and wound on us”. Participants reported that blackflies most often bite in the morning especially between 06:00 am and 08:00 am and between 15:00 and 18:00 pm in the afternoon. A 29-year-old female from Dukaboro said: “From five o’clock to six o’clock is when they will start biting people” A 60-year-old male participant from Gira said: “from four o’clock to five and six o’clock”. It was reported that blackflies bite from May until November. A 40-year-old female from Dukaboro

Table 1 Biting Activity of *Simulium Damnosum* s.l. in Control and Intervention Sites Along the River Naam in Mvolo County, South Sudan

	Ento. indices	2023						2024	
		^a October		November		December		January	
		Contr.	Int.	Contr.	Int.	Contr.	Int.	Contr.	Int.
1	Person days worked	2		4		4		4	
2	Total number of flies caught	325	129	1531	476	534	226	76	37
3	Biting density of flies	893.7	709.5	4210.25	1309	1468.5	621.5	209	101.75
4	Daily Biting Rate (DBR)	81.25	64.5	382.75	119	133.5	56.5	19	9.25
5	Monthly Biting Rate (MBR)	2518.75	1999.5	11,482.5	3570	4138.5	1743.75	589	286.75

Notes: Ento, entomological; contr, control; int, intervention. ^aSlash and clear intervention initiated in the second week.

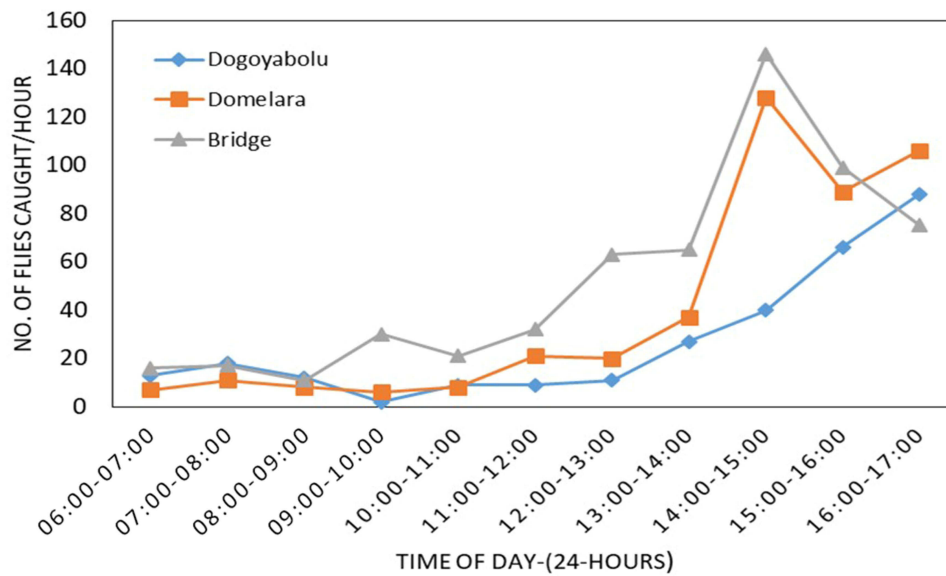


Figure 2 Biting activity of *Simulium damnosum* in Mvolo County (river Naam), Western Equatoria, South Sudan.

said “will start from May, June, and July, within this period they are doing us bad”. Participants did not know where blackflies live, however they see them around water, the bush, mountains, and streams. They reported being bitten during farming, around streams and boreholes. The following residential areas were mentioned as areas where blackflies often bite: Hai Koroson, Minimbere, Bogori 2, Sariwulayat, Domboro, Doseri, Mvolo center, Dukaboro, Domandede, and Fulupi. Children were also reported to be bitten by blackflies. A 33-year-old female from Mukaboro said “even when there are kids on the floor like this though they wear clothes like this, it will bite the child”. Participants from Gira and Dukaboro reported that certain households moved to another place because of frequent blackfly bites. A 39-year-old female in Dukaboro said “so many people leaving their home because of the flies, others are now staying in town”. However, other factors like presence of mosquitoes and lack of clean drinking water were also mentioned as reasons for displacement. A 29-year-old male from Minimbere said: “People shift because of the flies, lack of water and also too much mosquitoes”.

Coping Mechanisms for Blackfly Bites

Most participants cited the use of paraffin to repel blackflies from their bodies. Some would put on long-sleeve shirts and trousers while some use smoke to chase the blackflies. Some parents use mosquito nets to protect their children from blackfly bites. A 25-year-old participant from Dukaboro said “at the river side you have to put on a long sleeves shirt so that they cannot access to your body otherwise you will not work well” and a 46-year-old woman from Dukaboro said “if I want to go the garden I will smear paraffin on my body to keep the flies away and to help the body from itching”.

Table 2 *S. Damnosum* Parity Rate at Three Catching Sites Along the River Naam in Mvolo County, Western Equatoria, South Sudan

County	River	Catching Site	No. of Flies Dissected	No. of Flies Parous	% Parous	95%CI
Mvolo	Naam	Domilara	283	90	32	0.2236±0.04164
			270	89	33	0.2323±0.04277
		Dogoyabolu	207	54	26	0.1430±0.04030
TOTAL			760	233	31	0.2976±0.09176

Slash and Clear Strategy

Most participants accepted the idea of slashing and clearing the vegetation where blackflies breed. “This is good for us because we are human being and we need to be protected” one of the participants in Gira village said. Another 50-year-old male participant from Bogori 2 said “it is good, slashing the bush can reduce them”. Community members expressed their willingness to participate in the “slash and clear” activity. They said that they need to be notified whenever the activity is about to start. “We can do the work by ourselves” a female in Bogori 2 said. However, they mentioned that they will need the tools for slashing the vegetation “if they bring the working tools it will work, it needs things like face mask and gumboot so that they can do the work well” a 27-year-old female stated. Participants in Dukaboro indicated gumboots, slashers, panga, rain coat, over roll and face mask as working tools. Most participants expressed their willingness to support the S&C activity by involving themselves directly or indirectly. Some participants suggested that the selection of volunteers for the activity needs to be done through the community in collaboration with the chief. A 35-year-old female from Dukaboro said that the training provided will be enough to ensure the continuity of the S&C activity “we have really benefited a lot from their teachings and this can help us a lot in order to prevent ourselves from those flies biting us”.

Use of Ivermectin

Participants in Gira, Dukaboro, Minimbere and Bogori 2 agreed that it is beneficial to take ivermectin. A 47-year-old woman in Bogori 2 said “it is also good because once you are bitten by the blackflies your body can be itching and once you take the medicine, you become fine”. Most participants said they take ivermectin to stop the itching. A 37-year-old female in Dukaboro said “the reason why I took the drug was because my body was itching”. However, adverse effects like fever, headache and swollen body parts have discouraged some community members from taking ivermectin. Similarly, other participants from both Gira and Minimbere also experienced body swellings, high fever, and headache after the taking ivermectin.

Some community members mentioned that they were not informed when the mass drug distribution campaign was going to happen and therefore they missed taking ivermectin. A 23-year-old male from Dukaboro said “the bad thing is that they don't announce about the distribution of the drug that's why some people end up not getting”. It was reported that the community directed distributors (CDDs) only visit the households once and therefore people may miss the ivermectin because they were not at home. “they are always coming only once, so if you're not at home you will not get it” a 37-year-old male participant said in Dukaboro. Most participants agreed that they will take ivermectin when it is distributed twice a year, “yes if they distribute the drugs twice a year we can take” participants in Dukaboro responded. Also in Gira participants agreed that it is good to take ivermectin twice a year “we accept that when brought twice a year, we can still take it” a participants from Gira said.

Slash and Clear Implementation

The S&C community vector control was implemented in October 2023 for two days by six volunteers at Dogoyabolu site (Figure 1, blue circle; Figure 3). This was the only site that was fairly favorable for this strategy despite predominant outcrop of rocks in the middle of the river and at the banks. However, this site was not very far away from the two control sites. Breeding of *S. damnosum* in Mvolo is apparently confined to a very short stretch measuring approximately 3–5 km, and in proximity to human settlements.

Discussion

The prospections activities conducted in the river Naam during the dry season in October 2023 confirmed the presence of *S. damnosum* s.l., for 74 years when Lewis visited this site and conducted the first entomological assessment.¹¹ This is an indication that this vector species earlier reported by Lewis¹¹ has persisted in the area (Mvolo) despite various environmental changes that might have taken place both in the river Naam and its surroundings. There was however, only one site (Dogoyabolu) where the immature stages of *S. damnosum* was found in abundance on trailing vegetation. In the middle of the river dominated by rocks of various sizes, trailing vegetation was generally absent. There are strong possibilities that some of the breeding sites could be on the rocks as earlier observed by Lewis.¹¹ During the



Figure 3 Slash and clear at Dogoyabolu site along the river Naam (A) before slash and clear (B) during slash and clear (C) after slash and clear implementation.

prospections, the field team could not detect any young stages on the rocks probably because of the high-water level that had covered most of them. It was only in Domelara site where the exposed rock was seen coated with algae and no larvae or pupae was collected from this site. This kind of scenario was reported in Maridi¹⁴ and Victoria Nile focus in central Uganda.²⁵ Algae is known to reduce the amount of oxygen and makes the site unfavorable for *S. damnosum* breeding.

The monthly biting density of flies recorded in Mvolo along the river Naam is far above the maximum range reported previously by Crosskey²⁶ in Africa, that the highest biting density of *S. damnosum* complex are usually not above 30–60 flies/month/h in savanna and 200 flies/month/hour in forest areas. This is in agreement with earlier observation by Lewis¹¹ in 1949 where he reported a biting rate of over 200 flies/h during the month of May in Mvolo. Comparatively, both the biting density and MBR observed in Mvolo is still lower than the fly density of 9444 flies/man/h and MBR of 58,552.8 bites/person/month reported in Abu Hamad focus in Sudan.²⁷ However, for the case of Mvolo, it is apparent that the blackfly breeding season of May to December earlier reported by Lewis¹¹ seems to have been maintained. This argument is further supported by views of the communities about the blackfly in the area. It was reported that blackflies bite from May until November. A 40-year-old female from Dukaboro said “will start from May, June, and July, within this period they are doing us bad”. Participants did not know where blackflies live, however they see them around water, the bush, mountains, and streams. In 1949, Lewis¹¹ further reported that the river Naam ceased to flow by 13 December and by the end of January there were hardly any blackfly seen along the river Naam. This was slightly different in the current study because biting density of 209 flies/month/h and MBR of 589 flies/man/month were still recorded in January 2024, indicating some slight seasonal change in blackfly breeding in the Mvolo area.

The diurnal biting pattern of the flies in Mvolo showed a bimodal peak of activity – a morning peak and an evening peak. The morning peaks occurred between 09:00 and 10:00 only at one site (at bridge) and the evening peaks occurred between 14:00 and 15:00, however, in one site apparently there was no clear pattern observed. This is consistent with what was earlier reported by Lewis¹¹ that *S. damnosum* in Mvolo bit from before dawn till after sunset and usually more during the 2 h before sunset than other times. These findings are in agreement with other studies conducted in Maridi,¹⁴ Nigeria,²⁸ and Sudan.²⁷ It has been reported that hourly biting patterns varied among sibling species.²⁷ *S. damnosum* s.s./*S. sirbanum* showed a distinct bimodal pattern, peaking in the morning between 07:00 and 09:00, and in the afternoon between 15:00 and 18:00. These basic patterns reported from various sites, however, were found subject to some seasonal variation. In the rainy or cold weather, the morning peak may be suppressed and much smaller than the afternoon peak, and the midday lull may not be clearly defined. For the case of Mvolo, it was observed that the lowest proportion of flies came to feed in the morning peak and more in the afternoon. This findings on biting peaks in Mvolo agreed very well with the results obtained during FGD with communities along the river Naam. Participants reported that blackflies most often bite in the morning especially between 06:00 and 08:00 and between 15:00 and 18:00 in the afternoon. A 29-year-

old female from Dukaboro said “from five o'clock to six o'clock is when they will start biting people”. A 60-year-old male participant from Gira said “from four o'clock to five and six o'clock”. The cause of biting activity peaks is still poorly understood, but it has been suggested that an innate clock rhythm may be involved.²⁶ This apparent biting pattern has epidemiological implications because the biting peaks correspond almost exactly to the working habit of the people living along the river Naam in Mvolo, most importantly the subsistence farmers and fishers. During the FGD the communities also confirmed that they are bitten during farming, around streams and boreholes. The following residential areas were mentioned as areas where blackflies often bite: Hai Koroson, Minimbere, Bogori 2, Sariwulayat, Domboro, Doseri, Mvolo center, Dukaboro, Domandede, and Fulupi. Children were also reported to be bitten by blackflies. A 33-year-old female from Mukaboro said “even when there are kids on the floor like this though they wear clothes like this, it will bite the child”. This correspondence between the biting activity pattern of the parous flies and the working habit of the communities maintain an uninterrupted link in man-fly contact and the risk of disease transmission by infected parous flies. Lewis¹¹ during his visit in Mvolo reported that most settlements were close to the river Naam (<700 m) including a police post. However, due to the intense bite of blackflies, all these settlements had to be moved away. During the FGD this was also alluded by the participants. From Gira and Dukaboro participants reported that certain households moved to another place because of frequent blackfly bites. A 39-year-old female in Dukaboro said “so many people leaving their home because of the flies, others are now staying in town”. Despite some of the communities moving away, during peak transmission season they need to have a coping mechanism to prevent the high number of fly bites. A 25-year-old participant from Dukaboro said “at the river side you have to put on a long sleeves shirt so that they cannot access to your body otherwise you will not work well” and a 46-year-old woman from Dukaboro said “if I want to go to the garden I will smear paraffin on my body to keep the flies away and to help the body from itching”.

The mean parity rate observed in the three sites along the river Naam in Mvolo was above 30%; this was higher than the 13% earlier reported by Lakwo et al in Maridi.¹⁴ This low proportion of the parous flies earlier observed in Maridi and recently along the river Naam in Mvolo could be a reflection of local production of the flies. However, in contrary, the Mvolo parity rate (31%) is lower than the 56% reported by Baker and Abdelnur²⁹ along the river Bussere in Wau, Barh el Ghazal, South Sudan, 42.5% in Asubende village, Brong-Ahafo region, Ghana³⁰ and 46.1% along the river. Osun in Nigeria.³¹ These variations in parity rate are mainly attributed to seasons, parity rates have been observed to be higher in the dry season than the rainy season.³¹ In Mvolo, entomological assessment was conducted towards the end of the dry season (October) but the water level was still relatively high, therefore the parity rate reported may still be within that of rainy season. In the FGD with communities along the river. Naam, they were quite knowledgeable about the seasons when the blackflies are many. It was reported that blackflies bite from May until November. A 40-year-old female from Dukaboro said “will start from May, June, and July, within this period they are doing us bad”.

The S&C method was shown to be very successful in Maridi County¹⁵ and Uganda.²³ However, its impact on areas with multiple breeding sites remains to be determined. The current S&C site on the river Naam is a typical example where *S. damnosum* breeding is on trailing vegetation, on the rocks and other immersed substrates under water. The communities along the river Naam seem to have embraced this strategy and are ready to implement it once provided with the necessary tools. A 50-year-old male participant from Bogori 2 said “it is good, slashing the bush can reduce them”. Community members expressed their willingness to participate in the “slash and clear” activity. They said that they need to be notified whenever the activity is about to start. “We can do the work by ourselves” a female in Bogori 2 said. However, they mentioned they will need the tools for slashing the vegetation “if they bring the working tools it will work, it needs things like face mask and gumboot so that they can do the work well” a 27-years-old female stated. The impact of this intervention that is being evaluated prospectively will be important to guide programs in scaling up this strategy. However, other vector control strategies might even be more challenging to implement, not only due to the rainy season²⁹ but also due to their substantial costs, especially considering the poverty in South Sudan. For instance, the vector control trial using temephos (Abate[®]) by Baker and Abdelnur in 1983–1984 on the river Bussere in Wau, Bahr el Ghazal, south Western Sudan was over USD \$15,000, and the cost would increase each year.²⁹ Thus, increasing the frequency of CDTI may be the most practical approach to potentially preventing the onset of OAE. This view on medication was similarly expressed by communities in Mvolo. Most participants agreed that they will take ivermectin when it is distributed twice a year, “yes if they distribute the drugs twice a year we can take” participants in Dukaboro

responded. Also in Gira participants agreed that it is good to take ivermectin twice a year “we accept that when brought twice a year, we can still take it” a participant from Gira said.

Despite the availability of ivermectin given twice a year, the communities residing along the river Naam can still adopt some preventive measures against blackflies. This will include, building homes away from the river, avoid visiting river during peak biting hours and putting on long-sleeve clothes.

Conclusions

S. damnosum active breeding was confirmed along the river Naam in a stretch of 3–5 km close to human settlements. A high biting density was recorded with two diurnal biting peaks. FGD showed that local communities were very willing to support a S&C intervention. Therefore, community vector control by S&C was implemented at one site, and will be prospectively monitored. The S&C intervention may protect the local population from blackfly bites but reinfestation from nearby sites may reduce the impact of the intervention. More support from national and international organizations will be crucial in addressing the issue of onchocerciasis control and elimination in this area.

Abbreviations

AMREF, African Medical and Research Foundation; CDTI, community directed treatment with ivermectin; CI, confidence interval; DBR, daily biting rate; FGD, focus group discussions; FMH, flies per man hour; GPS, global positioning system; HLC, human landing catches; Lat, latitude; Long, longitude; MBD, monthly biting density; MBR, monthly biting rate; MDP, monthly population density; MOH, Ministry of Health; NTD, neglected tropical diseases; OAE, onchocerciasis-associated epilepsy; PCR, polymerase chain reaction; PPF, proportion of parous flies; R2HC, research for health in humanitarian crises; RSS, Republic of South Sudan; S&C, slash and clear; UNHCR, United Nations High Commission for Refugees; WHO, World Health Organization.

Data Sharing Statement

The data reported in the submitted manuscript are provided as part of the submitted article.

Ethics Approval and Informed Consent

This study obtained ethical approval from the Ministry of Health of South Sudan (MOH/RERB/P35//15/05/2023-MOH/RERB/A/35/2023) and the University of Antwerp, Belgium (BUN B3002023000045). The study was conducted in conformity to the Declaration of Helsinki and the South Sudan Ministry of Health Ethical guidelines regarding handling of human subjects. There were community engagements before commencement of the two study components. Written informed consents were obtained from individuals who participated in key informant interviews and FGD after detailed explanations in their local languages about the study. The participants' informed consent included the publication of anonymized responses.

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Author Contributions

All authors made a significant contribution to the work reported, whether that is in the conception, study design, execution, acquisition of data, analysis and interpretation, or in all these areas; took part in drafting, revising or critically

reviewing the article; gave final approval of the version to be published; have agreed on the journal to which the article has been submitted; and agree to be accountable for all aspects of the work.

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Disclosure

The authors have no competing interest in this work.

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