STUDIES ON THE ANTHROPOPHILIC SPECIES OF *SIMULIUM* IN THE GHIBE RIVER VALLEY

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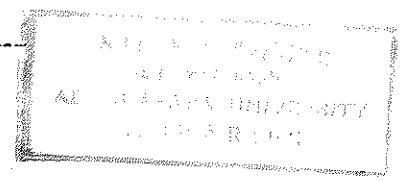

by
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ABSTRACT

Studies on the anthropophilic Simulium species of the Ghibe River Valley were carried out from December, 1981 to November, 1982.

Monthly pupal surveys revealed the presence of nine Simulium species, of which S. damnosum and S. gibense were dominant. These two species were found to be man-biting, the former being strongly anthropophilic, while the latter was highly zoophilic. S. gibense was encountered only in the wet season (June-October), whereas S. damnosum was present during most of the year with a peak in October, at the end of the wet season.

The diurnal biting activity of S. damnosum had a bipeaked fluctuation, one in the morning from 0800-1200 and the other in the afternoon from 1700-1900 hrs.

Dissection of 3418 female S. damnosum revealed an annual parous rate of 64.2% (2193); 107 (4.9%) of these carried developing filarial infections while 69 (3.1%) and 37 (1.7%) harboured infective larvae of Onchocerca volvulus and other filariae, respectively. Dissection of 95 female S. gibense collected on human bait showed an annual parous rate of 94.7% (90). None of these were infected. Of the 1492 S. gibense collected on human bait among grazing cattle, 13(0.87%) and 4(0.27%) carried developing and infective larvae, respectively.

Transmission of O. volvulus seems to be mainly due to S. damnosum with a peak in October. About 3567 O. volvulus larvae were estimated to be transmitted per man per year by S. damnosum.

Filarial larvae resembling those of O. volvulus have been isolated from S. gibense biting man among cattle. The exact origin and identity of these parasites require further study.

1. INTRODUCTION

Blood sucking blackflies (Diptera: Simuliidae) are known to affect the health of man and animals in several ways. They exert their importance by acting as vectors of disease, as a biting nuisance, and as annoying pests (Jamnback, 1976).

One of the most important human diseases that is transmitted by blackflies is onchocerciasis. It is caused by the filarial worm, Onchocerca volvulus (Leuckart) (Nelson, 1970). Onchocerciasis is a serious disease throughout the greater part of tropical Africa, parts of central and south America, Yemen and Saudi Arabia (WHO, 1966, 1976; Chumbley, 1980). The seriousness of the disease can simply be realized from its other name, "River blindness".

In Africa, the disease is widespread and some 90% of the onchocerciasis cases in the world are restricted to tropical Africa alone (WHO/AFR, 1978). The largest and the worst endemic areas of onchocerciasis occur in west Africa, particularly in the savanna belt. Here, ocular complications including blindness due to the disease, are so high that the socio-economic repercussions are most common and serious. The fear of blindness and also the bite of Simulium have contributed to the depopulation of many fertile river valleys (WHO, 1966). For example, 10% of the territory of the upper Volta Republic, comprising the most fertile and best irrigated soils, are actually devoid of humans (Le Berre et al., 1978).

Over most of Africa, the vector is the man-biting S. damnosum s. l. which breeds in turbulent waters of small to large rivers. In some central and east African regions S. neavei is also man-biting

and an important vector; locally other species may also be involved in transmission of onchocerciasis (Crosskey, 1973).

In Ethiopia, onchocerciasis is widespread and endemic in south-western, western, and north-western parts of the country. Its distribution extends from the Omo valley in the south to the Atbara river system in the north and stretching westwards to the Sudan border (Torrey, 1966; Oomen, 1969a,b; Iwamoto et al., 1973; TenEyck, 1973; Lester and Tsega, 1974; Mengesha and Jembere, 1975; DeSole and Walton, 1976; DeSole and Kloos, 1976; Mengesha and Tiruneh, 1977; Woodruff et al., 1977). In the south-west, the disease is highly endemic in the coffee growing areas (Oomen, 1969a,b; DeSole and Kloos, 1976).

Among the several anthropophilic Simulium species recorded in various parts of Ethiopia (Grenier and Ovazza, 1956; Oomen 1969a,b; Ogata et al., 1970a; Kaneko et al., 1973; Tanaka et al., 1973; Uemoto et al., 1977; White, 1977) only S. damnosum s. l. and S. ethiopiense (a neavei group member) are regarded as the most important vectors of onchocerciasis (Raybould and White, 1979). In addition, S. gibense has recently been documented as the third man-biting species in Ethiopia (Uemoto et al., 1977). Other species with some anthropophilic tendencies are also known to occur (Van Someren, 1944; Ogata et al., 1970a; White, 1977).

On epidemiological grounds, Grenier and Ovazza (1956) and Oomen (1969a,b) postulated S. damnosum s. l. as the most important vector of onchocerciasis in Ethiopia. Their conclusion was presumably

based on the high biting density of this species as well as on its occurrence in large parts of the endemic areas.

The first strong evidence for S. damnosum s. l. to be a vector in Ethiopia came from the work of Tanaka et al. (1973) who isolated infective O. volvulus larvae from natural populations. This was later confirmed by White (1977). Other than these, data on O. volvulus infection in S. damnosum s. l. is lacking

On epidemiological grounds again, S. ethiopiense is also believed to play a secondary role in the transmission of O. volvulus, particularly at high altitudes in south-west Ethiopia (Oomen, 1969a, b; Raybould and White, 1979). No O. volvulus larvae have yet been isolated from natural populations of this species. However, its vector potentiality for onchocerciasis has been proven experimentally (Schmidt, M.L., pers. comm. to White, G.B., 1977).

With regards to S. gibense, no further study has been done since it was first reported by Uemoto et al. (1977). Because of its occurrence in endemic areas, Uemoto et al. (1977) and Raybould and White (1979) have emphasized the need for future work towards elucidating the vector status of this species.

Before considering any control programme, it would be necessary to establish the vector status of all the anthropophilic Simulium spp. that are found in a given area. In addition, knowledge of the identity, biology and seasonality of both the immature and adult stages will be required. Such information will be crucial to

determine where and when to apply insecticides as well as to estimate costs that will be required for the operation.

As far as I know, systematic studies on seasonal variations of the man-biting Simulium spp. have not been made in Ethiopia up to now. Kaneko et al. (1973) and White (1977) have made some observations on the biting cycles and density of S. damnosum s.l. and S. ethiopiense during some selected seasons of the year. Although these observations are useful, they are not quite sufficient to give a clear picture of the seasonal variations in the biting behaviour and density throughout the year.

Observations on O. volvulus infection rate in the fly populations as well as its seasonal course are essential to understand the importance of the biting Simulium and the epidemiology the disease in an area (WHO, 1976). Such data on the man-biting species of Simulium in Ethiopia are lacking, and that is one of the reasons why the role of some of the potential vectors (e.g. S. ethiopiense) in nature remain unclear. The only available data in this respect are those of Tanaka et al. (1973) and White (1977) both of which are results of brief observations.

The Ghibe river valley (Addis Abeba - Jimma main road) and the areas around lie in the endemic area of onchocerciasis in Ethiopia (Grenier and Ovazza, 1956; Oomen, 1969a,b; WHO, 1976; Raybould and White, 1979). During a reconnaissance survey in October, 1981, I personally observed a few cases (4) from labourers in this area, although the cases may well have acquired infection elsewhere. In

this river valley, the presence of man-biting S. damnosum s. l. has been previously reported by various workers (Grenier and Ovazza, 1956; Ogata et al., 1970b; Kaneko et al., 1973; Mebrahtu et al., 1980). Furthermore, a new man-biting species, S. gibense has been also reported from this river valley (Uemoto et al., 1977).

The present investigation was carried out in the Ghibe river valley with the aim of determining the anthropophilic Simulium spp. of the area; their seasonal variation, biting cycles, infection rates with Onchocerca or any other filariae, and their role in the transmission of onchocerciasis.

2. LITERATURE REVIEW

2.1 Simuliids-Recognition, Biology and Ecology

Simuliids, also known as "blackflies" are small and stout flies measuring between 1 and 5 mm in length. They may be recognized by the strongly humped thorax, broad and transparent wings and short horn-like antennae. They are usually black, although some species in central and south America are conspicously pale yellow or orange (Dalmat, 1955; Gordon and Lavoipierre, 1962; Crosskey, 1973).

Simuliids have a world wide distribution and are usually found where there are rapid streams and rivers for the immature forms. Most members need a slow or fast flowing water, but a few African species area capable of development in stagnant waters (Crosskey, 1973). The type of river or stream inhabited by Simulium differs markedly from one species to another (Laurence, 1976).

The eggs of Simulium are often laid in masses of about 150 to 600 on partially submerged substrates (eg. vegetation or stones) in running water (Crosskey, 1973). The larvae are found attached to the substrates by circlets of hooks on the posterior ends of their abdomens. They hang downstream, trapping food by means of a pair of brush-like structures (cephalic fans) on the head (Laurence, 1976). Pupation takes place in a cocoon, constructed from silk by the salivary glands of the mature larva (Crosskey, 1973). On the substrates, the pupae orient themselves in such a way that the respiratory filaments and the open end of

the cocoon point downstream. This is believed to be an adaptation to their way of life (Laurence, 1976). Some members have their larvae and pupae always attached to other creatures such as crabs and prawns (Raybould, 1969; Lewis et al., 1969; Disney, 1969). This association is believed to be an adaptation for transporting the fixed larvae and pupae (McCrae, 1969). Adults emerge from pupae, usually with males first (Jamnback, 1976). The life cycle from oviposition to adult emergence in tropical conditions is about 2 weeks. In temperate regions this will take about 4 to 8 weeks, although in severe winters there may be only one generation (Gordon and Lavoipierre, 1962; Crosskey, 1973). The maximum longevity of adult simuliids is believed to be about 1 month, although some species (eg. S. metallicum) can live much longer (Crosskey, 1973).

The male simuliids do not feed on blood as they cannot bite, but the great majority of females are blood-suckers from a number of animal hosts, and herein lies the importance of these insects (Crosskey, 1973).

2.2. Simuliids as Biting Pests

All simuliids that take blood-meals may be considered as pests (Fallis, 1964).

In North America, the bites of Simulium venustum is known to cause lowered milk production and loss of weight

(Fallis, 1964). Attack by S. canonicolum is also associated with decreased egg production in poultry in California (Anderson and Voskuil, 1963). In Europe, Eichler (1971) noted cattle licking their umbilici during periods of heavy attack by S. ornatum. In certain animals the bites of Simulium may result in illness or death (Fallis, 1964).

Man is also considerably affected by the bite of these insects throughout the world. The mass-biting of Simulium spp. at certain times affect the daily activities of the local people. In Nigeria, for example, the flies are locally named "ankura", referring to the irritation of the bite (Laurence, 1976). Severe skin reactions including dermatitis may also result from the bites of some species (Crosskey, 1973). In Africa, the depopulation of many fertile river valleys have been attributed partly to the bites of these insects alone (WHO, 1966).

2.3. Simuliids as Vectors of Disease

Simuliids are more important as vectors of disease than they are biting pests. They are responsible for a number of diseases in man and other animals.

The role of simuliids as vectors of avian trypanosomes and Leucocytozoon species have been documented in several parts of the world (Fallis et al., 1951; Bennet and Fallis, 1960; Raybould et al., 1974). Infections with these protozoan

parasites are known to cause severe diseases and mortalities in many birds (Fallis, 1964; Baker, 1969).

Apart from their role as transmitters of protozoan parasites to birds, simuliids are also very well known to transmit various filarial nematodes to birds and mammals. They have been shown to be vectors of Ornithofilaria fallisensis Anderson, a common parasite of both wild and domestic ducks in Canada (Anderson, 1956). Among bovine filariae, Onchocerca gutturosa, which is a common parasite of cattle in many temperate and tropical regions, is transmitted by several species of Simulium (Steward, 1937; Eichler, 1971; Poinar, 1977). In Africa also, although evidence is lacking, this parasite is suspected to be transmitted by simuliids (Raybould et al., 1974). More recently S. damnosum has been shown to be a potential vector of O. ochengi, an intradermal parasite of cattle in Africa (Omar et al., 1979). It is now known that some 9 animal Onchocerca are known to be transmitted by simuliids (Muller, 1979).

Simuliids are not known to transmit any protozoan parasite or viral infections to man (Crosskey, 1973), but their medical importance lies in their ability to transmit two human filariae. One of these, Mansonella ozzardi which is restricted to tropical America has recently been shown to be transmitted by Simulium amazonicum in Brazil (Shelley and Shelley, 1976; Shelley et al., 1980). This non-pathogenic filaria is known to be transmitted by Culicoides spp. eleswhere in that region (Kettle, 1965).

However, the greatest importance of simuliids lies in their role as vectors of Onchocerca volvulus, the cause of human onchocerciasis in the tropical regions of the world.

2.4. Human Onchocerciasis - its Distribution and the Magnitude of the Problem.

Human onchocerciasis is a serious tropical disease that is recognized as one of the most important public health problems (WHO, 1966, 1976). It is characterized by severe itching, various kinds of skin changes, the occurrence of subcutaneous nodules, and worst of all, a severe and irreversible blindness (WHO, 1976).

Transmission of onchocerciasis occurs locally and foci of the disease are found in the neighbourhood of water courses where blackflies breed. The disease is endemic in Africa, tropical America, Yemen and Saudi Arabia (WHO, 1976; Chumbley, 1980).

In the tropical Americas, foci of the disease are scattered and are small. It occurs in Mexico, Guatemala, Venezuela, Colombia and Brazil (WHO, 1976). Approximately 50,000 people are thought to be infected in Guatemala and Mexico (Crosskey, 1973) where blindness is also common (Duke, 1968a).

In Africa, onchocerciasis has a wide distribution, covering the entire region between 15°N and 15°S (Le Berre et al., 1978).

Of the estimated 20 million people infected with this disease in the world, more than 90% live in tropical Africa alone (WHO/AFR, 1978). The worst endemic areas of onchocerciasis occur in west Africa being more serious in the savanna than in the forest areas. Savanna foci are characterized by high blindness rates and may reach alarming levels of 10-15% of the population (Duke and Anderson, 1972a). In the forest, blindness is seldom over 1 percent (Duke and Anderson, 1972a). This has been shown to be due to the existence in west Africa of forest and savanna strains of O. volvulus adapted to forest and savanna zone strains of S. damnosum (Duke et al., 1966), where the savanna strain of O. volvulus is found to be more pathogenic and invasive (Duke and Anderson, 1972b). It is estimated that some 70,000 people are blind in the savanna areas in west Africa (Davies et al., 1978). Because of the severity of the disease in the savanna areas, many fertile river valleys have been abandoned.

2.5. The Vectors of Human Onchocerciasis

Different species of Simulium are known to transmit the disease in different geographical regions. They will be discussed only briefly here.

In Guatemala and Mexico, three species, S. ochraceum, S. metallicum and S. callidum were incriminated as vectors (Dalmat, 1955), but recent studies (Garms, 1975; Garms and

OchoaA, 1979) have firmly established S. ochraceum to be the most important vector. S. metallicum and S. callidum are more zoophilic and are considered to be of minor importance.

In Venezuela, however, S. metallicum is surprisingly highly anthropophilic and is the main vector of onchocerciasis (Crosskey, 1973; WHO, 1976). S. exiguum is believed to be the vector in Colombia (WHO, 1976). Recent work in Brazil suggest S. sanguineum as the principal vector (Shelley et al., 1979).

In Africa, onchocerciasis is transmitted by species belonging to the Simulium (Edwardsellum) damnosum Theobald complex and the S. (Lewisellum) neavei Roubaud group (WHO, 1976). S. damnosum s. l. is the most widely distributed and hence the most important vector in the whole continent. The S. neavei group are limited in their distribution to eastern Africa and are only important in this region (WHO, 1976; Raybould and White, 1979).

However, the presence of anthropophilic and non-anthropophilic members in S. damnosum, and also of epidemiological differences between savanna and forest forms of the disease, led to cytotaxonomic work (Dunbar, 1966, 1969; Vajime and Dunbar, 1975, 1977), by which to date at least 26 cytological forms are recognized (Townson and Meredith, 1979). Some members from west Africa have been given specific names, and of these S. damnosum s.s., S. sirbanum, S. soubrense, S. squamosum and

S. yahense are the most important vectors (Townson and Meredith, 1979). Two of these, S. damnosum s.s. and S. sirbanum occur and are also vectors in east Africa. Most members, particularly those found in eastern Africa, are not yet given specific names and are referred to by locality of collection. Thus in Ethiopia, two forms, "Jimma"and "Kulfo" are known, where the "Jimma" form is considered the vector (Raybould and White, 1979; Townson and Meredith, 1979).

The distribution of S. damnosum s.l. is far more extensive than that of the disease (McCrae, 1969; Crosskey,1973; WHO, 1976). This is due to the presence of non-anthropophilic members, which are most common in east and southern Africa (McCrae, 1969).

The S. neavei group, comprising the second important African vectors of onchocerciasis, includes all those blackflies whose larvae and pupae attach themselves to freshwater river crabs (Potamidae) and prawns (Atyidae) (Raybould and White, 1979). The group is limited in distribution and is largely found in eastern Africa. Some 10 members are known, of which three are regarded as vectors of the disease. One of these, S. neavei s.s. is an important vector of onchocerciasis in Uganda and was formerly so in Kenya (Crosskey,1973). S. woodi is responsible for transmission in the Amani area of Tanzania (Raybould, 1967). S. ethiopiense is reported to be strongly anthropophilic and is thought to be involved in transmission

in the southern highlands of Ethiopia (White, 1977; Raybould and White, 1979).

Other possible African vectors of onchocerciasis include S. bovis which has been found to harbour infective larvae resembling O. volvulus in Nigeria (Crosskey, 1957). S. vorax, S. adersi and S. nyasalandicum developed infective larvae of O. volvulus after feeding on an infected carrier (Wegesa, 1967, 1970). S. dukei (Lewis, Disney and Crosskey, 1969) was also implicated by Duke (1962) (as S. aureosimile).

Outside Africa, Garms and Kerner (1982) have demonstrated for the first time S. damnosum s. l. to be anthrophophilic and also to be a vector in Yemen. They have been able to isolate infective larvae of O. volvulus from wild-caught flies. Regarding the vector in Saudi Arabia, no information is yet available, although it is most likely to be an anthropophilic member of the S. damnosum complex.

2.6. Factors Affecting Transmission of Onchocerciasis.

The transmission of onchocerciasis is determined by various environmental and biological factors which in many cases exert their influence on the behaviour and vector status of the biting Simulium. Only the major ones will be considered herein.

Like most vector populations, the density of the man-biting Simulium in an area is an essential factor in determining the

intensity of transmission of onchocerciasis. Dalmat (1955) was able to incriminate S. ochraceum as the major vector of onchocerciasis in Guatemala partly from its biting density. He considered S. metallicum and S. callidum as relatively unimportant because of their low biting density. In Brazil, S. sanguincum was also regarded as important in the transmission of onchocerciasis mainly for its higher biting density than S. guianense (Shelley et al., 1979). In Ethiopia, S. damnosum is also considered as the main vector because of its higher biting density than S. ethiopiense, although there is very little data on filarial infection (White, 1977).

The density of biting simuliids may be determined by its degree of anthropophily. Some species (eg. S. vorax, S. adersi) are weakly anthropophilic and have low biting densities and are thus relatively unimportant in transmission.

Biting densities may also be influenced by seasonal changes of the climate. During the dry months, temperatures are very hot, rivers may dry up and breeding areas may be restricted. As a result, adult populations of Simulium may be scarce so that transmission during that period is minimal. In the rainy season the converse is true.

Another factor related to the biting of Simulium, and important in transmission, is the time of biting. Although the great majority of blackflies are diurnal, biting from dawn

to dusk (Crosskey, 1973), different species exhibit diurnal fluctuations in host-seeking activities. The African vectors, S. damnosum s. l. and S. neavei group usually show two peaks of biting activity, one in the morning and another in the afternoon, particularly during the dry season when temperatures are hot (McMahon et al., 1958; Le Berre et al., 1964; Le Berre, 1966; Hauserman, 1969). These two peaks of biting activity may be modified into one peak in the afternoon during the rainy season (Hocking and Hocking, 1962; Hauserman, 1969; WHO, 1976). In the Latin American vectors, no marked biting peaks are observed, although S. ochraceum exhibits a single peak between 0800 - 1000 hrs (Dalmat, 1955; Collins et al., 1981). It has now been shown that these peak periods of biting activity are associated with periods of highest density of O. volvulus microfilariae in an infected individual (Duke et al., 1967; Anderson et al , 1975). This periodicity appears to be an adaptation to the biting cycle of the local vectors, thus enhancing effective transmission of the disease.

Longevity of female Simulium is essential for effective transmission of the disease. In general, the older the fly, the higher the chance of its being a carrier of an infective pathogen. Therefore, parous flies (i.e. females that have previously fed and laid eggs) are older than nulliparous flies (i.e. females that have not taken their first blood meal or laid eggs) and are the only possible carriers of infective

stages. In west Africa, Le Berre (1966) and Duke (1975) have shown that in the savanna parous flies concentrate more along the banks of the rivers than nulliparous flies, which disperse further away. In the forest however, no obvious differential dispersal is observed. It follows from this that people living close to the river banks in the savanna are exposed to more intense transmission of onchocerciasis than those living in the forest. This feature may partly explain the great epidemiological differences in the savanna and forest forms of the disease in west Africa (Duke, 1975; Laurence, 1976).

Furthermore, for effective transmission of the disease, the intake of the microfilariae by the simuliid must be successful. This depends on the fly biting on the appropriate area of the skin where microfilariae are abundantly found. The African vectors, S. damnosum s. l. and S. neavei group are predominantly low biters (Crosskey, 1973) and are well adapted for picking up the microfilariae which are mainly found below the trunk (Nelson, 1970). Likewise, the Central Amerioan vector, S. ochraceum, bites on the upper part of the body where the microfilariae are found (Nelson, 1970). In addition, even if the parasite is taken in it may not develop to the infective stage unless it is in the appropriate local vector (Duke et al., 1966; DeLeon and Duke, 1966). Furthermore, temperature will also affect the development of the parasite even if it is taken in by the appropriate vector. Nelson and Pester (1962) observed

that in S. neavei, the larvae of O. volvulus reached the infective stage in 6-7 days at a temperature of about 21°C. In Tanzania, Wegesa (1966 - cited in Raybould and White, 1979) had reported that in S. woodi the optimum temperature range for O. volvulus development was 23-25°C and that no development occurred at temperatures below 18°C. In S. damnosum the optimum temperature is reported to be about 25°C (WHO, 1976).

2.7. Problems in the Identification of O. volvulus

The correct identification of the filarial larvae found in Simulium vectors is one of the main problems in the study of the epidemiology of human onchocerciasis. This is because not all filariae found in the anthropophilic Simulium spp. are of human origin (Nelson and Pester, 1962; Duke, 1967; Voelker and Garms, 1977). Nelson and Pester (1962) reported for the first time unknown filarial larvae provisionally designated as Types A and C in addition to O. volvulus (= Type B) from S. neavei in Kenya. Later, Duke (1967) described three more unknown filariae from S. damnosum in Cameroon and designated them as Types D, E, and F, continuing the sequence of Nelson and Pester (1962). This was followed by the discovery of five more unknown filariae from S. damnosum in Liberia by Garms and Voelker (1969) who later (Voelker and Garms, 1977) named them as Agamofilaria Type III, A. Type IV, A. Type V, A. Type VI and A. type VII. The term Agamofilariae is a collective group

to mean all the immature forms of Filarioidea in the final and intermediate host which do not permit identification (Voelker and Garms, 1977). These various records of filariae indicate that simuliid vectors can feed on other animals also, although the vertebrate hosts of these unknown filariae still remain unknown (Voelker and Garms, 1977). However, the occurrence of these filariae in man-biting species may complicate interpretation of infection rates and transmission rates unless correctly distinguished from O. volvulus.

In most epidemiological studies of onchocerciasis, filarial infection rate in man-biting species is based on isolation and identification of the infective forms. The developing larvae (i.e. 1st and 2nd stages) cannot be identified for certain (Omar and Kuhlov, 1978) because the life cycle of the unknown filariae and other Onchocerca spp. are not yet worked out. However, the infective larvae of O. volvulus are relatively easy to distinguish from those of the unknown filariae because of their distinct size differences (Voelker and Garms, 1977).

Currently, the commonest method of distinguishing infective O. volvulus larvae from others in Simulium is a morphological one. The main criteria are the total length of the larvae, the relative length of the oesophagus to the intestine, and the caudal morphology including the anal ratio (Nelson and Pester, 1962). Unfortunately, some of these dimensions do not clearly distinguish O. volvulus from other species of Onchocerca (Dr. R.

Garms, 1983, pers. Comm.). There are some 25 animal filariae in the genus Onchocerca, some of which are transmitted by simuliids (Muller, 1979). For example, the infective larvae of O. gutturosa, O. ochengi, O. cervicalis are 427-572 µ, 470-620 µ and 600-700 µ long, respectively (Nelson and Pester, 1962; Nelson, 1970; Omar et al., 1979). Thus these dimensions overlap with that of O. volvulus, which has a length ranging from 440 - 731 µ (Nelson and Pester, 1962; Duke, 1967; Voelker and Garms, 1977).

So, all infective larvae of Onchocerca found in Simulium can only be classified into "Morphologically indistinguishable from O. volvulus" (Nelson and Pester, 1962; Voelker and Garms, 1977). Obviously those that are morphologically indistinguishable from O. volvulus may also consist of species of animal Onchocerca.

From the foregoing, it is obvious that accurate assessment of the transmission rate of O. volvulus and determination of vector status of man-biting Simulium species based on morphological differentiation is seriously handicapped. To tackle this problem, alternative means of identification are being developed. Recently, limited progress has been achieved by employing histochemical methods (Omar and Kuhlow, 1978; Omar et al., 1979; Muller, 1979; Omar and Garms, 1981).

2.8. Onchocerciasis in Ethiopia

Human onchocerciasis has been known to exist in Ethiopia since 1939, when Jacono et al. (cited in Oomen 1969a,b) demonstrated for the first time the presence of Onchocerca volvulus infections in Kaffa region, south-west Ethiopia. Since then the disease in Ethiopia remained largely unrecognized and recieved little attention until about the late 1960's. Around this time extensive epidemiological studies carried out by different workers (Torrey, 1966; Oomen, 1967a,b; Oomen 1969a, b; Iwamoto et al., 1973; TenEyck, 1973; DeSole and Walton, 1976; DeSole and Kloos, 1976; Woodruff et al., 1977) clearly established onchocerciasis as an endemic and widespread disease in South-west and western Ethiopia. It is believed that in much of these areas, the vegetation, perennial rivers and streams and the climate offer favorable conditions for the presence of Simulium as well as for the transmission of the disease (Schaller, 1972).

Relatively recently, the presence of the disease in the Humera area, north-western part of Ethiopia has been reported (Lester and Tsega, 1974; Mengesha and Jembere, 1975; Mengesha and Tiruneh, 1977) from patients who have been to the Metema - Humera lowlands. The discovery of a new focus of onchocerciasis in the Sudan close to the Ethiopian border along the Atbara river by Abdella and Abubakr (1975) strongly suggested the presence of the disease in this part of Ethiopia.

According to Oomen (1969a,b) and DeSole and Kloos (1976) the endemicity of onchocerciasis in Ethiopia is generally low except in some coffee growing areas. The coffee areas seem to offer favourable ecological conditions for the vector as well as for the dense human populations associated with coffee, thus forming a large reservoir for the parasite (DeSole and Kloos, 1976).

The prevailing clinical picture of the disease in Ethiopia involves various degrees of skin changes (Oomen, 1967a, 1968, 1969a, b; Iwamoto et al., 1973; DeSole and Walton, 1976; Woodruff et al., 1977) and appears to be more common in the highlands (above 1000m) than in the lowlands (below 1000m) (Oomen, 1969a,b). Eye lesions and blindness due to onchocerciasis are absent or uncommon. Whether this is due to a low level of transmission or a mild nature of the parasite in Ethiopia remains to be investigated.

2.8.1. The vectors of onchocerciasis in Ethiopia

Various investigators (VanSomeren, 1944; Grenier and Ovazza, 1956; Ogata et al., 1970b; Tanaka et al., 1973) have recorded at least seven anthropophilic species of Simulium in various parts of the country. Of these, S. damnosum s.l. and S. ethiopiense are considered as the most important vectors of onchocerciasis in Ethiopia. The latter, a member of

the neavei group, was earlier considered as a subspecies of S. woodi (Fain and Oomen, 1968 - cited in White, 1977), but has now been accepted as quite a distinct species (WHO, 1978; Raybould and White, 1979; Townson and Meredith, 1979).

Although Grenier and Ovazza (1956) and Oomen (1969a,b) had long suspected S. damnosum s.l. and S. ethiopiense to be vectors of onchocerciasis in Ethiopia, the first evidence for the vector status S. damnosum s.l. came from the work of Tanaka et al. (1973) who isolated O. volvulus larvae from natural populations of the fly in Gojeb and Didessa rivers. The infection rate (including developing infections) in Gojeb and Didessa ranged from 10.2% to 12.9% and from 20% to 40%, respectively. This was later confirmed by White (1977) when he isolated a few Onchocerca larvae from 2000 of the flies examined in Gilgel Ghibe river.

The role of S. ethiopiense in the transmission of the disease still remains to be clarified. This species bites man in sufficient numbers (Grenier and Ovazza, 1956;Oomen, 1969a,b; TenEyck, 1973; White, 1977) to be of importance, although its biting densities are very low compared with those of S. damnosum s.l. For example, in the Gilgel Ghibe river, the mean daily total of S. damnosum caught by a pair of catchers was 9556, while that of S. ethiopiense was 86 (White, 1977). No natural infections with Onchocerca larvae have yet been encountered in this species. Attempts by

White (1977) did not reveal any from the small number (46) dissected from the Gilgel Ghibe river. However, it has experimentally been shown to be a potential vector, since development of O. volvulus microfilariae to the infective stage was demonstrated in flies fed on infected men (Schmidt, M. L., Pers. Comm. to White, G.B., 1977).

Other anthropophilic species are also known to exist, but their biting behaviour and their importance in transmission in general are not yet clarified. One of these is S. gibense Uemoto, Ogata and Mebrahtu, 1977, whose biting habit is associated with the rainy season. S. dentulosum is believed to be unimportant, since it is unable to bite the human skin (White, 1977). Three other species (i.e., S. adersi and two unidentified species) also show anthropophilic tendencies but have not been observed biting man (Ogata et al.,1970a).

Furthermore, a checklist of Simuliidae in Ethiopia by Mebrahtu et al. (1980) reveals the presence of S. nyasalandicum, S. bovis and S. vorax which are accassional man-biters elsewhere in Africa and have been shown to be potential vectors (Crosskey, 1957; Wegesa, 1967, 1970). However, nothing is known about the biting habits of these species in Ethiopia.

As far as the distribution of Simulium vectors in Ethiopia is concerned, S. ethiopiense is known to be restricted to smaller streams and rivers of the south-western and western

highlands of Kaffa, Illubabor and Wollega regions (Oomen 1969a,b; Ogata et al., 1970b; TenEyck, 1973; White, 1977; Mebrahtu et al., 1980). Its highland distribution in the south-west is consistent with the behaviour of the neavei group which as a whole prefer well-wooded environments (WHO, 1978). On the other hand S. damnosum is known to occur along the major rivers of the south-western, western, north western and eastern part of Ethiopia (Grenier and Ovazza, 1956; Ogata et al., 1970b; TenEyck, 1973; Tanaka et al., 1973; White, 1977; Mebrahtu et al., 1980). The presence of a non-anthropophilic S. damnosum s.l. (Grenier and Ovazza, 1956) was not however verified when Ogata et al. (1970b) conducted an extensive survey throughout most of the country. The third man-biting species, S. gibense is so far reported from the Ghibe river, crossing the road from Addis Abeba to Jimma, and also from the Didessa river, crossing the road from the Nekempt to Ghimbi in Wollega region (Uemoto et al., 1977).

3. MATERIALS AND METHODS

3.1 Study Area

The Ghibe river, also known as the upper Omo, together with other tributaries, makes up the Omo river system. This river system runs for more than 608 km from its source in the western plateau in Wollega administrative region all the way to Lake Rudolf (now Lake Turkana) in Kenya and southern Ethiopia (Wodemariam, 1972).

The study area is in a valley at the boundary between Shoa and Kaffa administrative regions in the south-western part of Ethiopia. The Ghibe river actually forms a natural boundary (Fig 1). The study area is situated at about 180 km south-west of Addis Abeba near the Ghibe bridge on the Addis Abeba - Jimma main road. It lies between latitudes $8^{o}13'$ and $8^{o}14'$ N and longitudes $37^{o}33'$ and $37^{o}35'$ E. Here, the Ghibe flows at altitudes of 1040 to 1080 meters above sea level.

The valley above the bridge where this study was undertaken is broad and wide open and contrasts sharply with the part of the valley below the bridge which is closed and deeply incised (Fig. 1). The valley ascends to some 1600 meters above sea level on the escarpments on either side and the valley is about 30 km wide. In the study area up-river from the bridge, the river forms two large pools about 1.5 km apart and water enters and leaves these pools rather fast, forming gentler rapids. These and other rapids seem to form suitable breeding places for *Simulium*.

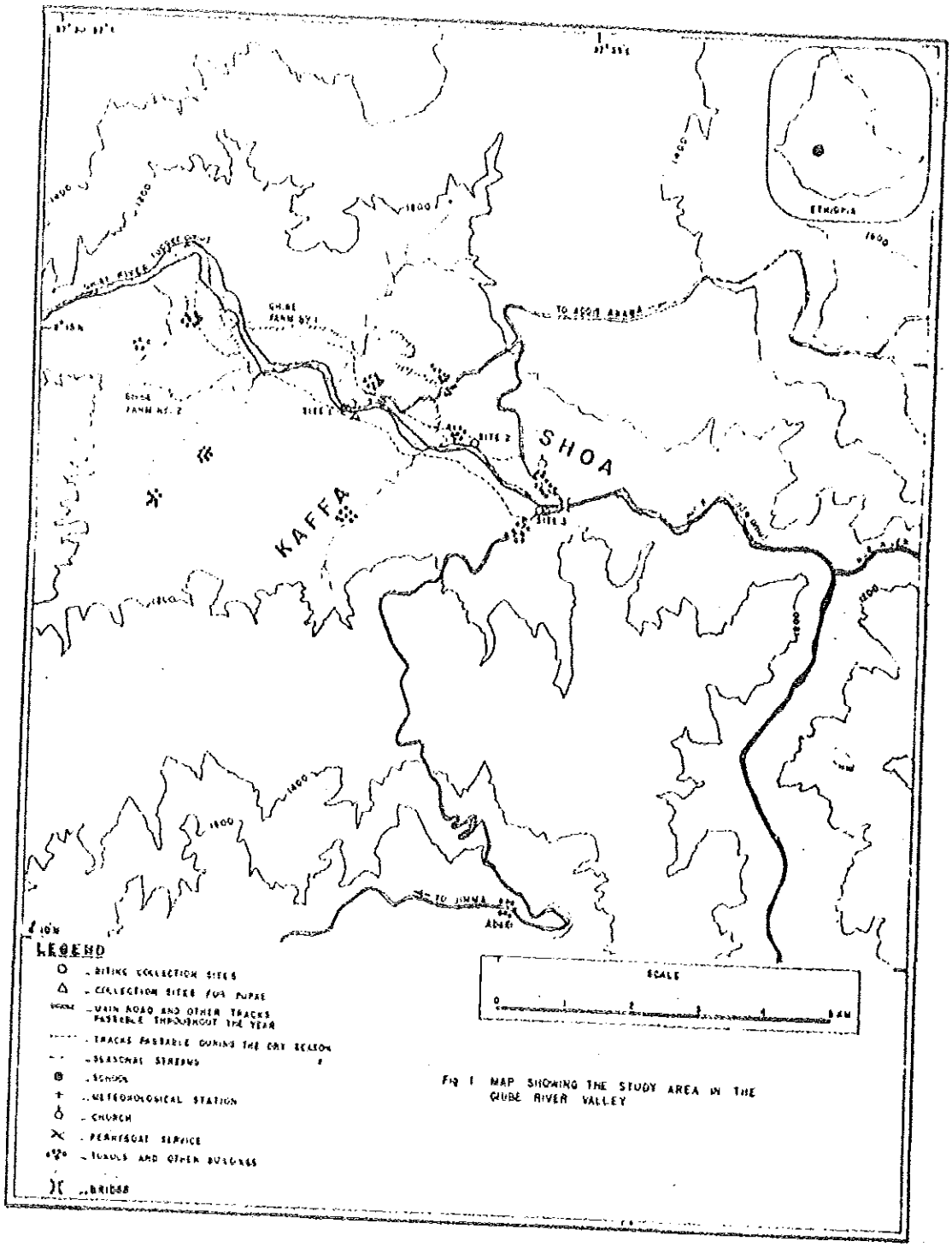


FIG 1 MAP SHOWING THE STUDY AREA IN THE GUBE RIVER VALLEY

According to Gamachu (1977), this area lies within the dry subhumid regions of the country. It also lies in the region where there are seven rainy months, with annual rainfall ranging from 600 to 1000 mm. The small and main rains occur from March to May and June to September, respectively. October to February are usually the dry months.

The natural vegetation of this area is grossly disturbed by increasing human activity. The primary vegetation would have ranged from riverine forest along the edges of the river to woodland and woodland savanna further up the escarpment. At present, the vegetation consists of shrubs and grasses with scattered Acacia seyal, Acacia sieberiana, Acacia tortilis, and Tamarindus indica as the outstanding species.

As regards the wildlife of this area, Hippopotamus (Hippopotamus amphibus), Crocodile (Crocodilus niloticus), Nile monitor (Varanus niloticus), Colobus monkey (Colobus polykomos), Anubis baboon (Papio anubis), Waterbuck (Kobus defassa), Bushpig (Potamochoerus porcus) and Warthog (Phacochoerus aethiopicus) are the most commonly encountered animals. A number of fish-eating birds have also been observed.

Most of the land in this open valley is being cultivated by the Horticultural Development Corporation, Ministry of Agriculture, where two small farms exist on a total of about 690 hectars of land. Some 380 permanent and 200 seasonal labourers, most of

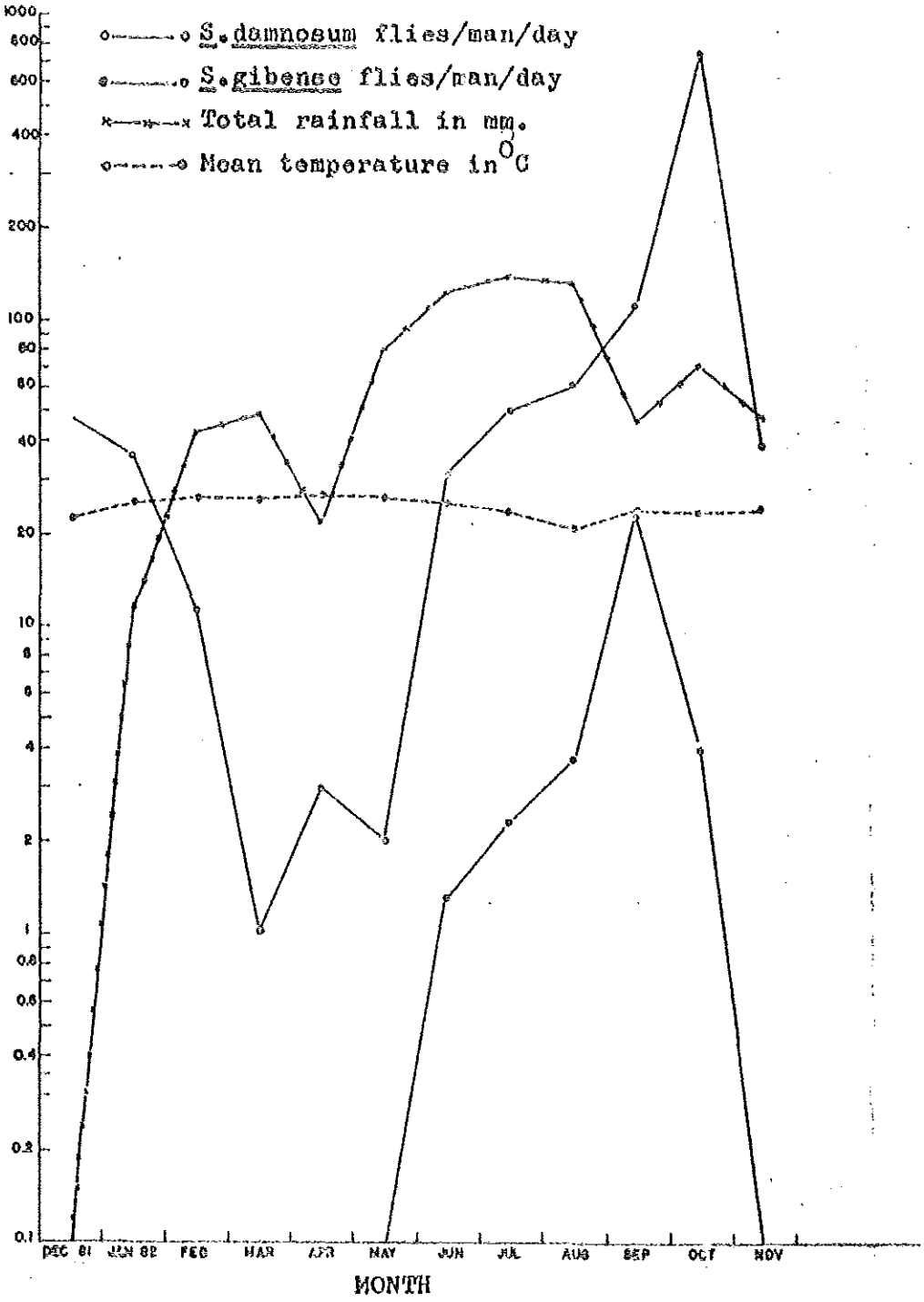


Fig. 3. Seasonal fluctuations in the population density of adult S. dammosum and S. gibense in the Gibe River Valley (Dec. 1981-Nov. 1982). (Prepared on 4-cycle semilogarithmic paper)

and distinct seasonal patterns were apparent (Fig. 3). It may be seen that at least a few S. damnosum were present throughout the year. The lowest biting density of S. damnosum, 1.3 flies/man/day or 0.1 flies/man/hour, was in March and the highest, 735.3 flies/man/day or 56.6 flies/man/hour, was in October. During the dry months (November - April) the biting density of S. damnosum ranged between 1.3 - 48.3 flies/man/day or 0.1 - 3.7 flies/man/hour. In the wet months (May - October) biting densities ranged between 2.0 - 735.3 flies/man/day or 0.2 - 56.6 flies/man/hour (Table 3).

On the other hand, adults of S. gibense were completely absent from collections and were caught biting man only during the wet season, between June and October (Table 3 and Fig. 3). The lowest biting density during this period was 1.3 flies/man/day or 0.1 flies/man/hour in June and the highest was 20.3 flies/man/day or 1.6 flies/man/hour in September.

During a routine biting catch at site 2 on June 25, 1982 (when S. gibense was caught for the first time) flies were seen hovering around cattle that came to the river by this site. Six S. gibense were caught biting the ears of the cattle. It was also observed then that these flies tended to bite man more when in the vicinity of cattle. This feeding habit of S. gibense then led to sampling of the flies on human bait among cattle during the subsequent surveys (July to November, 1982), in addition to the routine collections at the three sites,

Flies coming to bite a pair of human bait among cattle were collected by a pair of catchers for a period of about 2 hours. The number of S. gibense caught biting each month are shown in Table 4. Since two bait-subjects were involved for 2 hours, the number of flies per man per hour was 4 times less the mean number of flies per biting catch. To compare the biting densities of S. damnosum with that of S. gibense, figures obtained for S. damnosum from the three sites (Table 3) during the same period (July - November) are also shown in Table 4.

It must be noted here that the highest number of S. gibense caught, in September (Table 3 and Fig. 3), was mainly due to the presence of a large number of cattle by the river (between 1300 - 1400 hrs) close to site 3 (Fig 1) where a normal biting catch was being conducted.

It may be seen in Table 4 that the biting densities of S. gibense among cattle during July to November ranged from 6.6 to 37.5 flies /man/hour with an average density of 26.7 flies /man/hour. The corresponding densities of S. damnosum during these periods ranged from 3.1 to 56.6 flies /man/hour (average = 15.5). The density of S. gibense was actually much higher than that indicated as numerous flies were seen hovering around man and briefly landing and taking off without biting. A few S. damnosum were also collected during these catching periods but were never seen feeding on cattle.

TABLE 4

Monthly Biting Catches of S.gibense on Human Bait Among Cattle and Their Comparison with S.damnosum caught the 3 sites During the Wet Season of 1982 in the Ghibe River Valley

Month	No. of biting catches*	Total No. of S.gibense caught	Mean no. of S.gibese/biting catch	No. of S.gibense/man/hour	No. of S.damnosum/man/hour
July	3	328	109.3	27.3	4.1
August	5	750	150	37.5	5.0
September	3	361	120.3	30.1	8.7
October	2	53	26.5	6.6	56.6
November	1	0	0	0	3.1
Total	14	1492	106.6	26.7	15.5

*Each biting catch lasted for about 2 hours.

The preferred biting sites of S. gibense on cattle were mainly the umbilicus and the udder region of the cow. Few were also seen feeding on the ears.

On man, the preferred biting sites of S. gibense seemed to differ from person to person. On baits having thin and curly hair, the head seemed to be the preferred site. On baits with thick hair the legs seemed to be the preferred sites. However, flies would also bite on the arms, the ear lobes and around the eyes.

4.4. Age Structure of the Biting Simuliids

4.3.1. Age structure of S. damnosum

During the process of age determination in S. damnosum the general appearance of the fat bodies, the Malpighian tubules and the ovaries were quite sufficient to establish whether the fly was nulliparous or parous. The presence or absence of follicular relios were checked during the first two months of this study, and only in doubtfull specimens during the rest of the study period. This was decided because the observed changes in the Malpighian tubules, fat bodies and ovaries during the first two months were found almost always correlated with the presence of follioular relics.

From a total of 3,418 female S. damnosum collected and dissected during the study period, 2,193 (64.2%) were

parous and 1,225 (35.8%) nulliparous (Table 5). The parous rates ranged between 11.3% in July and 83.3% in May. The parous rates followed a seasonal trend (Fig. 4), in that most flies were nulliparous at the peak of the rains (June to August), while in the other months the parous rates were generally much higher throughout.

4.3.2. Age structure of S. gibense

All S. gibense that were collected among cattle were not age-graded. However, those routinely collected from the 3 catching sites were dissected for age determination. Of the total 95 flies caught and dissected between June and October, 90 (94.7%) were found to be parous (Table 6). Determination of parity in these flies was based on the observation of follicular relics in all cases, since other features were not found reliable. Most had relatively large follicular relics when compared to those of S. damnosum.

4.4. The Diurnal Biting Cycle

To determine the general form of the diurnal biting cycle over the year, the mean total hourly collections of fly populations (i.e., total flies, both nulliparous and parous flies) from the three sites were calculated for each month. The hourly

Table 5

Age Structure and Infection Rate of *S. damnosum* Collected in the Ghibe River Valley (Dec. 1991 — Nov. 1992).

Year	Month	No. of flies caught & dissected	Nulliparous No. (%)	Parous No. (%)	Parous infected No. (%)	Parous with dev. larvae No. (%)	Parous with infective *O. volvulus* larvae No. (%)	Parous with other infective larvae			
								** A. Type VI	** A. Type III	Un-identified	Total with other infective larvae No. (%)
1991	December	145	63(43.4)	82(56.6)	19(23.2)	15(18.3)	3(3.7)	1	-	-	1(1.2)
1992	January	108	26(24.1)	82(75.9)	15(18.3)	7(8.5)	4(4.9)	2	1	1	4(4.8)
"	February	34	13(38.2)	21(61.8)	5(23.8)	2(9.5)	1(4.8)	1	1	-	2(9.5)
"	March	4	2(50.0)	2(50.0)	1(50.0)	1(50.0)	-	-	-	-	-
"	April	9	2(22.2)	7(77.8)	2(28.6)	-	1(14.3)	1	-	-	1(14.3)
"	May	6	1(16.7)	5(83.3)	-	-	-	-	-	-	-
"	June	94	69(73.4)	25(26.6)	5(20.0)	3(12.0)	2(8.0)	-	-	-	-
"	July	159	141(88.7)	18(11.3)	4(22.2)	2(11.1)	1(5.5)	1	-	-	1(5.6)
"	August	193	161(83.4)	32(16.6)	4(12.5)	2(6.3)	2(6.3)	-	-	-	- -
"	September	341	153(44.9)	188(55.1)	9(4.8)	6(3.2)	3(1)*(1.6)	-	-	-	- -
"	October	2206	573(26.0)	1633(74.0)	173(8.3)	63(3.9)	47(7)*(2.9)	9	-	18	27(1.7)
"	November	119	21(17.6)	98(82.4)	12(12.2)	6(6.1)	5(5.1)	-	-	1	1(1.0)
Total		3418	1225(35.8)	2193(64.2)	213(9.7)	107(4.9)	69(8)*(3.1)	15(.7)	2(.1)	20(.9)	37(1.7)

*Figures in brackets = Number of double infections (i.e. also harbour developing larvae)

** Agamofilaria

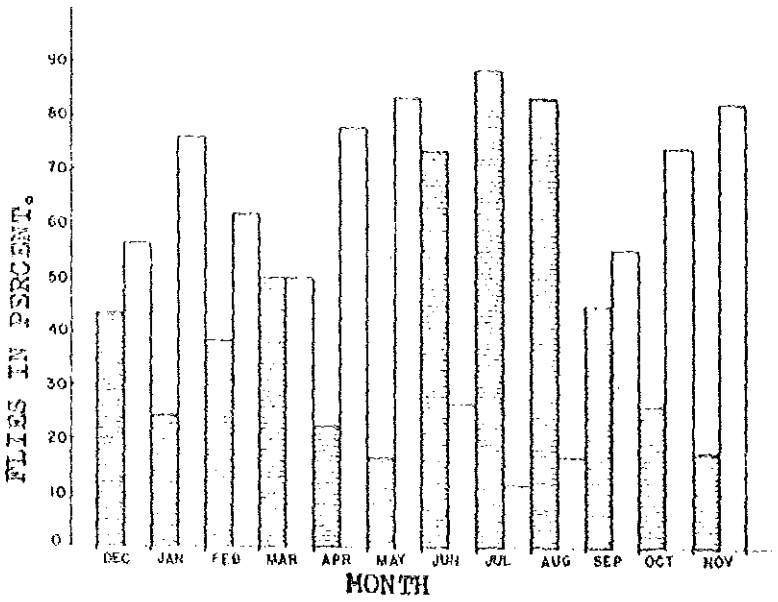


Fig. 4 The seasonal trends of parous (☐) and nulliparous (▤) S. damnosum population in the Ghibe River Valley (Dec. 1981-Nov. 1982)

TABLE 6

Age Structure and Infection Rate of S. gibense Collected in the Ghibe River Valley (Dec. 1981 - Nov. 1982)

Year	Month	No. of flies caught and dissected	No. (%) nulliparous	No. (%) Parous	No. (%) of Parous infected
1981	December	-	-	-	-
1982	January	-	-	-	-
"	February	-	-	-	-
"	March	-	-	-	-
"	April	-	-	-	-
"	May	-	-	-	-
"	June	4	1(25%)	3(75%)	0
"	July	7	0	7(100%)	0
"	August	11	1(9.1%)	10(10.9%)	0
"	September	61	3(4.9%)	58(95.1%)	0
"	October	12	0	12(100%)	0
"	November	-	-	-	-
Total		95	5(5.3%)	90(94.7%)	0

results for the 12 months were then pooled and the mean taken for each fly population. Similar calculation were made of meteorological conditions recorded during catching session. The results for S. damnosum are shown in Table 7 and Fig. 5.

It may be seen in Fig. 5 that S. damnosum flies were found biting from dawn to dusk. The diurnal biting cycle showed two peaks, one in the morning (0800 - 1200 hrs) and the other in the afternoon (1700 - 1900 hrs), the latter being the more pronounced. The favourable air temperatures and relative humidities under which most biting females were caught lie between 21.4 and 27.6°C and 53.3% and 73.8%, respectively. Biting activity was suppressed between midday and much of the afternoon (1200 - 1600 hrs), when temperatures were above 28°C and relative humidities below 47%. The curve for parous flies followed the same basic pattern as the total population while nulliparous flies had a much suppressed peak in the morning but rose steadily in the afternoon.

This basic pattern of the diurnal biting cycle was observed to change at different seasons (Fig. 6a,b). During the dry season (Fig. 6a), both peaks were separated by several hours of very low biting activity. During and immediately after the rainy season (June - September), when temperatures are cold, the morning peak was somewhat reduced but was much smaller than the one in the afternoon (Fig. 6b). Furthermore, biting activity was also observed between the two peaks.

whom are migrants from Wollo and Gojjam adminstrative regions, work on these farms. The seasonal labourers spend the rainy season here and go to Kaffa and surrounding areas to work on Coffee collection during the dry season. Orange, mandarine, lemon, banana, mango are planted as the main horticultural cash crops. However, some spices like ginger, turmeric and pepper are also grown. Maize is grown as the main food crop. On the slopes and close to the bridge there are few farmers growing maize and sorghum as food crops, and cotton and sesame as cash crops. These farmers and the labourers also keep cattle, sheep, goats and poultry. Some fishing is also practiced.

3.2. Selection of Study Sites

Before the actual study began, two reconnaissance trips were made in September and October 1981 to Ghibe, in order to select sampling sites and to recruit and train local assistants to be involved in biting catches. Three catching sites at intervals of about 1.5 km from each other were selected (Fig.1). They were selected on WHO's (1976) recommendation of their proximity to breeding sites, presence of relatively large number of biting simuliids during reconnaissance surveys, and the degree of human activity in the area. All the sites were on the banks of the Ghibe river and were shaded for most of the day.

3.3. Collection and Identification of *Simulium*

3.3.1. Adults

A monthly biting catch for a period of one year (from December 1981 to November 1982) was performed at each site for a whole day from 0600 to 1900 hours. Catches at the three sites were carried out on three consecutive days for most of the time. However, this regularity was sometimes affected by bad weather conditions (eg. heavy rain) or high fly catches on a previous day. The sum of all the flies collected from the three sites was then taken as an index of *Simulium* populations for a given month.

Flies coming to bite the hands and legs of a seated human bait were regularly removed with an aspirator by a second person seated next to him. Both sat on low chairs. The bait often wore shorts and short-sleeved shirt, but when the bait wore trousers and long-sleeved shirts, these were rolled up well above the knees and elbows, respectively. To minimize boredom and avoid bias in sampling (i.e. due to possible differences in host attractiveness), two baits were alternated every two hours. On the other hand, the fly catcher always wore trousers and long-sleeved shirt and was sometimes replaced by me when fly density became very low. At the end of every 15 or 30 minutes, depending on

abundance, flies were transferred to glass tubes plugged with cotton wool to await identification and dissection for age determination. Flies were identified under a binocular dissecting microscope using a key provided by Crosskey (1973) and published descriptions by Uemoto et al. (1977).

3.3.2. Pupae

A supplementary survey was also carried out of Simulium pupae, with the view to determining the species composition and their seasonal changes. Two sites were chosen for pupal collection, close to sites 1 and 3 of the biting collection (Fig. 1). Every month, collection of pupae was made by me for a period of 1 to 2 hours at each site. Pupae were either picked up from various supports (eg. rocks, sticks) using fine forceps or were sometimes removed together with their supports by cutting with scissors. All collections were preserved in 80% alcohol in vials and jars. In the laboratory in Addis Abeba these were sorted out and identified under a binocular dissecting microscope using keys and illustrations of Freeman and DeMeillon (1953), Crosskey (1969) and Uemoto et al. (1977).

For confirmation of identifications, samples of the biting simuliids and pupae were sent to Dr. G.B. White (LSHTM) and the British Museum (Natural History).

3.4. *Simulium* Dissection for Age Determination

To separate nulliparous from parous, flies where dissected in the field on the day of collection. If few flies were collected, dissection was often completed within a very short time. If complete dissection was not possible on the same day, either due to bad weather or high fly catches towards the evening, then the remaining flies were kept overnight in a field refrigerator to be dissected first thing the following morning.

Just before dissection, flies were killed with carbon tetrachloride vapour and transferred into a petri dish with damp cotton wool. Each fly was identified (Section 3.3.1.) before dissection. For dissection, a fly was placed in a drop of normal saline on a slide. Dissection was more or less performed using the method described by Duke (1968b). Using fine entomological needles, the abdomen of the fly was severed near the last two or three posterior tergites and the contents of the abdomen drown out under a binocular dissecting microscope to determine whether the fly was nulliparous or parous.

Determination of female *Simulium* as nulliparous or parous was based on characteristics that were observed on some organs of *Simulium damnosum* by various workers (Lewis, 1953; 1957; Duke, 1968b; Hauserman, 1969). Some of the criteria set by the above authors, and recommended by WHO (1976) were used during the present study. These were:-

a) The presence or absence of follicular relics in the ovaries (i.e., follicular relics are present only in parous flies),

b) The appearance of the ovaries (i.e. they are somewhat transparent in nulliparous flies and yellowish in parous flies),

c) The stretchability of the ovaries (i.e. they can be easily spread and stretched with needles in parous flies while those of nulliparous flies are fragile),

d) The retention of few eggs in some parous flies,

e) The appearance of fat bodies (i.e the fat bodies are large, opaque and sometimes multilobed in nulliparous flies while they are reduced to small translucent relics or become completely absent in parous flies), and

f) The appearance of the Malpighian tubules (i.e, in nulliparous flies, the Malpighian tubules are opaque throughout, except towards the end of the proctedeum, while in parous flies they are either totally transparent or have transparent and opaque sections).

During the process of age determination any nematode encountered, including mermithids, were preserved in glycerol-ethanol (70% alcohol plus 10% glycerol) in small vials for later identification in the laboratory.

The hourly catches which were dissected and divided into nulliparous and parous, were counted and separately preserved in 80% alcohol, in vials, for later staining and dissection for filarial parasites in the laboratory in Addis Abeba.

3.5. Simulium Staining and Dissection for Filarial Parasites

All females, including the nullipars but with the exception of nullipars of October 1982, were dissected in order to make sure that no filarial parasites were overlooked in the few parous flies which might have been mistaken for nulliparous.

In the laboratory in Addis Abeba all flies were stained according to the method recommended by WHO (1966). The alcohol-preserved flies were first washed in descending dilutions of ethanol (i.e., 70%, 50%, 30% and 15%) and finally in water for about 40 minutes in each case. Afterwards, the flies were stained for 3 days in Mayer's acid haemalum and then differentiated in water for 3 days. Finally, the flies were transferred to glycerol to await dissection. To search for Onchocerca volvulus or any other filarial parasite, head, thorax and abdomen were separately dissected in separate drops of glycerol on a slide, using fine entomological needles under a binocular dissecting microscope. The filariae isolated from each region were mounted in glycerol on slides and their number was recorded.

3.6. Identification of the Filarial Parasites

Identification of the filarial larvae was made by morphological examination and measurements of the infective (3rd stage) larvae found in proboscis, head, thorax and abdomen. The developing stages (1st and 2nd stages), which are difficult to identify morphologically, were simply noted and their number recorded.

The following morphological features and measurements were made from camera lucida drawings of the infective larvae. These were:- (i) total length, (ii) maximum body width, (iii) nerve ring (its distance from apex), (iv) length of the anterior oesophagus, (v) length of the posterior oesophagus, (vi) length of the intestine, (vii) tail length, and (viii) tail width (at the level of the anus). From these measurements also, the ratio of the whole oesophagus to the length of the intestine (= Oesophageal/intestinal ratio) and the ratio of the tail length to the width (= anal ratio) were calculated. The results were then compared with dimensions of *O. volvulus* documented by various workers (Nelson and Pester, 1962; Duke, 1967; Voelker and Garms, 1977) (Table 1). All infective larvae with dimensions falling in the ranges shown in Table 1 were identified as "morphologically indistinguishable from *O. volvulus*". Other filariae that were morphologically distinguishable from *O. volvulus* were identified by comparing the dimensions with

TABLE 1

Dimensions (in microns) of O. volvulus Infective Larvae Observed by Different Authors

Structures measured	Nelson & Pester (1962)		Duke (1967)		Voelker & Garms (1977)	
	Mean	Range	Mean	Range	Mean	Range
Total length	566	510 - 630	630	440 - 700	634	545 - 731
Max. width	18.7	17 - 20	18	15 - 20	18	17 - 21
Head	9	8.5 - 11	8	8	13	12 - 14
Nerve ring	80	66 - 90	-	-	81	70 - 94
Ant. Oesophagus	108	100 - 130	120	90 - 140	132	112 - 158
Post.Oesophagus	254	170 - 300	270	190 - 320	281	202 - 352
Intestine	163	130 - 250	190	130 - 220	172	122 - 252
Oeso. Int. ratio	2.2	1.4 - 3.1	2.1	2.0 - 2.6	2.4	1.3 - 3.1
Tail length	31.2	25 - 38	35	25 - 40	34	25 - 42
Tail width	13.3	10 - 16	-	-	-	-
Anal ratio	2.3	2.0 - 3.1	2.1	2.0 - 2.4	-	-

those of the unknown filarial types also documented by Nelson and Pester (1962), Duke (1967) and Voelker and Garms (1977). For confirmation of the identification of the larvae, samples of the infective larvae were sent to Dr. Ralph Muller, Director, Commonwealth Institute of Parasitology, U.K.

3.7. Observations on Meterological Conditions

During each catching session every month, air temperature and relative humidity were measured from dry and wet-bulb temperature readings of a whirling psychrometer. Readings were made every half an hour from 0600 to 1300 hours at each site. The mean values were taken as the hourly temperature* and relative humidity. In addition, the total rainfall and the mean temperature of the area for each month were obtained from National Meteorological Services Agency, Addis Abeba.

* The dry-bulb temperature reading was taken as the air temperature of the hour.

4. RESULTS

4.1. The Species Composition of Simuliidae in the Ghibe River Valley

Pupae of some 9 species of Simulium were collected during the regular monthly surveys carried out in the Ghibe river valley from December 1981 to November 1982 (Table 2). These species have been grouped into five subgenera (Crosskey, 1969) below. They are:-

A. Subgenus Edwardsellum Enderlein, 1921

S.(E.) damnosum Theobald, 1903.

B. Subgenus Metomphalus Enderlein, 1935

S.(M.) gibense Uemoto, Ogata & Mebrahtu, 1977

S.(M.) hargreavesi Gibbins, 1934.

S.(M.) vorax Pomeroy,1922.

S.(M.) (?) dawaense Uemoto, Ogata & Mebrahtu, 1977.

C. Subgenus Pomeroyellum Rubzov, 1962

S.(P.) (?) awashense Uemoto, Ogata & Mebrahtu, 1977.

S.(P.) cervicornutum Pomeroy, 1920.

D. Subgenus Meilloniellum Rubzov, 1962

S.(M.) hirusutum Pomeroy, 1922.

E. Subgenus Eusimulium Roubaud, 1906.

S.(E.) (?) nigritarsis Coquillett, 1902.

Of these, S.(E.) damnosum and S. (M.) gibense were the dominant species followed by S. (P.) (?) awashense (Table 2). The others can be regarded as rare species.

TABLE 2

Species of *Simulium* in the Ghibe River Based on Pupal Collection (Dec. 1981-Nov. 1982)

Year	Month	*S.* damnosum	*S.* gibense	*S.*(?) awashense	*S.* hargreavesi	*S.* hirusutum	*S.*(?) cervicornutum	*S.* vorax	*S.*(?) dawaense	*S.*(?) nigritarsis
1981	December	269	47	-	3	4	-	-	-	-
1982	January	434	1	6	-	2	-	-	-	-
"	February	139	1	1	-	2	-	-	-	-
"	March	111	2	-	-	-	-	-	-	-
"	April	80	18	-	-	3	-	-	-	-
"	May*	4	-	-	-	-	-	-	-	-
"	June*	7	-	7	-	-	-	-	-	-
"	July*	-	-	-	-	-	-	-	-	-
"	August	30	67	30	2	-	4	-	4	2
"	September	69	1995	-	-	-	-	1	-	-
"	October	193	1096	-	15	-	-	3	-	-
"	November	1418	88	89	20	2	-	-	-	-
Total		2754	3315	133	40	13	4	4	4	2

? Identification doubtful.

* Surveys were difficult due to flooding.

In general, an increase in the numbers of pupae was observed towards the end of the rainy season, when S. gibense seemed to be the most abundant. Later, and during most of the dry season, S. damnosum became more abundant. Expressed as number of pupae per man per hour, the seasonal fluctuations of the two common species, S. damnosum and S. gibense are shown in Fig. 2. Both of these species seem to breed continuously except during the heavy rains (May - July) when pupal surveys were made difficult due to constant flooding of the river. However, a dramatic increase in pupae was observed just after the heavy rains in September and October (Table 2 and Fig. 2).

4.2. The Man-biting Simulium Species in the Ghibe River Valley and Their Seasonal Fluctuations

During the course of this study, only two-man biting species, S. damnosum and S. gibense were found in the Ghibe river valley. However, the dominant anthropophilic species was S. damnosum. The total number of adults of S. damnosum and S. gibense caught on human bait at the three catching sites (Fig. 1) were 3418 and 95, respectively (Table 3).

Expressed as number of flies per man per day or flies per man per hour, the monthly biting densities are shown in Table 3 and Fig. 3. In spite of the fact that catches from the different sites produced very different fly totals (Appendix) the catches could be regarded as representative for the area

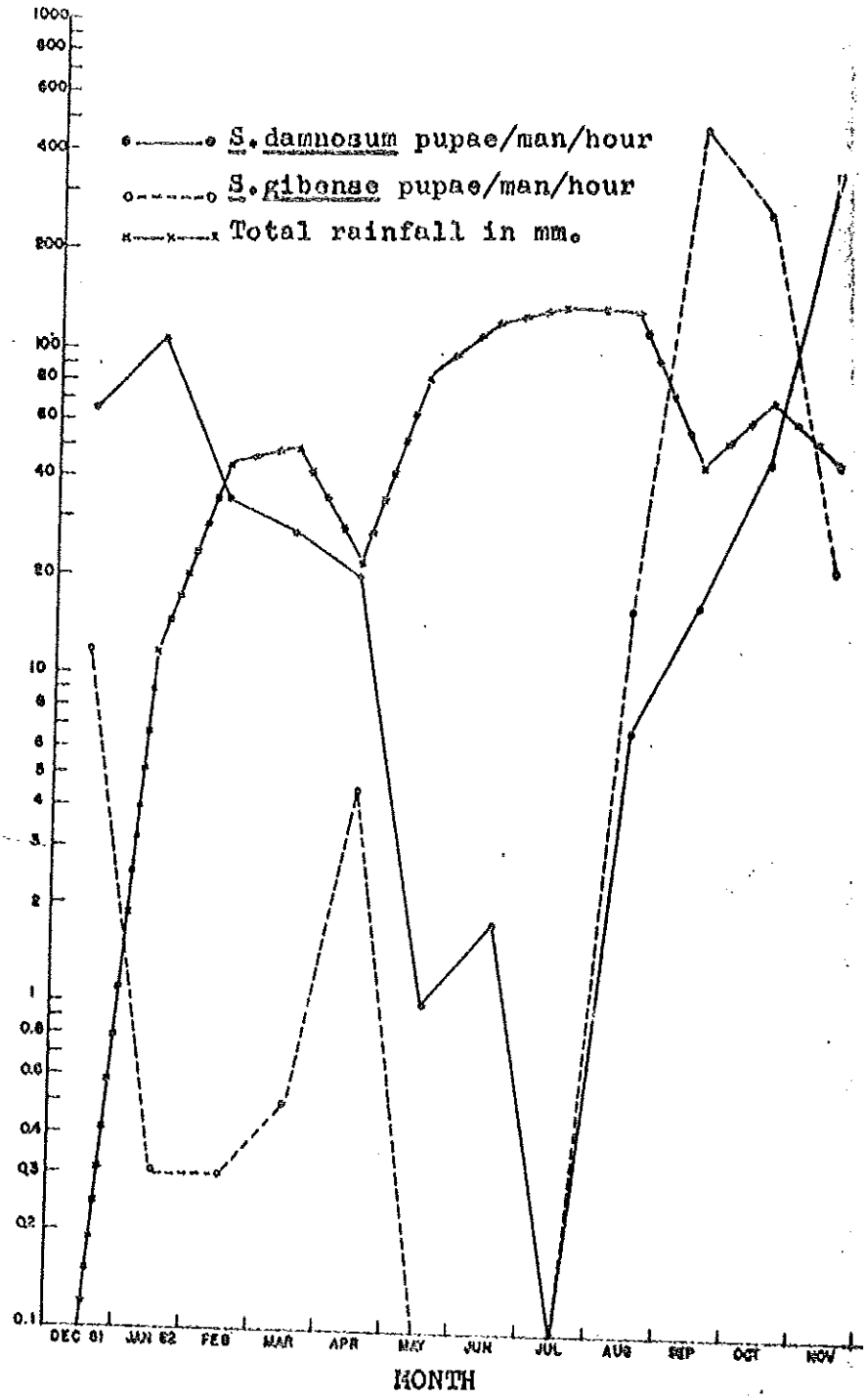


Fig. 2 Seasonal fluctuations in numbers of pupae of S. damnosum and S. gibense in the Ghibe River Valley (Dec. 1981-Nov. 1982).
(Prepared on 4-cycle semilogarithmic paper)

TABLE 3

Biting Densities of S. damnosum and S. gibense in the Ghibe River Valley and the Monthly Meteorological Data (Dec. 1981 - Nov. 1982).

		Meteorological conditions			Biting densities							
Year	Month	Mean Temp. °C	RH (%)	Rainfall in mm	Total No. of S. damnosum caught	Total No. of S. gibense caught	Number of man-days	Number of man-hours	S. damnosum/man/day	S. damnosum/man/hour	S. gibense/man/day	S. gibense/man/hour
1981	December	22.8	43.4	0.0	145	0	3	39	48.3	3.7	0	0
1982	January	25.4	35.3	11.9	108	0	3	39	36.0	2.8	0	0
"	February	26.9	52.5	43.4	34	0	3	39	11.3	0.9	0	0
"	March	26.5	54.2	49.3	4	0	3	39	1.3	0.1	0	0
"	April	27.0	52.0	22.1	9	0	3	39	3.0	0.2	0	0
"	May	26.6	52.5	84.3	6	0	3	39	2.0	0.2	0	0
"	June	25.8	64.9	123.0	94	4	3	39	31.3	2.4	1.3	0.1
"	July	24.0	74.6	139.5	159	7	3	39	53.0	4.1	2.3	0.2
"	August	21.0	82.2	132.5	193	11	3	39	64.3	5.0	3.7	0.3
"	September	24.3	65.8	46.3	341	61	3	39	113.7	8.7	20.3	1.6
"	October	23.5	56.1	70.2	2206	12	3	39	735.3	56.6	4.0	0.3
"	November	24.3	72.8	47.5	119	0	3	39	39.7	3.1	0	0
Total					3418	95						

Table 7

The Hourly Mean Biting Density per Man of A. dorsalis
Population in the White River Valley (Dec. 1981 – Nov. 1982)

Hour of the day	6-7	7-8	8-9	9-10	10-11	11-12	12-13	13-14	14-15	15-16	16-17	17-18	18-19	Total
All flies	3.30	3.38	7.20	10.25	9.94	9.36	4.61	4.11	2.58	3.36	5.11	20.05	15.08	94.91
Nulliparous flies	0.20	1.44	2.55	2.17	2.28	1.61	1.11	1.03	0.67	1.14	1.86	9.03	8.97	34.04
Parous flies	3.10	1.44	4.79	8.08	7.66	7.75	3.50	3.08	1.91	2.22	3.25	11.02	6.11	60.87
Proportion of flies parous	0.33	0.50	0.65	0.79	0.77	0.83	0.76	0.75	0.74	0.66	0.64	0.55	0.41	
Temperature (°C)	17.2	17.6	21.4	23.7	25.9	27.6	29.1	31.1	31.0	29.6	28.2	27.5	25	
Relative humidity	92.2	84.6	73.3	64.5	57.6	53.5	47.1	44.5	42.0	45.2	47.4	53.3	59.3	

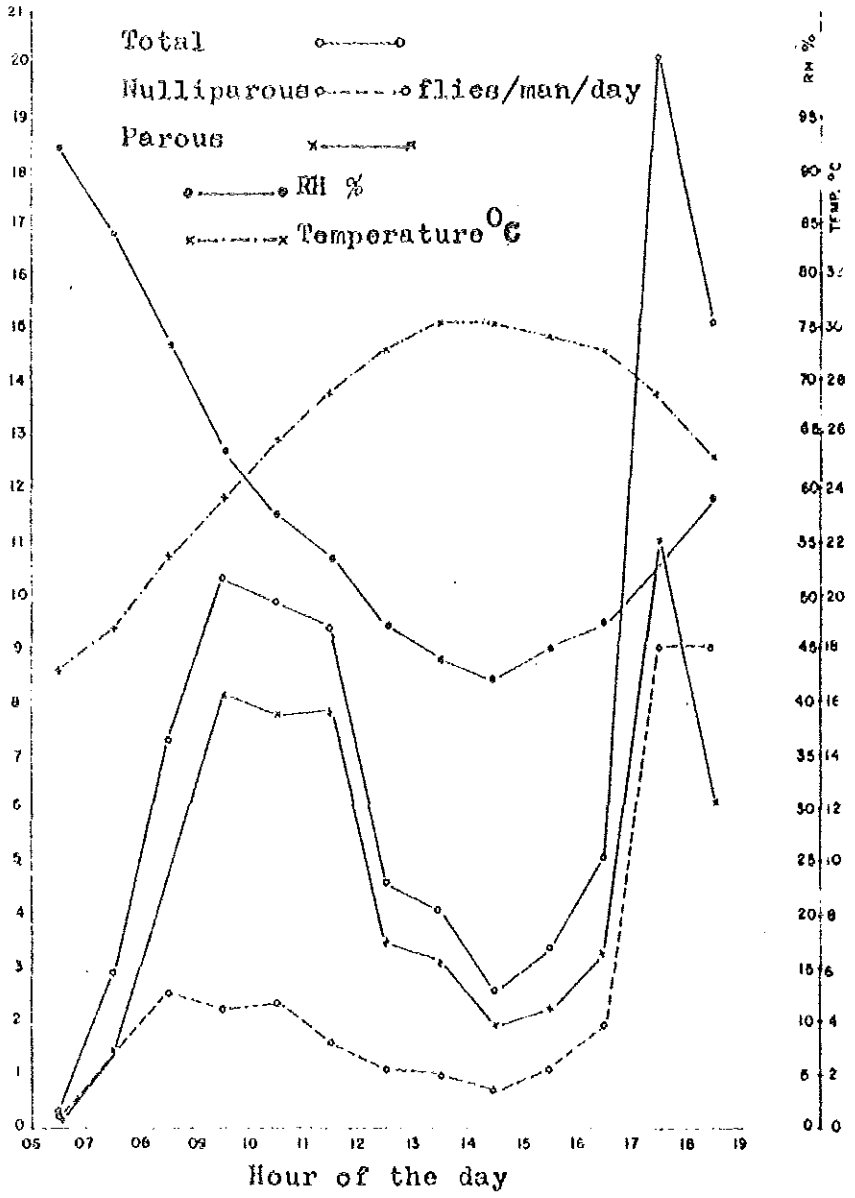


Fig.5 The diurnal biting cycle of different populations of S. damnosum in the Ghibe River Valley (Dec. 1981-Nov. 1982)

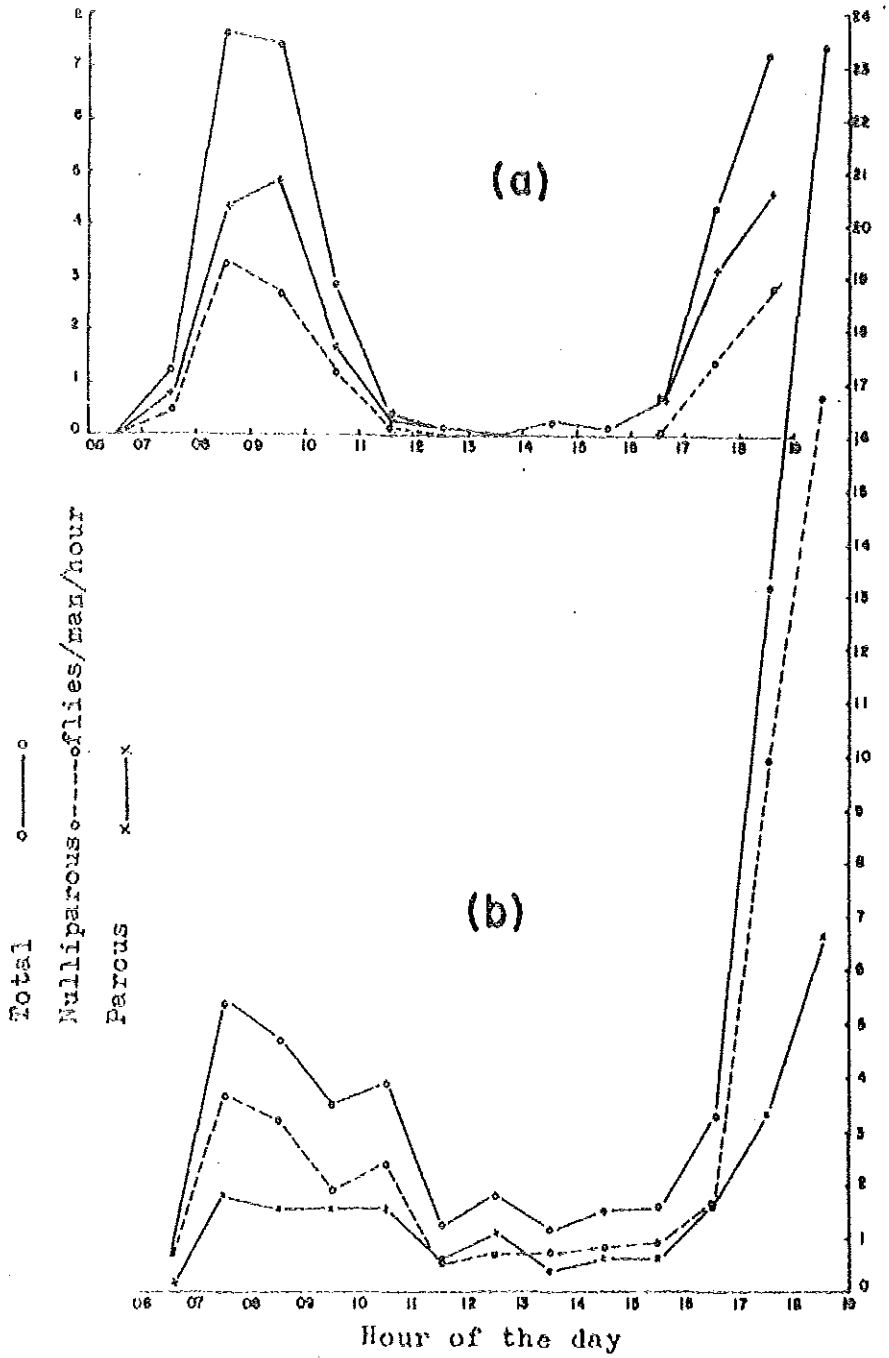


Fig. 6 The diurnal biting cycle of S. damnosum populations during

(a) three dry months (Dec. 1981-1982)

(b) four wet months (Jun. 1982-Sep. 1982)

The biting cycle of S. gibense (Fig. 7) also had a morning and an afternoon peak, but also included a much more pronounced peak in the middle of the day between 1300 and 1400 hours.

4.5. Infection Rates

4.5.1. Filarial infection rates in S. damnosum

Of 2,193 parous flies dissected, 213 (9.7%) were infected both with developing and infective larvae (Table 5). The infection rate per month varied from 4.8% to 50% and showed some seasonal trend (Table 5 and Fig. 8), in that it seemed lower when the biting density was high in September and October (Table 3 and Fig. 3).

Developing larvae (Fig. 9a,b) were found singly in 107 (4.9%) of the parous flies dissected (Table 5). The number of these present in each fly ranged from 1 to 28 (Table 8). All were found in the thorax.

Infective larvae (3rd stage larvae), morphologically indistinguishable from O. volvulus (Fig. 10a and Table 9) were found in 69 (3.1%) of the parous flies dissected (Table 5). 8 of these were also carrying double infections. The number of O. volvulus infective larvae per fly ranged between 1 and 34 (Table 8) with mean 3.5 (Table 14). Most of these larvae were located in the proboscis and head (Table 10) and were thus infective.

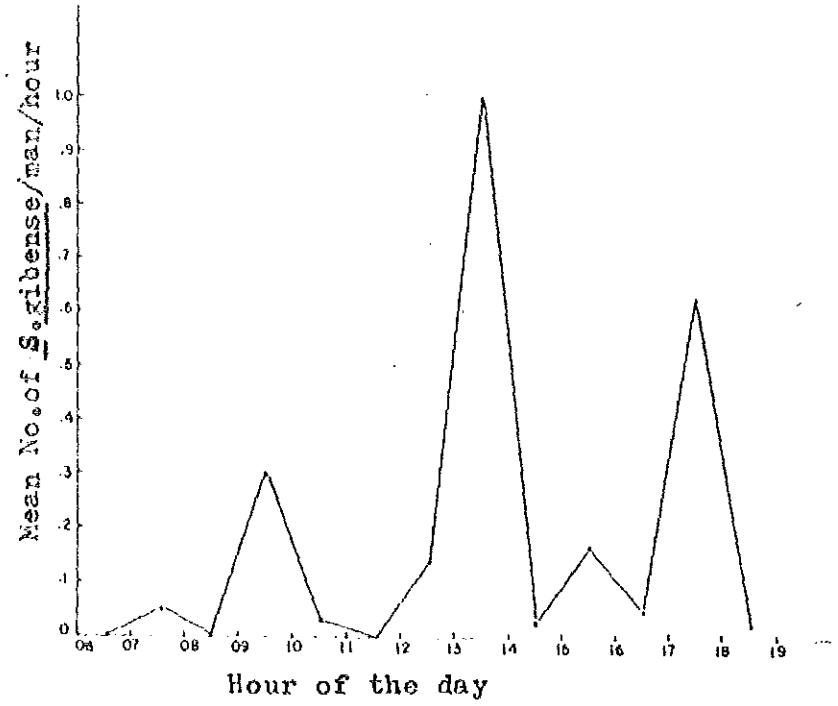


Fig. 7 The diurnal biting cycle of S. gibense in the Ghibe River Valley(Dec.1981-Nov.1982)

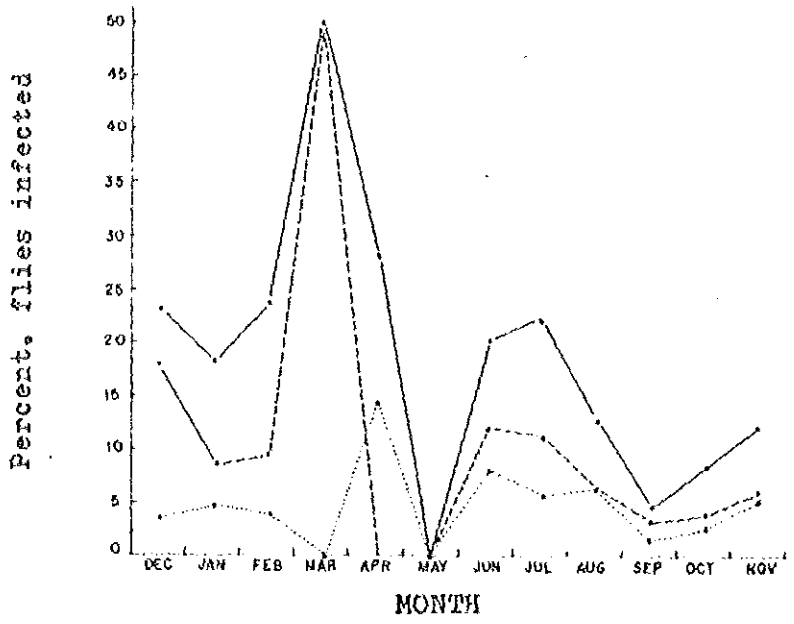
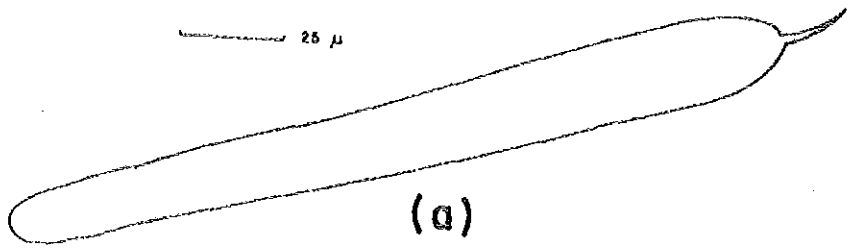
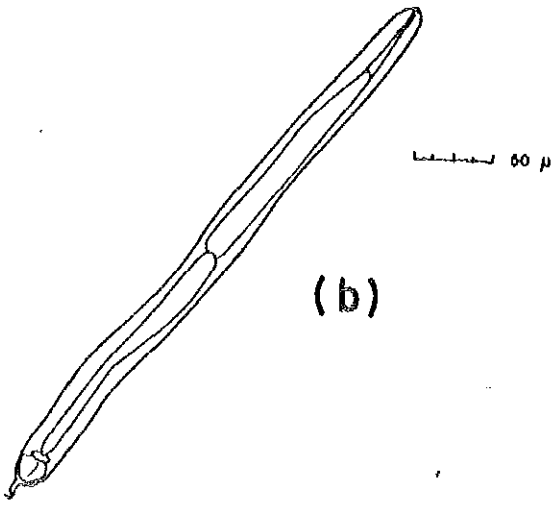


Fig.8 Seasonal trends of parous infected(———), parous with developing larvae(– – – –) and parous with infective O.volvulus larvae (.....) in S. damnosum in the Ghibe River Valley(Dec.1981-Nov.1982)



(a)



(b)

Fig. 9 (a) DEVELOPING LARVA (1ST STAGE) ISOLATED FROM S. DAMNOSUM
(b) DEVELOPING LARVA (2nd STAGE) ISOLATED FROM S. DAMNOSUM

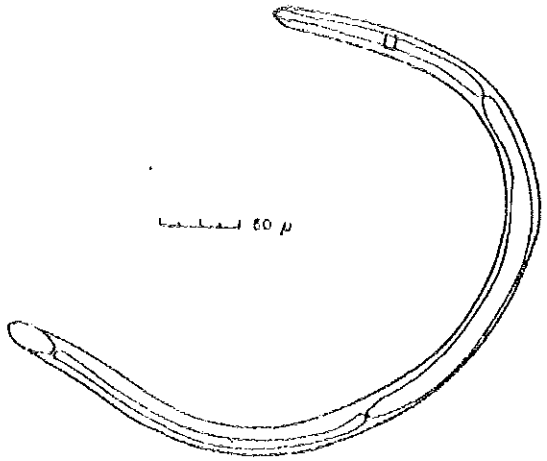
Table 6

Mean Number and Range of Filarial Parasites Found in 227 Infected S. arnoldi Flies

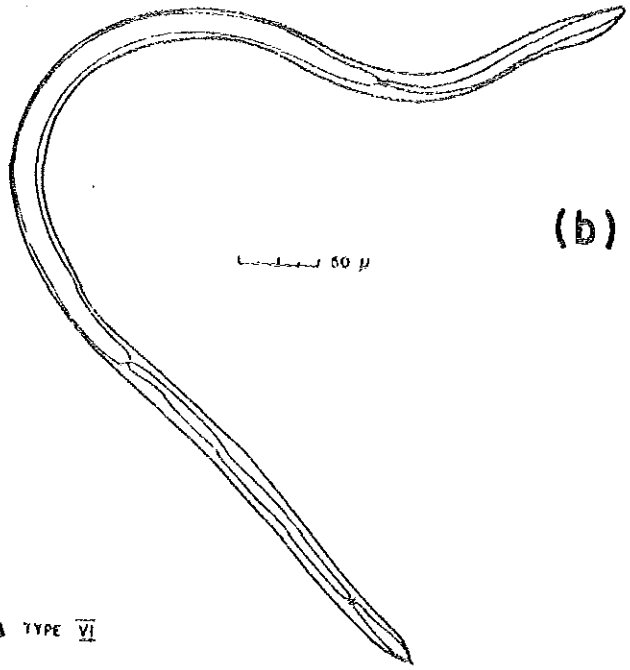
No. of Flies infected	Developing larvae	Infective larvae			
		O. volvulus	Parafilaria Type VI (eType D of Duke 1967)	Parafilaria Type III (eType B of Duke 1967)	Unidentified
107	6.9(1-20)	-	-	-	-
61	-	5.11(1-34)	-	-	-
8	4.5(1-10)	4.5(1-11)	-	-	-
20	-	-	-	-	6.65(1-17)
15	-	-	1.66(1-4)	-	-
2	-	-	-	1(1)	-

Few had larvae in their abdomen. Young third-stage larvae which had not yet left the thorax were also present. The monthly infection rate with infective O. volvulus ranged from 1.6% to 14%. No infection was detected in the months of March and May (Table 5). The observed infection rates were found to be low (Table 5 and Fig. 8) in September and October when biting densities were higher during these months.

Three other types of infective larvae, morphologically distinguishable from O. volvulus were detected in 37 (1.7%) of the parous flies dissected (Table 5). All were distinguished from O. volvulus by their size (Table 9). Two of these, Agamofilaria Type VI (Fig. 10b) and Agamofilaria Type III (Fig. 11) were present in 15 (0.7%) and in 2 (0.1%) of the parous flies dissected, respectively. The average number of A. Type VI per fly was 1.66 (Table 8). Only 1 larva of A. Type III was found in each of two flies infected. Other infective larvae temporarily referred to here as unidentified (Fig. 10c) were found to be more common than the other two. They were isolated from 20 (0.87%) of the parous flies (Table 5). Although the anal ratio is identical to O. volvulus, they were generally smaller in size (Table 9). The number of these larvae per fly ranged from 1 to 17 (mean 6.7) (Table 8).



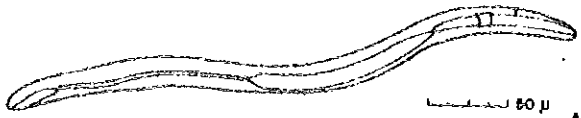
(a)



(b)

FIG. 10 (a) O. VOLVULUS
(b) AGAMOFILARIA TYPE VI
(c) UNIDENTIFIED

ISOLATED FROM S. DAMNOSUM



(c)

Table 9

[illegible] (in microns) of different infective larvae

isolated from S. damnosum in the Ghibe River Valley

Structures measured	50 O. volvulus		25 unidentified		[illegible] Type [illegible] of Duke's [illegible]		[illegible] Type III*
	Mean	Range	Mean	Range	Mean	Range	
Total length	590	446.3-725	401	355.3 - 436.5	[illegible]7.4	765.[illegible] - 9[illegible]	1593.13
Maximum width	18.8	16.3 - 20.3	17	15.6 - 21.8	19	16.3 - 21	35.63
Nerve ring	74	65.6 - 93	65	50 - 74.7	-	-	101.25
Anterior oesophagus	115.2	88 - 150	92	63 - 109.4	158.2	143.8 - 270	222.50
Posterior oesophagus	247.0	161 - 350	152	133 - 180	408.5	360 - 462.5	1061.25
Intestine	100.6	124 - 250	123	83.8 - 154.4	214.8	175 - 200	240.63
Oesophageal: intestinal ratio	1.9	1.5 - 2.5	2.0	1.6 - 3.2	2.7	2.0 - 3.3	5.33
Tail length	33.4	26.8 - 40	31.5	27.5 - 37.5	49.5	46.3 - 56.3	68.75
Tail width	13.7	10 - 16	13.6	12.5 - 15.6	14.0	12.2 - 15.6	23.13
Anal ratio	2.4	2.0 - 2.8	2.3	2.00- 2.20	3.5	3.2 - 3.9	2.97

* The only other specimen of this type (length = 1680 μ. max. width = 46.75 μ), was sent to Dr. Muller for identification before sufficient examination of the larva.

TABLE 19

The Distribution of Different Filarial Infective Larvae in Different Body Regions of S. damnosum

Infective Larvae	No. of parous flies containing infective larvae	Distribution of larvae				Total no. of larvae
		Proboscis	Head	Thorax	Abdomen	
O. volvulus	69	31	214	97	6	348
Acanthocheilonema Type VI	15	0	17	3	5	25
Acanthocheilonema Type III	2	0	1	0	1	2
Unidentified	20	17	104	11	1	133

Identification of the larvae were confirmed by Dr. R. Muller. The identity of the smaller larvae awaits further examination by Dr. Muller.

No infection was detected in the 652 nulliparous flies dissected to check for the accuracy of age determination in the field. However nullipars (573) of October were disoarded in the field because of the high number of flies caught in that month.

4.5.2. Other nematodes in S. damnosum

Among the 3,418 females of S. damnosum dissected for age-determination, only 4 (0.12%) were found infected with mermithids (Fig. 12) in the abdominal cavity. Only 1 nematode per fly was found. The nematodes were found in 1 fly in August, in 2 in September, and in 1 in October. All flies infected with these mermithids were nulliparous. The average length and maximum body width of these were 5347.5 μ (5260 - 5400 μ) and 103.5 μ (103 - 107 μ), respectively. These dimensions were consistent with those given by Poinar (1977) for mermithids.

4.5.3. Filarial infections in S. gibense

The results of dissection of S. gibense collected from man at the three catching sites are shown in Table 6. None were found infected.

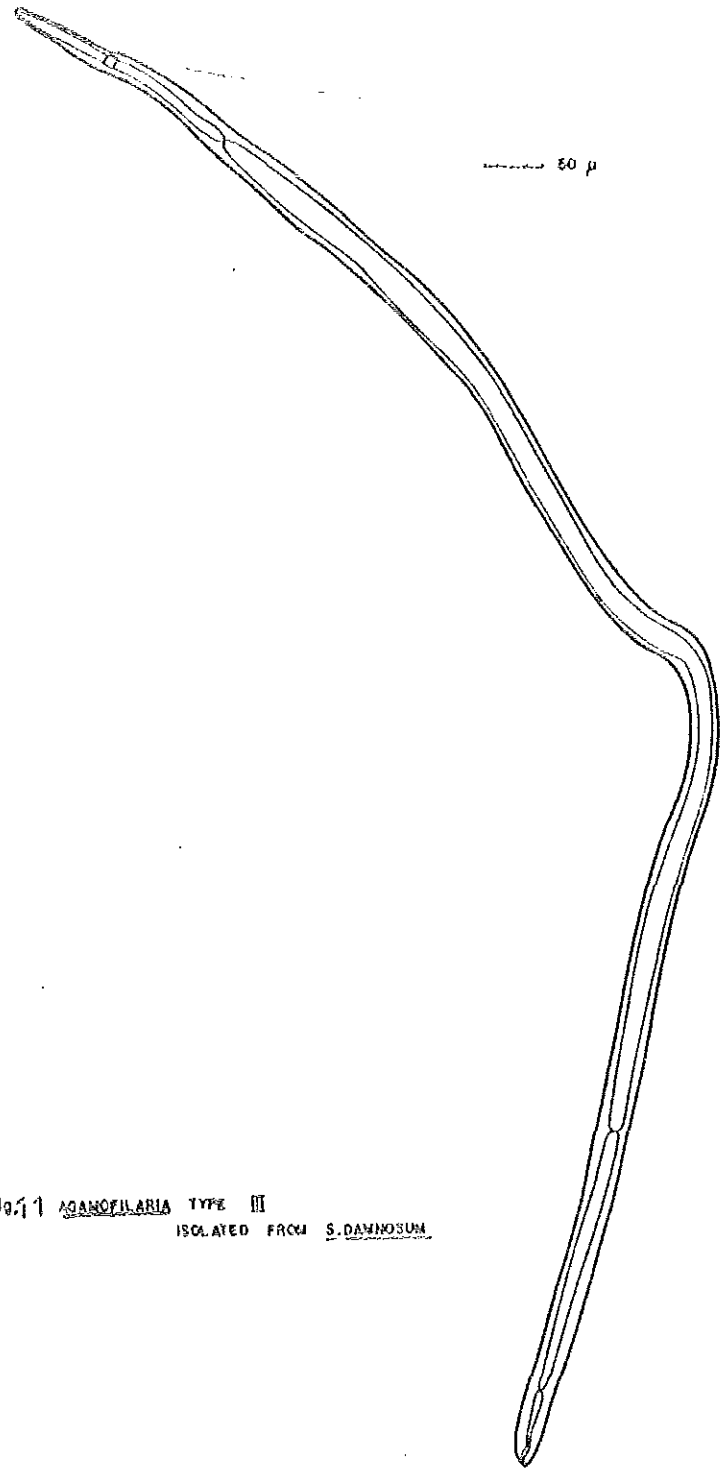


Fig. 1 ADANOFILARIA TYPE III
ISOLATED FROM S. DAMNOSUM

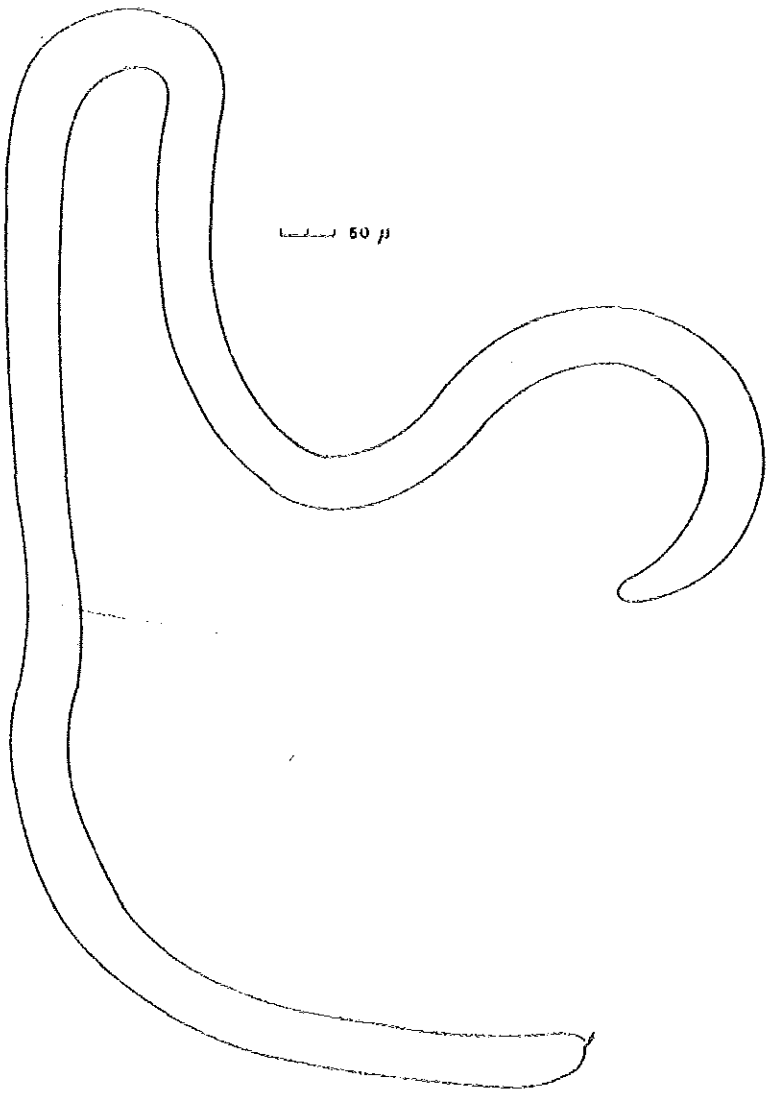


Fig.12 MERMITHID ISOLATED FROM S.DAMIOSUM.

However, of the total 1492 S. gibense that were collected from man among cattle and dissected, only 17(1.4%) were found infected (Table 11). Of these, 13(0.87%) and 4 (0.27%) were found infected with developing stages and infective larvae, respectively. The lowest infection rate, 0.61% was observed in July and the highest, 1.89% in October.

The number of developing forms per fly ranged 1 to 10 (mean 4) and that of infective forms ranged from 1 to 5 (mean 2.25). All the infective larvae were located in the head (Table 12).

Morphological examinations and measurements of most infective larvae (Table 13) isolated from S. gibense show that they are morphologically indistingushable from O. volvulus that were isolated from S. damnosum (Table 10). The larvae had an average length of 616.6 μ, a maximum body width of 17.8 μ and an anal ratio of 2.6. The larvae (isolated from 1 fly) sent to Dr. Muller were identified as morphologically indistinguishable from O. volvulus (Fig.13).

TABLE 11

Simulium gibense Collected on Human Bait Among Cattle and Dissected for Filarial infection

Year	Month	No. of flies dissected	No.(%) of flies infected	No.(%) of flies dinfected with developing larvae	No.(%) of flies infected with infective larvae
1982	July	328	2(0.61%)	2(0.61%)	-
"	August	750	8(1.07%)	7(0.93%)	1(0.13%)
"	September	361	6(1.67%)	4(1.11%)	2(0.55%)
"	October	53	1(1.89%)	-	1(1.89%)
Total		1492	17(1.14%)	13(0.87%)	4(0.27%)

TABLE 12

Mean Number, Range and Distribution of Filarial Parasites Found in 17 Infected *G. gibense* flies Collected on Human Bait Among Cattle.

No. of flies infected	Developing larvae mean(range)	Infective larvae mean(range)	Distribution of Larvae				Total no. of larvae
			Proboscis	Head	Thorax	Abdomen	
13	4(1-10)	-	-	-	52	-	52
4	-	2.25(1-5)	-	9	-	-	9

TABLE 13

Dimension (in microns) of 6 Infective Larvae Isolated from *S.gibense* caught in Three Different Months

Structures measured	August (1) (Haemetoxylin stained)	September (2) (haematoxylin stained)		October (3) (Unstained, glycerol mounted)			Mean dimensions of 6 infective larvae
	1	1	2	1	2	3	
Total width	594.4	571.9	555.0	640.0	658.1	680.0	616.6
Max. width	17.5	18.8	17.5	17.5	17.5	18.1	17.8
Nerve ring	66.3	61.3	67.5	83.8	-	-	-
Ant. Oesophagus	86.3	118.1	112.5	150.0	138.8	135.0	123.4
Post Oesophagus	305.0	292.5	251.4	287.5	287.5	266.9	282.3
Intestine	162.5	122.5	153.8	162.5	192.5	247.5	173.5
Oeso.-intestinal ratio	2.4	3.4	2.4	2.7	2.2	1.6	2.3
Tail length	31.3	38.8	28.1	38.8	35.6	33.8	34.4
Tail width	13.1	12.5	13.8	13.8	12.5	14.4	13.3
Anal ratio	2.4	3.1	2.0	2.8	2.8	2.4	2.6

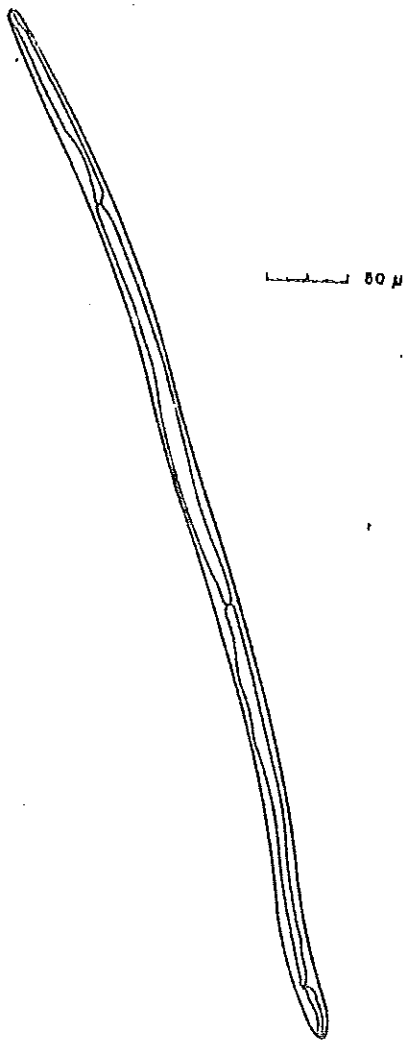


Fig. 13 INFECTIVE LARVA ISOLATED FROM *S. GIBSONI*.

4.5.4. Transmission of O. volvulus by S. damnosum

In order to see the intensity of transmission that was possible in the Ghibe river valley, daily, monthly or annually, the concept of transmission potential (Duke, 1968b) has been used. This is defined as the number of infective O. volvulus which theoretically could be inoculated into one man in unit time. Mathematically, it is the product of the infective biting density on man (flies harbouring infective O. volvulus larvae) in a unit time (daily or monthly) and the mean number of infective larvae per infective fly. Table 14 shows the results of these estimates from the samples obtained. It can be seen (Fig. 14) that the observed transmission intensities followed a seasonal trend. The highest peak of transmission was observed in October when about 92 infective larvae could be transmitted per man per day. The lowest was observed in April when only 0.3 larvae/man/day could be transmitted. In general, transmission potentially could to take place throught most of the year, although on a very low level. The annual transmission potential which is the sum of the monthly transmission, was found to be 3567.3 infective larvae/man/year (Table 14).

TABLE 14

Estimated transmission potential of *O. volvulus* by *S. damnosum* in the Ghibe River Valley (Dec.1981–Nov.1982)

Symbol	Parameter	Derivation	DEC.	JAN.	FEB.	MAR.	APR.	MAY	JUN.	JUL.	AUG.	SEP.	OCT.	NOV.	Total
a	No. of parous flies infected with infective *O. volvulus* larvae		3	4	1	0	1	0	2	1	2	3	47	5	69
b	Total no. of infective *O. volvulus* larvae isolated		13	6	3	0	1	0	9	5	2	20	279	10	348
c	Average no. of *O. volvulus* per infective fly	b/a	4.3	1.5	3	0	1	0	4.5	5	1	6.7	5.9	2	5.5
d	No. of man-days		3	3	3	3	3	3	3	3	3	3	3	3	
e	No. of days in month		31	31	28	31	30	31	30	31	31	30	31	30	
f	Total no. of infective flies/man/day	a/d	1	1.3	0.3	0	0.3	0	0.7	0.3	0.7	1	15.7	1.7	
g	Total no. of infective *O. volvulus* per man per day	cf	4.3	2.0	0.9	0	0.3	0	3.2	1.5	0.7	6.7	92.0	3.4	
h	Total no. of infective flies/man/month	of	31	40.3	8.4	0	9	0	21	9.3	21.7	30	486.7	51	708.4*
i	Total no. of infective *O. volvulus* larvae/man/month	cg	133.3	62	25.2	0	9	0	96	46.5	21.7	201	2870.0	102	3567.3**

* Annual infective biting density

** Annual transmission potential

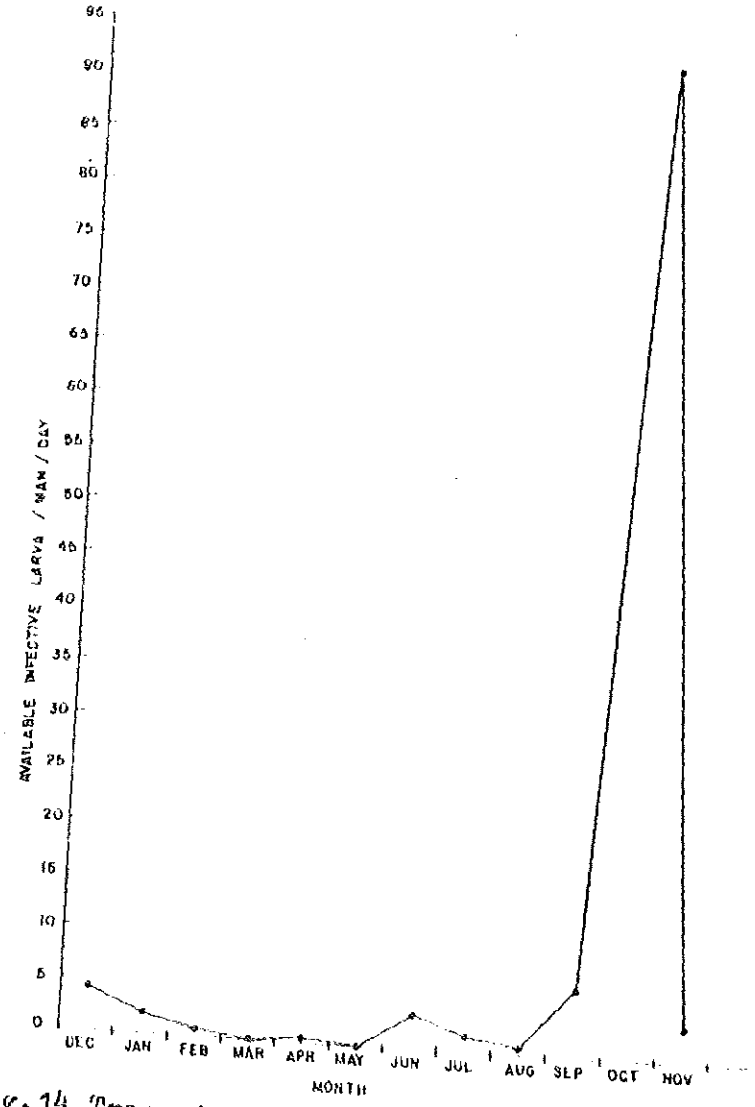


Fig. 14 Transmission potential of O. volvulus in the Ghibe River Valley (Dec. 1981-Nov. 1982)

5. DISCUSSION

5.1. Species Composition, Seasonal Fluctuations, Parous Rates and Biting Cycles.

Four of the 9 species of Simulium recorded in the Ghibe river valley during this study (Table 2), S. vorax, S.(?) dawaense, S. hirusutum, and S. cervicornutum appear to be new records for this stretch of the Ghibe River. Whilst S. cervicornutum and S. hirusutum have been recorded elsewhere on the Ghibe - Omo River system (Mebrahtu et al., 1980), S. vorax and S. (?) dawaense have not been recorded from this river system previously.

The specimen identified as S. (?) awashense in this work could very well contain specimen of S. macmahoni, which is also known to occur in this area (Mebrahtu et al., 1980). Distinction between these two species could not easily be made on pupal characteristics alone since both species can have 12 respiratory filaments, although 8 is the usual number for S. macmahoni (Freeman and De Meillon, 1953; Crosskey, 1969). No pupa with 8 respiratory filaments was collected during this study. The exact identity of most of the pupae is pending confirmation by Dr. R.W. Crosskey, British Museum (Natural History).

Of all the species found, S. damnosum and S. gibense can be regarded as the most common and important species in the valley. Furthermore, they are the only man-biting blackflies, although S. damnosum is the predominant and most important

anthropophilic species. The observation of S. gibense biting man, though occasionally, further confirms a previous report by Uemoto et al. (1977).

In swarms of simuliids, surrounding and biting cattle in this valley, predominantly S. gibense and occasionally S. damnosum were caught on human bait. While S. gibense has been observed feeding mainly on cattle, S. damnosum was never seen doing so. S. bovis, which is also cattle-feeder (Freeman and De Meillon, 1953; Crosskey, 1957) and with which S. gibense may be confused in the adult stage, has not been found in pupal collections during this study. There is no past record of S. bovis occurring in the Ghibe-Omo River system (Mebrahtu et al., 1980). The identity of S. gibense in this study was also sometimes confirmed by breeding adults from pupae which were very easily distinguished from pupae of S. bovis and comparing with wild caught flies using descriptions of S. gibense (Uemoto et al., 1970). Specimens have also been sent to Dr. R.W. Crosskey, British Museum, for confirmation of identfication. S. vorax was represented by only 4 pupal specimens (Table 2) and was not encountered biting cattle or man in this study. According to Raybould (1967), S. vrax feeds mainly on cattle but will also bite man when in close proximity to cows. It has also been reported as a potential vector of O. volvulus in Tanzania (Wegesa, 1967). In Ethiopia, S. vorax is known to occur in a few localities (Mebrahtu et al., 1980), but nothing

is yet known about its biting habits. However, this species seems unimportant at least in the Ghibe river valley.

The seasonal data of this study reflect only broad trends because collections were made at about monthly intervals. More frequent collections are apparently required to observe detailed seasonal patterns in the immature stages, biting densities, age structures and filarial infection rates. It should be noted that in tropical conditions, most species of Simulium, including S. damnosum, have a short life cycle of about two weeks and adult longevity of about three to four weeks (Crosskey, 1973; WHO, 1976).

From Fig. 3 and Table 3,it may be seen that there is some association between seasonal incidence and rainfall. Although adults of S. damnosum were found in every month of the year, a reduced biting density was observed during most of the dry season (November - April) with densities of 0.1 to 3.7 flies /man/hour. Higher biting densities were observed in the wet season (May - October), the highest being noted in October with 56.6 flies /man/hour. In Northern Nigeria, where there are also dry and wet seasons, Crosskey (1955) observed very small numbers of S. damnosum biting, ranging between 0.01 and 0.2 flies/man/hour during the dry months (November - April). Lewis (1953) also noted very few S. damnosum biting during the dry months (November - April) in Southern Sudan. The low biting densities during the dry months may be attributed to the

scarcity of breeding sites, whereas in the wet months, abundant and favorable breeding sites are created with a consequent rise in fly populations. The highest peak observed for S. damnosum in October (Fig. 3) may be due to the fact that towards the end of the rainy season, river levels become more or less stable or slowly decrease thus favoring maximum breeding with consequent rise in adult population. During the months of high rainfall (June - August), frequent flooding of the river may be less favorable for breeding.

The observation of S. gibense biting only from June - October (Table 3 and Fig. 3) indicates that it is a wet-season species. However, since man does not seem to be its natural host, Fig. 3 may show a false impression of its relative abundance during this period. As a cattle-feeder mainly, biting catches of the species in the vicinity of cattle (Table 4) clearly showed that it was much more numerous and abundant than indicated in Table 3. Moreover, when compared with biting densities of S. damnosum during the wet season, S. gibense was more abundant than S. damnosum from July to September (Table 4) when its biting density was 27 - 30 flies/man/hour. The corresponding values for S. damnosum was about 4 - 8 flies/man/hour. For a very closely related species, S. bovis, Crosskey (1957) noted similar observations in Northern Nigeria. Not only was S. bovis a wet season species but also it was found to be much more common than S. damnosum during this period.

The fluctuation in numbers of S. damnosum and S. gibense pupae seem also to be related to changes in the seasons (Fig. 2). Both these species continously bred throughout the year except May - July when the river was in deep flood and surveying was difficult to make (Table 2). During the dry season, S. gibense bred on a very small scale compared to S. damnosum. This in part explains the absence of adult S. gibense from the biting catches during the dry season. However, whether S. gibense occured among cattle in small numbers has not been studied, although this appears to be so. The high number of S. gibense pupae during August - October (Fig. 2) is also manifested by the large number of adults (Table 4) and indicates that it is a wet-season species. The reason why S. damnosum pupae were much fewer than S. gibense during these periods (Fig. 2), particularly in October when its adult population was at its highest peak (Fig. 3), is not clear. During these periods S. gibense pupae were predominantly found on several of the substrates examined. The possibility of S. damnosum breeding in some inaccessible substrates below the surface of the water (Lewis, 1953) might explain this discrepancy in pupae/adult population. In Tanzania, Hauserman (1969) also noted S. damnosum being out-numbered by mass-breeding of S. vorax on habitats with fast flowing water. Members of the subgenus Metomphalus are known to be dominant in fast-flowing and turbulent rivers (Crosskey, 1969). This might also explain the decreased breeding activity of S. gibense during the dry season when water speed

markedly declined. Survival of this species during the dry season therefore seems to be assured by a reduced breeding in a few places with moderate water speed.

The observed high parous rate (64.2%) for S. damnosum (Table 5) seems similar to those of the savanna zones in west Africa (Le Berre et al., 1964; Le Berre, 1966; Lewis, 1965; Duke, 1975). Duke (1975) has shown that, in the savanna, parous flies tend to concentrate along the banks of the river while nulliparous flies disperse further away from the river. It is interesting to note that the vegetation, altitude and the semi-arid or dry-subhumid climate in the Ghibe River valley are savanna or semi-savanna types (Gamachu, 1977). The high proportion of nulliparous flies during the rainy months (June - August) when the population started to increase could be explained by the fact that at this time favorable air humidity and air temperature induced increasing reproductive activity with corresponding increase in nulliparous flies. However, according to Garms (1973) infestations with parasites, especially fungi which are usually very high in the rainy season, might contribute to low parous rates during these periods. In this study also, mermithids were found during the wet months (August - October).

The reason why S. gibense had a high parous rate, 94.7% (Table 6) during the wet-season when it is mass-breeding is not quite clear. Most had large follicular relics probably

resulting from recent oviposition and were probably feeding on the same day of oviposition (Lewis, 1960a). The high parous rate may be due to nullipars dispersing great distance in search of their preferred hosts (cattle). The possibility of a change in host specificity with increasing age has also been postulated by Disney (1972) so that what started as a primarily zoophilic population would tend to be more anthropophilic or vice versa.

The bipeaked diurnal biting cycle of S. damnosum (Fig. 5) was probably due to high temperatures ($>28^{o}C$) and low humidities (<47 RH%) around the middle of the day. The favorable range of temperature for fly activity in the present study was $21.4^{o}C$-$27.6^{o}C$ and is similar to $22^{o}C$-$26^{o}C$ reported by Hauserman (1969) for S. damnosum in Tanzania. It was also observed that the proportion of parous flies was higher in the morning hours than in the afternoon (Table 7 and Fig. 5). Several authors (Lewis, 1956, 1960a, 1960b, 1965; McCrae and Prentice, 1963; Duke, 1968b) have also reported similar differences in the biting cycle of parous and nulliparous flies. Although the reasons are unclear, this is significant in the epidemiology of onchocerciasis in that persons exposed to Simulium bites in the morning hours are at a greater risk of infection than those in the afternoon hours. In addition, if flies caught in the afternoon were dissected for filarial infection, much time would be spent dissecting nulliparous flies which are

free of infection (Lewis, 1957). However, this differential biting cycle between parous and nulliparous flies was not apparent in the curves for dry and wet seasons (Fig. 6a,b). Perhaps data of a few months may not be sufficient to appreciate such differences, for little will be understood without a prolonged study (Lewis, 1960b).

The biting cycle of S. gibense (Fig. 7) also seems to be basically bipeaked, one peak in the morning (0900 - 1000 hrs) and another in the afternoon (1700 - 1800 hrs). But the highest peak occured in the early afternoon (1300 - 1400 hrs) and coincided with cattle coming to the river, when most flies were collected as a result. However, since man does not seem to be the preferred host and differences are likely to occur in the biting cycles of flies caught on man and animals (Disney, 1972), the exact picture of the biting activity of S. gibense must be determined in the future on its normal host (cattle).

5.2.1 Transmission of Onchocerciasis

The presence of anthropophilic Simulium species in essential for transmission of human onchocerciasis to take place. Of the two man-biting species encountered in the Ghibe River Valley, S. damnosum is the predominant anthropophilic species and seems responsible for much of the transmission of human onchocerciasis in the area. Infective larvae morphologically

indistinguishable from O. volvulus were found in 3.1% of the parous flies dissected. Because of the strong anthropophilic behaviour of the flies, the filarial larvae are assumed to be of human origin. It may be seen (Fig. 8) that the infection rate with O. volvulus showed some seasonal variation. Lowest infection rates were observed in September and October, when biting densities were actually high during these months, and higher infection rates were observed during most of the dry season when biting densities were low (Fig. 3). This would probably give a wrong impression of the transmission of the disease to follow the same seasonal trend. However, estimates of the daily or monthly transmission potential (Table 14 and Fig. 14) indicate that this was not the case. It is observed that the highest transmission potential took place when highest biting densities were recorded, in October (Fig. 14 and Fig. 3). If a man was exposed to infection during 13 hours of the day, he would be exposed to about 92 O. volvulus larvae per day in that month. Throughout most of the year low transmission intensities were observed and this coincided with low biting densities. It follows from this that any decrease in the total fly population resulting from control operations would bring about a reduction in transmission.

The annual transmission potential (ATP) which is the number of infective O. volvulus larvae which would be innoculated into one man in one year (WHO,1976) has been found to be about 3567

for the Ghibe River Valley during this study (Table 14). This figure does not indicate the actual amount of transmission to which an individual would be exposed in the Ghibe River Valley. In general, ATP levels do not indicate the actual number of larvae transmitted to any person in the local population, since fly catches are carried out at selected sites (Duke, 1968b; Garms, 1973; WHO, 1976). Moreover, no person is normally fully exposed to Simulium bites all day. Added to that, only some of third stage larvae are actually infective and not all of them leave the vector during a blood meal (Garms, 1973). ATP figures thus only represent the theoretical maximum transmission potential to which an individual would be exposed (Duke, 1968b).

In forest villages in Cameroon, Duke et al. (1972) found ATP levels of 897, 2806, 10241 and 87846, where the prevalence of onchocerciasis was 31%, 64%, 77% and 71%, respectively. They concluded that, under forest conditions, ATP's of 3000 or more are associated with a high prevalence of onchocerciasis. This notion was, however, refuted by Garms (1973) when ATP levels as low as 900 were also associated with a high prevalence of onchocerciasis (64%). In a savanna zone in Cameroon, Duke et al. (1975) associated ATP levels of 500 - 18000 with 100% prevalence of onchocerciasis and concluded that ATP levels above 1500 are associated with high rates of blindness.

Unfortunately, no figure is available for human cases in the Ghibe river valley to make comparisons with those of west Africa.

Although S. damnosum is considered to b strongly anthropophilic, three types of infective larvae, other than O. volvulus have been isolated from this fly during this study. Two of these, Agamofilaria Type VI (Fig. 10b) and A. Type II (Fig. 11) are known from S. damnosum elsewhere in Africa (Duke, 1967; Voelker and Garms, 1977). The other, a smaller larva (Fig. 10c), was also detected in about 20% of the parous flies. These small larvae could possibly be O. volvulus larvae, since staining processess may lead to shrinkage and distortions (WHO, 1966). Whatever the case may be, further investigations are required to clearly establish whether these are in fact O. colvulus or other unknown larvae. The occurrence of such unknown filarial larvae in S. damnosum indicates that it is not strictly anthropophilic. In some areas (eg. Liberia), the incidence of A. Type III was recorded to be very high (3.8%) and exceeded that of O. volvulus (2.2%) (Garms and Voelker, 1969). This suggests that S. damnosum can predominantly be zoophilic.

The detection of mermithid nematodes (Fig. 12) in S. damnosum is also of significance. Mermithids are known to be parasites of many insects and invariably kill their hosts (Gordon et al., 1973). The isolation of these nematodes from nulliparous flies in this study may suggest that flies infected with these

nematodes may not survive long enough to reproduce or serve as vectors of O. volvulus. Hauserman (1969) has also recorded these nematodes from nulliparous flies.

The role of S. gibense in the transmission of human onchocerciasis in the Ghibe river valley is uncertain. This species bites man occasionally when its density is high, and only during the rains. The species is found to be strongly zoophilic, feeding mainly on cattle. However, it bites man in quite sufficient numbers when man is in the vicinity of cattle.

Although infective larvae resembling O. volvulus have been detected, it is by no means certain that they are O. volvulus. In fact they are most likely to be species of animal Onchocerca, since the flies are associated more with cattle than with man. The morphological resemblance of animal Onchocerca to O. volvulus is well known (Nelson and Pester, 1962; Nelson, 1970; Omar, et al., 1979) and is practically impossible to distinguish morphologically between human and animal Onchocerca (Garms, 1983; Pers. Comm.).

No information is available on the exact prevalence and distribution of animal Onchocerca in Ethiopia. However, the presence of O. armillata, O. gutturosa, O. ochengi and O. gibsoni is documented (Bwangamoi, 1969; Graber, 1973). The vectors of these in Ethiopia and elsewhere in Africa are not yet known

although both Simulium and Culicoides species are suspected (Raybould et al., 1974; Muller, 1983, Pers. Comm.).

Although the filariae isolated from S. gibense may or may not have been O. volvulus, the fact that infective larvae were isolated suggests the ability of this species to serve as a vector of human or animal Onchocerca. The exact role of S. gibense can only be ascertained by knowledge of the prevalence and intensity of both human and animal onchocerciasis in the area, for which no information is yet available. Furthermore, the use of histochemical methods (Omar and Kuhlov, 1978; Omar et al., 1979; Muller, 1979; Omar and Garms, 1981) may be required for the identification of the larvae isolated from S. gibense.

Even if S. gibense proves to be a vector of O. volvulus, it is unlikely to be a very important one, for it is mainly a zoophilic species. However, with the absence of its normal host (Cattle) due to mechanization or otherwise in this valley, the possibility of this species resorting to man for blood-meals cannot be ruled out. In such a case S. gibense might become as important as S. damnosum in the transmission of human onchocerciasis in the area. The observations in this study suggest that transmission of human onchocerciasis by S. gibense is at least a possibility. Much more comprehensive studies are therefore required to ascertain this possibility.

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APPENDIX

The Monthly Collections of *S. damnosum* and *S. gibense* at the three Sites in the Ghibe River Valley (Dec. 1981 - Nov. 1982).

Year	Month	*S. damnosum*				*S. gibense*			
		Site 1	Site 2	Site 3	Total	Site 1	Site 2	Site 3	Total
1981	December	110	21	14	145	-	-	-	-
1982	January	94	7	7	108	-	-	-	-
"	February	26	4	4	34	-	-	-	-
"	March	3	1	0	4	-	-	-	-
"	April	6	3	0	9	-	-	-	-
"	May	2	2	2	6	-	-	-	-
"	June	80	9	5	94	2	1	1	4
"	July	87	52	20	159	0	6	1	7
"	August	109	43	41	193	5	5	1	11
"	September	154	120	67	341	9	16	36	61
"	October	1003	420	783	2206	1	1	10	12
"	November	70	48	1	119	-	-	-	-