THE IMPACT OF TEN YEARS OF IVERMECTIN MASS DRUG ADMINISTRATION ON THE LEVEL OF ENDEMICITY AND INTENSITY OF *ONCHOCERCA VOLVULUS* INFECTION IN THE AOWIN DISTRICT OF GHANA

BY

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DECLARATION

I do hereby declare that all the supervised works described in this thesis were done by myself towards the MPhil with the exception of references made to other people's works published or not have all been duly acknowledged. To the best of my knowledge, this thesis has never been submitted anywhere else for the award of similar or different degree neither in whole nor in part.

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DEDICATION

I dedicate this thesis to my lovely cousin, Dr. Emmanuel Morhe for challenging me to aspire higher.

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To God Almighty, thank you for being faithful.

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ABSTRACT

Ivermectin remains the only safe drug for the control and treatment of onchocercisis in endemic areas. Despite some reports on its sub-optimal response in some parts of Ghana, long term treatment has shown that it can eliminate the infection and interrupt transmission in Africa. Taking into account the feasibility of elimination of onchocerciasis infection and transmission interruption with ivermectin mass treatment alone, Joint Action Forum (JAF), and the governing body of African Programme for Onchocerciasis Control in 2011, has reaffirmed its endorsement for the Programme to pursue the elimination of onchocerciasis in Africa. Aowin district of the Western Region of Ghana is meso-endemic for onchocerciasis infection and had received ivermectin mass drug treatment for 10 years. This study was undertaken to assess the impact of 10 years of mass drug administration with ivermectin on the level of endemicity and intensity of *O. volvulus* infection in Aowin district following the claim made by the District Health Directorate that onchocerciasis transmission and infection had been eliminated in the district. A cross sectional survey was conducted in 20 endemic communities situated along rivers in the district. In all, 1,698 volunteers took part in the study. They were examined by palpation for onchocercal nodules or onchodermatitis. Of the 1,698 volunteers examined, 300 who were positive for palpable nodules or onchodermatitis were skin-snipped for microfilarial load assessment. Community microfilarial load (CMFL) for each of the studied community was also determined. Onchocercal nodules and microfilarial prevalence were used to measure the level of endemicity. The intensity of infection was measured using CMFL, a reference index used by the Onchocerciasis Control Programme. The results indicated that, 298 (17.6%) out of the 1,698 examined had palpable nodules. A significant difference was observed ($p=0.0001$) in the nodule prevalence between males and females. Out of the 300 skin snipped, 173 (57.7%) were microfilarial-positive. The microfilarial prevalence and community microfilarial load in the study communities ranged from 8.3-88.9% and 1.0-5.2mf/mg respectively. The overall nodule and microfilarial prevalence recorded suggest strong evidence of on-going transmission of onchocerciasis despite 10 years of annual mass treatment. Halting ivermectin treatment in the district is not recommended as it might risk recrudescence of transmission.

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ABBREVIATIONS

APOC	Africa Programme for Onchocerciasis Control
CDC	Centre for Disease Control
CDD	Community-Directed Distributor
CDTI	Community-Directed Treatment with Ivermectin
CMFL	Community microfilarial load
DALY	Disability Adjusted Life Years
DEC	Diethylcarbamazine
DHD	District Health Directorate
DIA-BA	Dot blot immunobinding assay
ELISA	Enzyme-linked immunosorbent assay
GPELF	Global Programme for Elimination of Lymphatic Filariasis
IVM	Ivermectin
JAF	Joint Action Forum
KCCR	Kumasi Centre for Collaborative Research in Tropical Medicine
L_3	Third Stage Larvae
MDP	Mectizan Donation Program
MDA	Mass Drug Administration
mf/MF/Mf	Microfilariae
mf/mg	Microfilariae per milligram
NTDCP	Neglected Tropical Disease Control Programme
NTD	Neglected Tropical Disease
NGDOs	Non-Governmental Development Organizations

LIST OF

OCP	Onchocerciasis Control Programme in West Africa
OEPA	Onchocerciasis Elimination Programme for the Americas

ONCHOSIM	Onchocerciasis Simulation Model
Pgp	Permeability glycoprotein
PCR	Polymerase Chain Reaction
REMO	Rapid Epidemiological Mapping for Onchocerciasis
SOR	Sub-optimum Response
SIZ	Special Intervention Zone(s)
UN	United Nation
WHO	World Health Organization

CHAPTER ONE

INTRODUCTION

1.1: Background

Human onchocerciasis is a debilitating disease affecting about 37 million people in the world (Basáñez *et al*., 2006). About 500,000 people suffer from visual impairment and 270,000 have gone blind (WHO, 1995). It is also an important cause of severe dermatitis and musculoskeletal pains (WHO, 1995).

Onchocerca volvulus is the causative agent responsible for onchocerciasis (river blindness) and the various dermatological manifestations (Hoerauf *et al*., 2011). The infections are primarily restricted to Africa with about 96% cases mostly in the Western sub-Saharan region (Basáñez *et al*., 2006; WHO, 2011). *O. volvulus* is transmitted by *Simulium damnosum,* particularly *S. damnosum sensu lato* (s.l.) is the major species in Africa (Crosskey, 1990;
Crosskey and Howard, 2004). Humans are the only known natural reservoirs (Udall, 2007). The third stage filarial larvae, L_3, which is the infective larval stage are introduced by the black fly into the human host when it feeds on a blood meal, and the life cycle is completed when adult worms, residing in the nodules produce microfilariae (mf) that move throughout the dermis of the host are taken up by the black fly (Taylor *et al*., 2010). *Simulium* flies reproduce in rapidly flowing streams and rivers and the prevalence of infection and disease in a community depends upon how the community is close to the riverine breeding site of the black flies with communities adjacent to rivers serving as the highest burden of infection and disease (Taylor *et al*., 2010).

Onchocerciasis prevalence is highest among the age population between 20-30 years (Hailu *et al.*, 2002; Little *et al*., 2004a). Men compared to female generally have higher disease prevalence possibly because of their high exposure to the bites of the black flies during farming and fishing activities. Prevalence is lowest in children under 10 years as children have less

exposure to the bite of the *Simulium* flies whose biting activities are greatest in the morning (Hailu *et al.*, 2002; Little *et al*., 2004a).

Adult filarial worms present in the nodules beneath the skin of the infected host can survive up to 14 years (Plaisier *et al*., 1990; Winnen *et al*., 2002). During this period, millions of microfilariae are produced and released into the skin of the infected individuals. The presence of these microfilariae in the skin of the infected individuals is responsible for the main clinical complications due to inflammatory reactions to the macrofilariae in the dermis and in the eyes (WHO, 1995).

The disease is a chronic, multisystemic one that leads to severe itching, blindness, skin lesions, epilepsy as a result of heavy infection, and reduced life expectancy among sighted individuals with high microfilarial (WHO, 1995). Some other consequences of infection with *O. volvulus* includes disability leading to significant morbidity, social ostracism, reduced work and abandonment of fertile river valleys causing reduction in agricultural output among the disease affected populations (Prost, 1986; Vlassoff *et al*., 2000; Boussinesq *et al*., 2002; Pion *et al*., 2002; Little *et al*., 2004b;). Annually, Onchocerciasis causes approximately 1.5 million healthy life-years lost as a result of impairment and mortality (Evans, 1995; Oladepo *et al*., 1997; Remme, 2004; Remme *et al*., 2006). Over half of this result from dermal diseases that have negative impact on income generating capacity, health and socioeconomics of those that are affected and their dependents. (Evans, 1995; Oladepo *et al*., 1997; Remme, 2004; Remme *et al*., 2006).

Onchocerciasis is a serious public health issue because of the detrimental and devastating effects of the disease to both human and economic development (WHO, 1997). Over the past decades, governments, policy and decision makers in the world community have continuously made efforts to control and eliminate this infectious disease. In the year 1975, the World Health

Organization (WHO) launched the Onchocerciasis Control Programme (OCP) of West Africa. The aim of the programme was to eradicate onchocerciasis as a disease of public-health importance from the savana areas of 11 West African countries including Ghana (WHO, 1997). The core activities of the programme included aerial larvicide spraying of rapidly flowing streams and rivers to control the multiplication of the *Simulium* flies and treating infected individuals with Diethylcarbamazine (DEC) that can kill microfilariae (WHO, 1997). However, DEC treatment of the infected individuals led to severe adverse effects and therefore was not considered safe for mass drug administration (Hawking, 1979).

Onchocerciasis control under the umbrella of Onchocerciasis Control Programme in West Africa (OCP) in the 11 West African countries achieved some significant successes. By the end of the programme in 2002, it covered 1,200,000 square kilometres, protected 30 million people that were at risk of the infection and prevented 600,000 people from blindness making 25 million hectares of land safe for relocation (Hopkins, 2005). It has reduced the problem of millions of the world's poorest people (Harlem, 2002; Hopkins, 2005).

The efforts of various governments and policy makers to control and eliminate onchocerciasis as a public health burden currently rely on mass drug administration of ivermectin under the following programme; African Programme for Onchocerciasis Control (APOC), the former Onchocerciasis Control Programme (OCP) and Onchocerciasis Elimination Program for the Americas (OEPA) (Sturchio, 2001). The drug ivermectin or Mectizan® was donated by Merck & Co, in 1987.

Ivermectin was registered in 1987 for human use to treat onchocerciasis and later lymphatic filariasis (Thylefors and Lawrence, 2008; Ottesen *et al*., 2008). To date (2016), it continues to be the only safe drug for mass treatment of onchocercisis (Cupp *et al*., 2011). National control programmes in the former OCP and the WHO African Programme for Onchocerciasis Control

(APOC) provide ivermectin treatment periodically using community-based volunteers (Amazigo *et al*., 2002) to reduce and possibly eliminate the infection where feasible. The recommended dose is 150 μg/kg body weight per annum or three times in a year and has effect on the microfilariae in the skin and the eyes (Enk, 2006; Canga *et al*., 2008). The drug is able to reduce levels of microfilariae in the skin and the eye and maintain it for about 9-12 months. (Greene *et al*., 1985; Lariviere *et al*., 1985; Awadzi *et al*., 1985; Diallo *et al*., 1986; White *et al*., 1987). Reduction of microfilarial density in the skin can significantly reduce parasite transmission but transmission cannot be interrupted after the initial few years of administration of ivermectin (Remme, 2004; Ndyomugyenyi *et al*., 2004). The drug also has advantageous effect on symptoms and clinical presentations of the disease. The drug relieves the intense itching from the infection and progression towards blindness except in very advanced cases. However, ivermectin does not kill adult worms directly, but hinders the release of microfilariae from the adult female worm just after the initial dose (Cupp *et al*., 2004; Duke, 2005). This effect can last up to a period of one year after treatment leading to degeneration of intra-uterine microfilariae and multiple treatments with the drug may increase mortality of adult worms (Cupp *et al*., 2004; Duke, 2005). Multiple doses of the drug have been shown to also have effect on the development of the embryo of the worms and also cause progressive restitution of the cellular anti-filarial immune response (Schulz-Key *et al*., 1992).

Due to the safety profile of the drug, it has been extensively used in mass drug administration since the mid-1990 and it is the WHO recommended approach for the control of onchocerciasis (WHO, 2006). As the drug has limited effect on the adult onchocercal worms, it means that persistent treatment is required in order to repress the manifestations of the infection over time (WHO, 2006). While early computer models estimated 25 years treatment to interrupt transmission, recent evidence has proposed that 5 to 15 years of ivermectin mass treatment

might interrupt transmission depending on the treatment method and the precontrol endemicity level (Winnen *et al*., 2002; Cupp and Cupp, 2007).

Within the past 29 years of Mass Drug Administration (MDA) using ivermectin, a billion treatments have so far been administered and currently, Mectizan Donation Program (MDP) approves an average of 140 million treatments for onchocerciasis and lymphatic filariasis per year (MDP, 2016). Treatment of onchocerciasis using ivermectin mass drug administration has brought significant reduction in the transmission of the disease in the endemic areas (WHO, 2012). By mid-2012, bi-annual mass treatments with ivermectin had eliminated or interrupted transmission of the disease in 10 onchocerciasis foci in the Americas (WHO, 2012).

Many communities in Africa and Latin America have also benefitted from the infrastructure and distribution systems that were developed for the onchocerciasis control and elimination programmes. The successful nature of these ivermectin-based programmes worldwide created a model for health care. Its successful policies and plans have proven the possibility of putting forth activities and efforts to target other diseases that are chronic in poor and remote localities of the world to effect improvements in productivity, morbidity and long-term mortality (Hotez *et al*., 2007).

1.2: Rationale

In order to attain the goal of eliminating onchocerciasis in an endemic area, ivermectin treatment is recommended for several years due to the long life expectancy of the adult onchocercal parasites. The number of years of treatment that are needed in an endemic focus depends on the pre-control endemicity level (the initial worm load and the pre-control intensity of transmission) of the area and the scope of treatment that is reached during the control period (APOC, 2010). According to APOC (2010), the pre-control intensity of transmission is subject to the density of the local vector and intensity of human/vector contact and it is therefore also

an indicator of the local potential of transmission during and when the control period is over. When the pre-control endemicity is higher, it becomes more difficult to bring the parasite population below negligible levels and to interrupt transmission. Computer simulation with Onchocerciasis Simulation Model (ONCHOSIM) showed that a pre-control Community microfilarial load (CMFL) of 10mf/s requires 10 years of treatment to be 95% sure of elimination. But when the pre-control CMFL is as high as 50mf/s, the model predicated 20 years with 80% coverage to have a high probability of elimination. Similarly, the model predicted that with 80% treatment coverage, elimination can be attained several years earlier than with 65% coverage (Plaisier *et al.*, 1990).

Eradication of onchocerciasis in America where the disease is much localized was feasible with ivermectin bi-annual treatment policy (Dadzie *et al*.,2003). WHO confirmed the elimination and interruption of transmission in Colombia (2013), Ecuador (2014) and Mexico (2015) (WHO, 2016). There has also been an immense success in the control of the disease in Africa where 99% of the global cases of the infection is found (WHO, 2016). More than 25 million people in these high risk areas that have received ivermectin treatment now have their infection levels to be low to the extent that the disease is no more considered as a public health burden (Remme, 2004). Despite these enormous achievements, there have been uncertainties among many scientists as to whether onchocerciasis elimination and reduction in transmission levels with ivermectin is possible to a point where treatment could safely be stopped in endemic African countries (Tekle *et al*., 2012).

In Ghana, onchocerciasis infection is known to be endemic in nine regions (Taylor *et al*., 2009). Greater Accra is the only region that is not endemic for the disease. Over 3,200 communities in more than 60 districts in these endemic regions are affected. Brong Ahafo and Ashanti regions have about 247 communities labeled as special intervention zones (SIZ)

because these communities are hyper-endemic to the onchocerciasis infection. The SIZ designated areas are located in the Pru River basin and they serve as foci of Community Directed Treatment (CDT). The estimated population of Ghanaians at risk of the onchocerciasis infection is about 3.4 million people (Taylor *et al*., 2009). Although reports from some foci in Ghana have recorded sub-optimal response of annual IVM treatment defined as 'higher than normal rate of skin mf repopulation by *O. volvulus* adult female worms' (Awadzi *et al*., 2004a; Osei-Atweneboana *et al*., 2007), treatment with ivermectin generally has been effective in Ghana with a minimum of 80% coverage required for elimination (WHO, 2015).

Aowin district started Mass Drug Administration (MDA) about 10 years ago according to the District Health Directorate (DHD). Since Aowin district is a meso-endemic district, 10 years of MDA should be sufficient for transmission and infection of onchocerciasis to be eliminated. Last year (2015), the district did not have mass drug administration of ivermectin because the District Health Directorate claimed onchocerciasis transmission and infection had been eliminated. Therefore there is the need for an independent assessment of the impact of the MDA in the district.

Hence, this study was undertaken to determine if after a decade of mass treatment with ivermectin, there is interruption in the transmission of onchocerciasis in the district. To do this, onchocercal palpable nodules and skin snip microfilarial prevalence was assessed for *O. volvulus* endemicity (Osei-Atweneboana *et al*., 2007). The intensity of *O. volvulus* infection was also measured using community microfilarial load (CMFL) (the reference index used in OCP) to assess the impact of annual ivermectin treatment on onchocercisis infection.

1.3: Aim

The aim of the study was to assess the impact of annual ivermectin mass treatment on onchocerciasis infection after 10 years in the Aowin district of Western Region of Ghana.

1.4: Specific Objectives

1. To determine the level of endemicity of *O. volvulus* infection among the inhabitant of Aowin district using:
    i. onchocercal palpable nodule prevalence and ii. skin snip microfilarial prevalence.
2. To determine the intensity of *O. volvulus* infection in the study area using Community Microfilarial Load (CMFL).

CHAPTER TWO

LITERATURE REVIEW

2.1: Introduction

Onchocerciasis also known as river blindness is a parasitic disease that is caused by one of the nine worldwide filarial nematode parasites *Onchocerca volvulus* (Onchocercidae: Filarioidea). The disease is transmitted from host to host via the bite of blood-feeding female blackflies, *Simulium damnosum* (Mackenzie *et al*., 2012). The blackflies are found in areas where there are rapidly flowing rivers and streams hence the disease is also commonly called river blindness. Onchocerciasis mainly causes infection in the skin and the eye. Humans are the definitive host of the *Onchocerca volvulus*.

2.2: Epidemiology

Worldwide, the disease affects millions of people in which more than 99% of the disease burden is in Africa (Ken *et al*., 2011). Onchocercal belt of the African stretched from Senegal in the west to Ethiopia in the east (Figure 2.2). Apart from the disease being endemic across the entire continent of Africa, onchocerciasis also occurs in a much lesser extent in Central and South America, in Yemen, and in Saudi Arabia (Figure 2.1) (WHO, 1995). Transmission of the disease is now eliminated or interrupted in 11 of the 13 foci in the Americas and is ongoing in one focus in Venezuela and one in Brazil (WHO, 2013).

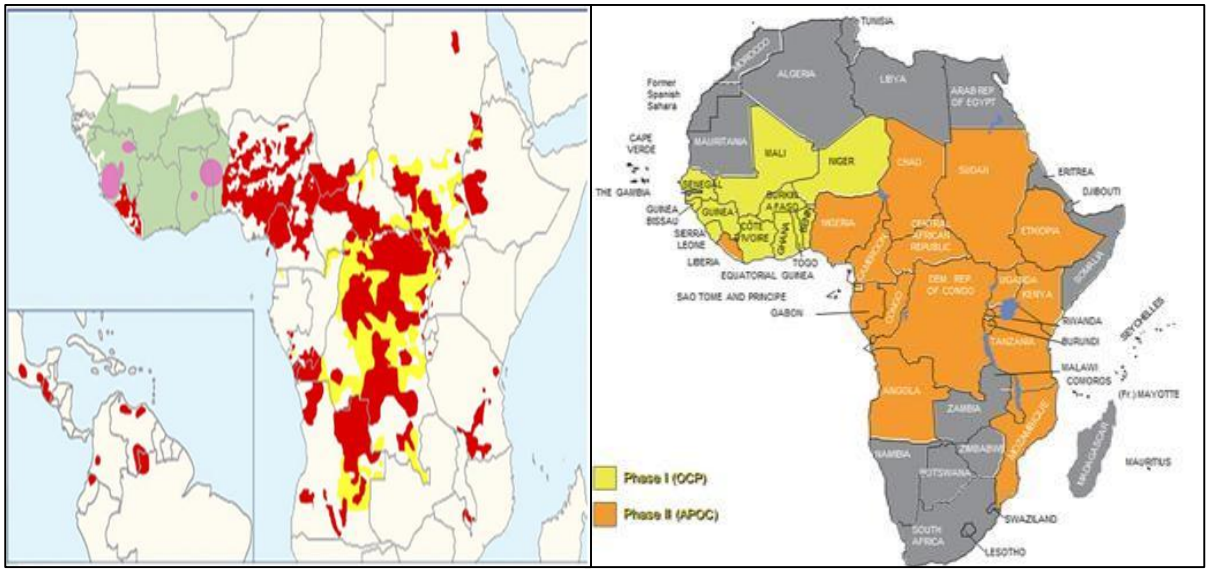

Figure 2.1: (Basáñez *et al*., 2006) **Figure 2.2:** (Fobi *et al.*, 2015)

Figure 2.1: Global epidemiological distribution of onchocerciasis infection *(*Basáñez *et al.*, 2006*) Red: Areas receiving ivermectin treatment: Yellow: Areas requiring further epidemiological surveys: Green: Areas covered by OCP in West Africa: Pink: special intervention zones (i.e., previous OCP areas receiving ivermectin and some vector control)*

Figure 2.2: Endemic onchocerciasis countries in Africa *(*Fobi *et al.,* 2015*) Yellow indicated countries covered by OCP. Peach indicated countries initially covered by APOC.*

The total estimated population in the world that is at risk of contracting the disease is more than 120 million with 34 million people already infected (Gustavsen *et al*., 2011). Of these, 4 million patients have skin manifestations and 2 million are blind or severely visually impaired. Onchocerciasis patients have varied symptoms and different clinical manifestations depending on their geographical locations, parasites strains and their pathogenicity, differences of vectors as well as their biting proclivities, host factors such as those associated with genetic susceptibility or immunity and a history of co-infection by other parasites. Manifestation of onchocerciasis is mostly in two forms and usually termed as forest and savana. High levels of blindness are seen among patients living in the western savana woodland (Woodruff *et al*.,

1977 and Duke, 1981). In the tropical rainforest and East African highlands stretching from Ethiopia to Malawi, cutaneous symptoms turn to be more prevalent (Woodruff *et al*., 1977 and Duke, 1981). Patients in East Africa are less commonly seen with onchocercal depigmentation. Lymphadenopathy mostly manifest in patients living in rainforest area. Severe skin atrophy is usually detected in the savana belt where the microfilarial load tends to be greater.

Onchocercal disease is found in remote, rural, poor communities. The disease affect the working age populations, infested areas with high agricultural potentials are abandoned which causes serious socio-economic problems (Bockarie *et al*., 2009).

In the sub-Saharan Africa where the disease mainly causes blindness and skin infection, they are together responsible for the loss of over one million Disability Adjusted Life Years (DALYS) every year (WHO, 2008). The third leading preventable blindness that occurs in the tropics is due to infections from onchocerciasis (Narita and Taylor, 1993), and skin disease is responsible for 60% of lost DALYs. Research has shown that onchocerciasis may be associated with some diseases such as epilepsy and dwarfism (Basáñez *et al*., 2006). Also, it may probably increase vulnerability to malaria and lowers efficacy of vaccinations (Druilhe *et al*., 2005). Aside the effect onchocerciasis infections have on quality of life; it also plays roles in shortening it. Work done by Little *et al* (2004a), showed an association between microfilarial load of *O. volvulus* and all causes of mortality in the temporal and regional boundaries of the study area claiming 5% of the deaths attributable to this infection.

Progress has been made to control onchocerciasis during the last three decades in sub Saharan African countries endemic to the infection through programmes such as Onchocerciasis Control Programme in West Africa (OCP) which was later replaced by African Programme for Onchocherciasis Control (APOC). Millions of individuals living in infected communities in countries like Ghana, Nigeria, Burkina Faso, Cameroon, and Mali have been relieved from the

infection (Borsboom *et al*., 2003).. Previously abandoned villages have been restored, there have also been considerable decrease of onchocerca induced blindness and other clinical manifestations of the disease (Borsboom *et al*., 2003).

2.3: Life Cycle and Transmission

The life cycle of the causal nematode; *O. volvulus* is illustrated in Figure 2.3. Blackflies are the only vectors that transmit the disease from person to person (Blacklock, 1927; Buttner *et al*., 1982) and humans are the only known definite host (Dozie *et al*., 2005; Krueger, 2006; Trpis, 2006). Adult worms reside in the subcutaneous nodules for up to 15 years. The female worms are larger than the male worms and can produce microfilariae for approximately 9 years (Ranganathan, 2012). Microfilariae are sheathless with a life span of 2 years (Ranganathan, 2012). Development of first-stage larvae (L_1) and finally into the third stage infective larvae (L_3) over a period of about 2 weeks takes place inside the body of the black fly which ingest the microfilariae when it feed on a blood meal from an infected person (Ranganathan, 2012).

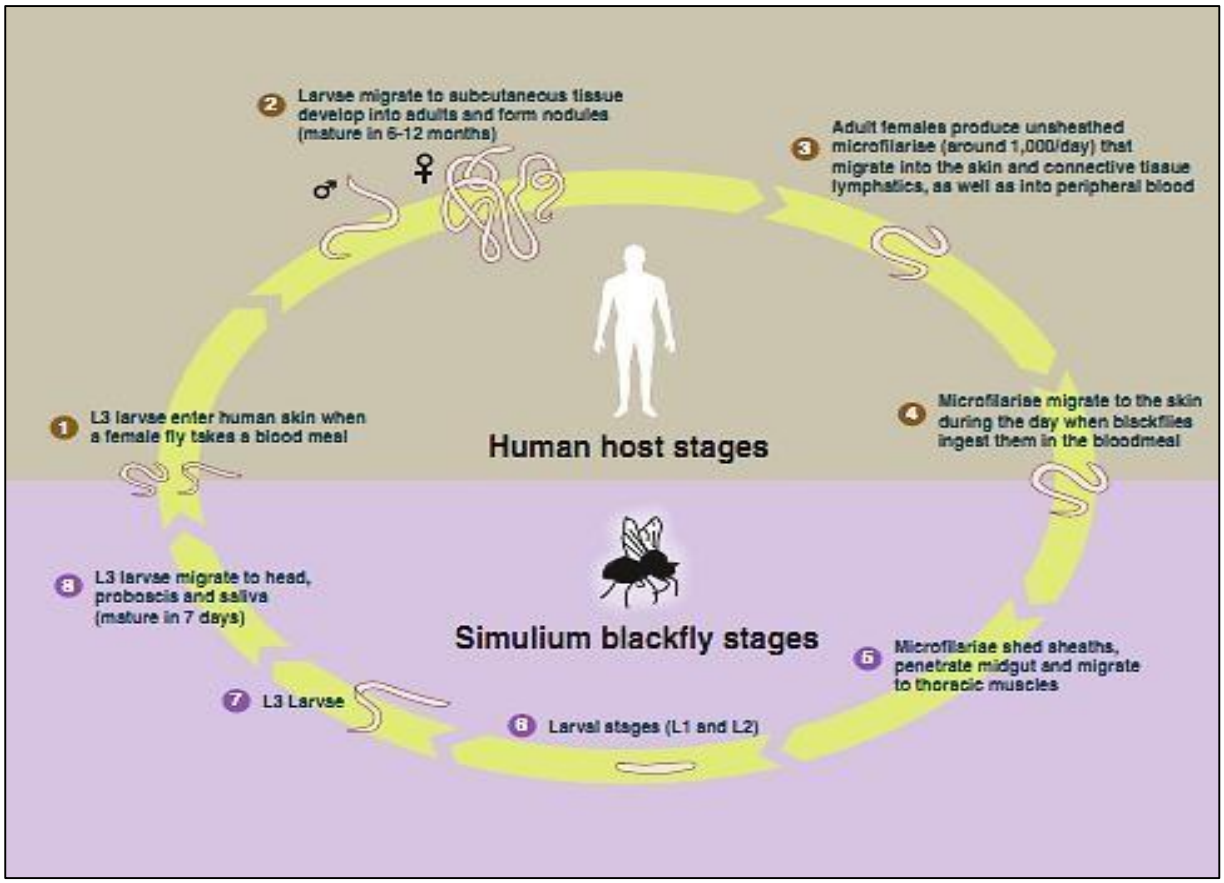

Figure 2.3: Life cycle of *Onchocerca volvulus* Source: (Crump and Mura, 2011)

Infection occurs during a blood meal when an infected *Simulium* blackfly inject the third stage infective larvae (L_3) into the skin of the human host and they enter into the bite wound (Little *et al*., 2004b). The larvae present in the connective tissues molt two times and then develop into adult filariae within 18 months duration and normally inhabit in a fibrous capsule (nodules) in subcutaneous connective tissues. Females measure 33 to 50 cm in length and 270 to 400 μm in diameter and confined permanently in fibrous capsule (nodules) in the subcutaneous connective tissues whiles the males measure 19 to 42 mm by 130 to 210 μm and move freely throughout the skin and subcutaneous spaces (Little *et al*., 2004b). Within 10-15 months after the introduction of infective larvae, the female adult worms start producing microfilariae of about 500–1500 per female per day for approximately 9 years

(Little *et al*., 2004b). Maximum production of microfilariae occurs within the first five years of the female fertile life and later decline linearly (Buttner *et al*., 1982; Maso *et al*., 1987; Duke, 1993; Trpis, 2006). The microfilariae are sheathless and measure 220 to 360 µm by 5 to 9 µm with an expanded head free of nuclei and a sharply pointed tail (Little *et al*., 2004b).

Microfilariae are mainly present in the upper dermis and in nodules and can invade the eyes (Duke, 1993). They can also be found in blood, urine or sputum especially when there is a heavy infection (Maso *et al*., 1987). Microfilarial loads can be as high as 2000/mg of skin, and individuals that are heavily infected can harbour >100 000 000 microfilariae (Simonsen, 2008). It has a life span of 1-2 years (Simonsen, 2008).

The life cycle continues when *Simulium* ingest microfilariae present in the skin during a blood meal. Afterwards, some of the microfilariae move from the midgut of the blackfly into the thoracic muscles via the hemocoel (Simonsen, 2008).. In the fly's thoracic muscles, the microfilariae mature into first-stage larvae and then into third-stage infective larvae over a period of 6–12 days (Trpis, 2006). The infective third-stage larvae migrate to the proboscis of the blackfly where they can be transmitted to humans when the fly takes a blood meal (Buttner *et al*., 1982).

Vertical transmission of microfilariae of *O. volvulus* has been reported but they do not undergo further development (Simonsen, 2009).

2.4: Pathogenesis and Clinical Manifestations of *O. volvulus*

Adult *O. volvulus* worms and the microfilariae contribute to the pathogenesis of onchocerciasis. Adult worms present in the subcutaneous nodules are generally asymptomatic, but the destruction of their progeny (microfilariae) in the dermis of the skin, the eye and lymph nodes are associated with the clinical changes in humans (Murdoch,

1992). Inflammatory and immune reaction of the host's response to dead and dying microfilariae leads to clinical disease (Murdoch, 1992). Endotoxin-like molecules from *Wolbachia* (a bacterial symbiont of *O. volvulus*) also contribute to the pathogenesis of onchocerca disease and in adverse reactions after treatment (Hoerauf *et al*., 2003).

Clinical presentations of the disease are mostly noticed in the skin and the eyes which develop after long exposure to infection (Basáñe *et al*., 2006). Severity is subject to the intensity of infection. In some endemic areas of Africa, symptoms such as epilepsy, growth arrest and general malaise and debilitation can be present (Basáñe *et al*., 2006).

Cutaneous pathology with pruritus is the first commonest symptoms of the disease in infected individuals. Various degrees of dermal and epidermal pathology result from secondary infection following scratching (Vlassoff *et al*., 2000). Onchocerciais also causes ocular pathology which occurs after several years of repeated severe infection. It can affect many part of the eye and sometimes can progress from irreversible blindness from more serious lesions (Basáñe *et al*., 2006).

2.4.1: Onchocercal Skin Diseases

Onchocercal skin diseases are usually the first and the commonest manifestations of onchocerciasis resulting from the inflammatory responses from dead and decayed microfilarial present in the dermis. Dead and decayed microfilariae release circulating antigens that can be deposited into the blood vessels and tissues causing skin pathology (Murdoch, 1992). Aggregation of eosinophils, neutrophils and macrophages surround dying or dead microfilariae (Mackenzie *et al*., 1987). Skin pathology is dependent on the dermal microfilarial load, duration of the infection, the immune responses from the human host and that of inherited factors (Mackenzie *et al*., 1987; Murdoch, 1992; Soboslay *et al*., 1997). Initial or early symptoms are itching and rash. The rash consists of many raised papules due to microabscess

formation, and can either spread or disappear within a few days. The papules may be small and densely populated or large and separated. The rush is often associated with intense itching and excessive scratching can lead to bleeding, ulceration, and secondary infection (a condition known in West Africans as craw craw). In the later stages there may be heavy lichenification and thickening of the skin (lizard skin) (Figure 2.4, Left).

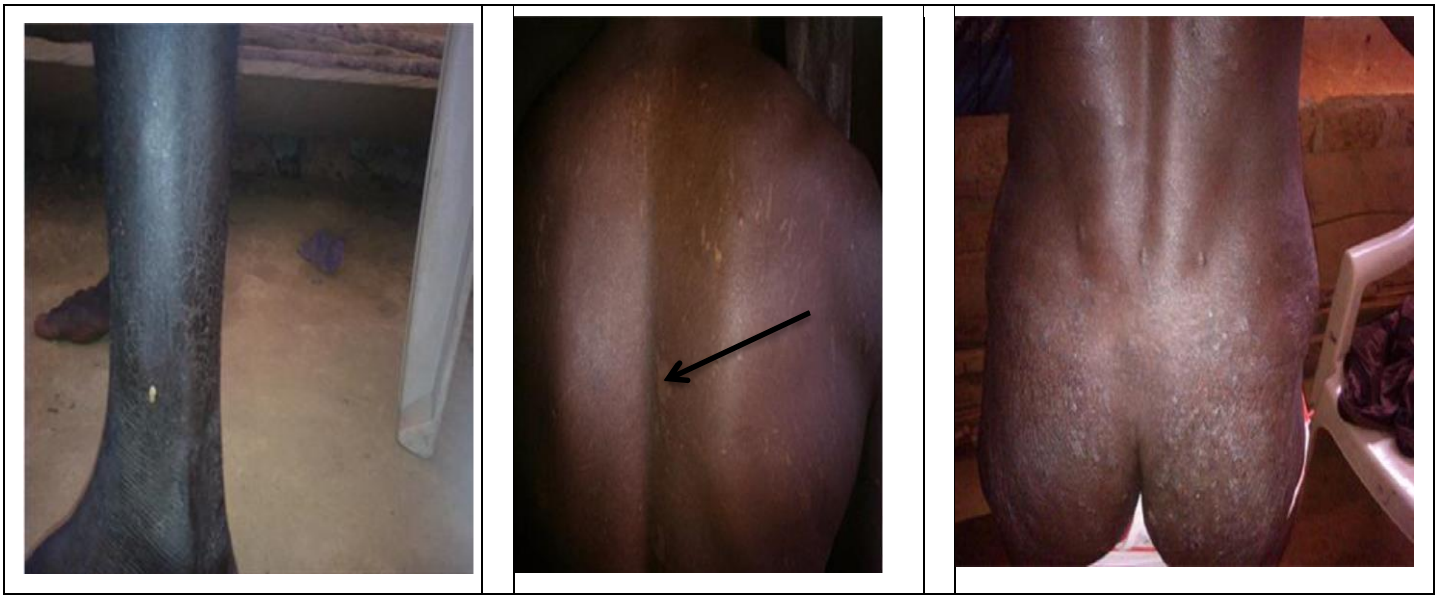

Figure 2.4: Left: Chronic onchodermatitis producing lizard skin in a patient; Middle: Chronic onchodermatitis with leopard spotting; Right: Chronic papular onchodermatitis (Source: Author, 2015**)**

Murdoch *et al* (1993) categorized and graded onchocercal skin infections as acute papular onchodermatitis that presents with small pruritic papules which may develop into pustules or vesicles. Manifestations are mostly seen in the face, the trunk, and the extremities (Enk, 2006; Okoye and Onwuliri, 2007). Chronic papular onchodermatitis affect the shoulders, the buttocks, and the extremities. It manifest as a dispersed, pruritic, hyperpigmented, and uniform topped papulomacular rash with papules diameter of 3mm with or without excoriations (Murdoch *et al*., 1993; Okoye and Onwuliri, 2007). Lichenified onchodermatitis is less common but it is the severe hyper-reactive form of the onchocercal skin diseases characterised

by mosaic patterns commonly referred to as “lizard skin”, “crocodile skin” or sowda (Enk, 2006; Okoye and Onwuliri, 2007). Depigmentation also referred to as “Leopard skin”, and atrophy with loss of elasticity (hanging groin) are the advanced or late stages of the condition (Murdoch *et al*., 1993; Okoye and Onwuliri, 2007). Advanced and late stages can be related to the repeated occurrence of local pathology around dying parasites (Murdoch *et al*., 1993). Skin atrophy with loss of elasticity, gives rise to a premature aged appearance (presbyderma) (Okoye and Onwuliri, 2007). Leopard skin results from loss of pigment, degeneration of the dermal collagen and thinning of the epidermis (Simonsen, 2008). It affects the pretibial regions and more often seen in older infected people (Simonsen, 2008).

Sowda is an Arabic word for black or dark (Fawdry, 1957). It is a localized form of the disease present in a small group of patients who have no or low live microfilariae in the dermis and a small number of nodules that contains adult worms (Fawdry, 1957). Sowda is a result of a strong immune response on the part of the host (Bartlett *et al*., 1978). The affected area of the skin which mostly involves only one lower extremity is severely pruritic, dark and thickened (Ghalib *et al*., 1987). The pruritic nature of the condition normally affects the wellbeing of the infected persons resulting to observable weight loss in these individuals that are found in hyper-endemic areas (Burnham, 1991).

Sowda was reported first in Yamen (Fawdry, 1957) and later in West Africa and Eastern Sudan (Bartlett *et al*., 1978; Ghalib *et al*.,1987).

2.4.2: Subcutaneous Nodules (Onchocercomata)

Nodules are subcutaneous granulomas resulting from the host body reactions around adult worms (WHO, 2001). Whiles some nodules can be found to lie deeper in tissues and nonpalpable, 80% of palpable nodules occurring in Africa are generally seen above bony protuberances of the pelvic girdle and others in the abdomen, chest wall, head or limbs (Figure

2.5) (Dozie *et al*., 2005). In Americas, where the disease is sometimes called "Robles disease" (WHO, 2001), the dominant strains mostly produce palpable nodules located in the scalp and shoulders, and are mostly less in numbers (Wolf *et al*., 2003). Cases of nodules formation in the breast mostly presenting as a breast mass and deeper pelvis nodules have been reported (Zavieh, 2004; Okulicz, 2004). Nodules formations in Yamen onchocerciasis' patients are uncommon (Buttner *et al*., 1982). When nodules are not pressing on vital organs, they do not cause medical problems (WHO, 2010). They are characteristically firm, mobile and measuring 0.5-1.0 cm in width (Krupp and Chatton, 1978). Nodules mostly have 2-3 female adults and 1-2 male adults. But when they form a cluster of 6-8 cm in width, they can contain 10-15 individual worm oodles (Krupp and Chatton, 1978). Nodules are often surrounded by lymphocytes and macrophages (Hoerauf *et al*., 2003), and it has been thought that an angiogenic protein production by the female adult worm contribute to the formation of the nodules (Udall, 2007).

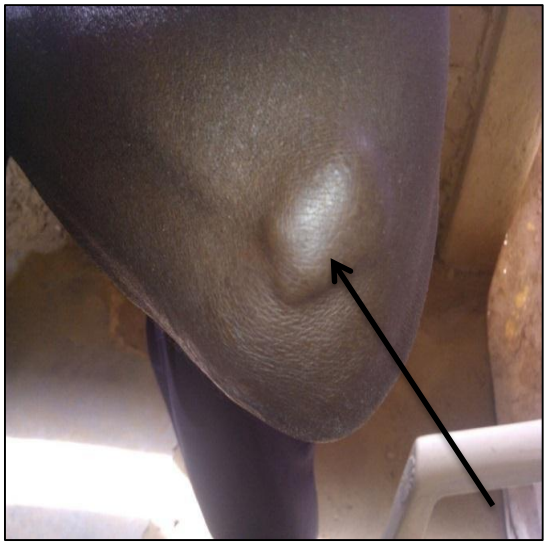

Figure 2.5: Onchocercal nodule indicated by the arrow head (Source: Author, 2015)

2.4.3: Ocular Onchocerciasis

Ocular lesions of the eye can be caused by direct infestation with microfilariae and the toxic reaction that are associated with their death (Basáñez *et al*., 2006). With the exception of the

lens, the lesion can affect the anterior and posterior parts of the eye together (Taylor *et al*., 2010). More serious lesions can lead to irreversible blindness (Basáñez *et al*., 2006; Taylor *et al*., 2010; WHO, 2010). Living microfilariae are distributed and usually coiled in the periphery of the cornea (Basáñez *et al*., 2006). The death of the microfiraliae results in local inflammation such as circumscribed oedema and cellular infiltration around the microfilarial body (WHO, 2010). When they disintegrate and become lysed later, they left a blurred greyish opacity called "snowflake" or "fluffy" opacities which disappears finally. This lesion occurs in the anterior part of the eye and it is called punctate keratitis (Thylefors, 1978). It is a sign of early and light infection which is reversible with treatment (Thylefors, 1978). Long term infections result in anterior segment sclerosing keratitis which occurs when the cornea is permanently damaged by presence of massive microfilariae (Newland *et al*., 1991). Patients with sclerosing keratitis also show signs of iridocyclitis, anterior synechiae, with consequent obstruction of the chamber angle, and glaucoma (Basáñez *et al*., 2006). Lesions such as optic nerve atrophy and choroidoretinitis of the posterior segment may follow and can result in blindness (Newland *et al*., 1991).

Onchocerca blindness prevalence depends on the geographical location (Simonsen, 2009). It occurs more commonly in the savana areas of West Africa with 2-15% of the inhabitants of hyper-endemic area than in West Africa tropical rain forest areas (blindness < 2%). Other factors contribute to the occurrence of ocular pathology. Such factors includes presence of palpable nodules in the upper part of the body (Simonsen, 2009), the intensity of the infection (Little, 2004b), parasite variants (Zimmerman, 1992), vector species (Baker, 1986) and higher *Wolbachia* load in the more virulent savana strain (Higazi *et al*., 2005).

Sex, race, and nutrition are also additional factors to the intensity of the infection and the severity of the ocular manifestations (Thylefors, 1978). Genetic and immunological differences

also serve as basis for ocular pathology (Pearlman and Hall, 2000). Ocular onchocerciasis is predominant in males than females, Thylefors (1978) associated this factor to increase exposure coupled with hormonal and immunological factors in males than females.

Available evidence suggests that responses to antigen specific T cell and antibody are important for *O. volvulus* keratitis (Pearlman and Hall, 2000). These immune cells are needed for the movement of inflammatory cells to the cornea resulting in the failure of cornea brightness (Pearlman and Hall, 2000). Studies from Gillette-Ferguson and colleagues (2004) shows contribution of *Wolbachia* activated neutrophil to the pathogenesis of ocular onchocerciasis.

2.5: Socio-economic and Psychological (psycho-social) Impacts of Onchocerciasis

Onchocerciasis is normally a major risk to public health and impedes socio-economic growth in areas with high intensity and endemicity of the disease (WHO, 1995). According to WHO, adults that are infected with skin diseases as a result of onchocerciasis have serious socio cultural impact on them (WHO, 1995). They have low self-esteem, experience isolation and worry that they will never get married (Okoye and Onwuliri, 2007; Mbanefo *et al*., 2010). The unbearable pruritus and scratching among susceptible individuals may be uncomfortable for weeks. Sometimes the itching and scratching may be so strong for insomnia (Nwoke *et al*., 1987; Wogu and Okaka, 2008). Children on the other hand are distracted in school due to constant itching.

The various skin lesions associated with onchocerciasis like rashes, desquamation, edema and depigmentation put a lot of stress on the lifestyles of infected persons (Nwoke, 1986; Nwoke *et al*., 1987) and can at times cause destitute (Nwoke, 1990). The existence of hanging groin and elephantiasis of the genitalia mostly found in some adult males and genital distortion in some females' causes the infected persons to seclude from other people in his or her environ (Nwoke, 1986). Sexual life of people that are affected with pendulous sacs is greatly impaired

or can be completely disadvantaged (Nwoke, 1986; Nwoke *et al*., 1987). Socioeconomically, skin lesions have been a heavy burden in terms of disability-adjusted life years (Kale, 1998). Constant skin itchings affect productivity as infected individuals has difficulties attending to their jobs (Nwoke *et al*., 1987). It can also lead to complete absenteeism from work (Alonso *et al*., 2009). Large number of nodules particularly around the hip hampers the farmer from farm work. (Nwoke, 1986; Nwoke *et al*., 1987). Research revealed that people infected with onchocercal skin diseases are 15% less productive than those who are not infected because they earned 15% lower in daily wages than those not infected (Kim *et al*., 1997).

Visual impairment incapacitates and turns affected segment of the community into an economic burden (Hamon and Kartman, 1973). As a result of disability and impaired vision, the affected individual is not able to maintain any type of effective work for long, (Ubachukwu *et al*., 2006) that leads into extreme poverty and high numbers of beggars among affected segment of the communities. Their life expectancy also reduces compared to normal people. (Vajime, 1982). Children are made to withdraw from school in order to assist their blind relatives. Mortality among the blind can be four times higher than among the people who are not blind of similar age in the same locality eliminating the supply of individual years of work in the future (Samba, 1994).

The major manifestations of the disease usually restrict human use of fertile lands most often near rivers where the vectors breed (Hamon and Kartman, 1973). As a result, these fertile agricultural lands become desolated, while less fertile highlands become overpopulated due to onchocerciasis. (Hamon and Kartman, 1973). According to Budden (1956), Bradley (1976) and Nwoke (1990), onchocerciasis leads to the reduction in the number of inhabitants and abandonment of lots of productive river lowlands in the savana belt of West Africa. Distorted distribution of population due to depopulation poses serious economic and social setbacks such

as overuse of onchocerciasis free lands, division of family life and labour as men affected by onchocercal blindness leave the villages while women and children remain (Bradley, 1976).

2.6: Diagnosis

The precise and specific diagnosis is important in determining the prevalence of infection with onchocerciasis to identify individuals who require treatment, to monitor the efficacy and effectiveness of treatment and to evaluate the impact of control efforts element. However, this element remains a challenge due to the wide spectrum of clinical manifestations associated with this infection and ineffective diagnostic tools to differentiate past infections from current ones. Parasitological diagnosis that detects microfilariae from a skin snip samples under the microscope is considered to be the traditional diagnostic method. Other diagnostic methods that employ clinical, immunological, molecular and epidemiological techniques are also used in diagnosing onchocerciasis.

2.6.1: Clinical Diagnosis

Presence of onchocercia skin lesions, eye lesions and or subcutaneous nodules with a travel history in an endemic area may be indicative of onchocerciasis. Microfilariae in the cornea and the front cavity of the eye can be diagnosed by a slit lamp. Onchocecal nodules are diagnosed using ultrasonography. Ultrasonography is a useful technique for detecting deep non-palpable nodules. Also, it can be used to evaluate effects of drug on adult worms and as a complement to histological assessment (Mand *et al*., 2005). Certain parasitic infestations can resemble onchocerciasis. Infections caused by *Mansonella streptocerca,* scabies, prickly heat; and contact dermatitis must be distinguished from pruritic onchodermatitis. Presentations for instance sebaceous and dermoid cysts, lipomas, foreign bodies and granulomas that are similar to onchocercal nodules must always be distinguished (Simonsen, 2008).

2.6.2: Parasitological Diagnosis

This is the traditional method of diagnosing onchocerciasis by detecting microfilariae in skin snips under a microscope. Three to five milligram skin snips from an affected area are taken with the help of a razor blade or Walser-type corneoscleral punch. The snips are incubated in sodium chloride solution. Microfilariae that have emerged after 0.5-24 hours are counted under the low power of a compound microscope. Skin snips obtained from the pelvic girdle provide higher chances for detection. The method is widely used for point of care diagnosis and it is very specific but not very sensitive for detecting early, light or pre-patent infections. Another disadvantage of this method is that it is painful and has high risk of blood-borne infections such as human immune deficiency virus (Hagan, 1998; Boatin *et al*., 1998).

2.6.3: Immunological Diagnosis

Mazzotti test is an allergy based reaction test for diagnosing patients with negative skin snips results. The test involves administration of 6mg of diethylcarbamazine (DEC). DEC inhibits neuromuscular transmission in nematode and positive results in two hours generate severe itching and sometimes, intense inflammation in the areas of dying microfilariae. The test may precipitate serious complications for example hypotension, vomiting, conjunctivitis, albuminuria, and sudden death (rare) that restrict its usability. Skin patch test that involves the application of DEC directly onto the skin producing local reaction to dying microfilariae at the patch site is more recently used. The test is non-invasive, simple, cheap, specific, but is not sensitive like skin snip test. Skin patch diagnostic technique could be useful in the future in detecting recrudescence of infection in onchocerciasis free zones (Stingl *et al*., 1984; Toè *et al*., 2000; Boatin *et al*., 2002). Also Enzyme-linked immunosorbent assay (ELISA) has been developed for immunodiagnosis of onchocerciasis. The test requires only a finger prick sample to recognize specific microfilarial antigens. ELISA sensitivity is better than skin snip tests. It is also less invasive than skin snip tests and feasible in the diagnoses of pre-patent infections

or in those with strong immune responses and patients with the reactive forms of the disease. ELISA system assay is also suitable in those below the ages of 15 years and can easily be incorporated into other disease programmes which monitors blood tests such as malaria (Botto *et al*., 1999; Ayong *et al*., 2005). The test however cannot distinguish between past and present infections making its use to be less valuable in endemic areas. Dot blot immunobinding assay (DIA-BA) has recently been suggested. The assay is dependent on the biotin-avidin binding system for detecting specific antigens of *O. volvulus* in body fluids (Ayong *et al*., 2005). The utilization of other species of onchocerca parasite antigens as sources of immunological diagnostic assays has also been reported. (Cho-Ngwa *et al*., 2003).

2.6.4: Molecular Diagnosis

Molecular diagnosis of onchocerciasis uses polymerase chain reaction (PCR)-based methods for the detection of *O. volvulus* DNA in the skin of an infected humans or in the vectors. PCR methods have higher sensitivity in patients whose infection levels are low than skin snip microscopy. The method is also capable of distinguishing between various strains of the parasite. One major disadvantage of PCR is its high cost (Buckley, 1964; Toé *et al*., 1998).

2.6.5: Epidemiological Diagnosis

Rapid epidemiological mapping for Onchocerciasis (REMO) is a standardized epidemiological procedure that is based on nodule palpation and leopard skin search in individuals that might have the disease. The person who performs the technique need to be knowledgeable and must be highly experienced in the field of REMO. REMO is simple, rapid, non-invasive, and cheap. The method is practical and can easily be applied regardless of other ecological conditions. It is also a reliable technique irrespective of the gravity and the period of the infection, non-technological, high acceptability to the rural dwellers with no risk of contaminating any other infections. It greatly helps in impact observation and assessment. A helpful tool in pilot

screening to determine endemicity for control programmes to be started (Edungbola *et al*., 1993; Whitworth and Gemade, 1999; Noma *et al*., 2002).

2.7: Onchocerciasis Treatment

Appropriate treatment of *Onchocerca volvulus* infection needs to be directed towards drugs that can kill the microfilariae, macrofilariae and *Wolbachia* bacterial endosymbionts that are in the endodermis and uteri of adult female worms with no or negligible adverse effects on those that are treated. Problem have been surrounded with the approaches that are used in treating onchocerciasis both in the past and present and these are yet to be resolved. Historically, onchocerciasis treatment until ivermectin discovering was plagued with adverse host reactions basically due to reactions associated with the death of microfilariae. The history of treatment includes nodulectomy, DEC, suramin, ivermectin (Mectizan®) and doxycycline. Presently, ivermectin is the preferred drug for treating and controlling onchocerciasis.

Originally, nodulectomy which involves surgical removal of adult worms in palpable nodules is used to treat onchocerciasis. This method is still in use but partially effective as many worms are outside the nodules. Also, there could be presence of extra nodules deeper in the body that are nonpalpable. (Guderian, 1988). Removal of palpable nodules surgically is common among the Central Americans after the disease has been discovered there (Fernández, 1979). Nodulectomy in cases of recent infections such as infected individuals living in non-endemic areas may be more effective as there is likely to be single nodules present (Fernández, 1979). Nodulectomy of head nodules also help in reducing the risk of eye disease and blindness.

Chemotherapy treatment with DEC came into play with the work of Luis Mazzotti in his ground-breaking research studies in Central America (Mazzotti, 1951). DEC is an oral therapy that is administered in a multiple dose regimes but the characteristic negative skin reactions associated with this drug in patients after treatment led to its discontinues use. Adverse dermal

reactions such as pruritus, papular eruptions, and hyperemia, are linked with the death of the microfilariae in the skin. In patients that has ocular lesions, DEC provokes the situation and can cause loss of vision completely (Bird *et al*., 1980). Suramin, a polyanionic compound is another toxic agent like DEC. It has both microfilaricidal and macrofilaricidal effects. The drug is administered intravenously in repeated doses (6 months) and it is excreted very slowly with severe cumulative toxicity effects (Ghalal, 1985). Problems such as toxicity that are associated with the use of suramin finally brought its use to a stop and it is no more recommended for this indication. The currently approved drug, ivermictin is a compound obtained from *Streptomyces avermitilis* (Njoku *et al*., 2013). Ivermactin therapy does not cause the severe adverse effect associated with DEC. It eliminates the need for 6 weekly injections of suramin and also very suitable for both clinical use and mass distribution in endemic areas. The drug prevents ocular disease, improves and eliminates skin diseases except depigmentation. Ivermectin drug is also known to inhibit the release of late phases of developing mf from the reproductive tract of female worms but does not kill adult worms (Nana-Djeunga *et al*., 2014). This effect last for many months but the underlying mechanism is unknown. A single dose of ivermectin, 150µg/kg brings dermal mf loads to negligible levels for at least 6 months in most people. Adverse reactions associated with IVM treatment include fever, body pains, pruritus, edema and lymphadenitis. They are related to the responses of the body to dying microfilariae, but the intensity and rate of these adverse effects development increases with the treatment. The duration and frequency of ivermactin therapy is still debatable. The only other documented efficacious filaricidal agent for onchocerciasis treatment is doxycycline (Debrah *et al*., 2006). Doxycycline is an antibiotic with a daily dose administration of 100–200 mg for 4–6 weeks (Debrah *et al*., 2006; Hoerauf *et al*., 2009; Tamarozzi *et al*., 2012). The drug eliminates the endosymbiont

Wolbachia required for the worm's survival (Tamarozzi *et al*., 2012). It is the only macrofilaricide available for human use. The drug however requires long duration of treatment and is contraindicated in patients less than 9 years and pregnant women.

2.8: Ivermectin as an Onchocerciasis Control and Preventive Chemotherapy 2.8.1: General uses of Ivermectin

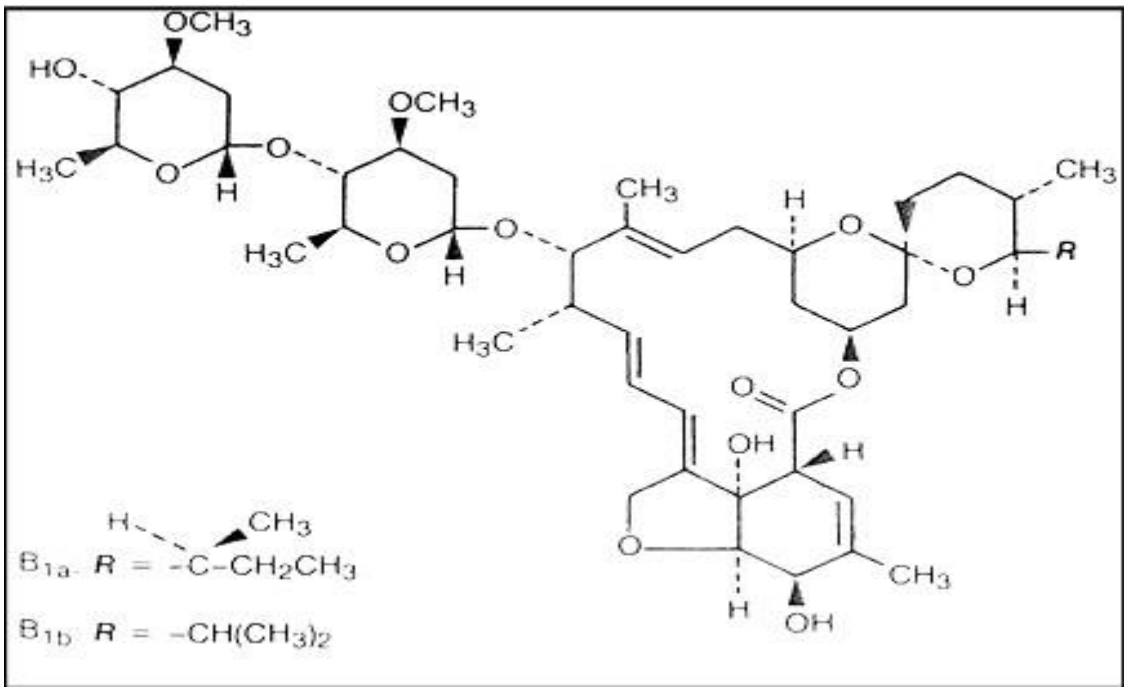

Figure 2.6: Chemical structure of ivermectin (Tanya *et al*., 2013)

Ivermectin is an antiparasitic agent that is useful in veterinary medicine because it has a broad spectrum of activity with high efficacy and wide margin of safety (Fisher and Mrozik, 1989; Steel, 1993). Structurally, it is similar to that of macrolide antibiotics but has no antibacterial activity (Figure 2.2). The drug was developed by Merck & Co. and currently, it is being sold with brand names of Mectizan in Canada by Merck, Stromectol in the United States, Ivomec in Europe by Merial Animal Health, and Ivexterm in Mexico by Valeant Pharmaceuticals International (Pampiglione *et al*., 1985). Ivermectin is a semisynthetic derivative of macro cyclic lactones avermectin B_1 and has original code name MK-933 by

Merck whiles in development (Pampiglione *et al*., 1985). It consists of the equipotent homologous 22, 23 dehydro B_{1a} and B_{1b} in a ratio of 80:20 mixture. Avermectin B was first isolated by means of fermentation of a soil microorganism, the actinomycete *Streptomyces avermitilis.* (Del Guidice and Marty, 1999).

Therapeutically, ivermectin is used in dermatology against some parasitic infestations in humans with cutaneous tropism (Dourmishev *et al*., 2005). The drug is efficacious against different forms of scabies, human body lice, head lice, demodecicosis, cutaneous larva migrans, cutaneous larva currens, myiasis and globally, it is used in the control and elimination of onchocerciasis and to prevent the transmission of lymphatic Filariasis (Dourmishev *et al*., 2005 ; WHO, 2013; TCC, 2013a; TCC, 2013b). In the area of veterinary medicine, ivermectin is used to treat a broad spectrum of animal parasites administered either alone or in combination with other medicine (ASHGI, 2013).

2.8.2: Effect of Ivermectin on *Onchocerca volvulus*

The mode of action of ivermectin involves inhibition of glutamate-gated chloride ion mediums (Glu_Cl) and ϒ_ aminobutyric acid (GABA)-activated chloride mediums existing in nerve and muscle cells of nematodes, bugs and ticks (Wang and Pong, 1982; Campbell *et al*., 1983). This inhibition enables the cell membrane to be more permeable to chloride ions and causes the cell to be hyperpolarized leading to muscular paralysis and death of the parasite (Vuong *et al*., 1992). The paralyzed and dead microfilariae are drained by way of the lymphatic process and destroyed by the immune system, mainly with the aid of macrophages at the level of the lymph nodes (Knab *et al*., 1997). This mechanism additionally results in the disruption of the ingestion of nutrients and explains the speedy drop of microfilaridermia after a specified dose of ivermectin (Vuong *et al*., 1992; Knab *et al*., 1997). The effect results in slow death of the parasite, minimizing inflammation to the host, and alleviates the clinical symptoms of the

disease (Wildenburg *et al*., 1998). Ivermectin also has prolonged effect on the uterus muscles of the adult female worm, which impedes the discharge of microfilariae out of the uterus. These microfilariae are piled up in the genital tract and degenerate in situ (Wildenburg *et al*., 1998).

The only approved route of administration of ivermectin in human is oral and must be taken in an empty stomach (Edwards *et al*., 1988). The drug is absorbed rapidly after intake and metabolized in the liver. 98% of the metabolised drug is excreted in the faeces and 1% in the urine (Edwards *et al*., 1988). Ivermectin has minimal concentration in human breast milk (Fink and Porra, 1989). Blood concentration is at a peak of 30 - 46 ng /mL around 4 hours post doses, and thereafter decreases slowly (Edwards *et al*., 1988; Fink and Porra, 1989). The drug metabolised has longer duration of peak plasma concentration compared to the parent drug, which suggest enterohepatic recycling. The drug was observed to be present in fat, skin, subcutaneous fascia and nodules (Aranzazu *et al*., 2008). Ivermectin reaches peak plasma levels 5 hours after oral administration and has a half-life of 36 hours (Baraka *et al*., 1996). The peak concentration of the drug in squames, sebum and sweat on the forehead and the antithenar was 8 hours after a single oral dose of 12-mg and declined after 24 hours (Haas *et al*.,2002).

Ivermectin does not normally cross the blood-brain barrier of mammals at the normal recommended dosage due to the presence of Permeability glycoprotein (Pgp) and therefore the drug can be used in mammals against invertebrate parasite that either has the Pgp or if present may permit ivermectin to reach all aspects of their neurons (Borst and Schinkel, 1996). The drug is therefore contraindicated in humans and mammals with defect in their Pgp gene (Borst and Schinkel, 1996). It should not be used together with drugs that inhibit Cytochrome P450 3A4 (CYP3A4) enzymes because of their possible inhibition of Pgp transport intensifying the risk of increased absorption of ivermectin across the blood-brain

barrier. Examples of such drugs include statins, many calcium channel blockers, HIV protease inhibitors, and glucocorticoids like benzodiazepine (Brunton *et al*., 2005). Ivermectin is also contraindicated in pregnant women for the period of the first month of lactation, children below the age of 5 years, or patients in bad health (Dourmishev *et al*, 2005).

2.8.3: Resistance and Sub-optimum Response of *O. volvulus* to Ivermectin

The impressive public health impact of MDA programmes on onchocercersis treatment has been well documented (Prichard, 2005). However, these programmes face some challenges as the goals are being translated from control to elimination. One of the challenges is the growing evidence for resistance and sub-optimum response (SOP) of *O. volvulus* to IVM (Prichard, 2005).

O. volvulus resistance and SOR to annual ivermectin treatment is a higher than normal rate of skin repopulation by the microfilariae of the parasites (Taylor *et al*., 2009). It is measured using the skin microfilariae prevalence and or the intensity of infection and confirmed by direct examination of the adult worms. *O. volvulus* resistance to IVM treatment could possibly be in two forms: Lack of parasitological response to IVM treatment or selection for IVM drug resistance in the parasite (Boussinesq and Gardon., 1999; Grant, 2000). Grant (2000) suggested that the manifestation of *O. volvulus*' resistance to IVM in the adult parasite leads to the adult female worms to regain their fertility sooner after IVM treatment and repopulate the skin more rapidly than would occur in a susceptible population (Grant, 2000). The consequences of this manifestation of resistance are more dangerous than a loss in microfilaricidal effect.

Sub- optimal responses of *O. volvulus* to IVM treatment as a result of higher repopulation of skin microfilariae have been reported in Sudan and Ghana (Ali *et al*., 2002; Awadzi *et al*., 2004a, b). The former authors speculated that the observed sub-optimal responses in Sudan

could possibly be due to host immuno-competence. However, when Awadzi *et al* investigated host, pharmaceutical and parasite parameters in some onchocerciasis patients in Ghana, they concluded that the sub-optimal responses were as a result of the development of tolerance or the selection for resistance to IVM in the adult female worms.

Additionally, studies have linked IVM resistance with genetic markers (Huang and Prichard, 1999; Kohler, 2001; Ardelli and Prichard, 2004; Ardelli *et al*., 2005; Eng and Prichard, 2005; Bourguinat *et al*., 2006; Ardelli *et al*., 2006a, b), notably the *β-tubulin* gene in human *O. volvulus* and the livestock nematode parasite *Haemonchus contortus* (Ardelli and Prichard, 2004; Eng and Prichard, 2005; Eng *et al*., 2006; Ardelli *et al*., 2006a, b).

2.9: Control and Treatment of Onchocerciasis in Ghana

Treatment and control of onchocerciasis in Ghana started in the years 1953-1954 where the medical field units of Ministry of Health of Ghana which was then called Gold Coast attempted to control the ocular forms of the disease in the northern territories by means of Antrypol (suramin) (McLean, 1959). In the year 1975 under OCP, vector control of the disease was implemented in West Africa including Ghana (Remme, 2004). Onchocerciasis savana foci of northern and central Ghana were beneficiaries of this program but not the southern forest foci (WHO, 1987). OCP in Ghana started ivermectin distribution in 1987 when the microfilaricidal drug was licensed for human use with the help of mobile teams. By the year 1988, the community directed treatment with ivermectin (CDTI) was accepted for the MDA (Meredith and Dull, 1998; Dull and Meredith, 1998). CDTI remains the delivery approach under the onchocerciasis control program (OCP) and later under APOC when OCP was brought to a closure in 2002. The treatment option for onchocerciasis and lymphatic filariasis co endemic areas is Albendazole and IVM combined. Albendazole and IVM combined treatment started in 2001 in five pilot districts and by 2005; it has reached 61 districts under the initiative of the

Global Programme for the Elimination of Lymphatic Filariasis (GPELF) (WHO, 2008). Through CDTI, population of 3.4 million were treated with a coverage starting from 48.4 to 79.1% between the years 2002-2007.

Onchocerciasis control since 2006 has been carried out in the context of the Neglected Tropical Diseases Control Programme (NTDCP). NTDCP is designated for 5 years with the aim of scaling up delivery of preventive chemotherapy for 5 targeted neglected tropical diseases (NTDs) including onchocerciasis (Taylor *et al*., 2009). The programme was implemented and started on a pilot basis in the Northern, Upper East, Upper West, Western and Brong Ahafo Regions (GHS, 2008).

Additionally, there were joint programmes around the community-directed mass drug treatment that have included vitamin A, schistosomiasis and lymphatic filariasis distribution (GHS, 2008). In 2007, the NTDCP delivered more than 25.5 million ivermectin tablets and more than 8 million tablets of Albendazole for distribution to the endemic areas in Ghana (APOC, 2010).

CHAPTER THREE

MATERIALS AND METHODS

3.1: Study Area

The study took place in 20 endemic communities in the Aowin district in the Western Region. The district is located in the mid-western part of the Western Region of Ghana with a total land area of approximately 2,610.301 square kilometres (PHS, 2010). It has five
Town/Area councils and about 134 communities bordered in the East by the Wassa Amenfi West District, in the Northwest by Suaman-Akontombra District in the North, and Sefwi

Wiawso Districts in the North east and in the South by the Jomoro District. The Republic of La Cote D'Ivoire also shares a common boundary to the Southwest with the District. Enchi is the District capital (Figure 3.1). The district is mainly characterised by low lands with very few hills and has many water bodies such as Disue, Boin and Susan rivers and other streams. River Tano forms a natural boundary between the District and Wassa Amenfi West District. Most of the communities lie closely to these water bodies that serve as breeding sites for the vector blackfly (*Simulium* spp).

The district is located in the rain forest belt of the country and experience high amount of rainfalls throughout the year (about eight months annually). It has eight forest reserves namely Tano Ehuro, Tano Anwia, Tano Nimire, Boin Tano, Jema Assemkrom, Boi River, Disue River and Yoyo. The major occupation of the inhabitant of the district is farming and the district is one of the major producers of cocoa in the country. Cassava is the major food crops grown in the district.

The district has a population of 117,886 which represent 5% of the Western region's total population of 2,376,021 (PHS, 2010). Males constitute 52.0% and females 48%. About

40.8% of the population is below 15 years of age. Majority of the population (90%) live in the rural areas (PHC, 2010).

The communities of the district have houses mostly made of mud and roofed with aluminium sheets or palm and bamboo leaves. The main indigenous ethnic group is the Brusas but other ethnic groups of Ghana such as Northerners, Krobos, Ewes, Fantis and Akans together with Ivorians are found in the district. Brusa is the main traditional language spoken in the district (PHS, 2010).

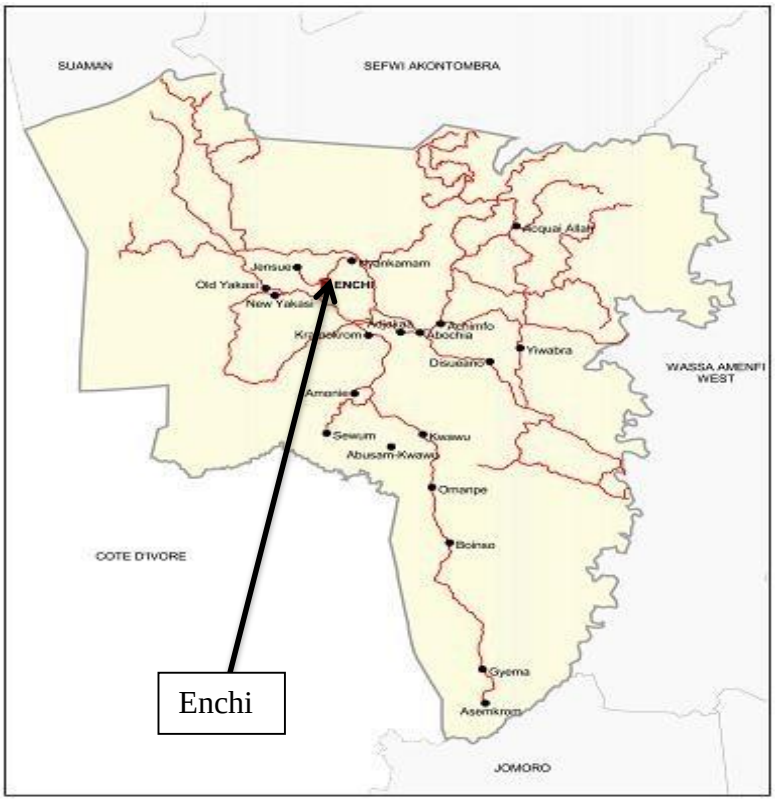

Figure 3.1: Geographic location of the study area (Aowin District) (**Source**: PHS, 2010)

According to the mapping of onchocerciasis carried out by the National NTD Programme of the Ghana Health Service (GHS, unpublished), the district is considered as meso-endemic for onchocerciasis infection. According to the District Health Directorate, ivermectin mass drug administration started in 2003 and ended in 2014 and was distributed by community

volunteers.

3.2: Ethical Approval

Ethical approval was given by the Committee on Human Research, Publication, and Ethics of the School of Medical Sciences of the Kwame Nkrumah University of Science and Technology (KNUST). The study was conducted based on the WHO protocol for epidemiological surveillance and analysis for onchocerciasis control (WHO, 2012).

Additional permission to conduct the study in the selected communities was sought from the Aowin District Health Directorate. Prior to the commencement of the study in each community, meetings were held with the local authorities and community participants to sensitize them on the value of the study and educated them on their right to make a decision to participate or not in the study in their local Twi language. Written informed consent was obtained from all participants either by signing or thumb-printing at the point of registration for the study.

3.3: Study Design

The study was a cross sectional study that evaluated the endemicity and intensity of *O. volvulus* infection in Aowin district of Ghana. Impact of ivermectin treatment on the infection was also assessed. A total of 1,698 volunteers from 20 endemic communities along the main rivers were chosen for the study by purposive sampling (Ngomou and Walsh, 1993). A three month field and laboratory work was conducted from October to December 2015. Volunteers who satisfied the inclusion criteria described in section 3.3.1 were examined for presence of palpable nodules (onchocercoma) and onchodermatitis. Volunteers who had palpable nodules (onchocercoma) or onchodermatitis were snipped for microfilarial load assessment using skin snip (skin biopsy) microscopy.

3.3.1: Inclusion Criteria for the Study

- Be resident in the study area for at least one year
- Both male and females of ages between 18-60 years
- Ability to understand and co-operate with the study principles
- Voluntarily accepting to participate in the study through thumb-print or signing of Informed Consent Form (ICF)

3.3.2: Exclusion criteria for the study

- Non-resident or those who resided in the study area for less than one year

- Volunteers with ages less than 18 or more than 60 years
- People who were not capable of giving Informed Consent

3.4: Study Procedure

3.4.1 Enrolment of the Study Participants

Prior to the enrolment of the study participants, a visit was first made to the chiefs and opinion leaders in each of the 20 selected communities to explain to them the purpose of the study, the need of the community members to participate and how to mobilize the community members for participation. Permission to proceed on the study was then sought. On the day of enrolment of the participants in each community, the community health volunteers helped mobilized the people and the research team explained to them the purpose of the study and the procedures involved. The community members were allowed to ask questions for further clarifications by the research team. Volunteers who met the inclusion criteria were one after the other recruited as they reported and after signing or thumb-printing the Informed Consent Form (ICF) (Plate 3.1) to participate in the study. Thus all the participants in this study were recruited based on their own willingness to participate.

Plate 3.1: Enrolment of the study participant (Source: Author, 2015**)**

3.4.2: Examination for onchocercal palpable nodules

Presence of palpable subcutaneous nodules was determined according to the World Health Organization (WHO) standard protocol (WHO, 1994). The prevalence was determined based on the description by Osei-Atweneboana *et al*.(2007). Examination was done in an enclosed and private area. Onchocercal nodules are firm, often flattened or bean-shaped mobile nontender and up to several centimeters in diameter favoring bony-prominences outside the inguinal and cervical regions (Albiez *et al*., 1988). Before the standardized physical assessment for nodules on each participant, the characteristic nature of the nodules was explained to the participants and each participant was asked if they were aware of any nodules present in their skin. Each volunteer's body was systematically examined by palpation following standardized routine with concentration on the bony prominences for the presence of subcutaneous onchocercal nodules (onchocercoma). While examining for the mobile nodules, the lower ribs,

back, waist, iliac crest, sacrum and hips, as well as the head, legs and arms of the body were searched for onchodermatitis. Onchocercal nodule examinations were done by an experienced oncho specialist from Kumasi Centre for Collaborative Research into Tropical Medicine (KCCR), Ghana.

3.5: Parasitological Examination and Analysis 3.5.1:

Skin Snipping

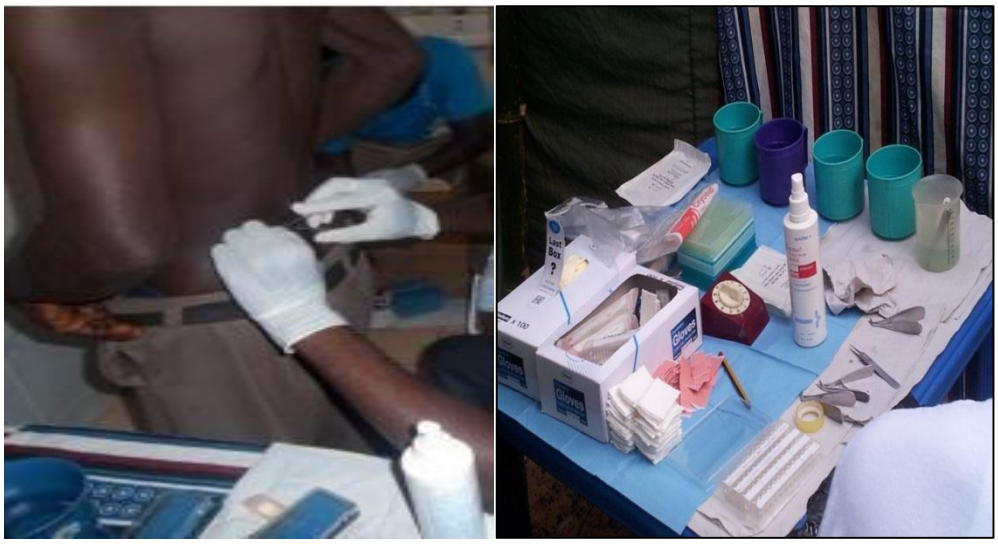

Plate 3.2: Skin snipping (Left). Skin biopsy setup (Right) (Source: Author, 2015**)** The skin to be snipped at the left and right iliac crest were disinfected with 70% alcohol with swaps and allowed to air dry (Plate 3.2, Left). Holth-corneoscleral punches were used to take bloodless skin biopsies from the upper part of the right and left buttocks (iliac crest) from each volunteer. Each biopsy was immersed in 100µl of 0.9% sodium chloride (saline) solution in a separate well of a 96 well round bottom microtitre plate (Nunc, Roskilde,
Denmark) and labeled with the volunteer's unique identification code number. The snipped area was dressed using sterile plaster and the punches sterilized using 10%
Mucocit™ solution (1 in 10 dilution of stock solution with distilled water) for between 5-10 minutes according to the manufacturer's protocol (Plate 3.2, Right).

The wells of the plates were then covered with adhesive tape and safely transported to Aowin District Hospital laboratory. The skin snips were kept at normal temperature in the microtitre plate in sodium chloride solution for 12 -24 hours to enable any mf present to emerge from the skin into the saline solution.

3.5.2: Microfilariae Count

The saline solution in each of the microtitre plate was carefully mixed and pipetted onto a glass slide and was examined unstained under a microscope. Microfilaria counting was made using 10-fold magnification of a compound microscope (Axiostar Plus, Gottingen, Germany).

Each skin biopsy was blotted to remove excess moisture and weighed using OHAUS Adventurer Pro analytical electronic balance (OHAUS, New Jersey, USA). The number of microfilaria from each biopsy was determined as mf per milligram (mf/mg) of skin. The geometric mean of the mf from the two skin biopsies from each patient was calculated and used as a measure of intensity of infection.

3.5.3: Skin Microfilarial load determination

The microfilariae per milligram of skin was determined using skin biopsies taken from the left and right buttocks of the volunteers and this was used to assess the prevalence of microfilaria and the intensity of *O. volvulus* infection. Skin biopsy microscopy is the 'gold standard' in detecting the presence of *O. volvulus* microfilariae in infected populations (Boatin *et al*., 1998).

3.5.4: Community Microfilarial Load determination (CMFL)

The community microfilarial loads (CMFL) were determined and used to evaluate the intensity of *O. volvulus* in the study communities. CMFL is the geometric mean of the individual microfilarial loads (including zero counts) in volunteers aged 20 years or older. This calculation was done using log (n+1) transformation, where n is the individual microfilaria load per snip (mf/mg) (Remme *et al*., 1986).

3.6: Statistical Analysis

The statistical analyses were done using Microsoft Excel® 2010, StatView® 5.0 and GraphPad Prism 5 software programmes. The raw data was entered using Microsoft Excel® and all the other statistics were done using StatView® 5.0 and GraphPad Prism 5. General descriptive information such as the mean and standard deviation from the data were obtained using descriptive statistics. Pearson chi square analysis was used to compare two proportions or groups. For non- parametric data set, analysis was done using Spearman rank correlation. A p-value of less than 0.05 ($P < 0.05$) was considered statistically significant.

CHAPTER FOUR

RESULTS

4.1: Age and Sex Distribution of the Study Participants

The participants were made up of 894 (52.7%) males and 804 (47.4%) females. The mean ages were 38.1 years (male) and 35.5 years (female) and an overall mean age of 36.9years.

The age range was between 18- 60 for both sexes (Table 4.1).

Table 4.1: Demographic data of the study participants

Sex of volunteers examined	Number examined	Age (mean ±SD)/years
Male	894	38.1 ± 12.3
Female	804	35.5 ± 12.0
Total	1698	36.9 ± 12.2

Out of the total 1698 study participants, 300 were skin snipped for microfilarial load assessment. The snipped participants were made up of 210 (70%) males and 90 (30%) females.

4.2: Onchocercal Nodule Prevalence in the Study Participants

Out of the total 1,698 volunteers examined from the 20 communities of the Aowin districts, 298 were positive for palpable nodules (onchocercomas) representing 17.6% (Table 4.2). All the study communities had palpable nodule prevalence range between 6.1-28.9% indicating hypo-endemicity. The lowest prevalence was recorded in Nkwanta No2 (6.1%) and the highest in Kwaku Atta No1 (28.9%).

Table 4.2: *O. volvulus* nodule prevalence in the selected communities

Community	Total number examined	Number with Nodules (%)	Number without Nodules (%)
Abotareye	76	20 (26.3)	56 (73.7)

Agyeikrom	82	16 (19.5)	66 (80.5)
Aleobo	97	15 (15.5)	82 (84.5)
Amoakrom	106	11 (10.4)	95 (89.6)
Amowie	123	32 (26.0)	91 (73.8)
Anwiafutu	98	11 (11.2)	87 (88.8)
Anwiafutu No2	82	19 (23.2)	63 (76.8)
Bodiewu	85	19 (22.4)	66 (77.7)
Bowohomoden	83	21 (25.3)	62 (74.7)
Camp 4	71	9 (12.7)	62 (87.3)
Disueano	89	8 (9.0)	81 (91.0)
Eboebo	60	14 (23.3)	46 (76.7)
Gyasikrom	111	12 (10.8)	99 (89.2)
James Adom	65	5 (7.7)	60 (92.3)
Kojo Bortey	86	15 (17.4)	71 (82.6)
Kordjour	62	8 (12.9)	54 (87.1)
Kwaku Atta No1	83	24 (28.9)	59 (71.1)
Kwaku Atta No2	84	23 (27.4)	61 (72.6)
Nkwanta No2	115	7 (6.1)	108 (93.9)
Pillar 3	40	9 (22.5)	31 (77.5)
Total	1698	298 (17.6)	1400 (82.5)

4.3: Age and Gender of Onchocercal Nodule Carriers

Table 4.3 shows gender related prevalence of onchocercal nodule grouped in ages. In all, 298 volunteers were positive for palpable nodules and out of this number, 208 (23.3%) were males whiles 90 (11.2%) were females. The difference between male and female nodule carriers was statistically significant ($p = 0.0001$). Onchocercal prevalence in relation to agegroup and sex is presented in a histogram (Figure 4.1). From Figure 4.1, it was observed that all the age groups in both sexes had nodules. Nodule prevalence was highest in males than females in all the age groups with the highest observed to be in the age group of 41-50 years. The graph (Figure 4.1) showed general progressive increase in nodule prevalence with increase in age. Figure 4.2 shows the correlation between the number of palpable nodules and age of the volunteers with coefficient of correlation being $r=0.145$ which was significant ($p=0.012$). From the correlation graph, the number of palpable nodules from 1-4 were observed to be equally distributed among all the ages (18-60 years) of the positive volunteers. Older positive individuals above 30 years

had palpable nodules ranging from 4-6. The highest number of nodule (9) was recorded in a volunteer of 30 years of age.

Table 4.3: Age and gender of onchocercal nodule carriers

Age group	Total Palpated	Nodule positive (Total) (%)	Male Palpated	Nodule Positive (Male) (%)	Female Palpated	Nodule Positive (Female) (%)
18-30	630	67 (10.6)	291	47 (16.2)	339	20 (5.9)
31-40	454	94 (20.7)	236	61 (25.8)	218	33 (15.1)
41-50	328	72 (22)	188	53 (28.2)	140	19 (13.6)
51-60	286	65 (22.7)	179	47 (26.3)	107	18 (16.8)
Total	1,698	298 (17.6)	894	208 (23.3)	804	90 (11.2)

($p = 0.0001$)

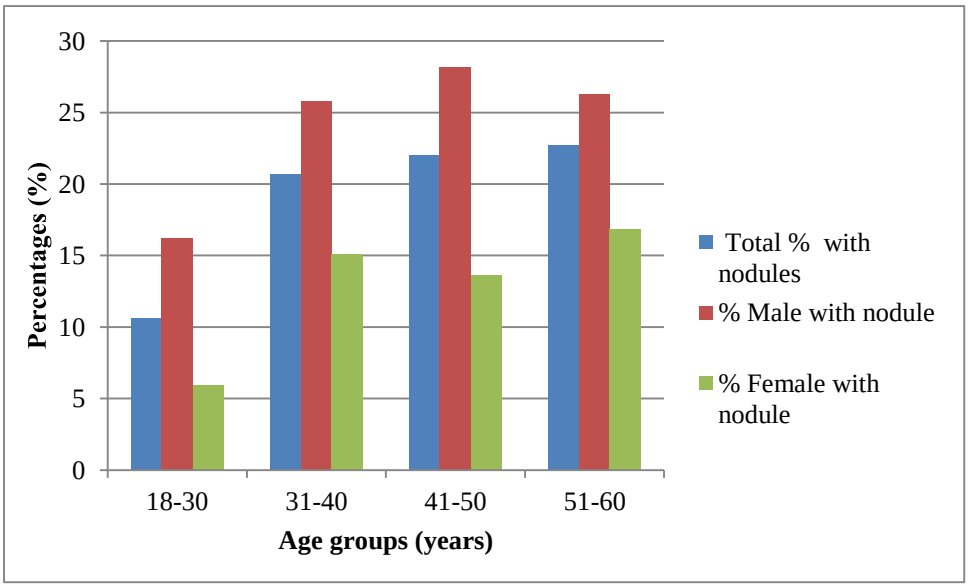

Figure 4.1: Histogram showing the prevalence of nodules on the basis of age groups and gender of volunteers

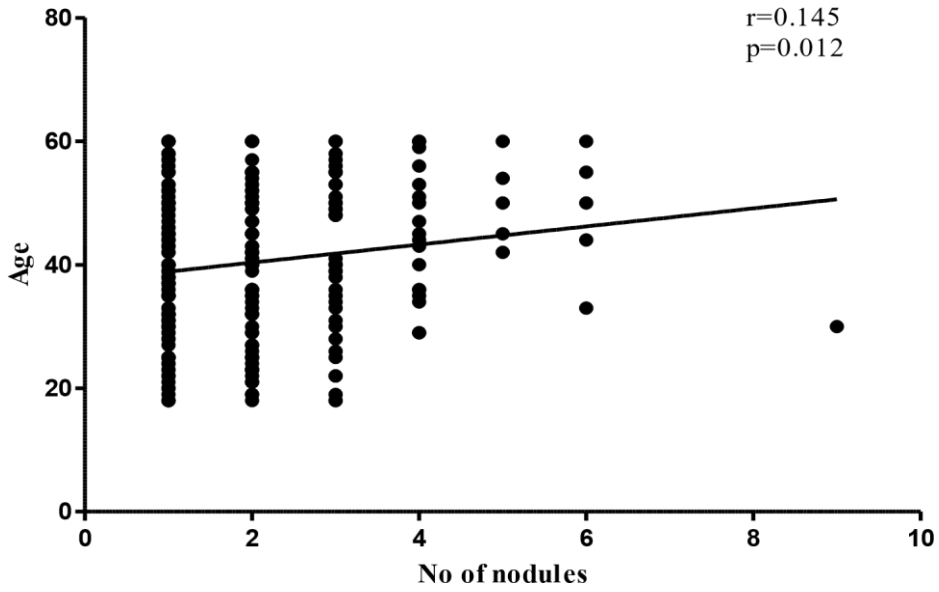

Figure 4.2: Correlation between number of onchocerca nodules and age of volunteers

4.4: Prevalence of *O. volvulus* Microfilariae among Nodule Carriers in the Study Communities

Out of the 298 palpable nodule positive volunteers recorded, 290 individuals took part in the skin snip microfilarial load assessment (Table 4.4). From the snipped volunteers, 169 with overall prevalence of 58.3% were microfilariae positive.

Table 4.4: Microfilarial prevalence among nodule carriers in the study communities

Communities	Total number of volunteers snipped	Number of volunteers with mf (%)	Number of volunteers with no mf (%)
Abotareye	**18**	**8 (44.4)**	**10 (55.6)**
Agyeikrom	**16**	**13 (81.3)**	**3 (18.8)**
Aleobo	**15**	**6 (40.0)**	**9 (60.0)**
Amoakrom	**11**	**6 (54.6)**	**5 (45.5)**
Amowie	**31**	**27 (87.1)**	**4 (12.9)**
Anwiafutu	**12**	**1 (8.3)**	**11 (91.7)**
Anwiafutu No2	**19**	**7 (36.8)**	**12 (63.2)**
Bodiewu	**18**	**14 (77.8)**	**4 (22.2)**
Bowohomoden	**21**	**14 (66.8)**	**7 (33.3)**
Camp 4	**9**	**7 (77.8)**	**2 (22.2)**
Disueano	**8**	**1 (12.5)**	**7 (87.5)**
Eboebo	**14**	**10 (71.4)**	**4 (28.6)**
Gyasikrom	**12**	**3 (25.0)**	**9 (75.0)**
James Adom	**5**	**1 (20.0)**	**4 (80.0)**
Kojo Bortey	**13**	**6 (46.2)**	**7 (53.9)**
Kordjour	**9**	**8 (88.9)**	**1 (11.1)**
Kwaku Atta No1	**22**	**16 (72.7)**	**6 (27.3)**
Kwaku Atta No2	**22**	**13 (59.1)**	**9 (40.9)**
Nkwata No2	**6**	**2 (33.3)**	**4 (66.7)**
Pillar 3	**9**	**6 (66.7)**	**3 (33.3)**
Total	**290**	**169 (58.3)**	**121 (41.7)**

The microfilarial prevalence among the nodule carriers recorded from the study communities ranges from 8.3-88.9% with Anwiafutu recording the least and Kordjour, recording the highest. From Table 4.4, 4 communities were observed to be hypoendemic as they had microfilarial prevalence of less than 30%. Eleven (11) communities had microfilarial prevalence between

30-75% indicating mesoendemicity and 5 recording more than 75% microfilarial prevalence indicating hyperendemicity.

4.5: Actual Microfilarial Prevalence in the Selected Communities.

A total of 300 individuals consisting 290 nodule carriers and 10 onchodermatitis positive individuals from the 20 endemic communities were snipped for skin snip microfilarial load assessment. They were also used to determine the actual microfilarial prevalence in each of the study communities (Table 4.5). Out of the 300 volunteers snipped, 173 were microfilarial positive giving an overall prevalence of 10.2%. From Table 4.5, Anwiafutu recorded the least (1.0%) and Amowie recorded the highest (22.0%). None of the communities studied had microfilarial prevalence that was < 1% that could meet the criteria of potential evidence for possible elimination of the infection in any of the studied communities.

Table :

4.5 Actual microfilarial prevalence in the study communities

Communities	Total number of volunteers examined	Number of volunteers with mf (%)	Number of volunteers with no mf (%)
Abotareye	76	8 (10.5)	68 (89.5)
Agyeikrom	82	13 (15.9)	69 (84.1)
Aleobo	97	6 (6.2)	91(93.8)
Amoakrom	106	6 (5.7)	100 (94.3)
Amowie	123	27 (22.0)	96 (78.1)
Anwiafutu	98	1 (1.0)	97 (99.0)
Anwiafutu No2	82	8 (9.8)	74 (90.2)
Bodiewu	85	14 (16.5)	71 (83.5)
Bowohomoden	83	14 (16.9)	76 (91.6)
Camp 4	71	9 (12.7)	62 (87.3)
Disueano	89	1 (1.1)	88 (98.9)
Eboebo	60	10 (16.7)	50 (83.3)
Gyasikrom	111	3 (2.7)	108 (97.3)
James Adom	65	1 (1.5)	64 (98.5)
Kojo Bortey	86	6 (7.0)	80 (93.0)
Kordjour	62	8 (12.9)	54 (87.1)
Kwaku Atta No1	83	17 (20.5)	66 (79.5)
Kwaku Atta No2	84	13 (15.5)	71 (84.5)
Nkwata No2	115	2 (1.8)	113 (98.3)
Pillar 3	40	6 (15.0)	34 (85.0)
Total	1698	173 (10.2)	1525 (89.8)

4.6: Age and Sex of Microfilariae Positive Volunteers

From Table 4.6, 173 (57.7%) participants were positive for *O. volvulus* microfilariae. Out of this number, 123 (58.6%) were males while 50 (55.6%) were females. The overall microfilarial prevalence was highest in males than females but with no significant difference (p=0.6280). Figure 4.3, shows the prevalence of microfilariae on the basis of age groups and gender of volunteers. It was observed that all the age groups recorded positive for microfilariae. Males in the age group of 18-30 and 31-40 had highest microfilaria prevalence than females. But in the age group of 41-50 and 51-60, it was the females who recorded the highest mf prevalence.

Table :

4.6 Gender dependent prevalence of microfilariae grouped in ages

Age group	Total Snipped	mf positive (Total) (%)	Males snipped	mf positive (Males) (%)	Females snipped	mf positive (Females) (%)
18-30	65	43 (66.2)	47	33 (70.2)	18	10 (55.6)
31-40	95	50 (52.6)	60	35 (58.3)	35	15 (42.9)
41-50	75	46 (61.3)	55	31 (56.4)	20	15 (75.0)
51-60	65	34 (52.3)	48	24 (50)	17	10 (58.8)
Total	300	173 (57.7)	210	123 (58.6)	90	50 (55.6)

(Chi square test for male against female, p= 0.6280)

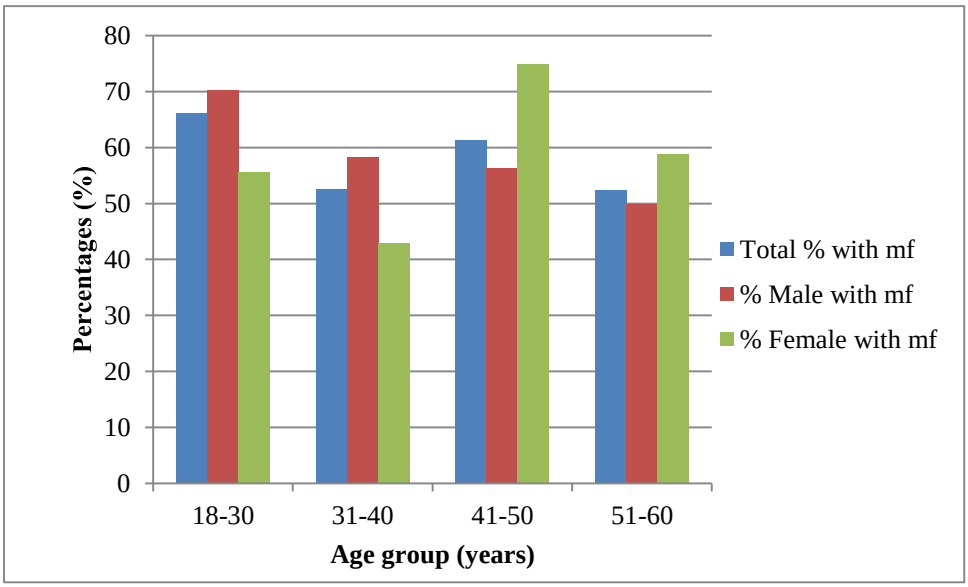

Figure 4.3: Histogram showing the prevalence of microfilariae on the basis of age groups and gender of volunteers

4.7: Community Microfilarial Load of the Selected Communities

The 300 individual snipped volunteers from the 20 studied communities were used to determine the community microfilarial load (CMFL). From Table 4.7, the overall CMFL for Aowin district was 2.3mf/mg and it ranged between 1.0-5.2mf/mg for the 20 communities studied. James Adom had the lowest while Bodiewu had the highest.

4.7 Community Microfilarial Load of the selected communities

Communities	Total number of volunteers examined	CMFL

Table :

Abotareye	19	1.8
Agyeikrom	17	2.5
Aleobo	16	1.7
Amoakrom	11	2.3
Amowie	31	4.2
Anwiafutu	12	1.1
Anwiafutu No2	21	1.9
Bodiewu	19	5.2
Bowohomoden	21	3.2
Camp 4	11	1.6
Disueano	8	1.3
Eboebo	14	2.8
Gyasikrom	13	1.3
James Adom	5	1.0
Kojo Bortey	13	1.3
Kordjour	9	4.4
Kwaku Atta No1	23	2.7
Kwaku Atta No2	22	2.8
Nkwata No2	6	1.3
Pillar 3	9	2.4
Total	300	2.3

:

Table 4.8 Variation of microfilarial load densities among the positive volunteers in the study communities Communities Total number Number of volunteers with microfilariae

	of participants with mf	per mg of snip of (%)					
		0.01-4.9	5-9.9	10-29.9	30-49.9	50-79.9	≥ 80.0
Abotareye	8	5 (62.5)	1 (12.5)	2 (25.0)	0 (0.0)	0 (0.0)	0 (0.0)
Agyeikrom	13	10 (76.9)	2 (15.4)	0 (0.)	1 (7.7)	0 (0.0)	0 (0.0)
Aleobo	6	3 (50)	1 (16.7)	2 (33.3)	0 (0.0)	0 (0.0)	0 (0.0)
Amoakrom	6	3 (50)	1 (16.7)	1 (16.7)	1 (16.7)	0 (0.0)	0 (0.0)
Amowie	27	17 (63)	3 (11.1)	4 (14.8)	1 (3.7)	0 (0.0)	2 (7.4)
Anwiafutu	1	1 (100)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
Anwiafutu No2	8	5 (62.5)	1 (12.5)	1 (12.5)	0 (0.0)	1 (12.5)	0 (0.0)
Bodiewu	14	6 (42.9)	2 (14.3)	2 (14.3)	2 (14.3)	1 (7.1)	1 (7.1)
Bowohomoden	14	8 (57.1)	1(7.1)	3 (21.4)	1 (7.1)	0 (0.0)	1 (7.1)
Camp 4	9	9 (100)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
Disueano	1	1 (100)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
Eboebo	10	6 (60)	1 (10.0)	3 (30.0)	0 (0.0)	0 (0.0)	0 (0.0)
Gyasikrom	3	3 (100)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
James Adom	1	1 (100)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
Kojo Bortey	6	6 (100)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
Kordjour	8	4 (50)	2 (25.0)	2 (25.0)	0 (0.0)	0 (0.0)	0 (0.0)
Kwaku Atta No1	17	13 (76.5)	0 (0.0)	3 (17.7)	1 (5.9)	0 (0.0)	0 (0.0)
Kwaku Atta No2	13	7 (53.9)	3 (23.1)	1 (7.7)	2 (15.4)	0 (0.0)	0 (0.0)
Nkwata No2	2	2 (100)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
Pillar 3	6	5 (83.3)	1 (16.2)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
Total	173 (100)	115(66.5)	19 (11)	24 (13.9)	9 (5.2)	2 (1.2)	4 (2.3)

4.8: Microfilarial Load Densities in the Communities

Table 4.8 shows the variations of microfilarial load densities among the microfilarial-positive individuals in the study communities. Of the 6 microfilarial loads density ranges, majority of the participants (66.5%) were in the density range 0.01-4.9mf/mg and seven of the 20 communities consisting of 22 positive individuals had microfilarial density belonging to only this range. The 50-79.9 range was the least group with only 2 (1.2%) mf-positive individuals from Anwiafutu No2 and Bodiewu communities. Only Amowie, Bodiewu and Bowohomoden communities recorded 4 mf positive volunteers belonging to the highest mf

density of ≥80.0mf/mg and Bodiewu was the only community with positive mf volunteers belonging to all the 6 mf density ranges.

4.9: Comparison of Number of Nodules and Microfilarial Status of the Volunteers Snipped

Table 4.9 compared the number of palpable nodules recorded for each volunteer to microfilarial status. Among the 300 volunteers snipped, 10 (3.3%) had no palpable nodules but only onchodermatitis whiles 290 (96.7%) had palpable nodules. Out of the 10 onchodermatitis individuals, 4 (40.0%) had microfilariae. Those with palpable nodules had only one volunteer with 9 nodules and also positive for microfilariae. Volunteers with 6 palpable nodules had 4 (80.0%) out of 5 individuals to be positive for microfilariae. Generally, the observation made from the table indicated that as the number of palpable nodule counts increased, the percentage of individuals with positive microfilariae count also increased.

Table 4.9: Number of nodules compared with microflarial status of the snipped volunteers

Number of nodules	Number of volunteers snipped (%)	Number of volunteers with: (%)	
		no microfilariae	microfilariae
0 (onchodermatitis)	10 (3.3)	6 (60.0)	4 (40.0)
	(49.3)	(47.3)	(52.7)
	(24.7)	(40.5)	(59.5)
3	(13)	(28.2)	(71.8)
4	(6.0)	(38.4)	(61.1)
5	(1.7)	(40.0)	(60.0)
6	(1.7)	(20.0)	(80.0)
9	(0.3)	(0.0)	(100)
	(100)	(42.3)	(57.7)

1
2

4. 10: Comparison of Number of Rounds of IVM intake against Onchocercal Nodules and Microfilariae Positivity

Figure 4.4 shows a graph of nodules and microfilariae positive volunteers and their IVM treatment rounds. IVM treatment rounds ranged from 0 (those who had never taken IVM before) up to ≥ 10 (those who took IVM 10 times or more). From the graph, 61 (20.5%) nodule

positive volunteers and 32 (51.6%) mf positive volunteers took IVM for 3 rounds and had the highest number out of the total positive for nodules and mfs respectively. There was no volunteer with IVM round of 9. The general trend observed from the graph was that as the number of rounds of IVM intake increased, nodule and microfilarial positivity decreased.

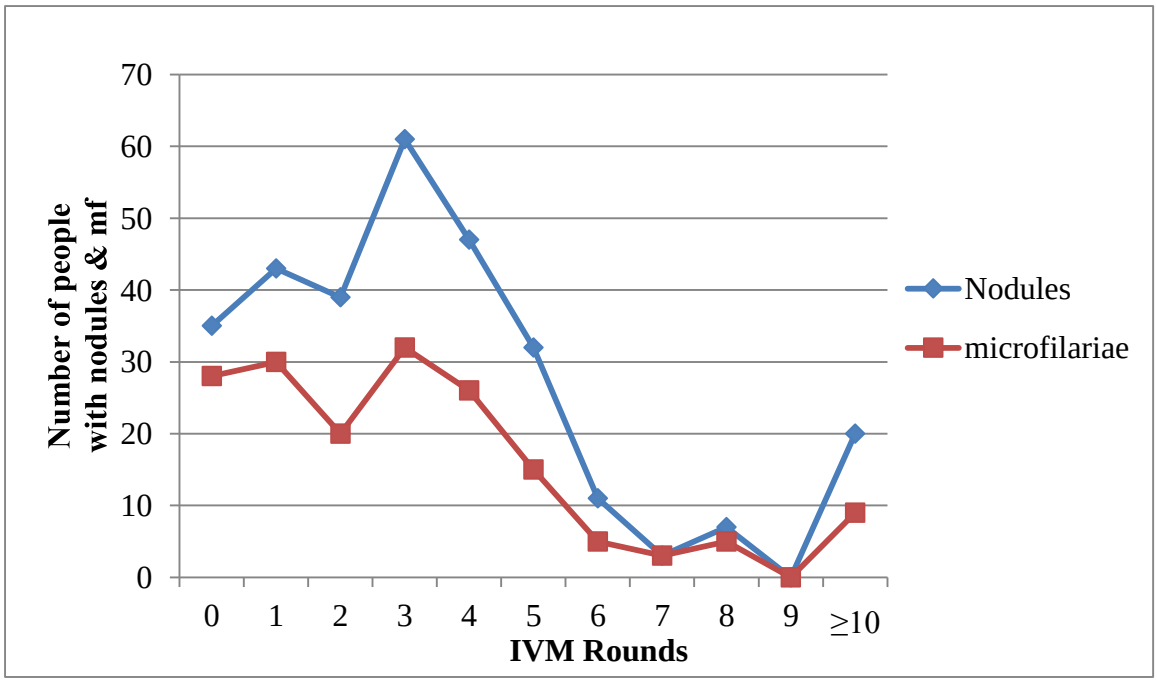

Figure 4.4: Comparison of number of rounds of ivermectin treatment against nodules and microfilarial positivity of snipped volunteers

4.11: Correlation between IVM Treatment Rounds and Number of Nodules

From the correlation graph (Figure 4.5), those who had never taken IVM before had at least 1- 6 palpable nodules. Only one volunteer who had taken IVM for 6 rounds had 9 palpable nodules. Generally, as the number of IVM rounds intake increased, the number of palpable nodules counted decreased with r = - 0.05438. The correlation coefficient was not significant (p = 0.3495).

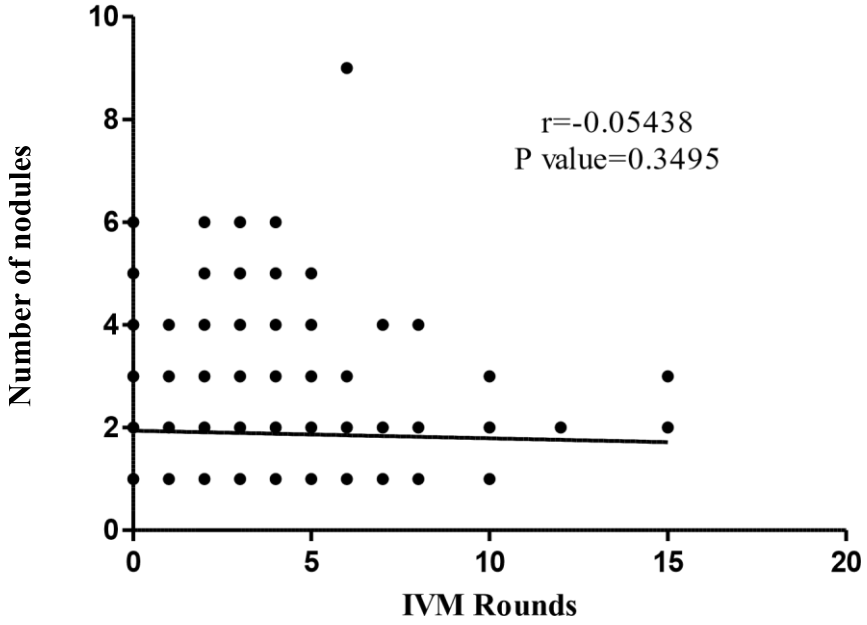

Figure 4.5: Correlation between IVM treatment rounds and number of nodules

4.12: Correlation between IVM Treatment Rounds against Mean Microfilariae (mf/mg)

Figure 4.6 shows the correlation between the IVM treatment rounds and the mean microfilarial per mg of skin snip for the volunteers who were positive for microfilariae. From the graph and Table 4.10, those who had never taken IVM before had their mean microfilariae per mg of skin snip ranging from 0.3-106.7mf/mg.

The highest mf/mg of 116.7 was recorded in only one volunteer who had taken IVM for 6 rounds. Generally, as the IVM treatment rounds increased, the mf/mg decreased with coefficient of correlation being r = -0.3636 ($p < 0.0001$).

Table 4.10: IVM treatment rounds and microfilariae load range (mf/mg)

IVM Treatment	mf load range (mf/mg) rounds
2	0.01 - 36.7

3		0.05 - 41.7
4		0.05 - 25.7
0	0.3 - 106.7	0.05 - 51.5
1	0.1 - 81.2	
6		
		0.8 - 116.7
7		0.2 - 21.7
8		0.2 - 12.57
9		0
≥10		0.06 - 6.7

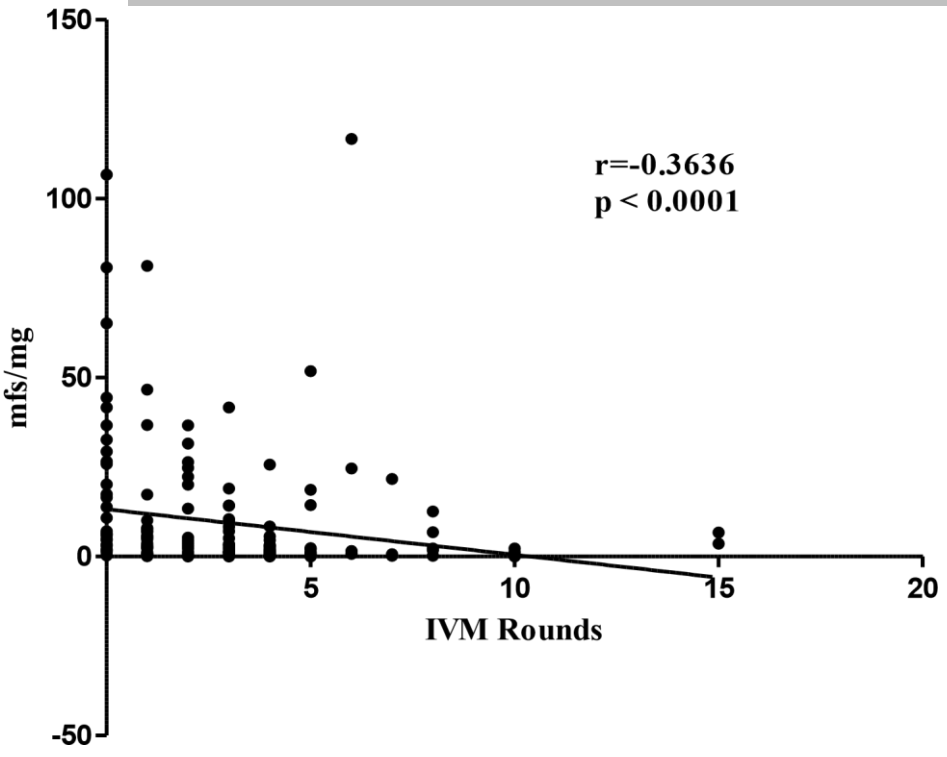

Figure 4.6: Correlation between IVM treatment rounds and microfilariae load (mf/mg)

4.13: Number of Ivermectin Rounds Intake against Microfilarial Load Densities

The number of rounds of IVM intake was compared to microfilarial load densities in Table 4.11. From the table, it can be observed that only volunteers who had never taken IVM before recorded microfilarial load densities across all the 6 mf density groups. No volunteer who had taken IVM for 9 rounds had mf in the skin. All the IVM treatment rounds with the exception of 9 have representatives in 0.01-4.9 and 10-29.9 mf density groups. The highest mf density group of ≥ 80 had 4 representatives belonging to 0 IVM (2 mf positive individuals), 1 IVM (1 mf positive individuals) and 6 IVM (1 mf positive individuals) treatment rounds. The microfilarial load densities generally declined as the IVM treatment rounds increased.

Table 4.11: Comparison of number of rounds of ivermectin intake with microfilarial load densities IVM Volunteers Total Number of volunteers with microfilariae per snip (mf/mg) of: Treatment examined number of (%)

rounds	(%)	mf positives						
			0.01-4.9	5-9.9	10-29.9	30-49.9	50-79.9	≥80
0	35 (11.7)	28	9 (32.1)	4 (14.3)	8 (28.6)	4 (14.3)	1 (3.6)	2 (7.1)
1	43 (14.3)	30	18 (60.0)	7 (23.3)	2 (6.7)	2 (6.7)	0 (0.0)	1 (3.3)
2	40 (13.3)	20	12 (60.0)	1 (5.0)	5 (25.0)	2 (10.0)	0 (0.0)	0 (0.0)
3	62 (20.7)	32	23 (71.9)	4 (12.5)	4 (12.5)	1 (3.1)	0 (0.0)	0 (0.0)
4	46 (15.3)	26	23 (88.5)	2 (7.7)	1 (3.9)	0 (0.0)	0 (0.0)	0 (0.0)
5	33 (11.0)	15	12 (80.0)	0 (0.0)	2 (13.3)	0 (0.0)	1 (6.7)	0 (0.0)
6	11 (3.7)	5	3 (60.0)	0 (0.0)	1 (20.0)	0 (0.0)	0 (0.0)	1 (20.0)
7	3 (1.0)	3	2 (66.7)	0 (0.0)	1 (33.3)	0 (0.0)	0 (0.0)	0 (0.0)
8	7 (2.3)	5	3 (60.0)	1 (20.0)	1 (20.0)	0 (0.0)	0 (0.0)	0 (0.0)
9	0 (0.0)	0	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
≥10	20 (6.7)	9	8 (88.9)	1 (11.1)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
Total	300 (100)	173						

CHAPTER FIVE

DISCUSSION

5.1: Introduction

Elimination of *Onchocerca volvulus* infection with long term ivermectin mass drug treatment (IVM) from endemic areas in Africa might be unlikely according to some authors (Borsboom *et al*., 2003). However, studies in Mali, Nigeria and Senegal gave evidence of successful elimination of the disease transmission (Diawara *et al*., 2009; Tekle *et al*., 2012). Following these successes, the possibility of eliminating onchocerciasis from Africa by mass treatment with ivermectin has been rejuvenated (Coffeng *et al*., 2013). In Ghana, mass drug treatment with ivermectin started in 1987 with the aim of reducing the community microfilarial load (CMFL) below 0.05mf/mg by the year 2015 (GHS, 2008). In this study, the impact of IVM MDA on the level of endemicity and intensity of infection was examined in the Aowin District in the Western Region of Ghana where mass treatment with IVM was administered for 10 years and the District Health Directorate (DHD) claimed the infection had been eliminated.

The endemicity of *O. volvulus* infection was determined using nodules and microfilariae prevalence. Community microfilarial load (CMFL), a method described by OseiAtweneboana *et al*., (2007) was used to determine the intensity of the infection. A nodule and microfilariae prevalence of 75% or more is indicative of hyperendemicity and that between 30 and 75% indicate mesoendemicity and below 30% is hypoendemicity (Vivas-Martinez *et al*., 2000). Also, CMFL value of more than 5mf/mg would mean that the infection still remains a public health problem (APOC, 2010).

5.2: The Endemicity of *O. volvulus* in the Study Communities 5.2.1: Endemicity using Nodule and Microfilarial Prevalence

The average prevalence of onchocercal nodules in the 20 selected communities was 17.6% ranging from 6.1-28.9%. Comparing the average nodule prevalence result with that of the threshold needed to launch mass treatment programmes (nodule prevalence > 20% or mf

prevalence > 40%), the prevalence is high and uncomfortably close to the threshold despite more than a decade of mass treatment with ivermectin. Nine of the 20 communities studied had nodule prevalence above the 20% threshold (Table 4.2). The high prevalence of palpable onchocercal nodules as recorded in this study is indicative of ongoing *O. volvulus* infection transmission as it is a highly significant risk factor for all forms of onchocercal skin disease (Ozoh *et al*., 2011).

Palpable nodules was significantly higher in males than females (23.3% versus 11.2%; p=0.0001) (Table 4.3 and Figure 4.1). Higher onchocercal nodule prevalence recorded in males than females in this study agreed with the patterns of *O. volvulus* infection which have been shown to vary markedly with locality, sex- and age-dependent exposure to the vector (Filipe *et al*., 2005). *O. volvulus* establishment in humans, according to Basanez and Boussinesq (1999), is determined by exposure to infective stages of the parasite. Members of the various communities where this study was performed were mainly cocoa farmers and the males were mostly the ones that were actively engaged in farming activities over the years implying that their risks of exposure to the blackfly bites were higher than those of females in the same communities. This higher risk of exposure may explain why the male participants recorded significantly higher onchocercal nodule prevalence than the females.

Prevalence of onchocercal nodules was recorded in all the age groups. The result showed a general increase in prevalence with increase in age (Figure 4.1). The observed progressive increase in nodule prevalence with increase in age may be that the individuals in these communities have been exposed to the bite of the black flies throughout their lives due to their farming activities. The observed nodule prevalence recorded in all the age groups in both sexes indicated that both males and females of different age groups had similar tendency to the infection. Notably the prevalence was higher in males at all the age groups than females (Figure

4.1). Higher nodule prevalence in males than females is often common as a result of occupational differences between males and females. Males unlike females are always engaged in active farming activities right from childhood. Moreover, females' farming activities are even reduced during their child bearing ages when they have to assumed roles of motherhood and spend most of their times at home performing household activities preventing them from being exposed to the bite of the black flies. The coefficient of correlation between number of nodules and age of the participants was weak but statistically significant (Figure 4.2). The weak correlation between number of nodules and age of the participants suggests that, aside age being a factor to the acquisition and formation of onchocercal nodules, other factors such as the intensity of infection coupled with hormonal and immunological factors of the host may play a role as suggested by other studies (Mackenzie *et al*. 1985; Wilson *et al*., 2003).

The overall microfilarial prevalence recorded among nodule carriers in the study communities (58.3%) indicated the presence of live female worms, inseminated female worms in the nodules and living male worms, which were producing microfilariae into the dermis of the skin. This increased the risk of infecting black flies for transmission of the infection to continue. The actual average microfilarial assessment among the volunteers who participated in the study revealed a prevalence of 10.2% indicating hypo-endemicity. The change from meso-endemicity before the launch of the MDA of ivermectin programme in 2003 to hypo-endemicity status could be due to the impact of ivermectin MDA in eliminating the disease as a public-health burden but the prevalence recorded remained unacceptably high for transmission of the disease to be eliminated in the study area. The effects of Ivermectin treatment are primarily to reduce mf in the skin. This reduction lasts for a period of three to six months (Basanez *et al*., 2008; Pion *et al*., 2011). The drug also has effect on the number of

microfilariae that the vector is able to ingest in the course of a blood meal but the treatment effects will not necessarily impact the adult worms that reside in the nodules (Pion *et al*., 2011). However, long term treatment with the drug is able to significantly reduce the endemicity level of *O. volvulus* infection and it is expected that with repeated rounds of ivermectin MDA, the infection can be reduced to below transmissible levels (Opara and Fagbemi, 2008). The high rate of microfilariae recorded despite more than 10 years of ivermectin mass drug administration suggested repopulation of microfilariae into the skin from the progeny of resistant adult parasite that might emerge (Osei-Atweneboana *et al*.,
2007). It could also be due to failure to achieve adequate drug coverage (Cupp *et al*., 2007; Mackenzie, 2007) or possibly from highly fecund worms that were able to recover more quickly from treatment (Remme *et al*., 2007). Even though there has been a change from meso-endemicity to hypo-endemicity in the study area, stopping CDTI mass treatment activities can result in disease recrudescence.

In this study onchocerca infection using microfilariae detection was higher in males than females but there was no significance difference between the genders (p=0.6280). The prevalence peaked in volunteers aged 18-30 years (Figure 4. 3). This observation is in conformity to previous studies that have indicated that onchocerca infection using microfilariae detection peaks in individuals aged 20-30 years and is higher in males than females (Hailu *et al*., 2002). However, this study did not agree with the findings by AntwiBerko, (2014) who worked in two onchocerca endemic districts in the Ashanti Region of Ghana and observed microfilariae peak in volunteers aged 31 to 40 years. The difference in the peaking age groups in this study compared to that observed by Antwi-Berko, (2014) may be due to difference in geographical locations and the patterns of exposure to blackflies and parasite acquisition of the locals (Filipe *et al*., 2005).

5.3: The Intensity of *O. volvulus* Infection in the Study Communities

The community mf load (CMFL) which is equal to the geometric mean mf load per snip in adults aged ≥ 20 years is the standard measure of intensity of *O. volvulus* infection (Remme *et al*., 1986). The infection in a community is considered to be a public health importance when the CMFL is more than 5mf/mg (APOC, 2010) as high microfilarial load negatively affects its host life expectancy (Little *et al*., 2004a). According to Duerr *et al*., (2005), to achieve ≥ 90% probability of elimination, meso endemic area with mean pre- control stimulated value of CMFL 30mf/mg after the last treatment round should be 0.2mf/mg and that of hyper-endemic area with mean pre-control stimulated value of CMFL of about 70mf/mg after the last treatment round should be 0.1mf/mg. In Ghana, the ultimate intervention goal is aimed at reducing CMFL below 0.05mf/mg by 2015 in order to completely eliminate this infection. This present study indicated an overall CMFL in the 20 communities to be 2.3mf/mg. One of the communities, Bodiewu, recorded CMFL of 5.2mf/mg a threshold above APOC recommended CMFL value for the infection in a community to be considered a public health problem. None of the communities recorded CMFL below the threshold such that when intervention is stopped, the parasite populations can be believed to be permanently moving to its demise. Even though using CMFL alone as an indicator suggested that the burden of *O. volvulus* infection as a public health may have declined, considering the IVM MDA treatment period and the pre-control endemicity, the situation is alarming and critical attention has to be paid to check the infection in this area. A study conducted by Osei Atweneboana *et al*., (2007) in some savana areas of Ghana with annual ivermectin mass drug treatment of 6-18 years recorded CMFL ranges between

0.06mf/mg and 2.85mf/mg which was lower than what was recorded in this present study and Osei Atweneboana *et al*., (2007) concluded possible emergence of resistant parasite populations, which were not responding as anticipated to ivermectin treatment.

It was also observed from this present study that as the number of onchocerca palpable nodules increased, there was accompanied increase in the percentage of volunteers with microfilariae (Table 4.9). These nodules contained adult worms which produce millions of mf (Little *et al*., 2004b). According to Osei-Atweneboana and colleagues (2007), ivermectin treatments have minimal effect on the adult worms. Therefore, to completely eliminate this infection, macrofilaricidal drugs should be developed to complement the use of ivermectin (Debrah *et al*., 2006).

5.4. Impact of Ivermectin Mass Drug Administration on Onchocerciasis Infection

Onchocerciasis elimination is defined as the reduction of local onchocerca infection and transmission to low or declining levels such that transmission can no longer sustain itself and treatment can safely be stopped without risk of recrudescence of infection and transmission (APOC, 2010). To determine whether elimination of infection has been achieved, low or declining levels of epidemiological indices such as nodule prevalence, mf prevalence and CMFL are required (GHS, 2008). Many years of ivermectin treatment is also an important requirement (Walsh *et al*., 1978; Diawara *et al*., 2009).

Findings from this present study showed little impact of ivermectin mass drug administration on onchocerciasis despite prolonged (10 years) mass treatment interventions. Ivermectin is an antiparasitic agent that is currently used for treating and controlling onchocerciasis (Fisher and Mrozik, 1989; Steel, 1993). The drug is able to reduce the winding and coiling motility of microfilaria even after the first dose (Soboslay *et al*., 1987). Aside the immediate action of ivermectin on microfilaria, the drug also suppresses parasite transmission (OseiAtweneboana *et al*., 2007). The foremost outcome of ivermectin treatment on onchocerciasis is its prolonged inhibition of microfilariae production by the female adult worms resulting in no or negligible amounts of skin microfilaria repopulation after treatment for 6 to 9 months (Duke *et al*., 1992;

Chavasse *et al*., 1992; Plaisier *et al*., 1995). In addition, studies have shown a rise in the inhibition of reproduction by the adult worm with increasing rounds of ivermectin treatment (Boatin *et al*., 1998; Osei-Atweneboana *et al*., 2007). A study by Basanez *et al*., (2008) reported a 99% efficacy of microfilaricidal effect after each IVM dose and a reduction in mf production by fertile worms. Antwi-Berko (2014), demonstrated from his study in some onchocerca endemic parts of Ashanti region of Ghana that after a single IVM treatment round, there was drastic decline in mf density (even after 8 months of last intake) as compared to volunteers who had never taken IVM before. The author also observed complete microfilariae clearance in all volunteers with 6 to 9 rounds of ivermectin intake from his study. This present study was done after one year of ivermectin last intake and has shown that with repeated rounds of IVM treatment, the drug remains a potent microfilaricide because generally, as the number of IVM intake increased, the percentage of volunteers with no mf increased and intensity decreased (Figure 4.4 and Table 4.10). However, there was a higher mf density observed after one round of IVM compared to ivermectin naïve volunteers. Moreover, volunteers with 6 rounds of IVM and beyond had individuals with microfilariae, an observation which was not in conformity with the findings by Antwi-Berko, (2014). One major reason that could be assigned to this finding is possible early repopulation of skin with microfilariae from *O. volvulus* adult populations which might become insusceptible to ivermectin treatment (Osei-Atweneboana *et al*., 2007).

This study showed that about 11.7% have never taken ivermectin before (non-compliance) despite 10 years of IVM MDA in the study area (Table 4.11). IVM compliance study done in Nigeria and Cameroon reported 6% non-compliance despite 8 rounds of ivermectin MDA (Brieger *et al*., 2011). These non-compliant individuals remain an essential group for retransmission of this infection. Some of the reasons that can be attributed to the noncompliance recorded from this

study include lack of understanding of the importance of the IVM treatment. The drug is distributed by Community Directed Distributers (CDD) often referred to as 'Organizers'. They are mostly non-health workers with minimal or no training. As a result, they are not able to explain to the local community members the reason and importance of participating in the IVM mass drug treatment. Also, due to the adverse effects (AEs) of IVM that some people develop after the first drug intake with minimal or no intervention, they often stay away from taking subsequent rounds of IVM treatment because of fear of developing AEs again. This may discourage others who need to be enrolled into the IVM treatment programme in the communities for their first time. Another most important reason is absence of CDD in the smaller communities such that they have to depend on the bigger communities for their drug supply limiting the accessibility of the drugs to inhabitants in smaller communities.

The CMFL of 2.3mf/mg found in this study is higher than the national target of 0.05mf/mg for elimination of the disease. Therefore the claim made by the DHD that onchocerciais infections have been eliminated from the district is far from the reality and cannot be sustained. . In order to achieve the national target of a CMFL below 0.05mf/mg, CDTI mass treatment activities should not cease.

CHAPTER SIX

CONCLUSION AND RECOMMENDATION

6.1: Conclusion

This study in 20 onchocherciasis-endemic communities in Aowin district in the Western Region of Ghana concluded that 10 years of annual ivermectin mass treatment of onchocerciasis might have reduced the infection below public health importance but the infection has not been eliminated. The epidemiological indices (nodule prevalence,

microfilaria prevalence and CMFL) established in this study suggest an on-going transmission and as a consequence danger of infection increasing if interventions are discontinued. . Since elimination and transmission interruption of the infection in Africa have become the goal, it is important that focus should be placed on new innovative and flexible approaches to achieve this goal.

6.2: Recommendation

It is therefore recommended that:

i. If transmission interruption is a goal in Ghana, annual or bi-annual treatments with ivermectin must continue, or a new strategy that aimed at complete elimination should be implemented to avoid onchocerciasis recrudescence in the future.

ii. Since reports from some parts of Ghana suggested that adult female *O. volvulus* worms are becoming resistant to ivermectin treatment, resuming mf reproduction more rapidly after IVM treatment than would normally be anticipated, further studies that evaluate the susceptibility of parasites should be undertaken in these communities as well.

iii. MDA with ivermectin should be complemented with macrofilaricidal drugs so that the reservior of the infection can be eliminated

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