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Investigation of *Simulium* Specie Infestation in Veritas University Metropolis, Abuja, Nigeria

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Abstract:

Simuliumdamnosum, commonly known as black flies, are the mechanical vectors of *Onchocerca volvulus*. Onchocerciasis (or river blindness), is a disease caused by this specie of microfilariae in humans in the tropical and subtropical regions of the globe. Critical public health and socio-economic problems arise due to Onchocerciasis, an insidious non- fatal filarial infection. With itching and disfiguring of skin with nodules on bony areas, serious eye lesions even blindness happen due to this. This study therefore investigated *Simulium*specie infestation in the campus of Veritas University Abuja and its environs with a view to establishing the possible occurrence of river blindness disease in the area. The study involved identification of possible breeding sites, searching and collecting eggs, larvae and adult blackflies, identification of the species through examination of the eggs, larvae, pupae and adult flies and dissection of the adults for presence of microfilariae of *Onchocerca volvulus*. The result showed that 365 adult flies were caught which were identified into 244 females and 131 males, 209 eggs and 113 larvae were also collected. The presence of the eggs, larvae and adult blackflies indicated not only their presence but also the active reproduction of these flies in the environment. The adult flies were then dissected for presence of microfilariae. 60/49.6% of the 131 adults *Simuliumdamnosum* complex were found to be infected with microfilariae in their head and thoracic regions meaning that the *Simulium* species present are infective and can bring about river blindness disease in the area and should therefore be checked.

Keywords: Investigation, simulium, infestation, onchocerciasis, veritas metropolis

1. Introduction

Morbidity and mortality happen every year due to insect-transmitted diseases in tropical countries. African countries, especially river blindness (onchocerciasis) has been caused by *Simulium* species transmit *Onchocerca volvulus* (5). Many problems of skins and eyes happen due to the parasite of onchocerciasis (4, 23). Onchocerciasis has been described as an insidious non-fatal filarial infection that causes not only blindness but also life-long human suffering and grave socio-economic problems (23). The disease is commonly called 'river blindness', because the blackflies which transmit the disease are strictly associated with riverine areas. The disease affects millions of people in the world, but mostly in Africa centuries, and therefore it is considered to be of high economic importance (19). According to WHO (14) report, Nigeria has the highest number of onchocerciasis victims than any other country in the world, and makes about 100,000 out of the 268,000 global known cases. Infection has spread on 37-86 million people in 35 Onchocerciasis endemic countries of Tropical Africa, Latin America and Yemen are infected with *Onchocerca volvulus*, while 2 million and 50,000 of these exposed individuals are blind and visually-impaired, respectively [21,19]. One quarter of the global infection has been caused by onchocerciasis in Nigeria (14). Landslides of rivers and streams had been noticed for immense morbidity of the infection in the country (9). Most rivers and fast flowing streams had become breeding sites for *Simuliumdamnosum* complex (22, 11). More than 20 million people infected, and millions blinded in West Africa and South America (17). A study by Hazarika *et al.*, (8) found that even in countries like India, where the parasitic load of *Onchocerca volvulus* is low, *Simulium* can still be found to bite and crawl on the skin of individuals, causing intolerable nuisance. Similarly, besides their painful bites which often lead to loss of significant amount of blood, the wounds can also carry secondary infections by serving as route for bacteria, viruses, protozoa, nematodes, etc., which the fly sometimes carry on their bodies (26). Furthermore, in humans, the bites of some blackfly species can cause allergic reactions known as "blackfly fever" or simuliotoxicosis. This condition is characterized by swelling, itching, haemorrhage and oedema which in severe cases, requires medical attention (17). Onchocerciasis has equally been regarded as a disease of rural areas, even though it is found also in urban centers due to rural-urban migration. Those most at risk of contracting the infection are mostly travelers, the missionaries and peace-corps volunteers who are often exposed to bites of black-fly in endemic areas.

The incidence of the disease increases with increase in human activities along the river valleys, which are the habitats of *Simulium* vectors. Human activities which could boost the infestation of *Simulium* have been identified as bathing, swimming, farming, hunting, and other recreational activities carried out along and in the vicinity of the streams. The effects of onchocercal infection are not only reflected in individuals, but also on the economy of the countries at large, since the disease normally leave debilitating effects on humans, incapacitating a huge number of working population, and thus render them incapable of working again.

Some of the ways of eradicating this vector-borne infection include destruction of the larvae of the vector (the blackfly), destruction of the adult parasite (*Onchocercus volvolus*) in hosts' body, and prevention of vector-host contacts (12, 26). Various synthetic chemical agents have been used to destroy the larvae of black flies, but this chemical control method does not usually produce impressive result (8). Besides, this effort requires treatment of all potential breeding sites or rivers with suitable insecticides. But, the major problem with such control method is that non-target organisms are also affected, thus leaving a negative feedback on the ecosystem (16). Secondly, such methods are daunting when a large area is infested with blackfly larvae. Although control of adult blackflies is said to be most desirable, it is less feasible because of their frequent migration and the long flight range of the vector (5).

Simuliids-biting generally occurs outdoor during daytime hours, and prefer to bite the lower exposed body parts (20, 8). Different simuliid species may have their own preferred biting periods (20).

In animals, blackflies have also been incriminated in the spread of leucocytozoonosis, bovine onchocercosis, the cytoplasmic polyhedrosis virus, the iridescent virus and vesicular stomatitis virus (17).

Deaths of animals, especially livestock had been happened due to attack of blackflies like tsetseflies (*Glossinaspp*). Many economic slowdowns had been caused by haematophagous *Simulium* species (28). Many studies also had been conducted South Africa (27). Other reports confirmed that cows can lose their udders and sheep their ears as a result of secondary infections arising from blackfly wounds (17). Ground larviciding had been mentioned as one of the most effective means for control. This concentrates containing up to 33 % DDT have given the best results. They have been applied at variable rates between 0.03 ppm/30 min and 0.5 ppm/30 min, depending on local conditions, and it has been found that at these levels, DDT has proved to be non-toxic to fish. Rivers need to be disinfected once in 7-10 days even throughout the rainy season, and for periods of not less than 3 months.

Crosskey, (15) in his study highlighted that the presence of hard Precambrian with rocks, outcrops in the streambeds as forming favorable breeding sites for especially *Simuliumdamnosum* complex. The challenge associated any form of control involving the use of aircraft is expensiveness, and might well be precluded on these grounds, except in exceptional circumstances. Female adults of some simuliid species generally act as vectors of some parasites and pathogens, like *Onchocerca* spp. in humans and cattle and *Leucocytozoon* spp. in birds (28). Generally, America, Mexico, Yemen, Brazil, Venezuela, Ecuador, Colombia and Africa (13) had been seen most suffering nations due to the flies. West Africa and also in some countries in Eastern and Southern Africa, *Simuliumdamnosum* has been noticed as a major vector which has been mentioned by Mustapha *et al.*, (18,9). Most of the previous researches had focused on the northern part of the Nigeria (11).

In 1974, World Health organization (WHO) had launched Onchocerciasis Control Programme (OCP). It started with the control of Onchocerciasis through eradication of the black fly vector in seven endemic countries of West Africa including Nigeria, using spraying dichlorodiphenyltrichloroethane (DDT) mainly in river breeding sites. But due to high toxic agents many side effects had been noticed inside human body which control the scourge of onchocerciasis using diethyl carbomazine (7, 27). Later, in 1987, ivermectin (Mectizan) had been introduced by a pharmaceutical firm (Merck) as a safest measurement (14). 32 endemics States and the Federal Capital Territory (FCT) (19) started the mass ivermectin (Mectizan) treatment of onchocerciasis.

2. Materials and Methods

2.1. Study Area

This study was carried out in Veritas University Campus, Bwari Area Council, Abuja, and its environs, Zuma I and II, and Garba, from February to July. Bwari Area Council is one of the six Area Councils in the Federal Capital Territory in the North Central Geopolitical Region of Nigeria. Veritas University was founded by the Catholic Bishops Conference of Nigeria. However, the University commenced admission of students in October Nigeria, in 2008 and today it has a population of over 3,000 people. Bwari Area Council where the Veritas University is located is found in the North Eastern part of the F. C. T., and is situated between latitude 7°23' 0" North of the Equator and Longitude 9°17' 0" East with a vast land of 1100km². It has a guinea savannah type of vegetation, with raining season stretching from April to October. From November to March, dry season has been noticed and the temperature ranges from 30-37°C yearly with the highest temperature experienced in the month of March. The indigenes of Bwari Area Council are the Gbagi's and Gwari's and they are mostly farmers by occupation.

2.2. Sources and Method of *Simulium* Specimen Collection

Samples were collected from the fast-flowing streams that transverse the Veritas University Campus and flowing behind the male hostels, the auditorium and the Vice Chancellor's office, and the ones are also found in Zuma I, Zuma II and Garba. The streams were marked A, B, C, D, E and F, respectively, for the purpose of this research.

The *Simulium* species were collected by scooping the stream vegetation and those at the shore or bank with butterfly nets. The *Simulium* specimens caught were immobilized by dipping the scooping nets in water, and removing

same after approximately 10 sec., after which the flies were handpicked, and dropped in labeled specimen bottles, containing formalin, and conveyed to the laboratory.

2.3. Sex and Species Identification of Simulium

The adult blackflies collected were subjected to sex and species identification using the morphometric and cytological identification keys described by Crosskey (15), Meredith *et al.*, (10) and Post *et al.*, (25). Using a combination of these keys, only *Simuliumdamnosum complex* was identified in the collection based on the possession of tubercles on the dorsal region of their larvae, possession of scales on the thoracic proleg as well as chromosomal variation in the 6th and 7th instar larvae of the blackfly since according to Post *et al.*, (25) 6th and 7th instar larvae give better information on the chromosomes. Ventral dissection of the head, thorax and abdomen was done under a dissecting microscope. The polytene chromosomes were prepared from the larval silk glands and examined using the oil immersion lens. The banding patterns of the three pairs of chromosomes were accordingly scored. The sexes of the *Simuliumdamnosum* were identified by the fact that male adults possess toothed mandibles and maxillae, eyes which are much larger touching above antennae, hence described as holoptic, compact and inconspicuous male Terminalia, a blacker body appearance and a reflective silvery mark on their abdomen to aid recognition by the female partner during mating. The female adults were identified by possession of larger mandibles bearing a row teeth for cutting and bloodletting as described by Crosskey (15), possession of smaller ommatidia which makeup the eyes, possession of dichoptic eyes (in other words, eyes which are well separated from the antennae that lie below the eyes), possession of single subspherical spermathecal and a grayish body appearance.

2.4. Dissection and Microscopic Examination Of The Adult Females Flies

The head, thorax, and abdomen of the identified female blackflies were dissected using dissecting microscope and pins, and examined for the presence of microfilariae of *Onchocerca volvulus* using X10 objectives lens. This process was repeated with all the females. The larval stages (L₂-L₃) of *Onchocerca volvulus* were isolated. The Microfilariae isolated from each morphological segment of the flies dissected were stained with haematoxylin, and then identified, counted and recorded appropriately.

3. Results

3.1. Result of Simulium Collection

Sample collection was carried between February and July, 2019. A total of 244 adult *Simulium* were collected during the field survey of the six streams, and this number comprised of 131/ 53.7% females and 113/46.3% males. Also harvested were 209 eggs and 113 larvae of the flies (Table 1).

Location	Time	Eggs	Larvae	Adult Flies	
				Male	Female
Stream A	Feb-July	25	19	24	28
Stream B	Feb-July	66	20	20	26
Stream C	Feb-July	30	22	23	23
Stream D	Feb-July	28	21	17	20
Stream E	Feb-July	42	16	14	18
Stream F	Feb-July	36	15	13	16
Total	Feb-July	209	113	113	131

Table 1: The Number of Eggs, Larvae and Adult Simulium Flies Collected During the Study

Key: Stream A = the stream behind the Vice Chancellors office; Stream B = the stream flowing from behind the Auditorium; Stream C = the stream behind the male Hostels; Stream D = the stream in Zuma I village; Stream E = the stream in Zuma II village; Stream; F = the stream in Garba village.

3.2. Result of Simuliumdamnosum dissection for microfilaria

The result of dissection of the different body regions of the 131 female adult *Simuliumdamnosum* showed that only the heads region of 23/17.6% blackflies contained onchocerca microfilariae, 37/28.2% were found with onchocerca microfilariae in their thorax while the abdomen had none (Table 2).

Body Parts	Positive	Negative	Total
Head	23/17.6%	98/80.9%	131
Thorax	37/28.2%	84/69.4%	131
Abdomen	0	131/100%	131
Total	60/49.7%	313/250.3%	393/300%

Table 2: The Result of Dissection of the Different Body Regions of the Flies for the Presence of Microfilariae of *O. Voluvulus*

The monthly collections of *Simuliumdamnosum complex* specie showed that while 39 eggs, 21 larvae and 29 adults were collected in the month of February, in March 43 eggs, 28 larvae and 57 adults were collected. In April 48 eggs, 26

larvae and 75 adults' blackflies were collected whereas in May 47 eggs, 24 larvae and 54 adults were obtained. Collection in June showed 32 eggs, 14 larvae and 24 adults *Simuliumdamnosum* while in July there were no eggs, no larvae but only 5 adults.

Month	Egg	Larvae	Adult Female	Adult Male	Total
February	39	21	18	11	89
March	43	24	33	28	128
April	48	26	40	35	149
May	47	28	26	24	125
June	32	14	12	12	70
July	-	-	2	3	5
Total	209	113	131	113	566

Table 3: Monthly Collections of *Simulium Species*

4. Discussion

The results obtained have shown that *Simulium* specie is exist in the streams on the campus of Veritas University, and streams in Zuma I, Zuma II and Garba, *Simuliumdamnosum* was identified as the species that exist in the streams surveyed, and no other species co-exist with it. The black flies dissected and examined under the microscope for the presence of microfilariae of *Onchocerca volvulus*, of 131 female adults dissected 60/49.7% were found infected with microfilariae in their head or thoracic regions. The proportion of *Simuliumdamnosum* complex found in the study area is a clear indication that the streams and the areas are highly infested with blackflies. The low number collected in the month of February can be attributable to the fact that there were no frequent human activities such as farming activities in and around the streams because it is usually human activities in a place that attract them. This assertion agreed with the finding of research conducted by (2) on the abundance of the *Simulium* species in the river Muvur, Mubi in Adamawa state, where he recorded 310 flies with 89 infected with *O.volvulus*. This research has equally confirmed earlier reports that the high numbers of *Simulium* in March and April may have been due to high level of farming activities and other human activities such as swimming and fishing all of which were ongoing in and around the river, besides, there was no rainfall. The contrary was the case in the onset of the rains in May and June because the rains normally come with violent windstorm which probably sweeps away the flies thereby making it difficult for their eggs and larvae of the fly to survive. Another factor that may have contributed to the reduced fly number in the month of July might be heavy rain falls and the accompanying windstorm which may have compelled the adult flies to flee from the streams to a more secured habitat to hide. It can also be argued that probably because the streams used in this study are shallow, therefore they were not able to adequately support the flies to breed abundantly. This observation has collaborated the findings of previous studies on the distribution of black flies in Bahia region of Brazil by Landeiro, *et al.*, (1) they asserted that there is a relationship between the number of *Simulium* species and the size and depth of the breeding sites.

Besides onchocerciasis disease which is associated with infestation of *Simuliumdamnosum*, another fear about the presence of blackflies in the streams surveyed in Veritas University Campus, Zuma I, Zuma II and Garba is the possible existence in the area of another species of blackfly which attacks sheep and other forms of livestock and which together with the human species normally cause high economic losses through reduced efficiency of agricultural and industrial workers, in addition to interference in recreation and the consequent reduction of real estate values. It could be this kind of fear that made Njebuome *et al.*, (19) and Tekie *et al.*, (21) to warn that annoyance and discomfort caused by haematophagous *Simulium* species are severe enough to warrant large-scale control operations. How physical destruction of the teats of some cows and reduction in milk production of up to 35 kg milk per week per cow (30-50% reduction) and 10-15% reduction in egg production in poultry has been noticed in Western Nigeria had been highlighted by Basanex *et al.*, (29). Moreover, the infections of udders and sheep their ears as a result of secondary infections caused from blackfly wounds (17) also have been mentioned. Similar calamity can befall animal farmers in this part of the world and that is why so something must be done urgently before it would be too late. Regarding the intimacy between *Simulium* species generally with warm-blooded vertebrates (humans and other mammals) it could be suggested that odors from these animal skin are attractive to blackflies. This may mean that blackflies have efficient olfactory system which plays important role in their host-detection process. Perhaps, the olfactory sensilla of the antennae of blackflies carry olfactory receptor neuron which is responsible for detecting human odors.

5. Conclusion

The study has established the presence of *Simuliumdamnosum* complex in the streams in Veritas University Campus and those in its environs, and the presence of microfilariae of *O. volvulus* in the head, thorax all of which are clear indications that both humans and livestock are potentially at risk of contracting onchocerciasis any moment since it is infected blackflies that transmit onchocerciasis to humans and other mammals, if human, recreational and agricultural activities proliferate in the area.

6. Recommendations

The use of biological control methods such as *Bacillus thurengensisisraelensis*(Bti) in the form of mosquito dunks or aquabacs can destroy the blackfly eggs in their breeding sites to prevent them from multiplying. Fumigation of the

infested streams and streamline areas can help reduce *Simulium* infestation in the area. Periodic administration of metizan drug (an anti-filaria drug) can help prevent the development of the microfilariae in infected humans.

Human activities in and around the streams must be reduced to stop by discouraging individuals from visiting the streams but, if they must do so, then they must wear light-colour protective clothes.

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