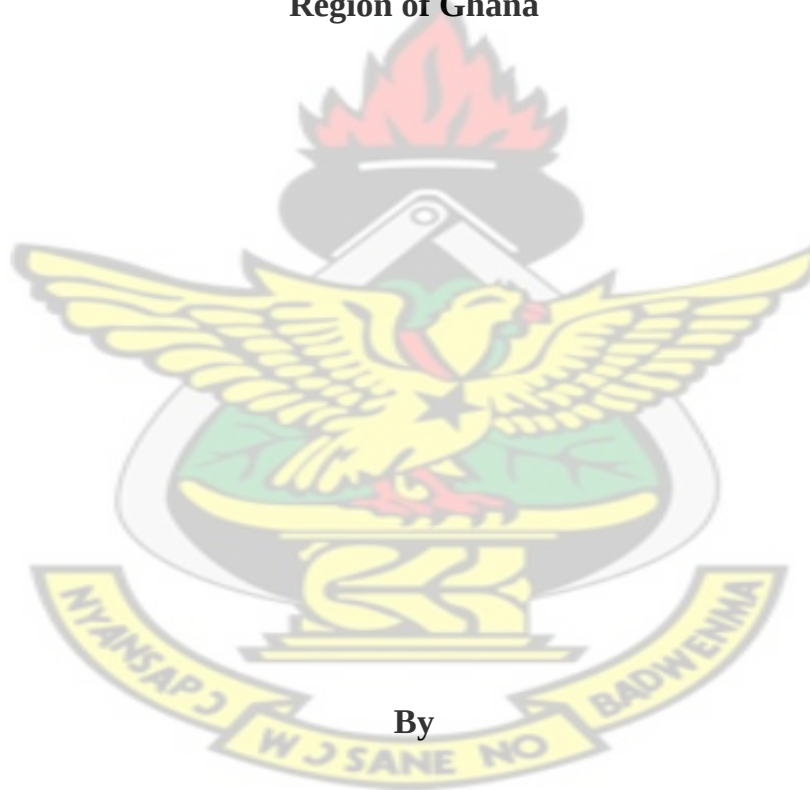


KWAME NKRUMAH UNIVERSITY OF SCIENCE AND TECHNOLOGY
COLLEGE OF HEALTH AND ALLIED SCIENCES
DEPARTMENT OF CLINICAL MICROBIOLOGY

**Studies of Entomological Parameters and Perception of Malaria Transmission on
the Kwame Nkrumah University of Science and Technology campus, in the Ashanti
Region of Ghana**



By

SYLVESTER COLEMAN

May 2009

**KWAME NKRUMAH UNIVERSITY OF SCIENCE AND TECHNOLOGY,
KUMASI, GHANA**

**Studies of Entomological Parameters and Perception on Malaria Transmission on
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Region of Ghana**

**A THESIS SUBMITTED FOR THE AWARD OF
A DEGREE OF MASTER OF SCIENCE IN CLINICAL MICROBIOLOGY**

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DECLARATION

This is to declare that this thesis is the candidate's own account of his research.

This thesis was supervised and approved for submission by:

.....
Signature of Supervisor

.....
Signature of Head of Department

.....
Signature of Student



DEDICATION

This work is dedicated to the memory of my late Grandparents,
Mr. and Mrs. Sylvester Benjamin Eshun and to my family for their love and care.

KNUST



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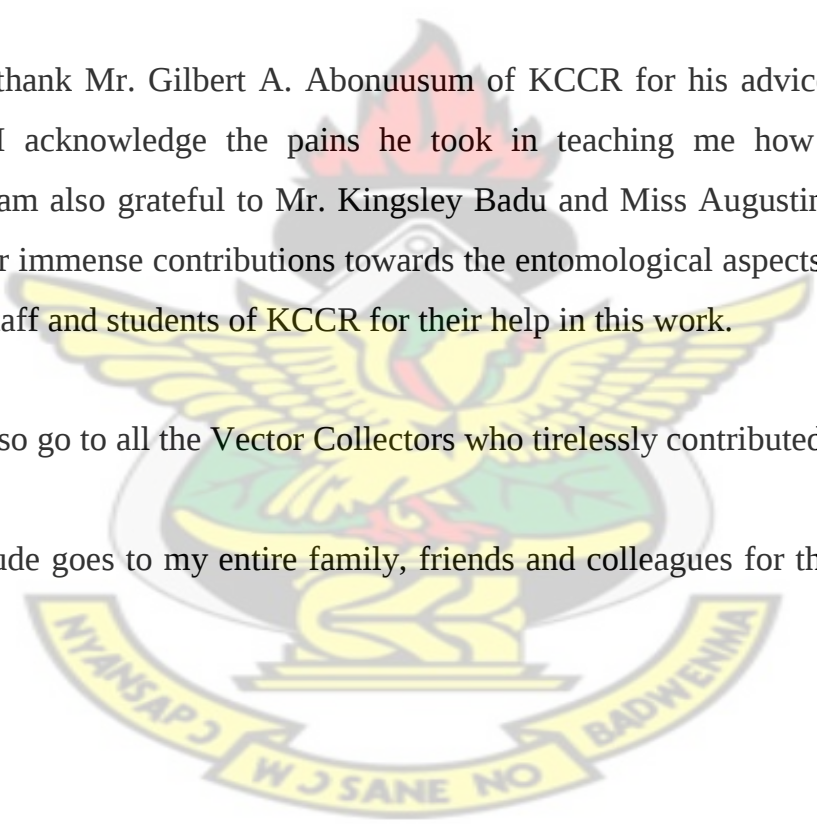
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ABBREVIATIONS

ARDS	Acute Respiratory Distress Syndrome
ACT	Artemisinin-based Combination Therapies
AGA	AngloGold Ashanti
CDC	Centers for Disease Control and Prevention
CSP	Circumsporozoite Protein
EIR	Entomological Inoculation Rate
ELISA	Enzyme-Linked Immunosorbent Assay
GBC	Global Business Coalition on HIV/AIDS, TB and Malaria
GDP	Gross Domestic Product
GHS	Ghana Health Service
GIS	Geographical Information Systems
GPI	Glucose Phosphate Isomerase
GPS	Global Positioning Systems
GSS	Ghana Statistical Service
ITN	Insecticide-Treated Net
IVM	Integrated Vector Management
KAP	Knowledge Attitudes and Practices
PBS	Phosphate Buffer Solution
KCCR	Kumasi Centre for Collaborative Research in Tropical Medicine
KNUST	Kwame Nkrumah University of Science and Technology
MARA	Mapping Malaria Risk in Africa
MBR	Man Biting Rates
MOH	Ministry of Health Government of Ghana
MSD	Meteorological Services Department
PCR	Polymerase Chain Reaction
RBM	Roll Back Malaria
UNICEF	United Nations Children's Fund
WHO	World Health Organization

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ABSTRACT

Malaria is a major public health problem in Ghana. Estimations are that 3.5 million people contract malaria every year. In order to develop effective control interventions targeted at reducing the malaria burden in any setting, it is important to understand the major factors that affect transmission and sustenance of the disease. The study was conducted on the Kwame Nkrumah University of Science and Technology (KNUST) campus to determine the vector species present and their roles in malaria transmission, map out areas of high malaria risk using GIS, and seek the perception of inhabitants of the KNUST campus on malaria.

Monthly mosquito sampling surveys revealed the existence of varied larval habitat types within the area, mostly characterised by clear or turbid shallow and sunlit conditions, with the greatest larval density contributed, mainly by vegetable gardens and irrigated farmlands. *A. gambiae* Giles complex, *A. funestus* Giles complex and *A. zieamanni* Grunberg were the three *Anopheles* species that were identified from 843 *Anopheles* spp that were caught from all night human landing collection during the survey. Of the 3 species *A. gambiae* proved to be the main species, with a sporozoite index of 1.01% and 0.57% for the dry and rainy seasons respectively. An average entomological inoculation rate (EIR) of 0.059 infective b/m/n and an annual EIR of 22 ib/m/yr were estimated. The faculty area was identified as area with the highest malaria risk, with respect to entomological parameters that were measured during the period. A KAP survey revealed that respondents had high malaria knowledge with 97.4% of respondents relating malaria to mosquito bites. Some respondents on the other hand also thought that eating too much oil and long exposure to sunshine caused malaria. However, high knowledge of malaria did not necessarily result in correct attitudes and practices.

This study reveals that though malaria transmission appeared low on the KNUST campus, misconceptions of some inhabitants on malaria transmission coupled with the high abundance of *Anopheles gambiae* Giles complex, on the KNUST campus could result in high levels of transmission if an infectious gametocyte pool comes into play. The study also provides a GIS based malaria information, which needs to be considered and integrated into the design and implementation of future malaria control interventions on the KNUST campus and its immediate surroundings. Educational programs aimed at increasing awareness on the correct attitudes and practices towards malaria transmission could promote community participation for effective malaria control in the study area.

CHAPTER ONE

1.0 INTRODUCTION

1.1.0 Background

Each year between 300 and 500 million cases of malaria are diagnosed worldwide, with 1.5 to 2.7 million deaths a year (WHO, 2002). Estimations are that Forty percent of the world's population (2.4 billion people) is exposed to the infection, especially people who live in tropical and subtropical countries. The World Health Organization (WHO) estimates that 60% of the cases of clinical malaria and over 80% of the deaths occur in Africa south of the Sahara. Of the more than 1 million Africans who die from malaria each year, most are children under 5 years of age (WHO/UNICEF, 2003). A report by WHO in 2006 revealed that malaria kills an African child every 30 seconds and most of these children who survive an episode of severe malaria may suffer from learning impairments or brain damage (WHO, 2006b). In 2007, UNICEF estimated that approximately 20,000 children died from malaria that year (25 per cent of the deaths of children under the age of five) (UNICEF, 2007).

The distribution of malaria in Ghana follows ecological zones, incidence being highest in the forest zone, followed by the coastal zone and then the northern savannah (Afari *et al.*, 1992). The National Malaria Control Programme of Ghana annual report in 2005 revealed that, malaria was responsible for up to 40% of daily out-patient consultations at hospitals and clinics and over 23% of deaths in children under five years of age. The report further indicated that it was also responsible for 38,000 deaths per annum and over 2,000 deaths in pregnant women, accounting for 9% of all deaths in pregnancy (NMCP, 2005). The world malaria report for 2008, indicates that Ghana had an estimated 7.2 million malaria cases in 2006, 3% of the total for the WHO African Region. There was no evidence of a reduction in malaria cases between 2001 and 2007, and reported deaths also increased in 2007 (WHO, 2008).

The Ashanti region, which falls within the forest zones of Ghana in the year 2000, was reported as the region with the greatest number of reported cases of malaria and the highest prevalence of malarial parasitaemia (Browne *et al*, 2000). In Obuasi for instance, malaria remains an important public health issue with parasitaemia prevalence of 68% in the human populace (Amoyaw, 2008). Studies conducted for the AngloGold Ashanti (AGA) Obuasi Mines in 2005, revealed that between 6,000 and 7,000 cases were diagnosed monthly among employees, contractors and employee dependants. In January 2006 alone, 11,800 malaria cases were reported in the Obuasi municipal district (GBC, 2008) and seven employees died as a result. Treatment costs for malaria at the AGA's Edwin Cade Hospital were estimated at US\$ 55,000 per month with an average of 7,400 shifts per month being lost in 2005 to malaria at the Obuasi Mine (GBC, 2008). In 2007, Obuasi municipality recorded 69,083 at all health facilities within the municipality; an average of 5,757 malaria cases per month (Amoyaw, 2008).

A survey conducted in Kumasi between April and May 2005, found *Plasmodium falciparum* prevalence rate of 37.8% among 296 children in 184 households from Moshie Zongo a suburb of Kumasi (Ronald *et al.*, 2006). According to the Ghana health services, as many as 15,999 residents in the Kumasi Metropolis suffered from uncomplicated malaria between 2005 and 2007, and were admitted to various public hospitals in the Kumasi metropolis; as compared to 3,858 residents who were also admitted during the same period due to cholera, diarrhoea and dysentery. Uncomplicated malaria accounted for 2,373 total admissions in 2005 (GHS, 2008). However the number of patients admitted increased to 6,175 in 2006, then to 7,451 in 2007 (GHS, 2008).

The socioeconomic impact of malaria in countries with intense transmission averages 1.3% loss of annual economic growth, leading to substantial differences in Gross Domestic Product (GDP) between countries with and without malaria. Malaria has been estimated to cost Africa more than US\$ 12 billion every year in lost GDP even though it could be controlled for a fraction of that sum. Cost towards control was close to US\$ 60

million in 2006 and US\$ 40 million in 2007 (WHO, 2008). The cost of malaria to countries with malaria manifests either directly or indirectly. The direct cost of malaria is estimated to include a combination of personal and public expenditures on both prevention and treatment of the disease. In malaria endemic countries as in Sub-Saharan Africa, the disease may account for as much as 40% of public health expenditure, 30-50% of inpatient admissions and up to 60% of outpatient visits (WHO, 2007).

The burden of malaria in Ghana in 2002 obtained through the cost of illness approach was estimated at US\$2.63 per capita or US\$13.51 per household and in 2003 the cost of work days lost to malaria was estimated as US\$8.4 (Asante and Asenso-Okyere, 2003). The permanent neurological and physical damages caused by severe episodes of the disease hamper children's schooling and their general well-being. This can directly affect their education and ability to learn in later life. Absenteeism from work also impacts on household income. Both direct and indirect costs associated with a malaria episode represent a substantial burden on the poorer households (European Alliance Against Malaria, 2007). A study in northern Ghana suggested that, while the cost of malaria care was just 1% of the income of the rich, it was 34% of the income of poor households (Akazili, 2002).

There are several reasons why sub-Saharan Africa suffers an overwhelming proportion of the malaria burden. Some of these reasons include; the occurrence of the most severe and life-threatening form of the disease and the presence of the most efficient vector in this region, drug resistance (Trape, 2001), more frequent exposure of less-immuned populations (pregnant women), climate and environmental change (Mouchet *et al*, 1998), changes in land-use patterns, and reductions in funding and manpower dedicated to control activities (Brêtas, 1996) Amongst these factors also is the emergence of HIV/AIDS; since HIV increases the risk of infection with malaria and decreases response to standard antimalarial treatment (WHO, 2005). The impact of malaria also takes its toll on the poorest – those least able to afford preventive measures and medical

treatment. The only parts of Africa that have significantly reduced malaria are the northern and southern most parts of Africa, home to the richest countries on the continent (Gallup and Sachs, 2001).

The lack of visual representation that seem to provide visual information on the, regional perspective of distribution, transmission intensity, seasonality, environmental determinants and populations at risk; is also seen as a contributing factor. Such information has been reported to have implications for the planning, targeting and implementation of control activities at continental, national and regional levels (MARA, 1998).

Although often considered a predominantly rural disease, in Africa malaria represents a leading cause of morbidity and mortality among many urban African populations (Donnelly *et al*, 2005). Keiser *et al.*, (2004) estimated that, 200 million urban residents were at risk of malaria, suggesting the need to identify risk factors for malaria in urban settings and subsequent control interventions. However it is well documented that malaria transmission is generally lower in urban compared with rural areas in Africa (Klinkenberg *et al*, 2005). This is largely attributed to such factors as lower vector density, higher human density, better quality housing and improved drainage in urban areas (Hay *et al*, 2005). Other research on malaria transmission in Africa also reveals that urban populations are better able to access healthcare facilities and consequently suffer lower malaria morbidity and mortality (Hay *et al*, 2005); and also reveals that malaria risk is unevenly distributed across urban environments (Klinkenberg *et al*, 2006). For this reason a better understanding of the individual, environmental, and community-based determinants and malariometric indices within specific communities are needed for formulation of control intervention (Donnelly *et al*, 2005 and Keiser *et al*, 2004).

A commonly used index of malaria transmission intensity is the Entomological Inoculation Rate (EIR) (WHO, 1975 and Beier *et al.*, 1999). The Entomological Inoculation Rate (EIR), is the product of the human biting rate (mosquito bites per person

per night) and the *Plasmodium falciparum* sporozoite rate (PfPR: the proportion of mosquitoes carrying the infectious sporozoite stage in their salivary glands) (Macdonald, 1957 and Smith *et al.*, 2005). MARA (1998) also confirmed that the distribution, transmission intensity and clinical consequences of malaria in Africa, vary greatly across the continent as well as within countries. Such variability could affect most quantitative aspects of malaria epidemiology on the African continent (Snow *et al.*, 2002), and could also result in duplication of control efforts. It is therefore necessary to document and aid the activities of control teams. Mapping the precise spatial extent of malaria transmission is seen by the World health Organization (WHO) as being central to control efforts within a particular community (Guerra *et al.*, 2006). Mapping offers an ideal way of documenting and displaying complex information in a way that will be understandable and intuitive, and also serves as an operational guide or model (Bettinger and Wing, 2004).

The best mapping tool that allows for modelling policy alternatives that also shows areas and regions that may be suitable and unsuitable for malaria transmission; and allows for a systematic and informed way of tackling malaria problems is the Geographic Information System (GIS) (Bettinger and Wing, 2004). Transmission parameters data gathered relating to anopheline (total mosquitoes captured per species, man biting rates (MBR), PfPR, and EIR) and some environmental parameters can be used to generate a geographical information system, which allows for establishment of an early warning system for control.

The global burden of malaria remains enormous, though access to malaria control interventions, especially Bed nets in Africa, increased sharply between 2004 and 2006. Malaria cases reported in 2006 was 247 million, and young children remain by far the most likely to die of the disease (WHO, 2008). For example; malaria cases recorded in Ghana in 2004 increased from 2,790,349 cases to 3,921,200 cases in 2005. Deaths due to malaria also increased from 2,688 in 2004 to 2,718 in 2005 (WHO, 2006b). These

findings suggest there is still much more to be done to control the malaria situation both locally and globally.

A multi-pronged approach to malaria control is seen as the effective tool in causing a significant impact on the disease (Eleanor, 2000). One such approach is the Roll Back Malaria programme of the World Health Organization, which is an approach to use whatever combination of available tools that best suits the control of malaria in a specific location rather than relying on one or two tools and applying them everywhere (Nabarro and Tayler 1998). The goal of the Roll back Malaria (RBM) Programme is to reduce malaria morbidity and mortality by 50% by 2010 through improved prevention, better access to care (i.e., early detection and rapid treatment of cases), higher quality and efficient service delivery, and increased partnership in the context of overall sector-wide development (WHO, 2000). Governments' over-reliance on curative services and their resulting problem of increasing spread of multi-drug resistant malaria consequently highlighted the importance of transmission reduction through vector control (USAID, 2007). Consequently the use of ITNs and ITMs increased since 2000, but its success in reducing malaria has varied widely. Integrated Vector Management (IVM), IVM then emerged, as a widely supported malaria control strategy (USAID, 2007). IVM is a decision-making process for the management of vector populations, so as to reduce or interrupt transmission of vector-borne diseases. A key component of its characteristic features is the selection of methods based on knowledge of local vector biology, disease transmission and morbidity (WHO, 2008). It is in the light of this that this study was conducted to set the stage for relevant, appropriate and cost effective intervention based on evidence.

1.2.0 Justification

Malaria risk is often associated with forest areas (WHO, 2006a). The situation in Ghana is no different; Brown *et al.* (2000) recorded an overall prevalence of malaria parasitaemia of 50.72% in children >2years for the forest zone of Ghana. The Kwame Nkrumah University of Science and Technology (KNUST) falls within this zone. The bio-ecological characteristics of the KNUST and increased land modifications within the area in recent times have the tendency to promote mosquito abundance for effective malaria transmission on the KNUST campus. However there is no information on vector bionomics and transmission dynamics. It is worth emphasizing that, the complex mix of people (including foreign students and other migrants settlers around campus) who make up the KNUST population, also serves as a likely malaria threat to life on the KNUST; since migration has been tagged as a probable precipitating cause of the most serious malaria problems in Africa (WHO, 1996).

Knowing the debilitating effect of human malaria and its threat especially to life on the KNUST campus, Professor Kwesi Andam, the former Vice-Chancellor of KNUST initiated a multi-disciplinary task force to control malaria and mosquito nuisance on the KNUST campus and its surroundings in 2003.

Information on the epidemiology of malaria is essential if the disease is to be controlled. Entomological, parasitological and clinical studies provide useful information on the characteristics of malaria transmission in an area as well as the habits and habitats of the specific vector species (WHO/CDS/CPE/SMT, 2002).

Malaria transmission studies conducted on the KNUST have over the years focused only on the parasitological and clinical indices of the disease. To date no entomological study on malaria transmission on the KNUST campus has been carried out, despite its importance in the characterisation of local malaria. Information from entomological studies, such as the types of vectors and the role these vectors play in the transmission of

infections is of essential importance in the determination of their public health importance and subsequent planning of any control programmes. It has been proposed that the important findings of such research allows researchers to gain knowledge of the local entomology so that subsequently the most appropriate vector control methods for each situation can be utilized, according to entomological and epidemiological criteria (Coura *et. al.*, 2006).

It is the first study to be carried out on vector bionomics and malaria transmission on the KNUST campus and its surroundings. It focuses on the entomological parameters of malaria transmission, exploits the application of geographical information systems (GIS) technology in malaria transmission studies, and seeks to know of the beliefs, knowledge, attitudes, and practices of the KNUST populations regarding malaria transmission. The inclusion of the knowledge, attitude and practices questionnaires, is to ascertain levels at which the community could be involved in control activities, that may be implemented and also to help deduce possible vulnerable groups and zones.

Proponents of Global Malaria Control Strategy (WHO, 1993) recognizing that the malaria problem varies enormously from epidemiological, ecological, social and operational view-points, recommended identification of easily recognizable eco-epidemiological types, namely malaria paradigms for local analysis for formulation of eco-friendly, cost-effective and sustainable control strategies. It is in this context that the GIS component was introduced to allow for an easy description of the malaria paradigm at the micro-level and the identification of eco-epidemiological characteristics that may favour local malaria transmission. The GIS component exploits data mining tools, which can prioritize the malaria risk zones for proper management of the disease.

The result of this work should serve as a comprehensive reference on malaria exposure on the KNUST campus and represent an important baseline data essential for implementing future malaria control programmes.

1.3.0 Objective

The main objective is:

To determine the vector species present on KNUST campus, their roles in malaria transmission, areas of high malaria risk, and perceptions of residents on malaria on the KNUST campus.

The specific objectives are;

- (i) To identify and map out vector breeding sites
- (ii) To identify *Anopheles* species on KNUST
- (iii) To determine Man Biting Rate,
- (iv) To determine Sporozoite Index of vectors caught on KNUST campus
- (v) To determine the Entomological Inoculation Rate (EIR) of vectors caught on KNUST campus
- (vi) To determine the perceptions of the KNUST community on malaria transmissions through knowledge, attitude and practice questionnaires
- (vii) To develop a malaria risk map of KNUST that reflects the spatial variability of the transmission using geographical data obtained from the study.

CHAPTER TWO

2.0 LITERATURE REVIEW

2.1.0 Entomological Studies

The role of the *Anopheles* vector in the malaria cycle makes it impossible to exempt entomology in any trustable analysis of the malaria situation in a given area and a sound programme of malaria control (Esposito and Habluetzel, 1997).

The importance of understanding malariological entomology, especially vector biology and behaviour prior to initiating control measures is evident when one considers the key questions that must be answered to determine which type of control measure is best for a given situation or area (Bloland and Williams, 2002). An important parameter required is the identification of the mosquito species responsible for most malaria transmission within an area. Identification of the mosquito species however, involves surveillance, and collection to determine *Anopheles* density and species, from various parts of the affected area (Bloland and Williams, 2002).

Larval and adult mosquito surveys should be the base of all control programs and are required to be conducted on a regular basis to determine the extent, type, and concentration of mosquito populations within a study area. The surveys are essential to the coordination and success of all control measures (Holub and Geery, 2007). Larval samples are the most basic, whereby potential mosquito breeding sources are inspected regularly within a 14 day period. Larval samples are taken from sites found breeding, and are identified by the laboratory staff to species. All potential sources are marked on permanent maps and numbered for reference. Individual source histories are maintained on computerized records (Holub and Geery, 2007).

2.1.1 Studies on the Mosquito Larvae

The larval aquatic habitat is an important part of the mosquito life cycle and strongly influences the distribution and abundance of malaria vectors. Although a range of mosquito habitats exist, the larval stage is mainly confined to stagnant water pools (Pfaehler *et al*, 2006). The proximity of houses to locations with suitable topography for mosquito breeding may be an important determinant of malaria risk (Carter *et al*, 2000). *Anopheles* breeding sites may occur where water collects and forms a pool for a period of time sufficient to permit larval development and adult emergence (Minakawa *et al.*, 2005). It is believed that topography has an effect on breeding selection, since small temporary pools and larger more permanent aquatic habitats are more likely to exist in flat, relatively low-lying regions (Carter *et al*, 2000).

Variable characteristics of *Anopheles* larval habitats have been identified; a common characteristic is a shallow larval habitat with the presence of algae, although such a correlation is not systematic (Gimnig *et al.* 2002, Oo *et al.*, 2002). Other habitats identified consist of animal hoof or human foot prints, or small ponds of still temporary water created by irrigation projects or rainfall. However the characteristics of the larval habitat that are adequate for a given species are still unclear (Pfaehler *et al*, 2006). Some malaria epidemiologists have proposed that environmental variables have a direct or indirect effect on anopheline oviposition (Sumba *et al.* 2004, Rejmànková *et al.* 2005), affects larval distribution, density, and development (Gimnig *et al.* 2001) and adult fitness (Briegel 2003). These contributions and influence of these variables on anopheline larval distribution and habitat types have been established in several studies on anopheline larvae. One of such numerous studies is that conducted by Pfaehler and others in Mbita, Western Kenya, to investigate how properties of the soil substrate of *Anopheles gambiae* breeding pools could influence development of this mosquito species. In this study *An. gambiae* eggs from an established colony were dispensed into experimental plastic troughs containing soil samples from a range of natural *Anopheles* larval habitats and filtered Lake Victoria water. The duration of larval development (8-15

days), pupation rate (0-79%), and adult body size (20.28-26.91 mm³) varied among different soil types. The total organic matter (3.61-21.25%), organic carbon (0.63-7.18%), and total nitrogen (0.06-0.58%) levels of the soils were positively correlated with pupation rate and negatively correlated with development time and adult body size. The study underscores the importance of larval ecology as an important aspect of the population dynamics of anopheline mosquitoes and proposes that adult fitness may be correlated with the nutritional conditions under which larvae develop.

In another study conducted by Matthys and others in 2006 on urban agricultural land use and characterization of mosquito larval habitats in a medium-sized town of Côte d'Ivoire to assess risk factors for productive *Anopheles* breeding sites. Typical *Anopheles* larval habitats were characterized by the presence of algae, and the absence of floating vegetation. The highest *Anopheles* larval productivity was observed in rice paddies, agricultural trenches between vegetable patches, and irrigation wells. The study established an indirect link between the occurrence of productive *Anopheles* breeding sites and agricultural land use through specific man-made habitats, in particular agricultural trenches, irrigation wells, and rice paddies.

A study on regulatory factors affecting larval mosquito populations in container and pool habitats by Washburn (1995) revealed that physical and biological features have significant implications for successful implementation of biological control agents. Knowledge gained from larval studies has important implications in developing control strategies.

2.1.2 Studies on the Adult mosquito

One of the most important epidemiological parameters for the assessment of malaria transmission is the prevalence, in a given sample, of female anopheline mosquitoes with salivary glands infected with malarial parasites, a measure usually called the 'sporozoite rate' (Mboera and Magesa, 2001). The understanding of the entomological aspects of the epidemiology of malaria depends, in part, on the accurate assessment of this rate, which

is generally estimated by salivary-gland dissection or by an Enzyme Linked Immunosorbent Assay ELISA for circumsporozoite protein (Beier *et al.*, 1999). It is usually measured using the anophelines caught on an individual who acts as bait. Coupled with the man-biting rate (the number of vector mosquitoes coming to feed each night), the sporozoite rate can be used to calculate the Entomological Inoculation Rate (EIR) which is the number of infective bites that an individual receives during a determined period of time (Cano *et al.*, 2006).

The criteria previously used to classify the malaria transmission levels were based on parasitological and clinical data, splenic index and prevalence of the parasitaemia (Gilles *et al.*, 1993). Today, the entomological inoculation rate (EIR) is considered a key factor when establishing the degree of endemicity or transmission level (Beier *et al.*, 1999).

2.1.3 Studies on Malaria Transmission

Several research on malaria in Africa seek an understanding of the relationships between malaria transmission by vector populations of mosquitoes and the outcome of measures of transmission in terms of malaria prevalence. Most of these studies explored malaria risk by seeking entomological evidence describing the average annual risk of receiving a *Plasmodium* infected bite from the local vector population (EIR).

One of such studies was conducted by Beier *et al.*, (1999) to assess entomological inoculation rates and *Plasmodium falciparum* malaria prevalence in Africa. This study confirmed EIR as a more direct measure of transmission intensity than the traditional measure of prevalence or hospital based measures of infection or disease incidence. The study evaluated data from 31 sites throughout Africa to ascertain basic relationships between annual EIRs and prevalence of *Plasmodium falciparum* malaria infection. Malaria transmission was found to be highly variable with annual ranges of <1 to >1000 infective bites per person per year, while the malaria prevalence rates ranged from 7.0%

to 94.5%. The majority of sites fitted a linear relationship ($r^2 = 0.71$) between malaria prevalence and the logarithm of the annual EIR. Some sites with EIRs < 5 infective bites per year had levels of *Plasmodium falciparum* exceeding 40%. There were no examples of sites that had prevalence rates < 50% when EIRs exceeded 15 infective bites per year. Annual EIRs of 200 or greater were consistently associated with prevalence rates >80%. As such, malaria field programs need to consider entomology as a major component of assessments of the efficacy of transmission control measures, since unacceptably high levels *P. falciparum* prevalence exist at very low levels of transmission by local vector populations.

EIR studies conducted by Killeen *et al.*, (2000) allowed for the prediction of malaria transmission intensity of the dominant malaria vector species at 3 malaria-endemic sites; Papua New Guinea, Tanzania and Nigeria. The study classified EIR as the product of the potential of individual vectors to transmit malaria, the vector emergence rate relative to human population size and the infectiousness of the human population to vectors. EIRs could be predicted using these measured parameters together with human biting rates and human reservoir infectiousness. This mathematical model EIRs ($\pm$ SD) that were 1.3 ± 0.37 (range 0.84 – 1.59) times those measured in the field. This model explains how effective malaria transmission is dependent on the vector, hence the need for measurement of entomological indices of transmission in epidemiological studies.

The relationship between variations of *P. falciparum* transmission and EIR in rural Gabon was assessed by Elissa *et al.*(2002) through entomological surveys carried out in Dienga and Benguia between May 1995 – April 1996 and May 1998 – April 1999. In Dienga, malaria transmission was found to be seasonal, with two transmission two (a major peak between December and March, and a minor peak between July and August) separated by two 3-month periods during which no transmission was detected. This phenomenon was related to the fact that malaria transmission in this zone was mostly by the *A. gambiae* s.l., none of which was collected during the dry season. An average

EIR of 0.28 infective bites/person/night was recorded. However, in Benguaia, transmission was identified to be perennial with seasonal fluctuations, and a mean EIR of 0.76 infective bites/person/night. Parasitological studies also carried out revealed that in each area, high biting densities preceded malaria attacks incidence with 1– or 2 – month time lag.

Wanji, *et al* (2003), reported on work carried out in the Mount Cameroon region on the biting habits, feeding behaviour and entomological inoculation rates (EIR) of malaria vectors, during two seasons of the year. The study revealed that out of a total of 2165 *Anopheles* species collected for both the rainy and dry season *A. funestus*, *A. nili* and *A. gambiae* s.s of the *A. gambiae* (Giles) complex were identified as the main malaria vector. The three vectors had the peak of their activities between 1am and 2 am. A human blood index of (HBI) of 98.29% was recorded for all fed *Anopheles*. The sporozoite rate, for all vectors together, was significantly higher in the rainy season (9.4%) than in the dry season (4.2%) with all species infected by *Plasmodium falciparum*. The average inoculation rate was 0.44 infective bites per person per night, which added up to 161 infective bites per year in the study area. Analyses of relative abundance and infection rates of malaria vectors at different sites situated along a transect of 20km during the dry season showed high heterogeneity in biting and sporozoite rates. Throughout this study the Mount Cameroon region was considered an area of high malaria transmission intensity.

Trung, *et al* (2004) studied malaria transmission and major malaria vectors in different geographical areas of Southeast Asia using similar entomological indices. The study was based on human landing collections conducted between April 1998 and November 2000 in six selected villages (four in Vietnam, one in Cambodia and one in Laos). The major *Anopheles* species identified were *Anopheles dirus*, *A. minimus* and *A. sundaicus*. In Cambodia and Vietnam *Anopheles dirus* played an important role in malaria transmission with a sporozoite rate of 2.8% for 480 man nights. In the same region *A. minimus*

showed a parous rate of 80% and a temporal and spatial variation in its role as a vector; suggesting that transmission occurred at anytime. In Mekong delta the study revealed that *A. sudaicus* occurred at high densities (up to 109 b/p/night) but none of the total number (11002) collected was positive for the circumsporozoite protein (CSP) and had a relatively low parous rate of 47% reflecting a low vectorial status even though they occurred in high numbers (109 b/p/night).

The distribution of malaria in Ghana follows ecological zones, incidence being highest in the forest zone, followed by the coastal zone and then the northern savannah (Afari *et al.*, 1992). Three species belonging to the *A. gambiae* complex - *A. gambiae* sensu stricto (s.s.) are distributed widely throughout the country, *A. melas* distributed along the coast, and *A. gambiae arabiensis* in the north (Appawu *et al.*, 1994); and the *A. funestus* (MOH, 2001) were identified in this study. All of these species are documented vectors of malaria transmission in Ghana (MOH, 2001).

An entomological study conducted by Afrane *et al* (2004) on the possible influence of urban irrigated agriculture on malaria transmission in the city of Kumasi, revealed that open space vegetable farms in Kumasi created and extended breeding sites for *Anopheles gambiae*. The major vectors identified were members of the *Anopheles gambiae* complex and *Anopheles funestus*. The study demonstrated seasonal variations in malaria risk with sporozoite rates being relatively lower (> 2.6 in the dry season and < 1.9 in the rainy season). Though the actual role played by irrigation agriculture activities could not be established, these agriculture activities contributed significantly to the occurrence of high densities of the vectors in the city of Kumasi. The occurrence of such high numbers was seen as a potential threat to human life in the city.

2.2.0 Geographical Information Systems

The geographic information system (GIS) and global positioning system (GPS) have been widely applied to health and epidemiology for malaria research and control in most sub-Saharan Africa countries (Hightower *et al.*, 1998; Hay *et al.*, 1998). Spatial point pattern analysis may help identify high-risk diseases areas, sources of diseases and high-risk populations (Gatrell and Bailey, 1996). These statistical techniques are based on case events and count data, where known geographic locations (x-y coordinates) of disease cases are commonly represented as points (Lawson and Denison, 2002)

Environmental factors such as topography, temperature, rainfall, land use, population movements, and degree of deforestation are believed to have a profound influence on the temporal and spatial distribution of malaria vectors and malaria (Brêtas, 1996). Despite its importance, the study of environmental determinants of malaria has been hampered by the difficulties related to collecting and analyzing environmental data over large areas, and to the speed of change in the malaria epidemiological situation (Brêtas, 1996). Remotely sensed data can also be employed to identify mosquito habitats and predict the likely range of vector (Hay *et al.*, 1998). The contribution of GIS to malaria research can be categorized as an operational planning aid for control programs; as a monitoring tool to assist with evaluation of control efforts; and as a research approach to investigate spatial associations of relevance to malaria epidemiology and control (Sweeney, 1998).

2.2.1 GIS as an Operational Planning Aid

GIS databases can be used as operational tools to support planning and implementation of control activities. For example, in insecticide treated bed net programs for community based malaria control, they can constitute simple and practical visual aids for detailed planning of bednet distribution and retreatment schedules. Similarly, in urban situations, where it is feasible to undertake larval control, purpose-generated large scale maps of larval habitats in relation to residential areas can guide the most efficient application of larvicides (Sweeney, 1998).

Large amounts of information are necessary for almost all aspects of malaria control programs. GIS offer the ability to process quantities of data beyond the capacities of manual systems. Data are stored in a structured digital format, which permits rapid retrieval and use. In addition, data may be quickly compiled into documents, using techniques such as automatic mapping and direct report printouts (Bernhardsen, 1992).

2.2.2 GIS as an Evaluation Tool

It is expected that changes in the global environment will lead to changes in malaria occurrence patterns. The occurrence of malaria under different land occupation strategies can be studied with the combination of remote sensing and GIS. The results could be used as guidelines for new development projects in areas receptive to malaria. Data on the distribution of vectors obtained from field surveys can be incorporated as GIS vector layers. Satellite imagery (Google earth, Landsat TM or Spot) reveals areas of environmental change which could be subjected to further investigation to analyse whether such factors might have an impact on malaria control efforts.

Thus, geographical clusters of malaria cases can assist with delineation of problem areas as a starting point for further analysis to identify possible reasons for the higher incidence of malaria in certain localities. This approach can therefore be of direct benefit to the evaluation of malaria control programs (Sweeny, 1998).

2.2.3 Use of GIS for Malaria Research

In Africa, extensive work on the use of GIS for malaria research has been done by Mapping Malaria Risk in Africa (MARA). MARA employs the use of GIS and other remote sensing techniques to establish a continental data base of the spatial distribution of the disease (MARA, 1998). To achieve this aim it relies on two complementary approaches: the first, involves the formation of a continental database of available malariometric data (EIR, infant parasite ratio etc.) of geographically positioned survey data; the second involves the use of environmentally determined models of continental limits of transmission risk. Malaria maps have been developed from these approaches

and have been used widely in Africa to provide fundamental resource for planners, donors and researchers.

In Australia GIS software was used to correlate the climatic attributes of the collection localities of mosquitoes with the presence or absence of the various species (Sweeney, 1997). In similar study that used a computer based GIS resource, it was possible to generate annual and seasonal estimates of key climate indicators (rainfall, relative humidity, maximum temperature and minimum temperature) for different collecting sites.

A retrospective surveillance study was conducted to examine the micro-geographic variation of malaria incidence in three malaria-endemic communities in the Northern Peruvian Amazon by Bautista and others from January to April in 1998–1999. The annual malaria risk rate (per 100) ranged from 38% to 47% for *Plasmodium vivax* and from 15% to 18% for *P. falciparum*. The study identified varying spatial clusters for *P. vivax* and for *P. falciparum*, and suggests a constant presence of high-risk areas (hotspots) for malaria infection in periods with high or low malaria incidence. Using GIS tools, the study suggested a modest targeted control efforts directed at identified high-risk areas may have significant impact on malaria transmission in the region (Bautista *et al.*, 2006).

The use of GIS as a malaria research tool is a worthy objective of research because it presents a wide spectrum of possibilities over which GIS can contribute to malaria control programs. Its application as an operational planning aid is an extension of geographical reconnaissance to promote better program management. Its use as an evaluation tool provides an additional means of spatially analysing outputs generated by health information systems in graphic visual formats which can be readily understood by field workers and program managers (Sweeney, 1998).

2.3.0 Knowledge, Attitudes And Practices About Malaria Transmission

Research findings have shown that environmental, behavioural and socio-economic factors are associated with ability to avoid mosquitoes and prevention of malaria attack (Macintyre *et al.*, 2002). Vector control programs are more effective with the involvement of the community, and prompt results are obtained from community-based programs compared with government-supported activities alone (Ruebush *et al.*, 1994). Klein *et al.*, (1995) identified that prior knowledge of the community beliefs and practices with respect to the disease in question is required to obtain and maintain its participation in surveillance and control activities. An understanding of local attitudes and beliefs is also important, since these can affect the acceptability of some interventions, particularly those that depend on changing human behaviour (Saiprasad and Banerjee 2003).

2.3.1 Studies of Knowledge Attitudes and Practices on Malaria Transmission

A cross-sectional survey was conducted by Rodríguez *et al.*, (2003), during May and June 1995 in Chiapas, Mexico to investigate the knowledge and beliefs about malaria transmission and practices for vector control. Questionnaires were used to investigate family structure, knowledge on malaria transmission, preventive measures and attitudes. It was established that malaria knowledge was poor and only 48% associated malaria with mosquito bites. Only 3% associated indoor residual spraying directly with the prevention of malaria transmission. The results also revealed that ninety nine percent of villagers had mosquito bednets, but 75.7% used them all year round. Other measures used by villagers to prevent mosquito bites were smoke and mosquito coils. Over 40% of villagers self-medicated when any member of the family had a fever episode, but 51% attended proper health services (community dispensary, private physician, health worker). It was concluded that educational programs aimed at increasing awareness on the participation of mosquitoes in malaria transmission and proper attitudes could promote community participation in malaria control in this region.

A cross-sectional study on malaria was carried out from 15 to 30 October 1999 firstly to assess the knowledge, perceptions and behaviour of married women in Sistan va Baluchestan province in the Islamic Republic of Iran about malaria and identify factors affecting these knowledge, perceptions and behaviour. The second aim was to describe the role of health providers in malaria education (Rakhshani *et al.*, 2003). A questionnaire about malaria knowledge, beliefs and practices was given to a random sample of 2168 married women from rural and urban areas of Sistan va Baluchestan, Islamic Republic of Iran. The mean knowledge score of subjects on malaria was identified to be low at 5.5 (maximum 15.0). Few respondents (37.6%) knew that malaria was an important disease in the area and only 58.4% knew that malaria was transmitted by mosquitoes. Most subjects (69.4%) never used a mosquito net. Only 49.9% of rural and 73.8% of urban residents would seek care for fever and chills from the local health centre. Community health workers were the main source of malaria information (29.5%) for rural women; the role of physicians in education was minimal. The results of this study helped to determine the educational needs of women and some of the associated factors in malaria prevention behaviour (Rakhshani *et al.*, 2003).

A baseline survey, was conducted between December 1999 and January 2000 by Okello-Ogojo, to investigate the level of knowledge, attitudes and practices about malaria and insecticide treated nets (ITNs) in four districts of Uganda (Mukono, Jinja, Mbarara and Arua). This study revealed that almost all respondents (96%) were troubled by mosquitoes and biting was cited by most (76.3%) as the main trouble, however there was limited knowledge and usage about nets treated with insecticide (14.1%) and also lack of the positive attitude towards bed-nets (Okello-Ogojo, 2001).

Oyewole and Ibidapo (2007) carried out a study in Ibadan the capital of Oyo state in southwestern Nigeria to investigate the basic factors responsible for human-mosquito interaction, attitudinal consequences of malaria treatment pattern and management strategies in an urban centre. This study identified late diagnosis, wrong medications,

incomplete doses, lack of knowledge about malaria episode and *Anopheles* mosquitoes as malaria vector, as some of the factors militating against prevention and proper management of the disease in this region. It also identified preventive measures adopted against mosquito bites included sleeping under net (treated and untreated) 17 (4.2%), door and window screening 37 (9.2%), cover cloth 55 (13.8%), mosquito repellent/insecticides spray 39 (9.8%). There was a significant difference between those that prevent malaria with chemoprophylaxis and other methods. Self-treatment (medication) accounted for 267 (66.8%) as against hospital treatment 93 (23.3%).

Onwujekwe *et al.*, (2008) investigated the link between socioeconomic status (SES) with differences in perceptions of ease of accessing and receiving treatment as well as with actual health seeking for treatment of malaria from different providers in a study towards equitable improvement of treatment-seeking in Enugu state, southeast Nigeria. Using structured questionnaires data was sought from 1,351 health providers in four malaria-endemic communities in Enugu state, southeast Nigeria. Data was collected on the peoples' perceptions of ease of accessibility and utilization of different providers of malaria treatment using a pre-tested questionnaire. A SES index was used to examine inequities in perceptions and health seeking. The results indicated that, patent medicine dealers (vendors) were the most perceived easily accessible providers, followed by private hospitals/clinics in two communities with full complement of healthcare providers: public hospital in the community with such a health provider and traditional healers in a community that is devoid of public healthcare facilities. This work identified that there were inequities in perception of accessibility and use of different providers, as well as in treatment-seeking for malaria. It was also established that the poor spent proportionally more to treat malaria. This study proposed that the differentials in perceptions of ease of access and use as well as health seeking for different malaria treatment providers among SES groups could be decreased by reducing barriers such as the cost of treatment by making health services accessible, available and at reduced cost for all groups.

Similar studies on knowledge attitude and practice towards malaria have been carried out in some communities in Ghana. One of such study was carried out in 13 communities in the Afram Plains District and 20 communities in the Asikuma-Odoben- Brakwa District of the Eastern and Central Regions of Ghana respectively (De La Cruz *et al.*, 2006). This study sought to identify factors and characteristics of women that affect bednet use among their children less than five years of age in Ghana. The study established that; region of residence; food security in the home (household); and caregivers' beliefs about symptoms, causation and groups most vulnerable to malaria, were factors most closely associated with bednet use. Most respondents knew mosquitoes caused malaria; however, 20.6% of doers (sleeping under a bednet) and 12.3% of non-doers ($p = .0228$) thought overworking oneself caused malaria. Ninety percent of doers and 77.0% of non-doers felt that sleeping under a net was protective against malaria ($p = .0040$). From their findings, the authors concluded that greater knowledge about malaria does not always translate into improved bednet use. The authors also recommended that, though culturally-based ideas about malaria may vary between communities, integrating them into traditional health education messages may enhance the effectiveness of public health efforts (De La Cruz *et al.*, 2006).



2.4.0 Epidemiology of Malaria

A familiarity with the technical aspects of malaria is necessary for decisions to be made about how to devise a locally appropriate malaria control strategy. The following section presents information on areas at risk of malaria transmission, the mechanics of how malaria is transmitted within a population, the range of clinical manifestations of malaria, and the mosquito vector.

2.4.1 Areas at Risk

Malaria occurs primarily in tropical and some subtropical regions of Africa, Central and South America, Asia, and Oceania (Figure 2.11). In areas where malaria occurs, there is tremendous variation in the intensity of transmission and risk of infection. For example, over 90 percent of clinical malaria infections and deaths occur in sub-Saharan Africa (WHO, 1996). However, even there (in sub-Saharan Africa) the risk varies widely. Highland (>1,500m above sea level) and arid areas (<1,000 mm rainfall/year) typically have less malaria, although these areas are prone to epidemic malaria if climactic conditions become favourable to mosquito development (WHO, 1996). Although urban areas have typically been at lower risk, explosive unplanned population growth has been a major factor in making urban or peri-urban transmission an increasing problem (Knudsen and Sloof, 1992).

Humans are commonly infected by four species of *Plasmodium* parasites: *Plasmodium falciparum*, *P. vivax*, *P. ovale*, and *P. malariae*. *P. falciparum*, the species most commonly associated with fatal malaria, is transmitted at some level in nearly all areas where malaria occurs. It accounts for over 90% of all malaria infections in sub-Saharan Africa, for nearly 100 % of infections in Haiti, and causes two-thirds or more of the malaria cases in Southeast Asia. *P. vivax* is a relatively uncommon infection in sub-Saharan but predominant in Central America and the Indian subcontinent. Some level of immunity is enjoyed by many ethnic groups, especially in West Africa; these people lack the “Duffy antigens”, which are required by the parasite to invade red blood cells (Miller *et al.*, 1976).

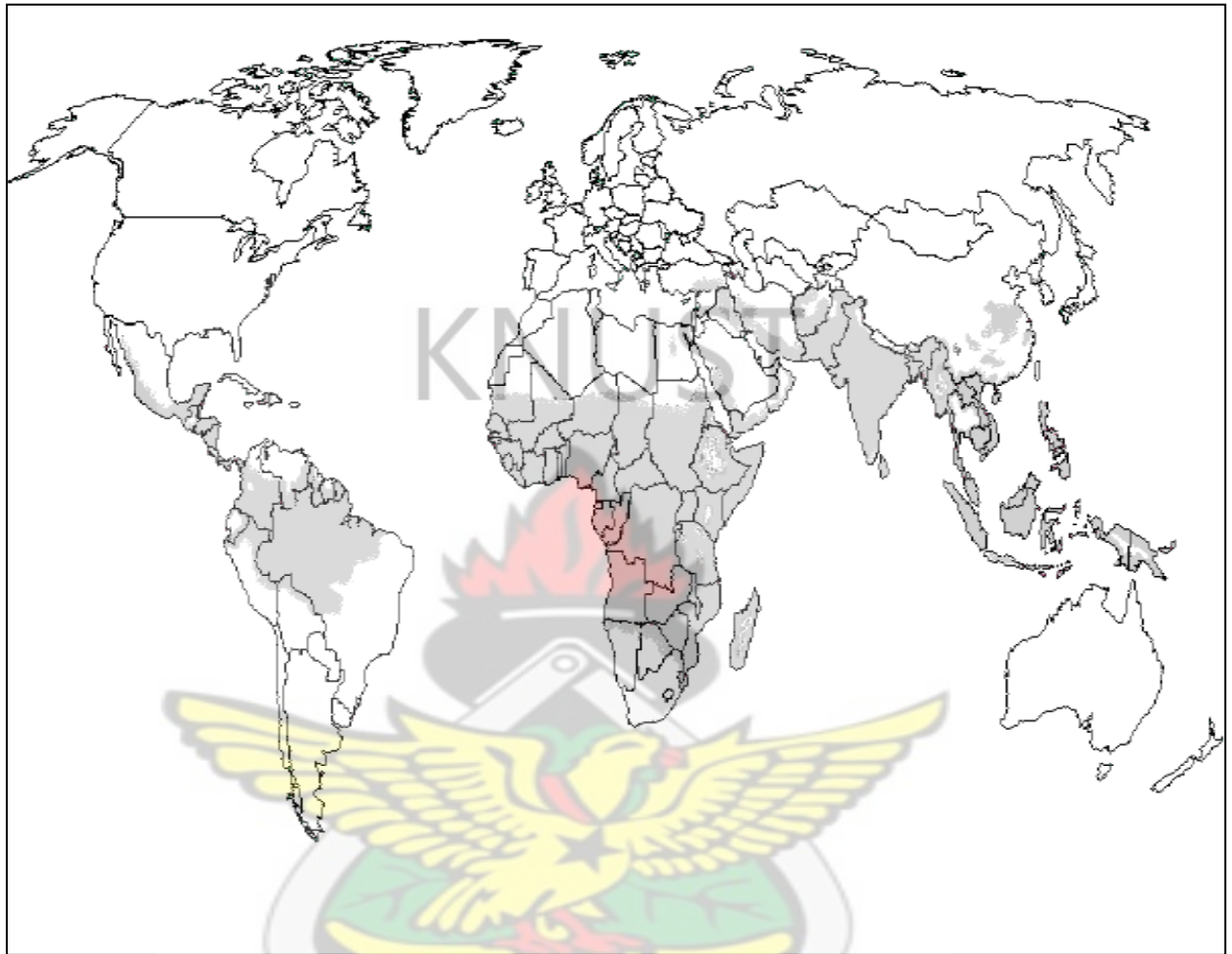


Figure 2.1 Malaria Distribution Map

Source: Adapted from World Health Organization (2002), and Centre for Disease Control and Prevention (2000).

2.4.2 Mechanisms Of Infection And Transmission

Malaria infection results from the bite of an infective female *Anopheles* mosquito. When an infected female *Anopheles* mosquito bites a human, she takes in blood. At the same time, she injects saliva that contains the infectious form of the parasite, the sporozoite, into a person's bloodstream (NIH,2002) The sporozoites rapidly (usually within one hour) enter parenchymal cells of the liver where the first stage of development in humans takes place (exo-erythrocytic phase of the life cycle, A from fig. 2.2). Subsequently, numerous asexual progeny, the merozoites, leave the liver cells and rupture into the blood stream, invading the erythrocytes. Parasites in the red blood cells multiply in a species-characteristic fashion, breaking out of their host cells synchronously. This is the erythrocytic cycle, with successive broods of merozoites appearing at 48-hour intervals (for *P. vivax*, *P. ovale* *P.* and *falciparum* infections) or every 72 hours (in *P. malariae* infections). For *P. vivax* and *P. falciparum*, this period is usually 10-15days, but it may lasts for weeks or months. The incubation period for *P. malariae* averages 28 days. In *P. falciparum* infections there is no return of merozoites from the blood into the liver cells. The other three species continue to multiply in the liver cells long enough after the initial bloodstream invasion, or there may be delayed multiplication in the liver. These exo-erythrocytic cycles coexist with erythrocytic cycles and in *P. vivax* and *P. ovale*, may persist as non-growing resting forms (hypnozoites) after the parasite has disappeared from the blood peripheral blood. Relapse occurs when merozoites from hypnozoites in the liver break out, to re-establish a red cell infection (Heyeneman, 2004).

Without treatment *P. vivax* and *P. ovale* infections may persists as periodic relapses for up to 5years. *P. malariae* infections lasting 40years have been reported; this is thought to be a cryptic erythrocytic rather than an exo-erythrocytic infection and it is termed recrudescence to distinguish it from a relapse. During the erythrocytic cycles, certain merozoites are differentiated into males and female gametocytes. The sexual cycle therefore begins in the vertebrate host (figure 2.2), but for its continuation into the sporogonic phase, the gametocytes must be taken up and ingested by blood sucking anopheles (Heyeneman, 2004).

2.5.0 Malaria Vectors And Vector Behaviour

Human malaria is transmitted by the bite of female mosquitoes belonging to the genus *Anopheles* (figure 2.3). Of the 400 species of *Anopheles* in the world, approximately 60 are important vectors of malaria. However, a particular species of *Anopheles* may be an important vector in one area of the world and of little or no consequence in another (National Center for Infectious Diseases, 2004).

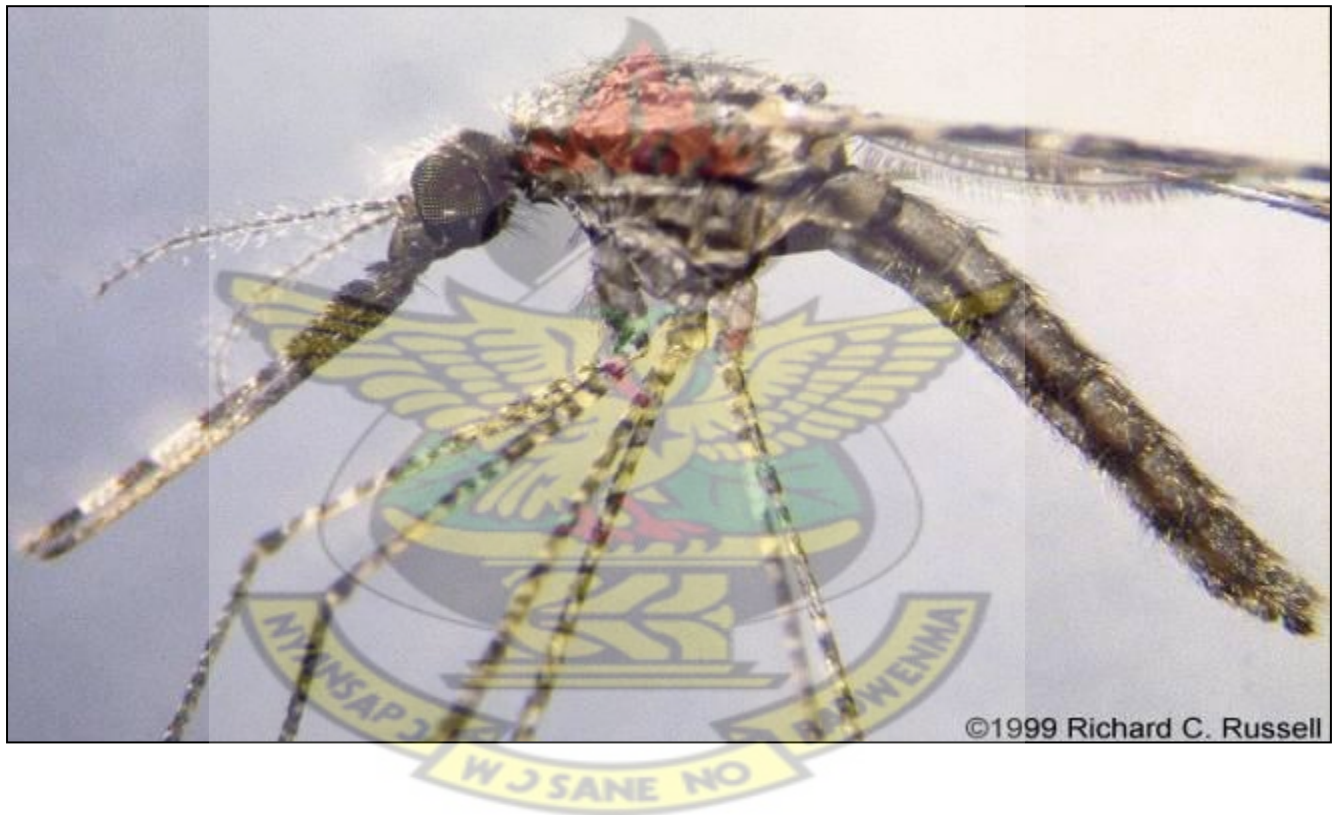


Figure 2.3 A Female *Anopheles gambiae*

Source: Richard C. Russell © 1999

Different species of *Anopheles* can behave differently. Mosquito behaviour can differ in terms of breeding or larval habitat (e.g., fresh vs. brackish water; flowing streams, still pools, or man-made habitats; shaded or sunny sites), feeding preferences (e.g., time of day when peak biting occurs, preferences for people over animals, feeding indoors or outside), and resting habits (resting indoors after feeding or leaving the house before resting). These differences in mosquito behaviour can affect both the epidemiology of malaria and the choice of malaria control strategy used. For example, *A. dirus* is an important vector in Southeast Asia and is primarily a forest dweller. This means that malaria control strategies aimed at preventing mosquito biting in the home (such as residual spraying or insecticide-treated bednets) would be of little value in preventing exposure and infection. *A. gambiae*, the most important vector in much of sub-Saharan Africa, breeds in small temporary pools of water (even as small and temporary as cattle hoof prints). Therefore, vector control strategies aimed at reducing or eliminating breeding sites will likely have little impact. For these reasons, expert advice relevant to the primary malaria vectors in a given area is essential for making sound decisions regarding control options (Bloland and Williams, 2002).

2.5.1 Life Cycle of the Vector

Like all mosquitoes, anophelines go through four stages in their life cycle: egg, larva, pupa, and adult.

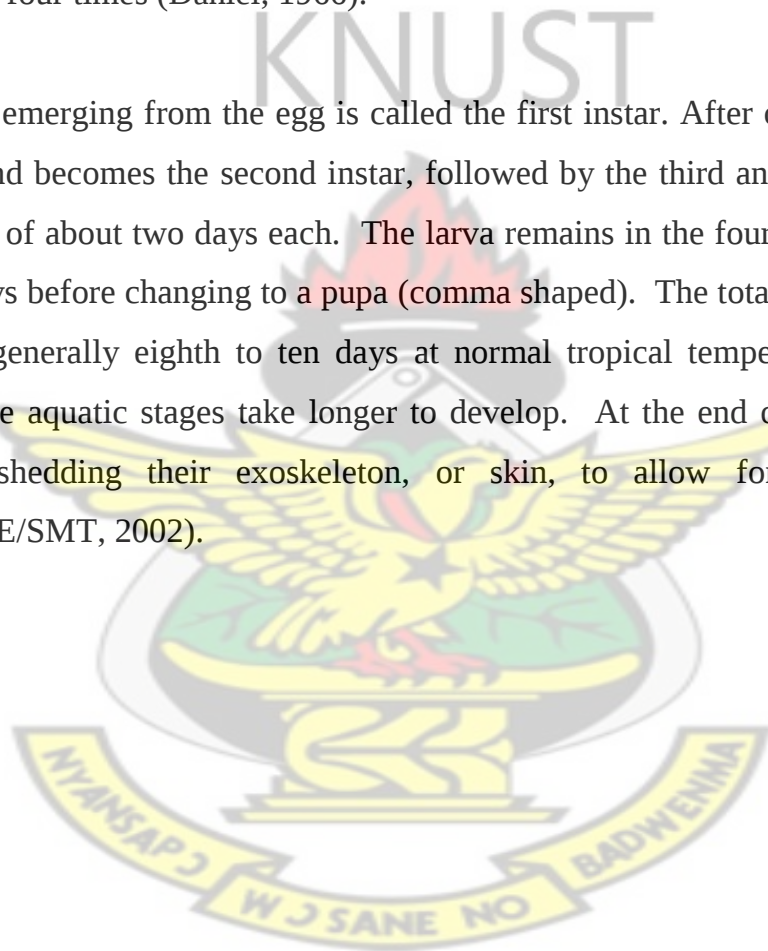
2.5.1.1 Egg

Adult females lay 50-200 eggs per oviposition. Eggs are laid singly directly on water and are unique in having floats on either side. Eggs are not resistant to drying and hatch within 2-3 days, although hatching may take up to 2-3 weeks in colder climates. Oviposition site may vary from small hoof prints and rain pools to streams swamps, canals, rivers, ponds, ponds, lakes and rice fields. Each species of mosquito prefers different habitats to lay eggs (WHO/CDS/CPE/SMT, 2002).

2.5.1.2 Larvae

A larva hatch from the egg after about one or two days and generally floats parallel under the water surface, since it needs to breathe atmospheric air. It feeds by taking up food from the water. When disturbed the larvae quickly swims towards the bottom but soon needs to return to the surface to breathe (WHO/CDS/CPE/SMT, 2002). About 24-hours after hatching, the young larva casts its skin. It repeats this process of feeding, growing, and skin-casting four times (Daniel, 1966).

The small larva emerging from the egg is called the first instar. After one or two days it sheds its skin and becomes the second instar, followed by the third and fourth instars at further intervals of about two days each. The larva remains in the fourth instar stage for three or four days before changing to a pupa (comma shaped). The total time spent in the larval stage is generally eighth to ten days at normal tropical temperatures. At lower temperatures, the aquatic stages take longer to develop. At the end of each instar, the larvae moult, shedding their exoskeleton, or skin, to allow for further growth (WHO/CDS/CPE/SMT, 2002).



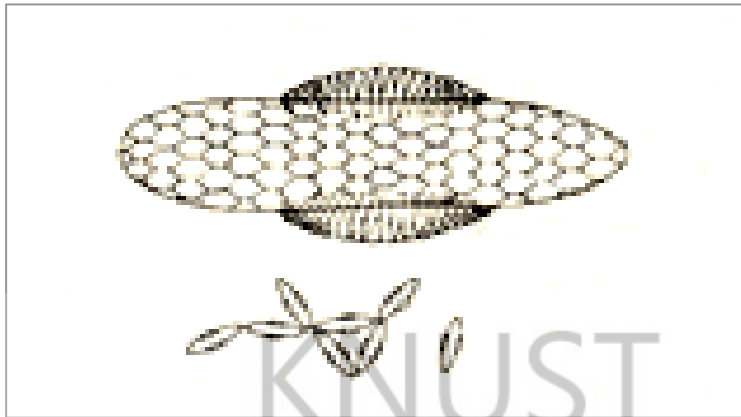


Figure 2.4 *Anopheles* Egg; note the lateral float :

Bottom: *Anopheles* eggs are laid singly

Source: <http://www.cdc.gov/malaria/biology>



Figure 2.5 *Anopheles* Larva. Note the position, parallel to the water surface

Source: <http://www.cdc.gov/malaria/biology>

2.5.1.3 Pupa

This is the stage during which a major transformation takes place, larval tissues are converted into adult tissues,. The pupa is shaped like a comma. It stays under the surface and swims down when disturbed but does not feed. The pupal stage lasts for two to three days after which the skin of the pupa splits. Then the adult mosquito emerges and rests temporarily on the water's surface until it is able to fly (WHO/CDS/CPE/SMT, 2002).

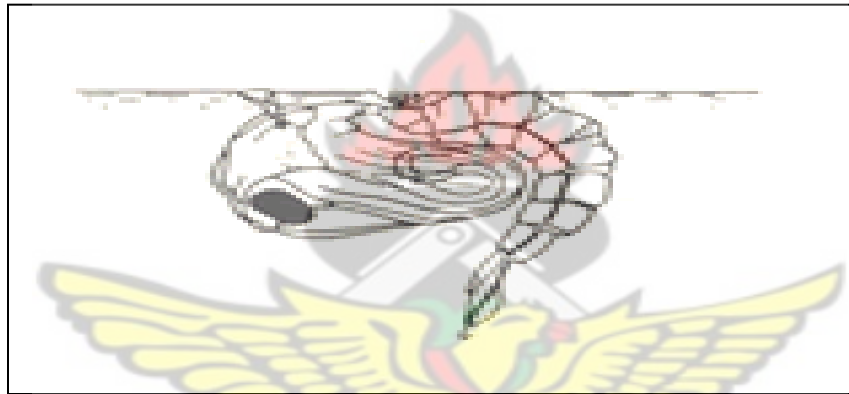


Figure 2.6: *Anopheles* Pupa.

Source: <http://www.cdc.gov/malaria/biology>

2.5.1.4 Adults

The pupal case eventually splits along the back and the adult mosquito emerges and works its way out on to the water surface. After the adult mosquito emerges, it seeks a protective environment in the surrounding vegetation to allow its wings to complete development. Male mosquitoes tend to emerge prior to the female mosquito (Renchie and Johnsen, 2007).

Newly emerged male mosquitoes are unfit for coupling with a female, as the external genitalia require a morphological change. This is accomplished by inversion of the terminalia within the first 24 hr following emergence; a process known as hypopygial circumversion. In many species, male accessory glands mature during the first few days of adult life, and this is needed before sperm can be successfully transferred (Clements 1999). In *Anopheles gambiae* Giles sensu stricto and *A. arabiensis* Patton optimal mating occurs with 5–7-day-old males (Reisen, 2003). Females, by contrast, are ready to mate almost as soon as they emerge from the pupal cases (Provost and Haeger, 1967).

As in other insects, the body of an adult anopheline mosquito consists of three recognizable divisions: head, thorax (fore-body) and abdomen (hind body). The head is equipped with a pair of long jointed antennae (feelers), which is bushy and hairy in the male, thinner and less hairy in the female. The head also has an elongated, forward-projecting proboscis used for feeding, and two sensory palps. The thorax is specialized for locomotion, with a pair of wings and 3 pairs of legs for resting. The hind body or abdomen consists of ten segments, of which only eight can be readily distinguished (Daniel, 1966).

Female and male mosquitoes both require carbohydrate sources (nectar, plant exudates) throughout their life to maintain energy for flying, mating and seeking hosts for blood meals. Only the female mosquito takes a blood meal; she needs the extra protein to develop eggs. The process of taking a blood meal is how the mosquito is able to vector

viruses, protozoans and helminthes (worms) to humans and animals. Male mosquitoes tend to live only a week or two while female mosquitoes can live for up to a month and produce multiple batches of eggs. Some mosquito species overwinter as blood fed females and can survive for multiple months (Renchie and Johnsen, 2007).

In the "freshly fed" females the abdomen is almost entirely filled with blood, but quite rapidly in the ensuing hours the maturation of the eggs entails blood digestion and a substantial increase of the volume occupied by the ovaries. The process is well discernible from the outside and, depending on the relative position of blood (or blood relics) and developing ovaries, mosquitoes are defined as "sub gravid" or "gravid". Usually, to lay the first batch of eggs a female mosquito requires 2 or even 3 blood meals, but subsequently blood taking and oviposition alternate regularly (Esposito and Habluetzel, 1997)

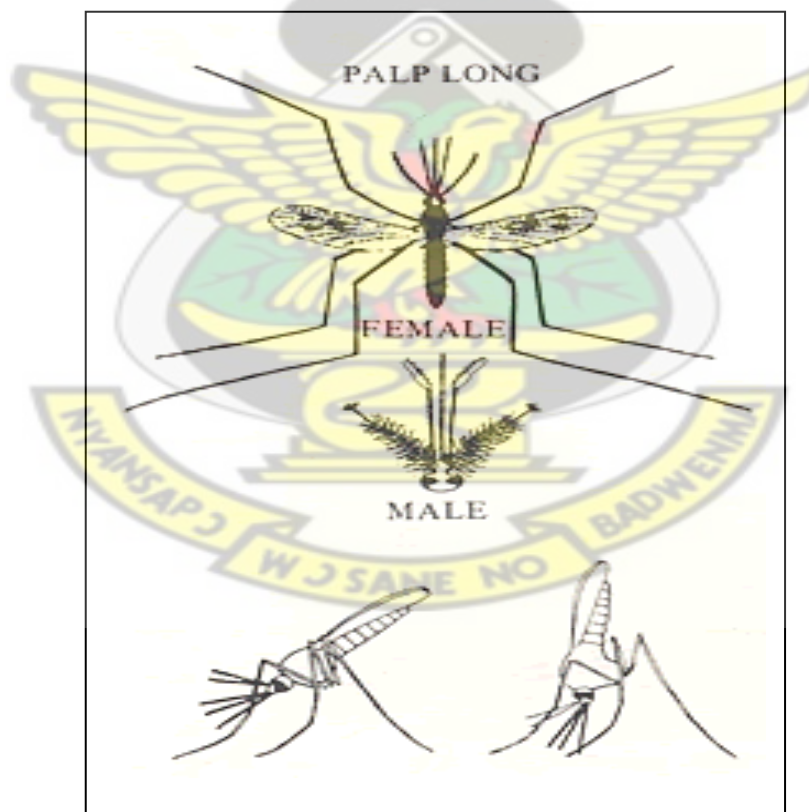


Figure 2.7: *Anopheles* Adults. Note (bottom row) the typical resting position.

Source: <http://www.cdc.gov/malaria/biology>

2.5.2 *Anopheles gambiae* complex

Anopheles gambiae refers to a complex of morphologically indistinguishable mosquitoes in the genus *Anopheles*, which contains the most important vectors of malaria in Sub-Saharan Africa, and the most efficient malaria vectors in the world. This species complex consists of: *Anopheles arabiensis*, *Anopheles bwambae*, *Anopheles merus*, *Anopheles melas*, *Anopheles quadriannulatus*, and *Anopheles gambiae* sensu stricto (Brunhes *et al*, 1998). Despite being morphologically indistinguishable, individual species of *Anopheles gambiae* complex exhibit different behavioural traits. For example, the *Anopheles quadriannulatus*, is generally considered to be zoophilic, (taking its blood meal from animals) whereas *Anopheles gambiae* sensu stricto is generally anthropophilic (Scott, 1993). *A. gambiae* s.s, M and S molecular forms (i.e. the Mopti (M) and Savannah (S)) have been discovered in the western savannah regions of Africa (WHO, 2006a).

2.5.3 Vectorial efficiency

The two most important members of the *A. gambiae* complex are the *A. arabiensis*, with females blood-feeding on livestock or humans plentifully indoors or outdoors, and *A. gambiae* s.s. with females more likely to bite humans indoors. Evidently these anophelines have co-adapted to human ecosystems in the Afro-tropical savannah where their combined contributions to malaria transmission have apparently facilitated the evolution of falciparum malaria (Coluzzi, 1999). Due to their endophilic and anthropophagic behaviour, *A. funestus* and *A. gambiae* s.s. seldom occur away from human habitations (WHO, 2006a). Malaria sporozoite rates are generally higher in populations of *A. gambiae* s.s., followed by *A. funestus*, than in *A. arabiensis*, making *A. gambiae* and *A. funestus* more effective vectors with a capacity to sustain malaria transmission at lower vector population densities than *A. arabiensis* (WHO, 2007). Their exceptionally high vectorial capacity can be attributed to their endophilic resting behaviour, allowing relatively longer survival rates than for exophilic adult mosquitoes, as well as their propensity to feed on humans repeatedly (WHO, 2006a).

2.6.0 Manifestation of the Disease

Following the infective bite by the *Anopheles* mosquito, the incubation period goes by before the first symptoms appear, and the disease condition may present as either uncomplicated or complicated malaria (Kakkilaya, 2006)

2.6.1 Uncomplicated Malaria

The classical malaria attack lasts 6-10 hours. It consists of: a cold stage, a hot stage (fever, headaches, vomiting; seizures in young children), and finally a sweating stage. The attacks occur every second day with the "tertian" parasites (*P. falciparum*, *P. vivax*, and *P. ovale*) and every third day with the "quartan" parasite (*P. malariae*). More commonly, the patient presents with a combination of the following symptoms: fever, chills, sweats, headaches, nausea and vomiting, body aches, and general malaise. These periodic paroxysms of malaria are closely related to events in the bloodstream (Heyeneman, 2004).

2.6.2 Severe Malaria

Severe malaria occurs when *P. falciparum* infections are complicated by serious organ failures or abnormalities in the patient's blood or metabolism. In Africa, the commonest complications precipitating admissions to hospitals are severe anaemia due to Haemoglobinuria (haemoglobin in the urine) due to haemolysis, abnormalities in blood coagulation and thrombocytopenia, and cerebral malaria, with abnormal behaviour, impairment of consciousness and convulsions (WHO, 2001), and acute respiratory distress syndrome (ARDS) (Gachot *et al.*, 1995). Other manifestations include acute kidney failure; hyperparasitemia, where more than 5% of the red blood cells are infected by malaria parasites; metabolic acidosis (excessive acidity in the blood and tissue fluids), often in association with hypoglycaemia (Greenwood *et al.* 1991); Hypoglycaemia (low blood glucose), which was found to be a common metabolic dysfunction occurring in 8.4% of the children admitted with malaria to Kenyan district hospital (Osier *et al.*, 2003)

2.6.3 Human Host and Malaria

2.6.3.1 Pathophysiology of Malaria

All the typical clinical symptoms and severe disease pathology associated with malaria is caused by the asexual erythrocytic or blood stage parasites.

When the parasite develops in the erythrocyte numerous known and unknown waste substances such as hemozoin pigment and other toxic factors accumulate in the infected red blood cell. These are dumped into the bloodstream when the infected cells lyse and release invasive merozoites. The hemozoin and other toxic factors such as glucose phosphate isomerase (GPI) stimulate macrophages and other cells to produce cytokines and other soluble factors which act to produce fever, rigors and probably influence other severe pathophysiology associated with malaria. *Plasmodium falciparum*-infected erythrocytes, particularly those with mature trophozoites, adhere to the vascular endothelium of venular blood vessel walls and do not freely circulate in the blood. When this sequestration of infected erythrocytes occurs in the vessels of the brain it is believed to be a factor in causing the severe disease syndrome known as cerebral malaria, which is associated with high mortality (National Center for Infectious Diseases, 2004). Anaemia may result from severe malaria infections and is frequently severe in children and pregnant women infected with *P. falciparum*. Intravascular haemolysis does not appear to be a major contributor to malarial anaemia except in the pathological state known as “black water” fever (National Center for Infectious Diseases, 2004).

2.6.3.2 Genetic Factors That Influence Malaria

Innate factors are those specific characteristics of a host that are present from birth. Several innate factors influence malaria infection. For example, persons who carry the sickle cell trait (heterozygotes for the abnormal haemoglobin gene HbS) will be relatively protected against severe disease and death caused by *P. falciparum* malaria. In general, the prevalence of haemoglobin-related disorders and other blood cell dyscrasias, such as Haemoglobin C, the thalassemias and G6PD deficiency, are more prevalent in malaria

endemic areas and are thought to provide protection from malarial disease. Another example of a genetic factor involves persons who do not have the Duffy blood on their erythrocytes. These Duffy negative individuals have red blood cells that are refractory to infection by *P. vivax*. Most of the people in West Africa and much of East Africa do not have this receptor and they are protected from *P. vivax* infection (National Center for Infectious Diseases, 2004). Point mutations in the mannose binding protein (MBP) and in the promoter regions of both the TNF- and NOS2 genes also been reported to influence the severity of disease due to infection with *P. falciparum* (Migot-Nabias, 2000).

2.6.3.3 Immune Responses to Malaria

The human body's defences against infection with malaria fall, in general terms, into natural (innate) resistance and acquired immunity. People residing in malaria-endemic regions acquire immunity to malaria through natural exposure to malaria parasites. Children living in areas of stable malaria transmission become infected early in life, and experience more severe disease symptoms during the first five years of life. But as immunity develops the disease becomes less severe and the number of parasites circulating in the blood declines. The acquired immune response to malaria is strain specific and is lost if a person moves away from a malaria endemic area (Cross, 2004).

Following infection with *Plasmodium* parasites, antibodies against schizont and merozoite antigens bind to infected red blood cells and to merozoites, and make them easier for macrophages and other immune cells to ingest. Antibodies also help other immune mediators, called complement proteins, to destroy parasites and they prevent merozoites infecting new red blood cells. Macrophages which have taken up *Plasmodium* express parasite antigens on their surface, and other immune cells called T cells recognize these antigens and bind to them. The T cells become activated and release molecules called cytokines that promote further cell activation, parasite killing and antibody production (Cross, 2004).

2.7.0 Burden of malaria on health in Africa

2.7.1 Mortality

Approximately 20,000 children die from Malaria every year (25 per cent of the deaths of children under the age of five) (UNICEF, 2007). There are three principal ways in which malaria can contribute to death in young children. First, an overwhelming acute infection, which frequently presents as seizures or coma (cerebral malaria), may kill a child directly and quickly. Second, repeated malaria infections contribute to the development of severe anaemia, which substantially increases the risk of death. Third, low birth weight - frequently the consequence of malaria infection in pregnant women - is the major risk factor for death in the first month of life. In addition, repeated malaria infections make young children more susceptible to other common childhood illnesses, such as diarrhoea and respiratory infections, and thus contribute indirectly to mortality (Adams *et al.*, 2004).

2.7.2 Morbidity and long-term disability

Children who survive malaria may suffer long-term consequences of the infection (UNICEF, 2007). Repeated episodes of fever and illness reduce appetite and restrict play, social interaction, and educational opportunities, thereby contributing to poor development. An estimated 2% of children who recover from malaria infections affecting the brain (cerebral malaria) suffer from learning impairments and disabilities due to brain damage, including epilepsy and spasticity (Murphy and Breman, 2001).

2.7.3 Burden of malaria on the poor

Poor people are at increased risk both of becoming infected with malaria and of becoming infected more frequently. Child mortality rates are known to be higher in poorer households and malaria is responsible for a substantial proportion of these deaths. In a demographic surveillance system in rural areas of the United Republic of Tanzania, under-5 mortality following acute fever (much of which would be expected to be due to malaria) was 39% higher in the poorest socioeconomic group than in the richest

(Mwageni *et al*, 2002). A survey in Zambia also found substantially high prevalence of malaria infection among the poorest population groups (RBM Zambia, 2001). Poor families live in dwellings that offer little protection against mosquitoes and are less able to afford insecticide-treated nets. Poor people are also less likely to be able to pay either for effective malaria treatment or for transportation to a health facility capable of treating the disease (WHO, 2001). Both direct and indirect costs associated with a malaria episode represent a substantial burden on the poorer households. A study in northern Ghana found that, while the cost of malaria care was just 1% of the income of the rich, it was 34% of the income of poor households (Akazili, 2002). A survey in Malawi showed that 4% of the income of very poor households is spent on malaria prevention, compared with 16% of the middle and upper income households. Inequity in access to health services and malaria interventions is a further disadvantage to the poor (WHO, 2006b).

2.8.0 Control of the Disease

A basic malaria control program combines five components: a public health surveillance system, curative services, preventive interventions, a program for community involvement, and a capacity to perform special studies (operational research) as needed (Bloland and Williams, 2002). WHO recommends that such a program should ultimately aim at the diagnosis and treatment of malaria cases, preventing mosquito bites, and killing mosquitoes (WHO/CDS/CPE/SMT, 2002).

2.8.1 A Public Health Surveillance System

A public health surveillance system is required to monitor temporal changes in disease incidence, to warn of epidemics, and to evaluate the impact of control efforts. This is obviously important for more than just malaria, and a well-designed and integrated surveillance system provides, but also an essential tool for monitoring a rapidly changeable health situation. A well-designed surveillance system allows for rapid identification of increases in cases of communicable diseases in an affected area, signalling the need for a specific response. Basic demographic data should be collected,

in addition to simple case counts using standardized case definitions. These include age, sex, pregnancy status, location of residence in the settlement area, and so forth. These data can be used to identify high-risk subgroups in the general population or areas in a settlement that might require specific attention (e.g., previously unrecognized vector breeding sites) (Bloland and Williams, 2002).

2.8.2 Curative services

Curative services include the provision of prompt diagnosis of malaria and an assessment of its severity, diagnosis of malaria-associated anaemia and other potential consequences of the disease, and prompt and effective treatment of both malaria and anaemia (Bloland and Williams, 2002). WHO now recommends combination therapies as the treatment policy for falciparum malaria in all countries experiencing widespread resistance of *P. falciparum* to monotherapy with conventional antimalarial drugs such as chloroquine and sulfadoxine–pyrimethamine. Artemisinin-based combination therapies (ACTs) are the most highly efficacious treatment regimen. It contains a derivative of the plant *Artemisia annua*, which is presently cultivated mainly in China and Viet Nam (WHO, 2005). The following ACTs combinations have been recommended artemether+amodiaquine, artesunate+mefloquine, artemether-sulfadoxine-pyrimethamine or arthemether-lumefantrine (Olumese, 2008).

2.8.3 Preventive Interventions

Preventive interventions are essential for limiting malaria-associated morbidity and mortality that would not be achievable through curative services alone. Prevention would include some combination of preventive use of anti-malarial drugs, encouraging use of personal protection measures which may include either the usage of insecticide-treated nets (ITN) or repellents, and vector control (Bloland and Williams, 2002).

2.8.3.1 Use of anti-malarial drugs

The use of chemoprophylactic drugs forms a major aspect in protection against malaria, and its use is being increasingly recommended in traveller's advice and used in control strategies against the infection (Corachan, 1997).

While in endemic areas only selected groups in specific situations (infants, pregnancy and the returning migrants) will use chemoprophylaxis, any tourist visiting these areas should use one or a combination of prophylactic drugs together with other personal protection (such as repellents, bednets, personal outfit etc) against mosquito bites.

The Epilepsy Research UK (2008) recommended some anti-malarial tablets mostly for travellers into malaria endemic countries to be started two days before arriving in the country where the risk of malaria exists.

2.8.3.2 Personal protection measure

Personal protection measures are based on insecticide-impregnated materials such as bed nets and curtains mainly. It has been demonstrated that if they are properly applied they can provide a 30 to 60 percent reduction in malaria morbidity, and can be useful in terms of preventing drug resistance (Touré, 2001). In areas of malaria transmission where sustained vector control is required, ITNs are the principal strategy for malaria prevention. Most National Malaria Control Programmes use various implementation methods to promote the usage of ITNs. The implementation methods may include: (i) stimulating the growth of commercial markets; (ii) reducing taxes and tariffs; (iii) cost-sharing; (iv) social marketing subsidies; and (v) ITN distribution free of charge among vulnerable groups such as children under 5 years of age, pregnant women and the poorest or most marginalized populations (WHO, 2005).

2.8.3.3 Vector controls

Vector control remains the most generally effective measure to prevent malaria transmission (WHO 2006a). The idea behind vector control is to reduce levels of mortality and morbidity by reducing transmission of the disease (Touré, 2001). WHO recommends a systematic approach to vector control based on evidence and knowledge of the local situation. This approach is called Integrated Vector Management (IVM). The IVM approach includes; eliminating breeding sites and killing the mosquito, to help reduce the number and, in the case of adults, the longevity of vectors. WHO recommends draining or filling areas where water collects or modifying the preferred habitats of

particular vector species as among several other ways by which breeding sites can be eliminated. Larval breeding can be reduced or prevented by either: petroleum oiling on the water surface to prevent larvae from breathing, covering the water surface with floating materials that deter the mosquitoes from laying eggs, treating the water with larvicides to kill larvae, and employing the use of larvivorous fish as predators (WHO/CDS/CPE/SMT, 2002 and WHO, 2006a). In some areas malaria transmitted by vectors that rest indoors can be prevented or controlled by spraying the insides of the house with residual insecticides. The principle employed by this method is that, if the surfaces on which indoor resting mosquitoes rest are sprayed with residual insecticides, the mosquito may eventually pick up a lethal dose and be prevented from transmitting the parasite (WHO/CDS/CPE/SMT, 2002).

2.8.4 Community involvement

The success or failure of any public health program is determined in large part by the public's acceptance and proper use of the services offered (Bloland and Williams, 2002). Community participation in malaria control in Eritrea where 50% of malaria cases were managed at community level by Community Health Agents (CHAs) contributed to the country meeting the Abuja target in 2004 by reducing overall mortality from malaria by nearly 60% compared to the 1999 baseline. Access to ITN's and their regular and proper use, re-treatment improved; 60%-70% of households had at least one ITN and about 66% of children aged below five years were sleeping under ITN (UNEP (2006).

2.8.5 Special Studies and Operational Research

Finally, agencies should develop a capacity to conduct simple operational research to answer specific question. In some situations, special studies might be required to more accurately estimate the prevalence or incidence of malaria-associated illness, to evaluate the efficacy of malaria therapy, to determine the principle malaria vectors in the area, or to test new interventions or monitor the effectiveness of existing ones. These operational research issues are necessary in order to make sound programmatic decisions (Waldman and Williams, 2001).

CHAPTER THREE

3.0 MATERIALS AND METHODS

3.1.1 Study Area

The Kwame Nkrumah University of Science and Technology, KNUST, located within the Kumasi metropolitan area, of the Ashanti region of Ghana, was the study area (fig. 3.1 and fig. 3.2).

The Ashanti Region is centrally located in the middle belt of Ghana (fig. 3.1a &b). It lies between longitudes $0^{\circ} 9'W$ and $2^{\circ} 15'W$, and latitudes $5^{\circ} 30'N$ and $7^{\circ} 27'N$, and occupies a total land area of 24,389 km² representing 10.2% of the total land area of Ghana. The region has a population density of 148.1 persons per km², compared with a national average of about 80 persons per km² (GSS, 2000).

Kumasi, which is the capital, is located 300 kilometres Northwest of Accra, and is the most populous district. The 2000 census recorded the region's population as about 3.5 million people, representing 19.1 per cent of the country's population. The urban population (51.3%) in the region exceeds that of the rural population (48.7%). The region is currently the second most urbanized in the country after Greater Accra (87.7%), the national capital. The housing stock in the region is 329,478, of which about 37% are in urban areas and 63% in rural areas (GSS, 2000). Due to its central location all road networks linking the northern sector and the southern sector of Ghana pass through Kumasi, resulting in a high daily influx of traders and civil workers.

Kumasi has a semi-humid tropical climate and lies in the tropical forest zone with an annual average rainfall of 1420 mm spread over about 120 days. The rainfall pattern of the town is bimodal with the major season falling between the months of March and July and a minor rainy season in September/October. The mean monthly temperature of the area ranges from 24°C to 27°C (MSD, 2001).

KNUST is about eight (8) Kilometres away from the centre of Kumasi, the Ashanti regional capital. The Study area lies between latitude 6°39' and 6°47' North and longitude 1°26' and 1°40' West and presents a scenery of buildings interspersed with luxuriant lawns, and covers an area of about eighteen square kilometres of undulating land (Facts and Figures of KNUST, 2008) (fig. 3.2).

KNUST has a population size estimated around 30,000; made up of 741 teaching staff and 2424 non teaching and a total student population size of 22,736 Undergraduate students and 1452 graduate students (at the time of this study) (Facts and Figures KNUST, 2008). KNUST being the only Science and Technology University in Ghana attracts scientists and technologists not only from Ghana but also from other African countries.



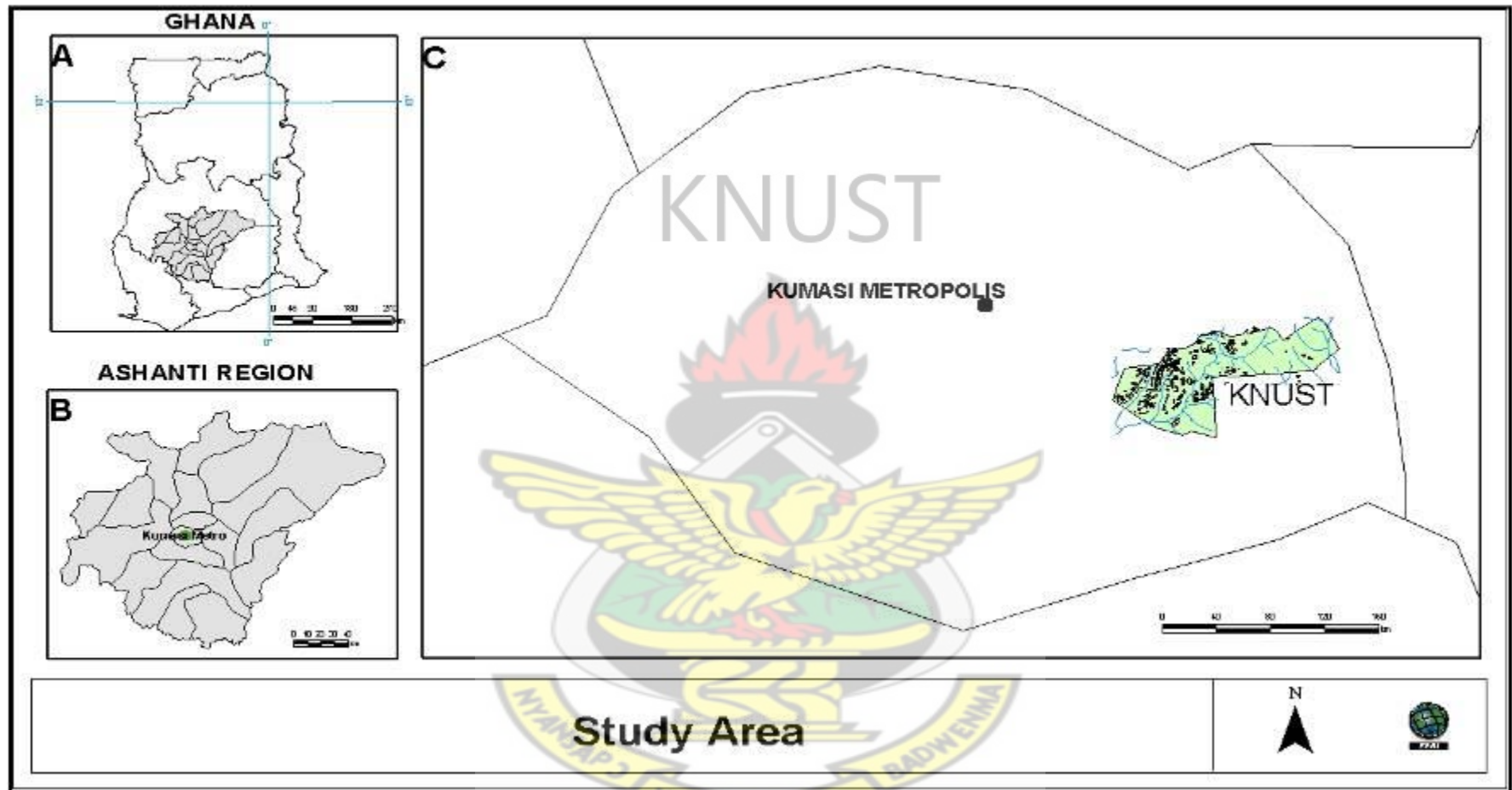


Figure 3.1: A. Map of Ghana; B. District Map of Ashanti region C. Map of the Kumasi Metropolis showing the location of the Study area (KNUST)

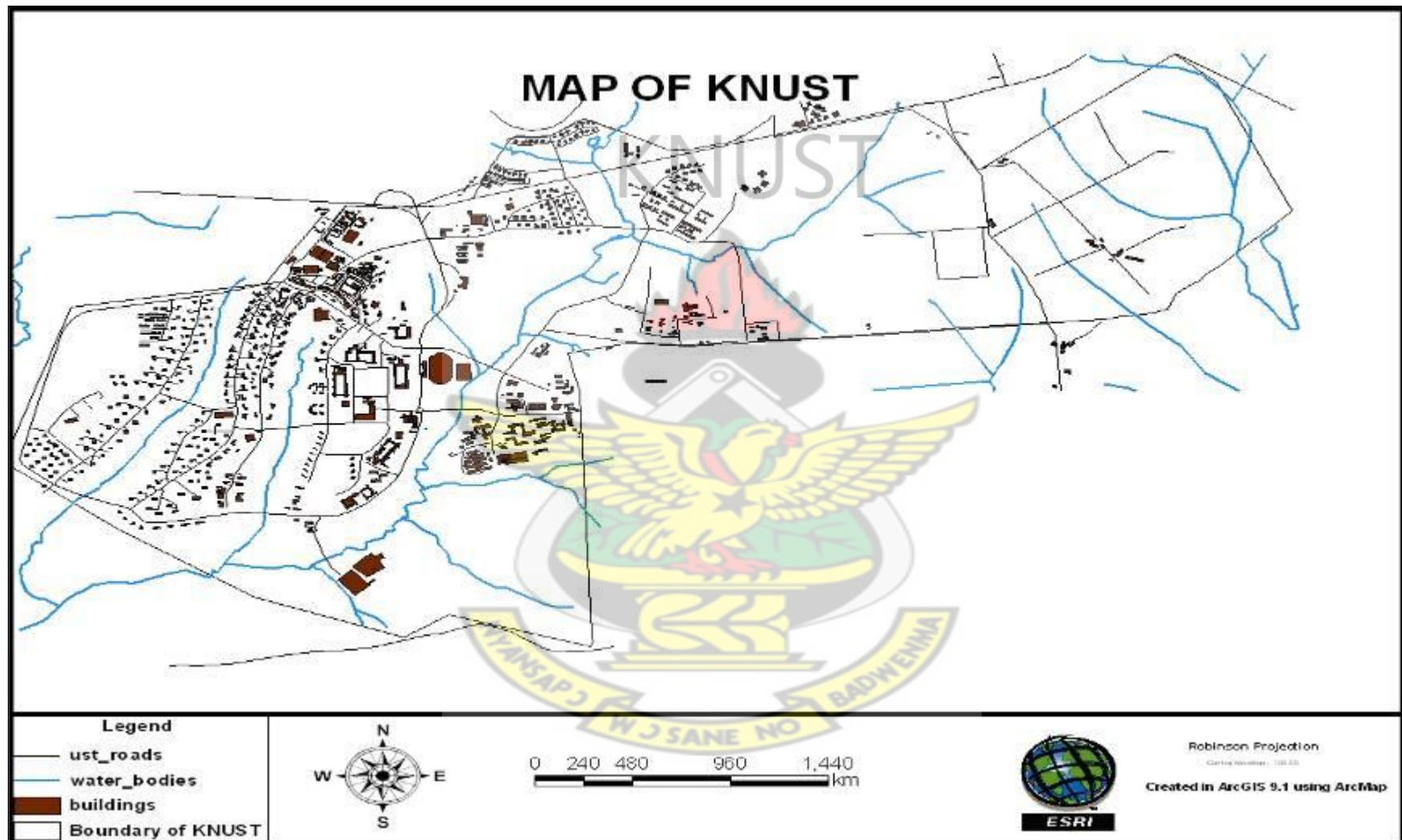


Figure 3.2: Map of Kwame Nkrumah University of Science and Technology (KNUST)

3.1.2 Collection Zones within the Study Area

Four different sites for collection were chosen based on the areas on campus where the residents centred their activities. These areas included two broad zones: the faculty area where most lecturers and students were mostly found in the daytime during the academic year. The second zone was the residential area for both staff and students of the University (see figure 3.3b).

3.1.3 Description of Selected Sites of Mosquito Collection

3.1.3.1 Zone A (Faculty Area)

In this zone most of the activities occur during the day, hence vector-human interaction was assumed to be minimal. One site of mosquito collection selected was to be an area where night activities were situated. The Non-residential facility was selected. From larval surveys this zone was identified to be surrounded by several breeding sites, several water bodies and with some areas with dense vegetation cover of about 60%.

3.1.3.2 Zone B (Residential Area)

Three different sites were selected for the Zone B. It included the Halls of Residence (Republic Hall), Senior and Junior Staff Residence (Hall 6) and the barracks of the University's Security Personnel.

3.2.0 Spatial Mapping

The process of mapping included creating a Geographical Information Systems (GIS) database for the area. A hand held GPS receiver unit, was used to capture spatial coordinates (i.e. longitude and latitude) to geo-reference landscape features, health facilities available, households and residential areas, and breeding sites. The coordinates were then converted into the Cartesian XY Plane and the point plotted on the KNUST map that was developed using ArcMap a component of the GIS desktop ArcView 9.1 software



Figure 3.3a: Aerial view of the Kwame Nkrumah University of Science and Technology and Surrounding communities at an eye altitude of 11648 ft. (6°40'31.89"N 1°34'24.68"W)

Source: Google Earth ©2008

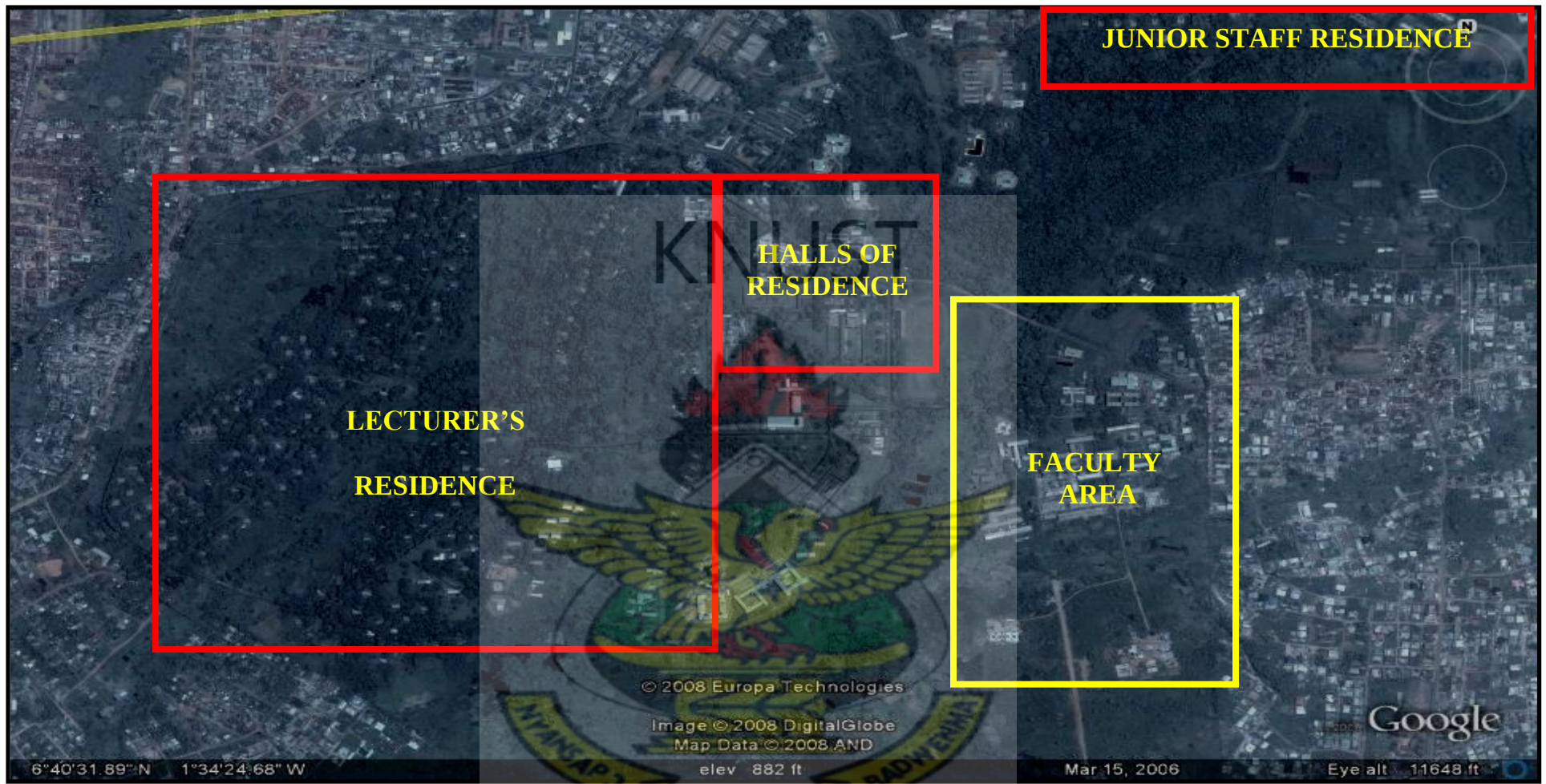


Figure 3.3b: Aerial view of KNUST showing the Study sites for larval and adult mosquito collection.: Zone A (the Faculty area), its boundary indicated by yellow outline, and Zone B (Halls of Residence, Senior and Junior Staff residence and the security barracks) boundary indicated by red outline (6°40'31.89"N 1°34'24.68"W). Source: Google Earth ©2008.

3.3.0 Mosquito Collection

3.3.1 Collection of Mosquito larvae

Through satellite imagery from Google Earth the major topographical features of the KNUST were determined (see Fig. 3.3a), so that the approximate location and extent of permanent sources of water could be marked before larval surveys began. Ground surveys were conducted monthly from October 2007 to February 2008 (dry season) and from March and September 2008 (rainy season). Aquatic habitats that posed as potential breeding places for mosquitoes were all identified. Some of these habitats included; drainage ditches, stream edges, swamps, footprints and other depressions that had collected rain water. The surveys were conducted at every fortnight for the rainy and dry seasons.

A white enamel bowl of a volume of about 350 ml was used to collect larvae from water samples in ditches, and the edges of other large bodies of water and swamps within the study area. For water bodies with surface area of about 0.5 m² 10 dips were used as a standard. However for small breeding sites, such as human footprints a ladle of about 10cm in diameter was used as the dipper. Water collected in either the enamel bowl or the ladle was examined carefully and the larvae coming to the surface were collected by a pipette. Each aquatic habitat that contained anopheline larvae (identified by the position with respect to the water surface) was then geo-referenced using a hand-held GPS.

The corresponding surface temperatures (°C) and pH of water from each sampling site were recorded on each sampling day. The mean and the standard deviation (s.d.) of all repeated measurements from each sampling sites was calculated. Regression analysis was carried out to account for physicochemical variables that determined the presence and abundance of *Anopheles* larvae at each breeding site (see Table 4.4).

The larvae were transferred into large labelled collection bottles half full of water from the breeding site. The larvae were then transported to the insectary, and reared to adulthood for identification.

Sample of water collected from each habitat, represented a single breeding site; hence the number of adult females that emerged from each water sample at the insectary per unit time, represent the habitat productivity. Reared mosquito larvae that matured into adulthood were then separated into their respective genera using morphological keys of Gillies and DeMeillon (1968), and Gillies and Coetzee (1987).

3.3.2 Collection of adult mosquitoes

The human landing catch method (HLC) (WHO, 1975; WHO/CDS/CPE/SMT, 2002) was used to collect adult mosquitoes. The principle of this method is that mosquitoes are collected while in the process of biting a human or vertebrate host. The night was divided into two 6-hour periods (18:00 – 00:00 and 00:00- 06:00). Four teams made of trained vector collectors caught adult female mosquitoes from 18:00 – 06:00 hours the next day, from all study sites. Each team consisted of two vector collectors, with each collector working for 6-hour periods. With all other parts of the body protected (covered) and with the aid of a torch light, the collectors caught mosquitoes that landed on their exposed legs into haemolysis plastic tubes of 1.5ml. Mosquitoes caught for each hour were placed in their corresponding moistened cotton bags referred to as hour bags and kept on ice in cooling boxes to immobilize the mosquitoes.

3.4.0 Processing of Adult Mosquitoes

Mosquito caught were processed in the laboratory at the Kumasi Centre for Collaborative Research in Tropical Medicine (KCCR) at KNUST, entomology laboratory. They were first separated into Anophelines and Culicines, using taxonomic keys of Gillies and De Meillon (1968), and Gillies and Coetzee (1987). The *Anopheles* were then identified as either *A. gambiae* s.l. or *A. funestus* with the aid of a stereo microscope using taxonomic keys of Gillies and Coetzee (1987) (as shown in Fig. 3.4), and stored at -20°C for subsequent tests and analysis.

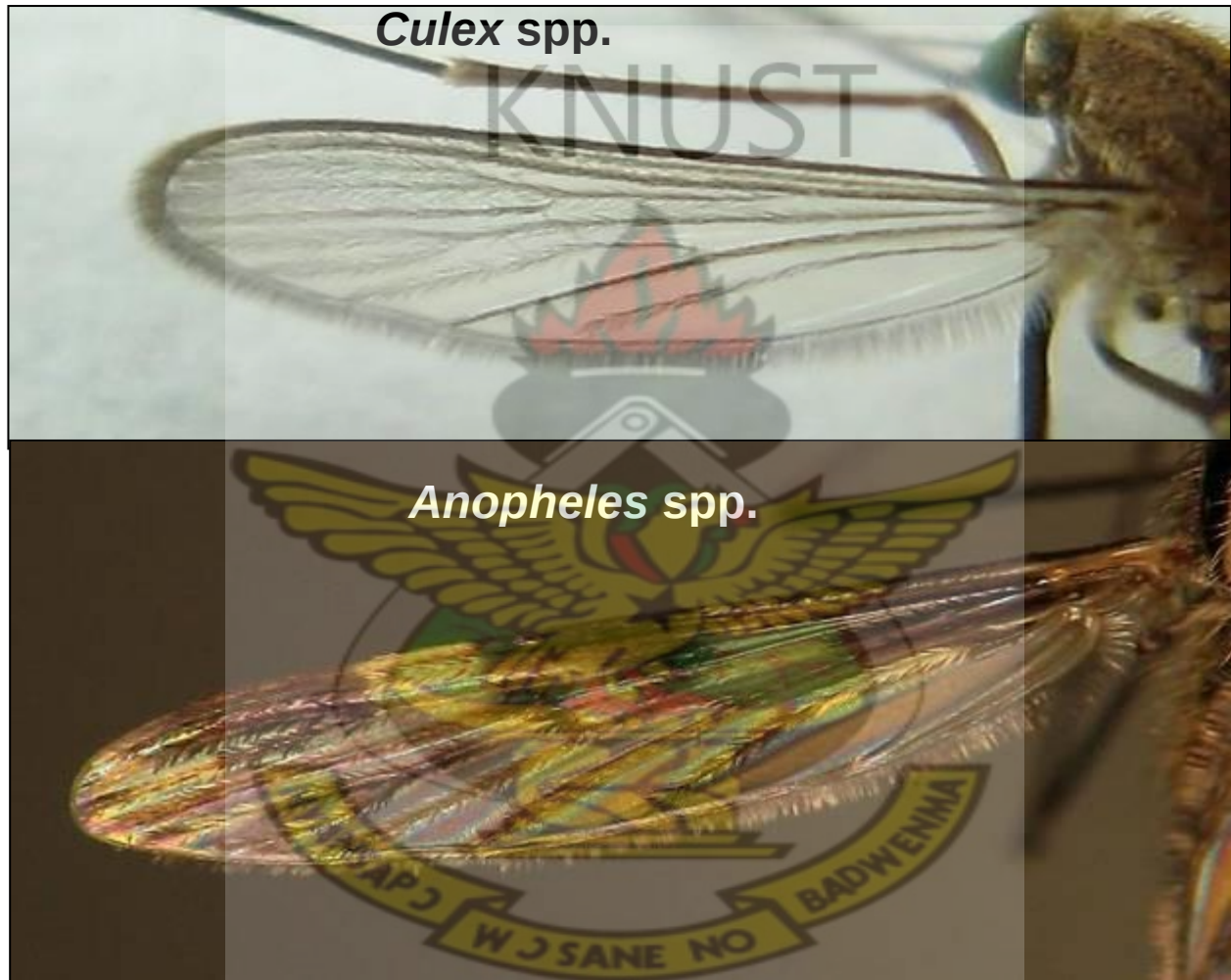


Figure 3.4: Some morphological differences between the wings of a Culicine (above) and an *Anopheles* spp. (bottom).

Source: Adapted from Stephen Luk Richmond Hill, Ontario, Canada

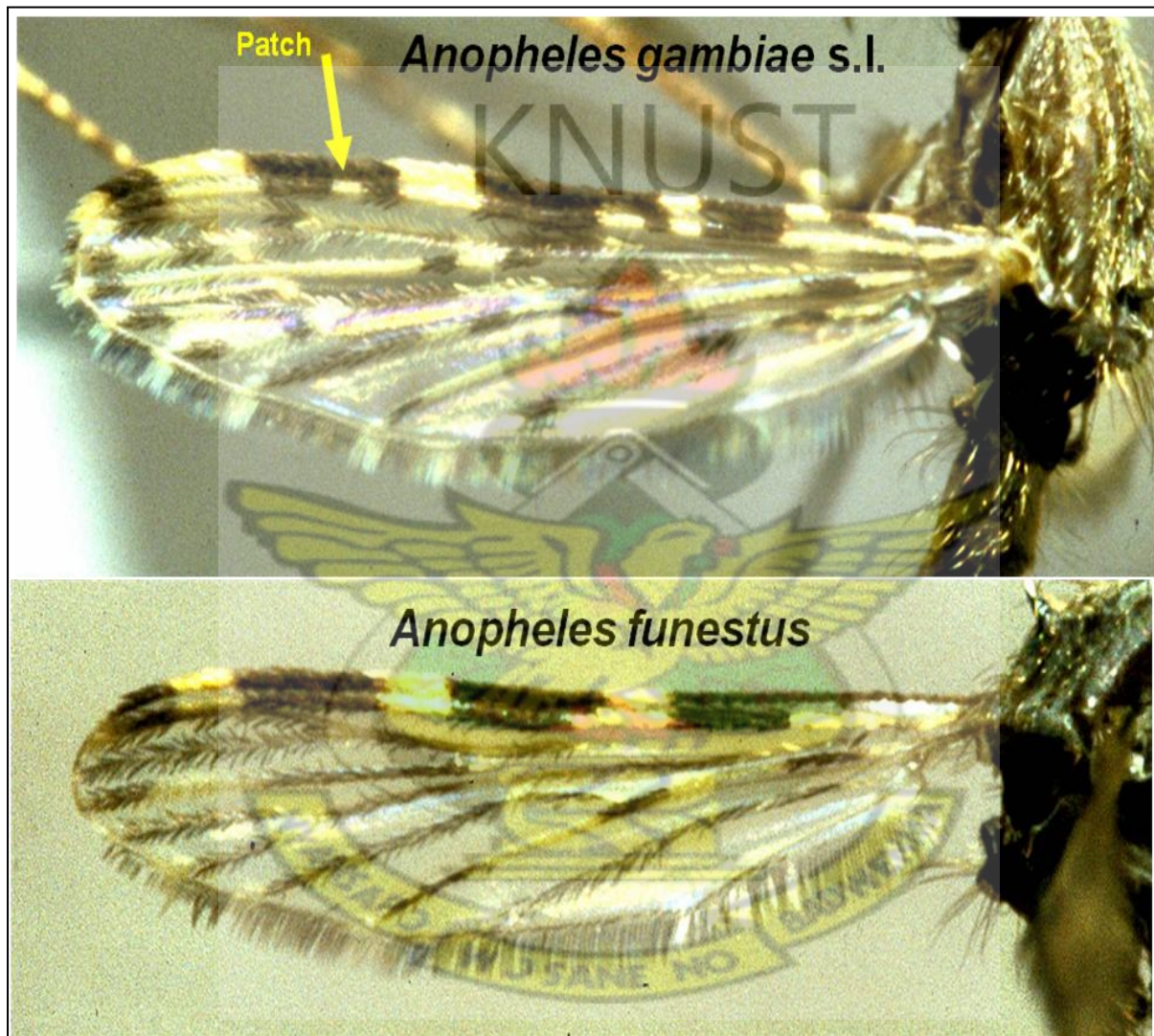


Figure 3.5: Differences in banding pattern between the wings of the *Anopheles gambiae* complex (top) and *Anopheles funestus* complex (bottom).

Source: Professor R. Garms, Bernhard Nocht Institute for Tropical Medicine, Hamburg, Germany.

3.4.1 Dissection of Mosquitoes

Dissection of adult mosquitoes was carried out to determine the proportion of infective mosquitoes (i.e. mosquitoes with sporozoites in their salivary gland), the proportion of mosquitoes with oocyst on the walls of midgut, and to determine the proportion of parous mosquitoes (parity). Dissection was performed only on fresh mosquitoes. Mosquitoes identified as female *Anopheles* spp. were immobilized at -20°C for ten minutes. Mosquitoes to be dissected were then arranged according to hour of collection on ice, while in the collection tubes. Each mosquito was placed on a clean labelled slide and, the legs and wings were removed and stored at -20°C in a micro-titre plate for further identification of *Anopheles* spp. into sibling species.

3.4.1.1 Salivary Gland Dissection and Examination

This was carried out to determine the sporozoite rate. Mosquitoes were arranged on the slide with the head pointing to the right. The needle on the left was placed on the neck of the mosquito without cutting it; then with light movements the head was gently detached from the body. A gentle traction on the thorax allows the glands to be expressed. The right needle was used to detach the glands from the head. Both glands were then transferred into a drop of x1 phosphate buffer solution (PBS). A cover slip was placed on the removed glands and then the slide transferred to the high power compound microscope for examination. Removed glands were then ruptured by pressing gently on the cover slip to disrupt gland cells to free the sporozoites present if the glands cells were infected. If present, the sporozoites were seen as very minute needle-like forms either in clusters or isolated.

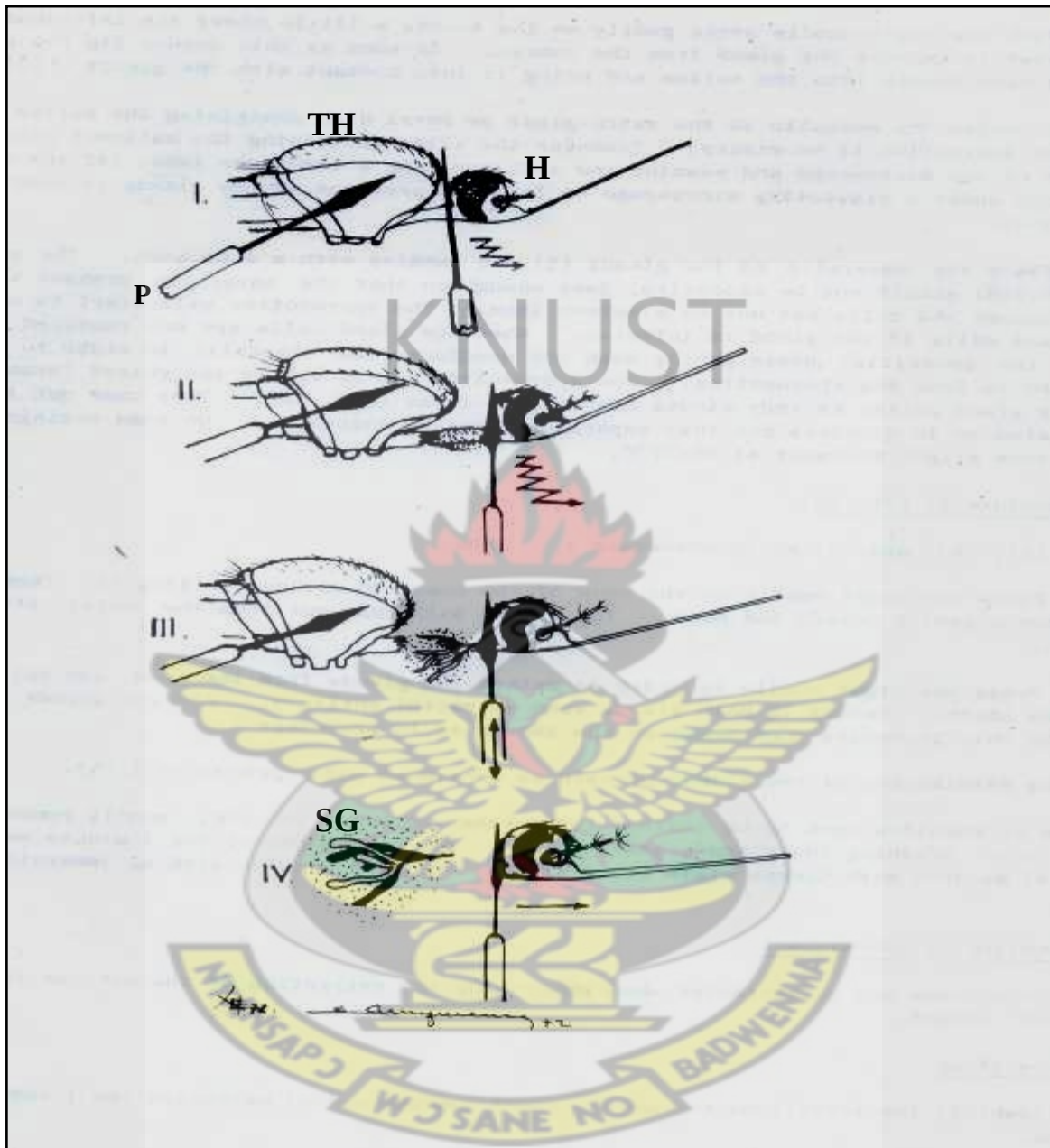


Figure 3.6: Dissection of an adult mosquito for its Salivary gland; H = head, P= dissecting pins, SG = Salivary gland and TH= thorax.

Source: WHO Medical Entomology Manual

3.4.1.2 Ovary Dissection and Examination

The abdomen was separated from the thorax, by placing the immobilized mosquito on its side. A drop of water was added to the extremity of the abdomen. One dissecting needle was placed in the thorax muscle while the other needle was used to make a small cut between VI and the VII sternite (terminal segments of the abdomen). The terminal segment was pulled away from the abdomen by a gentle traction until the ovaries emerged together with the mid gut. The mid gut was then examined for the presence of oocysts.

The ovaries were carefully transferred to the edge of the drop of water for drying. The dried ovaries were then examined under x40 magnification of a compound microscope.

Examination of the ovaries involved checking of the ovaries to see condition of tracheal skeins. Parity was determined by observing the degree of coiling in the ovarian tracheoles (Detinova, 1962). The nulliparous female were seen with coiled tracheolar 'skeins' in the ovaries, whereas the tracheoles. of the parous females appeared permanently stretched in the parous ovaries due to previous growth of eggs (Figure 3.7). The presence of eggs in the ovaries of dissected mosquitoes also suggested parity. The number of parous females was recorded and used to determine the parity rate.

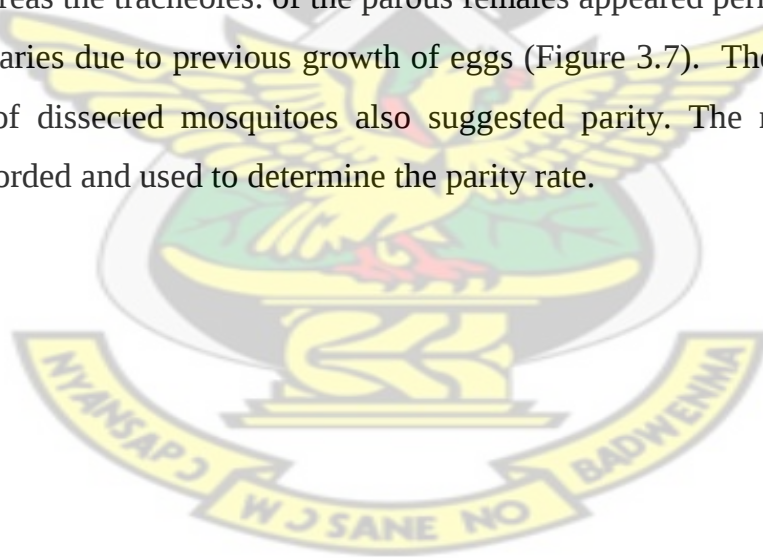




Figure 3.7: Ovaries of an Adult mosquito; **I.** Ovary of a nulliparous female: tracheolar system forming 'skeins'; **II.** End of fine trachea forming skein (T= trachea, Tr=trachioles); **III.** Ovary of a parous female with stretched tracheoles.

Source: WHO Medical Entomology Manual

3.4.2 Entomological Indices

3.4.2.1 Man Biting Rate

The Man Biting Rate (MBR) refers to the average number of bites per person per night by a vector species (WHO/CDS/CPE/SMT, 2002). This was determined directly by recording the number of mosquitoes collected by vector collectors and dividing this number by the number of collectors and number nights of activity.

$$\text{Man Biting Rate} = \frac{\text{Number of Anopheles collected}}{\text{Number of out door Collectors} \times \text{Number of nights of activity}}$$

3.4.2.2 Sporozoite rate (Sporozoite Index)

This was estimated as the percentage of *Anopheles* mosquitoes (collected by vector collectors within the study period from October 2007 to October 2008); that had sporozoite positive glands after dissection.

3.4.2.3 Entomological Inoculation Rate

Entomological Inoculation Rate (EIR) is defined as the number of infective bites received per person within a given period (WHO/CDS/CPE/SMT, 2002). It was calculated as the product of the man biting rate and the sporozoite rates give

$$EIR = \text{Sporozoite Rates} \times \text{Man Biting Rate}$$

3.4.2.4 Parity Rate Determination

The parity rate was determined by dissection, and the parous mosquitoes (those that have taken a blood meal at least once and laid eggs at least once) were separated from the nulliparous mosquitoes (those that had not taken a blood meal yet and had never laid eggs). It was determined as the proportion (percentage) of parous female *Anopheles* spp. amongst the total number of mosquitoes dissected.

3.4.2.5 Longevity and Expectation of Life of *Anopheles* spp. collected

The survival of a female mosquito after a blood meal (probability of surviving one day after blood meal, denoted as p and expectation of life for n days (n being the number of days for a sporogonic cycle to be completed) (WHO/CDS/CPE/SMT, 2002), was calculated as below:

Given an interval of two days between blood meals, the probability of surviving one day (denoted as p) can be estimated as:

$$\text{Probability of Survival } p = \sqrt[n]{\text{Proportion of Parous Anopheles}}$$

p assumes that the mosquito population has a stable size and age structure, and the death rate is independent of age. For this reason, the proportion parous is usually averaged over the whole population cycle, to eliminate the effect of seasonal fluctuation in population size and age structure (WHO/CDS/CPE/SMT, 2002).

Expectation of Life for a mosquito species is defined as the expectation of life for n days, where n represent the number of days the sporogonic cycle may be completed.

It was calculated as:

$$\text{Expectation of life} = \frac{1}{- \ln p}$$

3.5.0 Knowledge, Attitude and Practice (KAP) of Malaria

The questionnaire was designed to obtain personal information on the respondents, including occupation and level of education.

3.5.1 Selection of Study subjects

Two inclusion criteria were used in selecting subjects for the study, one for student respondents and the other for staff and all other respondent within the study area. The criterion for students required that a respondent should have stayed on campus for at least an academic year (2007/2008). For staff and all other respondents on the other hand, respondent who had lived on or around campus for the said academic year 2007/2008 or the last six 6 months, were interviewed.

3.5.2 Sample Size Estimation

The total number of persons chosen for this survey was 613. Using Statcalc from Epi-Info, with an estimated population size of 31,000 for the study area, an expected frequency 93.4% (% with high KAP in a previous study of the area; unpublished BSc dissertation by Arthur, May 2008) and an assumed worst acceptable value of 95.0%.

3.5.3 KAP Survey

In examining treatment-seeking behaviour, the questionnaire explored: How the respondents knew that they had malaria; treatment seeking behaviour; where they sought treatment and the reasons for doing so. To investigate the use of health services they were asked which options they usually resorted to in case of a fever episode, including: hospital attendance or self medication. The understanding and application of preventive practices and preferred preventive methods (such as bed-nets, smoke-fumigation, repellents etc.), were also investigated. Logistic regression analysis was used to explore the possible associations between schooling and knowledge of malaria transmission, measures used to prevent mosquito bites, and utilization of health services. All statistical analyses were carried out using the Minitab statistical package.

CHAPTER FOUR

4.0 RESULTS

4.1.0 Entomological Studies

4.1.1 Larval Survey

4.1.1.1 Anopheline larval habitat distribution

The anopheline larval habitat types identified were characterised by clear or turbid shallow and sunlit conditions. Some anopheline larvae were also found breeding in either puddles collected in human footprints or drying stream beds, conduits on some vegetable farms, rain-pools, or in swamps.

4.1.1.2 Zones of Larval Surveys

Faculty area (zone A), showed a relatively high degree of land-use changes such as deforestation for constructions projects and agricultural activities. The area is bordered by several vegetable farms on which *Anopheles* larvae were found breeding in either dugout wells, ditches of furrow systems between raised vegetable beds or in human foot prints. In most of the breeding pools on these farms mosquito larvae density were very high.

The breeding sites found within zone B (i.e. the halls of residence, the senior and the junior Staff residence (Hall 6), as well as the security barracks) had varied populations densities of *Anopheles* larval. Some of the habitats that were identified included sand pools or pond, edges of streams, drainage channels on sugar cane and vegetable cultivation site and temporary pools created after rains. Domestic waste water in chocked gutters within this area was characterized by high numbers of *Culex* but no *Anopheles* spp. larvae (figure 4.1 a & 4.1b).

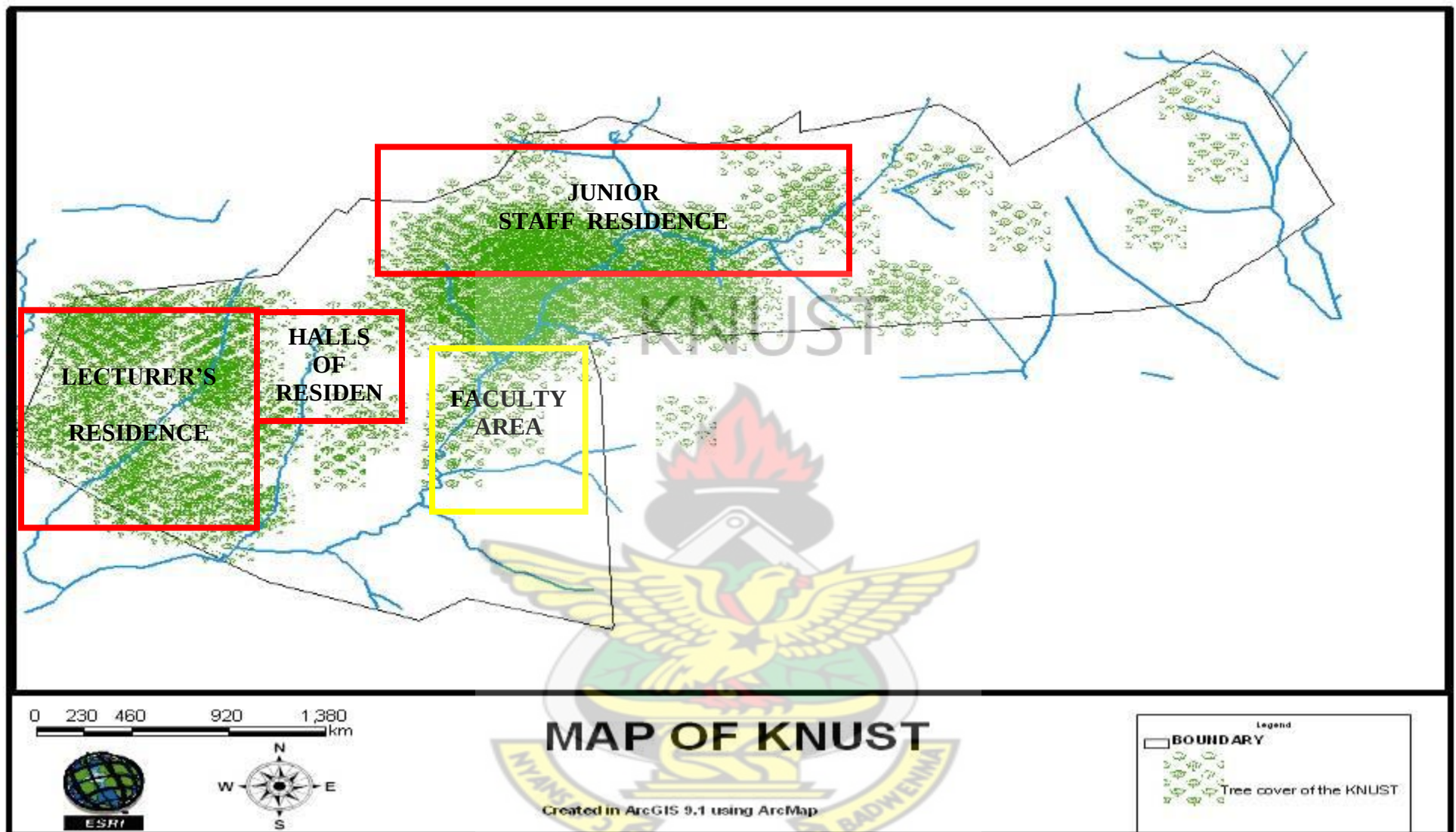


Figure 0.1: Map of KNUST showing the Study sites for larval and adult mosquito collections and the tree cover for each study site. Zone A (the Faculty area), its boundary indicated by yellow outline, and Zone B (Halls of Residence, Senior and Junior Staff residence and the security barracks) boundary indicated by red outline.

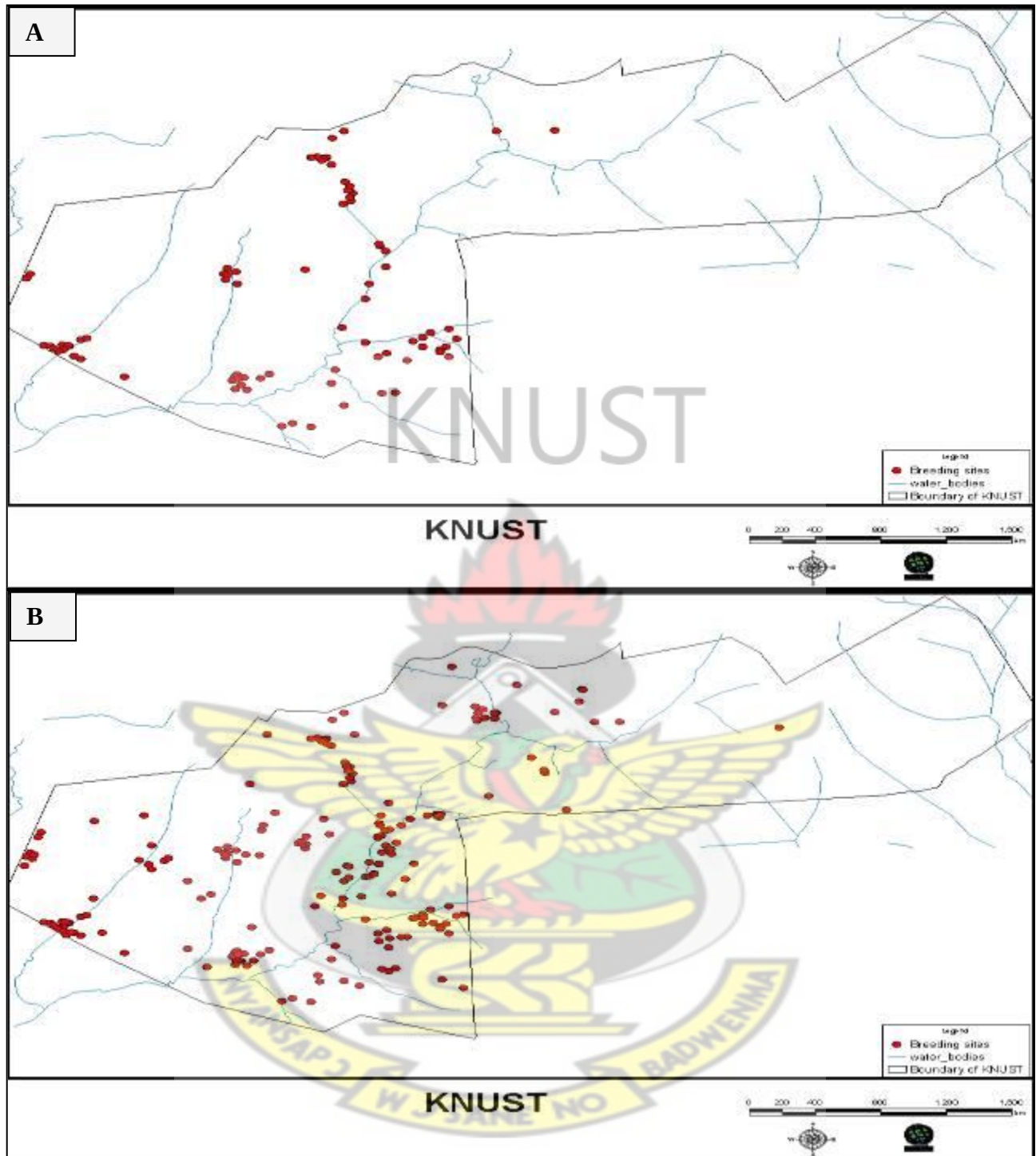


Figure 0.2A comparison of breeding site distribution during the dry and rainy seasons **a)** breeding sites identified during the dry season (October 2007 and February 2008); **b)** breeding sites identified during the rainy season (March 2008 to September 2008) (Map Created with ArcView 9.1).

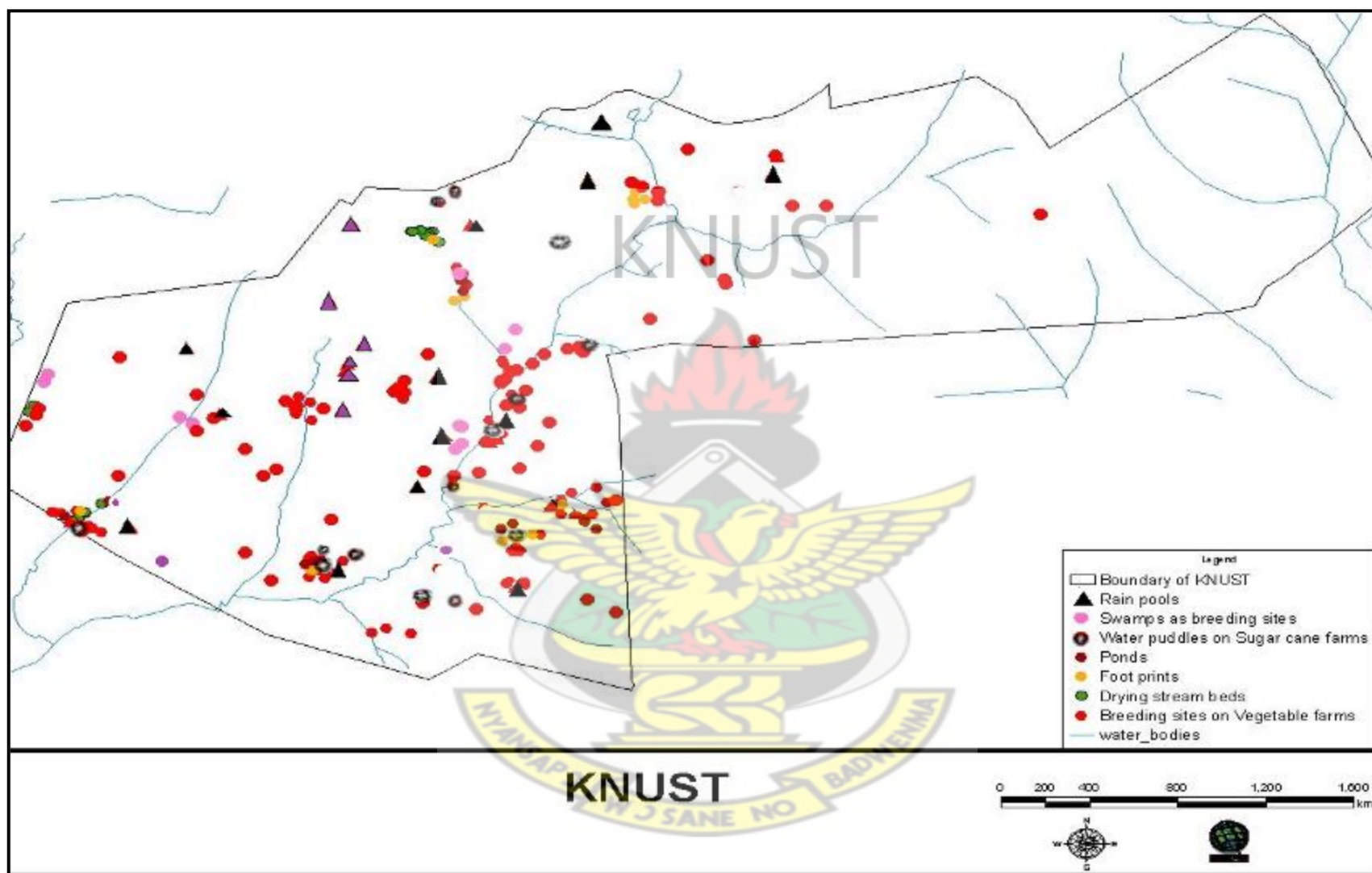


Figure 0.3: Map of KNUST showing the different types of breeding sites identified between October 2007 and October 2008. (Map Created with ArcView 9.1)

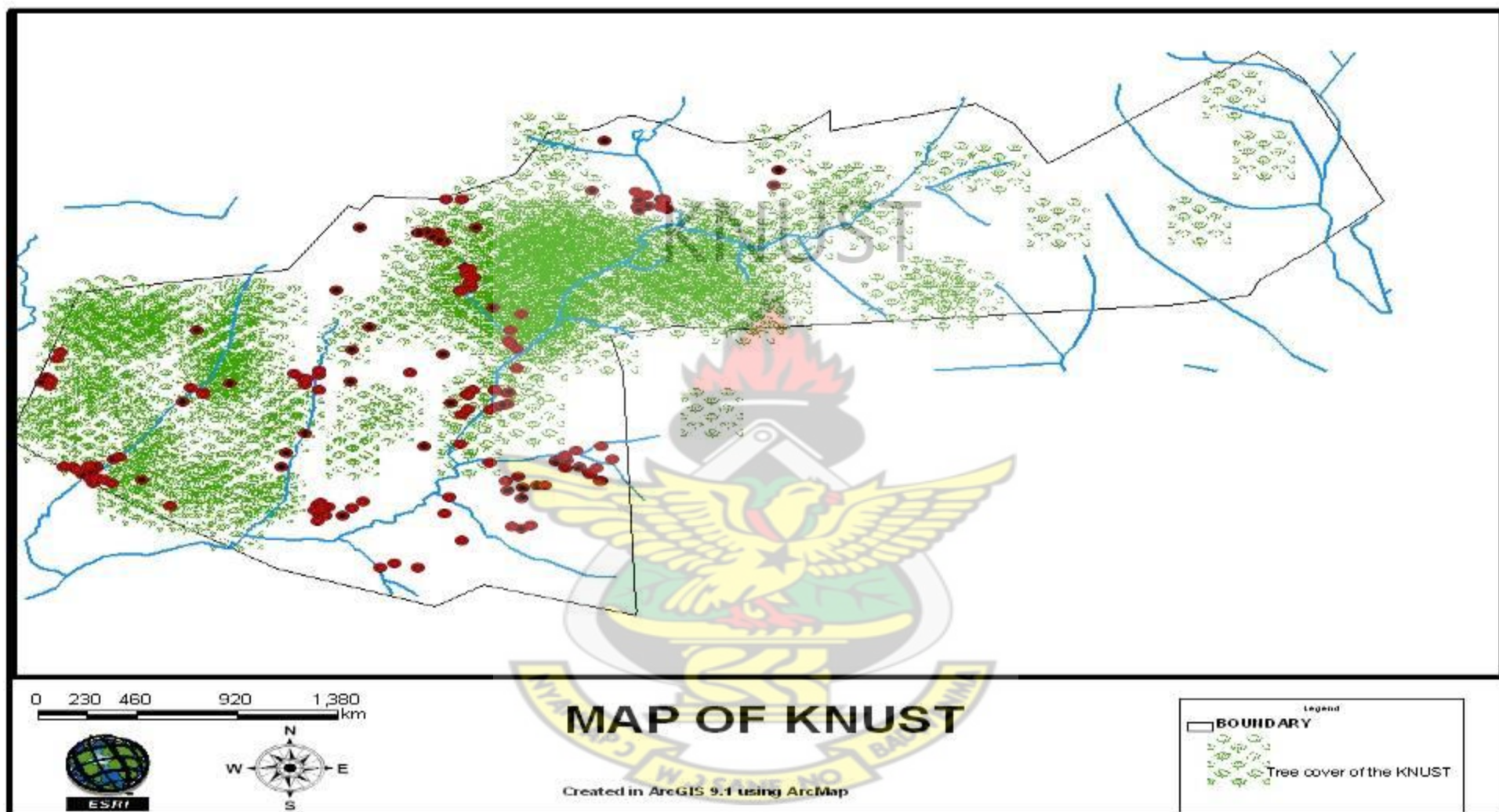


Figure 0.4: Map of KNUST showing *Anopheles* breeding site distribution and the tree cover of KNUST. (Map Created with ArcView 9.1)



Figure 0.5: A dug out well on a vegetable farm which served as a breeding site for *Anopheles* spp.



Figure 0.6: Conduits of the furrow system between raised beds with vegetables



Figure 0.7: Human foot prints (F) in the channels between raised vegetable beds which served as breeding sites for *Anopheles* spp larvae.



Figure 0.8: Drainage channels on a Sugar cane farm which served as breeding site for *Anopheles* spp.

4.1.1.3 *Anopheles* Larval Survey

Anopheline larvae were collected from conduits on vegetable farms (Fig. 4.6), edges of streams, drainage channels on sugar cane farms (Fig. 4.8), water puddles on drying stream-beds, ponds/dugout wells (Fig. 4.5), swamps, temporary pools such as rain pools and human foot prints (Fig. 4.7).

A range of factors that could affect the production of larvae and therefore explain variability in densities of larvae were observed (Table 4.1). Surface temperature appeared to have the greatest effect on anopheline larval density.

A total of 159 breeding sites were identified and geo-referenced during the larval surveys. Of this number, 61 (38.4%) were temporary breeding sites whereas 98 (61.60%) were permanent breeding sites such as, ponds, swamps, edges of streams, conduits on vegetable farms for irrigation. A total of 1632 *Anopheline* larvae were sampled from both temporal and permanent breeding sites. Of The permanent breeding sites contributed, 1287 (i.e. 78.9%) of the total number ($n = 1632$) of larvae collected (Table 4.1).

The breeding sites that were characterised by clear and shallow water produced higher densities of anopheline larvae. For example, of the total number ($n = 1525$) of adult mosquitoes bred from larval collections 1479 (96.9%) were identified as *Anopheles* spp. It was also observed that the densities of adult *A. gambiae* complex mosquitoes bred from larval collections in the rainy season were higher than densities recorded for the dry season. In the rainy season for example 579 adult *A. gambiae* complex were bred from larval collections from breeding sites identified within the faculty area, compared to 302 adult *A. gambiae* complex mosquitoes that emerged during the dry season (Table 4.2).

Table 0.1: Type of breeding sites identified within the different study sites on the KNUST campus between October 2007 - October 2008

Type of Breeding Site	ZONE A		ZONE B		Total Number of Breeding Sties	Total number of larvae collected
	Faculty	Lecturer's residence	Hall of Residence	Hall 6		
Temporary Breeding Site						
Rain pools	11	4	7	5	61 (38.4%)	345(21.1%)
Foot prints	8	2	3	4		
Water puddles on drying riverbeds	0	8	9	0		
Permanent Breeding Site						
Conduits on vegetable farms	32	13	4	0	98(61.6%)	1287 (78.9%)
Ponds	7	2	6	0		
Swamps	6	5	2			
Edges of streams	5	0	2	3		
Drainage Channels/ puddles on sugar cane farms	0	7	0	4		
	69	41	33	16	159	1632

Table 0.2: Seasonal densities of adult *Anopheles* spp bred from larval collections from the breeding sites within the study area.

Zone	Number of Emerged Adults female <i>Anopheles</i> spp		Total Emerged <i>Anopheles</i> spp
	Dry Season	Rainy Season	
A	408 (68.3%)	579 (65.7%)	987 (66.7%)
B	190 (31.7%)	302 (34.3%)	492 (33.2%)

Zone A = Faculty area =, Zone B =Residential Area;

* = reared mosquitoes from water samples collected from the breeding sites

Table 4.3 summarizes the contribution of the various study sites to the mosquito densities within the area. Zone A had the highest number of breeding sites and contributed a greater percentage (66.7%) to anophelines larval densities within the study area. In contrast the Zone B contributed relatively lower number (i.e. 33.2%) of breeding sites, though it covers a wider zone of the study area.

The different breeding sites identified were also assessed for their productivity. Significant difference ($F = 25.45$, $P < 0.001$) in habitat productivity was observed. The most productive habitats occurred on farm lands, and the predominant habitat type on these farmlands were mostly conduits or channels for irrigation purposes or drainage (see Table 4.3. Figure 4.3 shows the contribution of each habitat type to larval densities. Water samples from conduits or channels on irrigated farm within the faculty contributed 531 adult *Anopheles* spp. Other habitat types identified within the faculty, such as dugout ponds and wells also produced a relatively high densities compared to ponds or dugout wells found within zone B. Zone A produced the highest number of breeding sites compared to zone B (Table 4.3).

Table 0.3: Distribution of types of breeding sites/habitat and their contribution to larval densities within the study sites on the KNUST campus from October 2007- October 2008.

Type of Breeding site/ Habitat	ZONE A				ZONE B				Total
	Faculty		Lecturer's Residence		Halls of Residence		Junior Staff Residence		
	#	n	#	n	#	n	#	n	
Conduits on vegetable farms	32	531	13	95	4	84	0	0	710 (48.0%)
Rain pools	11	64	4	19	7	33	5	21	137 (9.2%)
Foot prints	8	31	3	12	2	19	4	13	75 (5%)
Ponds	7	118	2	39	6	51	0	0	208 (14.1%)
Water puddles on drying riverbeds	0	0	8	55	9	36	0	0	91 (6.2%)
Swamps	6	51	5	28	2	16	0	0	95 (6.4%)
Edges of streams	5	56	0	0	2	27	3	23	106 (7.2%)
Drainage Channels on sugar cane farms	0	0	7	57	0	0	4	0	57 (3.9%)
	69	851	42	305	32	266	16	57	

= Number of breeding sites for each locality/selected study site; n = total number of anopheline larvae collected

Total number of adult *Anopheles* spp bred from larval collections = **1479**

Table 4.4 shows temperature and pH of the water samples from the breeding sites. Surface temperature appeared to be an important characteristic of the larval habitats that played a role in determining the level of productivity of the habitat, since larval density was positively correlated to change in water temperature ($r = 0.689$, $P\text{-Value} = 0.059$). Most conduits on the vegetable farms were exposed to direct sunlight resulting in the relatively high water surface temperature (Table 4.4). High surface temperatures possibly favoured egg development, and larval density and could account for the high anopheline larval density collected from the conduits (48.0% of total larval density). It was also found that pH did not have much significant influence on larval densities ($r = -0.244$, $P\text{-Value} = 0.560$); a weak negative relationship existed (Table 4.4) between pH and larval densities.

Table 0.4: Temperature and pH measured from the different anopheline breeding sites identified on the KNUST campus between October 2007 – October 2008.

Type of Breeding Site/Habitat	Mean Temp. $^{\circ}\text{C}$	Mean pH
Foot prints	28.1 ± 0.11	7.31 ± 0.34
Ponds	26.9 ± 0.28	7.58 ± 0.12
Rain pools	25.9 ± 0.18	7.46 ± 0.31
Edges of streams	24.6 ± 0.25	7.18 ± 0.28
Swamps	26.1 ± 0.15	7.78 ± 0.16
Conduits on vegetable farms	27.2 ± 0.31	6.89 ± 0.53
Water puddles on drying riverbeds	26.6 ± 0.48	7.24 ± 0.20
Drainage Channels on sugar cane farms	26.3 ± 0.34	6.64 ± 0.27

In some breeding sites/ habitat tadpoles, dragonflies, and water bugs, were also found and most often very few mosquito larvae or no mosquito larvae were present when they were found.

4.1.2 Number of Adult female *Anopheles* caught biting humans on the KNUST

A total of 2461 adult mosquitoes comprised of 843 *Anopheles* spp and 1,618 Culicines were collected over the 12 months study period (from October 2007 to October 2008).

Relatively fewer numbers of mosquitoes were caught for the dry season (November and February) from all points of collection (Figure 4.9). Low numbers of adult mosquitoes were caught in February. However, the numbers increased from March to April. The highest mosquito density occurred during the months of the rainy period (March 2008-October 2008).

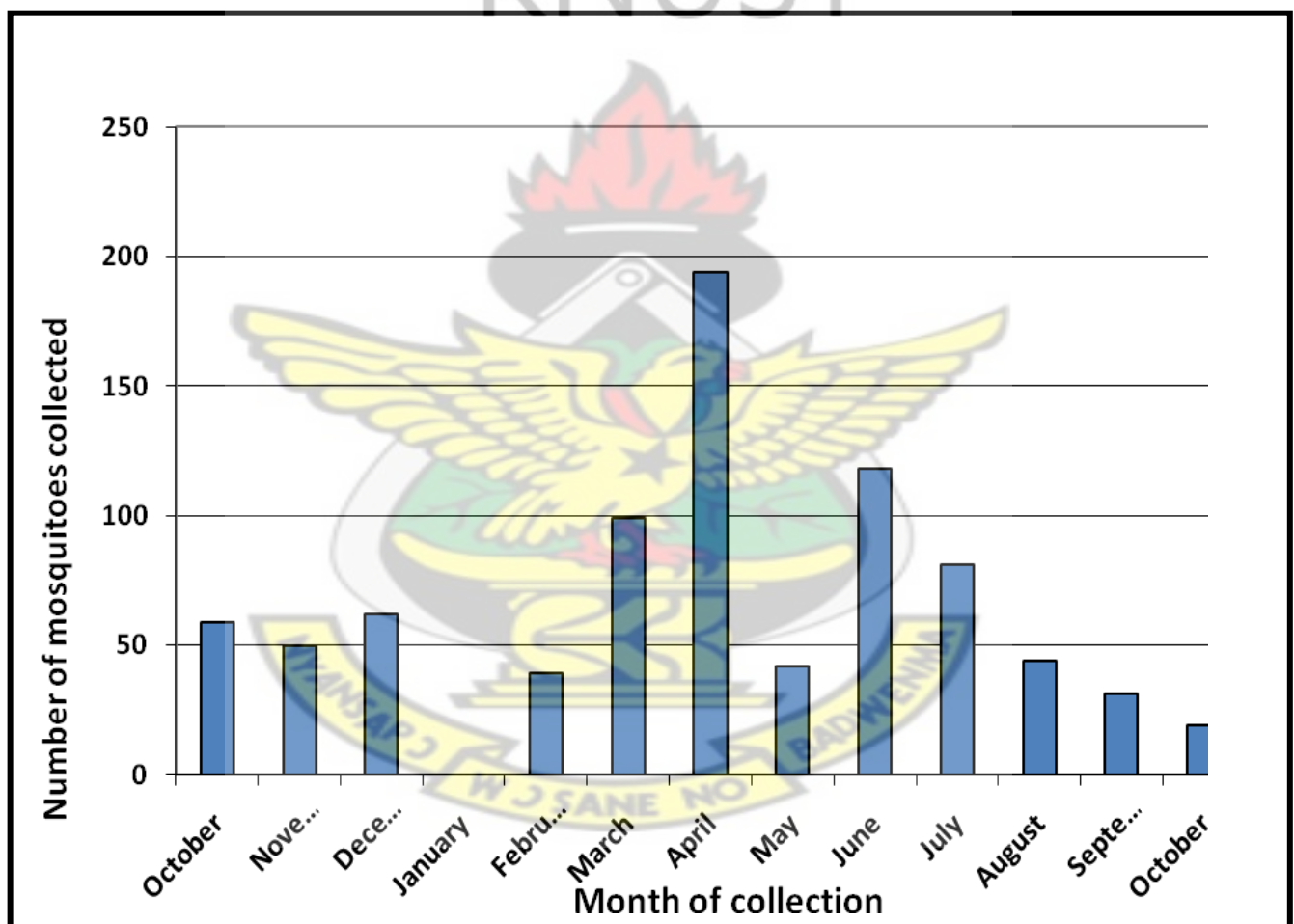


Figure 0.9: Number of female *Anopheles* spp caught by the Human Landing Collection (HLC) method from the four study sites on the KNUST campus for the dry and rainy seasons.

It was observed from figure 4.10 that, the anopheline biting density or activity was higher within the faculty area, compared to the three other study sites, from October 2007 – October 2008. The junior staff residence contributed the least number of *Anopheles* spp. Figure 4.10 shows that the anopheline biting was much greater in the rainy season compared to biting in the dry season in all the study sites.

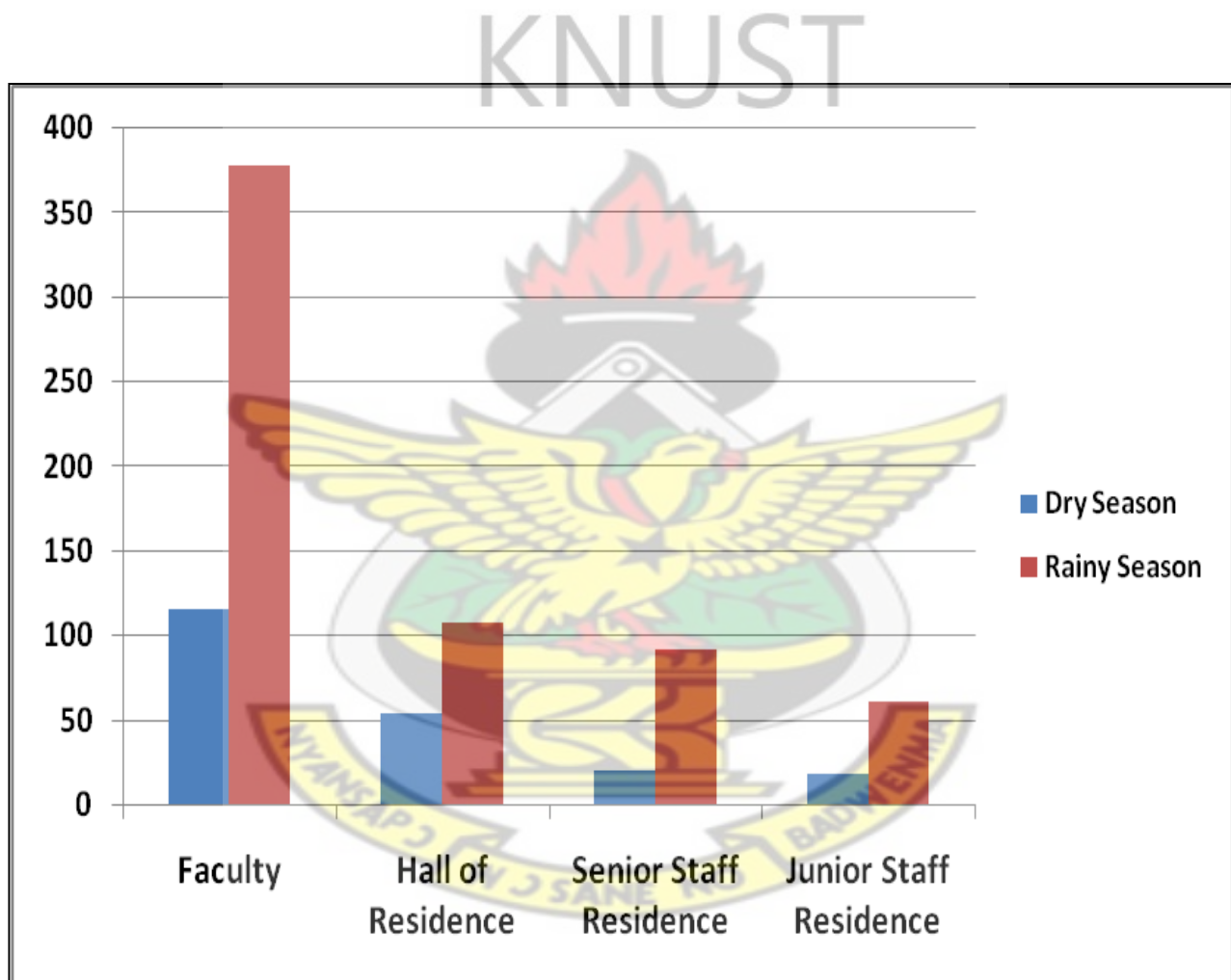


Figure 0.10: Monthly density of female *Anopheles* spp caught using the Human Landing Collections (HLC) method from the study sites on the KNUST campus from October 2007- October 2008.

The lowest adult anopheline density, were recorded during the dry season (November 2007 and February 2008), whereas the highest abundance was also recorded between March and August 2008 which also coincided with the presence of rains (Fig. 4.11).

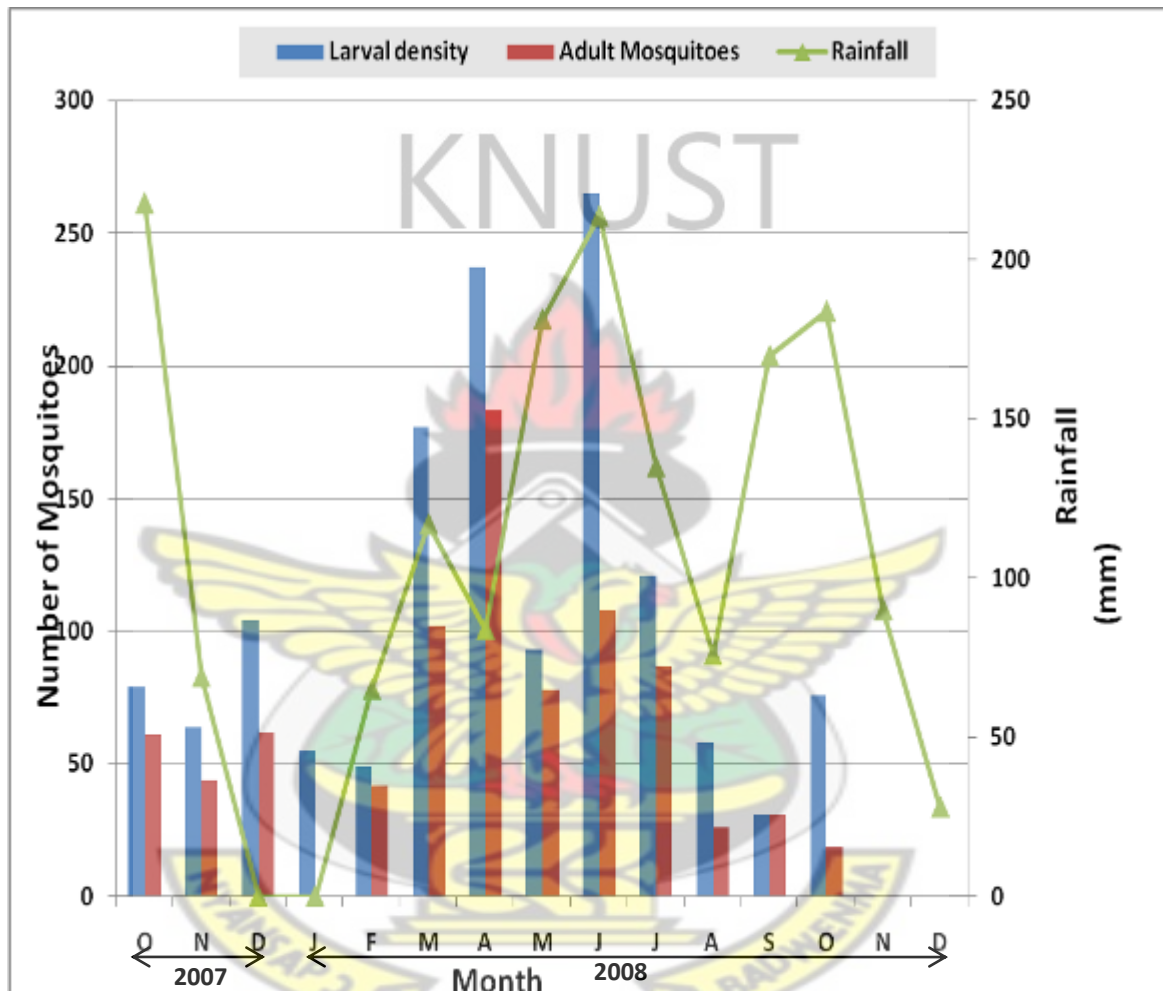


Figure 0.11: Monthly Rainfall recorded from October 2007 to December 2008 compared with larval densities and adult *Anopheles* females collected (by HLC method) from the KNUST campus BETWEEN October 2007 and October 2008. (Rainfall measurements were recorded at Kumasi Centre for Collaborative Research, KCCR)

Three *Anopheles* species were identified: *Anopheles gambiae* Giles complex, *Anopheles funestus* Giles complex and *Anopheles ziemanni* Grunberg. Of the total number of *Anopheles* spp that was caught, 825 (97.98%) were the *Anopheles gambiae* Giles complex group. *A. funestus* Giles complex also made up 2.01%. Only 1 *Anopheles ziemanni* Grunberg was caught; and it was caught in the dry season from the faculty area.

Table 4.5: Mosquitoes species and the total number of mosquitoes caught from the study sites on the KNUST campus during the Dry Season

Mosquito Spp	Number of Mosquitoes Caught per Site				Total
	Site A	Site B	Site C	Site D	
<i>A. gambiae</i>	102	54	20	18	194
<i>A. funestus</i>	12	0	0	0	12
<i>A. zieamanni</i>	1	0	0	0	1
Culicines	158	48	170	51	427

Site A= Faculty Area, Site B = Hall of Residence Site C = Senior Staff Residence
Site D = Junior Staff Residence (Hall 6)

Table 4.6: Mosquitoes species and the total number of mosquitoes caught from the study sites on the KNUST campus during the Rainy Season

Mosquito spp.	Number of Mosquitoes Caught per Site				Total
	Site A	Site B	Site C	Site D	
<i>Anopheles gambiae</i>	373	106	91	61	631
<i>Anopheles funestus</i>	4	1	0	0	5
Culicines	192	332	261	406	1191

Site A= Faculty Area, Site B = Hall of Residence Site C = Senior Staff Residence
Site D = Junior Staff Residence (Hall 6)

4.1.3 Biting Cycle

The hourly variations of the aggressiveness of the *Anopheles* species caught within the study area from October 2007 to October 2008 are shown in figures 4.12 and 4.13.

Biting activities of the female *Anopheles gambiae* began between 6pm and 7pm, and steadily increased and peaked just after midnight. The major peak of their activity occurred between 1 and 2 am. However the activity of *A. funestus* began after 10 pm. The only *A. ziemanni* that was collected was caught feeding after 10pm. The species varied in their biting behaviour during the two season, the activities of most anophelines caught during the dry season had an extended peak between 1am and 3.30 am. .

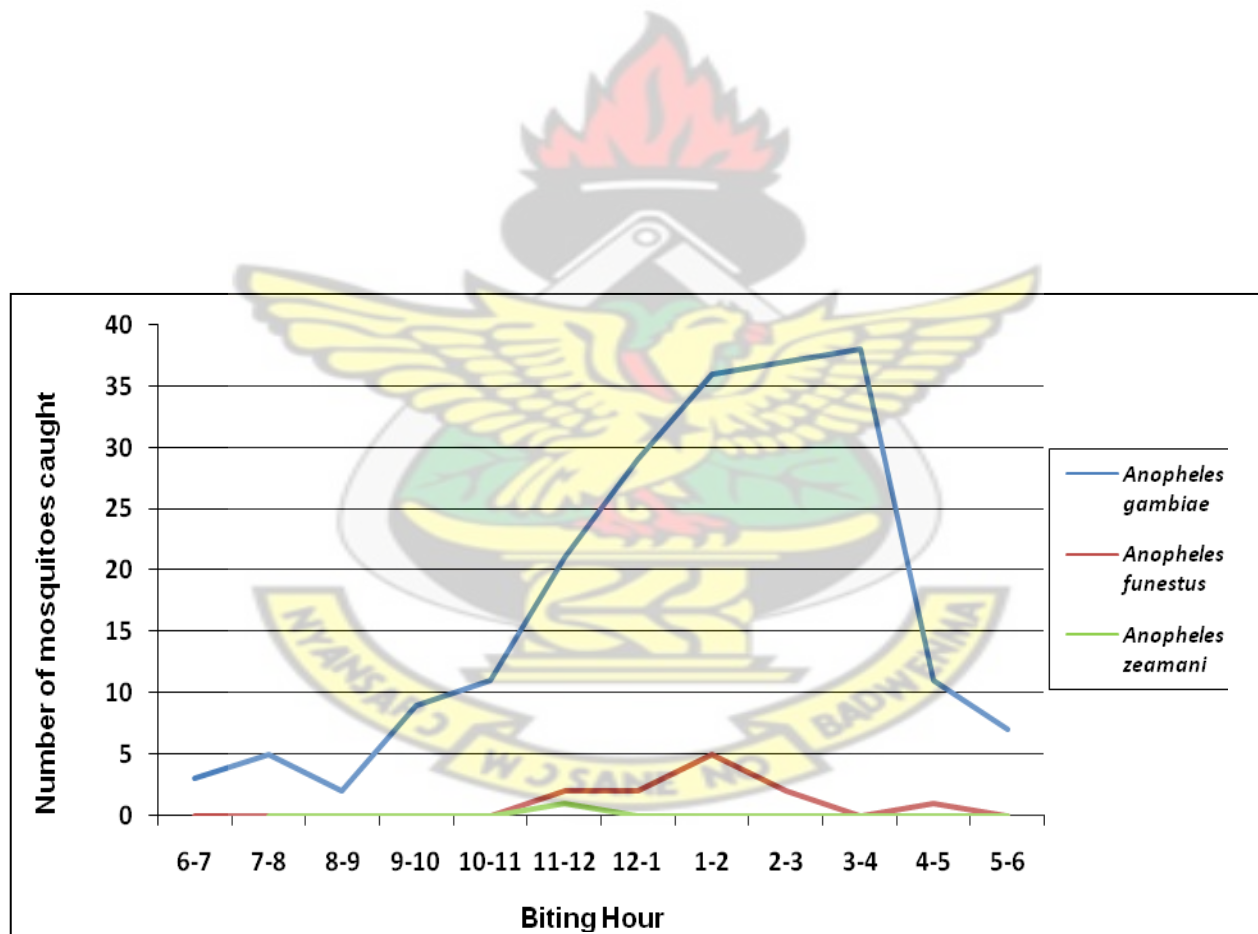


Figure 4.12: Night biting cycle of *Anopheles gambiae*, *A. funestus* and *A. ziemanni* caught from the study sites on the KNUST campus during the Dry season.

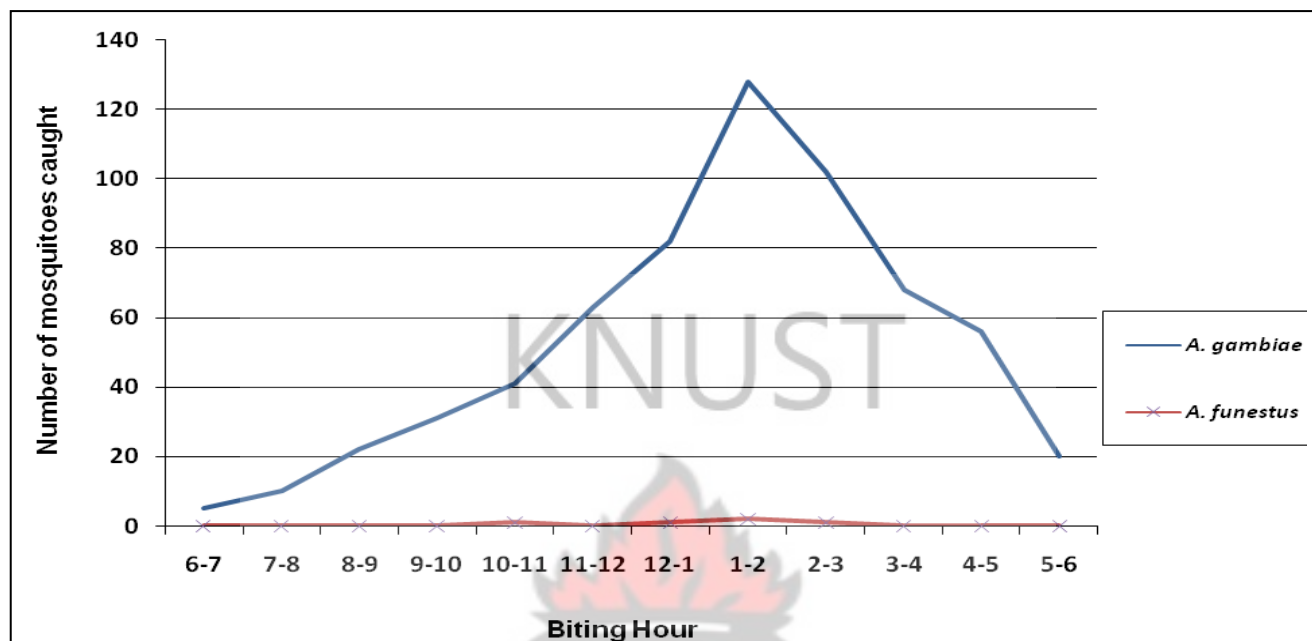


Figure 4.13: Night biting cycle of *Anopheles gambiae* and *Anopheles funestus* caught from the study sites on the KNUST campus during the Rainy season.

4.1.4 Man Biting Rate

The man biting rate (MBR); which is the average number of bites per man per night by vector species (WHO/CDS/CPE/SMT, 2002), was determined directly from the human landing collections. The aggressiveness of *A. gambiae* as a malaria vector was once again evidenced by higher man biting rate (MBR) than the other vectors observed for both the dry and rainy seasons. A total of 32 man nights of collection from the four study sites during the dry season yielded 194 *A. gambiae*, 12 *A. funestus* and 1 *A. ziamanni*. 64 man nights in the rainy season also yielded 631 *A. gambiae* and 5 *An. funestus* with a man biting rate of 9.86 and 0.08 bites/man/night (b/m/n) respectively (table 4.7). A high biting rate of 9.86 b/m/n was recorded in the rainy season for *A. gambiae* (table 4.8) compared to 6.60 b/m/n recorded during the dry season.

Table 4.7: Man Biting Rates of *Anopheles* species caught on the KNUST campus during the Dry season

<i>Anopheles</i> species	Number of Mosquitoes caught	b/m/n
<i>A. gambiae</i>	194	6.06
<i>A. funestus</i>	12	0.38
<i>A. ziamanni</i>	1	0.03

b/m/n = bites/man/night

Table 4.8: Man Biting Rates of *Anopheles* species caught on the KNUST campus during Rainy seasons

<i>Anopheles</i> species	Number of Mosquitoes caught	b/m/n
<i>A. gambiae</i>	631	9.86
<i>A. funestus</i>	5	0.08

b/m/n = bites/man/night

Biting rates were consistently higher within the faculty area throughout the year. In the dry season it recorded a high of 3.69b/m/n whereas the other collection sites; Hall of Residence, Senior Staff Residence and the Junior Staff residence recorded 1.69b/m/n, 0.63b/m/n and 0.56b/m/n respectively (see Table 4.9). Of the 4 collection sites, the faculty area still recorded the highest biting rate 5.89b/m/n in the rainy season (see Table 4.9).

Table 4.9: Mosquito distribution and human landing rates (number of mosquitoes per man per night of activity) at the different study sites.

Study Sites [†]	Number of Mosquitoes caught		Man Biting Rate(b/m/n)*	
	Dry Season	Rainy Season	Dry Season	Rainy Season
SITE A	118	377	3.69	5.89
SITE B	54	104	1.69	1.63
SITE C	20	91	0.63	1.42
SITE D	18	61	0.56	0.95

[†] Site A= Faculty Area, Site B = Hall of Residence, Site C = Senior Staff Residence
Site D = Junior Staff Residence (Hall 6)

*= 32 man nights during the dry season and 64 man nights during the rainy season,

4.1.5 Infection Rate amongst *Anopheles* species collected

Anopheles gambiae was the only *Anopheles* species that was infected with the sporozoite. No sporozoites were seen in the salivary glands of the other two species; *A. funestus* and *A. ziamanni*. Of the 728 *Anopheles* mosquitoes that were dissected, sporozoites were seen in the salivary glands of 5 *Anopheles gambiae* mosquitoes. Two (2) sporozoite infections were observed in mosquitoes from the faculty area in the dry season, whereas 3 sporozoite infections were observed in the rainy season (Table 4.10). Sporozoites in salivary glands of *A. gambiae* were observed in one month of the dry season (December) coinciding with the examination period on KNUST campus. In the rainy season however infections were observed in two months of the rainy season; 2 infections in March and 1 infection in June. The sporozoite index calculated were 1.01% and 0.57% for the dry and rainy seasons respectively.

The distribution of sporozoite positive bites of *A. gambiae* s.l. by hours showed that 1 inoculation occurred in the early hours of the night (8-9 pm), whereas 4 infective bites were received after midnight (12 – 4 am); which also coincided with the peak of mosquito activity (figure 4.14).

Oocysts infections were also seen in dissected mosquitoes, and included 1 and 6 infections in the dry and rainy months respectively (table 4.10). All mosquitoes with oocysts infection were caught feeding in the early hours of the morning (i.e. 3am and 4am).

KNUST



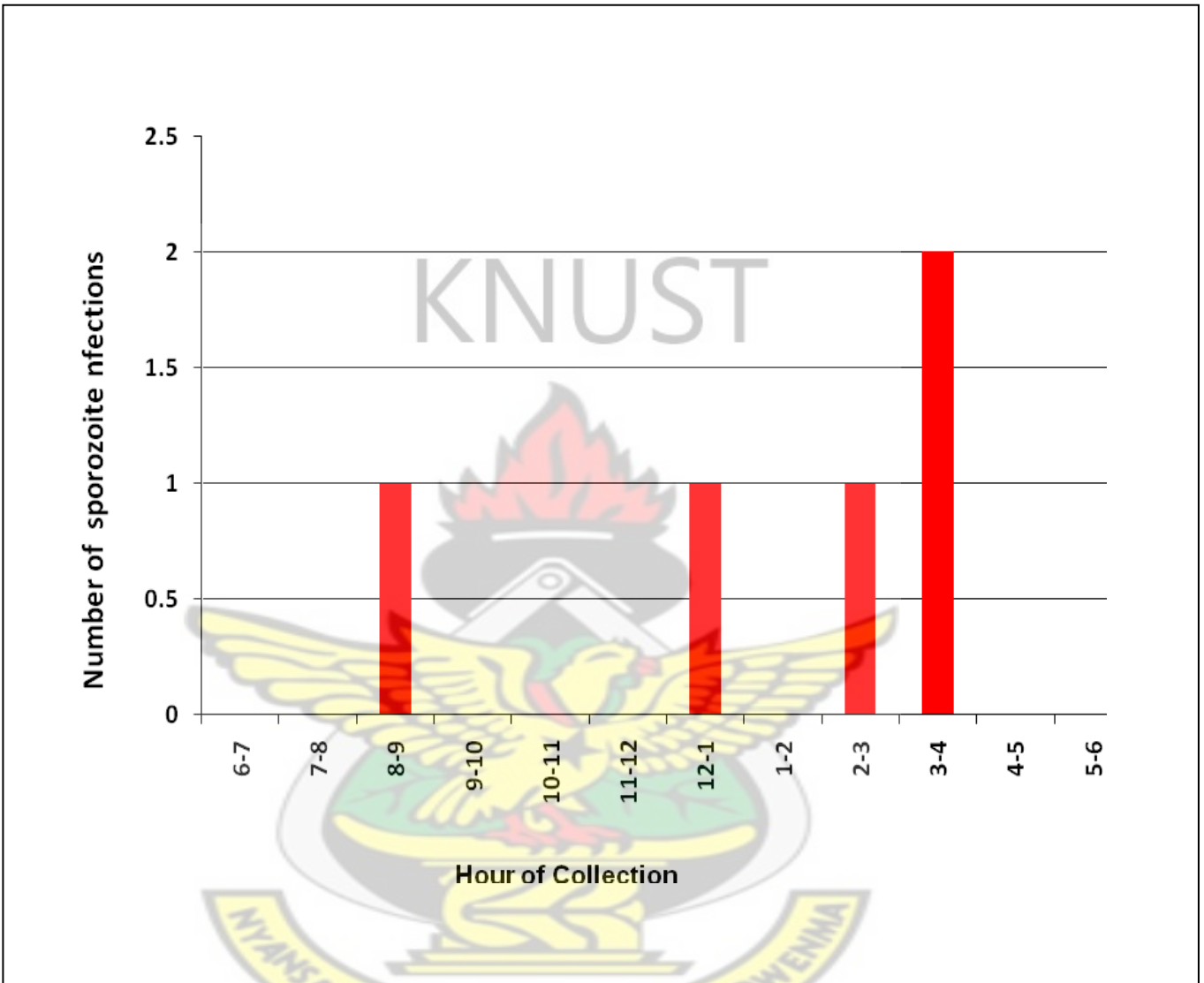


Figure 4.14: Distribution of infective bites *A. gambiae* experienced by collectors during the hours of the night on the KNUST campus between October 2007 and October 2008.

Table 4.10: Number of Sporozoite and Oocysts Infection observed after dissection of female *Anopheles* spp caught during the study period

	Number of Mosquitoes positive with Sporozoite		Number of Mosquitoes positive with oocysts	
	Dry season	Rainy Season	Dry season	Rainy Season
SITE A	2	2	1	2
SITE B	0	0	0	0
SITE C	0	0	0	0
SITE D	0	1	0	4

Site A= Faculty Area, Site B = Hall of Residence Site C = Senior Staff Residence

Site D = Junior Staff Residence (Hall 6)

4.1.6 Entomological Inoculation Rate

The entomological inoculation rate, defined as the mean number of infective bites received by a person within a period (WHO/CDS/CPE/SMT, 2002), was calculated as the product of the man biting rate and the sporozoite rate (sporozoite index). The average sporozoite inoculation rate for *A. gambiae* during the study period was 0.059 infective bites per man per night and a monthly risk of approximately 2 infective bites per man in a month (table 4.11). A slight variation in the risk of malaria transmission was observed between the two seasons. 0.061 and 0.056 infective bites per person per night were calculated for the dry and rainy months respectively (table 4.12). During the dryer months of 2007 it was estimated that an individual could receive 1.89 infective bites within the months, while in the rainy months of 2008, the monthly risk was 1.73 infective bites, and an estimated risk of 21.54 infective bites in the year.

Table 4.11 Entomological parameters of malaria transmission recorded for *Anopheles* species caught from the KNUST campus from October 2007 to October 2008

SPECIES	MBR	SPOROZOITE RATE			EIR		
	b/m/n *	Number Dissected	Number +ve	SI (%)	ib/m/n	ib/m/m	ib/m/yr
<i>A. gambiae</i>	8.59	728	5	0.69	0.059	1.84	21.5

* = 96 Man nights , b/m/n = bites/man/night, ib/m/n = infective bites/ man/night ,
ib/m/m = infective bites/ man/month; ib/m/yr = infective bites/ man/year

Table 4.12 Entomological parameters of malaria transmission recorded for *Anopheles gambiae* caught from the KNUST campus for the dry and rainy seasons

SEASON	MBR	SPOROZOITE RATE			EIR		% Parity
	b/m/n *	Number Dissected	Number +ve	SI (%)	ib/m/n	ib/m/m	
Dry	6.06	198	2	1.01	0.061	1.89	82.4
Rainy	9.86	530	3	0.57	0.056	1.73	68.1

*= 32 man nights for the dry season and 64 man nights for the rainy season,
b/m/n = bites/man/night, ib/m/m = infective bites/ man/month; ib/m/yr = infective bites/
man/year

4.1.7 Parity Rates

In order to assess the age and longevity of the vectors identified the proportions of parous females were determined. Seasonal variations in parity rates were observed, with more parous females being caught in the dry season than for the rainy season (Table. 4.13). Dissections of female *Anopheles spp* collected during the dry season showed parous females made up 82.3% (181 of 149) of mosquitoes caught whereas a relatively low percentage of the mosquitoes collected in the rainy season 68.1% (404 of 593) were parous. ($\chi^2=17.352$, df =3; p=0.001)

Table 4.13: Number of parous and nulliparous *Anopheles* mosquitoes dissected between October 2007 and October 2008

Dry Season			% Parity
Period of Study	No dissected	No. of Parous female <i>Anopheles spp</i>	
October '07	54	46	82.3%
November '07	38	29	
December '07	54	44	
February '08	35	30	
	181	149	
Rainy Season			
March '08	95	67	68.1%
April '08	170	106	
May '08	72	58	
June '08	100	71	
July '08	81	57	
August '08	24	16	
September '08	29	17	
October '08	18	12	
	593	404	

'07= 2007; '08 = 2008

4.1.8 Longevity

The longevity of *A. gambiae* affects the likelihood of being bitten by an infective mosquito. Two important factors that affect the longevity of vectors within an area are the expectation of life and the probability of survival (i.e. the probability of surviving 1 day after a blood meal). Calculated as:

$$\text{Probability of Survival, } p = n^{\sqrt{\text{Proportion of Parous Anopheles}}}$$

$$n = \text{days of blood meal interval}$$

$$\text{Expectation of life} = \frac{1}{\ln p}$$

The results (Table 4.14) indicate that the expectation of life for *Anopheles* species caught on campus between October 2007 and October 2008 varied. If a 3 –day blood meal interval is assumed, *Anopheles* species caught on campus within the study period would have a 0.937 probability of survival, and a 15.4 days expectation of life during the dry season compared to 0.825 probability of survival and an approximately 8days expectation of life in the rainy season. The expectation of life of *Anopheles* species caught on campus was thus higher in the dry season than in the rainy season, indicating that few anopheline collected had the higher probability of transmitting the disease than in the rainy season

Table 4.14: Survival of female *Anopheles* mosquitoes after a blood meal

		PROBABILITY OF SURVIVAL (<i>p</i>)		EXPECTATION OF LIFE (Days)	
	% Parous	Intervals Between Blood Meals		Intervals Between Blood Meals	
		1 day	3 days	1 day	3 days
Dry	82.3	0.907	0.937	10.2	15.4
Rainy	68.1	0.825	0.879	5.2	7.6

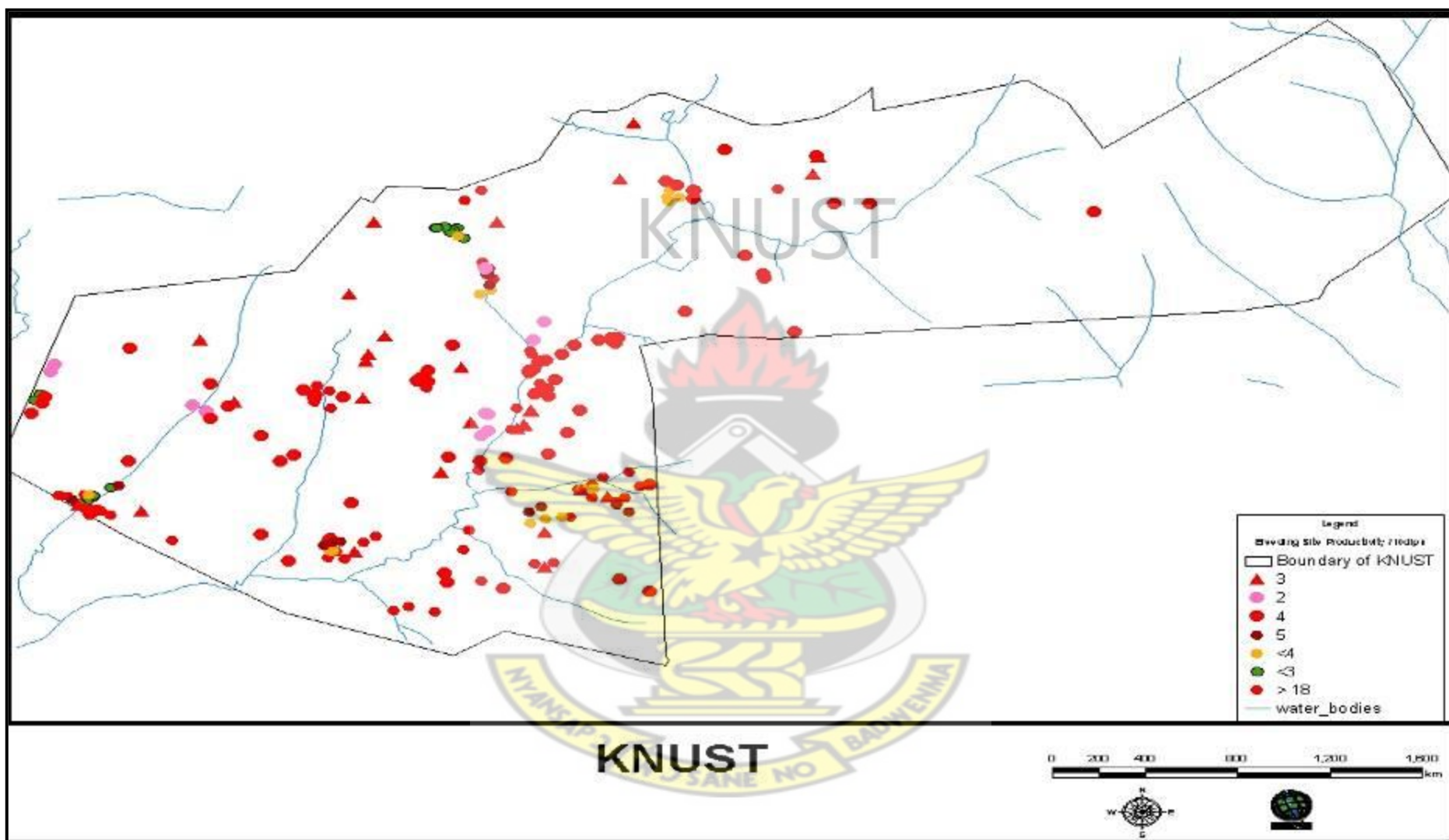


Figure 4. 15: Map of KNUST showing productivity of anopheline larval habitat identified between October 2007 and October 2008 on the KNUST campus.

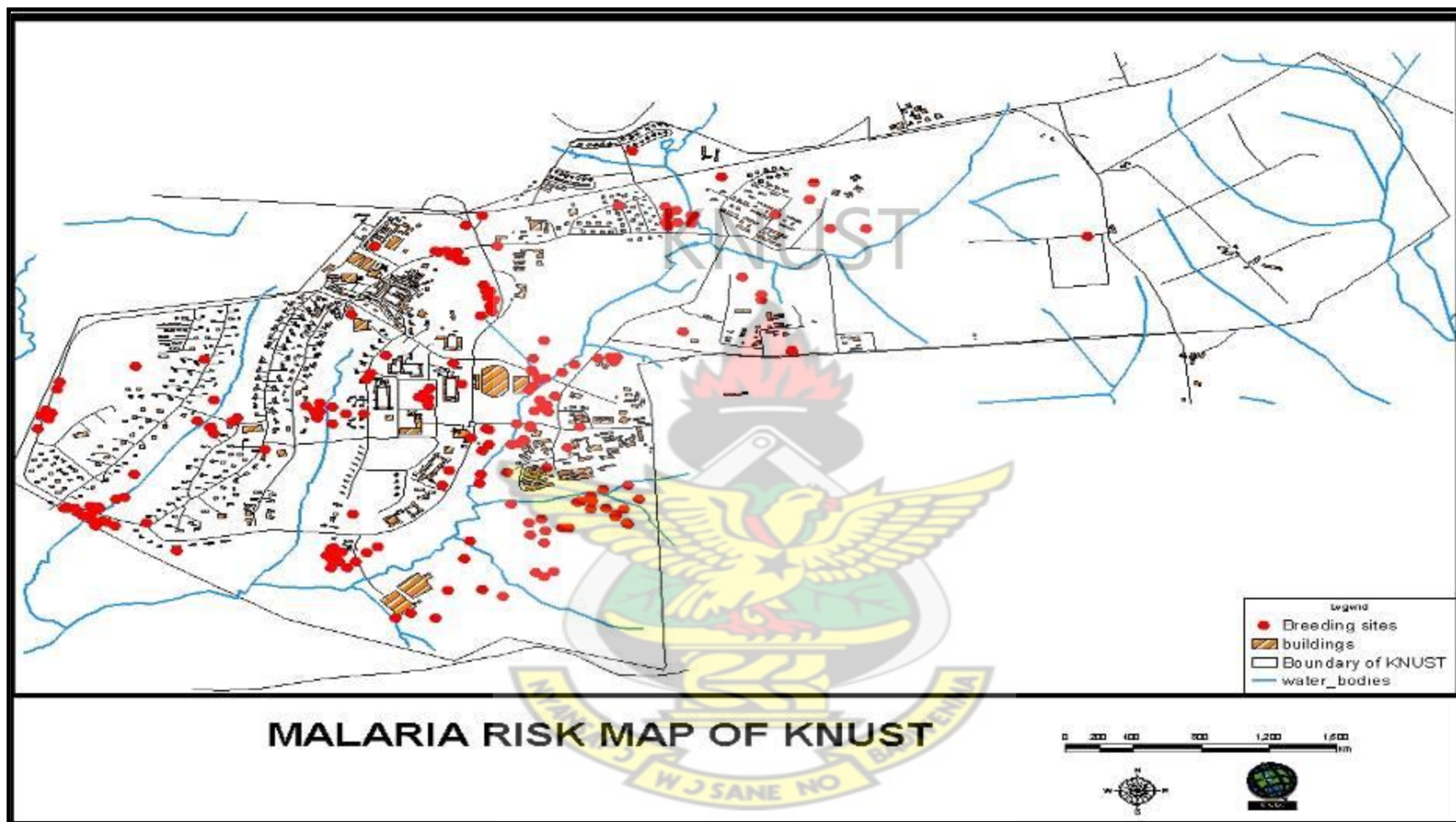


Figure 4.16: Malaria risk map of KNUST showing the distribution of breeding sites amongst the residential and faculty areas

4.2.0 Knowledge Attitude and Practices on Malaria

4.2.1 Demographic Data

Six hundred and twenty (620) questionnaires were administered. They comprised 402 (64.8%) males and 218 (35.2%) female respondents. The ages of respondents ranged from 18 to 60 years, with median age of 23 years (table 4.15).

The student population formed 67.7% (420 out of 620) of respondents interviewed (table 4.16). Members of staff (teaching as well as non-teaching personnel such as administrators, security personnel, librarians, manual workers etc.) formed 21.9%. The remaining 10.4% respondents were self employed (mostly traders).

Table 4.15: Percentage distribution of respondents' age groups by gender

Age group	Female (n=218 ;64.8%)	Male (n=402; 35.2%)
15-19	53.8	12.5
20-34	38.5	79.1
35-44	2.6	5.6
45 above	5.1	2.8

Table 4.16: Residence of respondents and occupation

Residence of respondents	KNUST		
	Students	Staff	Other Workers (Self employed)
<i>Residence of Respondent on Campus</i>			
Hall of Residence	300		
Lecturer's Residence	9	14	3
Junior Staff Quarters	4	65	13
Security Barracks		26	15
<i>Immediate surroundings of campus</i>			
	107	45	36
Total	420	136	64
	(67.7%)	(21.9%)	(10.4 %)

In assessing the educational level attained by respondents, the following observations were made: four hundred and ninety three (495) representing 79.8% had completed tertiary education or were in the tertiary institution, 76 (12.3%) had secondary education, 6.4% had only completed basic education while 9 (i.e. 1.5%) had never experienced any form of formal education (table 4.17). It was also observed that there was significant difference in the educational level between males and females, with more males being educated beyond the primary level ($\chi^2 = 15.15$; d.f. = 3; $p = 0.002$).

Table 4.17: Percentage distribution of respondents' education level groups by gender

Educational Background	Male		Female		Total	%
	(n=402)	%	(n=218)	%		
Never attended school	3	0.7	6	2.8	9	1.5
Basic	30	7.5	10	4.6	40	6.4
Some or completed secondary	61	15.2	15	6.9	76	12.3
Tertiary	308	76.6	187	85.8	495	79.8

4.2.2 Perception on Malaria Transmission

Figure 4.17 is a representation of the responses obtained on the perceived causes of malaria. It indicates that 97.4% of the respondents attributed the cause of malaria to mosquitoes. Other non-biological causes mentioned by some respondents were; dirty surroundings, dirty home surroundings, eating too much oil, and long exposure to the sun. Only one person did not know what caused malaria, 68.2% however, associated the cause of malaria with mosquito bites alone, whereas 29.4% associated it with mosquito bites and dirt.

The results revealed that education beyond primary school level increased the probability of respondents attributing the cause of malaria to mosquito bites. Respondents educated beyond primary school level were less likely to believe that long exposure to the sun and eating too much oil was responsible for catching malaria. No significant differences were observed in responses with respect to gender (table 4.18).



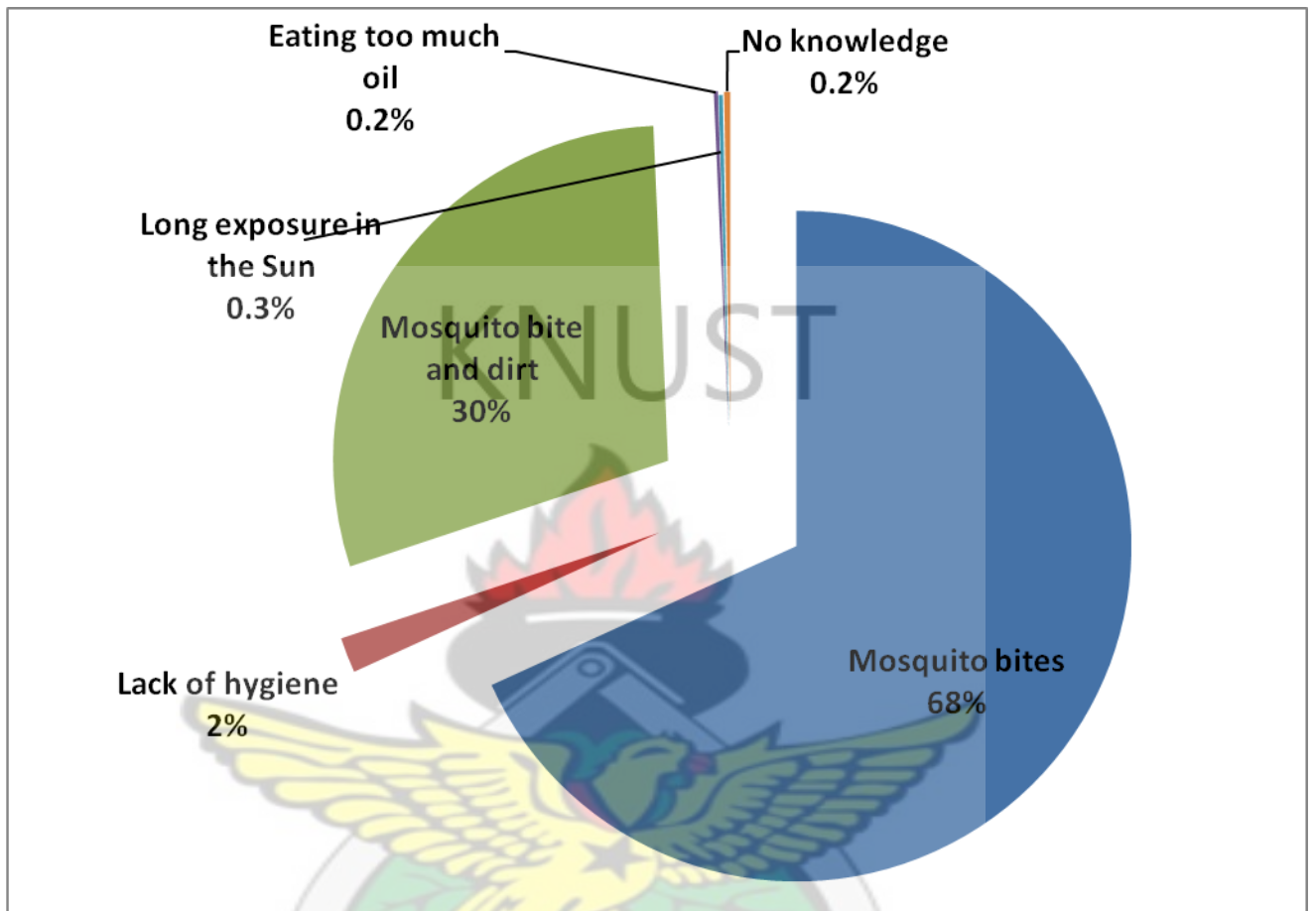


Figure 4.17: Percentage distribution of responses to perceived causes of malaria

Table 4.18: Impact of Educational background and Gender on correct knowledge and misconceptions on what causes malaria

Education	P-value	Odds Ratio	95% CI	
			Lower	Upper
None	0.501	0.43	5.13	0.04
Basic level	0.195	0.08	0.00	3.54
Secondary level	0.175	4.57	0.51	41.09
Tertiary level	0.232	0.94	0.84	1.04
Gender				
Males	0.123	0.60	0.32	1.15
Females	0.116	2.82	0.77	10.27

4.2.3 Malaria Protection and Control Practices

Respondent's knowledge on Protection practices was high. The practices mentioned included both personal protection practices as well as environmental management practices. Personal protection practices mentioned included use of ITN, Insecticide Spraying, chemoprophylaxis, use of mosquito repellents, wearing of protective clothing, window/door netting, periodic check-up, and burning mosquito coils at night. Other respondents either spread powdered camphor, or burned orange peels in their rooms at night. One person admitted not knowing what to do to prevent malaria infection and attack. Environmental management practices (cleanliness) mentioned by some respondents included proper waste disposal practices, clearing gutters and draining stagnant waters that served as breeding sites and bush clearing.

Varying prevention behaviour was observed amongst respondents. Knowledge about a particular method did not necessarily result in its usage as a preventive method. For example; though 120 respondents knew that Insecticide treated bed-nets (ITN) protected them from malaria 107 respondents used them. Also, of the 83 respondents who mentioned insecticide spray usage as a preventive method only 41 people used it. Chemoprophylaxis as protection measure was used by 16 people out of 43 respondents who knew about it. Similarly, 5 persons mentioned spreading of camphor in the room at night as a personal protection method but 3 would opt for a different protection method. Some respondents used a combination of these methods for protection.

Table 4.19: Personal and environmental protection methods applied by respondents

Prevention Strategies	Method Known	Method Used
ITN	120	107
Insecticide Spraying	83	41
Chemoprophylaxis	79	34
Repellents	71	60
Protective clothing	11	2
Window/Door netting	38	31
Periodic check-up	43	16
Burn Mosquito coils	23	9
Use camphor	5	2
Burn orange peels	1	1
Cleanliness	21	16

4.2.4 Reasons why Respondents Use a Particular Method

Respondents used a particular method because it either had no known side effects; convenience, cost, effectiveness, or believe that it is the approved protection method etc. More than half (58%) of the respondents would use a particular method because they felt it was effective in protecting them from malaria infections and/or attack. Only 1.9% of respondents agreed to use a method because it was an approved method.

Others chose a particular protection strategy or intervention for a combination of reasons; such as, effectiveness and convenience, cost, etc.

Table 4.20 Reasons guiding respondents' selection of a preventive method as a protection method or an intervention strategy.

Reasons	N=617	%
Convenience	37	6.0
Cheaper	49	7.9
Effective	358	58
No special reason	25	4.1
No side effects	19	3.1
Approved method	12	1.9
Two or more Reasons	117	19

4.2.5 Treatment Seeking Behaviour

Respondents were asked what they did when they experienced any of the symptoms (such as a fever) associated with malaria. Four hundred (400) respondents (65%) (Fig. 4.18) indicated that in the case of fever, they bought drugs from pharmaceutical shops for self treatment/medication. Of the number that practiced self medication, 88.3% (353) preferred Orthodox or Ministry of Health approved medication, whereas the remaining 11.7% (47), did not. Five and half percent (5.5%) of males treated their attack with herbal medication as compared to 2.1% for females. Significant difference in treatment seeking behaviour was observed ($\chi^2 = 199.63$; d.f. = 2; $P = 0.000$) amongst the two main

responses; hospital attendance and self medication. Only 35% of respondents attended hospitals in case of a fever. Table 4.21 reveals that most of these respondents that visited the hospital were females (i.e.157) and only 10.2% (63) respondents who visited the hospital for treatment were males.

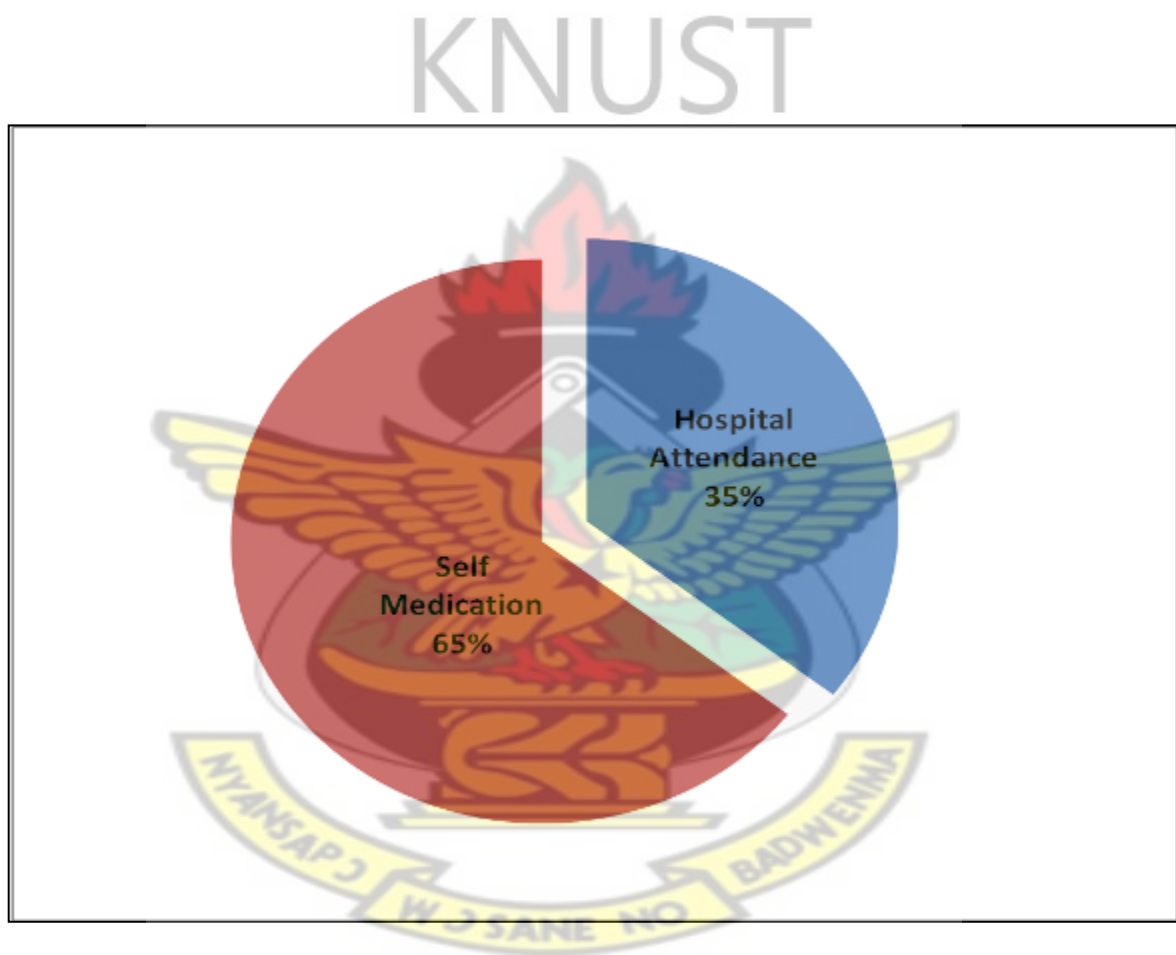


Figure 4.18: Percentage of respondents who practice self medication compared with respondents who visit the hospital for treatment.

Table 4.21: Variations in Respondents' treatment seeking behaviour by gender

Treatment Seeking Behaviour	Females		Males	
	n	%	N	%
<i>Hospital</i>	157	25.3	63	10.2
<i>Self Medication</i>				
- Orthodox Anti-malaria drugs	48	7.7	305	49.2
- Traditional/Herbal Medication	13	2.1	34	5.5



CHAPTER FIVE

5.0 DISCUSSION

This study was conducted primarily to determine the vector species present on campus, their roles in malaria transmission, and to map out areas of high malaria risk based on the entomological indices that were measured. The study sought to understand the knowledge attitudes and practices of inhabitants of the study area towards malaria.

5.1.0 Spatial distribution and types of breeding types

The aquatic habitats found were much varied (Fig. 4.3). *An. gambiae*, breeding in these different habitat types shows the versatility of this species and therefore its important vectorial status in malaria transmission.

Each larval habitat type contributed differently with regard to larval production with higher densities occurring on vegetable farms. This conforms with findings of Afrani et al., (2004), that open space vegetable farms in Kumasi contributed significantly to *Anopheles gambiae* density. In the areas where vegetable farms were abundant, more *Anopheles* larvae were collected. However, ponds were less productive for *Anopheles* larvae. This finding showed that small habitats were more productive for anopheline mosquitoes compared to large larval habitats. The possible explanation as to why *Anopheles* species larvae frequently occurred in puddles on these farms might be that the *Anopheles* females preferentially select small, open habitats for oviposition (Minakawa et al., 1999), or due to the absence larval predation in such temporary habitats (Sunahara, 2002) since most permanent breeding sites that were found with tadpoles and water bugs very few mosquito larvae or none.

Spatial analysis reveal that built up areas have less incidence of breeding, but a greater proportion of the breeding sites were clustered in areas with reduced tree cover due to modification of normal vegetation (Fig. 4.15 and Fig. 4.4). This shows the extent to which some transmission indices of malaria result mainly from human modification of the ecosystem. This accounts for the presence for the abundance of breeding sites in the faculty area. Farmlands and agriculture activities close to water bodies and on most

wetlands are abundant within and close to the faculty area. The land use for construction projects and cultivation of vegetables within zone A exposes the area to direct sunlight, creating optimum warmer micro-climatic condition, which favour the breeding of anophelines. This was evidenced in the variation in temperature recorded for the two study zones. Trape and Zooulani, (1987) also attribute land modifications or transformations to the creation of more man-made breeding grounds for *Anopheles* spp.

5.2.0 Species composition, abundance and distribution of *Anopheles* mosquitoes

The main *Anopheles* species that were caught and identified within the study period, using the morphological keys of Gillies and DeMeillon (1968) and Gillies and Coetzee (1987), were *Anopheles gambiae* Giles complex, *A. funestus* Giles complex and *A. ziemanni* Grunberg.

The unequal distribution of the *Anopheles* species within the area further suggests that, the occurrence of the species truly varies according to macro and micro environmental differences exhibited by different bio-ecological areas, as was found in studies conducted by Keating *et al* (2003). The predominance of *A. gambiae* could be attributed to the adaptability of this species.

The other species (i.e. *A. funestus* complex and *A. ziemanni*) collected occurred in very low densities. All the *A. funestus* caught, were collected at the beginning of the dry season. This is consistent with the observation reported by Appawu and others (1994) that *A. gambiae* breeds principally in temporal pools, abundant during the rainy seasons but dry up at the end of the rains, whereas *A. funestus* typically breeds in more permanent water which is mostly overgrown with vegetation.

The collection of *A. ziemanni* Grunberg mosquito confirms its occurrence over a wide area of Africa from Senegal eastwards to the Sudan and Ethiopia, and southwards to the Congo and Angola, it is predominantly a West African form (Gillies and De Meillon, 1968). However its low numbers could be due to its preference for blood meals other than human, since it has been observed to exhibit a high degree of zoophily (Gillies and De

Meillon, 1968). The proximity of KNUST cattle farm to the campus, could also account for the detection of such a rare species in the mosquitoes collected.

5.3.0 Seasonal abundance of *Anopheles* spp

Breeding of mosquitoes goes on all year round within the KNUST, though at a generally low level in the dry season. In the dry season, however, active breeding sites were maintained in proximity to water bodies as shown in figure 4.2a. The highest vector density recorded during the rainy season could possibly be due to the rains that sustained mosquito breeding sites, providing grounds for the larvae of the vector to develop. The occurrence of such high densities of both malaria and non-malaria vectors is indicative of how much of a nuisance these mosquitoes are to inhabitants of the KNUST and immediate surroundings.

Factors that account for the variation in anopheline densities between the two seasons seasonal and between study sites, as observed in this study ought to be critically looked at in designing intervention measures.

Abundance of the *Anopheles* mosquito species, the longevity of mosquitoes, the propensity and frequency of mosquitoes to bite host, the existence of malaria parasites amongst vector and hosts populations, population immunity as well as social factors like housing conditions and mosquito control measures, have been identified as being among some of the factors that determines transmission of malaria (Devi and Jauhari, 2006).

5.4.0 Transmission indices

The man biting rate (MBR) (bites/man/night) estimates of *Anopheles* species collected, are indicative of the nuisance these vectors pose to the community. In the dry season for example MBR recorded was 6.06 bites/man/night (b/p/n), however it increased to 9.86 b/p/n in the rainy season. The consistent variation in abundance and biting rates between the study sites on the KNUST campus could be explained by the abundance of active breeding sites within the faculty area than there were for other sites on campus within the study period, mostly as a result of the land modification or land use changes. Sharma and Kondrashin (1991) and Molyneux (1998) reported that ecosystem changes and land

transformations influence mosquito ecology and malaria epidemiology. Shililu (2001) also reported that these transformations affect habitat factors such as water turbidity, depth, current, and temperature, which in turn affect variation in larval densities. In this study the most important habitat factor affecting variation in habitat productivity was temperature. Larval density was positively correlated to change in water temperature ($r = 0.689$, $P\text{-Value} = 0.059$). The average water temperature (26.6°C) of breeding habitats found within zone A was slightly higher than the temperature ($26.2^{\circ}\text{C} - 25.9^{\circ}\text{C}$) measured for breeding habitats in the other areas. The findings of Sharma *et al* (1991), Molyneux (1998), and Shililu (2001) explain observation made in this study regarding mosquito abundance and distribution. It was evident that areas of KNUST that had seen immense land transformation and modification for either agriculture or construction purposes harboured abundant *Anopheles* spp breeding sites. Zone A (the faculty area and its surroundings) and parts of zone B (residential area) have seen such transformation over the years (see figure 3.3, figure 4.1 a & 4.1 b, figure 4.4). Figure 4.4 showing the *Anopheles* breeding site distribution and the tree cover of KNUST demonstrates the contribution of these land modifications to the creation of active larval breeding habitats. These activities resulted in the creation of a higher number of larval breeding grounds within zone A (the faculty area and its surroundings) than in zone B. Fewer vegetable farms were present in zone B and also had a more intact tree cover providing a shaded zone. This attribute of zone B seem to have created unfavourable environment for anophelines, and explains the occurrence of the relatively few larval habitats in zone B(residential) compared to zone A (faculty area). This suggests that the initiation of activities that result in land modification on the university campus should correspond with control efforts targeted at elimination of breeding habitats that may be created.

Anopheles gambiae was the only vector species, found infected during the dry and rainy seasons. This confirms that *A. gambiae* continues to be an efficient malaria vector in Ghana; as was found in studies conducted by Appawu *et al.*, (1994) as well as by Afrane *et al* (2004), and other areas of Africa and Madagascar (Jambou *et al.*, 2001).

Considering that the main vector species, *A. gambiae*, was found to breed in different habitat types in the different zones, presents a major challenge to the control of malaria on the KNUST, because under appropriate conditions of temperature and humidity, and possibly the presence of infectious gametocyte pool in the population, malaria transmission could be enhanced by this species.

No sporozoite was detected in its salivary glands of *A. funestus* and *A. ziemanni*. The absence of sporozoite infection in *A. ziemanni* could be compared to finding findings by Vercruysse *et al.*, (1983), that *A. ziemanni* was of no importance in the transmission of malaria, in a work carried out in Senegal. It could also be due to its zoophilic feeding behavior making them less likely to come into contact with infectious gametocyte pools.

Though no sporozoites were detected in the *A. funestus* that were dissected, it has been found to be a very important vector species that sustained malaria transmission in other parts of the country (Appawu *et al.*, 1994).

However the relatively low densities of *A. ziemanni* and *A. funestus* caught within the period makes it difficult to explain their vectorial significance and roles in malaria transmission on the KNUST campus.

It is a well established fact that humans become infected with malaria as a result of their exposure to blood- feeding infective female *Anopheles* mosquitoes. To estimate the intensity of malaria parasite transmission under field conditions, it is a standard practice to determine the entomological inoculation rate EIR (Beier *et al.*, 1999). The EIR calculated represent the intensity of transmission to which the human population within KNUST was potentially exposed. It was estimated that individuals within KNUST could be exposed to 21.5 infective bites in the year. Students and other inhabitants living in settlements such as Ayeduase, close to the faculty area are at a higher risk of malaria infection, since most (4 out of the five) infective bites were recorded from this area. A sporozoite rate of 1.01 in and 0.56 the dry and rainy seasons respectively are relatively lower than findings of 2.6 in the dry season and < 1.9 in the rainy season in the city of Kumasi by Afrani *et al.*, (2004).

In a study on prevalence and intensity of malaria on the KNUST (between November 2007 and April 2008) a prevalence of 18.4% was recorded for Ayeduase a settlement close to faculty compared to an average prevalence of 9% from all halls on campus (unpublished dissertation by Arthur, 2008). The highest prevalence rate (33%) was recorded from Junior staff residence (Hall 6) in the study on malaria prevalence. However the entomological study revealed that the Hall 6 area had the lowest human biting rates (0.56 and 0.95 b/m/n for the dry and rainy seasons respectively) and no sporozoite infection. Comparing these parameters; anopheline biting rates and infection to parasite prevalence, within the Junior staff residence (Hall 6); a negative association is revealed. However, relatively high oocysts infections were recorded, indicating the existence of an infectious gametocyte pool amongst inhabitants of the Junior staff residence. The low vector abundance and the prevalence of malaria in this area (Junior staff residence) confirms the findings of Rosenberg *et al.*, (1990) that showed that a small population of vector species within an area could maintain a focally high level of malaria prevalence. Sanitary conditions around some houses within the Hall 6 area were relatively poor. This observation as well as the relatively lower temperatures due to high amount of shade as a result of high tree covers within the area, possibly account for the low abundance of the vectors within Junior Staff Residence. Bruce-Chwatt (1985) reported that the poor sanitary conditions lead to the contamination of the few breeding places that may be available with polluted materials, making them unsuitable for the development of *Anopheles* larvae or resulting in decreased larval survival when present. In contrast to the junior staff residence, the faculty area seems to have all the factors that support and favour malaria transmission.

Though high biting rates were observed for *A. gambiae* in the rainy season than observed during the dry season, the sporozoite rate of 1.01 recorded in the dry season was higher than the sporozoite rate of 0.57 recorded in the rainy season. Cooseman *et al.*, (1992) found that potential malaria vectors occurring at high densities have often very low survival rates for assuring effective malaria transmission, and it explains this occurrence.

Relatively low parous rate and low expectation of life of *Anopheles* species were recorded during the rainy season compared to observation made in the dry season; thus confirming the findings of Cooseman *et al* (1992). The parous rate observed were 82.4% and 68.1% for the dry and rainy seasons respectively, these could be explained by the abundance and proliferation of anopheline breeding sites during the rainy season, which contribute to emergence of very young female adults all emerging at the sametime. The corresponding expectation of life calculated were 15.4days and 7.6days for the dry and rainy seasons respectively (assuming a blood meal interval of 3 days).

The overall prevalence of malaria on the KNUST campus of 14.2% (unpublished dissertation by Arthur, 2008) is lower compared to the prevalence of 50.72 reported for the Ashanti region (Browne *et al.*, 2000). The low infectivity rates of malaria vectors caught and the low malaria prevalence amongst inhabitants on the KNUST campus could be as result of behavioural changes on the part of the inhabitants (host) and adaptation by the vector. Diuk-wasser *et al.*, (2005) explained that when densities are high, people protect themselves, and the mosquitoes are forced to alternate hosts.

In assessing the level of malaria risk, experienced by inhabitants within the faculty area in terms of propensity and frequency of mosquitoes to bite host, the average retiring time of students and the biting activity was compared. The average retiring time of many students especially during the examination periods, which incidentally coincided with the months of infections (fig. 4.14), was after 12 midnight. Comparing the average retiring time and the high biting activity of *Anopheles gambiae* suggest that student face the highest malaria risk during the examination period, when they stay up deep into the night. The result of this comparison indicates that the tendency and frequency of the mosquitoes to bite its host within this period is very high.

Apparently, as result of their long personal experience with the nuisance biting behaviour of mosquitoes most respondents in KNUST were familiar with vector activity. It was noted that 97.4% of the respondents recognized the role of mosquitoes in malaria

transmission. The high malaria awareness could also be attributed to the higher education levels observed amongst most respondents, and previous malaria experience. However some of the most striking results of the community survey on misconceptions of inhabitants on malaria transmission such as; long exposure of a person to the sun, being rained on, eating too much oil and lack of hygiene as causes of malaria could lead to ineffective malaria prevention. This points out that there is the need for malaria control educational programs aimed at strengthening the capacities of communities for malaria control.

5.5.0 Knowledge Attitudes and Practice survey on Malaria

It was observed in this study that about 39.2% of 617 respondents used multiple mosquito protection measures, most of them combining bednets and any other. This result may also indicate the levels of awareness and enlightenments concerning malaria transmission patterns. However, there was a significant difference between the doers (those who used bednets) and non-doers (those who did not use bednets). Most people preferred the use of bed-nets because they thought it was the most effective method, which conforms to findings of Gyapong *et al.*, (1996) in a study in the northern part of Ghana. This provides an excellent base for increased training on bed net use and availability of nets to protect the target population in order to reach the Abuja target (WHO, 2000). Factors that led to non usage of other methods such as insecticide spray and mosquito coils usage included associated irritations as side effects to their usage. Understanding such an attitude regarding to preferences to intervention measures is important, since non-compliance for adapted strategies on a larger scale could undermine the use and sustainability of such intervention measures aimed at controlling malaria.

On treatment seeking behaviour it was observed that; 400 respondents (64.5%) were involved in self medication syndrome while only 35% visited hospital for treatment. This behaviour has been reported to be a common practice in many endemic areas worldwide (Foster, 1995), which is believed to have a likely financial undertone as well as

availability and affordability of sorts of antimalarial medications in drug stores (Mnyika *et al.*, 1995). It is evident in this study that high knowledge did not necessarily result in good attitudes and practices towards malaria. There is the need therefore to understand the reasons for these differences in knowledge attitudes and practices towards malaria in order to seek the appropriate methods towards malaria education. The efforts of the inhabitants to reduce the expectation of life and longevity of the vectors coupled with attitudinal change and education on the risk students and other inhabitants face would go a long way to reduce further the prevalence of malaria on the KNUST campus and its surroundings to a low level.

This study also shows the capabilities of spatial analysis and GIS in displaying geographically referenced malaria vector data such as the abundance and distribution of breeding sites on the KNUST campus (figure 4.16). From figure 4.15 the breeding sites could be ranked according to larval productivity and abundance, and this provides a basis for consistent monitoring and targeting of specific breeding sites by use of appropriate larval control strategies on a temporal basis. The risk maps would serve as an operational guide in future control interventions that may be designed. If indoor residual spraying is selected as an intervention, the maps would enable the KNUST to be divided into more accurate spraying zones. The exact number of sprayers required per zone as well as an accurate assessment of the amount of insecticide per zone can thus be done, since all information of sprayable structures could be incorporated in the maps. By loading the appropriate GIS database on operations that may be generated into software such as ArcView 9.2, progress of the spraying campaign and other control interventions that will be implemented can be monitored on a regular basis and other logistical information extracted as required.

CHAPTER SIX

6.0 CONCLUSION AND RECOMMENDATION

This study identified three different *Anopheles* species on the KNUST campus, based on morphological keys. The high abundance of *Anopheles gambiae* s.l. and their resulting high man biting rates (MBR), the detection of very few sporozoites and oocysts infection only in *A. gambiae* s.l. mosquitoes coupled with their seemingly high expectation of life, indicates that *A. gambiae* s.l. is the most efficient malaria vector species on the KNUST and surroundings. The results of the study points to the fact that there is active malaria transmission on the KNUST campus; though low, with close to 2 infective bites in a month and an average annual risk of 22 infective bites. Thus the risk of malaria that inhabitants on the KNUST may face is relatively lower comparing the entomological inoculation rate (EIR) for KNUST to EIRs in other parts of Ghana and Africa as a whole.. The risk of infection is high within the faculty area than at the other three study sites.

Though malaria transmission appears to be low on the KNUST campus, misconceptions by some inhabitants on malaria transmission coupled with the high abundance of the most efficient vector species, *Anopheles gambiae* Giles complex on the KNUST campus tell of a looming malaria threat that could result if an infectious gametocyte pool comes into play. There is the need to introduce vector control measures as one of the strategies of fighting malaria on the KNUST campus.

Yasuoka *et al*, 2006 recommends that an effective control strategy could be best achieved through community participation. This study, confirms the necessity of community involvement in control measures. The findings suggest that the educational factor had a significant effect on preventive behaviour, which was a useful finding. The findings show that it is necessary to design community sensitive but evidence-based education interventions which take such important observations such as local misconceptions on transmission into account.

Seasonality of vector density also becomes an important factor towards understanding malaria transmission, thus suggesting that control programs initiated should consider the seasonal patterns of the target parameters. The seasonal occurrence, the diversity in breeding sites and its varying productivity, is a further reflection of the need for further accurate mapping of all breeding sites area and subsequent cataloguing of their productivity in order to establish the temporal significance of each type of breeding site, over a much longer period. Mapping that shows spatial variability of the transmission using geographical data obtained from this study provides a reference for future control programs that may seek to eliminate or reduce the risk and exposure to malaria on the KNUST campus and nearby communities. This study presents the possibility of using GIS and remote sensing tools for health research, in Ghana, where GIS application in the health sector has not been extensively employed.

The findings support the necessity to implement a system of epidemiological surveillance to halt any possible dispersion of risk within the area. Educational programs aimed at increasing awareness on the correct attitudes and practices towards malaria transmission could promote community participation for effective malaria control in the study area.

All sectors of the society, central government, education departments and policy makers on malaria control have a vital role to play in enabling proper implementation of information provided by such evidence-based research to control malaria effectively and also to ensure that interventions are sustainable, culturally appropriate and resources appropriately utilized; by aiming identified zones of interest.

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APPENDIX 1

MATERIALS, EQUIPMENTS USED IN LABORATORY AND FIELD

1.0.1 Study and sampling of *Anopheles* species breeding sites

Materials/equipment

pH/Cond 340i/SET

GPS machine

1.0.2 Night catching of adult mosquitoes

Materials/equipment

Torch light

Batteries

Hour bags

Cooling elements/ice

Cooling boxes

1.0.3 Dissection

Materials/equipment

Stereo microscope

Compound microscope

Clean slides

Microtitre plates

Dissecting kit

Reagents

1XPBS

Sterile water

APPENDIX 2

QUESTIONNAIRE SURVEY ON KNOWLEDGE, ATTITUDE AND PRACTICES ON MALARIA ON KNUST CAMPUS AND SURROUNDINGS

A. PERSONAL INFORMATION

1. Date: 2. Age of Respondent:
3. Residential address..... 4. Occupation:
5. Gender (please tick) Male ☐ Female ☐
6. Educational Background Basic ☐ Secondary ☐ Tertiary ☐

B. PERSONAL INFORMATION

7. Have you heard of Malaria (please tick) Yes ☐ No ☐
8. What causes malaria
9. How often in a year (academic year) do you experience malaria attacks?
10. Which of these preventive measures do you know of (please tick)?
- Insecticide Treated Bed net ☐ Insecticide Spray ☐ Mosquito Coil ☐
- Window Screens/door trap ☐ Anti-malaria drug ☐ Repellents ☐
- (i) If Other please specify
- (ii) Where did you learn about this method ?
11. Which of these preventive measures do you use (please tick)?
- Insecticide Treated Bed net ☐ Insecticide Spray ☐ Mosquito Coil ☐
- Window Screens/door trap ☐ Anti-malaria drug ☐ Repellents ☐
- (i) If Other please specify
- (ii) Where did you learn about this method ?
12. If you use repellent when/what time, do you apply it?
13. How can you tell if you have malaria?
14. How you would treat malaria attacks ?
15. What time do you go to bed (sleep)?

APPENDIX 3
LABORATORY RECORD FORM
For Entomological Study

Date of Collection:					Catch No.:		
Date of Dissection:					Plate No.:		
No.	Study Site	Hour of Collection	Type of <i>Anopheles</i> spp	Age	Salivary Gland	Location in well	Remarks
1						A1	
2						A2	
3						A3	
4						A4	
5						A5	
6						A6	
7						A7	
8						A8	
9						A9	
10						A10	
11						A11	
12						A12	
13						B1	
14						B2	
15						B3	
16						B4	
17						B5	
18						B6	
19						B7	
20						B8	
21						B9	
22						B10	
23						B11	
24						B12	
25						C1	
26						C2	
27						C3	
28						C4	
29						C5	
30						C6	
31						C7	
32						C8	
33						C9	
34						C10	

APPENDIX 4

FIELD RECORD FORM

Date of Collection

Catch No:

Hour of Collection	Site of Collection	No. of <i>Anopheles</i> spp caught	No. of Culicines caught	Average Temperature for the hour
18.00 - 19.00				
19.00 - 20.00				
20.00 - 21.00				
21.00 - 22.00				
22.00 - 23.00				
00.00 - 01.00				
01.00 - 02.00				
02.00 - 03.00				
03.00 - 04.00				
04.00 - 05.00				
05.00 - 06.00				