

KWAME NKRUMAH UNIVERSITY OF SCIENCE AND TECHNOLOGY,

KUMASI

COLLEGE OF SCIENCE

DEPARTMENT OF THEORETICAL AND APPLIED BIOLOGY

MSc. ENVIRONMENTAL SCIENCE

**GASEOUS POLLUTANT EMISSIONS AND MOSQUITO SUSCEPTIBILITY
FROM THE USE OF MOSQUITO COILS IN THE INDOOR
ENVIRONMENT**

THOMAS PEPRAH AGYEKUM

NOVEMBER, 2016

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ENVIRONMENT**

**A THESIS SUBMITTED TO THE SCHOOL OF GRADUATE STUDIES,
KWAME NKRUMAH UNIVERSITY OF SCIENCE AND TECHNOLOGY,
(KNUST) IN PARTIAL FULFILMENT OF THE REQUIREMENTS FOR THE
AWARD OF MSc. DEGREE IN ENVIRONMENTAL SCIENCE**

**BY
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BSc. (HONS) ENVIRONMENTAL SCIENCE

NOVEMBER, 2016

KNUST



DECLARATION

I, hereby declare that, except for references to other people's work which have been duly acknowledged, this write-up, submitted to the Department of Theoretical and Applied Biology, KNUST, Kumasi is the result of my own original research and that this thesis has not been presented for any degree elsewhere.

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Head of Department

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Date

DEDICATION

This work is dedicated to my parents Mr. Samuel Agyei Agyekum and Madam

Comfort Boakyewaa

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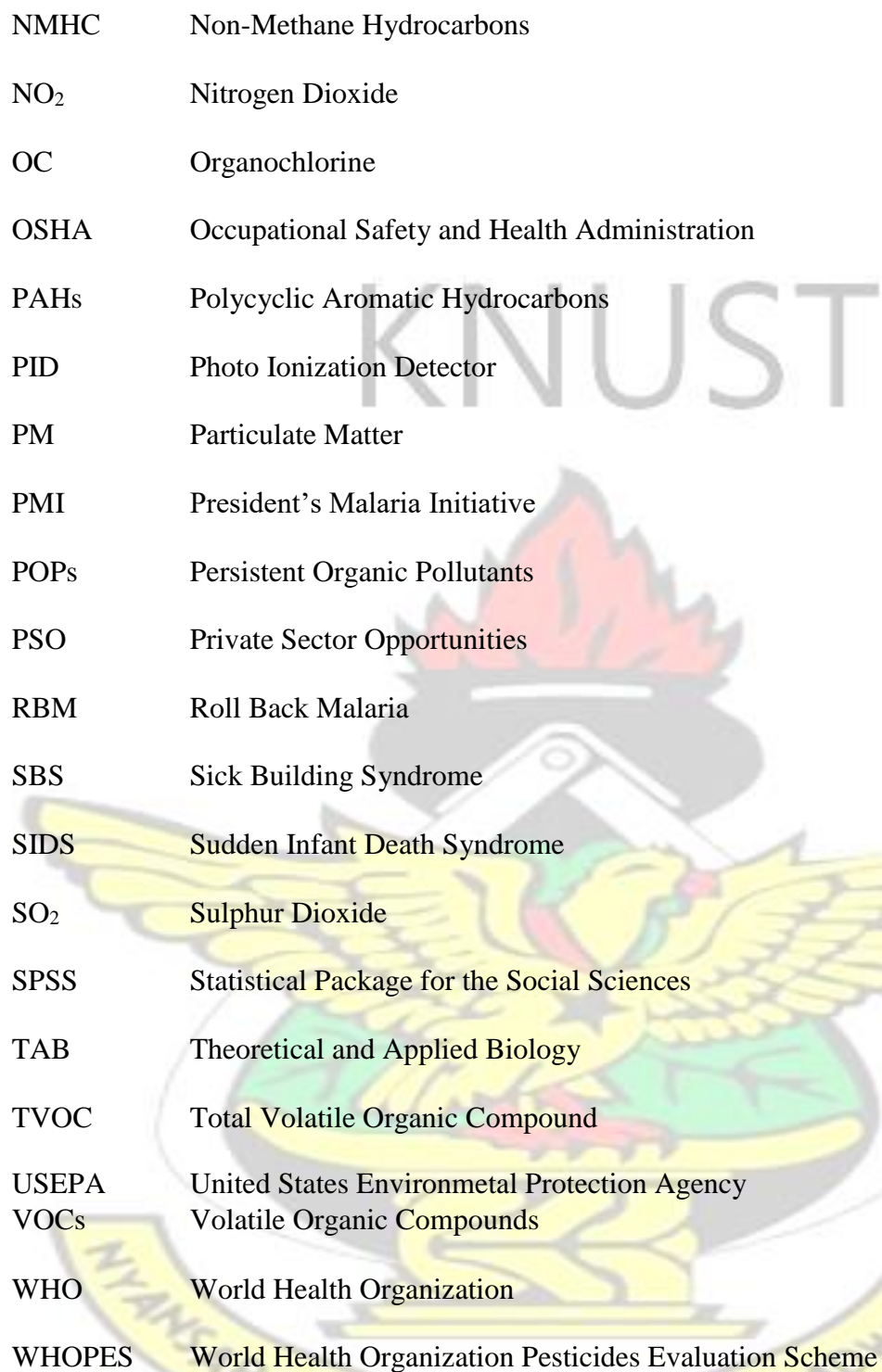
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LIST OF ABBREVIATIONS

AI	Active Ingredients
AIDS	Acquired Immune Deficiency Syndrome
BCME	Bis (chloromethyl) ether
BDL	Below Detection Limit
CDC	Centres for Disease Control and Prevention

CH ₄	Methane
CNS	Central Nervous System
CO	Carbon Monoxide
CO ₂	Carbon Dioxide
COHb	Carboxyhaemoglobin
DDD	Dichlorodiphenyldichloroethane)
DDT	Dichlorodiphenyltrichloroethane
GABA	Gamma-Aminobutyric Acid
GHS	Ghana Health Service
GMAP	Global Malaria Action Plan
GSE	Gas Sensitive Electrochemical
GSP	Ghana Spraying Performance
GSS	Ghana Statistical Service
H ₂ SO ₄	Sulphuric acid
HCB	Hexachloro-benzene
HCHs	Hexachlorocyclohexanes
HIV	Human Immunodeficiency Virus
IAPA	Industrial Accident Prevention Association
IRS	Indoor Residual Spraying
ITN	Insecticide-Treated Net
KCCR	Kumasi Centre for Collaborative Research in Tropical Medicine
KNUST	Kwame Nkrumah University of Science and Technology
LLIN	Long-Lasting Insecticide-treated Net
MC	Mosquito Coil
MDGs	Millennium Development Goals
MDH	Minnesota Department of Health



NMHC	Non-Methane Hydrocarbons
NO ₂	Nitrogen Dioxide
OC	Organochlorine
OSHA	Occupational Safety and Health Administration
PAHs	Polycyclic Aromatic Hydrocarbons
PID	Photo Ionization Detector
PM	Particulate Matter
PMI	President's Malaria Initiative
POPs	Persistent Organic Pollutants
PSO	Private Sector Opportunities
RBM	Roll Back Malaria
SBS	Sick Building Syndrome
SIDS	Sudden Infant Death Syndrome
SO ₂	Sulphur Dioxide
SPSS	Statistical Package for the Social Sciences
TAB	Theoretical and Applied Biology
TVOC	Total Volatile Organic Compound
USEPA	United States Environmental Protection Agency
VOCs	Volatile Organic Compounds
WHO	World Health Organization
WHOPES	World Health Organization Pesticides Evaluation Scheme

ABSTRACT

Malaria remains a challenge in sub-Saharan Africa and continues to be the primary cause of morbidity and mortality in Ghana. The use of mosquito coil is one of the ways people prevent themselves from mosquito bites. Burning these mosquito coils indoor produces smoke that can control mosquitoes effectively. This exercise is presently used not only in Africa but also in many households in Asia and South America. However, the smoke generated may contain pollutants of great health concern.

Efficacy and emission studies of five different commercial mosquito coils containing esbiothrin, dimefluthrin, d-allethrin and meperfluthrin were performed in three different experimental rooms under ventilated and poorly ventilated conditions in the Department of TAB, KNUST, Kumasi. All mortality tests were performed on 3-6 days old (sucrose fed) reared female *Anopheles* mosquitoes. Aeroqual Gas Monitor (S500) was used to determine the level of pollutants emitted from mosquito coil smoke (CO, TVOC, NO₂ and SO₂) and environmental factors like temperature and relative humidity. Ventilation rates were determined using a single-compartment mass balance model.

Mortality studies under poorly ventilated conditions were higher in all the three experimental rooms than under ventilated conditions. Statistically, there was a significant difference ($p < 0.05$) between the mortalities of mosquitoes under ventilated and poorly ventilated conditions. Mosquito coils containing esbiothrin recorded the highest insecticidal activity. There was statistically no significant difference ($p > 0.05$) between mortalities of the five tested coils under ventilated conditions but under the poorly ventilated conditions, there was a significant difference ($p < 0.05$) between the mortalities of MC 1 (esbiothrin) and MC 2 (dimefluthrin). Efficacy was found to decrease with an increase in room size from 8.5 m³ to 19 m³ and then to 34 m³ however there was no statistical difference between the mortalities recorded in 8.5 m³ and 19 m³ room as well as 19 m³ and 34 m³ room. The results also showed that active ingredients of the coils played a key role in the efficacy of the mosquito coils.

Pollutant concentrations resulting from burning mosquito coils especially under poorly ventilated conditions could substantially exceed health-based air quality standards or guidelines. Pollutant concentrations decreased as ventilation rates increased in the experimental rooms. The pollutant concentrations recorded under ventilated conditions were lower than concentrations under poorly ventilated conditions. The result of this study did not show any clearly defined trend of decreasing pollutant levels with increasing room sizes.

The findings from this study suggest that individuals sleeping in rooms with lit mosquito coils may be exposed to some undesirable levels of pollutants emitted from the coils. If it becomes necessary to use mosquito coil, the coil should first be burned in a closed indoor environment to achieve maximum insecticidal effect, following which the rooms should be well aerated prior to sleeping in them.

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CHAPTER ONE

INTRODUCTION

1.1 BACKGROUND OF THE STUDY

Malaria has been a global health issue for over 125 years since the establishment that plasmodium species was the causative organism (Walther and Walther, 2007). According to the World Malaria Report (WHO, 2014a), there is a global estimation of 3.2 billion people within 97 nations and territories that are at risk of malaria and emerging diseases infection and 1.2 billion are at high risk (>1 in 1000 chance) of getting malaria in a year. The problem is more profound in the WHO African State, where there is an estimation of 90% of all malaria deaths occurring and most of the deaths (about 78%) occur among children under five years (WHO, 2014a).

In Ghana, malaria is endemic and year-round in all parts of the country with seasonal changes that are more noticeable in the north. The entire Ghanaian population is at risk of malaria infection, but as a result of lowered immunity, children under five years and pregnant women are at greater risk of serious illness (PMI, 2014). A prime aspect of eradicating or controlling malaria and hence reducing its transmission is to control the infective mosquito from biting individuals. Controlling malaria vectors with insecticides remain a vital component in the attempt to reduce or probably eradicate malaria (WHO, 2009a).

Treated mosquito nets (Long-lasting Insecticide-treated Nets (LLIN) and Insecticides Treated Nets (ITN)) and indoor residual spraying (IRS) are the two main recommended vector control methods used to prevent individuals from mosquito bites (WHO, 2014a). In Ghana, the aim is to achieve 100% coverage of households owning a minimum of one

ITN, with 80% of the general populace sleeping under ITNs (GHS, 2013). According to Adjei and Gyimah (2012), only 48% of households own ITNs and 52.7% of the people owning the ITNs use it. This implies that a small fraction of Ghana's population is using mosquito nets to control the vector. With regards to the use of IRS, few households can afford them due to the high cost involved (Miller and Tren, 2012).

Mosquito coils, though not officially part of the Ghana's malaria control programmes, are highly patronized in the country mostly by rural and urban poor due to its affordability. The burning of mosquito coils in rooms produce smoke that can effectively repel mosquitoes. This exercise is ongoing in many homes in Africa as well as Asia and South America (Liu *et al.*, 2003). On the other hand, the mosquito coil smoke emissions may contain particulate matter, sulphur dioxide (SO₂), carbon dioxide (CO₂), nitrogen dioxide (NO₂), carbon monoxide (CO), ketones, aldehydes, polycyclic aromatic hydrocarbons (PAHs) and some volatile organic compounds (VOCs) and exposure to these cause both critical and chronic health hazards (Liu *et al.*, 2003). It has been suggested that exposure to the smoke generated from burning of mosquito coils may be a leading factor for the development of lung cancer (Shu-Chen *et al.*, 2008). Shu-Chen and co-workers found out that almost 50% of lung cancer deaths in Taiwan were not associated to cigarette smoking. Environmental exposure to smoke of mosquito coil burning may play a role in the development of the disease. Taiwanese households normally burn coils at home to deter mosquitoes and the risk of getting lung cancer was considerably higher in regular burners of mosquito coils (thrice per week) as compared to those who did not burn any mosquito coil. Given the undesirable smoke that may emanate from the burning of mosquito coils, vaporizer mats has been suggested as possible replacement (Ogoma *et al.*, 2012). The vaporizer mats contain entrenched repellent active ingredients that are volatilized using an

electric heating element. The use of electric heating can escalate the cost of production and hence unsuitable for a number of rural and urban dwellers to access the vaporizer mats.

A major gap in the current literature on mosquito coils is that it lacks clarity on whether the use of mosquito coils provide a net benefit or otherwise for the malaria control. If application of mosquito coils is a risky venture, then, people must be appropriately informed and the practice discouraged. On the other hand, if the benefits are overwhelming and the risks are within acceptable threshold, then, the mosquito coil could complement the current malaria control options, especially in poor communities.

1.2 PROBLEM STATEMENT

Malaria remains a challenge in Africa and continues to be the primary cause of illness and death in Ghana (Ronald *et al.*, 2006). Many attempts have been made to combat the disease in the country as well as many endemic countries but achievements have been minimal. Malaria accounted for about 40% of outpatient turnout with a yearly reported cases of 2.2 million from 1995 to 2001, and more than 10% were admitted at the hospital (Mba and Aboh, 2007). In 2010, the Statistics Department of the Kumasi Metropolitan Assembly (KMA) reported of 468 cases of malaria admissions in all ages and this increased to 712 cases in 2011 (KMA, 2011).

The control of malaria vectors depends mostly on the use of long-lasting insecticidetreated nets (LLINs) and indoor residual-spraying (IRS) (Sketetee and Campbell, 2010). However, in Africa and other developing countries, the use of mosquito coils is highly patronized for the control of malaria. The use of mosquito coils indoors may generate some amount of indoor smoke.

It is estimated that about 1.9 million individuals experience untimely deaths as a result of exposure to smoke generated indoors especially from solid fuel burning (Roehr, 2011).

Exposure to this indoor smoke is classified as the leading environmental risk factor accountable for 3.3 % of all deaths and 2.7 % of all disability-adjusted life years per year (WHO, 2009b).

Most of the mosquito coils available on the Ghanaian market contain pyrethrins since it is one of the most active ingredients (contributing to 0.3 – 0.4 %) in the mosquito coils (Lukwa and Chandiwana, 1998). People usually burn mosquito coils during the evenings to prevent mosquito bites and are therefore normally exposed to the smokes emanating from the coils for about 6 to 8 hours daily. When these coils are burned, the insecticidal contents of the coil vaporise with the smoke thereby preventing the malaria vector from entering the room or coming near the individual (Liu *et al.*, 2003). Regardless of its potential to repel the mosquitoes, it also generates smoke which may contain air pollutants (such as CO, PAHs, VOCs, PM, ketones and aldehydes) of great health concern (Liu *et al.*, 2003). Inhalation of mosquito coil smoke containing such pollutants has been said to cause asthma, bronchial irritation, eye irritation, itching, breathing difficulties and cough (Kurmi *et al.*, 2012).

1.3 JUSTIFICATION

The use of mosquito coils are not officially part of the malaria control programmes in Ghana yet they are highly patronized in the country mostly by rural and urban poor due to its affordability and accessibility. It is presumed that the use of mosquito coils has the ability to repel mosquitoes thereby reducing malaria transmission. Nevertheless, there are potential health implications that may arise as a result of the emissions from the smoke of the coils. Thus, although the use of the mosquito coil may be beneficial, there are concerns of environmental health risk. Therefore, there is the need to research into the level of pollutants generated from mosquito coils and also mosquito susceptibility from the use of

mosquito coils in the indoor environment. This study would provide knowledge on the efficacy and the level of pollutants generated from the burning of mosquito coils.

1.4 OBJECTIVES OF THE STUDY

The main objective of the study was to investigate the potential efficacy and gaseous emissions arising from the use of mosquito coils. The specific objectives were to:

- assess the efficacy of mosquito coils in terms of mosquito mortality.
- assess the concentration of pollutants (CO, TVOC, SO₂ and NO₂) due to emissions from mosquito coils.
- evaluate the effects from the mosquito coils under varying experimental indoor conditions in terms of different room sizes and ventilation rates.

CHAPTER TWO LITERATURE REVIEW

2.1 MOSQUITOES

Mosquitoes belong to the family *Culicidae* in the insect order of true-flies or two-winged flies called Diptera (Harbach, 2007). They are found globally except in areas that are permanently frozen. There are approximately 3,500 species of mosquitoes and out of this, almost three-quarters are found in the humid tropics and subtropics (Reiter, 2001).

Malaria parasites are transmitted to individuals from the bite of infected female *Anopheles* mosquitoes (Molavi, 2003) during the process of taking blood meal to help them develop their eggs. Mosquitoes are naturally part of the aquatic ecosystems and it should be expected that at least some mosquito activity will be experienced in the course of the year (warmer months) (Byun, 2012). However, agricultural practices such as storage of water

in reservoirs for animal husbandry and use of water for fishponds (Oladebo *et al.*, 2010), and poor sanitation (choked gutters, dirty and bushy surroundings) can provide a suitable breeding grounds for mosquitoes. This notwithstanding, climatic factors such as humidity, temperature and rainfall can greatly affect the ecology, development, behaviour, and survival of mosquitoes and the transmission dynamics (Reiter, 2001). According to a research conducted by Ntonfor *et al.* (2007), presently no sufficiently effective method has been found to completely destroy the malaria vector.

2.2 LIFE CYCLE OF MOSQUITO

The mosquito's life cycle is in four (4) stages and they are egg, larvae, pupa and the adult stage. The first three stages (egg, larva and pupa) of the cycle are spent in water, and it is only the final stage (adult stage) that negatively affects human wellbeing.

2.2.1 Eggs Stage

The female adult mosquito lays 200 – 300 eggs in one gonotrophic cycle. These eggs are laid separately and directly on water surfaces (Pwalia, 2014). The eggs are susceptible to desiccation and hatch within 2 to 3 days, even though hatching may take up to two to three weeks in colder climates (Coleman, 2009). When mosquito eggs are laid, they are white in colour but grow dark within 12 – 24 hours. With the exception of the *Anopheles* species whose eggs have floats attached to each side, the eggs of most species seem alike when viewed by the naked eye. The incubation period is contingent on genetic as well as environmental factors and differs greatly among different species (Dame and Brammer, 2002).

2.2.2 Larval stage

The larva of mosquito has a well-built head and a mouth with brushes for feeding, a large thorax and a segmented abdomen (Pwalia, 2014). *Anopheles* larvae breathe via a

congregate of small abdominal plates, which engenders them to lie flat near the basement of the water surface when not plunging. The larvae of some mosquito species are rapacious and prey on other invertebrates inclusive of mosquito larvae. Typical examples are *Toxorhynchites rutilus* and *Psorophora ciliata*. Aside this, some also feed by filtering microorganisms such as fungi, bacteria and protozoa (Renchie and Johnsen, 2007). Larvae evolve through four stages, after which they transform into pupae. The larvae moult and shed their skins to enable further growth at the end of each stage (Dame and Brammer, 2002).

2.2.3 Pupal stage

The shape of pupa is like a comma. Its head and thorax are fused into a cephalothorax, with the abdomen situated beneath. Major transformations happen during this stage, leading to the conversion of larval tissues into adult tissues (Coleman, 2009). They are relatively active and swiftly swim toward the bottom of their habitat on disruption. They do not feed and because of this they are not very active as compared to the larva. The pupa emerge into adult mosquitoes within 2 – 4 days and this development process commences with the removal of the pupal skin along the back (Goddard, 2009).

2.2.4 Adult stage

The adult mosquito looks out for a safe surrounding in the nearby flora to help its wings to fully develop after emerging from the pupal stage. Male mosquitoes come before the female mosquitoes and mate with them as soon as the females are ready. Plant exudates and nectar are the main carbohydrate sources of food for both the male and female mosquitoes which provides them with energy for their life activities such as flying and mating. In addition to the carbohydrates, the female mosquito alone takes a blood meal to

provide her with additional proteins for egg development. The life span of the male mosquito is only one week or two weeks whereas the female can live up to one month to produce more eggs (Renchie and Johnsen, 2007).

The female mosquitoes lay their eggs in aquatic habitats, in fissures in the soil, or on other convenient substrates or any unique niches that are likely to flood (e.g. containers and tree holes), and the whole mosquito life cycle reoccurs (Dame and Brammer, 2002). The female mosquito is responsible for the transmission of malaria parasite and this occurs through the course of taking a blood meal. Below is a simplified diagram of the mosquito's life cycle.

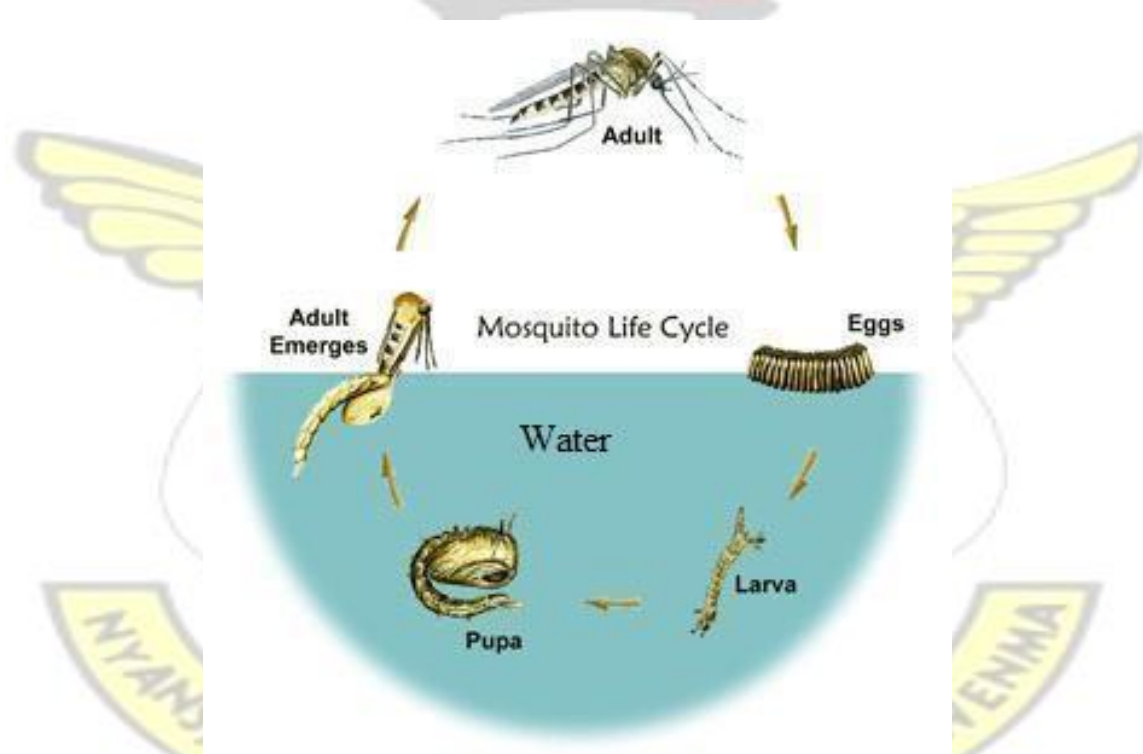


Figure 1: Life cycle of mosquito
(Source: www.mosquito.org/life-cycle)

2.3 EPIDEMIOLOGY OF MALARIA

Malaria is a communicable disease induced by a protozoan known as plasmodium and it's transferred from one individual to another via the bite of the female *Anopheles* mosquito

(Reiter, 2001). It can be transmitted by any of the four human malaria species which comprise *Plasmodium vivax*, *Plasmodium falciparum*, *Plasmodium ovale* and *Plasmodium malariae* (WHO, 2014b). Between these four species, *Plasmodium falciparum* is the most serious and dreaded malaria parasite and infection from it can lead to life threatening complexities (Otchere, 2014).

Malaria is classified as one of the six killer diseases in the world (Pal *et al.*, 2011). According to the latest estimates of the WHO, there were about 198 million global occurrences of malaria cases and this led to the death of 584000 individuals. Majority of the deaths (about 78 %) occurred among children under five years. The disease normally affects the poorest and most marginalized communities (WHO, 2014b).

The life cycle of malaria is very complex. It starts with the infected female *Anopheles* mosquito injecting sporozoites into the blood of its host during biting. The sporozoites then enter and proliferate in the liver cells of the infected host which proceeds to merozoites production. The merozoites attack the red blood cells and proliferation proceeds, with the rupture of red blood cells producing the indications from the disease. When a mosquito bites an infected host, the parasite is taken up again, matures in the stomach of the mosquito and then infects another host and the cycle continues. Infected individuals show symptoms of malaria infection approximately 9 to 14 days after been bitten by the mosquito. Some of the symptoms include: headache, vomiting, joint pains and fever (Cox, 2002). The cycle starts again when the mosquito bites another person. Below is a life cycle of malaria parasite.

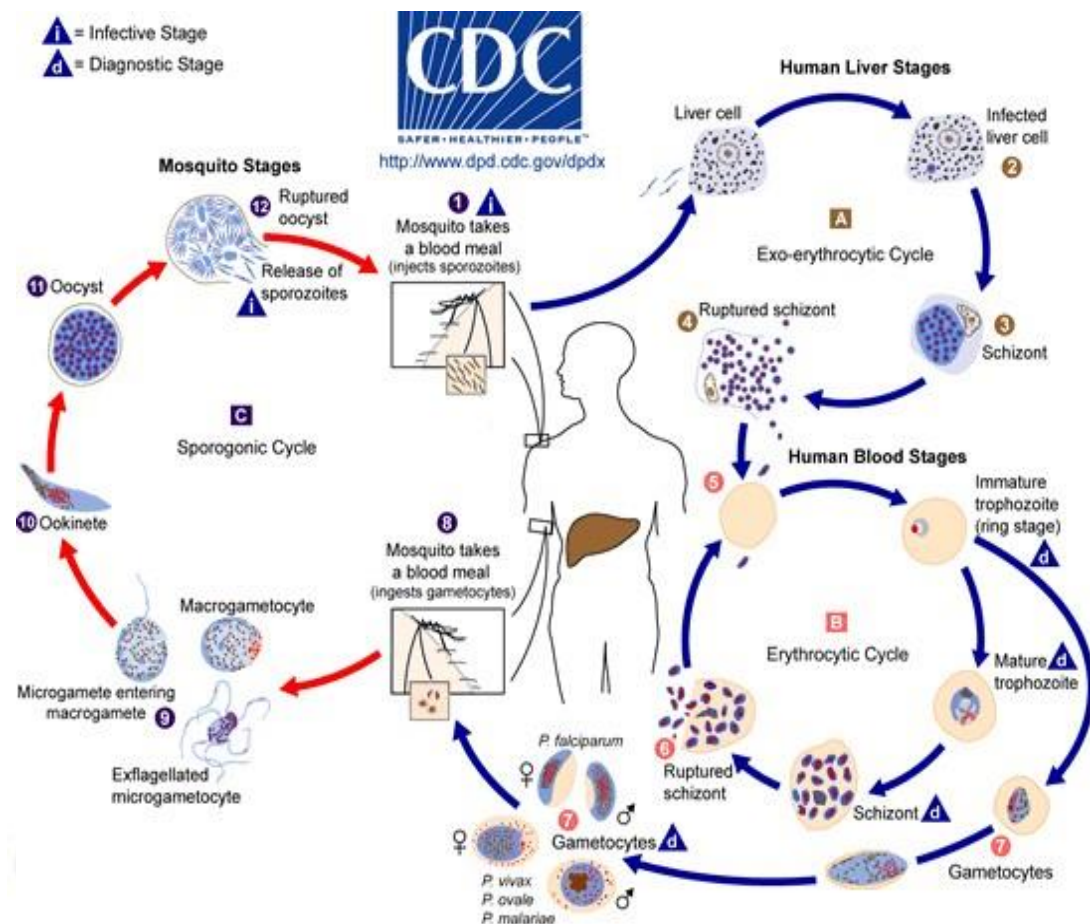


Figure 2: Life cycle of malaria parasite
(Source: www.cdc.gov/malaria/about/biology)

2.4 MALARIA VECTOR CONTROL

The control of malaria is urgently needed in realizing the Millennium Development Goals (MDGs). The MDG 6 talks about fighting malaria, HIV/AIDS and other diseases and this precisely addresses malaria. It also directly helps in the achievement of MDG 4 (minimising child death), and MDG 5 (enhancing maternal wellbeing) (Korenromp, 2005).

Vector control is an integral aspect of numerous vector-borne disease control programs. Its administration comprises targeted, site-specific usage of the accessible methods, based on practical and functioning achievability, resources and infrastructure. The application must be done in following the ideologies of integrated vector management, confirmed

decision-making process suitable to indigenous locales, which justifies the usage of vector control approaches and resources and also stresses on community participation (WHO, 2006a). The control of malaria vector is the prime intervention for worldwide reduction and eradication of malaria. It is crucial for the minimisation and also the disruption of malaria transmission. Presently, the two most common vector control mediations comprise Indoor Residual Spraying (IRS) and the use of insecticide treated nets (ITN or LLIN). Chemical insecticides for vector control are of four classes and they are pyrethroids, organophosphates, carbamates and organochlorines (chlorinated hydrocarbon) (WHO, 2006a).

2.4.1 Indoor Residual Spraying (IRS)

It is the use of a long-term, residual insecticide to potential mosquito hidden surfaces such as interior walls and ceilings of all houses where such mosquitoes might come into contact with the insecticide (WHO, 2013a). Indoor Residual Spraying (IRS) programmes remains the most extensively used technique for controlling mosquitoes (WHO, 2006a) and highly effective and can also drastically lower local malaria incidence and mortality, on condition that mosquito hide-outs in targeted communities are identified and sprayed (WHO, 2013b). This vector control method was introduced during the late 1940s when DDT was available and used to control mosquito vectors of malaria that entered houses. The Chagas disease in Latin America was also controlled using the same technique (Matthews *et al.*, 2012).

In reality, the success of house spraying for controlling malaria is dependent on adherence to the principles stated for the insecticide and the process of application, mass approval of spraying, the accessibility of well-kept equipment, well-trained spraying employees, effective supervision and strong fiscal support (WHO, 2006a). Not any insecticide is used

for the IRS programme. Based on that, the WHO has recommended some insecticides for IRS treatment against mosquito vectors and the table below gives the list of such insecticides.

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Table 1: WHO-endorsed insecticides for indoor residual treatment against mosquito vectors

Insecticide	Chemical type	Dosage of AI (g/m ²)	Duration of effective action (months)	Insecticide action	WHO hazard classification of AI ^c
Bendiocarb	Carbamate	0.1 – 0.4	2–6	Contact & airborne	II
Propoxur	Carbamate	1.0 – 2.0	3–6	Contact & airborne	II
DDT	Organochlorine	1.0 – 2.0	> 6	Contact	II
Fenitrothion	Organochlorine	2.0	3–6	Contact & airborne	II
Malathion	Organochlorine	2.0	2–3	Contact	III
Pirimiphos-methyl	Organochlorine	1.0 – 2.0	2–3	Contact & airborne	II
α-Cypermethrin	Pyrethroid	0.02 – 0.03	4–6	Contact	II
Bifenthrin	Pyrethroid	0.025 – 0.050	3–6	Contact	II
Cyfluthrin	Pyrethroid	0.02 – 0.05	3–6	Contact	II
Deltamethrin	Pyrethroid	0.020 – 0.025	3–6	Contact	II
Etofenprox	Pyrethroid	0.1 – 0.3	3–6	Contact	U
λ-Cyhalothrin	Pyrethroid	0.02 – 0.03	3–6	Contact	II

AI Active Ingredient

^c Class II, moderately hazardous; class III, slightly hazardous; class U, unlikely to produce an acute risk in normal use Source: WHOPES

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Among all the insecticides used for malaria control, DDT is the most effective IRS chemical used so far. Aside its ability to kill mosquitoes, it also reduces indoor mosquito populations and hence reducing the spread of malaria. According to literature, majority of the success of DDT is as a result of the excito-repellency present in them (Grieco *et al.*, 2007; Roberts and Tren, 2010; Sadasivaiah *et al.*, 2007). Excito-repellency results from insects coming into direct contact with insecticides on treated surfaces at a distance (Miller *et al.*, 2009). It is known that DDT which is an example of organochlorine affects the nervous system of insects by affecting the voltage-gated sodium channel proteins located in the insect's nerve cell membranes, distorting transmission of nerve impulses and finally leads to insect mortality (Glavan and Božič, 2013).

Many sub-Saharan African countries have included IRS in their extensive malaria control plan in agreement with the Global Malaria Action Plan (GMAP) instituted by the WHO and Roll Back Malaria (RBM) Partnership. Globally, 185 million persons (6 % of the global population at risk) were shielded from malaria through the use of IRS (Gething *et al.*, 2011). The total number of persons shielded from malaria by IRS in the WHO Africa state amplified from ten (10) million in 2005 to seventy-eight (78) million in 2010 (PSO, 2013). In Ghana, IRS activity was carried out in 8 districts in 2010. Out of the 8 districts, 6 were already part of the IRS programme and 2 new districts were added (Chereponi and Saboba). The activity protected 849,620 people from malaria (GSP, 2010). The prices of insecticides are one of the key factors that determine the total cost and also the coverage of an IRS programme (Miller and Tren, 2012). Due to the high cost of IRS chemicals, the poor people who are at higher risk to malaria cannot afford the insecticides.

2.4.2 Insecticide Treated Mosquito Nets

One of the major tools of the Roll Back Malaria (RBM) campaign is the use of insecticide treated mosquito bed nets (Skovmand *et al.*, 2008). An insecticide-treated net (ITN) is a bed net intended to physically obstruct the mosquito vectors, and also processed with residual insecticide for the purpose of repelling or killing mosquito vectors which cause malaria (Lengeler, 2004a). It is retreated with insecticides after some months of washing. A long-lasting insecticide-treated net (LLIN) on the other hand is an insecticide treated net made to remain effective for several years without retreatment (Jamison *et al.*, 2006) after washing. Insecticide treated mosquito nets (ITN and LLIN) come in different shapes and colours. Some of them have rectangular shapes while others are circular in shape. In terms of colours, the available mosquito nets on the market have white, light blue, dark blue and green colours.

Table 2: WHO-endorsed long-lasting insecticidal mosquito nets for use in public health

Product name	Product type	Status of WHO recommendation
DawaPlus® 2.0	Deltamethrin coated on polyester	Interim
Duranet®	Alpha-cypermethrin integrated into polyethylene	Interim
Interceptor®	Alpha-cypermethrin coated on polyester	Interim
Netprotect®	Deltamethrin integrated into polyethylene	Interim
Olyset®	Permethrin integrated into polyethylene	Full
PermaNet® 2.0	Deltamethrin coated on polyester	Full
PermaNet® 2.5	Deltamethrin coated on polyester with strengthened border	Interim
PermaNet® 3.0	Combination of deltamethrin coated on polyester with strengthened border (side panels) and deltamethrin and PBO integrated into polyethylene (roof)	Interim

Source: (Prato *et al.*, 2012)

The use of treated mosquito nets repels malaria vectors from biting people and thus significantly reduces malaria infections and its transmission (Mohammed, 2013). The nets are treated with insecticides capable of killing or repelling the mosquitoes. According to Lengeler (2004b), the control of mosquito vectors through the use of ITNs have been shown to greatly contributed to the reduction of malaria. In sub-Saharan Africa, the use of ITNs is estimated to lower malaria mortality rates in children below the age of five (5) by 55 % (Eisele *et al.*, 2010).

Regardless of the magnificent increases in malaria intervention coverage, an estimation in 2013 realized that 278 million out of the 840 million persons who were at risk of malaria infection in sub-Saharan Africa were living in their homes with no ITN (WHO, 2014b). Majority of nets (ITNs and LLINs) received by families at subsidized prices and also free of charge during mass distributions go unutilized (Fettene *et al.*, 2009; Ndjinga and Minakawa, 2010), thereby reducing the effectiveness of malaria control programs.

2.5 MOSQUITO COILS

The use of mosquito bed nets will be the most encouraging method for controlling malaria mosquitoes at low cost and high sustainability, as compared to residual spraying. Nonetheless, the exophagic behaviour of mosquitoes to change their biting preference from late to early evenings still poses a great problem. People who stay outdoors and indoors could still be introduced to threats of malaria transmission. The use of mosquito coils are able to prevent or repel mosquitoes from biting people (Kawada *et al.*, 2004).

Mosquito coil is a spiral coil which burns slowly and releases a smoke that prevents mosquitoes from biting. Mosquito coils are produced from a paste of granulated insecticide and a filler such as sawdust, and then extruded into a coiled shape (Lawrance and Croft,

2004). The coil is normally held at the middle of the spiral, suspending it in the air, or wedged by two pieces of heatproof nettings to enable constant smouldering. Burning generally starts from the outer part of the spiral and advances gradually toward the middle of the spiral, generating a mosquito-repellent smoke (McKean, 2005; Ogbonnia *et al.*, 2016). After lighting up the free end of a mosquito coil, it smoulders at a steady rate for 7 – 9 hours and releases insecticide into the air gradually. The insecticidal effect can only be reached after a certain concentration of the insecticide has accumulated in the chamber. The time required is dependent on the type and concentration of the active ingredients, the size of the room and the wind speed (Yin, 2009).



Plate 1: Mosquito Coil

Globally, 45 – 50 billion pieces of mosquito coils are used by about 2 billion persons in a year (Zhang *et al.*, 2010) and they are predominantly used in rural and semirural societies of developing nations like Asia, South America and Africa (Liu *et al.*, 2003) to prevent mosquito bites. The use of mosquito coils in Ghana is on its ascendancy. There are quite large a number of mosquito coil brands on the Ghanaian market and they are highly patronized. Most of these coils are mainly from China, Indonesia and Malaysia (Avicor *et*

al., 2013). Most people especially the poor patronize the coil due to its affordability and accessibility (Lawrance and Croft, 2004). About 30 % of households in the nation's capital use mosquito coils to control mosquitoes (Boakye *et al.*, 2009). Also in a study conducted in some urban, peri-urban and rural areas in Ghana, 62.9 % of the respondents use mosquito coils to prevent mosquitoes from biting them (Kudom *et al.*, 2013).

Pyrethrins (insecticidal compound that occurs in pyrethrum), which is the major active ingredient of the coil accounts for almost 0.3 – 0.4 % of the coil mass. Majority of coils are made up of plant-based constituents, such as joss powder, dyes, wood powder, oxidants (e.g., nitrates), binders, coconut shell powder and other extracts (Shu-Chen *et al.*, 2008). Subject to the size of the room in which the coil is burned, and the type of active ingredient used in the formulation of the coil, the biting rate of mosquito can be minimised by up to 80 % (Chavasse and Yap, 1997).

When the coil is burned, the chemicals vaporise with the smoke and inhibit the malaria vector by serving as a barrier that stop the mosquito from getting into the house. Massive sub-micrometre particles and gaseous pollutants are released from the combustion of the remaining materials used in the preparation of the coil. These particles have the potential to get to the lower respiratory tract and get covered with a vast array of organic compounds including PAHs (Shu-Chen *et al.*, 2008).

Mosquito coils that comprise of pyrethroids are able to prevent about 45 – 80 % mosquitoes from gaining access to houses (Ogoma *et al.*, 2012). As a result of the active ingredient (pyrethrum) used in the preparation of the coil, burning mosquito coil is able to repel the mosquitoes and also inhibit them from feeding. Below is a table showing the effect of burning mosquito coils on mosquitoes.

Table 3: Mosquito behavioural reactions induced by burning coils in experimental huts

Active Ingredient	Dose (w/w %)	Vector	Feeding Inhibition (%)	Non-contact irritancy (%)	Deterrence (%)	Mortality (%)
Pyrethrum	0.10	<i>Anopheles gambiae</i> Gillies	54	82	51	16
Pyrethrum	0.10	<i>Culex fatigans</i>	26	58	64	4
Pyrethrum	0.10	<i>Mansonia uniformis</i>	24	93	45	3
Pyrethrum	0.50	<i>Anopheles gambiae</i> Gillies	60	87	58	15
Pyrethrum	0.50	<i>Culex fatigans</i>	46	67	51	7
Pyrethrum	0.50	<i>Mansonia uniformis</i>	69	87	58	15

Source: (Ogoma *et al.*, 2012)

Pyrethroids are one of the major active ingredients used in the preparation of mosquito coils. They are man-made by-products of Pyrethrins from Pyrethrum. Pyrethroids were developed because of the relatively high cost, biodegradability and light instability of natural pyrethrum. Two major types of pyrethroids exist; they are Type I pyrethroids and Type II pyrethroids. The Type I pyrethroids include Permethrin, Allethrin and Lismethrin. They cause nerve axon discharge in insects by retarding sodium channel inactivation. The Type II pyrethroids include Cypermethrin, Deltamethrin and Fenvalerate which cause an even longer prolongation of the sodium influx along the axon leading to continuous nerve depolarization and blockage of axonal conduction. They may also impede inhibiting pathways through binding and altering GABA receptor-mediated chloride channels (Adam and Lawson, 2010).

Mosquito coils made from pyrethroid insecticides especially *d*-allethrin, may contain octachlorodipropyl ether (S-2, S-421) as an active or synergist ingredient. During burning of the coils, the S-2 may be released and this may cause contact and inhalation exposures which can pose serious health challenges. Also, people may be exposed to various levels

of Bis(chloromethyl)ether (BCME) during the usage of those mosquito coils. Bis(chloromethyl)ether (BCME) is made from formaldehyde and hydrogen chloride, burning products obtained from the gradual smouldering of the mosquito coils (Krieger *et al.*, 2003).

Apart from pyrethroids, another insecticide that was used in the formulation of mosquito coils is Organochlorine (OC). They are man-made organic insecticides that contain hydrogen, carbon, chlorine and at times oxygen (Afful *et al.*, 2010). Organochlorine (OC) insecticides are a group of chlorinated compounds that persist in the environment. Organochlorine insecticides are difficult to breakdown into less dangerous substances in the environment (Kang and Chang, 2011). There are three major kinds of Organochlorine insecticides and they are: DDT (dichlorodiphenyltrichloroethane) and its analogues (DDD – dichlorodiphenyldichloroethane), Chlorinated cyclodiene insecticides (e.g., Aldrine, Dieldrin, Endrin, Chlordane, Toxaphene, Heptachlor) and Hexachlorocyclohexanes (HCHs) such as Lindane (Adam and Lawson, 2010).

Though OC insecticides have been the most used pesticide they have now been substituted with Organophosphorus insecticides due to its environmental persistency. This has resulted in the banning of most OCs not merely as agrochemicals for pest control but also for the preparation of other pesticide such as mosquito coils (Bouwman, 2004). The current Stockholm Convention on Persistent Organic Pollutants (POPs) disallowed the use of most OCs. These banned OCs referred to as “dirty dozen” by the convention comprise hexachloro-benzene (HCB), aldrine, dieldrin, endrin, dichlorodiphenyltrichloroethane (DDT), chlordane and heptachlor, (Afful *et al.*, 2010).

Many OC residues and metabolites are immobile, with long half lives in the environment

(El-Mekkawi *et al.*, 2009). These have been linked with a wide array of adverse environmental and human health effects comprising reproduction and birth defects, cancer, immune system dysfunction and endocrine disruptions (Kafilzadeh *et al.*, 2012). According to Esmaili Sari (2002), DDT which is a type of OC is a hydrophobic molecule which disrupts ionic channels like $\text{Na}^+\text{-K}^+$ pumps in nervous cell membrane leading to automatic stimulation of neurons and involuntary contraction of muscles.

2.6 EMISSIONS FROM MOSQUITO COILS

About 1.9 million persons experience untimely death as a result of exposure to indoor smoke from solid fuel burning (Roehr, 2011). Exposure to this indoor smoke is classified as the leading environmental risk factor globally and it is accountable for 3.3 % of all deaths and 2.7 % of all disability-adjusted life years per year (WHO, 2009b). Most of the mosquito coils on the market have some amount of solid fuel products in them (Zhang *et al.*, 2010).

People in residences are usually protected from the inconveniences and disease-carrying mosquitoes by the vaporised insecticides or smoke produced from burning mosquito coils (Shu-Chen *et al.*, 2008). Regardless of its potential benefit as a mosquito repellent, the burning of the mosquito coil generates smoke which may contain air pollutants (such as carbon monoxide, PAHs, VOCs, particulate matter, ketones and aldehydes) of great health concern (Liu *et al.*, 2003).

In developing countries like Ghana, people burn mosquito coils in the evening to prevent mosquito bites and are therefore normally wide-open to the smokes generated from the coils for almost 6 – 8 hours daily. Inhalation of the smoke generated from the burning of the coils has been reported to cause breathing complications, bronchial irritation, eye irritation, asthma, itching and cough (Kurmi *et al.*, 2012).

2.6.1 Carbon Monoxide (CO)

It hinders the blood's potential to convey oxygen to body tissues including essential organs such as heart and brain. Carboxyhaemoglobin (COHb) is formed when the inhaled CO joins with the oxygen carrying haemoglobin. Carbon Monoxide (CO) is a chemical asphyxiant (prevents enough oxygen from reaching the body tissues) (IAPA, 2008). The level of CO will hinge on the effectiveness of fuel combustion and its moisture content (Demirbas, 2004), wet wood fuel generates more smoke, and consequently more carbon monoxide, due to incomplete oxidation of the carbon content (Kurmi *et al.*, 2012).

According to Fierro *et al.* (2001), health effects of CO may include Sudden Infant Death Syndrome (SIDS), early inception of cardiovascular disease, reduced birth weight, reduced exercise performance of young healthy men and upsurge daily death rate. The leading health effect of carbon monoxide is its potential to weaken the oxygen binding capacity of haemoglobin, which can result in dizziness, headaches, tiredness, nausea and breathing difficulty. High exposures can result in coma and death. The gravity of carbon monoxide poisoning depends on concentration, length of exposure, and the health condition of the exposed person. Since carboxyhaemoglobin concentrations in blood accumulate over time, continuous exposure to small levels for a long period can produce a substantial health effect (Weaver *et al.*, 2002).

2.6.2 Sulphur dioxide (SO₂)

Sulphur dioxide is an important gas and a key product resulting from the burning of sulphur compounds. Sulphur dioxide is usually termed as the “smell of burning sulphur. It is produced by volcanoes and in several manufacturing processes. Since coal and petroleum contain innumerable amounts of sulphur compounds, their combustion gives out sulphur dioxide. Additional oxidation of sulphur dioxide, normally in the company of

a catalyst like nitrogen dioxide, forms sulphuric acid (H_2SO_4), and consequently acid rain (Sfetcu, 2014).

When sulphur dioxide is inhaled or if it gets into contact with the eyes or skin, it can disturb the body. Inhalation of SO_2 can also lead to serious irritation of the throat and nose. The irritant effects of SO_2 are as a result of the velocity with which it forms H_2SO_4 on contact with wet membranes (White and Martin, 2010). Symptoms of sulphur dioxide inhalation consist of breathing difficulties, coughing, shortness of breath and tightness in the chest (Patocka and Kuca, 2014). About 90 % of all gasped SO_2 is absorbed in the upper respiratory tract, where it forms sulphurous acid which further oxidizes to form sulphuric acid. Levels between 6 to 12 ppm could lead to instant irritation of the throat and nose. Exposure to SO_2 beyond 20 ppm leads to irritation of the eyes, whereas concentrations of 10,000 ppm irritate wet skin within few minutes. Prolonged exposure to minimal concentrations may be hazardous for individuals with pre-existing cardiopulmonary diseases (Williams-Jones and Rymer, 2000). According to Tunnicliffe *et al.* (1994), acute exposure to SO_2 released during burning of biomass can proliferate bronchial reactivity in normal persons and result in bronchoconstriction in asthmatic persons at concentrations of ~100 ppb and prolong exposure may also escalate susceptibility to viral infections of the lung.

2.6.3 Nitrogen dioxide (NO_2)

Nitrogen dioxide is one of numerous nitrogen oxides. It is a reddish-brown toxic gas which has a characteristic sharp, biting odour and is one of the leading air pollutants (Khan, 2011). The key indoor sources of NO_2 include tobacco smoke and gas, wood, kerosene and coal burning machines such as stoves, ovens, space and water heaters and fireplaces, mostly unflued or poorly kept machines (WHO, 2014c). In the United States, high indoor

levels of NO₂ are more predominant in lesser income housing developments due to inadequate ventilation and small room size (Tunnicliffe *et al.*, 1994).

Nitrogen dioxide is engrossed along the whole respiratory tract, however research has shown that the main target site is the terminal bronchioles (Samoli *et al.*, 2006). The core effect of nitrogen dioxide in human exposure studies has been on bronchial responsiveness, generally observed at levels of $\geq 1,800 \mu\text{g}/\text{m}^3$ in strong people and $\sim 200\text{--}500 \mu\text{g}/\text{m}^3$ in people with asthma (Folinsbee, 1991) or Chronic Obstructive Pulmonary Disease (COPD) (Morrow *et al.*, 1992). Nitrogen dioxide has an increasing effect on the asthmatic response to allergen exposure. A short-term exposure (15 – 30 minutes) to $500 \mu\text{g}/\text{m}^3$ appears to intensify the reaction. Research has suggested that exposure to nitrogen dioxide at levels occurring in densely trafficked areas (15 minutes at $500 \mu\text{g}/\text{m}^3$) can increase allergic inflammatory reaction in the airways without producing indications or pulmonary dysfunction (Barck *et al.*, 2005).

A review of the health outcomes triggered by environmental nitrogen dioxide indicated that there was enough proof that short-term exposure (24 hours), even for average values, $50 \mu\text{g}/\text{m}^3$ NO₂, amplified both hospital cases and death (Latza *et al.*, 2009). The review also stated that there was enough proof of long-term exposure to nitrogen dioxide levels lower than the WHO suggested air quality annual mean guideline ($40 \mu\text{g}/\text{m}^3$) was connected with adverse health effects such as respiratory diseases, hospital admissions, mortality and otitis media. According to Esplugues *et al.* (2011), children are more prone to respiratory disease and more susceptible to indoor pollution, because their immune system and lungs are not fully developed. Symptoms of NO₂ exposure include headache, shortness of breath, chest tightness, cough and pulmonary oedema (Heuer and Scanlan,

2013). Nitrogen dioxide (NO₂) may be a critical concern in low-income generating homes, urban societies where asthma rates are excessively higher (Eggleston *et al.*, 1999) and also small apartment size and limited ventilation may intensify domestic exposure to NO₂ (Zota *et al.*, 2005).

2.6.4 Volatile Organic Compounds (VOCs)

They are organic chemical compounds that have high sufficient vapour pressures under normal conditions to greatly evaporate and go into the sky. Many of these are human-made chemicals and are used as industrial solvents (Ghazali *et al.*, 2012). Volatile organic compounds (VOCs) are a key category of indoor air pollutants, which greatly affect the quality of indoor air and hence affecting human well-being. Prolonged exposure to VOCs will be harmful to human well-being and can result in Sick Building Syndrome (SBS) (Wang *et al.*, 2007). According to research, the concentration of VOC in the indoor environment is usually two to five times greater than the concentration of VOC in the outdoor environment (USEPA, 2016).

Many VOCs are nephrotoxic, hepatotoxic or neurotoxic, or carcinogenic and many can impair the blood components and the cardiovascular system and cause gastrointestinal disturbances (Leslie, 2000). Exposure to volatile organic compounds could make symptoms poorer in asthmatic patients or individuals who are very delicate to chemicals. Common symptoms of volatile organic compounds exposure include eye irritation, headaches, vomiting, nose and throat irritation, dizziness, worsening of asthma symptoms which are as a result of short-term exposures. Prolonged exposures to volatile organic compounds can increase the risk of cancer, liver damage, kidney impairment and Central Nervous System damage (CNS) (MDH, 2011). Some of VOC sources include

formaldehyde and methane. Formaldehyde is categorised as a probable human carcinogen (Kumar *et al.*, 2011) and the emissions from burning one mosquito coil can be as high as that generated from burning fifty one (51) cigarettes (Liu *et al.*, 2003).

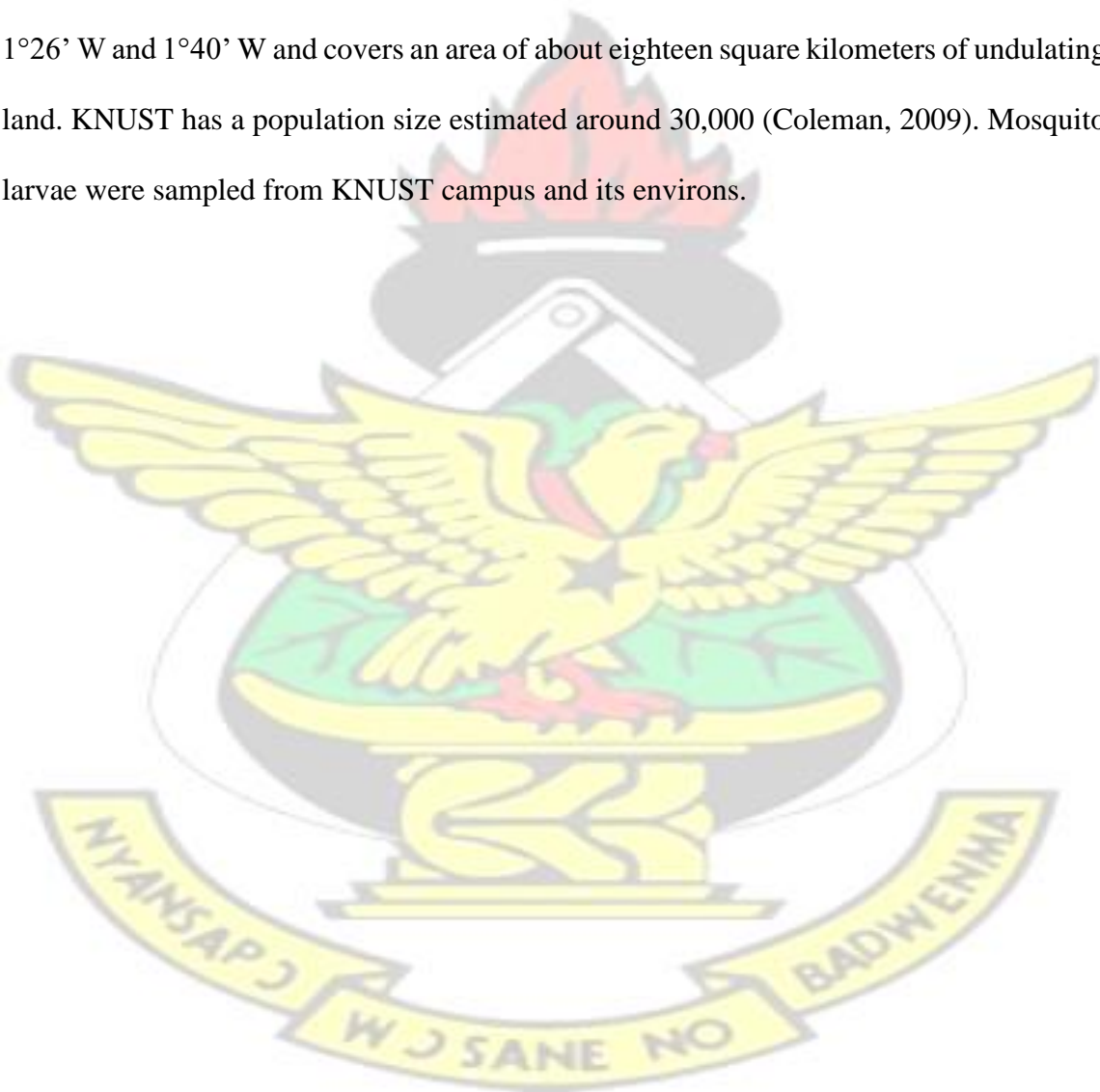
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CHAPTER THREE MATERIALS AND METHODS

3.1 STUDY AREA

The study was conducted at the Kwame Nkrumah University of Science and Technology (KNUST), situated within the Kumasi Metropolitan area, of the Ashanti region of Ghana. KNUST is about eight (8) kilometers away from the centre of Kumasi, the capital of Ashanti region. The study area lies between latitude 6°39' N and 6°47' N, and longitude 1°26' W and 1°40' W and covers an area of about eighteen square kilometers of undulating land. KNUST has a population size estimated around 30,000 (Coleman, 2009). Mosquito larvae were sampled from KNUST campus and its environs.



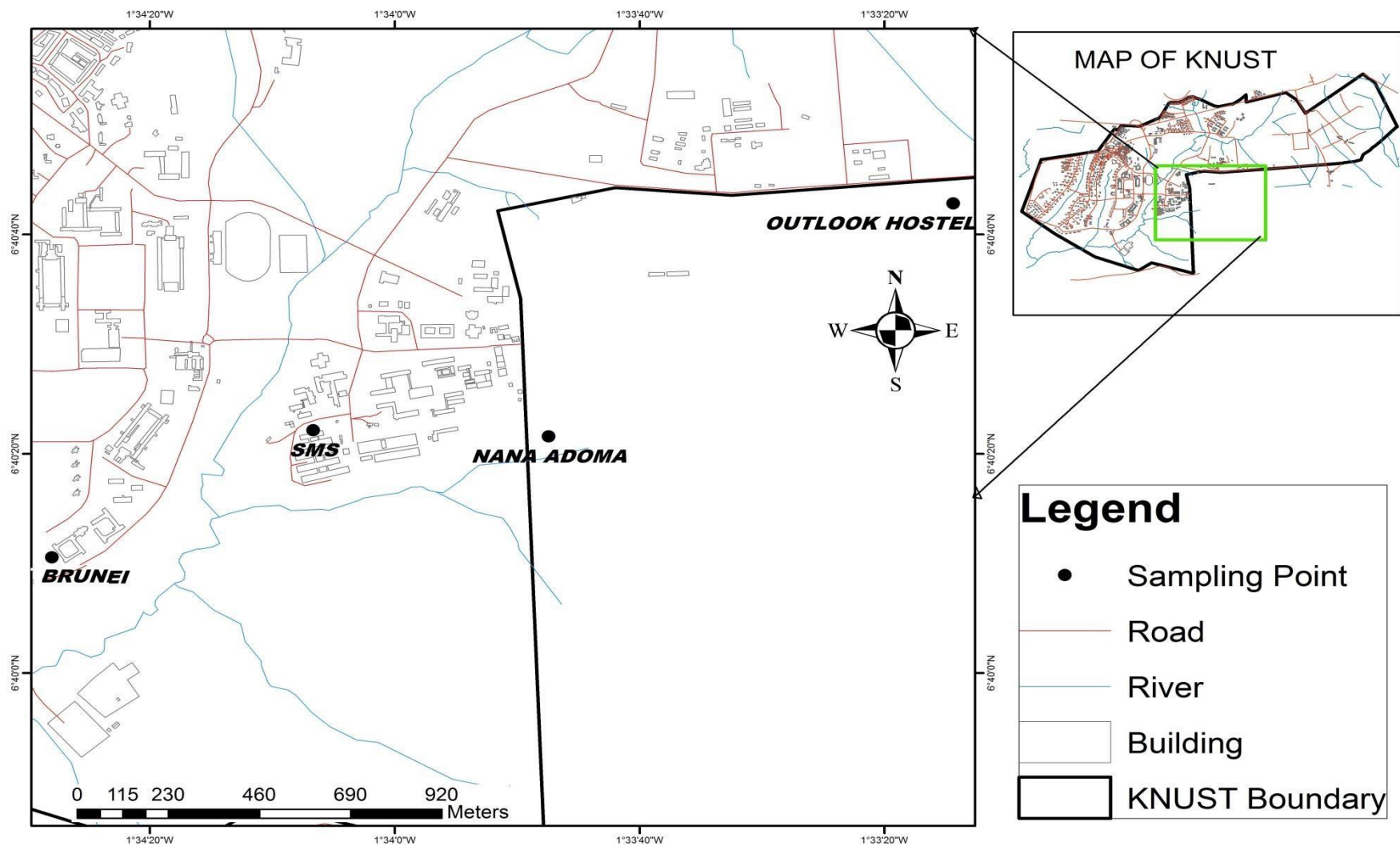


Figure 3: Map showing the sampling sites within the study area

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3.2 SAMPLING OF MOSQUITO LARVAE

Potential breeding sites of mosquitoes were surveyed from January 2016 to June 2016. The characteristics of a breeding site of *Anopheles*; aquatic, little to no pollution, temporal, not more than 2 km from human settlements, existence of vegetation, stagnant, shallow and well sunlit conditions (Baffour-Awuah, 2012) were noted which made the identification of larvae easy. There are many wetlands within the University (KNUST) campus which are mostly used by encroachers for vegetable cultivation. Most of these farmers created pools of water between the ridges of the beds on their farms to reduce the stress of getting water for irrigation (Baffour-Awuah, 2012). These provided breeding sites for mosquitoes. *Anopheles* larvae were collected from such breeding sites (Plates 3, 4 and 5) and reared in the insectary. They were identified by their parallel position on the water surface and were scooped with dippers into a rubber bucket (Plate 2).



Plate 2: Collection of mosquito larvae from a vegetable farm



Plate 3: *Anopheles* breeding sites in between two ridges on a vegetable farm



Plate 4: *Anopheles* breeding sites in stagnant water



Plate 5: *Anopheles* breeding sites in footprints

3.3 REARING OF MOSQUITOES IN INSECTARY

3.3.1 Larvae and Pupae

The larvae were brought to the insectary of Kumasi Centre for Collaborative Research (KCCR) at KNUST Campus. The door to the insectary was always firmly closed after entering or leaving it. The floor and insect rearing cages were cleaned daily to limit predators (ants and spiders) to the insectary. The cages were kept on a shelf which had its legs standing in petri dishes containing vegetable cooking oil to prevent ants and other predators from climbing.

At the insectary, the larvae were transferred into larval bowls and were further sorted to remove any other species apart from the *Anopheles*. The larvae were reared in their natural habitats' water since any abrupt change in their environment would be unfavourable and may cause mortality. The larvae were maintained in an insectary at 25°C and 75 – 80 % relative humidity with a photoperiod of 12:12 hours and fed with Elite fish flakes meal

which was ground and sprinkled evenly on the surface of their habitats' water daily. Extra caution was taken to ensure that the fish meal does not form foam over the surface of the water since this can prevent the larvae from inhaling oxygen which is needed for their survival. The larval bowls were covered with nets so as to prevent larvae that may emerge into pupa and then to adulthood from flying away. The mosquito larvae went through the four stages, 1st, 2nd, 3rd and 4th instar. After the 4th instar stage, it pupated. Pupae were then collected with a Pasteur pipette and placed in beakers containing water in the cages for them to emerge into adults.



Plate 6: Mosquito larval bowls covered with nets arranged on a shelf

3.3.2 Adult Mosquitoes

All adult mosquitoes were fed on 10 % sugar solution imbibed in cotton wool. The cotton wool was changed every 2 days to prevent it from being fermented. 3 – 6 days old nonblood fed female adult mosquitoes were then collected and their susceptibility tested against the mosquito coil. For the purpose of the susceptibility test, adult female *Anopheles* mosquitoes were sorted from the males. The proboscis of the female mosquito is

comparatively smooth, not bushy whereas the male mosquitoes have a feather-like proboscis.



Plate 7: Rearing cages for adult *Anopheles* species arranged on a shelf

3.4 SYNTHETIC MOSQUITO COIL

The effects of five different brands of synthetic mosquito coil on the mortality of female *Anopheles* mosquito were investigated. The mosquito coils were purchased from retail outlets at Adum, a key commercial centre in Kumasi, the capital of Ashanti Region of Ghana. The coils were also later evaluated for gaseous pollutant emissions that may emanate from their use. General information about the different brands of coils are summarized in Table 4.

Table 4: General information of tested mosquito coils

ID No.	Country of Origin	Shape	Colour	Mass per coil (g)	Active Ingredient	Burning Time (h)
MC 1	China	Spiral	Black	16.5	0.25 % Esbiothrin	7.50
MC 2	China	Spiral	Black	20.0	0.03 % Dimefluthrin	9.67
MC 3	China	Spiral	Black	17.5	0.03 % Dimefluthrin	8.00

MC 4	Spain	Spiral	Black	16.6	0.20 % D-allethrin	8.00
MC 5	China	Spiral	Black	17.0	0.08 % Meperfluthrin	8.00

3.5 EXPERIMENTAL CHAMBERS

The mosquito mortality test and the gaseous emissions test were undertaken in experimental chambers of varying room sizes and indoor conditions (Plate 8). The experimental chambers were constructed with wood. There were three different room sizes with dimensions 2 m × 2 m, 3 m × 3 m and 4 m × 4 m, and a uniform room height of 2.12 m. Each room was fixed with a door of dimensions 197 cm × 76 cm and for windows, each of dimension 87 cm × 65 cm. The doors and windows were protected with net. The floor of each room was laid with nylon carpet, over-laid with white papers. Experiments were conducted under ventilated conditions (with windows opened) and poor ventilation (with windows closed). The average temperature and relative humidity in the experimental rooms are indicated in Table 5.

Table 5: Average temperature and relative humidity in the experimental rooms

Room Size (m ³)	Ventilated Condition		Poorly Ventilated Condition	
	Temperature (°C)	Relative Humidity (%)	Temperature (°C)	Relative Humidity (%)
8.5	28.37 ± 1.96	70.77 ± 5.07	30.08 ± 1.90	66.24 ± 3.15
19	28.56 ± 0.81	71.77 ± 4.30	30.62 ± 1.40	66.10 ± 4.33
34	28.65 ± 0.81	69.53 ± 3.96	27.90 ± 0.54	67.09 ± 4.29



Plate 8: Experimental Rooms (34 m³, 19 m³ and 8.5 m³. From Left to Right)

3.6 SUSCEPTIBILITY TEST ON MOSQUITO COILS

The experiment was conducted under two conditions: (i) when the windows were opened to allow natural ventilation and (ii) when the windows were closed to provide poor ventilation. Prior to the beginning of the experiment, 50 female *Anopheles* mosquitoes (3 – 6 days old, sucrose-fed) were released into each of the rooms without the lit mosquito coil to serve as a control. The control mosquitoes were treated in the same way as the exposed mosquitoes; they were tested under the same conditions. The objective of the inclusion of the controls was to provide an estimate of natural mortality during the test and also to account for all variables that may induce mortality other than the insecticide in the mosquito coils being tested. Afterwards, one brand of the coil placed on a metal stand provided inside the coil packet was lit and placed at the centre of each room and 50 female *Anopheles* mosquitoes (3 – 6 days old, sucrose-fed) were gently transferred from their cages into the experimental room. The experiment was carried out at night (from 6pm until dawn of the next day) and was repeated in triplicates for each brand of mosquito coil. The burning time of each brand of mosquito coil was observed and recorded.

Considering all the experimental set-ups, a total of nine hundred (900) mosquitoes were exposed to each brand of mosquito coil. Four (4) days was allowed in between tests to ensure

a complete breakdown of insecticides from previous test that could influence new test. At the end of the burning period of the mosquito coils, the number of knock-down mosquitoes fallen unto the white floor of each room were counted and recorded. An adult mosquito was considered to be alive if it was able to fly, irrespective of the number of legs left. Mosquitoes that have lost their wings and could no longer fly were considered moribund and were therefore counted as dead.

3.7 MONITORING OF INDOOR AIR POLLUTANT CONCENTRATIONS

The quality of the indoor air was measured using Aeroqual Series 500 (S500) gas monitors (Aeroqual Limited; Auckland, New Zealand). First the portable gas monitors with the appropriate sensor heads were placed in the experimental rooms without lit mosquito coils to know the level of gases in the rooms before lighting mosquito coils in the rooms. This helped to determine the actual amount of emissions attributable to the coils. The gases monitored, sensor type, sensor range and the minimum detection limits are indicated in Table 6. The device was also capable of collecting meteorological data such as temperature and humidity.

Table 6: Sensor specifications for Aeroqual ambient sensors

Gas	Sensor Type*	Sensor Range (mg/m ³)	Minimum Detection Limit (mg/m ³)
CO	GSE	0 – 123	0.25
TVOC	PID	0 – 70	0.03
NO ₂	GSE	0 – 2	0.01
SO ₂	GSE	0 – 28	0.11

*GSE: Gas Sensitive Electrochemical; PID: Photo Ionization Detector

This instrument was preferred for its simplicity and reliability in setup, simplicity of handling, and rapidity in obtaining the gas concentration directly. The measurement units were in milligram per cubic meter (mg/m³). The sensors were warmed up to burn off any contaminants prior to usage. The monitors were also kept in Stand By mode when not in

use to keep the sensors heated and also prevented the build-up of contaminants. The Aeroqual S500 gas monitors were programmed to log at 5 minutes intervals. Thus, it recorded average concentrations of the gases as well as temperature and relative humidity continuously at 5 minutes interval during the burning and post burning of the mosquito coils.

Some earlier studies (Chan *et al.*, 2009; Song *et al.*, 2007; Wallace *et al.*, 1987) showed that the experimental device should be put between 1m to 1.5m above the floor level. However, according to the manufacturer of the Aeroqual 500 Series, the device can be placed anywhere in the experimental room (Ghazali *et al.*, 2012). The sampling period started from the beginning of the burning of the mosquito coils until one (1) hour after burning. The sampling period is dependent on the burning time of each tested mosquito coil. Logged data from the monitor's memory were downloaded onto a laptop computer using Aeroqual S500 gas monitor software version S500 V6.4.



Plate 9: Aeroqual S500 gas monitor, sensor heads and temperature/relative humidity sensor

3.8 ESTIMATION OF VENTILATION RATES

The ventilation rate of the experimental rooms was estimated using the CO removal rate. The gas generated from the burning of the mosquito coils were concurrently monitored during the sampling period. After extinguishing the coils, concentration started to decrease according to the rate of natural ventilation. The natural removal of the gas after the burning of the coils in the rooms with open and closed windows was used to estimate the natural ventilation rates of the experimental conditions. The diurnal mean natural ventilation rate was interpreted as the mean of the removal rates during the post burning period. The ventilation rates of the experimental rooms were estimated by using a single-compartment mass balance model (Fan and Zhang, 2001). It was projected that the elimination of the gas-phase compounds in the chambers was instigated only by ventilation; the removal rate of gas-phase compounds was equal to the air exchange rate in the chamber. Other factors such as diffusion, deposition and coagulation control the removal of gases (Liu *et al.*, 2003). The equation below was used to estimate the gas removal rate k . A real-time gas mass concentration in the rooms during the post-burning period was needed for the calculation.

$$C = C_{\max} (e^{-k(t-T)}), \text{ when } t > T \quad \text{Equation 1}$$

Where k is the total removal rate of pollutant (hr^{-1}), T is the time (hr) at which the coil was quenched, t is the time after T , C_{\max} is the maximum pollutant concentration at the time (T) when the mosquito coil was quenched and C is the pollutant concentration after T .

3.9 DATA ANALYSIS

Statistical analysis was performed using Statistical Package for the Social Sciences (SPSS) software package, version 23.0. The values were analysed by independent sample t-test

and one way analysis of variance (ANOVA). In all statistical test a value of $p < 0.05$ was considered significant. The corrected percentage mortality was calculated by using Abbott's formula (Abbott, 1987).

$$\text{Corrected mortality (\%)} = \frac{\text{Mortality in treatment (\%)} - \text{Mortality in control(\%)}}{100 - \text{Mortality in control (\%)}} \times 100$$



CHAPTER FOUR RESULTS

This section presents the results of the susceptibility test and emissions from the tested mosquito coils.

4.1 EFFICACY OF MOSQUITO COILS IN TERMS OF MOSQUITO MORTALITY

4.1.1 Effect of Ventilation on Mosquito Mortality

The corrected mortalities (%) of the tested mosquito coils recorded for the three experimental rooms under ventilated and poorly ventilated conditions are presented in Figures 4, 5 and 6. Mortalities in 34 m³ room ranged from 24.44 to 39.26 % and 33.33 to 53.33 % under ventilated and poorly ventilated conditions respectively. There was a significant difference ($p < 0.05$) in mortalities under the two experimental conditions.

Also, mortalities in 19 m³ room ranged from 34.78 to 44.93 % and 40.74 to 60 % under ventilated and poorly ventilated conditions respectively. Statistically, there was a significant difference ($p < 0.05$) in the mortalities resulting from the mosquito coils under both conditions. In the 8.5 m³ room, mortalities of mosquitoes ranged from 38.52 to 52.59 % and 46.21 to 63.64 % under ventilated and poorly ventilated conditions respectively. Statistically, there was a significant difference ($p < 0.05$) in the mortalities under the two experimental conditions.

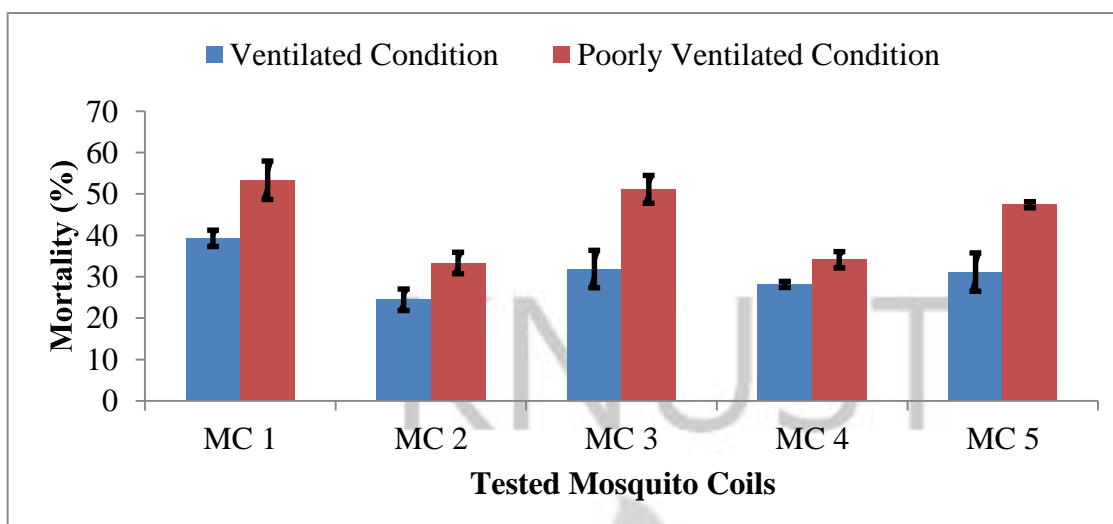


Figure 4: Mortality of mosquitoes in 34 m³ Room

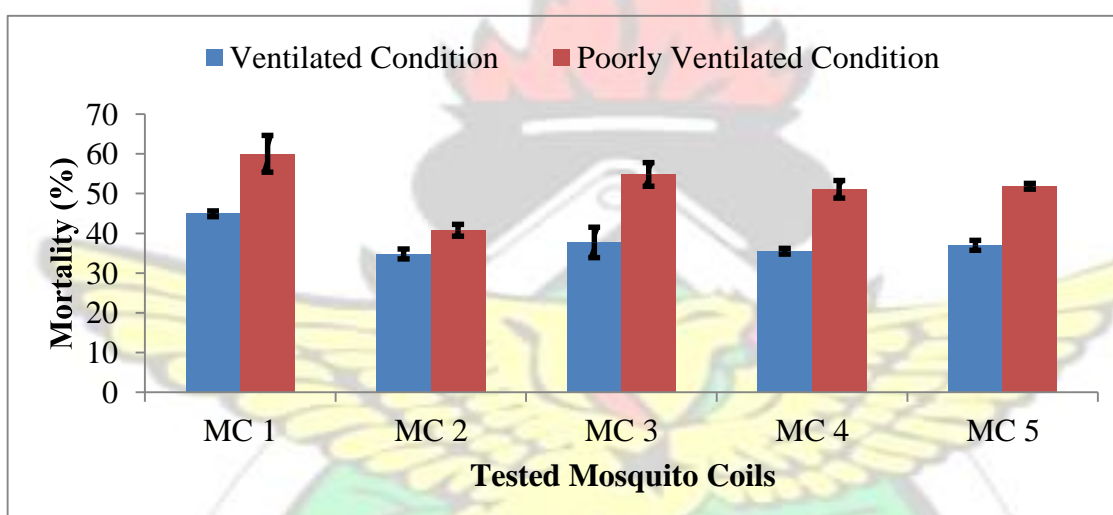


Figure 5: Mortality of mosquitoes in 19 m³ Room

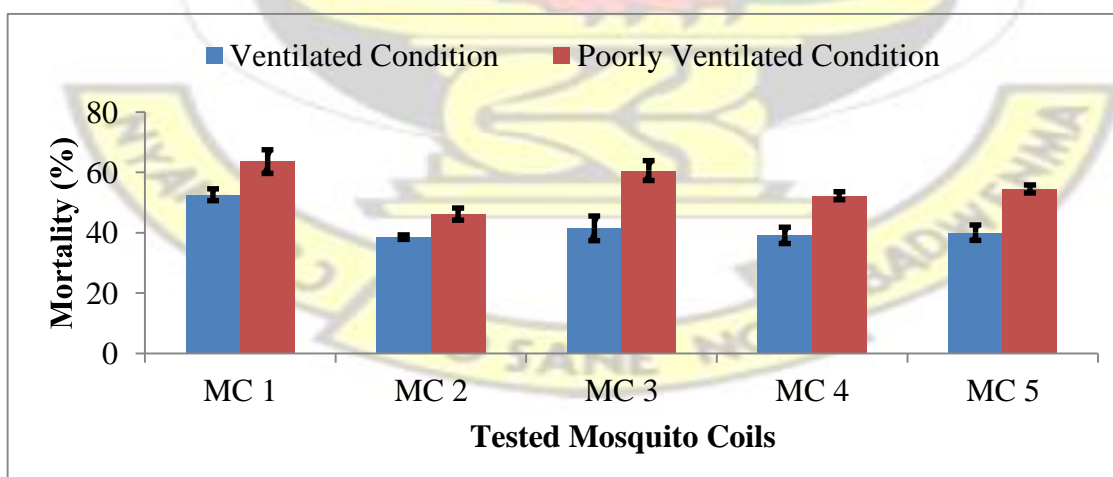


Figure 6: Mortality of mosquitoes in 8.5 m³ Room The highest mortality was recorded in MC 1 and the lowest mortality in MC 2 for all the three experimental rooms under both

conditions (Figures 4, 5 and 6). However, statistically there was no significant differences between mortalities among the five tested coils ($p > 0.05$) under ventilated conditions but under poorly ventilated conditions, statistical difference ($p < 0.05$) existed only between the mortalities resulting from MC 1 and MC 2.

The efficacies of the tested mosquito coils had a decreasing mortality trend of MC 1 > MC 3 > MC 5 > MC 4 > MC 2 in all the three experimental rooms under both ventilated and poorly ventilated conditions.

4.1.2 Effect of Room Sizes on Mosquito Mortality

Considering the effect of room sizes on mosquito mortality, 8.5 m³ room recorded the highest mortality followed by 19 m³ room and 34 m³ room recording the least mortality as shown in Table 7 for all the five tested coils in both the ventilated and poorly ventilated rooms. There was statistically no significant difference ($p > 0.05$) between mortalities in 8.5 m³ and 19 m³ rooms, and also 19 m³ and 34 m³ rooms. However, there was a significant difference ($p < 0.05$) between 8.5 m³ and 34 m³ rooms.

Table 7: Corrected mortality (% \pm SD) of mosquitoes to 5 different mosquito coils evaluated in 3 different room sizes

Coil ID (AI)*	Ventilated Condition			Poorly Ventilated Condition		
	8.5 m ³ Room	19 m ³ Room	34 m ³ Room	8.5 m ³ Room	19 m ³ Room	34 m ³ Room
MC 1 (0.25 % esbiothrin)	52.59 \pm 3.40	44.93 \pm 1.25	39.26 \pm 3.39	63.64 \pm 6.82	60.00 \pm 8.01	53.33 \pm 8.01
MC 2 (0.03 % dimefluthrin)	38.52 \pm 1.28	34.78 \pm 2.18	24.44 \pm 4.45	46.21 \pm 3.47	40.74 \pm 2.56	33.33 \pm 4.45
MC 3 (0.03 % dimefluthrin)	41.48 \pm 7.15	37.68 \pm 6.64	31.85 \pm 7.81	60.61 \pm 5.72	54.82 \pm 5.13	51.11 \pm 5.88
MC 4 (0.20 % d-allethrin)	39.13 \pm 4.69	35.51 \pm 1.26	28.15 \pm 1.28	52.27 \pm 2.28	51.11 \pm 3.85	34.07 \pm 3.40
MC 5 (0.08 % meperfluthrin)	40.00 \pm 4.44	36.96 \pm 2.18	31.11 \pm 8.01	54.55 \pm 2.28	51.85 \pm 1.28	47.41 \pm 1.28

* AI: Active Ingredient

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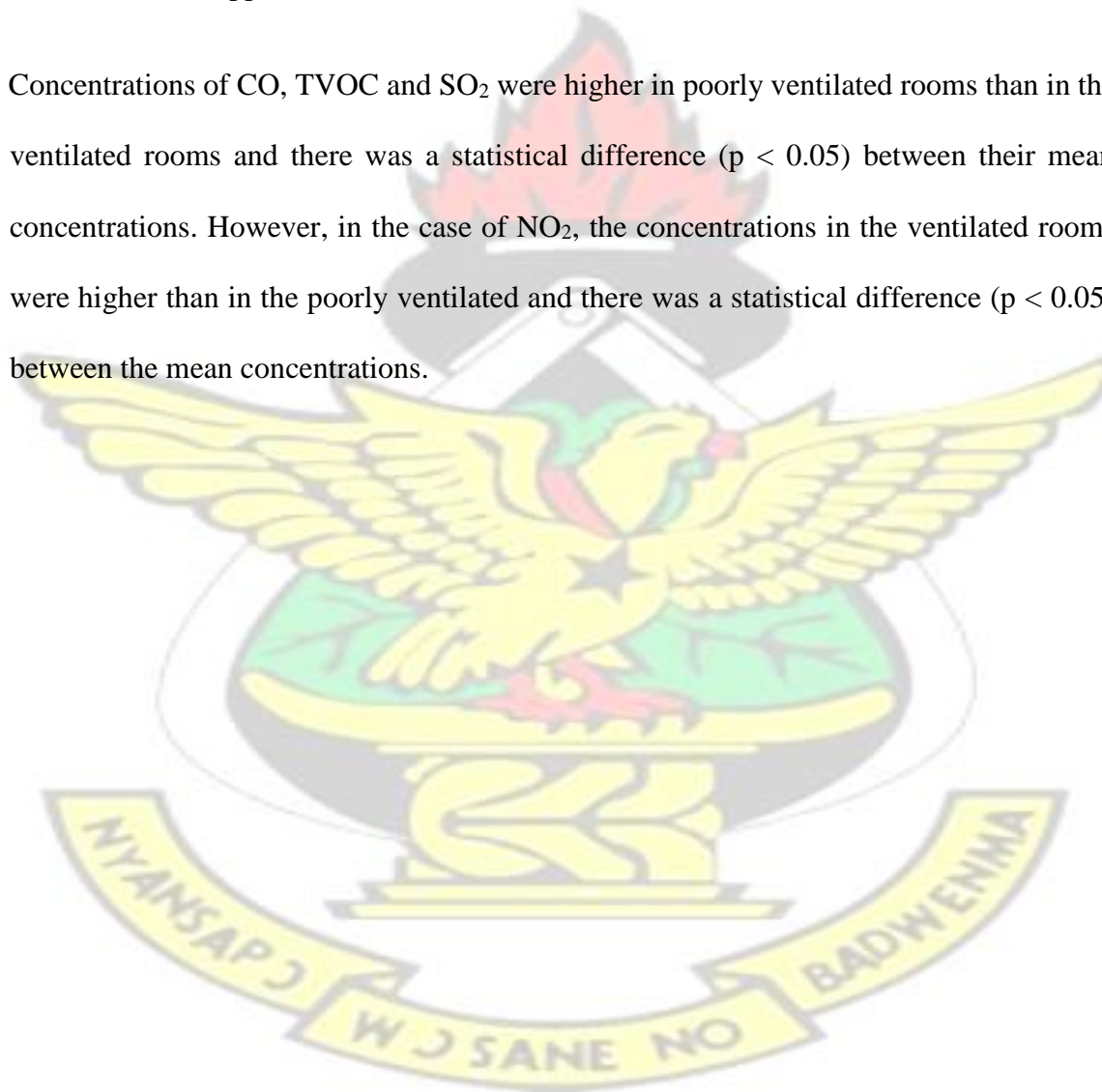


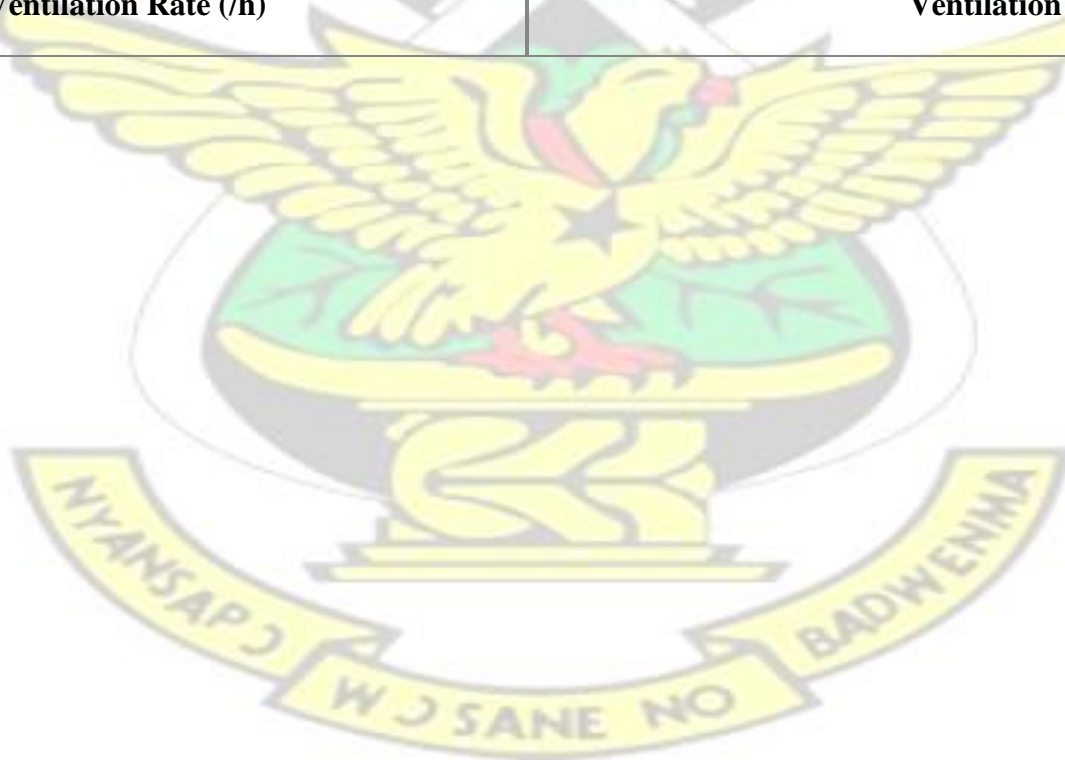
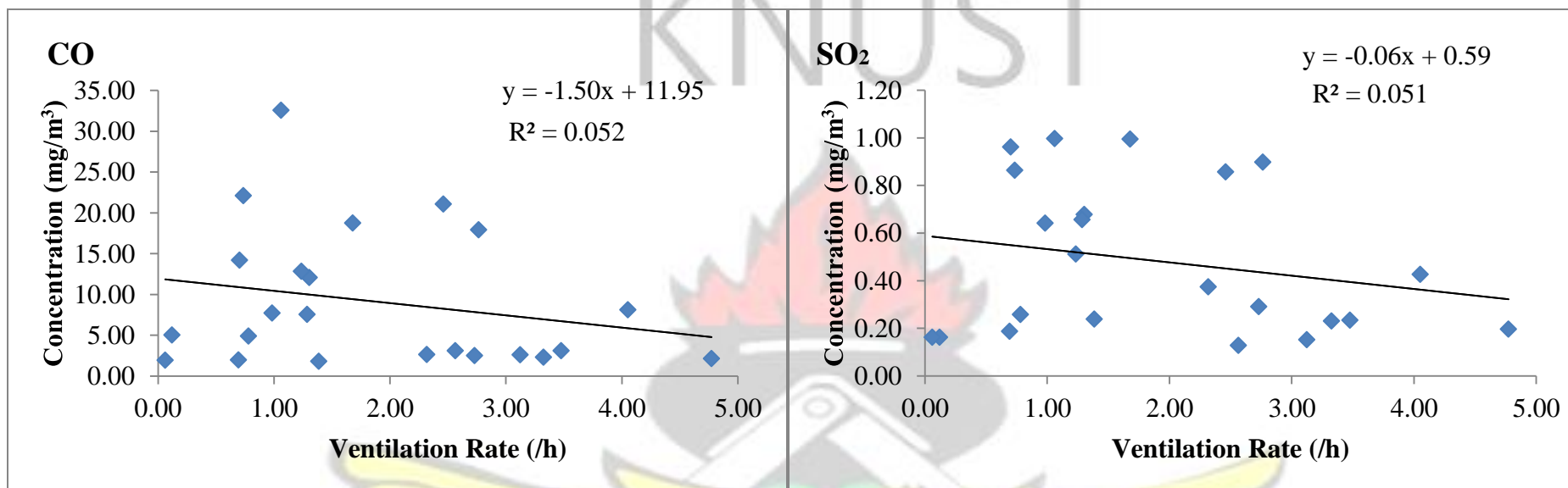
4.2 GASEOUS POLLUTANT EMISSIONS FROM MOSQUITO COILS

4.2.1 Effect of Ventilation on Mosquito Coil Emissions

Figure 7 shows the relationship between pollutant concentrations from mosquito coils and ventilation rates in the experimental room under ventilated and poorly ventilated conditions. Concentration of pollutants resulting from the burning of the mosquito coils decreased as ventilation rates increased except for NO_2 where the concentration and ventilation rates appeared to be on a horizontal line.

Concentrations of CO , TVOC and SO_2 were higher in poorly ventilated rooms than in the ventilated rooms and there was a statistical difference ($p < 0.05$) between their mean concentrations. However, in the case of NO_2 , the concentrations in the ventilated rooms were higher than in the poorly ventilated and there was a statistical difference ($p < 0.05$) between the mean concentrations.





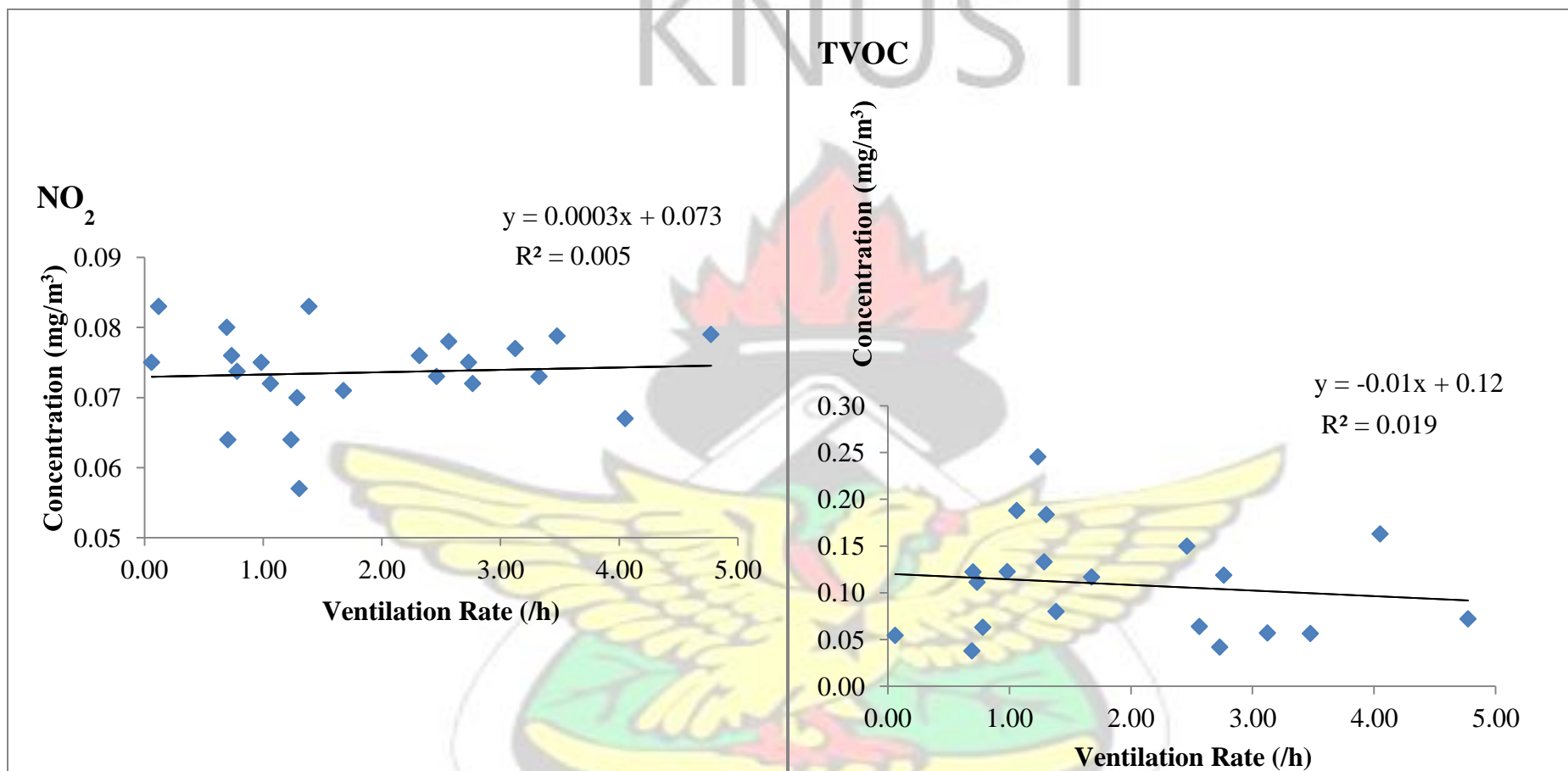


Figure 7: Relationship between pollutant concentrations and ventilation rates in experimental rooms

The concentration of pollutants recorded from five different mosquito coils in three different experimental rooms are given in Table 8. Concentrations of CO under ventilated conditions ranged from 1.30 to 5.04 mg/m³ and 7.56 to 32.60 mg/m³ under poorly ventilated conditions. There was a significant difference ($p < 0.05$) in CO concentrations generated from the coils and the control rooms (without lit coil) under both ventilated and poorly ventilated conditions. With the exception of MC 2 and MC 4 (in all the three rooms); MC 3 and MC 4 (only in 34 m³) which did not differ, there was a statistically significant difference ($p < 0.05$) in CO concentrations emitted from the coils in the 8.5 m³, 19 m³ and 34 m³ rooms under poorly ventilated conditions. Under ventilated conditions, CO emitted from most of the mosquito coils did not vary significantly ($p > 0.05$) especially in the 8.5 m³ room.

TVOC concentrations also ranged from 0.04 to 0.08 mg/m³ under ventilated conditions and 0.11 to 0.25 mg/m³ under the poorly ventilated conditions. There was a significant difference ($p < 0.05$) in TVOC concentrations generated from the coils and the control rooms (without lit coil) under both ventilated and poorly ventilated conditions. However, under both ventilated and poorly ventilated conditions, TVOC emitted from some of the mosquito coils did not vary significantly ($p > 0.05$) especially in the 34 m³ room.

The concentrations of NO₂ were slightly higher in the ventilated rooms (0.07 – 0.08 mg/m³) than in the poorly ventilated rooms (0.06 – 0.08 mg/m³). Some of the concentrations recorded in the control rooms did not differ ($p > 0.05$) from the emissions generated from the coils in the rooms under both ventilated and poorly ventilated conditions. Among the emissions from the individual coils, some of them did not differ ($p > 0.05$) in the rooms under both ventilated and poorly ventilated conditions.

Concentrations of SO₂ under the poorly ventilated conditions were higher (0.34 – 1.00 mg/m³) than concentrations in ventilated conditions (0.13 – 0.37 mg/m³). There was a

significant difference ($p < 0.05$) in SO_2 concentrations generated from the coils and concentrations in the control rooms (without lit coil) under both ventilated and poorly ventilated conditions. Apart from the emissions between MC 1 and MC 5 (in 8.5 m^3 and 19 m^3 rooms), there was a statistically significant difference ($p < 0.05$) between SO_2 concentrations recorded for the mosquito coils under the poorly ventilated conditions. Under ventilated conditions, there was a significant difference ($p < 0.05$) in the concentrations between the mosquito coils in the rooms except the 34 m^3 room where few mosquito coils (MC 1 and MC4; MC 1 and 5; MC 3 and MC 5; MC 4 and 5) did not differ in the emissions generated.

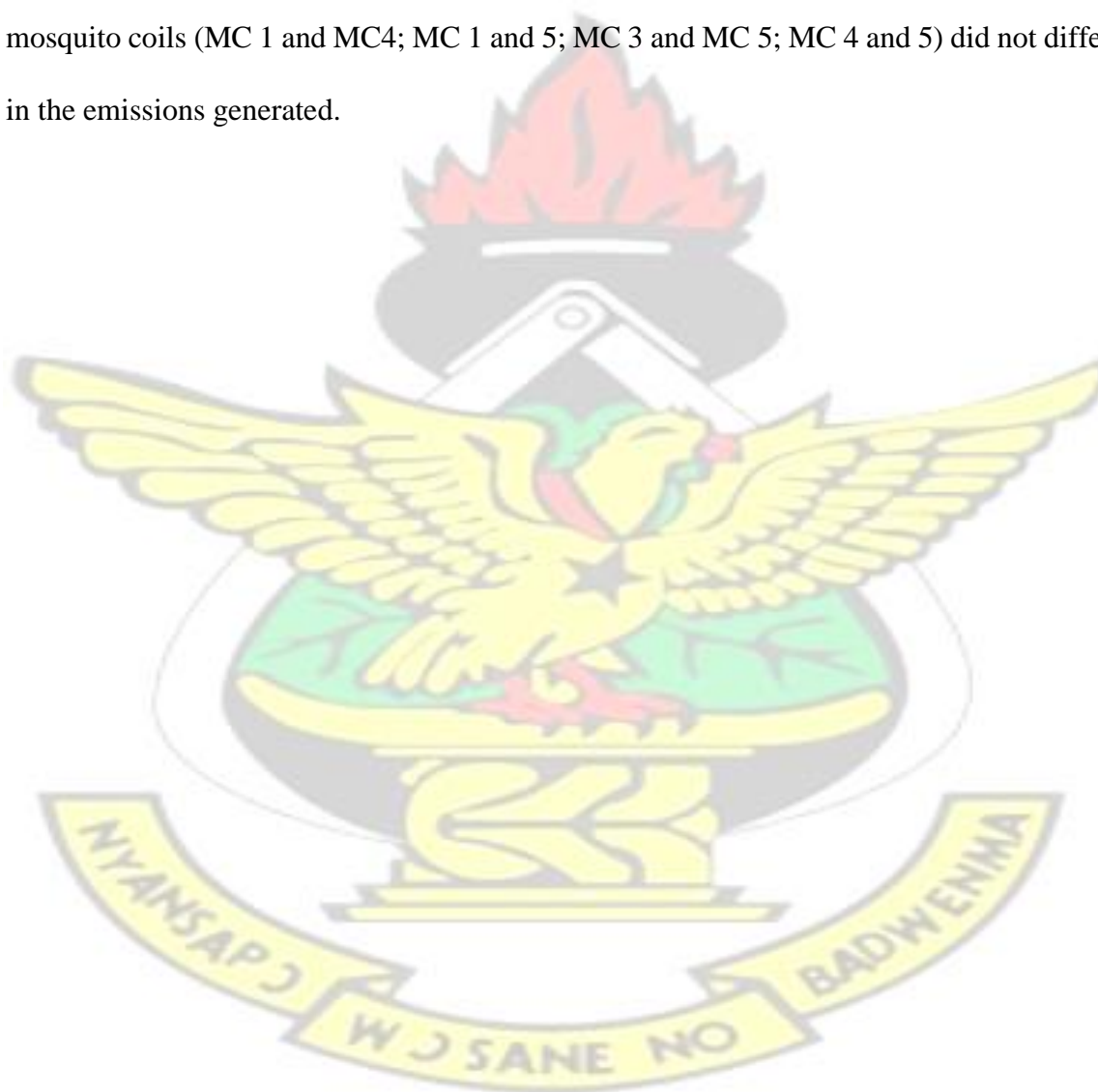


Table 8: Levels of pollutants (Mean \pm SD) mg/m³ recorded in the experimental rooms from five different mosquito coils

Mosquito coil	Ventilated Condition				Poorly Ventilated Condition			
	CO	TVOC	NO ₂	SO ₂	CO	TVOC	NO ₂	SO ₂
Room Size: 8.5 m ³								
MC 1	1.91 \pm 1.31	0.05 \pm 0.03	0.08 \pm 0.01	0.34 \pm 0.10	17.93 \pm 4.98	0.12 \pm 0.06	0.07 \pm 0.01	0.90 \pm 0.22
MC 2	1.95 \pm 1.13	0.05 \pm 0.03	0.07 \pm 0.01	0.16 \pm 0.03	7.56 \pm 2.20	0.13 \pm 0.02	0.07 \pm 0.01	0.66 \pm 0.13
MC 3	1.83 \pm 1.90	0.08 \pm 0.04	0.08 \pm 0.01	0.24 \pm 0.05	11.20 \pm 3.70	0.14 \pm 0.02	0.07 \pm 0.00	0.36 \pm 0.05
MC 4	2.51 \pm 1.15	0.04 \pm 0.02	0.07 \pm 0.00	0.29 \pm 0.08	8.12 \pm 2.13	0.16 \pm 0.01	0.07 \pm 0.01	0.43 \pm 0.05
MC 5	2.15 \pm 1.57	0.07 \pm 0.02	0.08 \pm 0.01	0.20 \pm 0.06	21.09 \pm 4.04	0.15 \pm 0.02	0.07 \pm 0.00	0.86 \pm 0.14
Control	*BDL	*BDL	0.07 \pm 0.01	*BDL	*BDL	0.05 \pm 0.03	0.06 \pm 0.02	0.16 \pm 0.02
Room Size: 19 m ³								
MC 1	2.66 \pm 1.61	*BDL	0.08 \pm 0.01	0.37 \pm 0.10	18.75 \pm 4.13	0.12 \pm 0.02	0.07 \pm 0.01	0.99 \pm 0.22
MC 2	1.99 \pm 0.82	0.04 \pm 0.02	0.08 \pm 0.01	0.19 \pm 0.04	12.09 \pm 4.92	0.18 \pm 0.02	0.06 \pm 0.00	0.68 \pm 0.18
MC 3	5.04 \pm 2.01	BDL	0.08 \pm 0.01	0.16 \pm 0.03	10.15 \pm 3.02	0.19 \pm 0.04	0.06 \pm 0.01	0.38 \pm 0.09
MC 4	1.30 \pm 0.99	0.04 \pm 0.02	0.08 \pm 0.01	0.26 \pm 0.05	12.85 \pm 2.78	0.25 \pm 0.02	0.06 \pm 0.01	0.51 \pm 0.12
MC 5	3.10 \pm 2.05	0.06 \pm 0.02	0.08 \pm 0.01	0.13 \pm 0.04	32.60 \pm 7.76	0.19 \pm 0.02	0.07 \pm 0.01	1.00 \pm 0.23
Control	*BDL	*BDL	0.07 \pm 0.01	*BDL	*BDL	0.05 \pm 0.03	0.05 \pm 0.02	0.25 \pm 0.11
Room Size: 34 m ³								
MC 1	2.33 \pm 1.32	*BDL	0.07 \pm 0.00	0.23 \pm 0.05	14.21 \pm 3.96	0.12 \pm 0.02	0.06 \pm 0.01	0.96 \pm 0.21
MC 2	2.61 \pm 0.96	0.06 \pm 0.01	0.08 \pm 0.01	0.15 \pm 0.03	7.73 \pm 1.51	0.12 \pm 0.02	0.07 \pm 0.01	0.64 \pm 0.16
MC 3	4.91 \pm 1.00	0.06 \pm 0.03	0.07 \pm 0.00	0.26 \pm 0.03	9.86 \pm 4.25	0.12 \pm 0.02	0.07 \pm 0.01	0.44 \pm 0.12
MC 4	3.13 \pm 0.92	0.06 \pm 0.02	0.08 \pm 0.01	0.23 \pm 0.04	8.25 \pm 2.38	0.12 \pm 0.01	0.07 \pm 0.00	0.38 \pm 0.12
MC 5	2.42 \pm 1.30	0.05 \pm 0.04	0.08 \pm 0.01	0.25 \pm 0.04	22.12 \pm 7.25	0.11 \pm 0.01	0.08 \pm 0.01	0.86 \pm 0.17
Control	*BDL	*BDL	0.07 \pm 0.00	*BDL	*BDL	0.06 \pm 0.02	0.06 \pm 0.00	0.15 \pm 0.08

* BDL: Below Detection Limit

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4.2.2 Effect of Room Sizes on Mosquito Coil Emissions

The concentration of pollutants (CO, TVOC, NO₂ and SO₂) recorded in the three experimental rooms are presented in Figure 8. The concentration of CO in the ventilated room increased from the 8.5 m³ to 34 m³ rooms. There was a statistical difference ($p < 0.05$) in the concentrations of CO between the three rooms under ventilated condition. Under poorly ventilated condition, the peak concentration was recorded in the 19 m³ room and there was no statistical difference ($p > 0.05$) in the concentrations between the 34 m³ and 8.5m³ rooms.

The TVOC concentration under ventilated conditions did not differ much across the three rooms. However, there was a statistically significant difference ($p < 0.05$) in the concentrations between the three rooms. The peak concentration of TVOC under poorly ventilated condition was recorded in 19 m³ room and the 34 m³ room recorded the least concentration. Significant difference ($p < 0.05$) existed in the concentrations between the three rooms.

Concentration of NO₂ under ventilated condition was the same (0.08 mg/m³) across the three rooms. Under poorly ventilated condition, NO₂ concentrations were also the same (0.07 mg/m³) in the 8.5 m³ and 34 m³ rooms. The least concentration was recorded in 19 m³ room (0.06 mg/m³). However, apart from the concentrations between the 8.5 m³ and 34 m³ rooms under both ventilated and poorly ventilated conditions, there was a significant difference ($p < 0.05$) in the concentrations of NO₂ recorded between the rooms (i.e. between 8.5 m³ and 19 m³; 19 m³ and 34 m³).

The highest concentration of SO₂ (0.24 mg/m³) was recorded in the 8.5 m³ room with the 19 m³ and 34 m³ rooms recording the same concentration (0.22 mg/m³) under ventilated

condition. Apart from the concentrations between the 19 m³ and 34 m³ rooms, there was a

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significant difference ($p < 0.05$) in the concentrations between the rooms (i.e. between 8.5 m³ and 19 m³; 8.5 m³ and 34 m³). Under poorly ventilated condition, the highest concentration was recorded in the 19 m³ room, followed by the 34 m³ and the 8.5 m³ rooms recorded the least concentration. Also, apart from the concentrations between the 8.5 m³ and 34 m³ rooms, statistically significant difference ($p < 0.05$) existed in the concentrations between the rooms (i.e. between 8.5 m³ and 19 m³; 19 m³ and 34 m³).



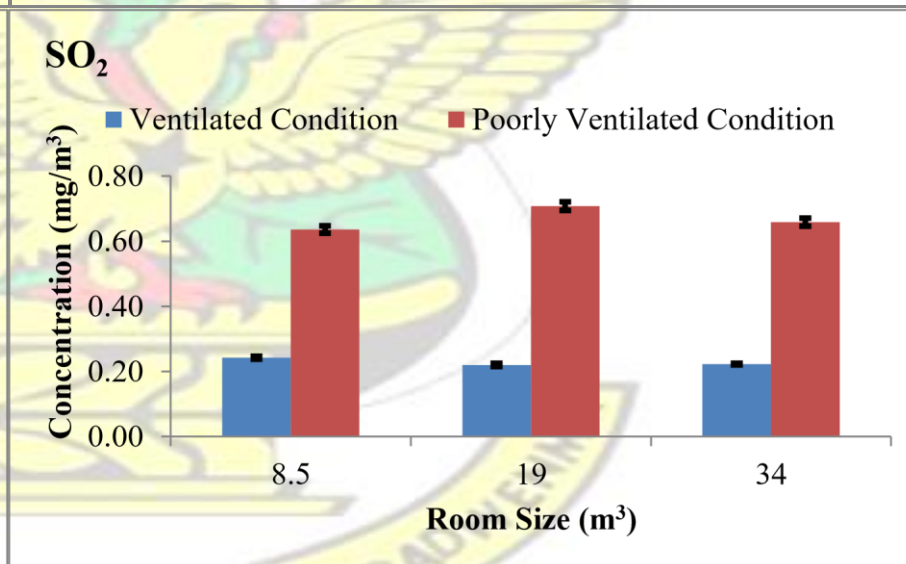
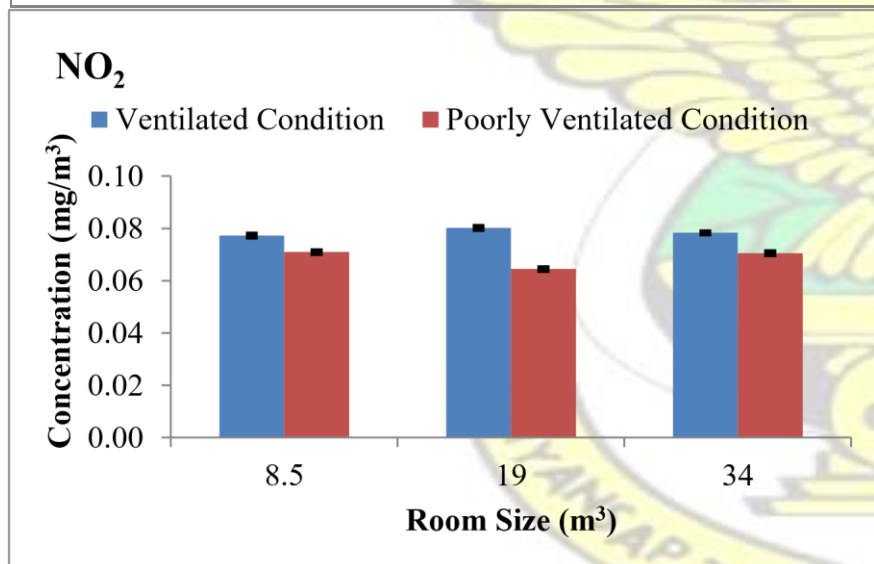
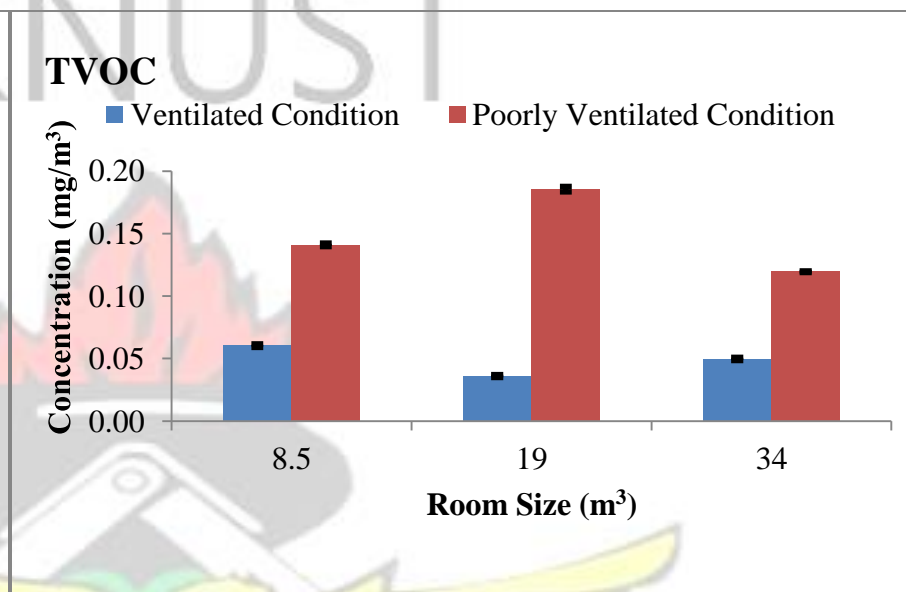
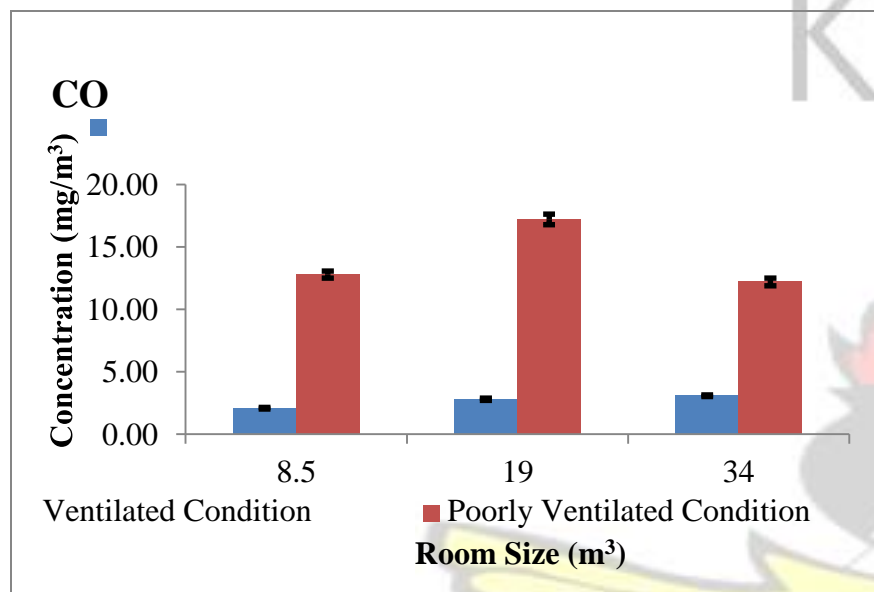


Figure 8: Concentration of pollutants emitted from mosquito coils under experimental room conditions

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CHAPTER FIVE DISCUSSION 5.1 EFFICACY OF MOSQUITO COILS IN TERMS OF MOSQUITO

MORTALITY

5.1.1 Efficacy of Active Ingredients on Mosquito Mortality

The usage of mosquito coil is very predominant in West African countries such as Ghana (Hogarh *et al.*, 2016), Nigeria (Efunshile *et al.*, 2011), Cote D'Ivoire (Doannio *et al.*, 2006; Essé *et al.*, 2008; Koudou *et al.*, 2010) and Burkina Faso (Samuelsen *et al.*, 2004; Yamamoto *et al.*, 2009). In Ghana, nearly 43 % of users of mosquito coils use them on a regular basis (Baume and Koh, 2011). The effectiveness of these mosquito coils is important in mosquito and malaria control. From the results, mosquito coil containing 0.25 % esbiothrin recorded the highest mortality and this can be attributed to the high insecticidal activities of esbiothrin. Esbiothrin is one of the most usually used active ingredients in mosquito coils, and its efficacy has been assessed in other studies (Mosha *et al.*, 1992; Msangi *et al.*, 2010). Mortality of mosquitoes had a trend of MC 1 (0.25 % Esbiothrin) > MC 3 (0.03 % Dimefluthrin) > MC 5 (0.08 % Meperfluthrin) > MC 4 (0.20 % D-allothrin) > MC 2 (0.03 % Dimefluthrin). This trend is contrary to the observation that mortality is strongly influenced by high doses of active ingredients used in their formulation (Ogoma *et al.*, 2012). The type of active ingredients used in the formulation of the coils may play a significant role in their effectiveness. Mosquito coils containing 0.08 % Meperfluthrin (MC 5) showed a strong insecticidal activity than MC 2 (0.03 % Dimefluthrin). This is consistent with the findings of Xue *et al.* (2012) where mosquito coils containing 0.08 % meperfluthrin showed strong insecticidal activity resulting in high mortality (> 90 %) of caged mosquitoes than mosquito coil containing 0.03 % dimefluthrin (< 90 %). However, MC 3 which also contains 0.03 % Dimefluthrin showed a strong insecticidal activity than MC 5 containing 0.08 % Meperfluthrin. The differences in

mortalities of MC 2 and MC 3 with the same percentage of active ingredient could be attributed to the differences in their smoke emissions. It was observed that MC 3 generated more smoke than MC 2. The production of smoke causes humidity to drop by reducing the moisture-carrying capacity of the air. This makes mosquitoes vulnerable to desiccation and lessens sensory input because mosquito chemoreceptors are more alert in the presence of moisture (Davis and Bowen, 1994).

This study recorded higher mortalities than those reported by Ogoma *et al.* (2012). They reported a very low mortality of *Anopheles* mosquitoes in field-assays (16 %). The increased mortalities in this study could be attributed to the type and percentage of active ingredients in mosquito coils used in this study. According to WHO criteria for susceptibility test, 98 – 100 % mortality means susceptibility; 80 – 97 % mortality means resistance suspected with more investigations required; 0 – 79 % mortality indicates resistance is confirmed (Adu-Acheampong *et al.*, 2014). The mortalities ranged from 24.44 to 63.64 % and hence indicated a resistance to pyrethroids in the *Anopheles* species. The study agrees with other studies in Ghana which reported resistance to pyrethroids based vector control methods (Adeniran, 2002; Achonduh *et al.*, 2008; Adu-Acheampong *et al.*, 2014; Boakye *et al.*, 2009). The use of pyrethroids based pesticides in the vegetable farms might be a contributing factor (Achonduh *et al.*, 2008; Adeniran, 2002; Boakye *et al.*, 2009) since mosquito larvae were taken from stagnant waters in vegetable farms.

5.1.2 Effect of Ventilation on Mosquito Coil Efficacy

Studies proved that increasing vaporisation rate of active ingredient may increase efficacy but can also lead to faster loss of active ingredients followed by reduced efficiency over time (Kawada *et al.*, 2006). Similar explanation could be linked to the mortalities under the ventilated conditions where most mosquitoes might have recuperated after the burning

period of the mosquito coils as a result of loss of the active ingredients. The spatial action of aerial insecticide is dependent on air current (i.e. air exchange), temperature, humidity and wind speed within the experimental room (Kawada *et al.*, 2006). A study carried out in Vietnam revealed an increase in the efficacy of emanators when used in rooms without eaves (Kawada *et al.*, 2006) compared to rooms with open eaves in Tanzania (Kawada *et al.*, 2008). This statement is similar to the findings of this study where mosquito exposure under poorly ventilated conditions recorded higher mortality than under ventilated conditions.

5.1.3 Effect of Room Sizes on Mosquito Coil Efficacy

Data from this study showed that room size and efficacy of mosquito coils are inversely proportional (Barnard *et al.*, 1998). The data also concur with a research conducted by Chadwick (1975) where mosquitoes were knocked-down at a faster rate in small rooms than large rooms (25 m³). It was reported by Kawada *et al.* (2008) that confined mosquitoes positioned closely to metofluthrin treated paper strips exhibited 100 % Knockdown within 30 minutes and 100 % mortality in post-exposure period of 24-hours, while mosquitoes positioned 1.5 m farther from the strip had slower knockdown and 70 % mortality and mosquitoes positioned 5 m away were not affected by the paper strips. Likewise, mortality of mosquitoes decreased as the room sizes increased from 8.5 m³ to 19 m³ and then to 34 m³ rooms. The decrease in mortality from 8.5 m³ rooms to 34 m³ rooms may be attributed to low airborne concentration of active ingredients in 34 m³ rooms especially under ventilated conditions. Detaining the test mosquitoes in the 8.5 m³ room might have exposed them to higher amounts of smoke (Ogoma *et al.*, 2012) and more concentrated active ingredients (Jeyalakshmi *et al.*, 2014) which in turn led to greater insecticidal activities of the mosquito coils. The higher mortality in the 8.5 m³ room than

in the 19 m³ and 34 m³ rooms discloses the significance of the room size on mosquito coil efficacy.

5.2 POTENTIAL HEALTH IMPACTS ASSOCIATED WITH EMISSIONS FROM MOSQUITO COILS

5.2.1 Emissions from Mosquito Coils

Epidemiological studies has revealed that prolonged exposure to mosquito coils might promote asthma and persistent wheeze in children (Azizi and Henry, 1991; Fagbule and Ekanem, 1993) and clinical signs (head shaking, scratching of nostrils, sneezing and ruffled fur) were also observed in smoke-exposed rats (Ogbonnia *et al.*, 2015).

The burning of mosquito coils introduced some levels of CO in all the rooms under both ventilated and poorly ventilated conditions. High CO concentration was measured from coils because they are made purposely to have very incomplete burning (smouldering effect) (Zhang *et al.*, 2000). Huge amount of incomplete burning products, consequently, would be released from the burning of mosquito coil. The results of this study agree with the levels of CO recorded from mosquito coils by Lee and Wang (2006). In their study, CO concentration was the highest pollutant recorded among other pollutants (Nonmethane Hydrocarbons (NMHC), Nitrogen Monoxide (NO), Nitrogen Dioxide (NO₂), Methane (CH₄) and Nitrogen Oxides (NO_x)). All the concentrations recorded in the rooms under ventilated conditions were within the WHO air quality standard of 10 mg/m³. However, with the exception of MC 2, MC 3 and MC 4 in the 8.5 m³ and 34 m³ rooms which were below the standard, all the concentrations recorded in rooms under the poorly ventilated conditions were higher than the standard. These high concentrations can result in adverse health effects by reducing oxygen transfer to the body's organs and tissues, as well as serious effects on the cardiovascular and CNS, it can also contribute to smog formation (ground-level ozone) which can cause respiratory problems (Singh *et al.*, 2016).

The burning of mosquito coils did not release significant levels of NO₂ into the experimental rooms. All the concentrations measured were around the levels of NO₂ measured in the ambient air (0.05 – 0.07 mg/m³). Concentrations of NO₂ recorded from this study were very low and this agrees with the findings of Lee and Wang (2006) where there was no noticeable NO₂ emissions from mosquito coils in Hong Kong. All concentrations were below the WHO air quality guideline of 0.2 mg/m³ and 0.04 mg/m³ for 1 hour and annual exposure respectively. Data from this study indicates that none of the monitored average concentrations of NO₂ generated from the burning of mosquito coils are close to the numbers reported to cause adverse effects; however, continuous human exposure to high levels of NO₂ might cause a respiratory health risk in the long run.

Most of the concentrations of SO₂ recorded were in the range (0.1 – 5 ppm; 0.3 – 14.1 mg/m³) that could result in some undesirable health effects among susceptible people, including asthmatics and others with respiratory problems (WHO, 2006b). The concentrations recorded in this study were within the United States Occupational Safety and Health Administration (OSHA) guideline value of 13 mg/m³ over an 8 hour period. However, they were higher than the WHO guideline value 0.02 mg/m³ over 24 hour period. According to the WHO Air Quality Guidelines (2000), the integral of a concentration over an extended period can have more effect on health than the pattern of highest exposure (WHO, 2000).

The burning of mosquito coils emitted some levels of TVOC into the rooms. According to Seifert *et al.* (1999), a long-term level of 1 – 3 mg/m³ TVOC should not be exceeded in rooms for permanent occupancy. Concentrations of TVOC recorded in this study were below 1 mg/m³ in all the experimental rooms hence may not pose any immediate health risks. There are no WHO air quality guideline values for TVOC but guideline values exist

for individual VOCs like benzene, toluene and so forth. This research did not consider the various forms of VOCs. However, Lee and Wang (2006) identified that relatively high concentrations of benzene, toluene and methylene chloride were emitted from mosquito coil smoke. VOCs exposure may cause potential health hazard like irritations in respiratory tract and eyes, headache, damage to liver, kidney and CNS. It is also a known potential carcinogen (Abogrean *et al.*, 2015).

A research conducted by Tharaphy and Chapman (2009) on the effects of household indoor pollution arising from the burning of mosquito coils on respiratory problems in Myanmar refugees in Mae Sot District, Tak Province, Thailand stated that mosquito coil use was positively connected to all the respiratory symptoms except shortness of breath. The use of mosquito coil was significantly associated with cough and phlegm prevalence, and marginally significantly associated with wheeze prevalence in the respondents. The differences in the levels of pollutants among the tested coils could be attributed to differences in the composition of the base materials (biomass) used in the formulation of the coils. Majority of mosquito coils comprises of plant-based materials, such as coconut shell powder, wood and joss powder, binders, oxidants (e.g., nitrates), dyes and other extracts making controlled smouldering possible through the 8 hour burning period (Shu-Chen *et al.*, 2008).

5.2.2 Effect of Ventilation on Mosquito Coil Emissions

The results from this study showed that increasing ventilation rates significantly reduce the concentration of pollutants. In rooms with restricted ventilation (poorly ventilated conditions), the concentration of pollutants were higher than concentrations in ventilated rooms except NO₂ where concentrations were slightly higher in the ventilated rooms than the poorly ventilated rooms. The NO₂ in the ambient air was high hence the higher levels in ventilated rooms. The burning of mosquito coils under poorly ventilated conditions is

likely to produce harmful levels of the indoor air pollutants, hence, in part resulting in adverse health effects. According to Ongwandee and Pipithakul (2010), the increased ventilation rate of a room is expected to minimize health risks of people exposed to the released air pollutants from the mosquito coils. They further used model simulations and it also indicated that burning these household products in an enclosed room is likely to produce harmful levels of the released air pollutants, thus in part resulting in adverse health effects. Increasing room ventilation rate could lessen customer exposure to these high pollutant levels (Ongwandee and Pipithakul, 2010). On the contrary, insufficient ventilation can also escalate indoor pollutant levels by not carrying sufficient outdoor air to dilute the emissions generated inside the rooms and by not transporting indoor air pollutants out of the room (Pillai *et al.*, 2010) as observed from mosquito coils burned under poorly ventilated conditions.

Noorhassim *et al.* (1995) reported that for asthmatic children in two rural communities of Malaysia, the occurrence of asthma was unaffected by environmental factors such as exposure to cigarette smoke, wood stoves and the use of mosquito coils. They attributed this to the fact that most rural houses usually have verandas and large windows, with sufficient ventilation than that of an urban residence. The larger distances between houses in the rural communities also helped general ventilation in the house. In this present study, emissions from mosquito coils burned under ventilated conditions were lower and hence may not pose serious health risks as compared to emissions under poorly ventilated conditions.

The results of this study are also consistent with those results reported by Lin and Lee (1997) where the concentrations of PAHs in the smoke of mosquito coils under different ventilation rates was investigated. In their study, concentrations of PAHs in rooms with higher ventilation rate ($Q = 20$) were lower than rooms with low ventilation rate ($Q = 10$).

5.2.3 Effect of Room Sizes on Mosquito Coil Emissions

The emissions from mosquito coils recorded in the experimental rooms did not fully follow the kinetic theory of James Clerk Maxwell and physicist Ludwig Boltzmann in 1865 which states that if a vessel, with a fixed number of molecules inside, is reduced in volume, more molecules will hit a given area of the sides of the container per unit time, leading to a greater impact (Kauzmann, 2012). Based on this theory, it was expected that the 8.5 m³ room would record the highest levels of pollutants followed by the 19 m³ room and the 34 m³ room recording the least levels of pollutants.

The result of this study did not show any clearly defined trend of decreasing pollutant levels with increasing room sizes. The concentration of NO₂ was similar in all the three rooms under both ventilated and poorly ventilated conditions. This may be attributed to the low levels of the pollutants generated from the coils. The inconsistency in the trend of pollutants may be attributed to the percentage of base materials (biomass) used in the formulation of the coils. It was also observed that, even for the same brand of coil, the burning time and the weight of individual coil differed and in terms of the smoke generated, some were higher than others. These inconsistencies could affect the levels of pollutants generated from the burning of the coils in the rooms. Room sizes alone did not play a key role in the removal of pollutants. However, the quantity of pollutants emitted from the coils may have also contributed to the concentrations in the rooms.

CHAPTER SIX CONCLUSION AND RECOMMENDATION

6.1 CONCLUSION

Five types of mosquito coils on the Ghanaian market were tested in experimental chambers with different room sizes and natural ventilation rates. The mortality studies indicated that mosquito coils containing esbiothrin produced better insecticidal effects on *Anopheles* mosquito than the other active ingredients in the mosquito coils. Mortality resulting from the tested mosquito coils had a decreasing trend of MC 1 (0.25 % Esbiothrin) > MC 3 (0.03 % Dimefluthrin) > MC 5 (0.08 % Meperfluthrin) > MC 4 (0.20 % D-allothrin) > MC 2 (0.03 % Dimefluthrin). The difference in mortalities of mosquito coils containing the same percentage of active ingredients also indicate that other factors such as smoke level of coils played a role in mosquito mortality. The highest mortality recorded fell within the WHO criteria for susceptibility test range of 0 – 79 %, which suggested resistance of mosquitoes to the pyrethroids.

Mortality of mosquitoes increased with decreasing room sizes and also insufficient ventilation. Generally, there was no significant difference in the mortalities of mosquitoes in the experimental rooms. Pollutant concentrations in the experimental rooms did not increase from 8.5m³ to 34m³ in all the experiment. This implies that room sizes alone do not play a key role in the removal of pollutants. The significant difference in mortalities and pollutant concentrations in ventilated and poorly ventilated rooms depicted the effect of ventilation on mosquito mortality and indoor air quality.

Among the pollutants that were monitored, carbon monoxide was the main gas pollutant and stemmed from the smouldering effect of the coils. Burning of mosquito coils does not generate significant levels of NO₂ into the indoor environment. The tested mosquito coils emitted considerable levels of TVOC into the rooms. There are no WHO air quality

guideline values for TVOC but guideline values exist for individual VOCs like benzene, toluene and so forth. This research did not consider the various forms of VOCs. The levels of TVOC recorded under ventilated conditions may not cause immediate health risks. The concentrations of CO and SO₂ especially under poorly ventilated conditions were above the WHO air quality guideline and could pose adverse health effects to users.

Under ventilated conditions, there was reduced mortality as well as reduced pollutant concentration unlike under the poor ventilation where there was high mortality and also high pollutant concentrations. It is therefore advisable that users of mosquito coils should not sleep in rooms with lit mosquito coils. Individuals sleeping in rooms with lit mosquito coils may be exposed to some undesirable levels of pollutants emitted from the coils. If it becomes necessary to use mosquito coil, the coil should first be burned in a closed indoor environment to achieve maximum insecticidal effect, following which the rooms should be well aerated prior to sleeping in them.

6.2 RECOMMENDATION

Users of mosquito coils should burn the products in rooms with good ventilation. Coils may also be burned outdoors since burning of coils in rooms with good ventilation reduces the concentrations of pollutants emitted.

Further studies are recommended to provide information on the form of resistance in the *Anopheles* mosquitoes in the study area and Kumasi as a whole.

Further studies are recommended in this area to provide information about the quantity of active ingredients in the solid mosquito coils and the vaporized mosquito coils, the levels of particulate matter, PAHs and individual VOCs such as formaldehydes and benzene generated from the burning of mosquito coils.

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APPENDICES

APPENDIX I: MORTALITY OF MOSQUITOES

Table 9: Mortality of mosquitoes resulting from mosquito moils in the three experimental rooms

Replicate	Total Mosquitoes		Under Ventilated Conditions		Under Poorly Ventilated Conditions		
	Exposed per Room		19m ³ Room	8.5m ³ Room	Exposed per Room		
	34m ³ Room				34m ³ Room	19m ³ Room	8.5m ³ Room
MC 1							
First	50	24	25	29	33	36	37
Second	50	21	24	27	26	29	31
Third	50	23	25	30	28	31	34
MC 2							
First	50	14	21	22	18	24	28
Second	50	16	19	23	22	24	25
Third	50	18	20	22	20	22	26
MC 3							
First	50	23	24	26	29	31	35
Second	50	19	22	25	25	27	30
Third	50	16	18	20	30	31	33
MC 4							
First	50	17	21	25	20	29	30
Second	50	18	20	22	22	29	28
Third	50	18	20	21	19	26	29
MC 5							
First	50	23	22	25	27	28	31
Second	50	18	21	23	26	29	30
Third	50	16	20	21	26	28	20
Control	50	5	4	5	5	5	6

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Calculations of Corrected Mortality under Ventilated Conditions

$$\frac{\text{Corrected Mortality in treatment (\%)} - \text{Mortality in control (\%)}}{100 - \text{Mortality in control (\%)}} \times 100 =$$

$$\% \text{ Mortality} = \frac{\text{number of mortality}}{\text{total number of mosquitoes exposed}} \times 100$$

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8.5 m³ Experimental Room

$$\text{Control: \% Mortality} = \frac{5}{50} \times 100 = 10\%$$

	MC 3	MC 1
	$\frac{29}{50} \times 100$	1st
1st Replicate:	$\% \text{ Mortality} = 58\%$	%
	$\frac{58(\%) - 10(\%)}{100 - 10(\%)} \times 100 = 53.33$	C.M
C.M (%) =	%	%

	2nd
2nd Replicate:	$\% \text{ Mortality} = \frac{27}{50} \times 100 = 54\%$
	C.M
C.M (%) =	$\frac{54(\%) - 10(\%)}{100 - 10(\%)} \times 100 = 48.89$
	%

	3rd
3rd Replicate:	$\% \text{ Mortality} = \frac{30}{50} \times 100 = 60\%$
	C.M
C.M (%) =	$\frac{60(\%) - 10(\%)}{100 - 10(\%)} \times 100 = 55.56$
	%

MC 2	MC 4
1st Replicate:	$\% \text{ Mortality} = \frac{22}{50} \times 100 = 44\%$
	C.M (%) =
	$\frac{44(\%) - 10(\%)}{100 - 10(\%)} \times 100 = 37.78$
	%

2nd Replicate:	$\% \text{ Mortality} = \frac{23}{50} \times 100 = 46\%$
	C.M (%) =
	$\frac{46(\%) - 10(\%)}{100 - 10(\%)} \times 100 = 40.00$
	%

3rd Replicate:	$\% \text{ Mortality} = \frac{22}{50} \times 100 = 44\%$
	C.M (%) =
	$\frac{44(\%) - 10(\%)}{100 - 10(\%)} \times 100 = 37.78$
	%

$$\text{Replicate: \% Mortality} = \frac{26}{50} \times 100 = 52$$

$$(\%) = \frac{52(\%) - 10(\%)}{100 - 10(\%)} \times 100 = 46.67$$

$$\frac{25}{50} \times 100 = 50$$

$$\text{Replicate: \% Mortality} = \%$$

$$\% (\%) = \frac{50(\%) - 10(\%)}{100 - 10(\%)} \times 100 = 44.44$$

$$\text{Replicate: \% Mortality} = \frac{20}{50} \times 100 = 40$$

$$(\%) = \frac{40(\%) - 10(\%)}{100 - 10(\%)} \times 100 = 33.33$$

$$44\%$$

$$\text{C.M (\%)} = \frac{44(\%) - 10(\%)}{100 - 10(\%)} \times 100 = 37.38$$

$$\%$$

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$$\text{1st Replicate: \% Mortality} = \frac{25}{50} \times 100 = 50\%$$

$$\text{C.M (\%)} = \frac{50(\%) - 10(\%)}{100 - 10(\%)} \times 100 = 44.44\%$$

$$\text{2nd Replicate: \% Mortality} = \frac{22}{50} \times 100 =$$

$$\text{C.M (\%)} = \frac{42(\%) - 10(\%)}{100 - 10(\%)} \times 100 = 35.56\%$$

MC 5 C.M

$$\text{1st Replicate: \% Mortality} = \frac{25}{50} \times 100 = 50\%$$

$$\text{C.M (\%)} = \frac{50(\%) - 10(\%)}{100 - 10(\%)} \times 100 = 44.44\%$$

$$\text{3rd Replicate: \% Mortality} = \frac{21}{50} \times 100 = 42\%$$

$$\text{C.M (\%)} = \frac{42(\%) - 10(\%)}{100 - 10(\%)} \times 100 = 35.56\%$$

$$\text{Mortality} = \frac{21}{50} \times 100 = 42\%$$

$$\text{C.M (\%)} = \frac{42(\%) - 10(\%)}{100 - 10(\%)} \times 100 = 35.56\%$$

19 m³ Experimental Room

$$\text{Control: \% Mortality} = \frac{8}{50} \times 100 = 8\%$$

$$\text{3rd Replicate: \% Mortality} = \frac{21}{50} \times 100 = 42\%$$

MC 1

$$\text{1st Replicate: \% Mortality} = \frac{25}{50} \times 100 = 50\%$$

$$\text{C.M (\%)} = \frac{50(\%) - 8(\%)}{100 - 8(\%)} \times 100 = 45.65\%$$

$$\text{2nd Replicate: \% Mortality} = \frac{24}{50} \times 100 = 48\%$$

$$\text{C.M (\%)} = \frac{48(\%) - 8(\%)}{100 - 8(\%)} \times 100 = 43.48\%$$

$$\text{3rd Replicate: \% Mortality} = \frac{25}{50} \times 100 = 50\%$$

$$\text{C.M (\%)} = \frac{50(\%) - 8(\%)}{100 - 8(\%)} \times 100 = 45.65\%$$

MC 2

$$\text{1st Replicate: \% Mortality} = \frac{21}{50} \times 100 = 42\%$$

$$\text{C.M (\%)} = \frac{42(\%) - 8(\%)}{100 - 8(\%)} \times 100 = 36.96\%$$

$$\text{2nd Replicate: \% Mortality} = \frac{19}{50} \times 100 = 38\%$$

$$\text{C.M (\%)} = \frac{38(\%) - 8(\%)}{100 - 8(\%)} \times 100 = 32.61\%$$

$$\text{3rd Replicate: \% Mortality} = \frac{20}{50} \times 100 = 40\%$$

$$\text{C.M (\%)} = \frac{40(\%) - 8(\%)}{100 - 8(\%)} \times 100 = 34.78\%$$

$$\text{1st Replicate: \% Mortality} = \frac{24}{50} \times 100 = 48\%$$

$$\text{C.M (\%)} = \frac{48(\%) - 8(\%)}{100 - 8(\%)} \times 100 = 43.48\%$$

$$\text{2nd Replicate: \% Mortality} = \frac{22}{50} \times 100 = 44\%$$

$$\text{C.M (\%)} = \frac{44(\%) - 8(\%)}{100 - 8(\%)} \times 100 = 39.13\%$$

$$\text{3rd Replicate: \% Mortality} = \frac{18}{50} \times 100 = 36\%$$

$$\text{C.M (\%)} = \frac{36(\%) - 8(\%)}{100 - 8(\%)} \times 100 = 30.43\%$$

MC 4

$$\text{1st Replicate: \% Mortality} = \frac{21}{50} \times 100 = 42\%$$

$$\text{C.M (\%)} = \frac{42(\%) - 8(\%)}{100 - 8(\%)} \times 100 = 36.96\%$$

$$\text{2nd Replicate: \% Mortality} = \frac{20}{50} \times 100 = 40\%$$

$$\text{C.M } (\%) = \frac{40(\%) - 8(\%)}{100 - 8(\%)} \times 100 = 34.78\%$$

$$\text{3rd Replicate: \% Mortality} = \frac{20}{50} \times 100 = 40\%$$

$$\text{MC 5} \quad \frac{42(\%) - 10(\%)}{\frac{22}{50} \times 100 = 44}$$

$$\text{1st} \quad \frac{44(\%) - 8(\%)}{100 - 8(\%)} \times 100 = 39.13 \quad \text{3rd}$$

$$\text{Replicate: \% Mortality} = \frac{21}{50} \times 100 = 42$$

$$\text{C.M } (\%) = \%$$

$$\text{C.M } (\%) = \frac{32(\%) - 10(\%)}{100 - 10(\%)} \times 100 = 24.44\%$$

$$\text{C.M } (\%) = \frac{\quad}{100 - 10(\%)} \times 100 = 35.56\%$$

$$\text{Replicate: \% Mortality} = \frac{20}{50} \times 100 = 40$$

$$(\%) = \frac{40(\%) - 8(\%)}{100 - 8(\%)} \times 100 = 34.78$$

$$\text{C.M } \%$$

$$\text{2nd Replicate: \% Mortality} = \%$$

34 m³ Experimental Room

$$\text{C.M } (\%) = \frac{40(\%) - 8(\%)}{100 - 8(\%)} \times 100 = 34.78\%$$

$$\text{3rd Replicate: \% Mortality} = \frac{18}{50} \times 100 = 36\%$$

$$\text{Control: \% Mortality} = \frac{5}{50} \times 100 = 10\%$$

MC 1

$$\text{1st Replicate: \% Mortality} = \frac{24}{50} \times 100 = 48\%$$

$$\text{C.M } (\%) = \frac{48(\%) - 10(\%)}{100 - 10(\%)} \times 100 = 42.22\%$$

$$\text{2nd Replicate: \% Mortality} = \frac{21}{50} \times 100 = 42\%$$

$$\text{C.M } (\%) = \frac{42(\%) - 10(\%)}{100 - 10(\%)} \times 100 = 35.56\%$$

$$\text{3rd Replicate: \% Mortality} = \frac{23}{50} \times 100 = 46\%$$

$$\text{C.M } (\%) = \frac{46(\%) - 10(\%)}{100 - 10(\%)} \times 100 = 40.00\%$$

MC 2

$$\text{1st Replicate: \% Mortality} = \frac{14}{50} \times 100 = 28\%$$

$$\text{C.M } (\%) = \frac{28(\%) - 10(\%)}{100 - 10(\%)} \times 100 = 20.00\%$$

$$\text{2nd Replicate: \% Mortality} = \frac{16}{50} \times 100 = 32\%$$

$$\text{C.M } (\%) = \frac{36(\%) - 10(\%)}{100 - 10(\%)} \times 100 = 28.89\%$$

$$\text{\% MC 3}$$

$$\text{1st Replicate: \% Mortality} = \frac{23}{50} \times 100 = 46\%$$

$$\text{C.M } (\%) = \frac{46(\%) - 10(\%)}{100 - 10(\%)} \times 100 = 40.00\%$$

$$\text{2nd Replicate: \% Mortality} = \frac{19}{50} \times 100 = 38\%$$

$$\text{C.M } (\%) = \frac{38(\%) - 10(\%)}{100 - 10(\%)} \times 100 = 31.11\%$$

$$\text{3rd Replicate: \% Mortality} = \frac{16}{50} \times 100 = 32\%$$

$$\text{C.M } (\%) = \frac{32(\%) - 10(\%)}{100 - 10(\%)} \times 100 = 24.44\%$$

MC 4

$$\text{1st Replicate: \% Mortality} = \frac{17}{50} \times 100 = 34\%$$

$$\text{C.M } (\%) = \frac{34(\%) - 10(\%)}{100 - 10(\%)} \times 100 = 26.67\%$$

$$\text{2nd Replicate: \% Mortality} = \frac{18}{50} \times 100 = 42\%$$

$$\text{C.M (\%)} = \frac{36(\%) - 10(\%)}{100 - 10(\%)} \times 100 = 28.89\%$$

$$\text{3rd Replicate: \% Mortality} = \frac{18}{50} \times 100 = 36\%$$

$$\text{C.M (\%)} = \frac{36(\%) - 10(\%)}{100 - 10(\%)} \times 100 = 28.89\%$$

MC 5

$$\text{C.M (\%)} = \frac{46(\%) - 10(\%)}{100 - 10(\%)} \times 100 = 40.00\%$$

$$\text{2nd Replicate: \% Mortality} = \frac{18}{50} \times 100 = 42\%$$

$$\text{C.M (\%)} = \frac{36(\%) - 10(\%)}{100 - 10(\%)} \times 100 = 28.89\%$$

$$\text{C.M (\%)} = \frac{68(\%) - 12(\%)}{100 - 12(\%)} \times 100 = 63.64\%$$

MC 2

$$\text{1st Replicate: \% Mortality} = \frac{28}{50} \times 100 = 56\%$$

$$\text{C.M (\%)} = \frac{56(\%) - 12(\%)}{100 - 12(\%)} \times 100 = 50.00\%$$

Calculations of Corrected Mortality under Poorly Ventilated Conditions

$$\text{Corrected mortality (\%)} = \frac{\text{Mortality in treatment (\%)} - \text{Mortality in control (\%)}}{100 - \text{Mortality in control (\%)}} \times 100$$

$$\% \text{ Mortality} = \frac{\text{number of mortality}}{\text{total number of mosquitoes exposed}} \times 100$$

8.5 m³ Experimental Room

$$\text{1st Replicate: \% Mortality} = \frac{23}{50} \times 100 = 46\%$$

$$\text{Control: \% Mortality} = \frac{6}{50} \times 100 = 12\%$$

MC 1

$$\text{1st Replicate: \% Mortality} = \frac{37}{50} \times 100 = 74\%$$

$$\text{C.M (\%)} = \frac{74(\%) - 12(\%)}{100 - 12(\%)} \times 100 = 70.45\%$$

$$\text{2nd Replicate: \% Mortality} = \frac{31}{50} \times 100 = 62\%$$

$$\text{C.M (\%)} = \frac{62(\%) - 12(\%)}{100 - 12(\%)} \times 100 = 56.82\%$$

$$\text{3rd Replicate: \% Mortality} = \frac{34}{50} \times 100 = 68\%$$

$$\text{2nd Replicate: \% Mortality} = \frac{25}{50} \times 100 = 50\%$$

$$\text{C.M (\%)} = \frac{50(\%) - 12(\%)}{100 - 12(\%)} \times 100 = 43.18\%$$

$$\text{\% 3rd Replicate: \% Mortality} = \frac{26}{50} \times 100 = 52\%$$

$$\text{C.M (\%)} = \frac{52(\%) - 12(\%)}{100 - 12(\%)} \times 100 = 45.45\%$$

MC 3

$$\text{1st Replicate: \% Mortality} = \frac{35}{50} \times 100 = 70\%$$

$$\text{C.M (\%)} = \frac{70(\%) - 12(\%)}{100 - 12(\%)} \times 100 = 65.91\%$$

$$\text{2nd Replicate: \% Mortality} = \frac{30}{50} \times 100 = 60\%$$

$$\text{C.M (\%)} = \frac{56(\%) - 12(\%)}{100 - 12(\%)} \times 100 = 50.00\%$$

$$\text{C.M (\%)} = 52.27\%$$

19 m³ Experimental Room

$$\text{Control: \% Mortality} = \frac{5}{50} \times 100 = 10\%$$

MC 3

$$\text{1st Replicate: \% Mortality} = \frac{31}{50} \times 100 = 62\%$$

MC 1

$$\text{1st Replicate: \% Mortality} = \frac{36}{50} \times 100 = 72\%$$

$$\text{C.M (\%)} = \frac{62(\%) - 10(\%)}{100 - 10(\%)} \times 100 = 57.78\%$$

$$\text{C.M (\%)} = \frac{72(\%) - 10(\%)}{100 - 10(\%)} \times 100 = 68.89\%$$

$$\text{2nd Replicate: \% Mortality} = \frac{27}{50} \times 100 = 54\%$$

$$\text{2nd Replicate: \% Mortality} = \frac{29}{50} \times 100 = 58\%$$

$$\text{C.M (\%)} = \frac{54(\%) - 10(\%)}{100 - 10(\%)} \times 100 = 48.89\%$$

$$\text{C.M (\%)} = \frac{58(\%) - 10(\%)}{100 - 10(\%)} \times 100 = 53.33\%$$

$$\text{3rd Replicate: \% Mortality} = \frac{31}{50} \times 100 = 62\%$$

$$\text{3rd Replicate: \% Mortality} = \frac{31}{50} \times 100 = 62\%$$

$$\text{C.M (\%)} = \frac{62(\%) - 10(\%)}{100 - 10(\%)} \times 100 = 57.78\%$$

$$\text{C.M (\%)} = \frac{60(\%) - 12(\%)}{100 - 12(\%)} \times 100 = 54.55\%$$

$$\text{C.M (\%)} = 58\%$$

$$\text{3rd Replicate: \% Mortality} = \frac{29}{50} \times 100 = 58\%$$

$$\text{3rd Replicate: \% Mortality} = \frac{33}{50} \times 100 = 66\%$$

$$\text{C.M (\%)} = \frac{58(\%) - 12(\%)}{100 - 12(\%)} \times 100 = 52.27\%$$

$$\text{C.M (\%)} = \frac{66(\%) - 12(\%)}{100 - 12(\%)} \times 100 = 61.36\%$$

MC 5

MC 4

$$\text{1st Replicate: \% Mortality} = \frac{30}{50} \times 100 = 60\%$$

$$\text{1st Replicate: \% Mortality} = \frac{31}{50} \times 100 = 62\%$$

$$\text{C.M (\%)} = \frac{60(\%) - 12(\%)}{100 - 12(\%)} \times 100 = 54.55\%$$

$$\text{C.M (\%)} = \frac{62(\%) - 12(\%)}{100 - 12(\%)} \times 100 = 56.82\%$$

$$\text{2nd Replicate: \% Mortality} = \frac{28}{50} \times 100 = 56\%$$

$$\text{2nd Replicate: \% Mortality} = \frac{30}{50} \times 100 = 60\%$$

$$\text{C.M (\%)} = \frac{60(\%) - 12(\%)}{100 - 12(\%)} \times 100 = 54.55\%$$

$$\text{3rd Replicate: \% Mortality} = \frac{29}{50} \times 100 = 58\%$$

$$\text{C.M (\%)} = \frac{58(\%) - 12(\%)}{100 - 12(\%)} \times 100 =$$

$$C.M (\%) = \frac{62(\%) - 10(\%)}{100 - 10(\%)} \times 100 = 57.78\%$$

MC 2

$$1^{st} \text{ Replicate: } \% \text{ Mortality} = \frac{24}{50} \times 100 = 48\%$$

$$C.M (\%) = \frac{48(\%) - 10(\%)}{100 - 10(\%)} \times 100 = 42.22\%$$

$$2^{nd} \text{ Replicate: } \% \text{ Mortality} = \frac{24}{50} \times 100 = 48\%$$

$$C.M (\%) = \frac{48(\%) - 10(\%)}{100 - 10(\%)} \times 100 = 42.22\%$$

$$3^{rd} \text{ Replicate: } \% \text{ Mortality} = \frac{22}{50} \times 100 = 44\%$$

$$C.M (\%) = \frac{44(\%) - 10(\%)}{100 - 10(\%)} \times 100 =$$

37.78% MC 4

$$1^{st} \text{ Replicate: } \% \text{ Mortality} = \frac{29}{50} \times 100 = 58\%$$

$$C.M (\%) = \frac{58(\%) - 10(\%)}{100 - 10(\%)} \times 100 = 53.33\%$$

$$2^{nd} \text{ Replicate: } \% \text{ Mortality} = \frac{29}{50} \times 100 = 58\%$$

$$C.M (\%) = \frac{58(\%) - 10(\%)}{100 - 10(\%)} \times 100 = 53.33\%$$

$$3^{rd} \text{ Replicate: } \% \text{ Mortality} = \frac{26}{50} \times 100 = 52\%$$

$$C.M (\%) = \frac{52(\%) - 10(\%)}{100 - 10(\%)} \times 100 = 46.67\%$$

MC 5

$$1^{st} \text{ Replicate: } \% \text{ Mortality} = \frac{28}{50} \times 100 = 56\%$$

$$C.M (\%) = \frac{56(\%) - 10(\%)}{100 - 10(\%)} \times 100 = 51.11\%$$

$$2^{nd} \text{ Replicate: } \% \text{ Mortality} = \frac{29}{50} \times 100 = 58\%$$

$$\text{C.M (\%)} = \frac{58(\%) - 10(\%)}{100 - 10(\%)} \times 100 =$$

53.33% 3rd Replicate: % Mortality =

$$\frac{28}{50} \times 100 = 56\% \text{ C.M}$$

m³ Experimental Room

Control: % Mortality = $\frac{5}{50} \times 100 = 10\%$

44.44%

C.M (%) = $\frac{50(\%) - 10(\%)}{100 - 10(\%)} \times 100 =$

1

1st Replicate: % Mortality $\frac{33}{50} \times 100 = 66\%$

(%) = $\frac{66(\%) - 10(\%)}{100 - 10(\%)} \times 100 = \mathbf{62.22}$

(%) = %

$\frac{30}{50} \times 100 =$

$\frac{60(\%) - 10(\%)}{100 - 10(\%)} \times 100 =$

4

2nd Replicate: % Mortality $\frac{26}{50} \times 100 = 52\%$

$\frac{20}{50} \times 100 = 40\%$

1st Replicate: % Mortality =

(%) = $\frac{52(\%) - 10(\%)}{100 - 10(\%)} \times 100 = \mathbf{46.67\%}$

C.M (%) = $\frac{40(\%) - 10(\%)}{100 - 10(\%)} \times 100 = \mathbf{33.33\%}$

3rd Replicate: % Mortality = $\frac{28}{50} \times 100 = 56\%$

(%) = $\frac{56(\%) - 10(\%)}{100 - 10(\%)} \times 100 = \mathbf{51.11\%}$

2nd Replicate: % Mortality = $\frac{22}{50} \times 100 = 44\%$

C.M (%) = $\frac{44(\%) - 10(\%)}{100 - 10(\%)} \times 100 = \mathbf{37.78\%}$

2

$\frac{18}{50} \times 100 = 36\%$

1st $\frac{36(\%) - 10(\%)}{100 - 10(\%)} \times 100 = \mathbf{28.89\%}$ **Replicate: % Mortality =**

(%) = $\mathbf{31.11\%}$ $\frac{22}{50} \times 100 =$

(%) = % $\frac{27}{50} \times 100 = 54\%$

5

2nd Replicate: % Mortality =

= 1st Replicate: % Mortality =

44%

(%) = $\frac{54(\%) - 10(\%)}{100 - 10(\%)} \times 100 = \mathbf{48.89\%}$

(%) = %

(%) = $\mathbf{37.78\%}$

(%) = $\frac{56(\%) - 10(\%)}{100 - 10(\%)} \times 100 = \mathbf{51.11\%}$

$$3^{\text{rd}} \quad \frac{44(\%) - 10(\%)}{100 - 10(\%)} \times 100 =$$

$$\frac{20}{50} \times 100 = 40\%$$

Replicate: % Mortality = 50

$$\frac{40(\%) - 10(\%)}{100 - 10(\%)} \times 100 = 33.33\%$$

C.M (%) = 33.33%

MC 3

$$1^{\text{st}} \text{ Replicate: } \% \text{ Mortality} = \frac{29}{50} \times 100 = 58\%$$

$$\frac{58(\%) - 10(\%)}{100 - 10(\%)} \times 100 = 53.33\%$$

C.M (%) = 53.33%

$$2^{\text{nd}} \text{ Replicate: } \% \text{ Mortality} = \frac{25}{50} \times 100 = 50\%$$

$$2^{\text{nd}} \text{ Replicate: } \% \text{ Mortality} = \frac{26}{50} \times 100 = 52\%$$

$$\frac{52(\%) - 10(\%)}{100 - 10(\%)} \times 100 = 46.67\%$$

C.M (%) = 46.67%

$$3^{\text{rd}} \text{ Replicate: } \% \text{ Mortality} = \frac{26}{50} \times 100 = 52\%$$

$$\frac{52(\%) - 10(\%)}{100 - 10(\%)} \times 100 = 46.67\%$$

C.M (%) = 46.67%

Note: C.M means Corrected Mortality

Statistical Analysis of mortality in 34 m³

Room under Ventilated and Poorly Ventilated Conditions Group Statistics

Conditions	N	Mean	Std. Deviation	Std. Error Mean
Mortality Ventilated Condition	5	30.9620	5.47976	2.45062
Poorly Ventilated Condition	5	43.8500	9.50750	4.25188

Independent Samples Test

		Levene's Test for Equality of Variances		t-test for Equality of Means						
		F	Sig.	t	df	Sig. (2tailed)	Mean Difference	Std. Error Difference	95% Confidence Interval of the Difference	
									Lower	Upper
Mortality	Equal variances assumed	4.675	0.063	-2.626	8	0.030	-12.88800	4.90755	-24.20484	-1.57116
	Equal variances not assumed			-2.626	6.393	0.037	-12.88800	4.90755	-24.71948	-1.05652

Statistical Analysis of mortality in 19 Room under Ventilated and Poorly Ventilated Conditions

Group Statistics

Conditions	N	Mean	Std. Deviation	Std. Error Mean
Mortality Ventilated Condition	5	37.9720	4.05504	1.81347
Poorly Ventilated Condition	5	51.7040	7.05543	3.15529

Independent Samples Test

		Levene's Test for Equality of Variances		t-test for Equality of Means						
		F	Sig.	t	df	Sig. (2tailed)	Mean Difference	Std. Error Difference	95% Confidence Interval of the Difference	
									Lower	Upper
Mortality	Equal variances assumed	0.568	0.473	-3.773	8	0.005	-13.73200	3.63930	-22.12424	-5.33976

Equal variances not assumed	m ³	-3.773	6.383	0.008	-13.73200	3.63930	-22.50923	-4.95477
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Statistical Analysis of mortality in 8.5

Room under Ventilated and Poorly Ventilated Conditions Group Statistics

Conditions	N	Mean	Std. Deviation	Std. Error Mean
Mortality Ventilated Condition	5	42.3440	5.83468	2.60935
Poorly Ventilated Condition	5	55.4560	6.89205	3.08222

Independent Samples Test

		Levene's Test for Equality of Variances		t-test for Equality of Means						
		F	Sig.	t	df	Sig. (2tailed)	Mean Difference	Std. Error Difference	95% Confidence Interval of the Difference	
									Lower	Upper
Mortality	Equal variances assumed	0.306	0.595	-3.247	8	0.012	-13.11200	4.03841	-22.42460	-3.79940

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Equal variances not assumed	m ³		-3.247	7.788	0.012	-13.11200	4.03841	-22.46893	-3.75507
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**Statistical Analysis of mortality from Tested Mosquito Coils under
Ventilated Conditions**

ANOVA

Mortalities

	Sum of Squares	df	Mean Square	F	Sig.
Between Groups	305.346	4	76.337	2.204	0.142
Within Groups	346.386	10	34.639		
Total	651.733	14			

Post Hoc Tests Multiple Comparisons

Dependent Variable: Mortalities

Tukey HSD

(I) Coils	(J) Coils	Mean Difference (I-J)	Std. Error	Sig.	95% Confidence Interval	
					Lower Bound	Upper Bound
MC 1	MC 2	13.01333	4.80546	0.122	-2.8018	28.8285
	MC 3	8.59000	4.80546	0.430	-7.2252	24.4052
	MC 4	11.33000	4.80546	0.204	-4.4852	27.1452
	MC 5	9.57000	4.80546	0.285	-6.2452	25.3852
MC 2	MC 1	-13.01333	4.80546	0.122	-28.8285	2.8018
	MC 3	-4.42333	4.80546	0.883	-20.2385	11.3918
	MC 4	-1.68333	4.80546	0.996	-17.4985	14.1318
	MC 5	-3.44333	4.80546	0.958	-19.2585	12.3718
MC 3	MC 1	-8.59000	4.80546	0.430	-24.4052	7.2252
	MC 2	4.42333	4.80546	0.883	-11.3918	20.2385
	MC 4	2.74000	4.80546	0.977	-13.0752	18.5552
	MC 5	.98000	4.80546	1.000	-14.8352	16.7952
MC 4	MC 1	-11.33000	4.80546	0.204	-27.1452	4.4852
	MC 2	1.68333	4.80546	0.996	-14.1318	17.4985
	MC 3	-2.74000	4.80546	0.977	-18.5552	13.0752
	MC 5	-1.76000	4.80546	0.996	-17.5752	14.0552
MC 5	MC 1	-9.57000	4.80546	0.285	-25.3852	6.2452
	MC 2	3.44333	4.80546	0.958	-12.3718	19.2585
	MC 3	-.98000	4.80546	1.000	-16.7952	14.8352
	MC 4	1.76000	4.80546	0.996	-14.0552	17.5752

Statistical Analysis of mortality from Tested Mosquito Coils under Poorly Ventilated Conditions

ANOVA

Mortalities

	Sum of Squares	df	Mean Square	F	Sig.
Between Groups	683.924	4	170.981	4.093	0.032
Within Groups	417.715	10	41.771		
Total	1101.638	14			

Post Hoc Tests Multiple Comparisons

Dependent Variable: Mortalities
Tukey
HSD

(I) Coils	(J) Coils	Mean Difference (I-J)	Std. Error	Sig.	95% Confidence Interval	
					Lower Bound	Upper Bound
MC 1	MC 2	18.89667*	5.27709	0.032	1.5293	36.2640
	MC 3	3.47000	5.27709	0.961	-13.8973	20.7473
	MC 4	13.17333	5.27709	0.167	-4.1940	30.4907
	MC 5	7.72000	5.27709	0.606	-9.6473	25.0873
MC 2	MC 1	-18.89667*	5.27709	0.032	-36.2640	-1.5293
	MC 3	-15.42667	5.27709	0.088	-32.7940	1.9407
	MC 4	-5.72333	5.27709	0.811	-23.0907	11.6440
	MC 5	-11.17667	5.27709	0.284	-28.5440	6.1907
MC 3	MC 1	-3.47000	5.27709	0.961	-20.7473	13.8973
	MC 2	15.42667	5.27709	0.088	-1.9407	32.7940
	MC 4	9.70333	5.27709	0.405	-7.6640	27.0707
	MC 5	4.25000	5.27709	0.883	-13.1173	21.6173
MC 4	MC 1	-13.17333	5.27709	0.167	-30.4907	4.1940
	MC 2	5.72333	5.27709	0.811	-11.6440	23.0907
	MC 3	-9.70333	5.27709	0.405	-27.0707	7.6640
	MC 5	-5.45333	5.27709	0.745	-22.8207	11.9140

MC 5	MC 1	-7.72000	5.27709	0.606	-25.0873	9.6473
	MC 2	11.17667	5.27709	0.284	-6.1907	28.5440
	MC 3	-4.25000	5.27709	0.883	-21.6173	13.1173
	MC 4	5.45333	5.27709	0.745	-11.9140	22.8207

*. The mean difference is significant at the 0.05 level.

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Statistical Analysis of mortality in the three Experimental Rooms

ANOVA

Mortality

	Sum of Squares	df	Mean Square	F	Sig.
Between Groups	679.533	2	339.767	3.839	0.034
Within Groups	2389.629	27	88.505		
Total	3069.162	29			

Post Hoc Tests Multiple Comparisons

Dependent Variable: Mortality Tukey
HSD

(I) Room Sizes	(J) Room Sizes	Mean Difference (I-J)	Std. Error	Sig.	95% Confidence Interval	
					Lower Bound	Upper Bound
8.5m ³	19m ³	4.06000	4.20725	0.605	-6.3715	14.4915
	34m ³	11.49400*	4.20725	0.029	1.0625	21.9255
19m ³	8.5m ³	-4.06000	4.20725	0.605	-14.4915	6.3715
	34m ³	7.43400	4.20725	0.200	-2.9975	17.8655
34m ³	8.5m ³	-11.49400*	4.20725	0.029	-21.9255	-1.0625
	19m ³	-7.43400	4.20725	0.200	-17.8655	2.9975

*. The mean difference is significant at the 0.05 level.

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APPENDIX II: EMISSIONS FROM TESTED MOSQUITO COILS

Statistical Analysis of pollutants emitted from mosquito coils under ventilated and poorly ventilated conditions Group Statistics

Condition	N	Mean	Std. Deviation	Std. Error Mean
CO Ventilated Condition	12	2.8508	1.07620	0.31067
CO Poorly Ventilated Condition	11	15.9136	7.61750	2.29676
TVOC Ventilated Condition	12	0.0488	0.02105	0.00608
TVOC Poorly Ventilated Condition	11	0.1505	0.04143	0.01249
SO ₂ Ventilated Condition	12	0.2175	0.06811	0.01966
SO ₂ Poorly Ventilated Condition	11	0.7718	0.19768	0.05960
NO ₂ Ventilated Condition	12	0.07767	0.003284	0.000948
NO ₂ Poorly Ventilated Condition	11	0.06918	0.005671	0.001710

Independent Samples Test

		Levene's Test for Equality of Variances		t-test for Equality of Means						
		F	Sig.	t	Df	Sig. (2tailed)	Mean Difference	Std. Error Difference	95% Confidence Interval of the Difference	
									Lower	Upper
CO	Equal variances assumed	16.970	0.000	-5.889	21	0.000	-13.06280	2.21818	-17.67575	-8.44985
	Equal variances not assumed			-5.636	10.366	0.000	-13.06280	2.31768	-18.20230	-7.92331
TVOC	Equal variances assumed	3.934	0.061	-7.521	21	0.000	-0.10170	0.01352	-0.12983	-0.07358
	Equal variances not assumed			-7.321	14.552	0.000	-0.10170	0.01389	-0.13139	-0.07202
SO ₂	Equal variances assumed	19.143	0.000	-9.156	21	0.000	-0.55432	0.06054	-0.68023	-0.42841
	Equal variances not assumed			-8.832	12.164	.000	-.55432	.06276	-.69086	-.41778

NO2	Equal variances assumed	3.041	.096	4.439	21	.000	.008485	.001911	.004510	.012460
	Equal variances not assumed			4.340	15.742	.001	.008485	.001955	.004334	.012635



Statistical Analysis of CO emissions in the experimental rooms under ventilated conditions

ANOVA

CO Concentration

	Sum of Squares	Df	Mean Square	F	Sig.
Between Groups	263.466	2	131.733	48.170	0.000
Within Groups	4025.548	1472	2.735		
Total	4289.014	1474			

Multiple Comparisons

Dependent Variable: CO Concentration Tukey
HSD

(I) Room Size (J) Room Size		Mean Difference (I-J)	Std. Error	Sig.	95% Confidence Interval	
					Lower Bound	Upper Bound
34m ³	19m ³	0.29851*	0.10549	0.013	0.0510	0.5460
	8.5m ³	1.00805*	0.10554	0.000	0.7604	1.2557
19m ³	34m ³	-0.29851*	0.10549	0.013	-0.5460	-0.0510
	8.5m ³	0.70953*	0.10538	0.000	0.4623	0.9568
8.5m ³	34m ³	-1.00805*	0.10554	0.000	-1.2557	-0.7604
	19m ³	-0.70953*	0.10538	0.000	-0.9568	-0.4623

*. The mean difference is significant at the 0.05 level.

Statistical Analysis of CO emissions in the experimental rooms under poorly ventilated conditions

ANOVA

CO Concentration

	Sum of Squares	df	Mean Square	F	Sig.
Between Groups	7459.477	2	3729.739	62.369	0.000
Within Groups	89223.482	1492	59.801		

Total	96682.959	1494			
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Multiple Comparisons

Dependent Variable: CO Concentration

Tukey HSD

(I) Room Size (J) Room Size		Mean Difference (I-J)	Std. Error	Sig.	95% Confidence Interval	
					Lower Bound	Upper Bound
34m ³	19m ³	-5.00647*	0.49132	0.000	-6.1591	-3.8538
	8.5m ³	-0.59151	0.49036	0.450	-1.7419	0.5589
19m ³	34m ³	5.00647*	0.49132	0.000	3.8538	6.1591
	8.5m ³	4.41496*	0.48811	0.000	3.2698	5.5601
8.5m ³	34m ³	0.59151	0.49036	0.450	-0.5589	1.7419
	19m ³	-4.41496*	0.48811	0.000	-5.5601	-3.2698

*. The mean difference is significant at the 0.05 level.

Statistical Analysis of TVOC emissions in the experimental rooms under ventilated conditions

ANOVA

TVOC Concentration

	Sum of Squares	Df	Mean Square	F	Sig.
Between Groups	0.148	2	0.074	93.094	0.000
Within Groups	1.183	1484	0.001		
Total	1.332	1486			

Multiple Comparisons

Dependent Variable: TVOC Concentration Tukey HSD

(I) Room Size (J) Room Size	Mean Difference		Sig.	95% Confidence Interval
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		(I-J)	Std. Error		Lower Bound	Upper Bound
34m ³	19m ³	0.01375*	0.00179	0.000	0.0096	0.0180
	8.5m ³	-0.01067*	0.00180	0.000	-0.0149	-0.0065
19m ³	34m ³	-0.01375*	0.00179	0.000	-0.0180	-0.0096
	8.5m ³	-0.02442*	0.00180	0.000	-0.0286	-0.0202
8.5m ³	34m ³	0.01067*	0.00180	0.000	0.0065	0.0149
	19m ³	0.02442*	0.00180	0.000	0.0202	0.0286

*. The mean difference is significant at the 0.05 level.

Statistical Analysis of TVOC emissions in the experimental rooms under poorly ventilated conditions

ANOVA

TVOC Concentration

	Sum of Squares	df	Mean Square	F	Sig.
Between Groups	1.131	2	0.565	476.442	0.000
Within Groups	1.765	1487	0.001		
Total	2.896	1489			

Multiple Comparisons

Dependent Variable: TVOC Concentration Tukey
HSD

		Mean Difference (I-J)	Std. Error	Sig.	95% Confidence Interval	
(I) Room Size	(J) Room Size				Lower Bound	Upper Bound
34m ³	19m ³	-.06619*	.00219	.000	-.0713	-.0611
	8.5m ³	-.02163*	.00219	.000	-.0268	-.0165
19m ³		.06619*	.00219	.000	.0611	.0713

34m ³		.04456*	.00219	.000	.0394	.0497
8.5m ³						
8.5m ³	34m ³	.02163*	.00219	.000	.0165	.0268
	19m ³	-.04456*	.00219	.000	-.0497	-.0394

*. The mean difference is significant at the 0.05 level.

Statistical Analysis of NO₂ emissions in the experimental rooms under ventilated conditions

ANOVA

NO₂ Concentration

	Sum of Squares	df	Mean Square	F	Sig.
Between Groups	0.002	2	0.001	14.588	0.000
Within Groups	0.124	1496	0.000		
Total	0.127	1498			

Multiple Comparisons

Dependent Variable: NO₂ Concentration Tukey
HSD

(I) Room Size (J) Room Size		Mean Difference (I-J)	Std. Error	Sig.	95% Confidence Interval	
					Lower Bound	Upper Bound
34m ³	19m ³	-0.00302*	0.00058	0.000	-0.0044	-0.0017
	8.5m ³	-0.00084	0.00058	0.312	-0.0022	0.0005
19m ³	34m ³	0.00302*	0.00058	0.000	0.0017	0.0044
	8.5m ³	0.00217*	0.00058	0.000	0.0008	0.0035
8.5m ³	34m ³	0.00084	0.00058	0.312	-0.0005	0.0022
	19m ³	-0.00217*	0.00058	0.000	-0.0035	-0.0008

*. The mean difference is significant at the 0.05 level.

Statistical Analysis of NO₂ emissions in the experimental rooms under poorly ventilated conditions

ANOVA

NO₂ Concentration

	Sum of Squares	df	Mean Square	F	Sig.
Between Groups	0.013	2	0.006	78.456	0.000
Within Groups	0.121	1502	0.000		
Total	0.133	1504			

Multiple Comparisons

Dependent Variable: NO₂ Concentration Tukey
HSD

(I) Room Size (J) Room Size		Mean Difference (I-J)	Std. Error	Sig.	95% Confidence Interval	
					Lower Bound	Upper Bound
34m ³	19m ³	0.00590*	0.00057	0.000	0.0046	0.0072
	8.5m ³	-0.00051	0.00056	0.632	-0.0018	0.0008
19m ³	34m ³	-0.00590*	0.00057	0.000	-0.0072	-0.0046
	8.5m ³	-0.00641*	0.00057	0.000	-0.0077	-0.0051
8.5m ³	34m ³	0.00051	0.00056	0.632	-0.0008	0.0018
	19m ³	0.00641*	0.00057	0.000	0.0051	0.0077

*. The mean difference is significant at the 0.05 level.

Statistical Analysis of SO₂ emissions in the experimental rooms under ventilated conditions

ANOVA

SO₂ Concentration

	Sum of Squares	df	Mean Square	F	Sig.
Between Groups	.151	2	.075	10.436	.000
Within Groups	10.716	1483	.007		

Total	10.867	1485			
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Multiple Comparisons

Dependent Variable: SO₂ Concentration Tukey
HSD

(I) Room Size (J) Room Size		Mean Difference (I-J)	Std. Error	Sig.	95% Confidence Interval	
					Lower Bound	Upper Bound
34m ³	19m ³	0.00216	0.00542	0.916	-0.0106	0.0149
	8.5m ³	-0.02016*	0.00538	0.001	-0.0328	-0.0075
19m ³	34m ³	-0.00216	0.00542	0.916	-0.0149	0.0106
	8.5m ³	-0.02232*	0.00540	0.000	-0.0350	-0.0096
8.5m ³	34m ³	0.02016*	0.00538	0.001	0.0075	0.0328
	19m ³	0.02232*	0.00540	0.000	0.0096	0.0350

*. The mean difference is significant at the 0.05 level.

Statistical Analysis of SO₂ emissions in the experimental rooms under poorly ventilated conditions

ANOVA

SO₂ Concentration

	Sum of Squares	df	Mean Square	F	Sig.
Between Groups	1.333	2	0.667	8.709	0.000
Within Groups	113.680	1485	0.077		
Total	115.014	1487			

Multiple Comparisons

Dependent Variable: SO₂ Concentration Tukey
HSD

(I) Room Size (J) Room Size		Mean Difference (I-J)	Std. Error	Sig.	95% Confidence Interval	
					Lower Bound	Upper Bound

34m ³	19m ³	-0.04903*	0.01762	0.015	-0.0904	-0.0077
	8.5m ³	0.02236	0.01761	0.412	-0.0189	0.0637
19m ³	34m ³	0.04903*	0.01762	0.015	0.0077	0.0904
	8.5m ³	0.07139*	0.01749	0.000	0.0304	0.1124
8.5m ³	34m ³	-0.02236	0.01761	0.412	-0.0637	0.0189
	19m ³	-0.07139*	0.01749	0.000	-0.1124	-0.0304

*. The mean difference is significant at the 0.05 level.

