MEASUREMENT OF MALARIA TRANSMISSION AND IMPACT OF MALARIA CONTROL INTERVENTIONS USING HEALTH FACILITY AND COMMUNITY-BASED ROUTINE REPORTING SYSTEMS

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ABSTRACT

Globally malaria still remains the most important parasitic disease of public health interest. In the recent past, most endemic countries have deployed and scaled up both preventive and curative interventions to reduce malaria transmission and, ideally, eliminate it. This has lead to global reductions in both mortality and incidence of malaria. These declines have been attributed to the reinvigoration of the global malaria control agenda by the explicit ambition of achieving elimination, which has lead to an increase in funding for national control programmes to increase coverage of preventive interventions, field compatible diagnostic tools for confirming parasite infection, and increased access to effective treatment. As a result of declines in malaria transmission, the focal nature of malaria transmission has become much more evident and has lead to consideration of surveillance as a key intervention for malaria control/elimination in its own right.

Surveillance systems have been well established in most formal health facilities but the incorporation of these systems at community level and operationalised by community health workers (CHWs) still remains limited. Additionally, these few examples of CHW-implemented surveillance systems have been typically only reporting indicators of malaria infection burden, without capturing indicators of intervention availability, deployment, coverage and utilisation, thus representing a missed opportunity for routine monitoring and evaluation of impact of interventions in “real time” to inform program planning and implementation.

The study was established as part of a multi-country study under the Malaria Transmission Consortium Project whose primary objective was to develop and evaluate new or improved methods for measuring malaria transmission. Thus the overall goal of this study was to

demonstrate how malaria transmission, and impact of interventions, could be routinely measured through a novel longitudinal community based surveillance system (CBSS) operationalised by modestly paid CHWs.

The CBSS included both passive and active surveillance activities using field – compatible test kits for *in situ* parasitological detection of malaria infections, based on which confirmed cases were treated with anti-malarial drugs, coupled with a detailed questionnaire on access and use of malaria control interventions and population characteristics. Passive surveillance was achieved conventionally whenever community members self-reported to the CHWs and active surveillance was achieved through monthly active visits to all households in their catchment populations to offer testing and treatment. In addition to recording detailed details of each patient contact in a paper patient register, weekly summaries of selected data elements were submitted by the CHWs using a mobile phone platform via short messaging system (SMS). The detailed reference data recorded in the patient register was then used to monitor malaria infection dynamics in the study population, evaluate the impact of preventative measures, such as indoor residual spraying (IRS) and long-lasting insecticidal nets (LLINs) and validate the electronic summaries submitted via SMS.

Overall, the CBSS did not routinely capture all malaria infections in the study population and was insufficient to eliminate the human parasite reservoir. This was primarily due to limited study participant participation with the monthly active testing and treatment offered by the CHWs. However, the CBSS clearly demonstrated the incremental and residual impact to supplementation of pyrethroid-treated LLINs with non - pyrethroid insecticides applied by IRS in areas where the dominant malaria vector is highly resistant to pyrethroids. The adequacy of the SMS reports submitted by the CHWs confirms the great potential of mobile

phone technology for facilitating and improving the effectiveness of community based reporting. Despite its limitations, the CBSS successfully provided programmatically relevant information regarding malaria infection dynamics across the large study area at a very affordable cost. The CHWs demonstrated their ability to not only provide treatment services but also adequately report their findings both electronically and on paper. CHWs are primarily tasked with providing routine health services at community level but clearly also have a valuable auxiliary role to play in “real time” surveillance of malaria, and most probably a range of other diseases. If the full potential of CHWs as agents of health surveillance can be realized, control programme progress can be measured through spatial and temporal mapping of transmission with greater sensitivity and at finer scales than is possible with health facilities alone, to enable improved, better-informed program planning, resource allocation and implementation.

DECLARATION

None of the contents in this thesis has been previously submitted for a degree in this or any other university. Where use has been made of the work of others, it is duly acknowledge in the text. Chapters 2, 3 and 4 have been submitted for publication or published already as papers in peer-reviewed journals in slightly different format from that presented here. The contributions of each of the various collaborators involved in each chapter are listed below:

Chapter 1: Introduction and literature review

Busiku Hamainza (BH) wrote the entire chapter. Dr. Gerry F Killeen (GFK) edited the chapter.

Chapter 2: Monitoring, characterization and control of chronic, symptomatic malaria infections in rural Zambia through monthly household visits by paid community health workers

BH, AS, CHS, HM, MK and GFK: Conceived, designed and supervised all field activities of the study. AB provided the map of the study area. AB, TE and JM: Assisted in developing the data analysis plan. BH and GFK: Drafted the manuscript in consultation with the other authors, all of whom reviewed it and provided comments. All authors read and approved the final version of the chapter.

Chapter 3: A comparison of mobile phone-based malaria reporting system with routine patient register data for capturing spatial and temporal trends in epidemiological indicators in rural Zambia

BH and GFK: Conceived and designed all field activities of the study. BH and MK supervised all field activities of the study. AB provided the map of the study area. BH, GFK, AB and JY: developed the data analysis plan. BH, GFK, AB and JY: Drafted the manuscript in consultation with the other author, who reviewed it and provided comments. All authors read and approved the final version of the manuscript.

Chapter 4 Incremental impact upon malaria transmission of supplementing pyrethroid-impregnated long lasting insecticidal nets with indoor residual spraying using pyrethroids or the organophosphate Pirimiphosmethyl

BH, CHS, HM, DC, JC, MM, MK, AS and GFK: Conceived, designed and supervised all field activities of the study. BH, CHS and GFK developed the data analysis plan. BH, CHS and GFK: Drafted the manuscript in consultation with the other authors, all of whom reviewed it and provided comments. All authors read and approved the final version of the chapter.

Chapter 5: General discussion and conclusions

BH wrote this chapter

Signed_____ (Candidate) Date_____

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DEDICATION

To my beloved family and friends, especially....

To my wife Khetiwe, you have been a rock, thank you for your love, prayers and patience

To my son and daughter, Nsondo and Chipego, you make it all worth it my babies, daddy loves you

To my father and mother, Goodfellow HR Hamainza and Beatrice EN Hamainza, thank you for your continued prayers, inspiration and encouragement, you guys are awesome, it's been a long time coming "Dr Geggez"

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To my mother-in law, Mrs Mary Chakulanda, thank you for your continued prayers and support

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"Success consists of going from failure to failure without loss of enthusiasm" – **Winston Churchill**

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LIST OF ABREVIATIONS

AL	Artemether Lumefantrine
CB	Community Based
CBSS	Community Based Surveillance System
CHW	Community Health Worker
DM-WG	Deltamethrin
DP	Diagnostic Positivity
EIR	Entomological Inoculation Rate
FTE	Full Time Equivalent
GLMM	Generalized Linear Mixed Model
HF	Health Facility
IRS	Indoor Residual House Spraying
ITT	Ifakara Tent Traps
LC-CS	Lambdacyhalothrin CS
LLINs	Long-lasting insecticidal nets
LOESS	Locally weighted scatter-plot smoother
LT	Centres for Disease Control and Prevention light traps
NMCC	National Malaria Control Centre
OR	Odds ratio
PCR	Polymerase chain reaction
PM-CS	Pirimiphosmethyl CS
PM-EC	Pirimiphosmethyl EC
RDT	Rapid Diagnostic Tests
SMS	Short Messaging System

CHAPTER ONE

GENERAL INTRODUCTION AND LITERATURE REVIEW

1.1 Introduction

Globally, there have been significant strides made in efforts to control malaria over the last decade (WHO 2013; WHO 2014). These positive developments have lead to the adoption of even more ambitious goals of malaria elimination from most endemic countries and, eventually, its global eradication (WHO 2013; WHO 2014). These positive gains have been attributed to the integrated approach of malaria control through cost-effective preventive and curative interventions, facilitated by a renewed global financial, technical and infrastructural support platform (Eisele, Larsen et al. 2012). The key vector control interventions are presently protection of sleeping spaces with long-lasting insecticidal nets (LLINs) and indoor residual spraying (IRS) of houses, and these are coupled with field compatible confirmatory diagnostic tools and artemisinin-based combination therapies for treatment (Eisele, Larsen et al. 2012; WHO 2014).

The use of curative drugs has been primarily focussed on addressing clinical manifestations of infection (McCombie 1996; Tanner and Vlassoff 1998; Najera 2001). However the majority of malaria parasite infections in the human populations exist as chronic infections that are often classified as “asymptomatic” because the infected human hosts do not feel ill enough to seek care, thus sustaining malaria transmission. Thus, chemotherapy for reduction of malaria transmission by such chronic carriers is being revisited as a complementary intervention with which to supplement the main-stream vector control and passive curative interventions (Okell, Drakeley et al. 2008; Okell, Ghani et al. 2009; Gosling, Okell et al. 2011) particularly in areas of low and seasonal transmission (Molineaux and Gramiccia 1980; von Seidlein, Walraven et al. 2003; WHO 2010). Historically, these approaches have been evaluated in the past in terms of their ability to interrupt malaria transmission. However, the impact of chemotherapy interventions has been mediocre and

short-lived at best, particularly when used without population wide vector control interventions (Molineaux and Gramiccia 1980; Jeffery 1984). Additionally, there have been concerns about the development of parasite resistance to the treatments due to wide scale and prolonged use (Greenwood 2010; Kaneko 2010). Malaria chemotherapy beyond routine passive curative service provision is broadly described as either mass drug administration (MDA) or mass screening and treatment (MSAT)(WHO 1963; Gosling, Okell et al. 2011). The former encompasses administration of anti-malarial drugs to entire targeted populations irrespective of confirmation of parasite infection, while the latter is provision of treatment to populations that have been subjected to a confirmatory diagnostic test and yield a positive test result. A major limitation of MSAT lies in the inability of currently available rapid diagnostic tests to detect low density infections, which can still contribute to the infection reservoir (Crowell, Briet et al. 2013). Currently, artemisinin-based combination therapies (ACTs) is the only available drug class that can potentially be used for MDA and MSAT. In addition to clearing asexual stage parasites, ACTs are also known to kill sexual gametocyte stages when they are immature so they may also reduce malaria transmission (von Seidlein and Greenwood 2003; Okell, Drakeley et al. 2008; Bousema, Okell et al. 2010). Chemotherapy interventions are thought to be most effective for transmission control when applied in low to moderate transmission areas, especially in the low transmission season when vector populations are at their lowest densities so the risk of re-infection is reduced (Griffin, Hollingsworth et al. 2010; Okell, Griffin et al. 2011).

Detecting temporal and spatial variations in malaria infection risk, to guide implementation of preventative and curative interventions, including chemotherapy, requires a viable surveillance system. These systems are usually weak in resource-limited malaria-endemic countries, thus limiting accurate assessment of disease trends and guide program planning

and implementation (WHO 2013). Additionally, they are mostly facility based and thus biased to reporting only acute symptomatic cases that seek care, which will therefore not be adequate to eliminate reservoirs of chronic, sub-acute, non-reporting infections. Thus in endemic countries the "reach" of the health facility, with regard to both service provision and derived reporting and surveillance functions may be increased with the use of community health workers (CHWs). These secondary reporting functions, integrated into an extended Health Management Information System (HMIS), should not distract in any way from the primary function of CHWs for providing diagnostic and curative services but should rather be viewed as an additional "spin-off" benefit that delivers enhanced surveillance to describe spatial and temporal disease trends at a fine scale. The use of community based surveillance systems (CBSS) have been underutilized globally with regard to malaria disease burden and service delivery indicators (WHO 2013), despite the availability of well-documented evidence of the ability of CHWs to both diagnose and treat malaria (Delacollette, Van der Stuyft et al. 1996; Yeboah-Antwi, Pilingana et al. 2010; Counihan, Harvey et al. 2012; Kalyango, Rutebemberwa et al. 2012), as well as report their findings (Hopkins, Talisuna et al. 2007; Alba, Hetzel et al. 2011; Kalyango, Rutebemberwa et al. 2012). In principle, these CBSS have potential to directly evaluate progress of program implementation with regard to intervention coverage, utilization and associated epidemiological outcomes in the catchment areas they cover. Such fine scale information would be invaluable to local health authorities for guiding resource prioritization and planning. Furthermore, recent advances in the availability of mobile phone technology in rural Africa suggest that it should be possible for such data to be reported electronically in real time so that they can be compiled, assessed and acted upon in a far timelier manner than existing paper-based HMIS platforms(Kamanga, Moono et al. 2010; Brieger 2012;

Zurovac, Talisuna et al. 2012).These innovations should be scalable and functional in low resource settings, easy to use and inexpensive.

However, the full benefits of such CBSS in a routine and sustainable program framework need to be rigorously evaluated and optimized before they can adopted at national scale up.

1.1.1 Goals and Objectives

1.1.2 Goal

The overall goal of the study was to demonstrate how paid CHWs offering malaria diagnosis and treatment services both passively and actively can be utilised to routinely monitor, characterize and control malaria infection burden and to assess the impact of supplementary vector control measures upon it.

1.1.3 Objectives

1.1.3.1 To evaluate the effectiveness of passively and actively provided malaria diagnosis and treatment services by CHWs for monitoring, characterizing and controlling infection burden.

1.1.3.2 To compare and contrast a mobile phone-based malaria reporting system with source participant register data for capturing spatial and temporal trends in epidemiological indicators of malaria transmission collected by community health workers in rural Zambia.

1.1.3.3 To demonstrate how community-based surveillance can be used to estimate the incremental protective efficacy provided by alternative IRS regimes in an area of high ITN coverage, upon diagnostic positivity and vector density.

1.2 Literature Review

1.2.1 Introduction to malaria

Malaria still remains the world's most important parasitic disease (WHO 2013; WHO 2014). Malaria is caused by a blood infection of protozoan parasites from the genus *Plasmodium*, which is transmitted from one human to another by female *Anopheles* mosquitoes. The species that infect humans are *Plasmodium falciparum, P. malariae, P.ovale and P.vivax* (McKenzie and Bossert 1997; Carter and Mendis 2002; Zimmerman, Mehlotra et al. 2004; Nadjm and Behrens 2012). A fifth species, *P. knowlesi*, has also been identified as a clinically significant pathogen in humans. This species, found in southeast Asia is typically a malaria parasite of the long and pig tailed monkeys (macaques) (Garnham, Lainson et al. 1957; Chin, Contacos et al. 1965; Singh, Sung et al. 2004; Cox-Singh, Davis et al. 2008; White 2008; Singh and Daneshvar 2010; Antinori, Galimberti et al. 2012). Globally, by far the most common of these are *P. falciparum* and *P.vivax*, with the former being the most lethal of the malaria parasites infecting humans and the latter being the most common and widespread across the Middle East, Asia, Central and South America (Table 1), where it is responsible for 70–90% of the global malaria burden (Carter and Mendis 2002; Hay, Guerra et al. 2004; Galinski and Barnwell 2008; Guerra, Howes et al. 2010; Prajapati, Joshi et al. 2011; Nadjm and Behrens 2012).

Table 1. Human malaria species distribution and clinical /pathologic features (Adapted from Nadjm & Behrens, 2012)

Species	Distribution	Clinical/Pathologic Details
P. falciparum	Widespread throughout the tropics	Causes the most fatalities Pronounced rosetting and sequestration
P. vivax	Widespread in tropics and subtropics, however quite rare in most parts of Africa	Relapses caused by hepatic hypnozoites Requires Red Blood Cell Duffy antigen for entry into cells
P. ovale	Patchy in West Africa and southwest Pacific	Relapses caused by hepatic hypnozoites
P. malariae	Patchy, worldwide	Late recrudescence and chronic infections Nephrotic syndrome (urinary protein, edema & increased cholesterol) (rarely)
P. knowlesi	Malaysia, Indonesia and other southeast Asia	Zoonosis (macaque monkeys are common hosts) Severe disease

Malaria is an acute febrile illness with symptoms that may appear from seven to fifteen days post exposure to an infective mosquito bite, depending on the causative parasitic species (Miller, Good et al. 1994; Miller, Baruch et al. 2002; Nadjm and Behrens 2012). The multiplication of the parasites in the red blood cells results in several symptoms which may include fever, chills and vomiting among others, with the possibility of progression to coma or even death in severe cases (WHO 1963; Bruce Chwatt 1985; Nadjm and Behrens 2012). The severity of these symptoms is dependent on the patient predisposition with regard to immunity to malaria (Smith and Schapira 2012) and the causative infective strain of the parasite (Jeffery and Eyles 1954; Muller, Genton et al. 2009).

A key feature of malaria parasite infections is the periodicity of the fevers they cause, which occur in multiples of 24 hour intervals (Table 2) (Cunha and Cunha 2008; Antinori, Galimberti et al. 2012). *P. falciparum* infection is characterized by acute cerebral, renal, or

gastrointestinal manifestations and severe paroxysms that reoccur every 2 days (48 hour interval) (malignant tertian) (Cunha and Cunha 2008; Antinori, Galimberti et al. 2012). If *P. falciparum* infection is not urgently treated, it may rapidly progress to severe disease and even death. Tertian fever or fever that occurs on a 3 day cycle (48 hour interval) is caused by *P.ovale* and *P. vivax* (Collins and Jeffery 2005; Antinori, Galimberti et al. 2012). Infection with these two parasites is characterised by relapses due to presence of the dormant liver stage hypnozoites, which are known to have long latent periods that may range from weeks, months to years (Lysenko and Beljaev 1969; Adak, Sharma et al. 1998; Collins and Jeffery 2005; Garcia 2010; Antinori, Galimberti et al. 2012). *P. malariae* is the causative agent for quartan fever, which is an acute febrile episode that returns every fourth day (72 hours interval) (Collins and Jeffery 2007; Cunha and Cunha 2008; Garcia 2010; Antinori, Galimberti et al. 2012). This infection has been known to be benign for up to 50 years in a few exceptional cases and is usually associated with low levels of parasitaemia infection (Collins and Jeffery 2007). *P.knowlesi* infection exhibits a unique daily or quotidian fever cycle, thus patients may not exhibit typical paroxysms patterns but rather have stable low-grade fever or an irregular lessening and then re- intensifying fever pattern (Chin, Contacos et al. 1965; White 2008; Ta, Salas et al. 2010).

Table 2. Characteristics of infection with the five human infection species (Adapted from Cunha & Cunha, Antinori *et al*., 2012, and White & Imwong 2012)

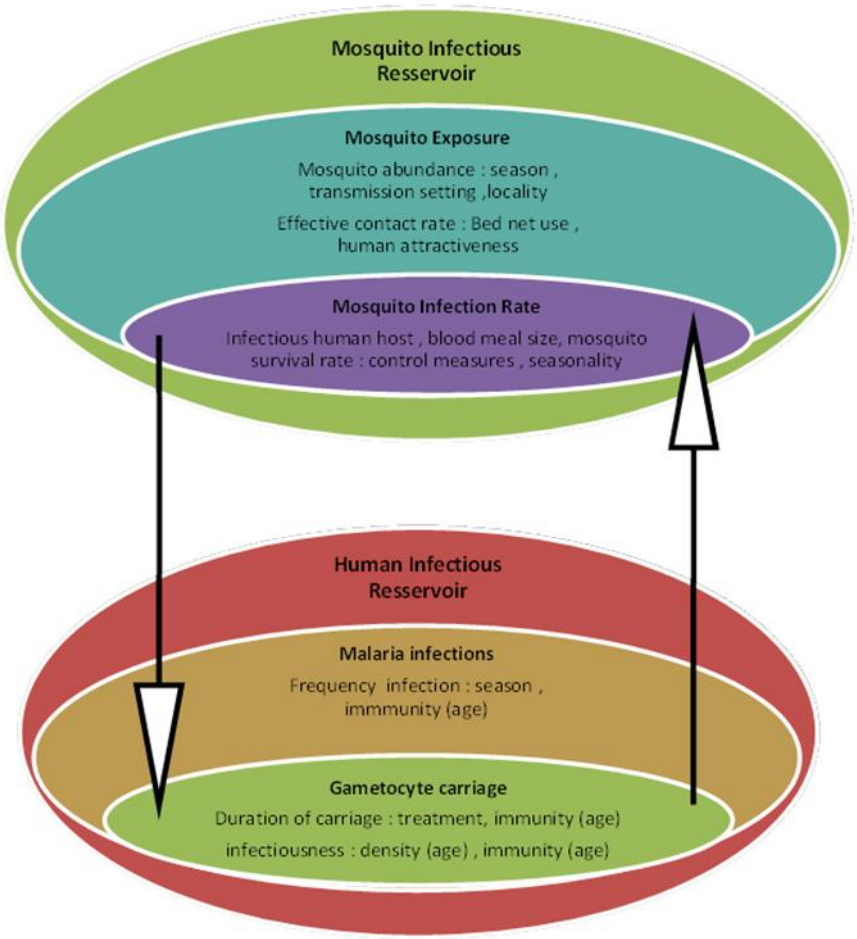
Characteristics	*P. falciparum*	*P. knowlesi*	*P. malariae*	*P.ovale*	*P.vivax*
Pre-erythrocytic stage (days)	5-7	8-9	14-16	9	6-8
Hypnozoite stage duration (months)	0	0	0	½ – 9 months (exceptional cases of up to 4 years have been reported)	¾ -9 months
Types of malaria	Malignant tertian / Pernicious tertian/ semi tertian	Quotidian	Quartan	Ovale tertian	Benign tertian
Pre-patent period (days)	9-10	9-12	15-16	10-14	11-13
Erythrocytic cycle (days)	48	24	72	50	48
Red cells affected	All	All	Mature erythrocytes	Reticulocytes	Reticulocytes
Parasitaemia per µL					
Average Maximum	20,000-500,000 2,000,000	600 -10,000 236,000	6,000 20,000	9,000 30,000	20,000 100,000
Febrile paroxysm (hours)	16-36 or longer	8-12	8-10	8-12	8-12
Severe malaria	Yes	Yes	No	No	Yes
Relapses from liver forms	No	No	No	Yes	Yes
Recurrences	Yes (treatment failure)	Yes	Yes (as long as 30-50 years after primary attack)	No	Yes (treatment failure)

1.2.2 Dynamics of Malaria transmission

Malaria has a complex epidemiology which is determined by four main factors which include; the environment, vector, parasite and host (Draper and Smith 1957; Hay, Guerra et al. 2009; Guerra, Howes et al. 2010). The interplay of these ecological and biological factors for both the human and vector hosts influences malaria transmission (Figure 1) (Drakeley 2013). For the malaria vector to transmit malaria, several key elements of the vector

population have to be in place and these include; parasite incubation period in the vector, number of blood meals taken per vector per day, density of vectors in relation to humans and the daily survival of the vector (Shililu, Ghebremeskel et al. 2003; Hay, Guerra et al. 2004; Drakeley 2013). The time required for the sporogonic cycle in the mosquito varies and is dependent on the *Plasmodium* species and external temperature. The later also affects mosquito longevity (Onori and Grab 1980; Molineaux, Muir et al. 1988; Beier 1998; Sinka, Bangs et al. 2012).

Figure 1.Factors that influence infectivity (Adapted from Drakeley 2013)



Optimal parasite development in the vector occurs between the temperatures 20°C to 30°C, however sporogony has been known to take place even at lower temperatures that range from 16°C to 40°C (Vanderberg and Yoeli 1966; LaPointe, Goff et al. 2010). Rainfall plays a part in providing habitat for breeding of the vector. In Africa *Anopheles gambiae* and *An. arabiensis* can utilise a wide variety of permanent and temporal water bodies that are exposed to strong sunlight, while *A. funestus* strongly prefer more permanently shaded water bodies (Gillies and Meillon 1968). Thus, higher temperatures in combination with high relative humidity, favour shortened parasite development, increased vector longevity and subsequent number of blood meals taken by the *Anopheles* vector (Onori and Grab 1980; Molineaux, Muir et al. 1988; Shililu, Ghebremeskel et al. 2003; Zhou, Minakawa et al. 2004; Reyburn, Mbatia et al. 2005; Hay, Guerra et al. 2009).

Only *Anopheline* mosquitoes that survive longer than the sporogonic phase are able to transmit malaria (Zucker 1996). However, only a small proportion of the vectors actually live long enough in nature to be able to transmit the infection. This is alluded to in the basic reproductive number (R_0),which is the expected number of secondary cases produced, in a completely susceptible population, by a typical infected individual during its entire period of infectiousness of vector borne infections (Diekmann, Heesterbeek et al. 1990; Dietz 1993; Smith, McKenzie et al. 2007). These infections are highly dependent on the longevity of the vector, specifically its infective life expectancy (Garrett-Jones 1964; Dye 1992; Zucker 1996). Thus, the number of parasite infected person's increases when R_0 is greater than 1 and decreases if R_0 is less than 1. In essence, the goal of infectious disease eradication is vested in the deployment of interventions aimed at keeping R_0 below 1 (Smith and Schapira 2012). The estimates of R_0 are based on the entomological inoculation rate (EIR), which is the mean number of infectious bites received by a person annually and the

parasite ratio (PR), which is the prevalence of malaria infection in humans (Smith, McKenzie et al. 2007). The relationship between EIR and PR varies as a result of the human population's heterogeneity due to the fact that some populations are bitten more than others and differ in levels of susceptibility to infection per bite due to among others mosquito feeding preferences and their proximity to larval habitats (20 % of the population receive 80 % of the infections) (Smith and Ellis McKenzie 2004; Smith, Dushoff et al. 2005).

Mathematical epidemiologic models provide insight into infectious disease transmission and their concepts have been utilized in design of various control programmes including the Global Malaria Eradication Programme in the 1950s and 60s (Farrington, Kanaan et al. 2001; Smith and Schapira 2012). One such model pioneered by Ross and later modified with contributions from Macdonald and others lead to an understanding of the linked phenomena for mosquito borne pathogen transmission (Ross 1911; Macdonald 1957; Smith, Battle et al. 2012). The model demonstrated the dependence of R_0 on the density of vectors, their preferred feeding behaviour, vector survival and the infectiousness of the host (Smith and Schapira 2012). The Ross-Macdonald model was defined as follows (Ross 1911; Macdonald 1957):

$$\frac{dx}{dt} = \left(\propto b\frac{M}{N}\right) y(1-x) - rx$$

$$\frac{dy}{dt} = \propto x(1-y) - \mu y$$

Where:

x is the proportion of the human population infected?

y is the proportion of the female mosquito population infected;

N is the size of the human population;

M is the size of the female mosquito population;

$m = \frac{M}{N}$ is the number of female mosquitoes per human host;

a is the rate of biting on man by a single mosquito (number of bites per unit time);

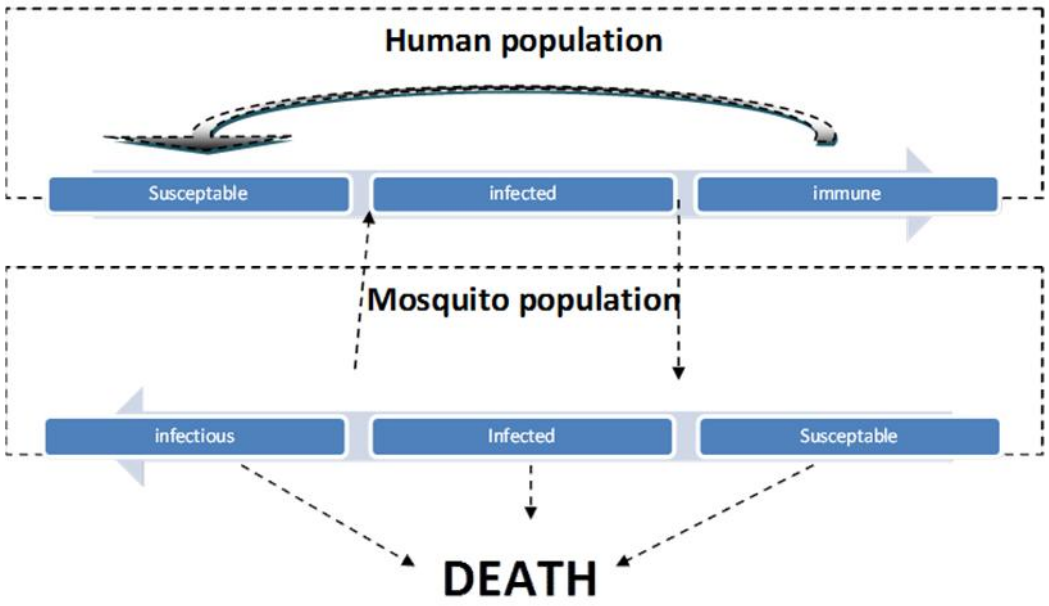
b is the proportion of infected bites on man that produce an infection;

r is the per capita rate of recovery for humans ($1/r$ is the average duration of infection in the human host);

μ is the per capita mortality rate for mosquitoes ($1/\mu$ is the average life time of a mosquito).

Despite its simplistic structure the model (Figure 2) allows for a comparison and subsequent interpretation of broad epidemiological patterns. A few assumptions were made which included ; the human host has no acquired immunity, humans are bitten by mosquitoes randomly, and mosquito and human populations are homogenous in nature (Koella 1991).

Figure 2.Structures of Ross-Macdonald model of malaria transmission (Adapted from Koella 1991)



R_0 best describes the basic results of the model. According to Aron and May, R_0 is derived using the formula (Aron and May 1982):

$$R_0 = \frac{M}{N}\frac{a^2 b}{\mu r}$$

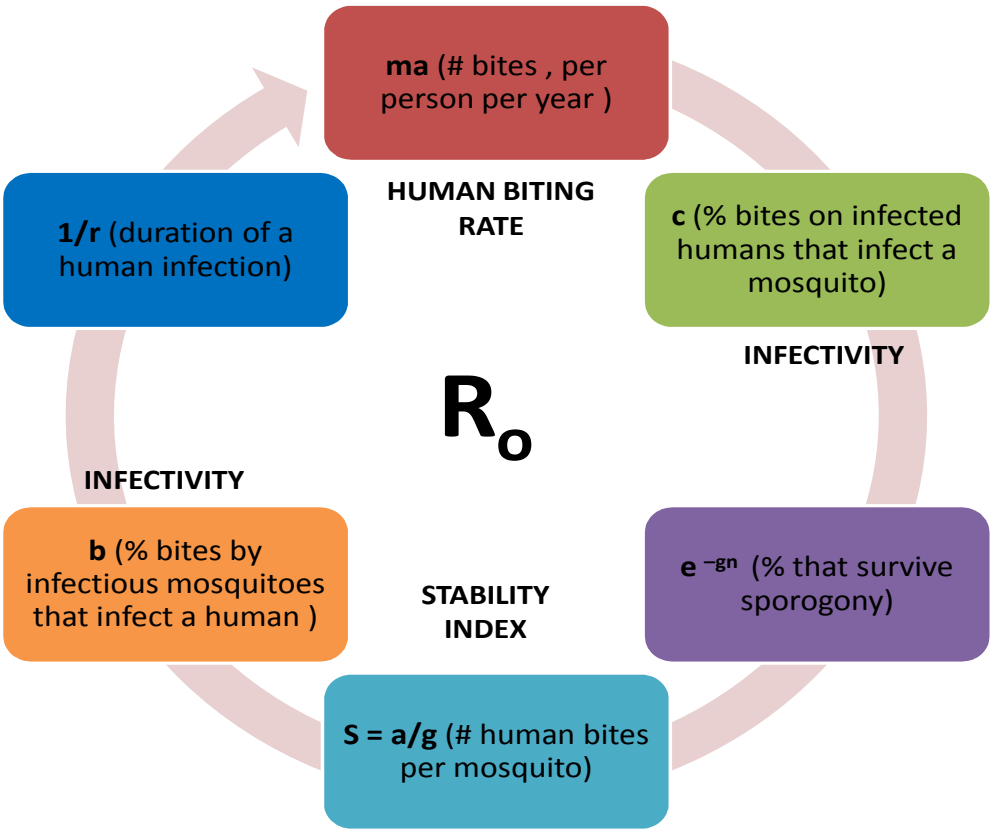
From the equation above it is clear that, malaria transmission is aided by a high number of contacts with mosquitoes that a single human has per unit time (high aM/N); a subsequent high number of mosquitoes that a single human infects over the entire infectious period (high $\left(\frac{M}{N}\right)\left(\frac{a}{r}\right)$); a high number of contacts with humans that one mosquito has per unit time (high a); a high probability of transmission from an infectious mosquito to a susceptible human (high b) ; a long mean infectious period of the mosquito (long $1/\mu$) and; high number of humans that a mosquito infects through its infectious lifetime (high ab/μ). The model assumes the probability from an infectious human to a susceptible mosquito is equal to 1 and $1/r$ is mean duration of the latent period of the human (Aron and May 1982; Koella 1991).

The model has some key elements missing such as the assumption that the population of humans and mosquitoes is constant and does not take into consideration the period of time a mosquito requires to develop into an adult for it to be able to transmit malaria (Smith, Battle et al. 2012) and that biting rate of the vectors per host is dependent on the biomass of the person, thus overestimating the challenge faced by small children (Smith, Killeen et al. 2004). However over the years, others have continued to work on this model to fully develop the key elements (Smith, Battle et al. 2012). Some of this work has been based on various assumptions which include ; there is a host population that is susceptible to infection, some members of the population are exposed to infection but are not able to pass

the infection on during the latent period, infected individuals can lead to more infected populations through interaction with susceptible groups, there is a part of the population that recovers from the infection, some infected population die due to infection and that the number of mosquitoes and humans is not constant (Smith, McKenzie et al. 2007; Mandal, Sarkar et al. 2011).

In the same vain as the Ross –Macdonald model, Smith and colleagues proposed a model based on the life cycle of *P. falciparum*, which forms the basis for the formula used to calculate R_0 as illustrated in Figure 3 (parameters described in table 3).

Figure 3.The cycle model and reproductive number (Adapted from Smith *et al.*, 2007)



This model is based on the assumption that all humans are bitten at the same rate and that the population is infinite. Thus, in such populations, humans that are bitten most absorb more parasites and infect more mosquitoes which results in an increase in R_0 (Smith and Ellis McKenzie 2004; Smith, McKenzie et al. 2007). The model assumes a stable relationship between the EIR, PR and vector capacity, which is the number of infectious bites that arise from all mosquitoes infected by one person on a single day, the human infectivity to mosquitoes (*c*), and the stability index (*S*), the number of bites a vector makes on a human in its lifetime (Smith, McKenzie et al. 2007).

Table 3. Parameters of life cycle model and reproductive number (Adapted from Smith *et al*., 2007)

Parameter	Description
a	Human feeding rate: the number of bites on a human, per mosquito, per day. Let f denote the feeding rate, i.e., the number of bites, per mosquito, per day, and Q the proportion of bites on humans. The human feeding rate is the product a= fQ.
b	Infectivity of mosquitoes to humans: the probability that a human becomes infected from a bite by an infectious mosquito. With pre-erythrocytic immunity, the infectivity of mosquitoes may depend on EIR, bE
c	Infectivity of humans to mosquitoes: the probability that a mosquito becomes infected from a bite on an infected human. Infected humans are not infectious all the time and infectious bites transmit less than perfectly. With transmission blocking immunity, infectivity of humans may depend on EIR, cE
g	Death rate of mosquitoes. The probability a mosquito survives one day is p = e-g, so g =-ln p. The expected lifespan of a mosquito is 1/g.
m	Number of mosquitoes per human. Assuming adult mosquitoes emerge at a constant rate λ, per human, then m = λ/g.
n	Number of days required for a mosquito to complete sporogony.
1/r	Expected waiting time to naturally clear a simple infection.

It is however important to note that malaria transmission, particularly *P. falciparum* malaria, does not reach its full potential primarily due to host naturally acquired immunity (Smith

and Schapira 2012). A key effect of acquired immunity is the reduction of asexual parasite stages, which consequently reduces the number of potential sexual stage parasites thus affecting the infectivity of humans to mosquitoes particularly in endemic areas (Alavi, Arai et al. 2003; Smith and Schapira 2012).This however does not prevent transmission from taking place, as it has been shown that even at low sexual stage densities transmission does occur, thus bottlenecking the parasite from reaching its full transmission potential (Alavi, Arai et al. 2003; Bousema and Drakeley 2011; Smith and Schapira 2012).

1.2.3 Global Burden of Malaria

Malaria still remains a major public health problem both globally and regionally. According to the World Health Organisation (WHO), there were 216 million cases of malaria globally in 2010, of which 81% were in the African region and an estimated 655,000 people (91% of these in the African region) died from the disease, many of whom were children under the age of five (WHO 2012). In the last decade there has been great progress made towards the prevention of malaria. According to the WHO, global malaria mortality and incidence rates have reduced by 26% and 17% respectively, since 2000 (WHO 2012). The declines in malaria deaths has been attributed to an increase in funding which has lead to increased population wide coverage of malaria control interventions coupled with effective confirmatory diagnosis and treatment (Eisele, Larsen et al. 2012). The accurate estimation of malaria attributable mortality still remains a challenge as most deaths occur at home and thus difficult to authenticate (Korenromp, Williams et al. 2003; de Savigny and Binka 2004). According to Murray and colleagues, 433,000 more deaths occurred worldwide in the population under the age of 5 years or older in 2010 than was suggested by the WHO estimates. Of these malaria deaths, 104,000 were estimated to have occurred outside Africa (Murray, Rosenfeld et al. 2012).

In addition to morbidity and mortality, malaria also exerts an economic burden where it occurs both at a national and household level (Sachs and Malaney 2002; Chima, Goodman et al. 2003; Laxminarayan 2004). These economic impacts at a national level may include, decreased productivity due to brain damage from cerebral malaria, working days lost due to sickness, cost of health care, days lost in education due to reduced cognitive development particularly in children and a loss of investments and tourism (Greenwood, Bojang et al. 2005; Fink, Olgiati et al. 2013). At the household level, these impacts may include, work time lost by the unwell household member, time spent giving care to sick family members, loss of productivity, costs associated with seeking care such as transportation and treatment and premature mortality among others (Sachs and Malaney 2002; Chima, Goodman et al. 2003; Laxminarayan 2004).

While Malaria transmission is geographically confined to the tropics and sub-tropics, it was formerly common in many parts of temperate northern Europe, America and Asia (Sachs and Malaney 2002; Kiszewski, Mellinger et al. 2004). The favourable climatic conditions (appropriate temperature and rainfall patterns) in the tropics and subtropics coupled with the human biting habits of the mosquito vectors and the presence of complementing malaria vectors (*An. gambiae* (wet season) and the *An. funestus* (dry *season*)) make these areas favourable for malaria transmission particularly in west and central parts of Africa (Bruce-Chwatt, Garrett-Jones et al. 1966; Gillies and Coetzee 1987; Killeen, McKenzie et al. 2001; Keiser, Utzinger et al. 2004; Kiszewski, Mellinger et al. 2004). Approximately 39% of the global malaria burden occurs in central and south-eastern Asia (Hay, Okiro et al. 2010). The Asian–Pacific region accounts for 88% of all malaria outside Africa (second to Africa). This has been attributed to the presence of a wide diversity of physiologically competent

vector species that exhibit varied behavioural patterns and preferences (Foley, Rueda et al. 2007; Hay, Okiro et al. 2010).

1.2.4 History of Discovery and Identification of Malaria Parasites

In 500 BC, Greek physician Hippocrates was first to describe the clinical symptoms of malaria and hypothesized on its linkage to stagnant water, which at the time was thought to be the causative agent (Cunha and Cunha 2008; Pappas, Kiriaze et al. 2008; George 2009; Wiser 2011). In 1879, Patrick Manson, a Scottish physician demonstrated that Bancroft's filarial (*Wuchereria bancrofti*) vector was a mosquito of the genus *Culex.* He, as Hippocrates, proposed that drinking water had something to do with the transmission of malaria (Dobson 1999; Capanna 2006). A year later in 1880, when Charles Laveran, a French army surgeon described malarial parasites in the blood of patients during malarial fever episodes, which he initially named as *Oscillaria malariae* (Bruce Chwatt 1985; Smith and Sanford 1985; Dobson 1999; Guillemin 2002; Schlagenhauf 2004). Ettore Marchiafava and Augusto Celli followed up on the work done by Laveran and proposed the name *Plasmodium* for this protozoan (Majori 1999). Between 1883 and 1885, the duo confirmed Laveran's work, and added further detail by differentiating tertian and benign fevers on the basis of their sexual stages of parasite development (Ascenzi 1999; Majori 1999; Cox 2010). They also studied the morphological differences of *Plasmodium* parasites in collaboration with a colleague named Bignami, which later on in 1892, lead to the identification of *P. falciparum* as a distinct parasite taxon (Majori 1999; Cox 2010; Ferroni, Jefferson et al. 2012). Prior to this in 1890, Giovanni Grassi and Raimondo Filetti introduced the names *P. vivax* and *P. malariae* for two other human malaria parasites (Collins and Jeffery 2007). Laveran's observations were further confirmed in 1886 by other scientists that included Camillo Golgi, George Sternberg, William Councilman, and William Osler (Smith and Sanford 1985; Cox

2010). Golgi related the clinical symptoms and fever episodes with the schizogonic phase of *Plasmodium* and showed that tertian and quartan intermittent fevers where due to two different *Plasmodium* species, namely *P. malariae* and *P. vivax* (Ascenzi 1999; Capanna 2006; Tognotti 2007). William Welch, continued to review and build on Laveran's work and, in 1897, formally named the malignant tertian malaria parasite *P. falciparum*. By the early 1890s, Laveran's germ was widely considered the true cause of malarial fever as a result of this painstaking investigation of the blood stages (Smith and Sanford 1985), however the mode of transmission between infected humans remained a mystery.

In 1897 Ronald Ross discovered that *culicine* mosquitoes transmitted *P. relictum,* an avian malaria parasite, and proposed that human malaria parasites may also be transmitted by mosquitoes. By 1899, Ross demonstrated that human malaria parasites were in fact transmitted by *anopheline* mosquitoes (Ross 1897; Bruce Chwatt 1985; Dobson 1999; Guillemin 2002; Schlagenhauf 2004; Cox 2010). John Stephens in 1922, described *P. ovale* as an additional human malaria parasite (Majori 1999) while *P. knowlesi* was first described in 1931 by Robert Knowles and Biraj Mohan Das Gupta (Garnham, Lainson et al. 1957; Singh, Sung et al. 2004; Singh and Daneshvar 2010).

1.2.5 Life Cycle of Human Malaria Parasites

The malaria parasite has an extraordinary complex, multistage life cycle occurring within two distinct hosts, the vector mosquito and the vertebrate (Bruce-Chwatt 1985). The life cycle of *Plasmodium* species comprises of three comprehensive stages, the exoerythrocitic and erythrocytic stages in the vertebrate host and the sporogonic cycle in the mosquito (Figure 4). The humans and other vertebrates act as the intermediate host for the parasite, while the mosquito, in which the sexual reproduction takes place, is considered to be the definitive host (Bruce Chwatt 1985).

The life cycle of malaria infection begins in the skin with an infected *Anopheles* female mosquito bite. When the mosquito takes a blood meal, its proboscis probes into the host's skin and deposits saliva to avoid blood coagulating (Beier 1998; Krettli and Miller 2001; Vaughan, Aly et al. 2008). A single mosquito ejects a few hundred skin infecting sporozoites which glide from the salivary gland cavities into the skin of the host. This is immediately followed by a rapid immuno-suppression of the host's immune system, which subsequently enables the establishment of early tolerance to pre-erythrocytic stages of the parasite (Guilbride, Guilbride et al. 2012). The sporozoites continue to glide through the skin until they reach a blood vessel where they then break the endothelial barrier to gain access to the blood circulatory system (Vaughan, Aly et al. 2008). Once in the blood stream, the sporozoites begin to migrate to the liver. The mechanism of locating the liver irrespective of where the sporozoites entered the host is guided by the unique properties of liver blood vessels that are rich in oxygen and nutrients (Vaughan, Aly et al. 2008). Once in the liver, the sporozoites initially invade and pass through a Kupffer cell after which they glide through several hepatocytes before finally taking residence in a hepatocyte, thus beginning the liver stage of development (Krettli and Miller 2001; Miller, Baruch et al. 2002; Vaughan, Aly et al. 2008). This movement of sporozoites through the hepatocytes leads to secretion of host hepatocyte growth factor, which makes hepatocytes susceptible to infection (Carrolo, Giordano et al. 2003). The mechanism that guides selection of the final hepatocyte to invade and take residence in to initiate the replication process is still not well understood (Vaughan, Aly et al. 2008). This invasion of the hepatocytes marks the beginning of the asexual exoerythrocytic schizogonic cycle, with each invading sporozoite developing into a schizont that contains approximately 10,000 – 30,000 merozoites within the hepatocyte, depending on the parasite species (Fujioka and Aikawa 2002). Each merozoite is able to

invade a red blood cell once released from the liver. A dormant stage called the hypnozoite has the ability to remain in the liver for several weeks to months to years before initiation of pre-erythrocytic schizogony occurs in *P.vivax* and *P. ovale.* These hypnozoites are responsible for relapses which characterise infections of these specific *Plasmodium* species (Fujioka and Aikawa 2002). Thus, malaria disease only occurs when the asexual parasite stage multiplies within the host red blood cell (Weatherall, Miller et al. 2002).

A series of extracellular recognition events between the erythrocyte receptors and merozoite ligands occur during the invasion process (Gaur, Mayer et al. 2004; Dvorin, Bei et al. 2010; Crosnier, Bustamante et al. 2011). The attachment / docking of the merozoite to the red blood cell occur between any of the surfaces of these two cells. A reorientation of the merozoite occurs with its apical end placed against the cell and resulting in the formation of a junction with the cell. The merozoite then pulls itself into the cell and a vacuole is formed in the process (Miller 1977; Krettli and Miller 2001; Fujioka and Aikawa 2002). When this attachment is occurring, the red blood cell undergoes a wave of deformation, followed by another resealing of the red blood cell membrane (Miller 1977; Hadley 1986). As the merozoite pushes its way into its host red blood cell, a parasitophorous vacuole is created that seals the invading merozoite from the host-cell cytoplasm to facilitate its continued development in an environment that is separated from the host cell's normal processing and presentation of its antigen mechanisms (Miller 1977; Greenwood, Fidock et al. 2008). This rapid invasion of the red blood cells by the merozoites ensures minimal exposure of the invading parasite, thus protecting these parasite forms from any host immune responses (Cowman and Crabb 2006; Greenwood, Fidock et al. 2008).

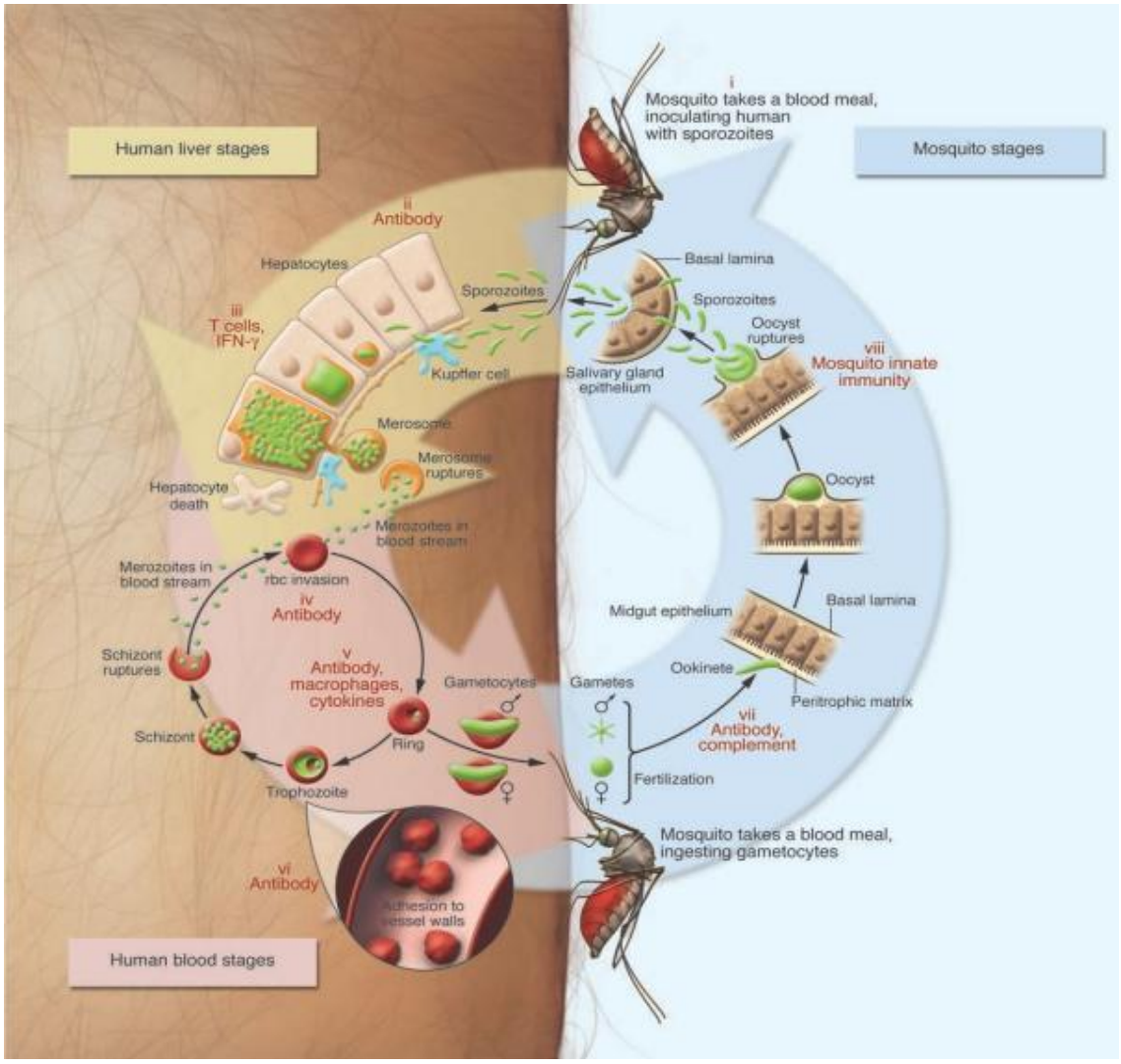
Once the parasite has entered the red blood cell, it undergoes a trophic period in which the parasite enlarges and this is followed by an asexual replication. The early stage or immature trophozoite is referred to as the ring form. The parasite continues its enlargement, losing its "ring" structure to become a mature trophozoite. During this trophic period, the parasite ingests the host cell cytoplasm and breaks down the haemoglobin into amino acids in a process known as proteolysis (Goldberg, Slater et al. 1990). The trophozoite stage ends when nuclear division begins and marks the beginning of the schizont stage (Bannister and Mitchell 2003; Cowman and Crabb 2006). During the process of red blood cell (erythrocytic) schizogony, there are three to five cycles of nuclear replication depending on the parasite species. The process of nuclear replication is followed by a budding process, in which the merozoites bud from a mature schizont or segmenter (Bannister and Mitchell 2003; Cowman and Crabb 2006). Once this process is completed, the infected red blood cell ruptures, thus releasing the merozoites. During this process, the parasitophorous-vacuole membrane ruptures first due to the action of proteases releasing the merozoites within the intact red blood cell. This leads to swelling of the red blood cell and weakening of its cytoskeleton resulting in rupture and subsequent release of the merozoites into free circulation within the blood plasma (Fujioka and Aikawa 2002; Bannister and Mitchell 2003; Cowman and Crabb 2006). The released merozoites invade new red blood cells initiating another cycle of schizogony. The invasion of a red blood cell by a merozoite results in the release of up to 32 new copies of itself in approximately 48 hours (Lew 2005). This dramatic increase in the parasite burden explains the rapid pathogenesis of malaria (Miller, Good et al. 1994; Miller, Baruch et al. 2002). The process of rupture of infected red blood cell occurs in a synchronous manner, releasing suddenly accumulated antigens and waste products from large numbers of parasites within hours or even minutes, thus causing the periodic

rapid onset of fever paroxysm that are associated with malaria infection. In *P. falciparum* infections, early ring stage and mature trophozoite - and schizont- infected red blood cells are not found in peripheral blood circulation as they adhere to the capillary epithelial cells and sequester in the microvasculature including that of vital organs, such as the brain, heart and lungs (Miller, Good et al. 1994; Miller, Baruch et al. 2002; Clark and Cowden 2003).

Some of the parasites differentiate into large sexual forms with only one nucleus which fills up the infected red blood cell, known as macro- or microgametocytes and are the infective stages to mosquitoes (Bruce Chwatt 1985; Miller, Baruch et al. 2002; Moore, Surgey et al. 2002). Ingestion of gametocytes by the mosquito vector induces the emergence of gametes from within these infected red blood cells (gametogenesis) (Ghosh, Edwards et al. 2000; Baton, Ranford-Cartwright et al. 2005; Vlachou, Schlegelmilch et al. 2006). This process is triggered by the decrease in temperature and dissolved carbon dioxide, the subsequent increase in pH from 7.4 (blood) to above 7.8 (mosquito gut) and the mosquito metabolites (mosquito-derived exflagellation factor) (Ghosh, Edwards et al. 2000; Baton, Ranford-Cartwright et al. 2005; Vlachou, Schlegelmilch et al. 2006). The microgametocyte undergoes several rounds of nuclear replication. The resulting nuclei become associated with flagella that emerge from the body of the microgametocyte (Ghosh, Edwards et al. 2000; Baton, Ranford-Cartwright et al. 2005; Vlachou, Schlegelmilch et al. 2006). These microgametes seek out and fuse with mature macrogametes resulting in a zygote, which in turn develops into an ookinete within 12-24 hours. The ookinete is motile and penetrates the mosquito gut epithelial cells and develops into an oocyst in the extracellular space between the basal lamina and the epithelial cells (Ghosh, Edwards et al. 2000; Baton, Ranford-Cartwright et al. 2005; Vlachou, Schlegelmilch et al. 2006). Sporogony then occurs, which is an asexual replication of the parasite DNA and the formation of sporozoites within the oocyst. The

length of the process of sporogony is dependent on the species and temperature and usually takes 10-28 days (Ghosh, Edwards et al. 2000; Baton, Ranford-Cartwright et al. 2005; Vlachou, Schlegelmilch et al. 2006). Once mature, the oocyts ruptures releasing the sporozoites into the body cavity (hemocoel) of the mosquito. The free sporozoites are motile and migrate to the salivary glands, where they invade and transverse the salivary gland epithelial cells and come to lie within its lumen, thus completing the life cycle (Ghosh, Edwards et al. 2000; Baton, Ranford-Cartwright et al. 2005; Vlachou, Schlegelmilch et al. 2006). While the hemocoel and salivary gland sporozoites are morphologically similar, they are functionally distinct. Salivary gland sporozoites efficiently invade liver cells, but cannot re-invade the salivary glands, whereas the hemocoel sporozoites readily invade salivary glands but are inefficient at invading liver cells (Ghosh, Edwards et al. 2000; Baton, Ranford-Cartwright et al. 2005; Vlachou, Schlegelmilch et al. 2006). During a blood meal some of the sporozoites are expelled into the vertebrate host, thus reinitiating the infection of the host (Bruce Chwatt 1985; Greenwood, Fidock et al. 2008).

Figure 4.The life cycle of malaria causing *Plasmodium* parasites (Greenwood *et al*., 2008)



1.2.6 Clinical Manifestations of Malaria Infection

Malaria parasite infections result in a wide variety of clinical outcomes varying from asymptomatic to mild to severe manifestations, and even to death. Depending on the severity of these symptoms, malaria can be classified as simple/uncomplicated or severe/complicated. All symptoms of malaria are caused by the erythrocytic stages of the parasite, when erythrocytes are destroyed. This results into the liberation of parasites and erythrocyte material into circulation and a host reaction (Miller, Good et al. 1994; Miller, Baruch et al. 2002) that destroys even more red blood cells than the parasite itself does

through triggering of a strong immune turnover of the red blood cells (Phillips and Pasvol 1992; Jakeman, Saul et al. 1999; Chang and Stevenson 2004). The materials released into the blood stream include toxins, such as haemozoin pigment and glucose phosphate isomerase. These induce production of cytokines or immune-modulating agents (which include among others, tumor-necrosis factor-a (TNF-a) and interferon-g (IFN-g)) by the macrophages and other immune cells, resulting in fever, rigors, and probably the other severe pathophysiologies associated with malaria (Kwiatkowski, Cannon et al. 1989; Karunaweera, Grau et al. 1992; Miller, Baruch et al. 2002; Clark and Cowden 2003; Clark, Budd et al. 2006; Tuteja 2007). In addition to these cytokine cascades, the sequestration of infected cells in tissue blood vessels also has a role in the manifestations of malaria particularly in its severe form (Marsh and Snow 1997). Severe or complicated malaria manifestations are complex and can be grouped into severe anaemia, respiratory distress and coma (Marsh and Snow 1997; Newton, Taylor et al. 1998). In a majority of cases, these syndromes reflect underlying metabolic acidosis associated with lactic acidaemia resulting from anaemia and anoxia compounded by the immune depletion of red blood cells (Marsh, English et al. 1996; Marsh, English et al. 1996; English, Sauerwein et al. 1997). Metabolic acidosis in severe malaria infection often manifests as respiratory distress and is the best independent predictor of a fatal outcome in all ages of patients (Taylor, Borgstein et al. 1993; Crawley and Nahlen 2004; Maitland and Marsh 2004; Maitland and Newton 2005). In uncomplicated malaria infections, infected persons typically experience fever, shivering, cough, respiratory distress, pain in the joints, headache, watery diarrhoea, vomiting and convulsions (Karunaweera, Carter et al. 1992; Miller, Baruch et al. 2002). The severity of these clinical attacks is dependent on various factors that include, the species and strain of the infecting parasite, level of transmission, age of the host, genetic constitution,

nutritional status (particularly in children) and malaria specific immunity (Idro, Jenkins et al. 2005; Crawley, Chu et al. 2010). The latter tends to be short lived and partial, unable to protect an individual against a new infection, due to the variability of the parasite proteins expressed during its complex life cycle (Mendis, David et al. 1991; Gamain, Miller et al. 2001; Obi, Okangda et al. 2010).

Clinical immunity to malaria appears to be acquired in distinct stages under suitable conditions. These stages include; (i) reduction of the likelihood of severe or fatal disease (anti-disease immunity); (ii) limitation of the clinical impact of mild malaria through conferring protection against parasitaemia (anti-parasite immunity); and (iii) maintaining a low-grade parasite density, which generally results in asymptomatic infection (premonition) (Schofield and Mueller 2006; Doolan, Dobano et al. 2009; Obi, Okangda et al. 2010; Van Den Steen, Deroost et al. 2011). This occurs in regions considered holo-endemic or hyper-endemic, where populations are routinely exposed to infection (Obi, Okangda et al. 2010). So acquired immunity is correlated to age of the host. Parasitaemia therefore often peaks in young children and subsequently declines in an age-dependent manner in areas of high endemicity (Baird 1995; Hviid 2005; Doolan, Dobano et al. 2009). This change in immunity is expressed as an increase in recovery and/or a decrease in trophozoite density with age thus reducing detectability by microscopy and /or decreasing susceptibility of the host. The reduction in parasite and trophozoite densities does not occur as fast as that of the gametocyte rate indicating that immunity reduces infectivity before increasing recovery and/or detectability by microscopy (Dietz, Molineaux et al. 1974; Maire, Smith et al. 2006; Smith and Schapira 2012). Thus, in such areas immunity reduces the frequency with which infections progress to clinical or severe malaria but however does not affect the rate of parasite infection itself (Hogh 1996; Smith and Schapira 2012). In regions of infrequent

exposure, the lack of protective immunity results in the spread of cases across age groups (Baird 1995; Hogh 1996; Doolan, Dobano et al. 2009). Malaria is then classified into two epidemiological malaria transmission patterns, thus stable and unstable malaria.

Stable malaria transmission occurs when a population is continuously exposed to malaria parasite inoculation (Kiszewski, Mellinger et al. 2004) leading to the development of a protective clinical immunity from malaria early in life usually around the age of 4 to 5 years. By contrast, in unstable malaria transmission areas immunity is gained slowly due to low to moderate mean malaria inoculations. These areas tend to be prone to epidemics. (Carter and Mendis 2002). These differences in stability are primarily due to differences in behaviours and other biological characteristics of the regional *Anopheles* vectors and their environment (Lindsay, Wilkins et al. 1991; Lindsay, Parson et al. 1998; Carter and Mendis 2002; Hay, Guerra et al. 2005; Reyburn, Mbatia et al. 2005). Stable transmission occurs in regions where strong human –biting preferences lead to uniform contact between the vectors and humans (Smith, Corvalan et al. 1999). This is coupled with favourable climatic conditions that are warm and humid supporting vector longevity and rapid development of the parasite stages within them (Smith, Corvalan et al. 1999). In unstable transmission regions the opposite is true with vectors preferring animal feeding sources and the climate not being conducive to support vector longevity and rapid parasite development within them. This leads to erratic contact between the vectors and humans and thus irregular malaria inoculations (Carter and Mendis 2002).

1.2.7 Malaria Case Management

1.2.7.1 Treatment

For thousands of years, various herbal remedies have been used for treatment of fevers associated with malaria. Quinine, as a component of the bark of the Cinchona (Quina-quina)

tree, was used to treat malaria from as early as the 1600s (Achan, Talisuna et al. 2011). As a result of their missionary work in Peru, the Jesuits learnt about the curative bark and brought back the information to Europe. The bark began to be referred to as the "Jesuits' bark," "cardinal's bark," or "sacred bark" (Butler, Khan et al. 2010; Achan, Talisuna et al. 2011). Through the activities of Sir Robert Talbot, the use of the bark became established in England. Talbot mixed the bark with rose leaves, lemon juice and wine (Butler, Khan et al. 2010). In 1820, the work of Pierre Pelletier and Jean Caventou lead to the isolation of a complex alkaloid structure, which they later named Quinine. Quinine remained the main treatment drug for malaria until the 1920s. During the First World War the supply of quinine dropped, leading to a search for alternatives which could be used by troops. The Germans offered the first alternative in the form of mepacrine, which was used for both treatment and prophylaxis (Butler, Khan et al. 2010). Later German scientists developed another antimalarial called sontoquine. The chemical variants of sontoquine were synthesized and tested for efficacy. The resulting drug was chloroquine which was found to be more effective and well tolerated. By the 1940s, chloroquine took over the role as the main treatment for malaria as it was considered a safer and cheaper option than quinine (Bruce Chwatt 1985; Achan, Talisuna et al. 2011). The efficacy of chloroquine begun to decrease and parasite resistance to the drug begun to spread by the early 1970s (Talisuna, Bloland et al. 2004). This was attributed to its wide and somewhat indiscriminate use particularly during the malaria eradication program in the 1950s and 1960s (Talisuna, Bloland et al. 2004). The American military also developed drugs with antimalarial properties which included primaquine, which was used for treatment of *vivax* malaria and much later tafenoquine (Greenwood 1995). During the same period, the British military developed proguanil which saved as a foundation for further development into pyrimethamine by

Burroughs Wellcome (Greenwood 1995). Sulphadoxine which was developed in the 1950s, had a long half life and improved toxicological profile compared to other sulphonamides (Schlitzer 2007). By the 1970s, pyrimethamine was being combined with sulphadoxine, resulting in a drug given the brand name fansidar and was widely used in Africa for treatment of uncomplicated malaria and prophylaxis (Meshnick and Dobson 2001; Schlitzer 2007). This was the next alternative to chloroquine in a large number of endemic countries, however the development of resistance rapidly hampered its efficacy (Black, Bygbjerg et al. 1981; Hurwitz, Johnson et al. 1981; Stahel, Degremont et al. 1982; Timmermanns, Hess et al. 1982; Hess, Timmermans et al. 1983; Sibley, Hyde et al. 2001).

The search for an alternative drug lead the world to China, where dating back over 2000 years ago, records showed the use of ginghaosu (Artemisia annua) for the treatment of fevers, which are believed to have been associated with malaria (Wright, Linley et al. 2010). A team lead by a Chinese scientist Professor Youyou Tu has been accredited with the discovery of artemisinin which is now recommended to be used in combination with other products as the mainstay of treatment of malaria (Miller and Su 2011).

Chemotherapy still remains a keystone of the full complement of malaria control activities. The primary effect of antimalarial treatment is to inhibit the multiplication of the parasite. Based on their chemical structure there are three main groups of antimalarials; i) the aryl amino-alcohol compounds (quinoline related i.e. amodiaquine, chloroquine, halofantrine, lumefantrine, mefloquine, piperaquine, pyronaridine, primaquine, quinine, quinidine, tafenoquine), ii) the antifols (chlorproguanil, proguanil, pyrimethamine, trimethoprim), and iii) the artemisinin compounds (artemisinin, artemether, artmotil, artesunate, dihydroartemisinin).

Of these, the artemisinin compounds produce the most rapid therapeutic responds due to their broad window of action on the asexual and sexual stages of the malaria parasite (Yayon, Vande Waa et al. 1983; ter Kuile, White et al. 1993; Drakeley, Jawara et al. 2004; Sutherland, Ord et al. 2005; Stepniewska, Price et al. 2008). They predominantly act on early trophozoites and schizonts before they can mature, therefore reducing infected erythrocytes sequestration in the micro-vessels of vital organs, thus preventing obstruction and subsequent associated complications (Schlitzer 2008; Dondorp, Fanello et al. 2010) There has been no anti malarial therapy that targets the brief extracellular merozoite stage of the parasite (Wilson, Langer et al. 2013). Anti malarial drugs can be further classified based on their mode of action which essentially targets the various stages of the parasite life cycle (Bruce-Chwatt 1962; Delves, Plouffe et al. 2012) as outlined in the table 4 below.

Table 4. Classification of antimalarial drugs based on mode of action (adapted from Bruce-Chwatt 1962, Plouffe *et al*., 2012)

Classification	Mode of action	Drugs
Blood Schizontocides	Anti-plasmodial activity is on the erythrocytic stages, thus preventing expansion of the infections	Artemisinin and its derivatives, chloroquine, quinine, mefloquine, halofantrine, pyrimethamine and sulphadoxine
Prophylactic Tissue Schizontocides	Act against the liver forms of Plasmodium occurring prior to the erythrocytic stage. Thus they prevent the onset and development thereafter of clinical symptoms	Primaquine and to some extent pyrimethamine
Anti-relapse drugs or secondary tissue schizontocides	Act by targeting the hypnozoites /dormozoites (dormant stages) of P vivax and P. ovale within host liver cells that lead to relapse and regeneration of disease	Primaquine
Gametocytocides	Target the sexual forms of the parasite in the blood thereby preventing transmission of the infection to the mosquito	Chloroquine and quinine against *P.vivax* and *P. malariae* ; artemisinin and its derivatives has activity against *P. falciparum*; primaquine against all *Plasmodia*, including *P. falciparum*
Sporontocides	Target and cease growth and/or development of the oocysts within the mosquito there by preventing transmission. Delivered through a mosquito blood meal	Primaquine, Ivermectin and chloroguanide

Due to the wide use of anti malarial therapies over time there has been great selective pressure on *P. falciparum* and leading to the spread of resistant parasites with the subsequent result of increased malaria morbidity and mortality (Bloland 2001; Barnes and White 2005; Mita, Tanabe et al. 2009; Khamsiriwatchara, Sudathip et al. 2012; Satimai, Sudathip et al. 2012) . To date resistance to all the commonly used antimalarials, including the most recent, artemisinin combination therapies has been reported (Delhaes, Benoit-Vical et al. 2003; Mita, Tanabe et al. 2009). Chloroquine (CQ) and sulphadoxine/pyrimethamine (SP) have been used for over 50 years for the treatment of uncomplicated *P. falciparum* malaria (Talisuna, Bloland et al. 2004; Mita, Tanabe et al. 2009). Most countries in endemic areas revised their treatment guidelines in the early 2000s to replace these two drugs with the more efficacious artemisinin combination therapies as per WHO recommendation. Quinine still remains the recommended treatment for severe malaria. However, there have been recommendations for the use of parenteral artesunate as an alternative to quinine for treatment of severe malaria (Dondorp, Nosten et al. 2005; Toovey 2006; Woodrow, Planche et al. 2006; Jones, Donegan et al. 2007; Dondorp, Fanello et al. 2010; Sinclair, Donegan et al. 2011; Sinclair, Donegan et al. 2012) due to its demonstrated quick reduction of mortality (Dondorp, Nosten et al. 2005; Dondorp, Fanello et al. 2010; Sinclair, Donegan et al. 2011; Sinclair, Donegan et al. 2012) and tolerability (Dondorp, Nosten et al. 2005). Additionally, the current recommendations promote the use of parasite-based diagnosis prior to prescription of antimalarial therapy.

1.2.7.2 Diagnosis

Several methods are used for malaria confirmatory diagnosis, as summarised in Table 5 (Tangpukdee, Duangdee et al. 2009). However, the main methods used for routine parasitological diagnosis are microscopy and rapid diagnostic tests (RDT). Microscopy is

considered as a sensitive method and recognised as the historically established gold standard for malaria diagnosis (Murray, Gasser et al. 2008), but it requires an appreciable time of expertise and labour and is dependent on good techniques, reagents, equipment and well-trained microscopists (WHO 1999). Some studies have shown that it has been a challenge to maintain accuracy of microscopy particularly in hard to reach areas that are usually poorly resourced (Durrheim, Becker et al. 1997; Kachur, Nicolas et al. 1998; Barat, Chipipa et al. 1999; Bell, Wongsrichanalai et al. 2006; Reyburn, Mbakilwa et al. 2007; Ngasala, Mubi et al. 2008; Batwala, Magnussen et al. 2010; Kahama-Maro, D'Acremont et al. 2011).

Table 5. Summary of malaria diagnostic tools and modalities (Adapted from Tangapukdee *et al*., 2009)

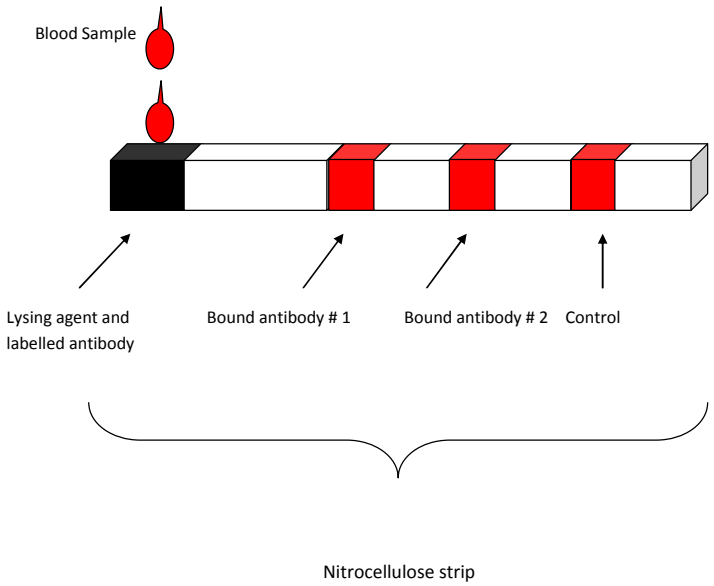
	Principal of method	Sensitivity and specificity	Time consumed (in mins)	Detection limit (parasites /µl)	Expertise require	Instrument cost
Clinical diagnosis	Malarial signs and symptoms	Dependent on malarial endemicity	Dependent on physicians skills	Undermined	High in non-endemic areas	n/a
Microscopy	Visualization of distinct morphological parasite stages under light microscopy using think & thin smears	Depends of good technique, reagent and microscopist skills	30-60	Expert 5-10, routinely 50	High in non-endemic areas	Low cost
QBC	Detection using epi-flourescent microscope detection after staining blood with acridine	Higher than microscopy	Less than 15	More than 5	Moderate	Moderate
RDTs	Detection of parasite antigens or antibodies	Moderate if more than 100 parasites / µl	10-15	50-100	Low	Moderate
Serological tests	Detection of antibodies against parasites	High but does not correlate to clinical symptoms of patients	30-60	Undermined	Moderate	Moderate
PCR	Amplification of parasite DNA	Very high	45-360, dependent on methods utilized	Greater or equal to 1	High	Expensive
LAMP	Detection of using turbidity test after parasite DNA amplification	Very high	Less than 60	More than 5	High	Moderate

Micro assays	Hybridization of DNA isolate and quantified by fluorescence based detection	High	Less than 60	Undetermined	High	Expensive
FCM	Detection of haemozoin by flow cytometer	Variable sensitivity, high specificity	Less 1 per sample / automated	Poor correlation with parasitaemia	High	Expensive
ACC	Detection of malarial pigment in activated monocyte	Variable sensitivity and specificity	Less 1 per sample / automated	5-20	High	Expensive
MS	Identification of heme by LDMS	Undetermined	Less 1 per sample / automated	100 for whole blood	High	Expensive

QBC – Quantitative buffy coat , RDTs - Rapid diagnostic tests, PCR – Polymerase chain reaction, LAMP - Loop mediated isothermal amplification, FCM - Flow cytometry, ACC - Automated blood cell counters, MS - Mass spectrophotometry

RDTs are immuno-chromatographic tests based on the detection of parasite antigen from peripheral blood using monoclonal antibodies against a parasite antigen target (Forney, Magill et al. 2001).They employ a lateral flow technology in which the liquid clinical sample, in this case blood, migrates using capillary action across a nitrocellulose membrane surface. On one end of the nitrocellulose surface, one or two target specific indicator labelled antigens (Table 5) are placed (Shiff, Premji et al. 1993; Piper, Lebras et al. 1999; Moody 2002; Murray, Gasser et al. 2008; Abba, Deeks et al. 2011). A control line is created by placing a specific antibody for the indicator labelled antibody complex. To perform the test, a blood sample mixed with a buffer solution is placed on one end of the nitrocellulose surface which contains a lysing agent and labelled antibody. This sample is drawn up the strip and if the antigen is present the test and control lines will trap some of the indicator-labelled antibody-antigen complexes and become visible (Figure 5) (Murray, Gasser et al. 2008).

Figure 5.Rapid diagnostic test schematic (Adapted from Murray *et al*., 2008)



There are currently 3 malaria antigens targeted by the available RDTs due to their abundance in all asexual and sexual stages of the parasite life cycle (Moody 2002). These are described in table 6.

Table 6.Parasite antigens targeted by RDTs (Schiff, Premji *et al*.,1993;Piper,Lebras *et al*., 1999; Moody 2002; Abba,Deeks *et al*., 2011)

Antigen	Description
histidine-rich protein II (HRP-II)	produced by trophozoites and young gametocytes of *P. falciparum* only and expressed in the erythrocyte membrane
Lactate dehydrogenease (pLDH)	Intracellular metabolic enzyme produced by all human infecting *Plasmodium* species and by all blood stage parasites (i.e. both asexual and sexual). Can distinguish *P. falciparum* from the non-*falciparum* species, but cannot differentiate between *P. vivax, P. ovale* and *P. malariae*.
Aldolase	Intracellular metabolic enzyme produced by all human infecting *Plasmodium* species however cant not differentiate between them

The most commonly targeted of the parasite antigens for RDTs are HRP-II and pLDH. HRP-II antigenaemia persistence after malaria treatment leads to false RDT positive results limits the use of this antigen for the clinical monitoring of the therapeutic response monitoring (Humar, Ohrt et al. 1997; Wongsrichanalai 2001; Gerstl, Dunkley et al. 2010). Further, HRP-II can only detect infections of *P. falciparum* (Gerstl, Dunkley et al. 2010). Assays based on pLDH only represent viable parasites and as such overcome this challenge (Moody 2002). However, production of pLDH from the gametocytes after clearance of asexual stages may lead to some tests giving falsely positive results for several days (Gerstl, Dunkley et al. 2010). pLDH assays parasite detection is closely related to malaria microscopy diagnosis as it is more specific but is however not as sensitive as HRP-II tests (Abba, Deeks et al. 2011). Thus RDTs provide a

qualitative result of infection as they do not information such as parasite stages and parasitaemic load (Marx, Pewsner et al. 2005; Hendriksen, Mtove et al. 2011). Studies have shown that RDTs are able to detect *P. falciparum* with sensitivities greater than an estimated 85% and with specificity of approximately 100% at parasitaemia greater than 100 parasites/µl of blood (Hanscheid 1999; Forney, Magill et al. 2001; Wongsrichanalai 2001; Moody 2002; Murray, Bell et al. 2003; Abba, Deeks et al. 2011). Some studies have demonstrated that RDTs have a higher sensitivity and specificity compared with microscopy (Beadle, Long et al. 1994; Craig and Sharp 1997; Makler, Palmer et al. 1998; Mendiratta, Bhutada et al. 2006; Ashley, Touabi et al. 2009). An advantage RDTs have over microscopy is that they do not require complex training, electricity, running water or any special equipment. Additionally, it has been demonstrated that RDTs can be utilized by community health workers with minimal supervision and ease (Bell, Go et al. 2001; Harvey, Jennings et al. 2008; Chanda, Hamainza et al. 2011; Counihan, Harvey et al. 2012; Rutta, Francis et al. 2012). In addition to those alluded to earlier, RDTs have some other limitations that include the lack of ability to detect all the species of *Plasmodium* and low concentrations of infections (Reyburn, Mbakilwa et al. 2007).The usefulness of RDTs in malaria case management has been demonstrated to reduce misdiagnosis and enhance cost savings through rational use of malaria treatments (Chanda, Hamainza et al. 2011; Yukich, Bennett et al. 2012).

Clinical diagnosis still remains the most wide spread method of malaria diagnosis particularly in the most peripheral health facilities in Africa. This has lead to an over estimation of malaria burden and over prescription of anti malarial drugs (Mkawagile and Kihamia 1986; Bassett, Taylor et al. 1991; O'Dempsey, McArdle et al. 1993; Font, Alonso Gonzalez et al. 2001; Nsimba,

Massele et al. 2002; Masanja, Selemani et al. 2012) due to the non – specific signs and symptoms that can be similar to those of many other febrile conditions (Redd, Bloland et al. 1992; Richens, Smith et al. 1992; O'Dempsey, McArdle et al. 1993; Berkley, Mwarumba et al. 1999; Kallander, Nsungwa-Sabiiti et al. 2004). There have been several efforts to improve clinical diagnosis of malaria based on signs and symptoms alone, leading to development of various clinical algorithms across all age groups and varying malaria endemicity (Gomes, Espino et al. 1994; Marsh, English et al. 1996; Olaleye, Williams et al. 1998; Bojang, Obaro et al. 2000; Oster, Krause et al. 2000; Chandramohan, Jaffar et al. 2002; Mwangi, Mohammed et al. 2005). However, remains unsatisfactory and reinforces the need for parasitological confirmation of infection to ensure accuracy of malaria diagnosis based on clinical symptoms alone (Mwangi, Mohammed et al. 2005).

1.2.8 The Vector and its distribution

Mosquitoes, family *Culicidae*, are made up of a taxon descended from a common evolutionary predecessor or ancestor (monophyletic) (Harbach and Kitching 1998) belonging to the order *Diptera*, Class *Insecta*, phylum *Arthropoda* (Darsie and Ward 2005). Culicidae is divided into the three subfamilies Toxorhynchitinae, Anopheline, and Culicine, collectively comprising 38 genera, 1 in subfamily Toxorhynchitinae,3 in Anopheline and 34 in Culicine (Foster and Walker 2002). There are some 3,490 species currently formally recognized (Harbach and Howard 2007), but some 3–5 times this number may exist if most of the known species, like many well studied *Anopheles,* prove to comprise species complexes morphologically (isomorphic) identical (Foster and Walker 2002; Fontenille and Simard 2004; Harbach 2011). All human malaria vectors belong to the genera *Anopheles* (Cellia) Myzomyia. These have been distributed globally in six

geographical regions, which include Palaearctic, Oriental, Australasian, Afro-tropical, Neo-arctic and Neo-tropical regions (Bruce Chwatt 1985). Of the 460 *Anopheles* mosquito species identified globally, only approximately 80 species are physiologically competent hosts for human malaria parasites , 70 species are known to act as vectors in natural conditions and 45 are dominant vector species/species complexes that are able to transmit malaria at a level of concern to public health (secondary vectors) (Foster and Walker 2002). In Africa there are *Anopheles* species that are considered the major vectors for malaria transmission are; *Anopheles gambiae sensu stricto* Giles*, Anopheles arabiensis* Patton and *Anopheles funestus* Giles (Gillies and Meillon 1968; White 1974; Gillies and Coetzee 1987; Levine, Peterson et al. 2004; Mouatcho, Hargreaves et al. 2007). *Anopheles moucheti, An. (Cel.) melas, An. (Cel) merus* and *An.(Cel) nili* are also considered locally important primary vectors in some regions of Africa with the former two occurring mostly in coastal areas (Awono-Ambene, Kengne et al. 2004; Fontenille and Simard 2004; Fontenille and Carnevale 2006; Sinka, Bangs et al. 2010). *Anopheles gambiae* and *An. arabiensis* belong to the *Anopheles gambiae* complex of 7 sibling species (Hunt, Coetzee et al. 1998). *An. gambiae* is considered the most widespread throughout sub-Saharan Africa and the world's leading vector for *P. falciparum* because of its longevity, preference for blood meal from human hosts (anthropophagic) and indoor resting behaviour (endophilic), resulting in it having great efficiency in transmission (Gillies 1955; Kiszewski, Mellinger et al. 2004).Through often found in the same geographical range with *An. gambiae*, *An. arabiensis* is able to tolerate higher temperatures and humidity (Kirby and Lindsay 2004), is widely distributed in arid areas (Coetzee, Craig et al. 2000) and tends to prefer to rest outside in

the open (exophilic) and blood meals from both animals (zoophagic) particularly cattle and humans (Gillies 1955).

The other members of the complex include *An. melas* (West Africa)*, An. merus* (East Africa), *An. bwambae* (Hot springs of Uganda)*, An. quadriannulatus* species A and B (White 1974; Gillies and Coetzee 1987; Fettene, Hunt et al. 2004; Parmakelis, Slotman et al. 2008; Parmakelis, Moustaka et al. 2010). All members of the complex are probable vectors of human malaria except *An. quadriannulatus* subspecies A and B which is primarily zoophilic or zoophagic (White 1974; Coetzee, Craig et al. 2000; Prior and Torr 2002; Coetzee 2004; Pates, Takken et al. 2006).

Anopheles funestus Giles is a significant malaria vector which is very focally distributed in many locations all across sub Saharan Africa and plays a more important role in malaria transmission than *An. gambiae* or *An. arabiensis* (Fontenille, Lochouarn et al. 1997; Cohuet, Dia et al. 2004). The *Anopheles funestus* group has nine members which are divided into two African subgroups i.e. the *funestus* subgroup (*Anopheles aruni* Sobti, *Anopheles confusus* Evans and Leeson, *Anopheles funestus sense strico* Giles, *Anopheles parensis* Gillies, *Anopheles vaneedeni* Gillies and Coetzee) and the *rivulorum* subgroup (*Anopheles brucei* Service*, Anopheles fuscivenosus* Leeson*, Anopheles leesoni* Evans, *Anopheles rivulorum* Leeson*,* and an "*An. rivulorum*-like" species)(Gillies and Coetzee 1987; Hargreaves, Koekemoer et al. 2000; Brooke, Kloke et al. 2001; Koekemoer, Kamau et al. 2002; Spillings, Brooke et al. 2009). From within this group there are two species implicated in malaria transmission, *An. funestus* (Mostly found in southern Africa) and *An. rivulorum,* a secondary vector in Tanzania (Wilkes, Matola et al. 1996; Coetzee, Craig et al. 2000). *An. funestus* prefer feeding indoors (endophagic) and are both

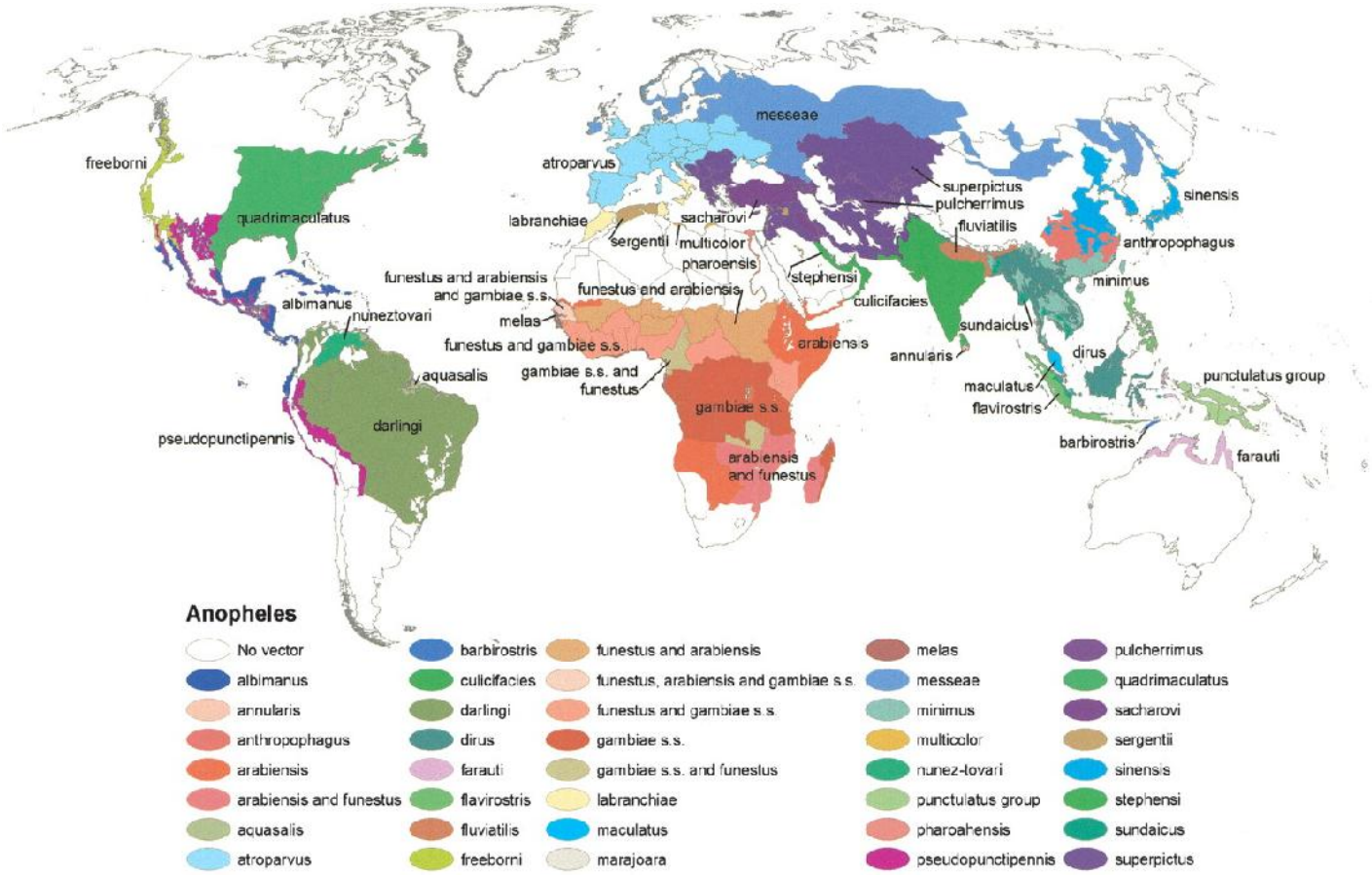
endophilic and anthropophilic. The other members of the group are typically limited in distribution and abundance and prefer feeding upon animals to humans (Bruce-Chwatt 1954; Hackett, Gimnig et al. 2000). However they have been known to act as secondary vectors in circumstances of scarcity of the preferred hosts (Gillies and Meillon 1968). *An. rivulorum* is primarily zoophilic and is the second most abundant and widespread species in the funestus group (Wilkes, Matola et al. 1996; Hackett, Gimnig et al. 2000).

Where both *An. gambiae* and *An. funestus* occur there are high levels of transmission (Coetzee, Craig et al. 2000; Mbogo, Mwangangi et al. 2003; Coetzee and Fontenille 2004; Hay, Guerra et al. 2005), primarily due to their different breeding habitats and preferences which peak at different times of the year (Mosha and Petrarca 1983; Bogh, Pedersen et al. 1998; Mwangangi, Mbogo et al. 2003; Mwangangi, Mbogo et al. 2004; Mwangangi, Mbogo et al. 2007). *An. gambiae* are most abundant during the rainy season (Coetzee, Craig et al. 2000; Coetzee and Fontenille 2004), while *An. funestus* is most abundant when the rainy season subsides and the dry season initiates, and thus has the potential to elongate the malaria transmission period where both species occur (Coetzee, Craig et al. 2000; Coetzee and Fontenille 2004). *An. nili* and *An. moucheti* are the major vectors of malaria in West and Central Africa (Awono-Ambene, Kengne et al. 2004; Fontenille and Simard 2004; Fontenille and Carnevale 2006).

The distribution of all malaria vectors is governed greatly by environmental and climatic factors, which influence their abundance and survival (Figure 6) (Lindsay, Parson et al. 1998; Smith, Corvalan et al. 1999; Kiszewski, Mellinger et al. 2004; Kaga and Ohta 2012) and ultimately impact malaria transmission intensity (Kiszewski, Mellinger et al. 2004; Kaga and Ohta 2012).

Many of the vector species occur in sympatry and their importance to malaria transmission is governed by their biting, feeding and resting preferences, in addition to seasonal prevalence and vectorial capacity. These differences are responsible for the varied malaria epidemiological patterns across Africa and result in a need for varied vector control approaches (Coluzzi 1984; Fontenille and Simard 2004).

Figure 6. Global distribution of dominant or potentially important malaria vectors (adapted from Kiszewski *et al*., 2004)



1.2.9 Early efforts to control the causes of malaria

1.2.9.1 Larval source management and therapeutic approaches

The observations by Laveran and Ross shaped the beginning of modern preventive strategies to control malaria (Bruce-Chwatt 1977; Guillemin 2002). Ross and others postulated that studies on malaria and its eventual control, should be centred on what he referred to as the third and last phase, prevention, through the extermination of the malaria bearing mosquitoes primarily by locating their breeding sites and applying measures to suppress their development (Jeffery 1976; Bruce-Chwatt 1977; Najera 2000). This source reduction leads to decrease in vector density to a level where vectorial capacity and subsequently, transmission is reduced significantly (Coosemans and Carnevale 1995). There are basically two main techniques in larval source management (LSM) and these include environmental management (temporary or permanent removal of *Anopheline* larval habitats) and larviciding with chemical or biological agents (Walker and Lynch 2007). The focus on aquatic stages was primarily due to the fact that these stages are less mobile compared to the adult ones and control measures could be instituted where breeding sites where identified (Killeen, Fillinger et al. 2002). This approach in certain areas showed some good results, particularly in Europe (Najera 2000). However these initial strategies evolved over time and prompted Ross in 1911 to propose an integrated approach to malaria eradication including personal protection, mosquito reduction, and treatment (Ross 1911). Ross further stated that it would be challenging for any general community to apply these measures in totality and noted that each could be useful alone under certain circumstances (Ross 1911). Confidence in this strategy was reinforced by the development of effective chemotherapeutic and chemoprophylactic agents and, the promise of

vector control through then modern insecticides (Beausoleil 1984). One of the most outstanding demonstrations of anti larval complimented with other interventions was in Havana, Cuba during the construction of the Panama Canal in 1904 to 1914. These efforts where overseen by William Gorgas and was an integrated program that was strictly enforced and included; drainage of pools and brush and grass cutting (rationale was that mosquitoes could not cross open areas more than 100 yards) in the environs surrounding residential areas; oiling of ponds and swamps to kill larvae ; larviciding using a mixture of carbolic acid, resin and caustic soda was used when oiling was considered insufficient; prophylaxis with quinine ; screening of buildings and; killing adult mosquitoes (Gorgas 1910; Marshall 1913). These interventions lead to the eradication of yellow fever and a dramatic decline in malaria morbidity and mortality. Similar large scale control efforts were conducted during the first world war which involved draining (pontine marshes in Italy), larval control using paris-green dust and oil (Brazil and Egypt) and spraying of houses with pyrethrum (Natal and Zululand) (Rieckmann 2006). The cost of these interventions was beyond what could be afforded by most communities in these malarious regions. The larval stage targeted interventions required locating breeding sites and targeting them with the appropriate interventions on a regular and indefinite time frame, which proved to be a challenge in most malarious areas (Killeen, Fillinger et al. 2002).

1.2.9.2 The Global Malaria Eradication Program using indoor residual spraying and drug therapy

The first global effort to eradicate malaria was initiated in the 1950s and 1960s, with the World Health Organization (WHO) spearheading the effort which was called the Global Malaria

Eradication Program (GMEP). Technically the program departed from the earlier larval control strategies to those providing for (a) an establishment of pilot projects in which methods were tested for their local suitability, (b) an attack phase where of dichloro-diphenyl trichloroethane (DDT) or other comparable long lasting residual insecticides were used for indoor residual spraying (IRS) and the resultant insecticidal coverage maintained for several years, (c) a consolidation phase during which insecticide spraying would be discontinued and any remaining infections would be actively sought out by screening whole populations for infection and treating all positive cases, and lastly (d) establishment of eradication, followed by strengthened surveillance until the risk of reinvasion disappeared (Macdonald 1965). The strategy essentially depended on chloroquine for treatment and prevention, DDT for vector control targeting adult mosquitoes and surveillance (Garrett-Jones 1964). Thus the targeting of adult mosquitoes and the knowledge of resting and feeding behaviours lead to the adoption of IRS as the key vector control intervention to suppress vector capacity by reducing their longevity and potentially disrupting the sporogonic stage of the parasite (Garrett-Jones 1964; Garrett-Jones and Shidrawi 1969; Mabaso, Sharp et al. 2004). IRS is defined as the application of long-acting chemical insecticides on the walls and roofs of all houses and domestic animal shelters in a given area, in order to kill the adult vector mosquitoes that land and rest on these surfaces (WHO 2006). The resulting application of IRS is thought to both repel mosquitoes from entering the house as well as kill fed female mosquitoes that rest on the walls of dwellings (Pluess, Tanser et al. 2010). However, there was an oversight in the strategy of targeting adult mosquitoes as it did not take into consideration the mobility of this stage and variability in behaviour between vectors (Killeen, Fillinger et al. 2002).

The GMEP, however, excluded most of sub-Saharan Africa due to the lack of infrastructure to implement the program, wars, post independence destabilization and the fact that the intense transmission in this region was likely to defeat eradication as a goal (Bruce-Chwatt 1987; Trigg and Kondrachine 1998). However by 1978, the program had successfully eliminated malaria in 37 (27 in the Americas or Europe) of the targeted 143 endemic countries (Bruce Chwatt 1985; WHO 2008). The success of the program in these areas which were mostly temperate, subtropical and had unstable transmission, engendered undue optimism that malaria elimination could be achieved anywhere. This stifled any additional thoughts on further research to improve anti malarial activities for almost 4 decades (Alilio, Bygbjerg et al. 2004). The initial goals of malaria eradication where not achieved as in some areas there was a resurgence of malaria to previous or higher levels in areas where it was thought malaria had been eliminated (Bruce-Chwatt 1987; Feachem and Sabot 2008). With the development of vector resistance to DDT, vector behavioural pre-existing resilience to the spraying programs, the emergence of resistance to the available anti malarial drugs, subsequent economic challenges and the realization that a time limited eradication campaign was not feasible in all areas, the elimination effort was abandoned by 1969 (Wright, Fritz et al. 1972; WHO 1974; Bruce-Chwatt 1987; Roberts and Andre 1994; Litsios 1996; Trigg and Kondrachine 1998; Feachem and Sabot 2008). This lead to a re-examination of the program focus to one of sustained control as opposed to that of eradication (Wright, Fritz et al. 1972; Bruce-Chwatt 1987; Trigg and Kondrachine 1998; WHO 2008).

1.2.9.3 Roll Back Malaria the role of insecticide treated bed nets, indoor residual spraying and drug therapy

The post global eradication campaign era, in the 1970s and '80s, saw the weakening of malaria programs due to challenges in integration into the mainstream health sector without a corresponding increase in resources (Nabarro and Tayler 1998). The growing increase in parasite and vector resistance to anti malarial drugs and insecticides respectively, coupled with the development of conflicts and humanitarian crisis in some endemic countries further exacerbated the problem. This period also saw a continued increase in the burden of malaria particularly in sub-Saharan Africa (Alilio, Bygbjerg et al. 2004). With increased knowledge of the transnational nature of malaria, Ministers of Health from malarious nations lead the process to develop the Global Malaria Control Strategy (GMCS) in 1992, which called for less focus on vector control and emphasis on case detection and treatment. This was a result of a realization that primary health systems had failed to deliver positive results and that new efforts were required to control malaria, particularly in Africa which was most affected (Alilio, Bygbjerg et al. 2004). The strategy further called for full integration of malaria control activities into general health services with reflection on the various socioeconomic development objectives (Roberts, Laughlin et al. 1997). There were various meetings and declarations that followed all calling for an accelerated implementation of malaria control efforts (Alilio, Bygbjerg et al. 2004). This provided a framework for the eventual creation of the roll back malaria (RBM) partnership to coordinate global efforts in combating malaria, initially, particularly in the deployment of insecticide treated bed nets (ITN) as a key strategy in proving child survival (Nabarro and Tayler 1998; Alilio, Bygbjerg et al. 2004; Breman, Alilio et al. 2004; Lengeler 2004). An ITN is a

mosquito net that repels disables and/or kills mosquitoes coming into contact with insecticide on the netting material. The ITN also provides a physical barrier thus preventing vector – human contact (Lines, Myamba et al. 1987; Lindsay, Wilkins et al. 1991; WHO 2007). The practice of applying insecticides to fabrics for personal protection against vector borne diseases began in the Second World War by German, Soviet and US military personnel. In the late 1970s, pyrethroids were the insecticide of choice for this, due to the low toxicity to humans and their high insecticidal activity. In the 1980s, studies were instituted to assess the use of this technology for disease control in the form of bed nets (Lengeler and Snow 1996). The rationale of ITNs was based on the understanding of vector behaviour of indoor biting late at night, the provision of both a physical barrier preventing man-vector contact and repellent/killing insecticide effect, thus impacting the vector feeding success, vector capacity and human –biting rates (Bermejo and Veeken 1992; Curtis and Mnzava 2000; Lengeler 2004). Various ITN trials conducted in Gambia, Burkina Faso, Kenya and Ghana demonstrated positive impact of ITNs on reducing morbidity and mortality in children (Alonso, Lindsay et al. 1991; Picard, Aikins et al. 1993; Binka, Kubaje et al. 1996; Cham, Olaleye et al. 1997; D'Alessandro, Olaleye et al. 1997; Binka, Indome et al. 1998; Phillips-Howard, Nahlen et al. 2003). These findings contributed to the reinvigoration of malaria control efforts through increased funding, scale up of both effective preventive and curative interventions with particular prioritisation of malaria prevention in pregnancy, and focus on under five children and vector control efforts. Much later in the early 2000s, the development of more efficacious treatments in the form of artemisinin combination therapies further strengthened the resolve that malaria could be controlled and eventually eradicated (White 2008).

Since the launch of the RBM, the global goals and approaches for malaria control have been modified to address expected positive impacts on disease transmission and respond quickly to any unexpected situations arising from social, economic, political or environmental trends among others in endemic areas (Nabarro and Tayler 1998; WHO 2005; WHO 2009). The revised focus is to expand coverage of cost effective key intervetions such as indoor residual spraying, insecticide treated bed nets, intermittent presumptive therapy and case management using artemisinin combination therapies (WHO 2005; WHO 2009; Eisele, Larsen et al. 2012). As a result of increased global resources (US$ 149million in 2000 to US$1.2 billion in 2008), coupled with renewed attention by governments towards malaria, these scale up efforts and high coverage have begun to show dividends, resulting into notable declines in malaria disease burden globally (WHO 2010; Eisele, Larsen et al. 2012; Murray, Rosenfeld et al. 2012); however more investments are required to achieve the goal of malaria eradication (WHO 2010; Murray, Rosenfeld et al. 2012).

1.2.10 Modern Era of Malaria Control Interventions

According to the World Health Organization, there are two main priorities of a malaria elimination programme which include: (i) to identify and treat malaria patients and all people carrying parasites, including those carrying gametocytes, to ensure that they become non-infectious as early as possible; and (ii) to sustainably reduce human–vector contact and the vectorial capacity of the local Anopheles mosquito population, to prevent new infections from occurring (WHO 2005; WHO 2006; WHO 2007; WHO 2009; WHO 2012; WHO 2012). These priority areas are therefore very broad based on and encompass provision of curative and

preventive interventions to all at risk populations. Essentially these interventions would include appropriate confirmatory diagnosis, prompt and effective treatment, provision of prophylaxis where appropriate and universal coverage of vector control measures (Bhattarai, Ali et al. 2007; Okiro, Hay et al. 2007; Ceesay, Casals-Pascual et al. 2008; Noor, Kirui et al. 2009; D'Acremont, Lengeler et al. 2010).

1.2.10.1 Contemporary Vector Control

Vector control methods are defined as measures of any kind directed against a vector of disease and intended to limit its ability to transmit the disease (Karunamoorthi 2011). Insecticide treated bed nets and indoor residual spraying are currently considered the most effective of these currently available (Okumu and Moore 2011). The universal coverage of these two interventions is considered as the key preventive strategy under the global malaria control and elimination plan (WHO 2005; WHO 2006; WHO 2009). Both IRS and ITNs primarily target adult stages of the mosquito thus reducing overall transmission and providing a community protective effect by reducing abundance and infectivity of malaria vectors (Lengeler 2004; Killeen, Smith et al. 2007; Okumu and Moore 2011).

They exploit behaviours of vector mosquitoes and have shown to be very effective in controlling *An. gambiae s.s* and *An. funestus* than *An. arabiensis* primarily due to its resting and feeding preferences (Gillies 1955; Mosha and Petrarca 1983; Bradley 1991; Lines and Nassor 1991; Bogh, Pedersen et al. 1998; Lengeler and Sharp 2003; Kiszewski, Mellinger et al. 2004). During the Pare-Taveta scheme, it was demonstrated that there was a dramatic decline of *An. gambiae s.s* and *An. funestus* populations in the area after implementation of IRS using dieldrin

(Bradley 1991). *An.arabiensis* tends to be more adaptive to interventions that focus particularly on residual insecticides and tends to alter its behaviour (Sharp, Le Sueur et al. 1990; Fettene, Hunt et al. 2004; Geissbuhler, Chaki et al. 2007) with variations in its host, feeding and resting preferences, replacing *An. gambiae s.s*. in some settings and becoming the sole malaria vector (Lindblade, Gimnig et al. 2006).

ITNs have been shown to reduce malaria morbidity and all causes of malaria mortality across a variety of transmission profiles (D'Alessandro, Olaleye et al. 1995; Binka, Indome et al. 1998; Hawley, Phillips-Howard et al. 2003; Lengeler 2004; Lengeler 2004; Killeen, Smith et al. 2007; O'Meara, Bejon et al. 2008; Kleinschmidt, Schwabe et al. 2009). IRS has similar impact on malaria mortality and morbidity (Payne, Grab et al. 1976; Curtis 1999; Misra, Webber et al. 1999; Curtis and Mnzava 2000; Bhatia, Fox-Rushby et al. 2004). IRS was initially mainly implemented in areas with seasonal transmission and epidemic prone, principally due to the logistical and resource aspects of conducting a spraying campaign, however with the development of longer lasting insecticides this has changed to allow for spraying in areas of perennial transmission (WHOPES 2007). Pyrethroids are the only permitted insecticide group for treating bed nets because of their low mammalian toxicity, a quick knock down rate and repellence effect. However, most modern insecticide formulations have very little repellency effect making the physical barrier the key deterrent. Repellency may have a negative effect on the community wide effect that results from high coverage of ITNs and IRS, as mosquitoes are not killed thus introducing the possibility of them diverting to non-protected communities (Killeen and Moore 2012). Thus, non repellent insecticides will not deter mosquitoes and will most likely only kill them after they have fed. In order to achieve high coverage, this implies

establishing aggressive campaigns to emphasize the nature of communal protection where such insecticides are utilized (Killeen, Chitnis et al. 2011). Insecticides used for IRS include organochlorines, pyrethroids, carbamates and organophosphates (Pemba and Namangale 2009). These four classes of insecticides are the most important in public health (WHO 1997). The effect of these insecticides is dependent on their intrinsic toxicity to the mosquito and residual effect which is dependent on the formulation used, stability, volatility and nature of surface sprayed (Najera and Zaim 2001). Due to selection pressure since the introduction of these insecticides, more than a hundred species have developed resistance to one or more of these insecticides (Hemingway and Ranson 2000; Najera and Zaim 2001; Coleman, Sharp et al. 2006). This poses a great challenge to malaria control as the essential characteristic of selection of an insecticide is the susceptibility of the vectors in the area of concern. This is compounded by the choice of insecticides suitable for IRS (dependent on local resistance profile) and the fact that only pyrethroids are the only class of insecticides that can be used for ITNs (Coleman, Sharp et al. 2006; Pluess, Tanser et al. 2010).

With the current scale up of both ITNs and IRS in sub-Saharan Africa, it is apparent that most communities will be protected by both these interventions (RBM 2010). Several studies have demonstrated that combing these interventions yields better results than singular implementation (Yadav, Sampath et al. 1998; Curtis and Mnzava 2000; Lengeler 2004; Kleinschmidt, Schwabe et al. 2009; Bekele, Belyhun et al. 2012) particularly in areas of perennial malaria transmission (Hamel, Otieno et al. 2011). Some other studies have demonstrated little or no positive impact of combining IRS and ITNs (Protopopoff, Van Bortel et al. 2007). Largely,

the optimal combination of ITNs and IRS still remains an area that requires further research to elucidate further (Okumu and Moore 2011).

With the focus on IRS and ITNs, LSM is often dismissed as a reliable vector control method in Africa (Kitron and Spielman 1989; Killeen, Fillinger et al. 2002; Killeen, Fillinger et al. 2002; Gu and Novak 2005). However some studies have shown that larval control is able to reduce the malaria mosquito larvae abundance and adult female populations by more than 90% including outdoor biting species (Fillinger and Lindsay 2006; Fillinger, Ndenga et al. 2009). Thus LSM has the capacity to supplement IRS and ITNs in sustaining low malaria transmission as it attacks both the indoor feeding mosquitoes and those less likely to be affected by IRS or ITNs such as *Anopheles arabiensis* (Killeen, Fillinger et al. 2002; Walker and Lynch 2007; Fillinger and Lindsay 2011; Worrall and Fillinger 2011). Some key challenges with large scale LSM deployment put forward included; the difficulty of estimating impact using a randomised control trail, which is impractical for this intervention; the belief that greatest impact on malaria transmission reduction should focus on reducing longevity of mosquito vectors through targeting adults and; spatial and temporal distribution of vector larvae may inhibit LSM efforts due to high cost and management (Fillinger, Kannady et al. 2008; Fillinger and Lindsay 2011). Thus, LSM has been relegated to be only implemented in specific circumstances as a compliment to IRS and ITNs. These settings have been described as those with few mosquito breeding sites that are easily identifiable and accessible (Fillinger and Lindsay 2006; WHO 2011). Additionally, these areas should be in moderate to low transmission, ideally in the consolidation phase towards control and elimination to allow for targeting in space and time (Fillinger and Lindsay 2006; Fillinger, Kannady et al. 2008; Fillinger, Ndenga et al. 2009; Fillinger and Lindsay 2011).

1.2.10.2 Scaling up access to case management beyond the facility: community case management

There are a substantial number of deaths that occur in homes in endemic areas due to poor access to health care services (Black, Morris et al. 2003; Rutebemberwa, Kallander et al. 2009; Black, Cousens et al. 2010; Kinney, Kerber et al. 2010). The concept of primary health care stresses the need to intergrate both preventive and curative interventions with close involvement of the community due to these access limitations (Haegeman, Wyffels et al. 1985; Riji 1992; O'Meara, Noor et al. 2009). Thus community based approaches built around the principal of providing prompt diagnosis and treatment for malaria as close to home as possible by community health workers become all the more important (Chanda, Hamainza et al. 2011; Counihan, Harvey et al. 2012; Kisia, Nelima et al. 2012; Rutta, Francis et al. 2012; Thiam, Thwing et al. 2012). In the 1970s, community based health workers were used as a condiut for delivery of chemoprophylactic treatment to communities. This at the time, in addition to treatment of symptomatic cases, was considered a key malaria control intervention in endemic areas (Haegeman, Wyffels et al. 1985; Haegeman, Wyffels et al. 1985; Greenwood, Greenwood et al. 1988). Based of this initial concept, various studies demonstrated the use of community based health workers in malaria control using variations of the initial concept. These variations included training community health workers to recognise and refer all patients with danger signs and treat all simple forms of malaria (Spencer, Kaseje et al. 1987; Spencer, Kaseje et al. 1987; Delacollette, Van der Stuyft et al. 1996) and /or training mothers to recognise danger signs and seek drugs from community health workers (Pagnoni, Convelbo et al. 1997; Sirima, Konate et al. 2003). These community-based treatment interventions were based on clinical

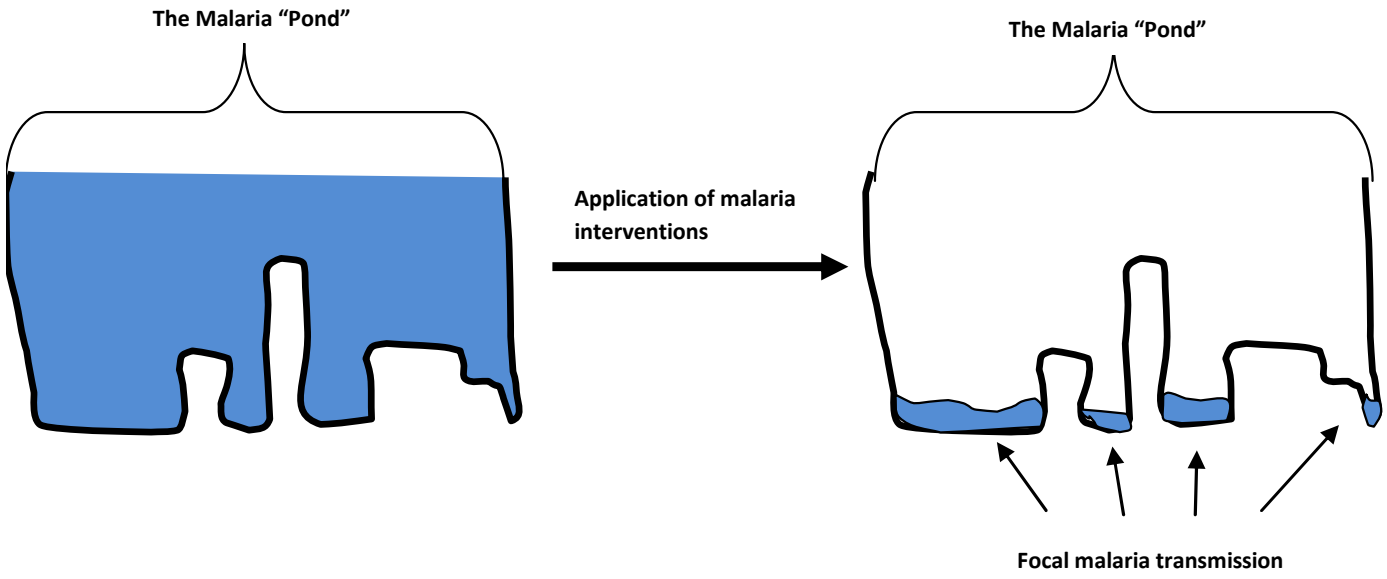
diagnosis based on fever and other common signs and symptoms (Font, Alonso Gonzalez et al. 2001; Othnigue, Wyss et al. 2006) which lead to over diagnosis of malaria. The current recommendations for malaria case management advocate for confirmatory diagnosis prior to initiation of malaria chemotherapy (WHO 2006; Shillcutt, Morel et al. 2008; WHO 2010) and for strengtherning of community based interventions as a cornerstone of increasing case management coverage (WHO 2006; WHO 2012). Several studies have shown that community health workers are able to use and interprete malaria rapid diagnostic tests and subsequently provide treatment or refer appropriately (Harvey, Jennings et al. 2008; Chanda, Hamainza et al. 2011; Counihan, Harvey et al. 2012; Ratsimbasoa, Ravony et al. 2012). Community health workers have also shown ability to provide integrated treatment for malaria, pneumonia and diahorea (Yeboah-Antwi, Pilingana et al. 2010; Kalyango, Rutebemberwa et al. 2012). This offers an oppurtunity to scale up malaria case management to the peripheral areas using commmunity health workers as part of the routine health system outreach in detecting mildly symptomatic malaria cases (Harvey, Jennings et al. 2008; Chanda, Hamainza et al. 2011; Counihan, Harvey et al. 2012; Ratsimbasoa, Ravony et al. 2012). Additionally, community case management is useful to achieve and sustain control and thus is a prerequisite to elimination of malaria (WHO 2012).

1.2.11 Eliminating cryptic malaria infections – The Role of Community Health Workers

Even though most countries have been successful in reducing the burden of malaria it still remains high in many remote and historically holoendemic areas. This variation occurs between countries, within short distances in same local areas and between age groups (Snow, Bastos de

Azevedo et al. 1994; Giha, Rosthoj et al. 2000; Snow and Marsh 2002; Bousema, Drakeley et al. 2010). The exposure to infected mosquitoes is likely the major reason for this variation (Bousema, Drakeley et al. 2010). This is exemplified in the anology of "draining the pond", which illustrates that the bottom of a pond is not always even and as such when water is drained out of it, some places within the pond continue to have water while others are dry. The areas with water represent areas where malaria transmission still occurs, thus intuitively, as transmission of malaria drops it becomes more focal (Figure 7) (Newman 2012).

Figure 7."Draining the pond" (Adapted from Newman 2012)



With most countries adopting an elimination agenda for malaria due to scale up of high impact interventions, these malaria hot spots are increasing becoming more important (Barnes,

Durrheim et al. 2005; Bhattarai, Ali et al. 2007; O'Meara, Bejon et al. 2008). Several studies have shown that in areas of low and seasonal malaria burden, transmission is usually sustained by clustered populations that are semi immune who are infectious to mosquitoes, carry asymptomatic infection for long periods of time and remain undetected and thus untreated (Hogh 1996; Freeman, Laserson et al. 1999; Drakeley, Akim et al. 2000; Abdel-Wahab, Abdel-Muhsin et al. 2002; Alves, Durlacher et al. 2002; Coleman, Kumpitak et al. 2004; Bousema, Schneider et al. 2006; Shekalaghe, Bousema et al. 2007; White 2008; Okell, Ghani et al. 2009; Ogutu, Tiono et al. 2010; Gosling, Okell et al. 2011). Currently malaria control curative activities are essentially targeted at symptomatic patients and are reliant on passive detection of patients reporting to health facilities, seeking care when they are unwell (Najera 2001). Strategies for control using chemotherapy are being revisited as they can be complimentary to the other primary interventions to further accelerate declines of disease burden (Bhattarai, Ali et al. 2007; Okell, Drakeley et al. 2008; Gosling, Okell et al. 2011; Kern, Tiono et al. 2011; Huho, Killeen et al. 2012) particularly in areas of low and seasonal transmission (Freeman, Laserson et al. 1999; von Seidlein, Walraven et al. 2003). Thus in order to reduce malaria transmission, populations with cryptic infections and associated parasitic biomass must be targeted (Drakeley, Akim et al. 2000; Okell, Ghani et al. 2009; Gosling, Okell et al. 2011; Shekalaghe, Drakeley et al. 2011). In high transmission areas, the burden of these infections is in the older age groups, while the young harbour the burden of clinical disease (White 2008; Gosling, Okell et al. 2011). This is due to the vector biting rates being dependent on the host biomass and hence age (Smith, Killeen et al. 2004) and development of immunity due to exposure over time.

In order to target asymptomatic parasite with curative drugs populations there are two plausible broad strategies that can be considered: mass drug administration (MDA) or mass screening & treatment (MSAT), historically referred to as mass blood examination (WHO 1963; Gosling, Okell et al. 2011). MDA is the administering of antimalarials to each member of a population, regardless of the presence of malaria detectable parasitaemia or symptoms, while MSAT targets only confirmed parasitaemic individuals after parasitological testing for infection status (refer to table 7). The success of these interventions lies in their ability to reduce prevalence of parasitaemia to thresholds where re-emergence is improbable and control maintained. The possibility of success increases when parasite prevalence in low in the general population and vector population density is also low. This usually occurs towards the end of the dry season in most endemic areas (Okell, Griffin et al. 2011) or can occur as a result of effective implementation of such as LLIN deployment and/or IRS (Gosling, Okell et al. 2011). MDA as opposed to routine malaria case management, is a high coverage intervention and may accelerate the spread of drug-resistant strains particularly when sub-therapeutic doses of antimalarials are administered leading to pressure on the parasite populations (Greenwood 2010; Kaneko 2010). MSAT has been proposed as a better alternative due to the addition of a confirmatory diagnostic tool which leads to provision of treatment only to those found positive for infection thus reducing selection pressure on the drugs (Ogutu, Tiono et al. 2010; Crowell, Briet et al. 2013). However, a challenge with MSAT is lies in the missing of sub patent infections which, as alluded to contribute to the reservoir of infection and its higher cost and logistical demands compared to MDA (Crowell, Briet et al. 2013). With increasing availability of more sensitive diagnostic tools such as RDTs, the possibility of implementing MSAT is logistically

possible using community based health workers (Kaneko 2010; Ogutu, Tiono et al. 2010). MSAT has been recommended for small, isolated communities as it relies on explicit, accurate population data and the ability to monitor the movements of people with access to a confirmatory diagnostic tool which is the essential foundation of a successful MSAT program (Macauley 2005) as an "end game" intervention in the overall continuum of malaria elimination

Table 7.Acronyms and strategies for mass drug administration (Adapted from Gosling, Okell *et al*., 2011)

Acronym	Name	Definition
MDA	Mass drug administration	Simultaneous drug administration, without testing, to the total population of an island, country, region or district
TMDA	Targeted mass drug administration	Drug administration to all people living within a small high-risk area, without testing, such as the population surrounding a case
MSAT or MBS	Mass screening and treatment or mass blood survey	Screening of all persons irrespective of symptoms and treatment of positives only
FSAT	Focal screening and treatment	As MSAT, but in a localized area such as a household, neighbourhood, or village
MFT	Mass fever treatment	Treatment of all people with fever, irrespective of diagnosis
ACD or MSFAT	Active case detection or mass screening of fever and treatment	Screening of all people with fever and treatment of positives
IPT	Intermittent preventive treatment	Giving a full treatment dose of an antimalarial to an asymptomatic individual, regardless of infection status, at an opportunistic time
IPTi	Intermittent preventive treatment of malaria in infants	IPT of infants (under the age of 1 year) at times of vaccination
IPTp	Intermittent preventive treatment of malaria in pregnancy	IPT of pregnant women at times of antenatal visits
IPTc or SPC	Intermittent preventive treatment of malaria in children or seasonal prophylaxis in children	IPT of children at defined time periods during the malaria season in seasonal transmission sites. Usually applies to children under the age of 5 years

The concept of community based management of malaria stems from the recognition that, there is a human resource deficit amongst clinically trained cadres, thus requiring interventions outside centralised, facility based health system services to improve access to appropriate

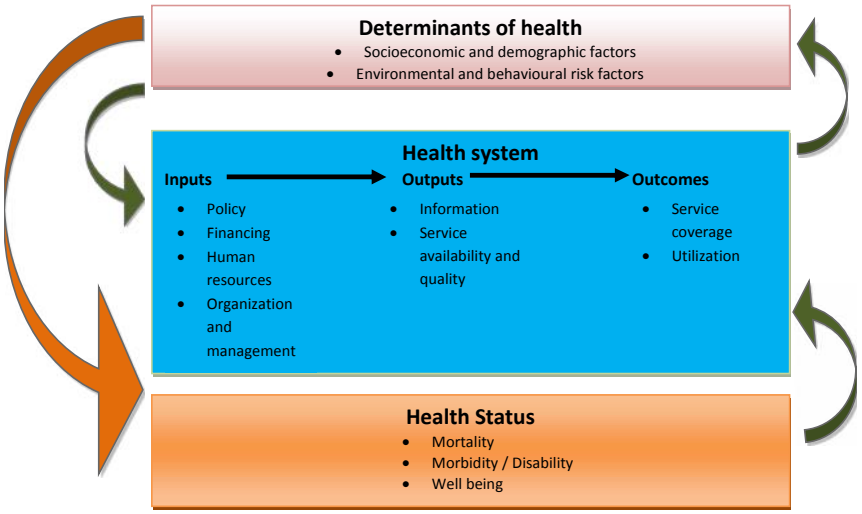
malaria case management (WHO 2004; WHO 2004; WHO 2005; WHO 2012). Community health workers have demonstrated the capacity to diagnose malaria and provide treatment according to the local guiding guidelines (Cunha, Piovesan-Alves et al. 2001; Harvey, Jennings et al. 2008; Elmardi, Malik et al. 2009; Hawkes, Katsuva et al. 2009; Mmbando, Segeja et al. 2009; Chanda, Hamainza et al. 2011; Counihan, Harvey et al. 2012; Kalyango, Rutebemberwa et al. 2012). The community approach has also been shown to be cost effective (Pang and Piovesan-Alves 2001; Chanda, Hamainza et al. 2011), improve delivery of malaria case management overall (Yeung, Van Damme et al. 2008; Elmardi, Malik et al. 2009; Hamer, Brooks et al. 2012) and is well accepted by communities where it has been implemented (Ajayi, Falade et al. 2008; Mukanga, Tibenderana et al. 2010; Yeboah-Antwi, Pilingana et al. 2010). The demonstrated decline in malaria burden in most parts of sub-Saharan Africa, has made the reliance on fever as a marker for diagnosis of malaria undesirable and hence the need for confirmatory diagnostic tests at all levels of care (Rutta, Francis et al. 2012). Community health workers have shown that they are able, with minimal training, of using and interpreting RDTs correctly (Counihan, Harvey et al. 2012), which is key to establishing effective high quality surveillance data (Hopkins, Talisuna et al. 2007; D'Acremont, Lengeler et al. 2010; WHO 2010) which can be used for decision making (Hopkins, Talisuna et al. 2007; WHO 2012)

1.2.12 The potential role of Community Health Workers in public health surveillance

Public health surveillance has been defined as the "ongoing systematic collection, analysis, and interpretation of data critical to the planning, implementation, and evaluation of public health interventions, and is closely integrated with timely dissemination of data generated to all

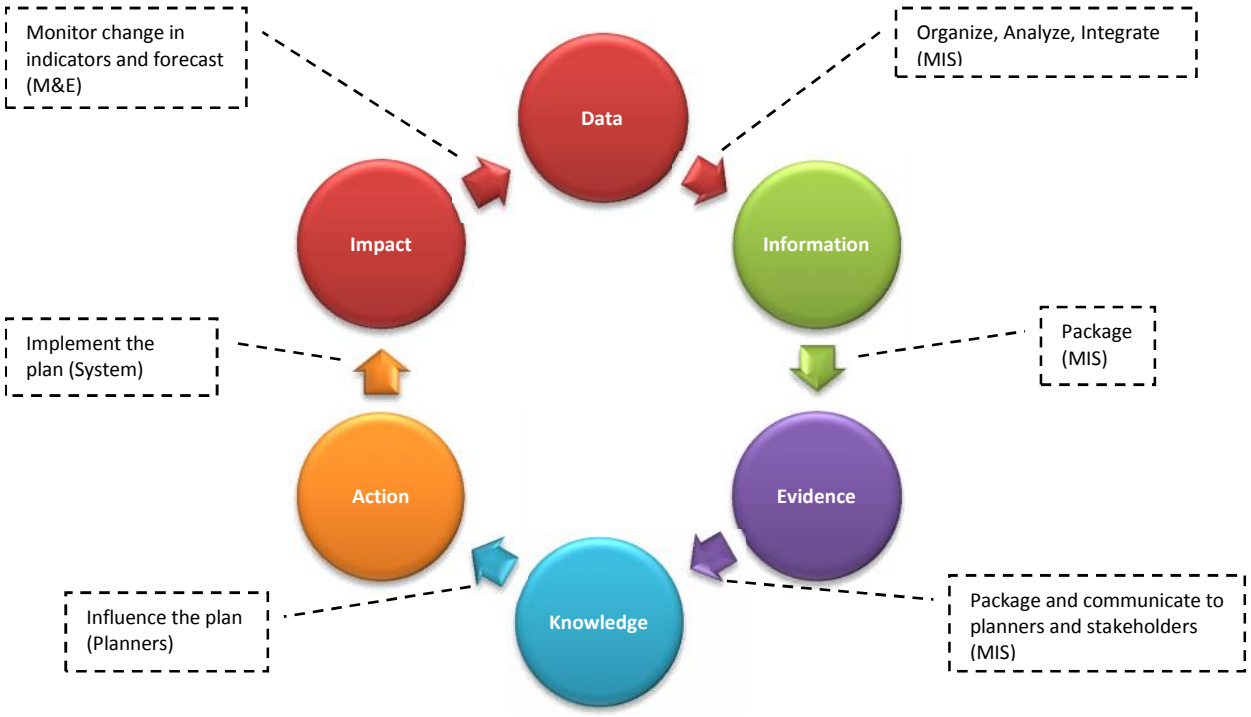
stakeholders" (WHO 1962; Thacker and Berkelman 1988). Timeliness is a key performance measure of public health reporting systems and may vary by disease, intended use of the data, and public health system level (Jajosky and Groseclose 2004). Surveillance stresses the need to collect routinely a limited amount of data relevant to describing the population's health status (Nelson, Thompson et al. 1997), primarily to detect any changes over time resulting from changes in health practices, infectious and environmental agents and control methods. There are four main areas of measurement in health information systems are; (1) Health determinants, (2) Health system inputs, (3) Health system outputs and (4) Health outcomes (as illustrated in Figure 8)(WHO 2008; WHO 2008). Thus the core indicators selected should be able to reflect changes over time and should be valid, reliable, specific, sensitive, feasible/affordable to measure, relevant and comparable (Lippeveld, Sauerborn et al. 2000; WHO 2008; WHO 2008)

Figure 8.Domains for measurement for health information systems (Adapted from WHO 2008; WHO 2008)



Thus public health surveillance aids in decision making, priority setting, targeting interventions and management of health problems through the tracking of set indicators (de Savigny and Binka 2004; Jajosky and Groseclose 2004; Odhiambo-Otieno 2005; Thacker, Qualters et al. 2012). In sub-Saharan Africa these routine information systems are inadequate and weak. A major reason for this is that, monitoring and evaluation is not considered an integral part of the information pathway and this lack of verification has lead to stakeholders having little or no confidence in utilising this information for decision making and planning . Often, the available information is also poorly packaged and communicated making its use for decision making a challenge by the health system (de Savigny and Binka 2004). This has been compounded by the lack of financial investment, human resources, infrastructure and institutional capacities, particularly in low income countries (St Louis 2012). For evidence based decision making to have a positive impact on the health system there must a translation of the data into information which subsequently needs to be packaged to reveal the evidence it embodies. This evidence adds to the body of knowledge which warrants some action by stakeholders which drives change or impact. This “change” alters the data and the cycle begins again (Figure 9) (de Savigny and Binka 2004).

Figure 9.Pathway for evidence based planning (Adapted from de Savigny & Binka 2004) – (M&E = Monitoring and evaluation, MIS= Management of information systems)



In poorly resourced countries, community-based surveillance systems (CBSS) can compliment health facility based public health surveillance. They are able to provide quantitative estimates of disease burden in a defined population and service delivery indicators (Baker and Ross 1996). This they have been able to demonstrate as part of their curative functions particularly for community based management of malaria and pneumonia predominantly in their local communities that are usually rural and hard to reach (Yeboah-Antwi, Pilingana et al. 2010; Kalyango, Rutebemberwa et al. 2012). Community health workers (CHWs) have also demonstrated that they are able to collect epidemiological data on various diseases provided they have the appropriate training (Hopkins, Talisuna et al. 2007; Alba, Hetzel et al. 2011;

Kalyango, Rutebemberwa et al. 2012). Studies have shown that CHWs are also able to implement routine programmatic community based mosquito surveillance (Chaki, Mlacha et al. 2012; Sikaala, Chinula et al. 2014). Information generated from community level efforts can be utilised to describe spatial and temporal discrepancies in health status and/or access to services which would in turn allow for targeted interventions and resource allocation on programmatic scales (Tatem, Campiz et al. 2011; Barclay, Smith et al. 2012).

1.2.13 Technology and Public Health Surveillance – What part can mobile phones play?

The effective control of most diseases, particularly malaria require a wide range of timely and routinely collected data which should encompass, (1) surveillance, monitoring and evaluation activities, (2) health commodities and consumables supply chain monitoring and (3) pharmacovigilance, safety and quality of antimalarial therapies monitoring (Zurovac, Talisuna et al. 2012). In most developing countries, health related information is collected routinely, usually passively, shared at intervals of a month or two at best, of poor quality, incomplete and contributes to poor management, resource allocation, priority setting and provision of health services (Safaie, Mousavi et al. 2006; Braa and Sahay 2012). It is important here to note that health information has a positive recurring endogenous relationship between timeliness, use, correction and quality (de Savigny and Binka 2004). These deficits certainly lead to delayed responses to threats and emergencies such as disease outbreaks, supply stock-outs of and undetected occurrences of severe adverse drug reactions and treatment failures (Talisuna, Staedke et al. 2006; Rowe, Kachur et al. 2009; Maokola, Willey et al. 2011). This has been primarily attributed to limitations in human, infrastructural and institutional capacity (Chretien,

Burkom et al. 2008; Ranck 2011). Perhaps the greatest challenge surveillance systems need to overcome is the need to relay data to and from the collection points in a timely manner (Mawudeku, Ruben et al. 2007). Unlike conventional public health surveillance which is dependent on the health worker with/without laboratory confirmation reporting data which may initiate a manual analysis (Siegrist 1999; Soto, Araujo-Castillo et al. 2008), electronic systems dramatically accelerate the collation, availability of health data and increase the automation of many standard analysis required for decision making (Soto, Araujo-Castillo et al. 2008; Ranck 2011). An electronic approach to surveillance may address principally four important objectives of surveillance which include: (1) outbreak detection, (2) strengthen local/ focal responses to outbreak detection, (3) monitoring and evaluation of disease patterns and trends and/or intervention effectiveness or impact, and (4) facilitate data-driven policy decision making through hypothesis testing (Henning 2004). In order to efficiently implement an electronic surveillance system, it is essential to have sufficiently trained specialist staff, infrastructure, technical support and equipment. There several proposed platforms that can be used in these types of electronic surveillance systems and which could provide real-time data collation and access (Johnson and Blazes 2007; Mawudeku, Ruben et al. 2007; Morse 2007; Siswoyo, Permana et al. 2008; Keller, Blench et al. 2009; Chunara, Freifeld et al. 2012; Morse 2012; Free, Phillips et al. 2013).These platforms compliment classical control, verification and analysis found in traditional surveillance, through increased coverage, transparency, timeliness and scalability (Freifeld, Chunara et al. 2010). However, most of these are largely based on email and internet availability, which still remains a challenge in many developing countries particularly in isolated rural settings (Johnson and Blazes 2007). In the recent past there has

been dramatic growth in mobile phone networks globally and Africa particularly, where approximately two-thirds of the population is covered by a mobile network with an estimated penetration rate of 50%, representing over half a billion mobile phone subscribers across the continent (ITU 2010). While third generation mobile phone networks capable of supporting high speed internet exchange have followed this trend, these lag behind regular second generation mobile phone networks, making the later the most important communication infrastructure for supporting surveillance reporting in the developing world (Johnson and Blazes 2007). Evidence shows that access and use of mobile phones is not strongly influenced by socio-economic lines, which is not the case with access to other communication technologies such the internet (Forestier, Grace et al. 2002). Thus, mobile phones can play a complimentary role in the provision of health system information needs in developing countries (Deglise, Suggs et al. 2012) such as data collection, training and access to reference material, communication between health workers, decision support, supervision, and health behaviours promotion (Derenzi, Borriello et al. 2011) including disease surveillance (Diero, Rotich et al. 2006; Fynn, de Jager et al. 2006; Shirima, Mukasa et al. 2007; Skinner, Rivette et al. 2007; Blake 2008; Ma, Zhou et al. 2009; Tatem, Qiu et al. 2009). However, the use of mobile phones in public health still remains in its infancy in most of the developing world (Krishna, Boren et al. 2009).

Similar to other electronic platforms, mobile phones can be used to directly record digital data at the point of collection, potentially adding efficiency, through automated aggregation and analysis, and versatility by accommodating various data forms for divergent purposes based on need (Fjeldsoe, Marshall et al. 2009; Whittaker, Borland et al. 2009; Wei, Hollin et al. 2011). Mobile phone technologies are now considered important communication infrastructures

across health, finance, education and marketing sectors (Tufano and Karras 2005; Levine, McCright et al. 2008; Fjeldsoe, Marshall et al. 2009; Lefebvre 2009; Ackerman, Filart et al. 2010; Derenzi, Borriello et al. 2011; Lim, Xue et al. 2011; WHO 2011). A summary of both the attractions and barriers of mobile phone technology based data collection tools is provided in table 8.

Table 8.Attractions and barriers to use of mobile phone technology based data collection systems

	Property	Description
Attraction	Low start up costs	Costs less to rollout over a large area has low training costs, can be used by any cadre and running costs are low (cheaper than voice)
	User friendly	Data inputting is relatively easy using SMS or any predefined tools on the mobile phone.
	Payment forms	Using the prepaid module allows for users to control their usage of airtime and indeed have access to more if need be.
Barriers	Cost issues	High cost of penetration of the mobile phones to remote parts of many developing countries
	Information carrying capacity	Particularly in SMS reporting where characters are limited to 160 and in areas on low band width
	Language and illiteracy	Particularly when information has to be transmitted in the form of text

Mobile phones can transmit information via both verbal and non-verbal methods (Fynn, de Jager et al. 2006; Patnaik, Brunskill et al. 2009), however the former is considered cost inhibitive, prone to errors and generally impractical in most developing countries (Fynn, de Jager et al. 2006). The most common types of non-verbal based data capture applications are

short messaging system (SMS), Java Micro Edition Platform (J2ME) and internet /web-based forms (Table 9)(Loudon 2009).

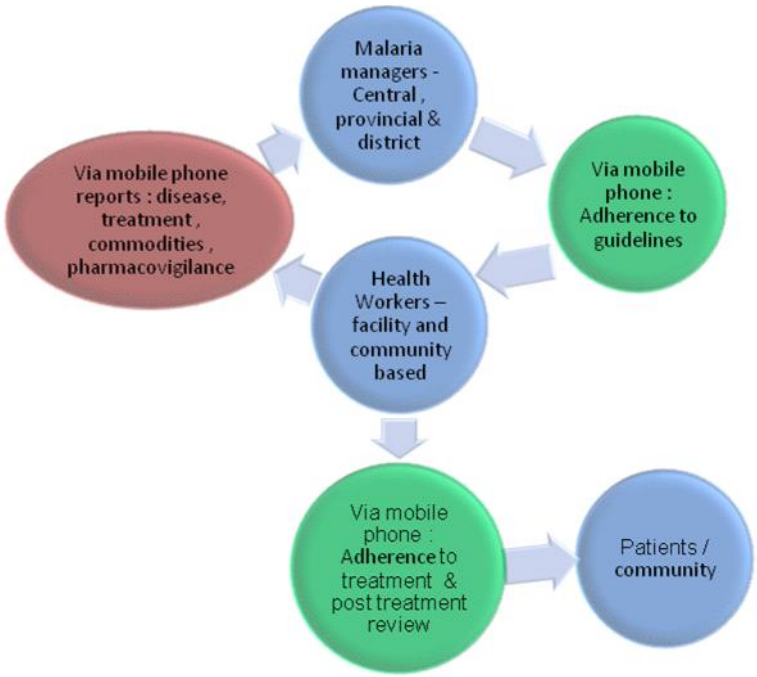
Table 9.Characteristics of client applications and data transmission (Adapted from Loudan 2009)

Client Application	Type of phone	Requirements for data collection	Data transfer methods available
Fixed format SMS	All SMS capable phones	No additional software needs. The user writes and SMS in a predefined format, representing answers to successive questions	SMS only
Java Micro Edition Platform (J2ME) application	All java phones	Data capture application has to be downloaded onto the phone via the web or bluetooth or cable. The data collector navigates through questions in an application on the phone, which saves the responses and transmits the completed form to a server.	SMS or General packet radio service (GPRS) (these can be asynchronous allowing for data saving and transmission when web access is available)
Web based form	All phones with a web browser	Phone must have access to the internet. The data form is filled out online, and saved directly online	GPRS

Of these three applications, the most readily available in many rural parts of most developing countries, cost effective and feasible is SMS, (Fynn, de Jager et al. 2006; Safaie, Mousavi et al. 2006; Fjeldsoe, Marshall et al. 2009; Cole-Lewis and Kershaw 2010; Wei, Hollin et al. 2011) particularly in resource constraint settings due to its minimal running costs and availability on almost all inexpensive “feature” or smart mobile phone handsets (Zurovac, Sudoi et al. 2011; Zurovac, Talisuna et al. 2012). J2ME has a higher set up cost, but due to its ability to transfer more data, tends to be cheaper as a long term investment compared to SMS which can send a maximum of 160 characters per single SMS message, thus inhibitive depending on the amount of data required to be transmitted (Cole-Lewis and Kershaw 2010).

Frequent and rapid inflow of data improves the understanding of local mapping of disease burden and decision-making abilities, enabling efficient targeting of interventions and commodities, particularly for diseases such as malaria (Kamanga, Moono et al. 2010). As the malaria burden becomes increasingly focal, community-level mapping techniques become important in order to be able to identify these "hot spots" of transmission (Bousema, Drakeley et al. 2010; Chizema-Kawesha, Miller et al. 2010; Davis, Kamanga et al. 2011; Bousema, Griffin et al. 2012). Mobile phones thus, offer a practical complimentary tool, that can be utilized for reporting data collected by community-based surveillance staff, in order to describe spatial and temporal malaria transmission in real time and guide timely programming (Kamanga, Moono et al. 2010; Zurovac, Talisuna et al. 2012). Figure 10 below illustrates some possible applications of mobile phone technology in malaria programmes.

Figure 10.Applications of mobile phone technology in malaria programming (Adapted from Zurovac, Talisuna *et al*., 2012)



1.2.14 A brief context of public health surveillance in Zambia

As part of the national health reforms, Zambia decentralised its health system in 1994 which receives support from government (37%), cooperating partners/donors (26%), households (21%), and private entities (13%) (Bossert, Chitah et al. 2000; Heywood, Nielsen et al. 2005; Mutemwa 2006; WHO 2006). The Zambian health care delivery system consists primarily of 4 levels; the Ministry of Health (MOH) (national coordination of the health service delivery systems); the provincial medical offices (liaisons between the MOH and the districts); the district medical office (oversee the district health facilities and manages the district health team); and the hospital (first, second and third level) management boards (management of primary, secondary and tertiary level hospitals)(Hoppenbrouwer and Kanyengo 2007). At health centre level, each facility has facility committees and neighbourhood committees to encourage community participation in their management (WHO 2006). As part of the health decentralisation process, the health information management system (HMIS) was adopted as a significant element of district-level health management practice, to ensure that concerns about long term sustainability, performance, proficiency and effectiveness were addressed (Mutemwa 2006; MOH 2008). The initial roll out of the HMIS had various challenges which led to several review processes (Heywood, Nielsen et al. 2005; MOH 2007). These reforms mainly considered reorganization of key performance indicators for measurement of the millennium development goals and national strategic plans and associated goals, including providing for local self-assessment at all levels of health care provision for evidence based decision making (Heywood, Nielsen et al. 2005; Mutemwa 2006). The current HMIS has been scaled up to all formal public health facilities in the country and integrates routine service activities, specialist

vertical programs including the malaria control programs and selected aspects of epidemiological surveillance for notifiable disease (Heywood, Nielsen et al. 2005; MOH 2008). Generally, HMIS has largely failed to deliver on its ultimate goals primarily because of, the lack of complete and timely reports, inadequate skilled labour and reporting infrastructure and negligible quality control or even quality assurance (Evans and Stansfield 2003; WHO 2013). Additionally, HMIS do not routinely report on laboratory, environmental health and administrative data such as finances, human resources and transport among others (Heywood, Nielsen et al. 2005; MOH 2008). Fundamental health decisions are made based on rough incomplete poor quality estimates of disease and treatment burdens (Siaga 1993; UNFPA 1995).

1.2.15 Study Design Considerations

The study was part of a larger multi country study, called the Malaria Transmission Consortium (MTC) which was funded by the Bill and Melinda Gates Foundation. The overall goal of this consortium project was to "develop a standardized set of tools that can be used to rapidly and accurately measure the malaria transmission intensity in a manner that will be suitable for use by control programs". The objectives of the MTC were "(1) The development of a set of standardized measures of malaria transmission (and the biological factors that contribute to transmission) that can be used across the range of malaria epidemiology, (2) The evaluation of the effectiveness of different transmission-reducing malaria control interventions, both alone and in various combinations in environments with varying parasite transmission levels, and (3) An assessment of the impact of biological factors associated with vector populations, such as variable levels or different mechanisms of insecticide resistance or variation in vector behaviours, on the effectiveness of malaria control interventions.". This study aimed to address

objectives 1 and 2, although some data collected to address objective 3 was also included as background descriptive data that strongly supports the selection of IRS with non-pyrethroid insecticides as the vector control measure to be evaluated. Malaria infection burden was assessed through using longitudinally in 14 clustered cohorts, each comprising of 165 households, centred around a selected health facility, so that the incremental impact of supplementary vector control with IRS could be evaluated by assigning them experimentally to these clusters using a randomised design. While CHWs were required to actively visit each household every month, and to test and treat all consenting and assenting individuals present at the time, no effort was made to insist that each individual participated in testing and treatment during each visit or that the CHW conduct follow up visits to ensure participation by all household members. These deliberate choices to ensure relatively easy, flexible interactions between the CHWs and the residents were driven by programmatic logistical considerations that aimed at obtaining valid data that were both (1) adequate for the purposes of monitoring and characterizing malaria infection burden, as well as the impact of experimentally-introduced supplementary IRS upon it, and (2) practically applicable routinely on national scale by the control programmes. The rigidity of control exerted by the research team was minimized so that the relevance of the results to programmatic applications under representative condition of management on much larger scales by the Ministry of Health was maximized (Habicht, Victora et al. 1999; Glasgow, Lichtenstein et al. 2003; Boffetta 2011). ,

While every effort was made to randomly assign the specific IRS treatments to these clusters in a controlled fashion, it was obvious from the outset that NMCC did not have full control over either the availability of insecticide products, nor over all the actual implementation operations

within these districts, because both procurement and implementation were both partially executed by contractors engaged directly by the funding agency. Given this situation, as well as the limited capacity of NMCC for conducting research at the time, it was decided to implement as closely controlled an experimental study as possible but accept that it failed to satisfy the criteria required to have it formally registered as a randomized controlled trial. Despite the effort to experimentally control the assignment of IRS treatments, it was therefore decided to nevertheless classify the study *a priori* as an observational study designed to deliver evidence of impact plausibility rather than probability. As an observational study, the inherent methodological limitations of vulnerability to selection bias and confounding variables, which may cause under-estimation or over–estimation of intervention impact must be accepted (Carlson and Morrison 2009; Boffetta 2011), even though many of the latter were recorded and taken into consideration during the implementation and analysis phase of the study. As many potential confounding variables as possible were recorded so that their contribution to the primary outcome, which in this study was malaria infection diagnostic positivity, could be parsimoniously (Sober 1981) accounted for in *post hoc* regression model analyses (Carlson and Morrison 2009; Starks, Diehr et al. 2009), thus minimizing biases in the estimation of the treatment effects (Starks, Diehr et al. 2009). .

However the study was conducted as an experimental evaluation of the adequacy, effectiveness and plausibility of a community based surveillance system that used both passive and active malaria testing and treatment participant contact mechanisms in a programmatically relevant setting. This approach as opposed to an efficacy evaluation which would require optimal conditions with sufficient resources, financial and otherwise allowed for an

understanding of program implementation in a real programmatic setting (Habicht, Victora et al. 1999; Glasgow, Lichtenstein et al. 2003; Flay, Biglan et al. 2005). So while the observational nature of the study has some disadvantages in terms of delivering evidence of impact plausibility, rather than probability, it also has the advantage of quantifying effectiveness under programmatically relevant conditions, rather than merely efficacy under controlled conditions of questionable relevance to the NMCC. In fact, this approach also has the advantage of identifying limitations in these prototype implementation systems by , in terms of coverage and utilization of these interventions in a real life setting (Carlson and Morrison 2009). Lessons learnt with regard to the practicality, scalability, generalizability and resource input requirements for program implementation may be highly valuable and relevant as they were evaluated in representative programmatic conditions (Habicht, Victora et al. 1999; Glasgow, Lichtenstein et al. 2003; Flay, Biglan et al. 2005).

CHAPTER TWO

MONITORING, CHARACTERIZATION AND CONTROL OF CHRONIC, SYMPTOMATIC MALARIA INFECTIONS IN RURAL ZAMBIA THROUGH MONTHLY HOUSEHOLD VISITS BY PAID COMMUNITY HEALTH WORKERS

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2.0 Abstract

Background

Active, population-wide mass screening and treatment (MSAT) for chronic *Plasmodium falciparum* carriage to eliminate infectious reservoirs of malaria transmission have proven difficult to apply on large national scales through trained clinicians from central health authorities.

Methodology

Fourteen population clusters of approximately 1,000 residents centred around health facilities (HF) in two rural Zambian districts were each provided with three modestly remunerated community health workers (CHWs) conducting active monthly household visits to screen and treat all consenting residents for malaria infection with rapid diagnostic tests (RDT). Both CHWs and HFs also conducted passive case detection among residents who self-reported for screening and treatment.

Results

Diagnostic positivity was higher among symptomatic patients self-reporting to CHWs (42.5%) and HFs (24%) than actively screened residents (20.3%), but spatial and temporal variations of diagnostic positivity were highly consistent across all three systems. However, most malaria infections (55.6%) were identified through active home visits by CHWs rather than self-reporting to CHWs or HFs. Most (62%) malaria infections detected actively by CHWs reported one or more symptoms of illness. Most reports of fever and vomiting, plus more than a quarter of history of fever, headache and diarrhoea, were attributable to malaria infection. The minority of residents who participated >12 times had lower rates of malaria infection and

associated symptoms in later contacts but most residents were tested <4 times and high malaria diagnostic positivity (32%) in active surveys, as well as incidence (1.7 detected infections per person per year) persisted in the population. Per capita cost for active service delivery by CHWs was US$5.14 but this would rise to US$10.68 with full community compliance with monthly testing at current levels of transmission, and US$6.25 if pre-elimination transmission levels and negligible treatment costs were achieved.

Conclusion

Monthly active home visits by CHWs equipped with RDTs were insufficient to eliminate the human infection reservoir in this typical African setting, despite reasonably high LLIN/IRS coverage. However, dramatic impact upon infection and morbidity burden might be attainable and cost-effective if community participation in regular testing can be improved and the substantial, but not necessarily prohibitive, costs are affordable to national programmes.

2.1 Background

A relatively large proportion of malaria infections are only mildly symptomatic, especially in endemic countries where acquired immunity moderates both parasitaemia and pathology (Smith, Schellenberg et al. 1994; Bottius, Guanzirolli et al. 1996; Alves, Durlacher et al. 2002). Even in areas with only modest, seasonally sporadic transmission where little immunity exists among the human populations, the many mildly symptomatic infections that persist chronically, often at sub-patent parasite densities below thresholds of detectability, are responsible for sustained malaria transmission (Babiker, Abdel-Muhsin et al. 1998; Bousema, Gouagna et al. 2004). As early as the 1930s, these populations have been targeted for treatment with anti-malarial drugs as a control measure for malaria transmission, particularly during the era of the Global Malaria Eradication Programme initiated in the 1950s (Molineaux and Gramiccia 1980; Jeffery 1984; Killeen 2013). Due to the emergence of drug resistance, which was attributed to mass drug administration campaigns (Kouznetsov 1987; Wellems and Plowe 2001; Parija and Praharaj 2011) and evidence that the impact on transmission may be limited, the use of therapeutic approaches to control transmission rather than burden was not considered as effective (Molineaux and Gramiccia 1980; Jeffery 1984; Wernsdorfer 1992; WHO 2010) until recently when interest in malaria elimination was rejuvenated (Moonen, Cohen et al. 2010; WHO 2010; Maude, Socheat et al. 2012).

Currently curative drugs are predominantly targeted at symptomatic individuals based on passive detection of acute infections among patients seeking care for fevers through the formal and informal health system (McCombie 1996; Tanner and Vlassoff 1998; Najera 2001). Strategies for control of human-to-mosquito transmission by providing chemotherapy to

chronic parasite carriers are now being revisited as they may be complimentary to front-line interventions for preventing mosquito-to-human transmission, such as long-lasting insecticidal nets (LLINs) and indoor residual spraying (IRS) of houses, to further accelerate declines in malaria burden (Okell, Drakeley et al. 2008; Okell, Ghani et al. 2009; Gosling, Okell et al. 2011) in areas of low and seasonal transmission (Molineaux and Gramiccia 1980; von Seidlein, Walraven et al. 2003; WHO 2010).

However, most of the infected fraction of the human population only exhibits sub-acute symptoms so they do not always obtain medical care, even where it is readily available (Lalchhuanawma and Murhekar 2012; Xu, Xu et al. 2012). These carriers of chronic, mildly symptomatic malaria infection thus act as the silent reservoir of infection because even low, often sub-patent levels of parasitaemia are sufficient to infect mosquitoes (Dunyo, Milligan et al. 2006; Bousema and Drakeley 2011; Okell, Bousema et al. 2012). Additionally, asymptomatic individuals risk developing chronic anaemia and symptomatic malaria (Prentice, Cox et al. 2010; Tiono, Ouedraogo et al. 2013). Across all levels of transmission, the majority of infections and onward transmission to mosquitoes occurs in older age groups, even though the young often harbour the highest parasite densities, because the former comprise the bulk of the human population and vector biting rates increase with host biomass and therefore age (Drakeley, Akim et al. 2000; Smith, Killeen et al. 2004; Gosling, Okell et al. 2011). In such populations, many individuals remain parasitaemic and infectious for one or more years (Bruce-Chwatt, Southgate et al. 1974; WHO 1987; Bretscher, Maire et al. 2011). Thus, in order to eliminate human-to-mosquito transmission with therapeutic drugs, all cryptic or asymptomatic infections within a human population must be successfully terminated, necessitating comprehensive

coverage of targeted communities (Okell, Ghani et al. 2009; Gosling, Okell et al. 2011; Shekalaghe, Drakeley et al. 2011; Tiono, Ouedraogo et al. 2013).

Essentially two broad strategies for taking malaria chemotherapy beyond routine case management have been described: mass drug administration (MDA) or mass screening & treatment (MSAT), historically referred to as mass blood examination (WHO 1963; Gosling, Okell et al. 2011). MDA entails administering anti-malarials to every traceable consenting member of a population, regardless of whether their malaria infection status is known or whether they exhibit symptoms (WHO 1963; WHO 2010; Gosling, Okell et al. 2011), while MSAT targets only confirmed parasitaemic individuals after parasitological testing for infection status (WHO 1963; Gosling, Okell et al. 2011). MDA necessitates comprehensive coverage and failure may accelerate the spread of drug-resistant strains selected by strong but incomplete selection pressure on the parasite populations (Greenwood 2010; Kaneko 2010). MSAT has been proposed as a better alternative because treatment is limited to those diagnostically confirmed to be infected, thus lowering treatment costs and risks of selection for resistance (Ogutu, Tiono et al. 2010; Crowell, Briet et al. 2013). However, the major limitation of MSAT lies in the challenge of detecting low-density infections, which contribute substantially to the reservoir of infection (Crowell, Briet et al. 2013). With increasing availability of more sensitive, scalable malaria rapid diagnostic tests (RDTs) that can be used by non-specialist community based health workers in low-resource settings, MSAT is now logistically feasible (Harvey, Jennings et al. 2008; Kaneko 2010; Ogutu, Tiono et al. 2010; Counihan, Harvey et al. 2012).

The concept of community-based management of malaria stems from the recognition that human resource deficits amongst clinically-trained professional cadres are commonplace, so

extending service delivery beyond centralized health facilities, by mobilizing through community health workers (CHW) will be required to improve access to appropriate management of uncomplicated malaria (WHO 2005; WHO 2012). CHWs have demonstrated the capacity to effectively diagnose malaria with RDTs and provide treatment according to the locally relevant policy and guidelines (Harvey, Jennings et al. 2008; Hawkes, Katsuva et al. 2009; Chanda, Hamainza et al. 2011; Counihan, Harvey et al. 2012). The community-based diagnosis and treatment approach has also been shown to be cost-effective (Pang and Piovesan-Alves 2001; Chanda, Hamainza et al. 2011), improve delivery of malaria case management overall (Yeung, Van Damme et al. 2008; Elmardi, Malik et al. 2009; Hamer, Brooks et al. 2012), is well accepted by communities (Ajayi, Falade et al. 2008; Mukanga, Tibenderana et al. 2010; Yeboah-Antwi, Pilingana et al. 2010) and also provides a potentially valuable population–wide platform for monitoring trends in human parasitaemia (Rutta, Francis et al. 2012).

However, this approach remains grossly underutilized and understudied, with only 15 million RDTs utilized at community level globally, mostly in India (WHO 2013). This study therefore evaluated the effectiveness of paid CHWs providing improved access to blood screening and treatment services to community members, not only when they self-reported because they felt ill, but also through regular monthly active visits to their homes.

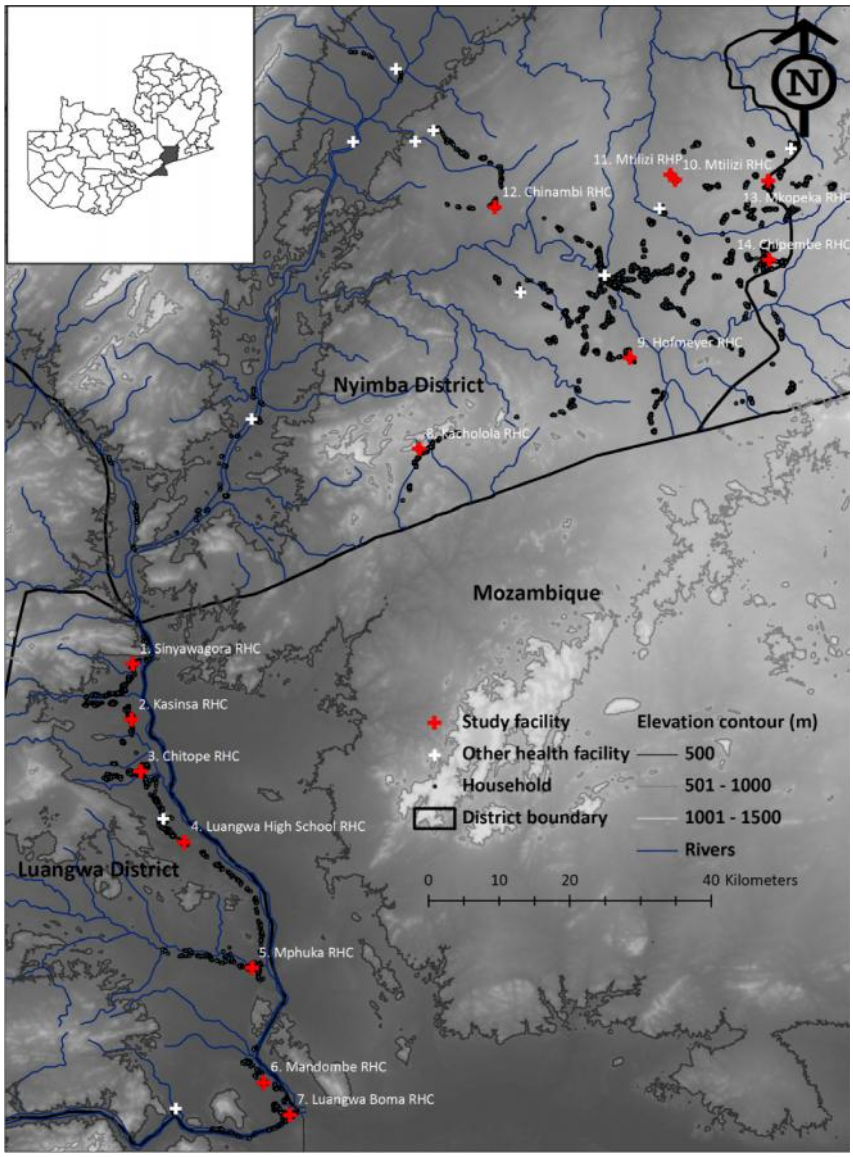
2.2 Methods

2.2.1 Study areas

The study was conducted in two adjacent rural districts of Zambia, Luangwa District in Central province and Nyimba District in Eastern province (Figure 11) where perennial transmission of

Plasmodium falciparum is predominantly mediated by *Anopheles funestus* (Seyoum, Sikaala et al. 2012; Sikaala, Killeen et al. 2013) at an entomologic inoculation rate (EIR) of approximately 70 infectious bites per unprotected person per year (Sikaala CH, Chinula D, Chanda J, Hamainza B,Mwenda M, Mukali I, Kamuliwo M, Lobo N, Seyoum A and Killeen GF, personal communication).

Figure 11.Map indicating location of health facilities and associated catchment populations enrolled in the study



Nyimba is located 350 kilometres east of the capital city Lusaka and covers an area of 10,943 km^2 with a population of approximately 86,000 residents, most of whom are involved in agriculture as their main livelihood. Nyimba is drained by three perennial rivers the Luangwa and its two tributaries the Lunsemfwa and the Lukusashi, as well several streams. Nyimba spans a range of altitudes from 400m to 1,200m along the Luangwa valley with temperatures averaging 30°C and mean annual rainfall of 1,000 mm (Kasali 2008). The population of Nyimba is served by 17 health centres and one first-level hospital located in the centre of Nyimba town.

Luangwa district is located at the confluence of the Zambezi and Luangwa rivers, 325 kilometres south-east of Lusaka. The population of the Luangwa is 25,000 residents, most of whom are settled along the river where the main economic activities are fishing and agriculture. The district covers a surface area of 3,468 km^2 and lies 350 to 500 metres above sea level. Luangwa has an average temperature in above 35°C and a mean annual rainfall of approximately 800mm (Kasali 2008). Luangwa has nine health centres, of which two have inpatient services, one of which is located in the district capital at Luangwa Boma and another at Katondwe Mission located about 40km further north.

2.2.2 Community health workers

In Zambia, community health workers (CHWs) are a formally accepted part of the health service delivery system and are selected by the communities they serve. In order to qualify, they must reside within the community, be literate in English, and be available and willing to provide basic care to their communities. Once selected, they are trained for a period of six weeks in basic primary healthcare, mainly focusing on aspects of prevention and treatment of common ailments as part of their health promotion duties. The major disease focus areas are the simple

forms of malaria, pneumonia and diarrhoea. When care is sought from the CHWs, the patient's symptoms are assessed and the CHWs follow the guidelines for diagnosis and treatment. The CHWs are also trained how to recognize clinical danger signs and facilitate referral of these cases to the HFs for further management (MOH 2010). Each CHW is attached to the HF nearest to their community and is normally responsible for approximately 500 of the resident inhabitants. The clinically-trained HF staff are responsible for supervising and mentoring the CHWs, and for supplying all their equipment and consumables. The CHWs that participated in this study were selected from available CHWs in their specific catchment areas and recruited through their supervising HFs. These recruited CHWs were remunerated at a rate of 350 Zambian Kwacha (approximately US$64) per month. A total of three CHWs per cluster (42 active CHWs) were selected to participate in the study and re-trained for a week in malaria diagnosis (use and interpretation of RDTs), treatment (prescription and dosage of AL), referral procedures (recognition of danger signs) and reporting.

2.2.3 Study design

This longitudinal study was conducted, from January to December 2011. In each study district, seven clusters of approximately 165 consenting households (average number of resident members per household was estimated at approximately six (CSO 2011)), each of which was selected and enrolled in order of proximity to the public-sector HF that defined the centre of the cluster. All members of the household were eligible to participate except pregnant women and children below six months of age. The exclusion of these groups was stipulated in accordance with national guidelines that CHWs are not allowed to manage any condition in these select groups. Each cluster received the standard Ministry of Health interventions for

malaria control in Zambia, which included long-lasting insecticidal nets (LLINs), diagnosis by either microscopy or rapid diagnostic tests (RDTs), treatment with artemether-lumefantrine (AL) and intermittent presumptive treatment with sulphadoxine-pyrimethamine. The LLINs were distributed through mass distribution campaigns in 2006, 2008 and again 2009 (Eisele, Miller et al. 2011), following a project-based distribution to selected villages in Luangwa in 2005 (Keating, Hutchinson et al. 2012) and routine delivery for pregnant women and under five children through the antenatal clinics at each of the health facilities in the study (NMCC 2011; NMCC 2012). Intermittent presumptive therapy in pregnancy IPTp) was also routinely provided through the antenatal clinics with a target of 3 doses during each pregnancy (NMCC 2011; NMCC 2012).

2.2.4 Data collection

Each CHW recorded all patient diagnoses and responses to pre-set questions in a malaria register book which detailed RDT outcomes, as well as details of age, symptoms, use of IPTp or LLINs, and treatment of their house with IRS in the previous six months. Information on access and use of interventions was collected at an individual level among all participants for those who gave consent to participate in the study. Active case detection was achieved through monthly home visits by the CHWs to each enrolled household within their assigned catchment areas, during which all consenting and assenting members were screened for malaria infection using HRP2-based RDTs from ICT Diagnostics (ICT Malaria Pf cassette test). Between these visits, passive case detection was accomplished by encouraging those who had symptoms to seek care from either their assigned local CHW or the nearest public sector HF. Individuals that

escorted patients who sought care were also tested if they were identified as members of the study clusters. During both active and passive visits, all study participants found to be positive for malaria by RDT received standard AL treatment for uncomplicated malaria. Those found to be febrile and negative by RDT through either active or passive testing systems were referred to the nearest healthcare facility. In both the active and passive systems, the CHWs reported weekly summaries via mobile phone short messaging system of the number of patients tested, RDT test results, AL and RDT stock status and the numbers of treatments of each pack size (6, 12, 18 or 24 tablets) they had dispensed.

2.2.5 Data management and statistical analysis

The principal data sources for this study were the CHW malaria registers and health facility reports sent to NMCC through the malaria rapid reporting system platform (NMCC 2011) via the Airtel^{TM} mobile phone network in the two districts. To ensure data quality and full disaggregation, the line-listed data from the malaria registers were double-entered and verified, reconciled and then cleaned following descriptive frequency analysis of the distributions of values for each variable. Statistical analysis was accomplished using SPSS version 20 (IBM) and R version 2.14.1 augmented with the lattice, Matrix and LME4 packages.

2.2.6 Association of malaria infection with age, sex, symptoms, interventions, cluster and season

Generalized linear mixed models (GLMM) were used to evaluate the association between observed RDT-determined malaria infection status as a binary dependent variable with an intercept, age category (<1, 1 to 4, 5 to 10, 11 to 14, 15 to 24, 25 to 44 and >45 years of age),

sex, access to an LLIN, use of an LLIN, having slept in a house that had been treated with IRS in the previous six months, and the three seasons (hot and wet from December to April, cool and dry from May to August, and hot and dry from September to November) as categorical independent variables. All models included household identity number nested within CHW catchment nested within cluster, as well as date of participant contact, as random effects, with the exception of one model lacking an intercept in which cluster was treated as a categorical independent variable to determine the mean diagnostic positivity of each cluster. The final model presented in Table 10 was selected by building the model through adding explanatory variables to the model one at a time, assessing improvements of the goodness of fit using log-likelihood tests and retaining only those parameters that either had significance in the model or which significantly improved the goodness of fit as per principle of parsimony (Sober 1981).

Table 10.Association of malaria infection with age, sex, symptoms, interventions, geographical location and season [i] [ii]

Category	Passive Surveillance				Active Surveillance			
	DP%[a]	n/N[b] (I)	OR [95%CI][c]	P[d]	DP%[a]	n/N[b] (I)	OR [95%CI][c]	P[d]
Overall	42.50	2236/5261(3804)	0.74[0.68,0.79]	<0.001	20.26	11851/58500 (17543)	0.25[0.23,0.27]	<0.001
Age								
<1	27.88	58/208 (145)	1.27[0.76,1.78]	0.352	12.82	204/1591 (635)	1.28[1.08,1.48]	0.015
1-4	42.11	480/1140 (768)	3.38[3.04,3.72]	<0.001	22.47	2211/9841 (2797)	3.03[2.92,3.15]	<0.001
5-10	59.10	799/1352 (935)	8.40[8.06,8.73]	<0.001	26.40	3794/14369 (3920)	4.16[4.04,4.27]	<0.001
11-14	53.55	294/549 (410)	7.05[6.68,7.43]	<0.001	25.19	1813/7198 (2265)	3.97[3.85,4.09]	<0.001
15-24	39.85	263/660 (510)	3.43[3.07,3.79]	<0.001	18.84	1690/8969 (3031)	2.26[2.14,2.38]	<0.001
25-44	27.32	244/893 (713)	1.81[1.46,2.17]	0.001	13.63	1470/10787 (3265)	1.36[1.25,1.48]	<0.001
≥45	20.41	79/387 (323)	1[NA][e]	NA[e]	11.70	617/5275 (1630)	1[NA][e]	NA[e]
Sex								
Male	44.32	1058/2387 (1699)	1[NA][e]	NA[e]	21.92	5802/26475 (7703)	1[NA][e]	NA[e]
Female	40.99	1164/2840 (2042)	0.83[0.67,0.99]	0.019	18.74	5901/31481 (8285)	0.86[0.81,0.92]	<0.001
Interventions								
ITN	39.02	1480/3793 (2738)	0.71[0.50,0.92]	0.002	17.14	7197/41978 (12295)	0.79[0.72,0.85]	<0.001
IRS	42.03	759/1806 (1224)	1.32[1.06,1.58]	0.036	15.39	2429/15788 (4323)	1.25[1.14,1.35]	<0.001
Number of preceding tests[f]	45.1[f]	2236/5261(3804)	0.92[0.90,0.93][f]	<0.001	30.7[f]	11851/58500 (17543)	0.91[0.88,0.95][f]	<0.001
Clusters								
Luangwa district								
Sinyawagora RHC	44.56	86/193 (178)	2.76[1.59,3.94]	0.090	23.23	809/3482 (1207)	1.75[0.96,2.54]	0.164
Kasinsa RHC	33.95	184/542 (454)	4.46[3.43,5.49]	0.005	13.51	684/5063 (1243)	2.39[1.61,3.18]	0.030
Chitope RHC	28.94	68/235 (179)	1.71[0.64,2.78]	0.325	17.58	1163/6614 (1030)	4.71[3.93,5.50]	<0.001
Luangwa High School RHC	44.14	644/1459 (732)	1.93[0.94,2.91]	0.191	18.57	1342/7225 (999)	2.08[1.30,2.86]	0.066
Mphuka RHC	39.10	217/555 (278)	2.79[1.72,3.87]	0.062	23.07	1249/5414 (1327)	1.59[0.81,2.37]	0.243
Mandombe RHC	36.22	205/566 (469)	1.22[0.22,2.22]	0.695	9.69	606/6252 (1206)	1.14[0.37,1.92]	0.734
Luangwa Boma RHC	22.03	39/177 (160)	1[NA][e]	NA[e]	5.90	368/6241 (1304)	1[NA][e]	NA[e]

Nyimba district								
Kacholola RHC	42.34	94/222 (194)	10.76[9.62,11.91]	<0.001	24.65	1198/4860 (1038)	6.87[6.09,7.66]	<0.001
Hofmeyer RHC	51.99	157/302 (280)	46.94[45.87,48.02]	<0.001	47.23	1050/2223 (1394)	13.25[12.46,14.04]	<0.001
Mtilizi RHC	66.94	164/245(193)	11.34[10.23,12.46]	<0.001	36.51	782/2142 (847)	5.93[5.16,6.70]	<0.001
Mtilizi RHP	50.00	19/38 (37)	9.24[7.89,10.58]	0.001	22.72	573/2522 (1227)	5.08[4.36,5.80]	<0.001
Chinambi RHC	57.35	39/68 (59)	10.44[9.01,11.86]	0.001	29.11	560/1924 (947)	6.90[6.02,7.78]	<0.001
Mkopeka RHC	54.90	112/204 (179)	12.03[10.89,13.16]	<0.001	35.41	1004/2835 (753)	6.27[5.43,7.11]	<0.001
Chipembe RHC	45.71	208/455 (333)	7.56[6.50,8.61]	<0.001	27.19	463/1703 (897)	5.89[5.07,6.71]	<0.001
Season								
Hot & wet (Dec – April)	50.47	1082/2144 (1865)	8.11[7.78,8.44]	<0.001	20.79	4615/22195 (9843)	4.92[4.71,5.13]	<0.001
Cool & dry (May - Aug)	46.26	929/2008 (1749)	4.99[4.67,5.31]	<0.001	25.38	6125/24137 (12025)	4.00[4.71,5.13]	<0.001
Hot & dry (Sept – Nov)	24.72	224/906 (858)	1[NA][e]	NA[e]	9.05	1091/12056 (7745)	1[NA][e]	NA[e]

[i] a – Diagnostic positivity, b – (n – Number RDT positive, N – Total number of testing events), I – number of individuals that participated , c – odds ratio with 95% confidence intervals, d – p-value,NA [e] –Not applicable /reference group,f – An integer continuous covariate so the diagnostic positivity presented represents that for the first visit calculated as the intercept of the model and the odds ratio presented represents proportional change of diagnostic positivity per additional preceding test.

[ii] The association of malaria infection with age, sex, symptoms, interventions, cluster, number of tests conducted per individual and season was determined using GLMM; with observed malaria RDT determined status as a binary dependent outcome with the independent categories of age, sex, symptoms, access and use of ITNs and/or IRS and seasons. The models included date and individual nested within CHW catchment nested within cluster as random effects except for one in which cluster was treated as a categorical variable to determine the effects of each cluster. Symptoms were removed from the final model for logical reasons, as they are effects and not causes of infection. The final model consisted of age, sex, access and use of ITNs and/or IRS, season, number of tests conducted per individual and geographical location as the determinants of malaria infection.

2.2.7 Association of malaria infection with clinical symptoms

While several symptoms were indeed positively associated with *P. falciparum* infection, these were excluded from the final model that captures the effects of most of the variables presented in Table 10 simply because they are an effect, rather than a cause, of malaria so they cannot be regarded as underlying independent determinants of malaria risk. Instead, the association of these symptoms with malaria infection was assessed as follows, using separate GLMMs treating the presence or absence of each symptom as the binary dependent variable and malaria infection diagnostic result as a binary, categorical, independent explanatory variable (Table 11). Each model also included age, sex and season as additional categorical independent variables and individual study participants nested within CHW catchment, nested again within clusters as random effects. Each model was selected to include only variables, which were significant or significantly improved the goodness of fit, as described above for the models of malaria infection risk described in Table 11.

Table 11.Association of symptoms of illness with RDT positivity, age and seasonality [iii] [iv]

Symptom	Passive surveillance		Active surveillance	
	OR [95%CI][c]	P[d]	OR[95%CI][c]	P[d]
Fever	5.98[5.79,6.16]	<0.001	14.63[14.55,14.71]	<0.001
History of fever	2.16[1.93,2.40]	<0.001	2.86[2.77,2.95]	<0.001
Headache	2.31[2.15,2.47]	<0.001	6.83[6.77,6.90]	<0.001
Cough	0.74[0.58,0.90]	<0.001	1.85[1.77,1.93]	<0.001
Diarrhoea	1.47[1.15,1.79]	0.017	2.04[1.86,2.21]	<0.001
Vomiting	3.01[2.74,3.27]	<0.001	6.61[6.43,6.80]	<0.001
Chest pain	0.88[0.56,1.20]	0.447	1.47[1.31,1.64]	<0.001
Breathing problems	1.78[1.40,2.16]	0.914	7.99[6.47,9.52]	0.008
Other symptoms	0.87[0.46,1.28]	0.510	0.98[0.81,1.15]	0.809

[iii] c – odds ratio with 95% confidence intervals, d – p-value.

[iv] The association of clinical symptoms with malaria infection as determined by RDT positivity was determined by logistic regression with GLMMs controlling for age and seasonality as fixed effects and for date, individual nested in CHW and CHW nested in cluster location as random effects.

2.2.8 Cost per case diagnosed and treated

The approximate cost of diagnosis and treatment of malaria parasite infection through either the CHW or health facility systems was calculated separately. For the CHWs, this was split into two arms: active and passive. The costs incorporated into our calculations were personnel time, RDTs, microscopy where available, anti-malarial drugs and sundry maintenance, transport and consumables. Personnel time included an assumed 30% full time equivalent (FTE) contribution to malaria diagnosis and treatment by all personnel based at the HF and the CHWs were assumed to have allocated 90% and 10% FTE contributions to the active and passive service delivery arms. The cost of diagnosis was estimated by the multiplying the unit cost (US$0.31 per RDT and US$1.30 per microscopy test) (Uzochukwu, Obikeze et al. 2009; USAID 2012) of each diagnostic method by the number of tests done. The cost of drugs was estimated by multiplying the total number of treatments dispensed by the average Zambian malaria programme unit cost of US$1.55. Annual remuneration costs for all the CHWs are calculated based on their monthly remuneration of US$71.58 (ZMW 350) per month per CHW and the number of person months for which they were engaged over the course of the study. Annual costs of remuneration for health facility staff were collated from the medium term expenditure frameworks of the two district medical offices, totalling US$281,150 (ZMW 1,377,600). Total personnel and other facility maintenance costs were calculated by dividing the total number of times all study participants/patients were appropriately diagnosed and treated. Subsequent estimates of cost per case diagnosed and treated were made by adding personnel time, costs of

diagnosis and treatment per case and facility maintenance costs. It was not possible to estimate capital costs for either the CHW or health facility systems, so these were excluded from these calculations. Calculations of directly observed costs were based on those recorded over the six months from April to September 2011 when all the CHW and HF were fully functional but before IRS was introduced as a possible confounding effect. A projected per capita cost estimate for a year was then estimated by doubling these six month cost estimates. Furthermore a projected potential cost per capita for implementing such an equivalent CHW programme but complete compliance of the catchment population with monthly testing was also calculated. Annual per capita cost estimates for the actual and enhanced-participation scenarios were also calculated for scenarios in which transmission is reduced to levels where treatment costs were negligible.

2.2.9 Population attributable fraction of symptoms to malaria infection

The proportion of cases of each specific clinical symptom identified within the population by the CHWs that could be attributed to RDT-detectable malaria infections was estimated as follows (Smith, Schellenberg et al. 1994):

$$\lambda = p(R-1)/R$$

Where λ is the attributable fraction, p is the diagnostic positivity and R is the odds ratio of symptom associated with malaria infection. This formula was separately applied to both the active and passive surveillance data obtained through the CHWs.

2.3 Ethical approval

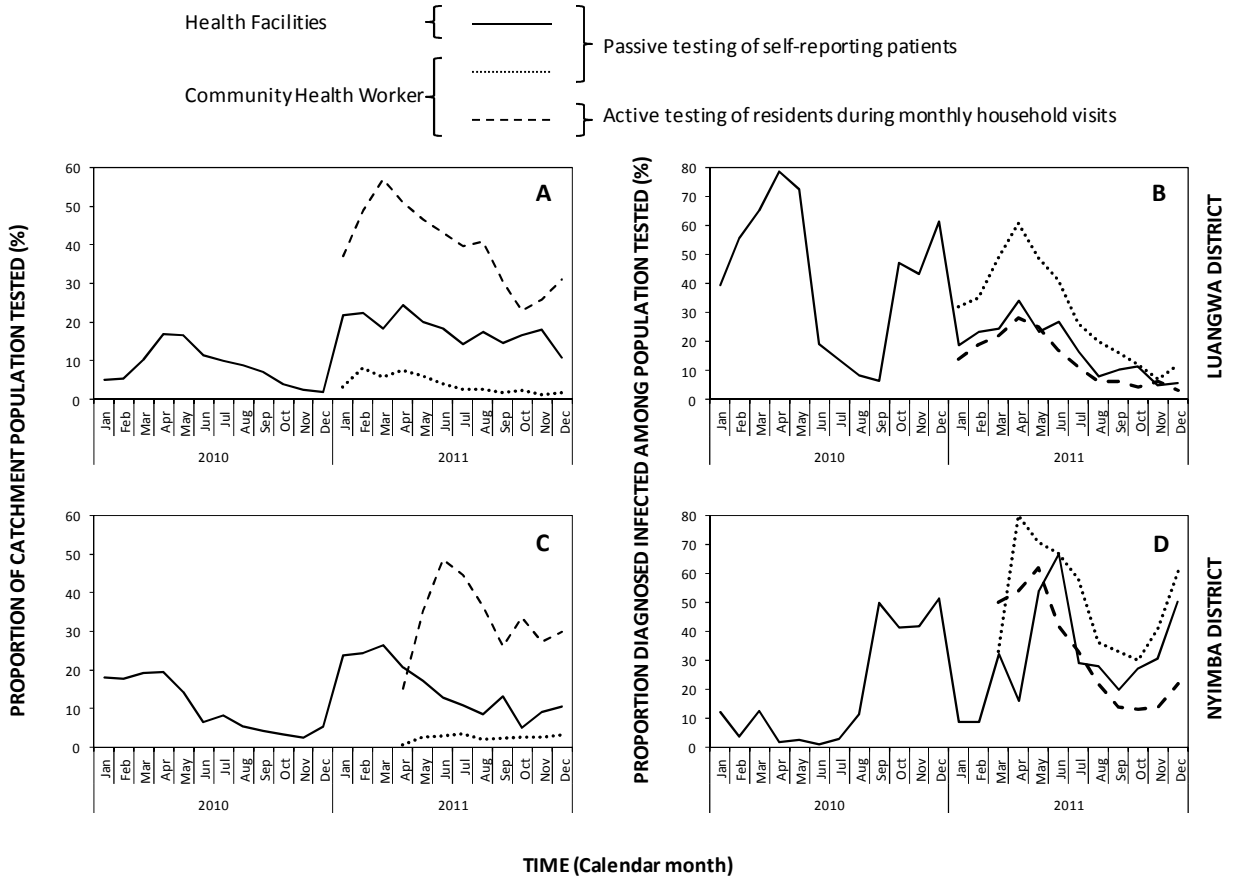
Ethical approval was obtained from the University of Zambia, Biomedical Research Ethics Committee (Reference 004-05-09) and the Research Ethics Committee of the Liverpool School of Tropical Medicine (Approval 09.60). Authority to conduct the study was also obtained from the Ministry of Health in Lusaka, Zambia.

2.4 Results

2.41Rates of participation and testing through health facilities and community health workers

From a combined catchment population of 77,754 for Luangwa and Nyimba, there were approximately 7, 5791 outpatients visits attended to at the 14 enrolled HFs in 2011. Seasonal patterns of testing for malaria at HFs were roughly similar and peaked at approximately the same time of the year in both districts, with an overall mean rate of testing for malaria infection of 56%. Mean monthly testing rates were 36.9% (range = 0% to 100%) for patients in Luangwa and 69.3% (range =0% to 100%) in Nyimba (Figure 12A and C) so the proportion of patients in which malaria was suspected and tested for was approximately twice as high in Nyimba as Luangwa.

Figure 12.Proportion of catchment population tested (A and C) and diagnostic positivity for malaria infection among residents (B and D) in Luangwa (A and B) and Nyimba (C and D)



A total of 14 population clusters centred around these HFs, with enrolled populations that ranged from 753 to 1,243, were established and followed up over a period of one full calendar year in 2011 from January to December for Luangwa District, and April to December for Nyimba district. A total of eight and 12 monthly rounds of active household visit surveys were conducted by each CHW in Nyimba and Luangwa, respectively, to test and treat all consenting residents within these population clusters. A total population of 17,543 individuals participated by consenting to testing during the active monthly household visits, of whom 20% were under the age of five. During the same period a total of 3,804 individuals, of whom 24% were under

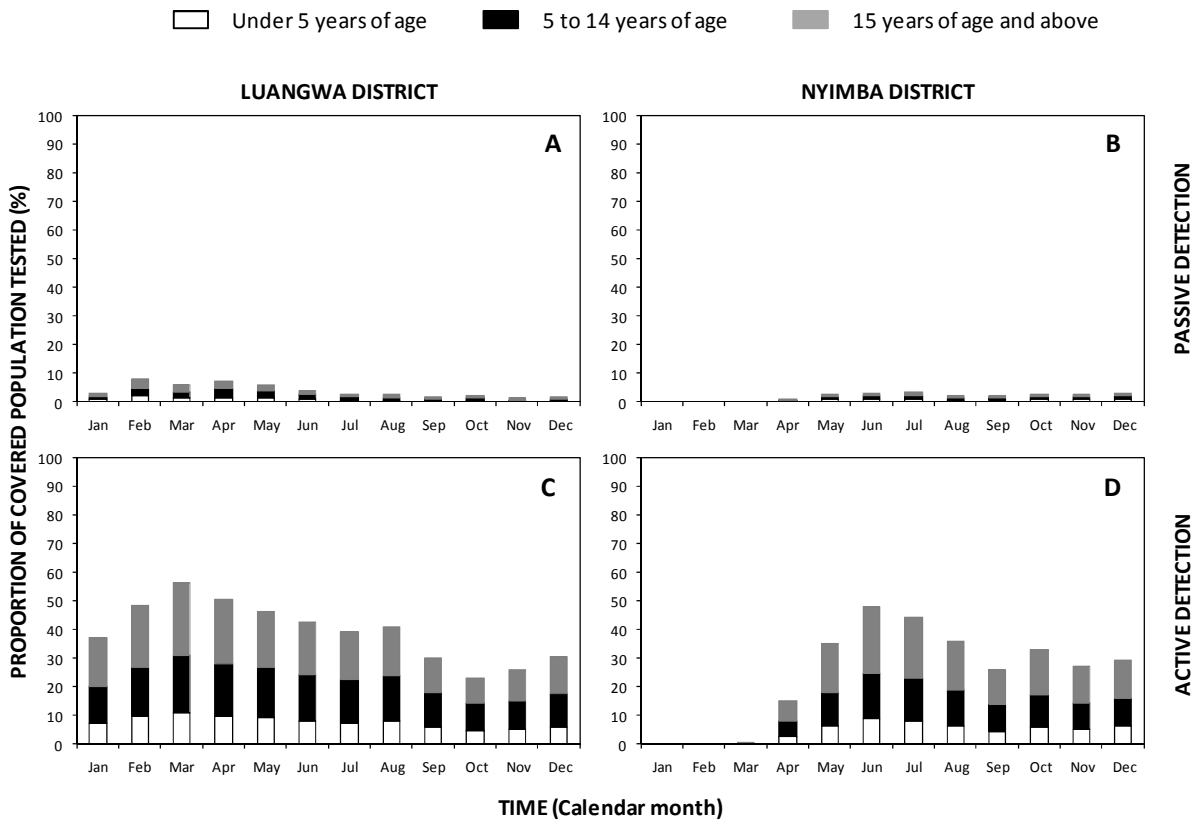
the age of five, sought care and were tested when they self-reported to the CHW, the results of which were then recorded as a passive surveillance indicator (Table 10). Slightly more females participated in both the active and passive visits than males (Table 10), presumably because females were more accessible during the active household visits, and sought care more frequently from CHWs through their passive service provision role, than males because they spent most of their time at home (Diallo, Nadam et al. 2012). The CHWs referred a total of 577 and 631 patients detected through passive and active surveillance systems, respectively, to the health facilities for further management. The main reasons provided for referral were the inability by the CHWs to manage some of the conditions presented to them by the patients, RDT positive results for pregnant women, lack of patient improvement while on malaria treatment and insufficient AL stocks in the hands of that CHW for him or her to provide treatment directly. Over the same period 42,389 and 932 suspected malaria cases were tested by RDT and microscopy, respectively, and 20,794 were treated for malaria through the health facilities in these clusters.

Introduction of CHWs for screening and treating of residents captured a higher proportion of the populations they covered than the HFs they were based near to, overwhelmingly through active monthly visits to the household rather than passive reporting (Figures 12A and C). This can be readily explained by the fact that these 14 HFs covered a total catchment population of 77,754 people, equivalent to 58.8% of the combined population of the two districts, while a total of 42 CHWs were assigned a total of 17,543 people or only 13.3 % of the combined population of the two districts. The introduction of this CB extension of primary healthcare services had no obvious impact upon attendance rates at health facilities in Luangwa, and a

simultaneous drop in HF attendance in Nyimba resembled seasonality patterns from the previous year (Figures 12A and C). Thus no obvious impact of CHW services upon the rates of reporting of suspected malaria to the local health facility was apparent.

The proportion of enrolled participants who actually consented to testing during active household visits rose rapidly and then peaked at approximately half during March in Luangwa (Figure 12A) and June in Nyimba (Figure 12C). The proportion of enrolled individuals who sought care and were passively tested for infection by CHWs was generally far lower, only exceeding 5% in an early peak in Luangwa but not in Nyimba (Figure 12A and C). In both CHW survey arms, older children (5-14 years) and the adults (≥15 years) comprised the highest proportion of those tested while young children (<5 years) comprised a minority, presumably because they comprise a small demographic proportion of the overall population (Figure 13). The age distribution of participants in testing and treatment were approximately comparable to data from the nationally representative malaria indicator surveys (MOH 2010; MOH 2012) but rigorous assessment of participation biases would require comparison with detailed census data.

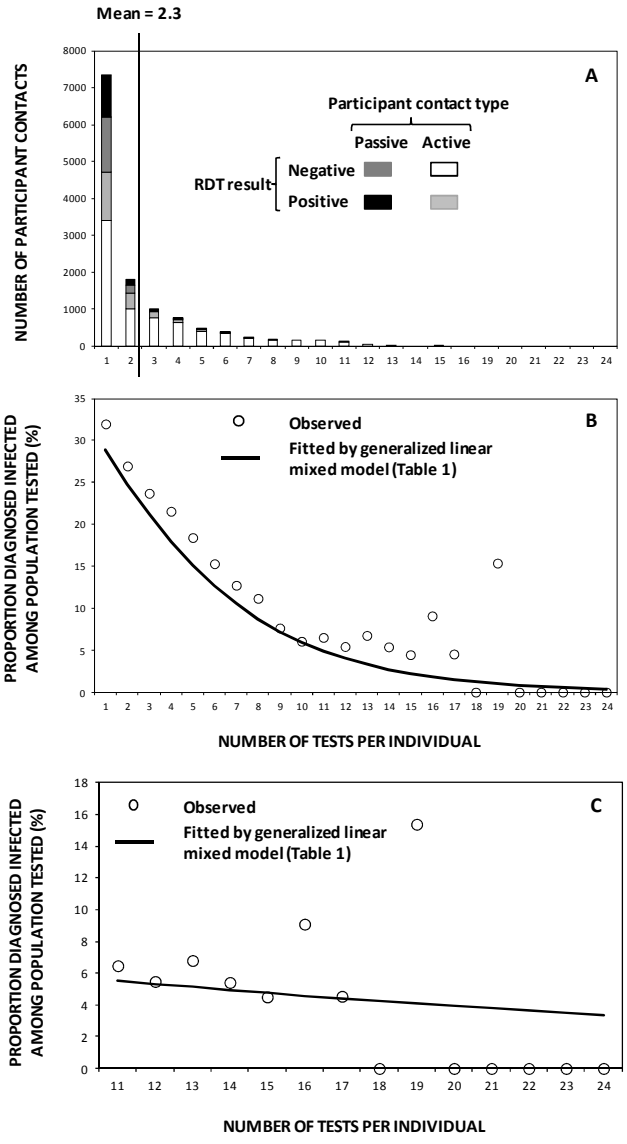
Figure 13.Age and proportion of covered population tested for malaria infection each month as contacted passively (A and B) and actively (C and D) by community health workers in Luangwa (A and C) and Nyimba (B and D)



Overall, less than 20% of the enrolled population was tested more than once by CHWs through either the active or passive surveys but the mean number of tests per individual (2.3) was much higher than the median (1 [range=1 to 24]) because the frequency distributions for the numbers of times individuals were tested through either mechanism were highly skewed (Figure 14A). The average number of tests per individual participant through both surveillance systems combined was understandably somewhat higher in Luangwa (4.4) than in Nyimba (3.2) because they were operational for 12 months in the former but only eight months in the latter. The number of passive patient contacts per individual through self-reporting to CHWs ranged from 0 to 24 times in both districts over the course of the study period, with a median of only

once, and an average of 1.2 times per individual. The number of active patient contacts per individual through monthly household visits by CHWs ranged from 1 to 12 times in both districts over the course of the study period, with a median of only once, and an average of 2.6 times per individual.

Figure 14.Frequency histogram of the number of study participant contacts for each total number of preceding malaria infection tests by community health workers per individual study participant (A), the relationship between the proportion of those participants diagnosed as being infected and the cumulative number of diagnostic tests for malaria infection per individual participant for all participants (B) and for the minority subset who had more than 10 cumulative diagnostic tests (C)



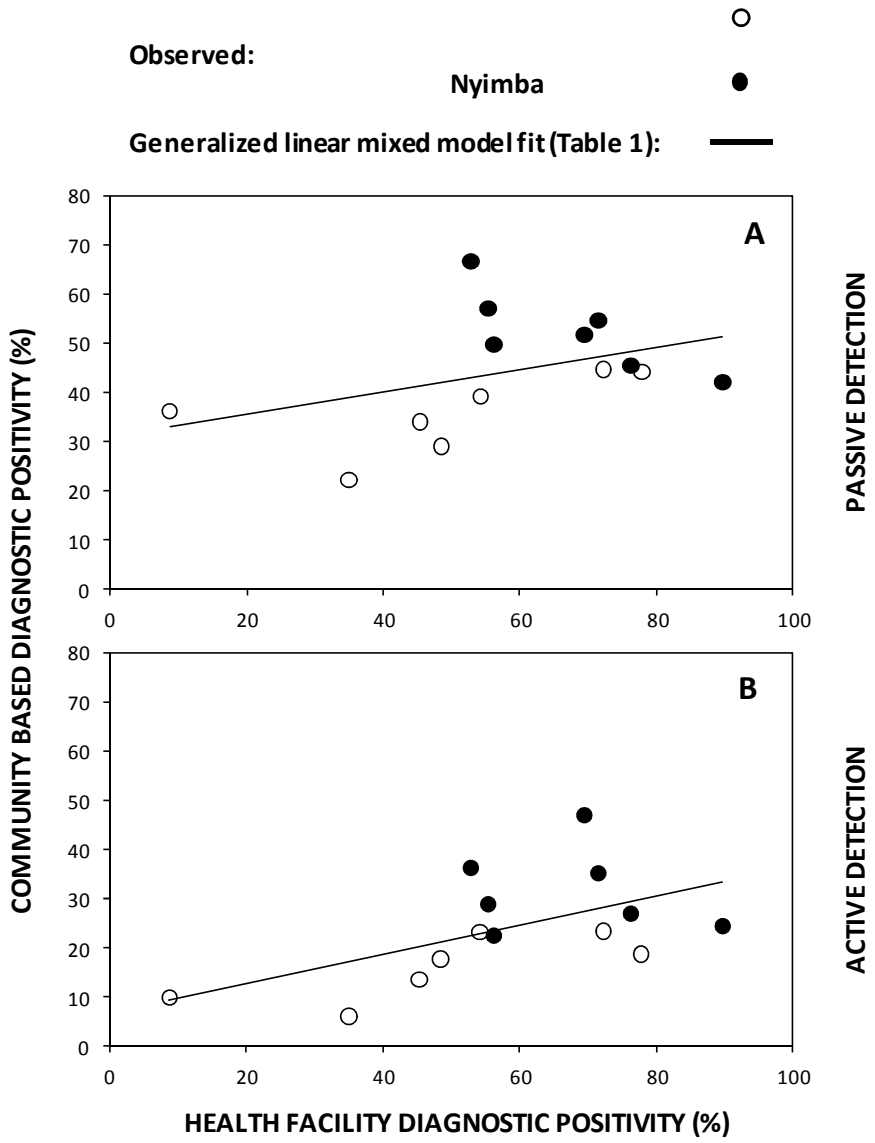
2.42 Comparative rates of malaria infection diagnosis by health facilities and community health workers

Overall, the majority of occasions when residents sought care through the passive detection systems of the HFs (44%) and CHWs (57.5%) did not have malaria parasite infections (Figure 2B and D). Nevertheless from the total study population, more than 14,000 uncomplicated malaria infections were identified by CHWs over the course of 2011 with the vast majority (84.1%) of these being detected through the active household visits rather than through self-reporting to a CHW for passively offered service (Table 10). While the HFs detected even more malaria infections over the course of the same year (37,204), these were drawn from much larger catchment populations so the overall rate of detection of cases of malaria infection per head of population covered was highest for the active surveys of the CHWs, followed by passive surveys at the HFs and CHWs (1.16, 0.40 and 0.14 diagnostically confirmed malaria infections per person per year) (Table 12). The overall incidence rates for detected and diagnostically confirmed malaria infections, broken down by district were 1.13 and 1.51 cases per person per year for Luangwa and Nyimba, respectively.

The overall diagnostic positivity, or proportion of diagnostic tests which confirmed malaria infection, was generally far higher among patients seeking diagnosis and treatment through the routine services offered passively by either the HF or the CHWs than among those screened actively through the monthly household visits of the CHWs (Table 10, Figure 12B and 12D). This was presumably because self-reporting patients obviously present a sample that is strongly biased towards those who are actually ill at the time. Diagnostic positivity observed at the healthcare facilities fluctuated seasonally, peaking at the end of the rainy season in April and

May and reaching its lowest point at the end of the dry season in September and October, with a mean of 17.2% in Luangwa (range 4.8% to 34.1%) and 31.0% in Nyimba (range 8% to 67%). The wide range of diagnostic positivity in these study sites is comparable to what has been observed in other malarious parts of Zambia (MOH 2010; MOH 2012) and may be considered reasonably representative of the range of transmission across most endemic parts of the country. Considerable geographical heterogeneity was also observed in the diagnostic positivity rates obtained through the CHWs, especially those from their active household surveys that were less biased towards infected individuals, with the lowest being in the two most urbanized clusters in the Luangwa Boma, the district capital at the south of Luangwa District (Table 10). Seasonal patterns of diagnostic positivity at HFs, expressed as the proportion of all patients tested diagnostically with RDTs or microscopy who were confirmed to be infected, differed appreciably between the two districts with no particularly consistent similarities or dissimilarities from 2010 to 2011 (Figure 12B and C). The seasonality patterns of diagnostic positivity among residents tested by CHWs closely paralleled those tested by HFs in Luangwa and even preceded them by a month or two in Nyimba (Figure 12) where access to HFs was is more challenging, especially during the rains. Furthermore, the estimated mean diagnostic positivity of both passive and active surveillance of the CHWs were positively correlated with those observed through passive surveillance at the HFs across both districts (Figure 15) and were, therefore, highly consistent with each other as measures of malaria infection. Interestingly, diagnostic positivity rates reported by CHWs were much more closely associated with those reported by HFs in Luangwa than in Nyimba (Figure 15) where access to HFs is far more difficult for this more scattered population.

Figure 15.Association of diagnostic positivity for malaria infection among patients attending health facilities with diagnostic positivity recoded by community health workers through passive (A) and active (B) participant contacts



2.43 Demographic, geographic and vector control determinants of malaria infection burden

Malaria infection among residents tested by the CHWs was associated with age, sex, season, geographical location and coverage with vector control in the form of LLINs and IRS, as well as the number of times each individual had been tested previously and, in most cases, treated for

malaria (Table 10). Malaria infection burden among patients self-reporting to the CHW through the passive surveys peaked in exactly the same age category as those tested during their active monthly household visits (Table 10), confirming that essentially the same population was being monitored by both systems. Risk of infection peaked in older children and was least among infants and the oldest adults and females were slightly at less risk than males (Table 10). Malaria infection probability was far higher in the hot and wet season, and the cool and dry season, than in the hot and dry season (Table 10).

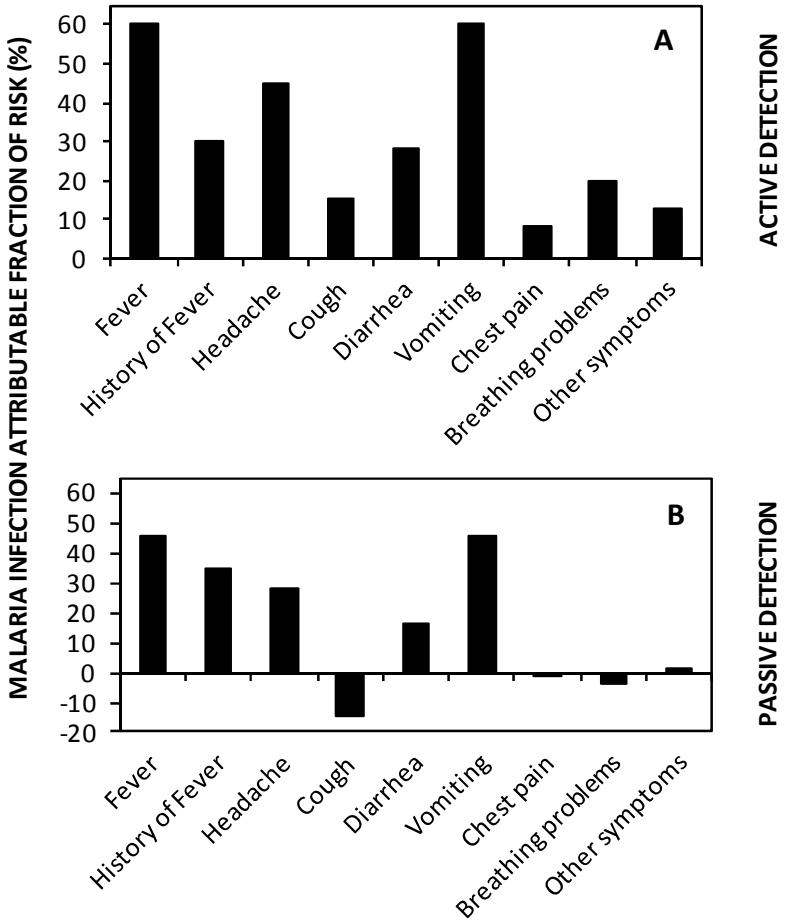
The majority of study participants who reported using an LLIN when they were tested by a CHW (72% (2738/3804) for passive contacts and 70.1% (12295/17543) for active contacts) had lower diagnostic positivity, consistent with the protective effect expected. However, individuals living in the 9 clusters that were treated with IRS towards the end of 2011, whose houses were actually sprayed, had higher diagnostic positivity, even when time, location and household effects were controlled for (Table 10). Rather than conclude that IRS actually increases malaria risk, it may be presumed that these estimates from best-fit models are probably a spurious artefact arising from endogeneity caused by logically and deliberately biased deployment of IRS to areas within each cluster with highest disease burden by the District Medical Offices tasked with implementing malaria control activities. Specifically, the IRS teams in both districts deliberately started spraying the most isolated villages at the fringes of the enrolled population clusters first so that these could be completed before arrival of the rains and associated limited access.

2.44 Association of malaria infection with clinical symptoms of illness

A substantial proportion of all residents who reported no symptoms whatsoever were found to carry malaria parasite infection; 12% (5123/42881) and 27% (286/1062) of the active and passive contacts, respectively. Discussions with CHWs confirmed that essentially all asymptomatics who were tested through passive contacts were those friends, relatives and caregivers who had escorted a patient to see the CHW and were also tested during such a visit. The overall number and proportion of all patient contacts which were classified as asymptomatic malaria infection detected by CHWs was approximately twice as high among residents tested through in active surveillance [8.8% (5,123/58,500)] rather than passive [5.4% (286/5,261)]. The proportion of confirmed malaria cases identified through active monthly surveys by CHWs who apparently exhibited no symptoms whatsoever was only 43% (5123/11851), confirming that most detectable malaria infections are chronic, but nevertheless associated with substantial, if non-severe, symptoms at the time they are surveyed. Malaria infection was associated with all specifically assessed symptoms, and even with the "other symptoms" category among residents screened during active household visits by CHWs, and most of these associations could also be detected using data collected passively from self-reporting patients (Table 10). The symptoms most strongly associated with malaria infection were fever, a history of fever in the last month, headache and vomiting, with the former being the highest reported in both surveillance arms (Table 10). The reverse was also found to be true as all symptoms were associated with RDT-detected malaria infection in both active and passive CHW surveys, except for breathing problems and sundry other symptoms, using the passive surveys data with limited sample size (Table 12). More than half of all cases of fever and

vomiting, and more than a quarter of all cases history of fever, headache and diarrhoea, among residents tested during active CHW visits to their households were attributable to malaria infection (Figure 16). The positive association of cough with detectable infection in the active visits, contrasting with a negative association in passive surveys that is more difficult to rationalize (Tables 10 and 12), may reflect an interactive effect upon patient reporting rather than the manifestation of the symptom itself, resulting in under-reporting of cough among patients reporting to CHWs because they were infected with malaria.

Figure 16.Fractions of risk for reported clinical symptoms which are attributable to malaria infection detected by community health workers through active (A) and passive (B) contact events

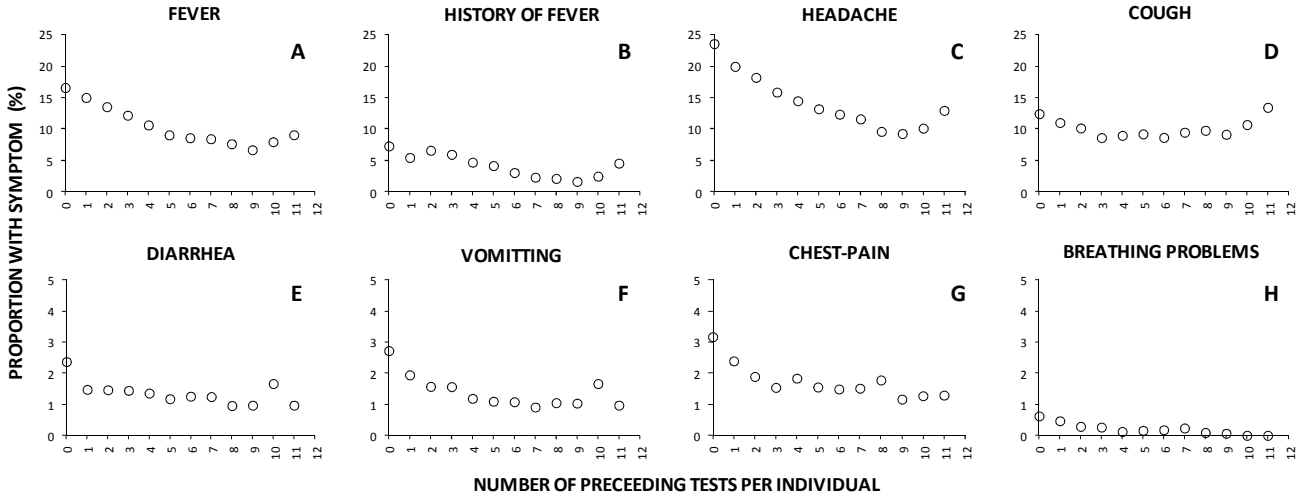


2.45 Association of malaria infection status and clinical symptoms of illness with malaria testing frequency

Only one study participant was tested 24 times by CHWs during both active and passive visits combined. Diagnostic positivity for malaria was negatively associated with the number of times that participant had been previously tested, and in most cases, treated (Table 10, Figure 14B). Sub-analysis of this relationship among the minority subset of 2376 participants who had more than 10 cumulative diagnostic tests for malaria infection (Figure 14C) confirmed this association (P=0.07) so diagnostic positivity does appear to decline continuously and indefinitely with intensity of testing and treatment. For the small proportion of individuals who received 12 tests during a single calendar year, diagnostic positivity through active surveys was estimated to be 5.4% in their last test, compared with 32% for those tested for the first time. Diagnostic positivity was therefore proportionally 83% lower for the 12^{th} test of those who participated at least that often, and this trend towards lower diagnostic positivity continued downward for who were tested even more frequently (Figure 14B). A similar phenomenon was observed with regard to manifestation of symptoms, with much lower rates of occurrence observed for all reported clinical symptoms except cough among individuals who had been repeatedly tested and treated for malaria (Figure 17). Interestingly, even when RDT-diagnosed infection status is accounted for by adding this independent variable to the models depicted in Figure 17, the number of times an individual had been previously tested remained predictive of fever ($P < 0.001$), history of fever ($P < 0.001$), headache ($P < 0.001$), diarrhoea ($P < 0.001$), chest pain ($P < 0.001$), breathing problems, vomiting ($P < 0.001$) and other symptoms ($P < 0.001$). Taken at face value, these observations appear to suggest that screening and treatment may not only

reduce probability of infection with malaria at detectable parasite densities (Figure 14B), but also persistent sub-patent infections that contribute to symptoms of illness despite parasite densities too low to be detected (Figure 17). However, testing frequency was not assigned to distinct treatment groups or experimentally controlled in any other way so these associations are purely observational and causality cannot be directly inferred. For example, these observations might also be explained by co-association of testing frequency, malaria infection and symptoms of illness with unrecorded health-conscious behaviours that are not accounted for in the model described in Table 10. Indeed, the test results at first active visit did seem to slightly influence of the number of subsequent tests so the trends observed in Figures 14B and 17 should be cautiously interpreted: individuals whose first test yielded a negative result had a slightly greater mean number of tests over the course of the study (4.65±0.03 versus 4.11±0.05, P<0.001 by GLMM).

Figure 17.Relationship between the proportion of participant contacts with community health workers in which they experienced fever (A), history of fever (B), headache(C), cough (D), diarrhoea (E), vomiting (F), chest pain (G) and breathing problems (H) and the cumulative number of preceding diagnostic tests for malaria infection per individual participant



Despite the impressive negative association of repeated testing and treatment with the probabilities of infection and symptoms among individuals, no dramatic impact upon these parasitological and clinical outcomes were obvious and a total incidence rate of 1.7 detected infections per head persisted (Figure 12,Table 12). As illustrated in Figure 14A, the mean number of times participants were tested was 2.3, so even if the relationship between number of preceding tests and diagnostic positivity is causal, rather than merely co-associated, too few participants were tested regularly enough for any dramatic impacts to be observed at population level: Those who had the mean number of tests would be expected to maintain a mean diagnostic positivity of 23.7% at the end of the year, only 17.9% lower proportionally than those tested only once (Figure 14B). Even if direct impact of testing and treatment upon infection probability is assumed, and comprehensive monthly testing could be achieved in the future, this would still be expected to leave sufficient levels of parasitaemia at population level to maintain endemic transmission (Figure 14A and 14B) (Smith and Hay 2009). The intensity of persisting transmission reflected in the measured EIR, despite considerable levels of vector control, is also reflected in measured rates of re-infection among humans: Over the course of the study period, CHWs detected as many as eight malaria infections in a single study participant detected through passive surveillance, while the maximum was nine infections in single participant as detected through active surveillance (Figure 14A).

2.46 Adherence of CHWs to diagnosis and treatment guidelines

Patterns of diagnosis and treatment differed between patients seeking care at HFs or from CHWs, as well as residents consenting to being tested by CHWs during their active household

visits (Figure 18, Table 12). Adherence to national guidelines for diagnosis and treatment were generally good among CHWs with 78% of all contacts that resulted in an RDT test being followed by an appropriate decision to treat or not (Figure 18). The remainder was primarily accounted for by diagnostically confirmed cases of malaria infection that could not be treated because the CHW had run out of the drug, and also because small proportions of patients were treated in the absence of a diagnostic result or despite a negative diagnostic result (Figure 18, Table 12). More worryingly, only 53% of patients attending HFs were tested and then treated or not treated appropriately to the test result, primarily because a substantial proportion were neither tested nor treated but also because small proportions were treated in the absence of a test or despite a negative test (Figure 18, Table 12). So, consistent with reports from other settings in Zambia (Harvey, Jennings et al. 2008; Chanda, Hamainza et al. 2011; Littrell, Gatakaa et al. 2011; Counihan, Harvey et al. 2012) and beyond (Pagnoni, Convelbo et al. 1997; Yeung, Van Damme et al. 2008; Thang, Erhart et al. 2009), the CHWs had greater adherence to policy guidelines on treatment practices in relation to diagnostic test results than specialist staff at HFs (Figure 18, Table 12). The proportion of patient contacts resulting in a negative RDT test result, or assessed only clinically without a confirmatory diagnostic test, that were treated with AL were both at least six times higher at HFs than CHWs (Figure 18). CHW provision of treatment to patients with a negative test result was twice as high in the active compared with passive but in both cases this occurred only very rarely. All confirmed cases of malaria infection reporting to health facilities received treatment with AL and the same was true for 92% of those identified passively by CHWs but only 59% among those identified through active household visits (Table 12, Figure 18). In most cases where CHWs did not provide treatment for

RDT-positive patients, this was because they lacked AL and those patients were referred to the HFs to collect curative drugs.

Figure 18.Community health worker and health facility staff treatment and diagnostic practice in relation to national guidelines

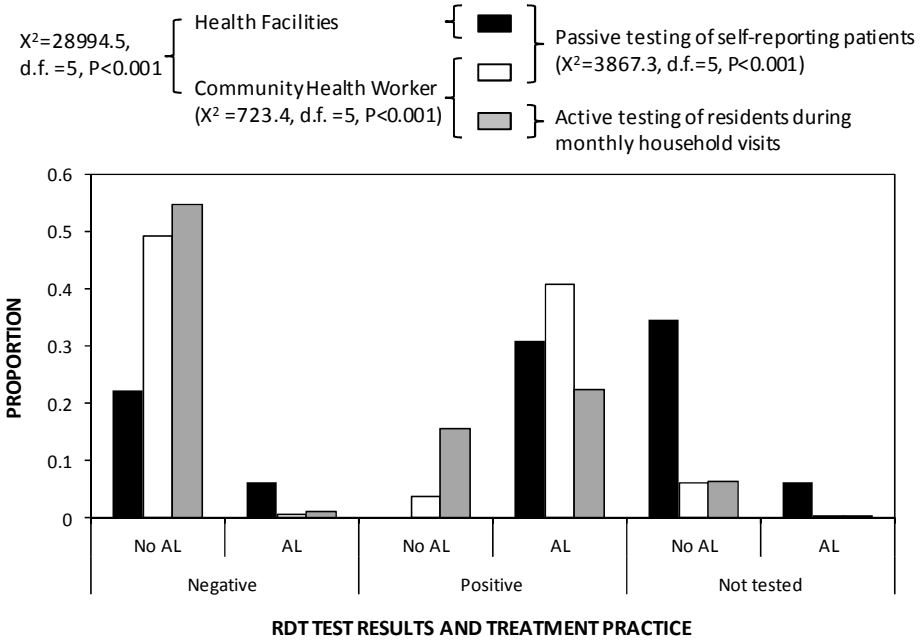


Table 12.Compliance to diagnostic and treatment policy - Community based and Facility Based (April to September 2011) [v]

		Facility based			Community based									Community & facility based		
		Passive			Passive			Active			Passive and active			Passive and active		
Al dispensed		**No**	**Yes**	**Total**	**No**	**Yes**	**Total**	**No**	**Yes**	**Total**	**No**	**Yes**	**Total**	**No**	**Yes**	**Total**
Tested by	**test result**															
RDT	**negative**	11,024	3,046	14,070	1,518	16	1,534	24,878	517	25,395	26,396	533	26,929	374,20	3,579	**40,999**
	positive	0	15,306	15,306	111	1,260	1,371	7,012	10,149	17,161	7,123	11,409	18,532	7,123	26,715	**33,838**
Microscopy	**negative**	204	55	259	n/a	n/a	n/a	n/a	n/a	n/a	n/a	n/a	n/a	204	55	**259**
	positive	0	262	262	n/a	n/a	n/a	n/a	n/a	n/a	n/a	n/a	n/a	0	262	**262**
RDT or microscopy	**negative**	11,228	3,101	14,329	1,518	16	1,534	24,878	517	25,395	26,396	533	26,929	37,624	3,634	**41,258**
	positive	0	15,568	15,568	111	1,260	1,371	7,012	10,149	17,161	7,123	11,409	18,532	7,123	26,977	**34,100**
Not tested		17,541	3,158	20,699	185	3	188	2,845	18	2,863	3,030	21	3,051	20,571	3,179	**23,750**
Total		**39,997**	**40,496**	**80,493**	**3,443**	**2,555**	**5,998**	**66,625**	**21,350**	**87,975**	**70,068**	**23,905**	**93,973**	**110,065**	**64,401**	**174,466**

[v] Treatment practice based on confirmatory RDT result was assessed between April and September as this was the period of time when all the CHW based active & passive surveillance and the health facility based passive system were all fully functional.

2.47 Testing and treatment service delivery, cost and cost-effectiveness

As detailed in Table 13, expenses associated with personnel and commodities are important components of the overall cost of providing malaria diagnosis and treatment services through either HFs (49%) or CHWs (25%). Sundry transport and maintenance expenses also contributed substantially to the overall cost of the HFs but not the CHWs. The overall cost of HFs was three-fold higher than the CHW approach but was less expensive than CHWs per head of population covered, because the HFs were assigned to cover far larger catchment populations while the CHWs simply provided far more frequent testing and treatment service to each resident through the active surveys conducted through monthly household visits (Table 13). If the same frequency of testing were implemented in a pre-elimination scenario (WHO 2007; WHO 2007) where improved vector control reduced drug treatment requirements to negligible levels, this would save only a quarter of the overall costs of providing these HF or CHW services. Because of the lower personnel, transport and maintenance costs of the CHWs, combined with their better compliance with national guidelines, both the passive and active services provided by the CHWs were almost twice as cost-effective in terms of cost per diagnostically confirmed case identified and treated.

Table 13.Observed and potential cost-effectiveness of cases appropriately diagnosed and treated [vi vii viii ix]

	Health Facility	Community based		
Directly observed process indicators over 6 months		**Passive**	**Active**	**Total**
Covered community based cluster or facility catchment	77754	17543	17543	17543
Diagnostic tests carried out over six months	29897	2652	42556	45208
Diagnostically confirmed and treated cases of malaria infection over 6 months	15568	1260	10149	11409
Diagnostically confirmed and treated malaria case per head population over 6 months	0.20	0.07	0.58	0.65
Diagnostic tests per head of population over 6 months	0.38	0.15	2.43	2.58
Directly observed costs over 6 months				
RDT tests conducted	29,376	2,652	42,556	45,208
Cost per test (US$)	0.31	0.31	0.31	0.31
Total RDT cost (US$)	**9,113**	**823**	**13,201**	**14,024**
Microscopy tests	521	n/a	n/a	n/a
Cost per test (US$)	1.30	n/a	n/a	n/a
Total cost of microscopy (US$)	**676**	**n/a**	**n/a**	**n/a**
AL treatments	21,827	1,279	10,684	11,963
Cost per treatment (US$)	1.38	1.38	1.38	1.38
Total cost of AL (US$)	**30,069**	**1,762**	**14,718**	**16,480**
Total personnel costs (US$)	281,150	18,001	18,001	18,001
Time commitment (% FTE)	30	10	90	100
Personnel costs of malaria testing and treatment	**84,345**	**1,800**	**16,201**	**18,001**
Sundry maintenance, transport and running costs for six months (US$)	**43,201**	**103**	**926**	**1,029**
Total cost for Six months (US$)	**167,404**	**4,488**	**45,046**	**49,533**
Total non-treatment costs over 6 months (US$)	**137,335**	**2,726**	**30,328**	**33,053**
Projected annual summaries at observed rates of testing & treatment				
Total cost per head of population covered per year (US$)	**4.31**	**0.51**	**5.14**	**5.65**
Total non-treatment costs per head of population covered per year (US$)	**3.53**	**0.31**	**3.46**	**3.77**
Total cost per confirmed case treated (US$)	**10.75**	**3.56**	**4.44**	**4.34**
Projected potential summaries at optimized rates of active testing & treatment				
Total cost per head of population covered per year (US$)			**10.68**	
Total non-treatment costs per head of population covered per year (US$)			**6.25**	
Total cost per confirmed case treated (US$)			**8.09**	

[vi] US$ 1 = ZMW 4.89, exchange rate has been rebased to fit current Zambian currency. Source: http://www.boz.zm/(S(keg4bza3j0p4fx2uixtmnibq))/FinanciaMarkestReport.aspx. Accessed 1st October 2013

[vii] FTE - Full Time Equivalent.

[viii] In the projected annual summaries, cost per head of population was calculated by dividing the total costs by the catchment population. The cost of non treatment is the cost of only testing such as in an elimination scenario. The cost per confirmed case if the total costs divided by the diagnostically confirmed and treated cases of malaria infection.

[ix] Projected potential summaries developed from assumptions that the HF and CHW passive will not change even in an optimized environment. Cost/head of population calculated by addition to observed cost of an average 9 missed active visits per person and 2 passive visits per person by the cost of RDTs

and Treatment respectively. The cost per confirmed case treated is the total cost per head divided by the mean number of infections per year of 1.3.

If community participation could be dramatically improved to ensuring the average resident is tested at least once per month, and the trend observed in Figure 14B is assumed to represent impact of frequent testing and treatment upon infection status, the cost and cost-effectiveness of detecting and treating this diminishing case load would approximately double, even in a pre-elimination scenario where improved vector control would negate treatment costs (Table 13).

2.5 Discussion

Despite the fact that only a quarter of the covered resident population agreed to be tested in each monthly round of household visits by the CHWs, these active surveys by modestly remunerated paid CB staff identified >11,000 malaria parasite infections in a population of <18,000 residents in a single calendar year, of whom more than half were symptomatic in or around the time they were visited and may not have otherwise sought care (Table 10). The far higher sensitivity with which these active household surveys by the CHWs detect cases of malaria infection is also reflected in the observation that this surveillance arm captured twice as high an incidence rate as the passive surveillance activities of the HFs and CHWs combined. The strong association of many symptoms, especially fever, headache and vomiting with malaria infection, particularly among individuals tested during active household visits by the CHWs, confirms previous reports that illustrate just how inaccurate the term *asymptomatic* is in relation to widespread chronic malaria infections (Bisoffi, Gobbi et al. 2012; Cucunuba, Guerra et al. 2013; Lindblade, Steinhardt et al. 2013) that clearly cause very large proportions of the

overall burden of clinical illness in the community (Table 12, Figure 16). Clearly a large proportion of the population are infected with malaria, and suffering from a range of mild symptoms of clinical illness as a consequence, but do not feel ill enough to report to a HF or even to a nearby CHW to seek care. In addition to representing a major proportion of overall morbidity burden among the population, these chronic infections also act as a reservoir for continued transmission (Babiker, Abdel-Muhsin et al. 1998; Bousema, Gouagna et al. 2004; Schneider, Bousema et al. 2007; Okell, Bousema et al. 2012; Lindblade, Steinhardt et al. 2013). Regularly scheduled household visits by CHWs, who presumably will need to be paid for such a full time commitment, may therefore be extremely useful for identifying, treating and mapping the individuals who harbour chronic malaria infections and constitute the infectious reservoir that sustain transmission (Kaneko 2010; Campbell and Steketee 2011; Sturrock, Hsiang et al. 2013).

However, as implemented in this study, even these monthly active household visits, repeated on a continuous monthly survey cycle, had no obvious impact on malaria infection burden, possibly because most participants did not consent to testing often enough to benefit from any impact upon malaria infection and associated symptoms that are suggested, but not proven by, Figures 14 and 17, respectively. While the AL treatment used here has well-documented gametocidal properties (Drakeley, Jawara et al. 2004; Sutherland, Ord et al. 2005; Delves, Plouffe et al. 2012), the limited sensitivity of RDTs or microscopy and considerable natural density fluctuations of circulating *P. falciparum* blood stages mean that approximately half of all malaria infections are sub-patent and escape detection by a single testing event (Dietz, Molineaux et al. 1974; Bousema, Schneider et al. 2006; Okell, Bousema et al. 2012). Such test-

and-treat approaches would undoubtedly benefit from the availability of more sensitive nucleic acid-based testing technologies to detect all infections in the given community (Bousema, Okell et al. 2014). However, this was not a feasible option at the time of this study because the potential of technologies such as loop-mediated isothermal amplification remain uncertain (Abdul-Ghani, Al-Mekhlafi et al. 2012). Furthermore, mosquito-to-human transmission remained remarkably high in the study area, measured as a mean EIR of approximately 70 infectious bites per unprotected person per year (Seyoum, Sikaala et al. 2012; Sikaala, Killeen et al. 2013). The most likely explanation of the persistence of such intense transmission, despite reasonably high rates of LLIN use, supplemented with IRS in selected clusters towards the end of this study, is probably the emergence of pyrethroid resistance among local populations of *An. funestus* (Chanda, Hemingway et al. 2011). The high rates of re-infection suggested by these entomological surveys are consistent with and confirmed by the high rates of infection incidence (Smith, Drakeley et al. 2010) recorded here (Table 13) despite the imperfect sensitivity of RDTs (Bisoffi, Sirima et al. 2010; Abba, Deeks et al. 2011). Given the imperfect detection sensitivity of RDTs (Bisoffi, Sirima et al. 2010; Abba, Deeks et al. 2011) and the rapid rates of re-infection that can be expected in a setting with such a high EIR (Charlwood, Smith et al. 1998; Beier, Killeen et al. 1999; Smith, Killeen et al. 2004; Smith, Dushoff et al. 2005; Smith, Maire et al. 2006), it is unsurprising that at least monthly screening and treatment is required to achieve dramatic reductions of malaria infection burden (Figure 14B), associated symptoms (Figure 17, and presumably transmission (Brenner and Gefeller 1997; Missinou, Lell et al. 2003; Okell, Bousema et al. 2012), even assuming these two Figures reflect genuine impact rather than mere association. However, it is certainly encouraging that the apparent impacts among

residents consenting to such frequent testing and treatment, which these associations suggest, compare very well with simulations and field data from annual mass screen and treat programmes (Crowell, Briet et al. 2013), and even simulations and field observations of year-long mass drug administration programmes with treatment cycles of only four or even two weeks (Okell, Drakeley et al. 2008). It may also be encouraging that, despite their known limited sensitivity, RDTs appear to be sensitive enough to detect persistent malaria infections if each individual is tested often enough (Figure 14B) so that the frequent sporadic surges of detectable parasitaemia characteristic of *P. falciparum* are captured (Collins and Jeffery 1999; Eichner, Diebner et al. 2001). If the observed association of parasitaemia with testing and treatment (Figure 14B) reflects genuine impact, this also suggests patient compliance with the AL treatment regime used in this study was probably comparable with high estimates (84.5%) from previous evaluations in Zambia (NMCC 2006).

While evaluating the efficacy of the anti malarial drug was beyond the scope of this study, another I conducted with colleagues from the NMCC shows that the AL product used here remained an efficacious drug for the treatment of uncomplicated *P. falciparum* malaria in Zambia at that time (Hamainza, Masaninga et al. 2014). Resistance to this drug is therefore most probably of negligible relevance to this study so the limitations of impact upon diagnostic positivity are probably largely determined by rates of testing and treatment, as well as the absolute sensitivity of the RDT tests, relative to the rate of re-infection mediated by the vector population.

Beyond extending delivery of diagnostic and therapeutic services to the grass roots community level, the CHWs also provided a remarkably informative source of surveillance data, including

the overall burden and distribution of malaria and associated clinical symptoms, a number of important demographic and geographic determinants of risk, and the rates utilization of preventive interventions, such as IRS and ITNs. It is particularly encouraging that, despite their known sensitivity limitations, RDTs (Bisoffi, Sirima et al. 2010; Abba, Deeks et al. 2011) appear to be more than adequate for monitoring disease burden through CHW extension systems that can guide programme implementation. Latent antigenaemia several weeks after successful clearance of infection can cause false positive results when using HRP2 based RDTs, and therefore over-prescription of anti-malarial drugs (Mayxay, Pukrittayakamee et al. 2001; Swarthout, Counihan et al. 2007; Baiden, Webster et al. 2012), so it is possible that estimates of cost and cost-effectiveness described in table 13 may be improved upon with better diagnostic technology. If scale up of such CHWs is affordable beyond this research setting and could be scaled up across entire districts, provinces or even whole countries, such routinely collected data reported in disaggregated form from such small population subdivisions could be invaluable at all levels of programmatic monitoring and evaluation.

The costs of providing this CB extension of primary health care services, to provide both active and passive screening and treatment for malaria were substantive (Table 13), corresponding to 11.1 % of the annual per capita health budget of Zambia in 2011 ($96) (WHO 2011). Furthermore, to achieve the full potential of this service by ensuring community-wide engagement in screening and treatment on at least a monthly basis (Figure 14B), these costs are likely to double, even if baseline levels of transmission were reduced to pre-elimination levels so that the costs of drug treatment were negated (Table 13). It is highly unlikely, or desirable, that such a cadre of CHWs would be mobilized to deal with surveillance and control

of malaria alone so these CB personnel would also be required to deal with uncomplicated forms of other common illnesses like diarrhoea and pneumonia (Yeboah-Antwi, Pilingana et al. 2010; Hamer, Brooks et al. 2012; Seidenberg, Hamer et al. 2012). Even the passively provided malaria diagnosis and treatment services described here would need to be augmented with a range of other clinical services to be supported at programmatic level (Yeboah-Antwi, Pilingana et al. 2010; Hamer, Brooks et al. 2012). It is therefore difficult to envisage CHWs effectively or sustainably taking on such substantive commitments on a purely voluntary basis with no remuneration whatsoever. Barely more than a third of the overall costs of the active household surveys by the CHWs were accounted for by the cost of their meagre remuneration, and this Figure would reduce to less than a fifth if the increased commodity costs of full compliance with monthly screening and treatment were to be incurred through improved community participation. Paying these CHWs is therefore not only likely to be essential to ensure their retention and effectiveness as full time agents of malaria infection surveillance and control, it is also a relatively minor fraction of the overall cost of actively delivering extended CB primary healthcare services.

Apart from its observational design, the most obvious limitation of this study is that the majority of enrolled residents were tested and, where appropriate, treated far less than once a month (Table 10, Figures 12A and C, 13C and D, and 14A) as originally envisaged. Future evaluations of CHWs, especially those engaged to conduct frequent active household surveys of their entire assigned populations, should include operational research studies to better understand and address the limitations of service uptake by community members observed in this study.

As described earlier, it was hoped that this study would demonstrate successful reduction of diagnostic positivity across entire populations, and by inference human-to-mosquito transmission, by frequent testing and treatment, especially through active contacts by CHWs to seek out chronic infections that would otherwise go undetected (Okell, Bousema et al. 2012). Under the near-programmatic conditions of this study, these procedures failed to achieve sufficient frequency of testing and treatment. It is therefore understandable that the goal of suppressing the infectious parasite reservoir was not achieved. If these procedures are to be developed further so that this strategy constitutes a transmission control intervention in its own right, several aspects have to be improved upon: (1) Dramatically improved community engagement and sensitisation to enable consistent acceptance by the target population of participation in testing and treatment during monthly household visits, (2) Incorporation of more sensitive point-of-care confirmatory diagnostic tests, which would probably need to be nucleic-acid based in nature(Perandin, Manca et al. 2004; Okell, Ghani et al. 2009; Veron, Simon et al. 2009), (3) Improved supply chain management to enable timely and sufficient provision of all commodity requirements of the CHWs, (4) Consistent incorporation of additional, complementary vector control measures beyond LLINs alone, such as IRS with organophosphates, which was only applied to some selected population clusters too infrequently to maintain effectiveness all year round (Chapter 4), and (5) Institutionalised supportive supervision (Macauley 2005). While the experimental allocation of IRS treatments, and participant recollections of whether their houses had been sprayed or not, were recorded and accounted for in this study (See also Chapter 4), none of these other issues were assessed so these all merit substantive operational research investigation in their own right.

2.6 Conclusions

The monthly active household visits to entire communities by CHWs equipped with existing field-compatible diagnostic tools were not sufficient to eliminate the human reservoir of malaria infection from this rural African setting with intense transmission despite reasonably high LLIN/IRS coverage. However, observed negative associations between infection status and frequency of testing and treatment suggest that dramatic impact upon malaria parasite infection risk and associated disease burden may be achievable through far more regular testing and treatment. Substantive alleviation of malaria may be attainable and cost-effective if the substantial, but not necessarily prohibitive, costs of implementing frequent active CB surveys for chronic malaria infections are affordable to national programmes and higher levels of community participation in regular testing opportunities are achievable.

CHAPTER THREE

A COMPARISON OF MOBILE PHONE-BASED MALARIA REPORTING SYSTEM WITH ROUTINE PATIENT REGISTER DATA FOR CAPTURING SPATIAL AND TEMPORAL TRENDS IN EPIDEMIOLOGICAL INDICATORS IN RURAL ZAMBIA

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3.0 Abstract

Background

Timeliness, completeness, and accuracy are key requirements for any surveillance system to reliably monitor disease burden and guide efficient resource prioritization. Evidence for the effectiveness of electronic reporting of malaria cases by community health workers (CHWs) remains limited.

Methodology

Residents of two adjacent rural districts in Zambia were provided with malaria testing and treatment services, through both passive and active mechanisms, by 42 CHWs who were provided with malaria rapid diagnostic tests (RDTs) and artemisinin-based combination therapy, and served 14 population clusters centred around public sector health facilities. Reference data describing total numbers of RDT- detected infections and diagnostic positivity (DP) were extracted from detailed participant register books kept by CHWs. These were statistically compared with equivalent weekly summaries relayed directly by the CHWs themselves through a mobile phone short messaging system (SMS) reporting platform.

Results

Slightly more RDT-detected malaria infections were recorded in extracted participant registers than were reported in weekly mobile phone summaries but the difference was equivalent to only 19.2% (31,665 *versus* 25,583, respectively). The majority (81%) of weekly SMS reports were received within one week and the remainder within one month. Overall mean [95% confidence limits] difference between the numbers of register-recorded and SMS-reported RDT-detected malaria infections per CHW per week as estimated by the Bland Altman method was only -2.3 [-21.9, 17.2]. The mean [range] for both the number of RDT-

detected malaria infections (86 [0, 463] *versus* 73.6 [0, 519], respectively)) and DP (22.8% [0.0 to 96.3%] *versus* 23.2% [0.4 to 75.8%], respectively) reported by SMS were generally very consistent with those recorded in the reference paper-based register data and exhibited similar seasonality patterns across all study clusters. Overall, mean relative differences in the SMS reports and reference register data were more consistent with each other for DP than for absolute numbers of RDT-detected infections, presumably because this indicator is robust to variations in patient reporting rates by location, weather, season and calendar event because these are included in both the nominator and denominator.

Discussion/Conclusion

The SMS reports captured malaria transmission trends with adequate accuracy and could be used for population-wide, continuous, longitudinal monitoring of malaria transmission.

3.1 Background

Zambia, like many endemic countries, is going through an epidemiological transition with regard to malaria disease burden, with a notable national decline in parasite prevalence and incidence (Masaninga, Chanda et al. 2013). Public health surveillance, which has been defined as the "ongoing systematic collection, analysis, and interpretation of data critical to the planning, implementation, and evaluation of public health interventions and is closely integrated with timely dissemination of data generated to all stakeholders" (WHO 1962; Thacker and Berkelman 1988) therefore has an increasingly important role to play as malaria control steadily progresses towards malaria elimination (Wetterhall, Pappaioanou et al. 1992; Barclay, Smith et al. 2012; WHO 2012).

A viable surveillance system for malaria in Zambia needs to routinely collect sufficient data to describe the population's health status (Nelson, Thompson et al. 1997), for the purpose of detecting temporal and spatial variations in epidemiological profile, including those arising directly from changes in clinical and public health practices. In Zambia, the standard national surveillance system is the integrated health management information system (HMIS) (MOH 2008). Currently the HMIS is operational at all established health facilities in the country and requires monthly reporting from all medical office teams that aggregate and summarize these facility reports. Currently, there are 22 malaria-specific indicators reported through the HMIS which encompass disease burden, as well as use and availability of commodities for prevention, diagnosis and treatment (MOH 2008). As in most countries in sub-Saharan Africa, the completeness, accuracy and timeliness of HMIS in Zambia are often inadequate or, at the very least, questionable (WHO 2013).These systematic

weaknesses undermine stakeholder confidence in the reliability of data and, consequently lead to under-utilization for decision-making and planning (de Savigny and Binka 2004).

In poorly-resourced countries, community-based surveillance systems (CBSS) can complement health facility (HF)-based surveillance. This is of particular relevance to malaria control because the infectious human reservoir is primarily comprised of chronic infections associated with sub-acute symptoms distributed across entire at-risk populations (Hamainza, Moonga et al. 2014) and both rapid diagnostic tests (RDTs) and artemisinin-based combination therapies (ACTs) for uncomplicated malaria can be utilised appropriately by community health workers (CHWs) with minimal general education and technical training (Delacollette, Van der Stuyft et al. 1996; Yeboah-Antwi, Pilingana et al. 2010; Chanda, Hamainza et al. 2011; Mukanga, Babirye et al. 2011; Counihan, Harvey et al. 2012; Kalyango, Rutebemberwa et al. 2012). CBSS can provide quantitative estimates of disease burden in a defined population and service delivery indicators for disease control measures (Baker and Ross 1996) but remain under-exploited in relation to malaria, with only 12 million RDTs being used by CHWs globally, 11 million of which are accounted for by India alone (WHO 2013). Several studies have demonstrated the ability of CHWs to collect epidemiological data on a variety of diseases, including malaria, provided they have appropriate training and can relay their observations through appropriately tailored reporting systems (Hopkins, Talisuna et al. 2007; Alba, Hetzel et al. 2011; Counihan, Harvey et al. 2012; Kalyango, Rutebemberwa et al. 2012; Rutta, Francis et al. 2012; Hamainza, Moonga et al. 2014). Data from CBSS can be utilised to describe spatial and temporal patterns of variation in infection or disease incidence, as well as access to and use of preventive services, so that interventions and resources can be rationally allocated at fine scales across large-scale programmes (Tatem, Campiz et al. 2011; Barclay, Smith et al. 2012; Hamainza, Moonga et

al. 2014). While there is widespread consensus about benefits of the use of electronic reporting systems for relaying and collating data from CBSS in programmatic contexts (Freifeld, Chunara et al. 2010; Deglise, Suggs et al. 2012; Zurovac, Talisuna et al. 2012), evidence to support this view remains limited to remarkably few studies (Oak 2007; Freifeld, Chunara et al. 2010; Randrianasolo, Raoelina et al. 2010).

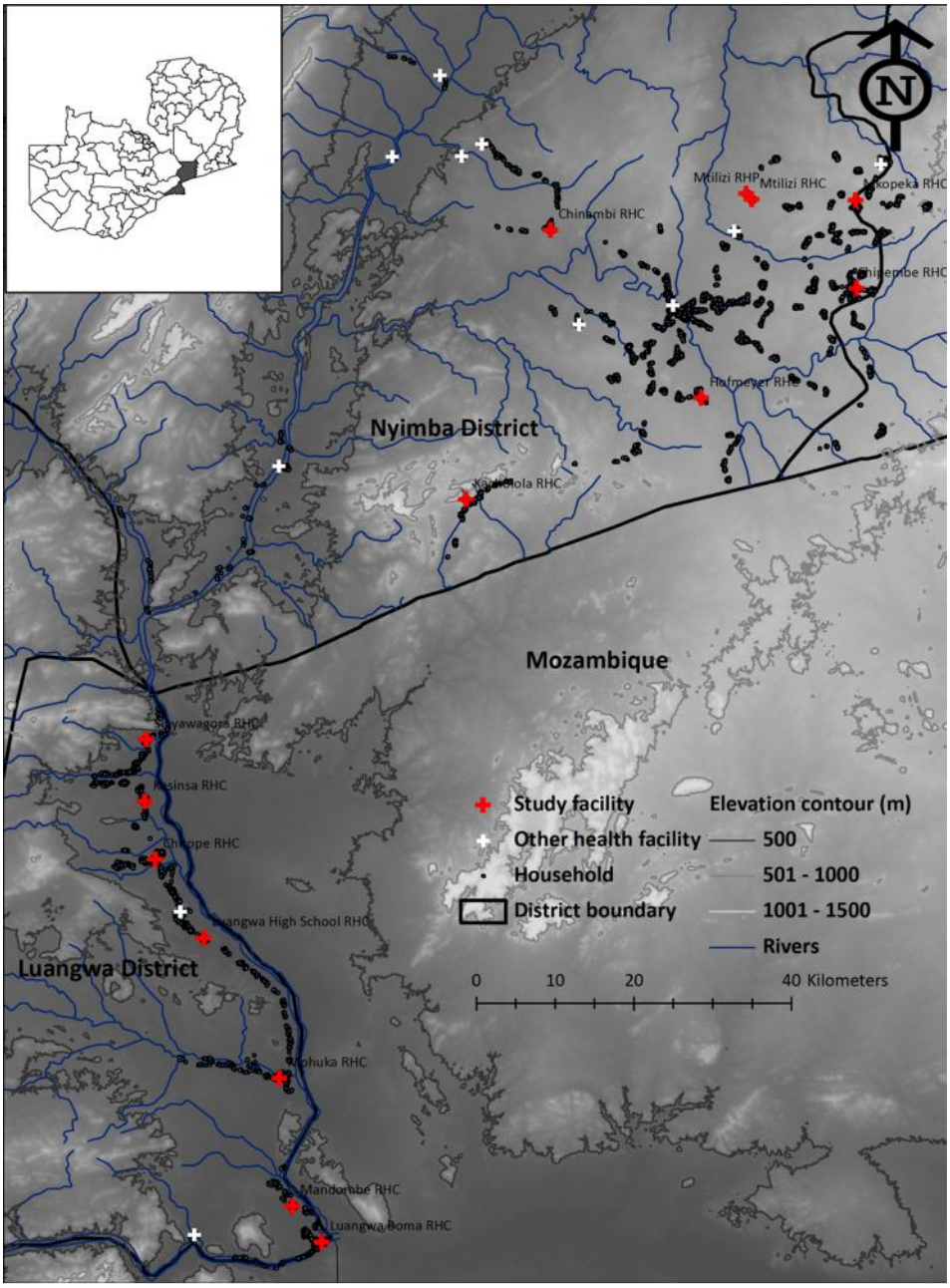
This study describes and evaluates a prototype mobile phone reporting platform for a CBSS in rural Zambia that was initially established as a program implemented by CHWs for community-wide passive and active testing with RDTs and treatment of all confirmed cases with Artemether-Lumefantrine (AL), which also allowed monitoring of malaria parasite infection burden as a secondary objective (Hamainza, Moonga et al. 2014).

3.2 Methods

3.2.1 Study sites

A CBSS was established in the rural districts of Luangwa and Nyimba, respectively located in Lusaka and Eastern provinces of Zambia (Figure 19) (Hamainza, Moonga et al. 2014).

Figure 19.Map showing the selected health facilities around which the community health workers and the catchment population they served were located



In these districts, perennial, intense transmission of *Plasmodium falciparum* was mediated by *Anopheles funestus* Giles at an estimated entomological inoculation rate of approximately 70 infectious bites per unprotected person per year (Sikaala, Chinula et al. 2014). Luangwa (3,468 km^2) is located 325 kilometres south-east of Lusaka, the capital city

of Zambia. The district's total population is approximately 27,560 residents and it has an annual estimated population growth rate of 2.9% (CSO 2011). Nyimba is a larger district (10,943 km^2), located 350 kilometres east of Lusaka, with an approximate population of 108,637 inhabitants and an estimated annual population growth rate of 3.4 % (CSO 2011).

3.2.2 Testing and treatment

As described in detail elsewhere (Hamainza, Moonga et al. 2014), 14 population clusters (7 in each district), consisting of the nearest consenting 165 households to a selected public sector HF were selected and enrolled to participate in longitudinal parasite surveys conducted by paid CHWs. Each cluster had 3 CHWs, resulting in a total of 42 CHWs, and thus a corresponding number of reporting units, distributed across the study area. Parasitological assessments were conducted continuously from January 2011 to March 2013 in Luangwa and from April 2011 to March 2013 in Nyimba district in all the selected clusters. All consenting households received monthly active visits from CHWs, which included parasitological surveys using RDTs detecting histidine-rich protein antigen (Malaria Pf cassette test, ICT Diagnostics), coupled with registers designed in a pre-defined questionnaire format (Hamainza, Moonga et al. 2014). Consent for household participation was given by the head of the household and then consent for participation in the RDT test was obtained from individual study participants, or guardians in the case of assenting minors. If they developed any symptoms in between these active visits, study participants were encouraged to seek care through passively-offered diagnosis and treatment services, either from the CHWs at their place of residence or at the nearest HF. All participants found positive for malaria infection were treated with AL as per national policy (NMCC 2010). All participants encountered in either the active or passive visits that were found to be negative

for malaria infection, but were febrile or complained of any other symptom of illness, were referred to the nearest HF.

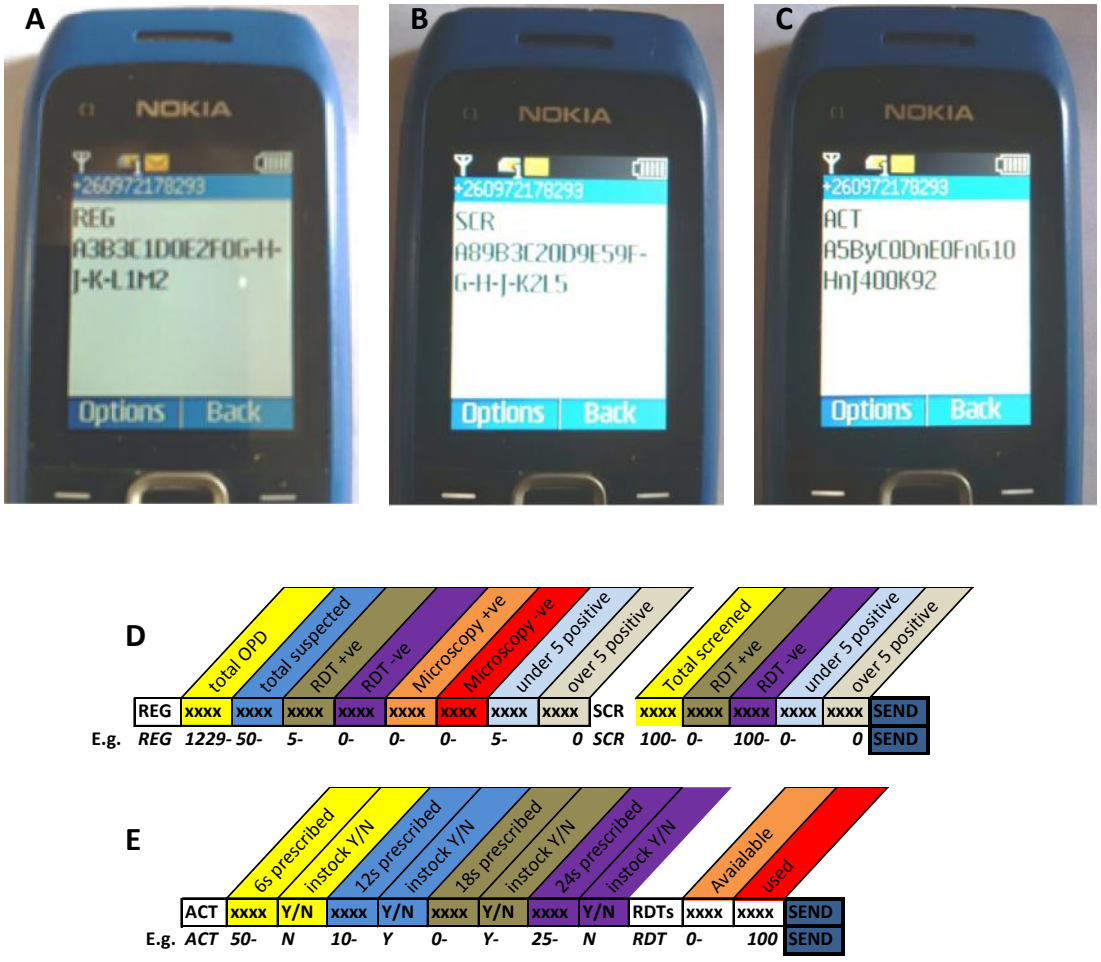
3.2.3 Questionnaire surveys

The CHWs' primary method of collecting study participant information was through the use of a paper-based, pre-defined questionnaire provided in the physical form of a register book. A total of 16 data elements were captured through this method and these included date, cluster, participant identification number, sex, age, visit type (active/ passive), village, axilliary temperature, RDT results, clinical symptoms of illness (fever, history of fever, headache, cough, diarrhoea, vomiting, breathing problems, chest pain and any other symptoms), treatment provided and participant outcome, in addition to access and utilization of specific preventive measures, namely long-lasting insecticidal nets (LLINs), indoor residual spraying (IRS) and intermittent preventive therapy. All the entries in the registers, with each line of data corresponding to a single participant contact, were made in duplicate with carbon paper. During the monthly supervisory visits, original copies of these reports were collected and transported to the NMCC for entry into a correspondingly structured Microsoft excel spreadsheet in fully explicit, line-listed electronic database format, while the CHWs retained the duplicate copies for their own records. All data from the registers were double-entered and verified, reconciled, and then cleaned following descriptive frequency analysis of the distributions of values for each variable.

3.3 Weekly summary reporting of aggregate data via short message service

In addition to the paper-based participant registers, weekly aggregate summary reports of these data were collated through mobile phone short messaging service (SMS) via the Airtel® network (Figure 20).

Figure 20. Screenshot examples of Community Health Worker mobile phone short messaging service (SMS) text before transmission to the National Malaria Control Centre and an Illustration of the interpretation of the code sent through the SMS reporting system. A: example of an SMS code for study participants that were attended to through the passive system when they self-reported to the CHW, B: example of an SMS code for study participants contacted through active monthly household visits, C: Example of an SMS code for reporting on use and availability of AL and RDTs, D: Schematic Illustration of the format and syntax of reports for diagnostic results from both passive and active visits, and E: Schematic Illustration of the format and syntax of reports for the availability of AL and RDTs. REG refers to passive participants data, SCR refers to active participant data, ACT refers to Artemether-Lumefantrine combination therapy stocks, RDT refers to rapid diagnostic test stocks, OPD refers to passive participant contact, and Y or N – refer to stock availability (Yes or No).



A paper-based form summarizing the aggregated data elements to be transmitted via SMS was completed by each CHW each week, based on the fully explicit data recorded in the participant register books. These weekly summaries were then transmitted to a mobile

phone at the NMCC via SMS text in a pre-determined format, every Friday where that was possible (Figure 20). A paper-based form summarizing the aggregated data elements to be transmitted via SMS was completed by each CHW each week, based on the fully explicit data recorded in the participant register books. These weekly summaries were then transmitted to a mobile phone at the NMCC via SMS text in a pre-determined format, every Friday (Figure 20). A total of 25 data elements were transmitted through this method, which included the total number of participants in both active and passive visits, number of positive and negative cases disaggregated by age and type of visit, number with and without fever, in addition to summaries of various AL pack sizes dispensed and remaining, and similar indicators for the use and availability of RDTs (Table 14).

Table 14.Data element definitions for all data transmitted through the SMS mobile reporting system

DATA ELEMENT	DEFINATION
REG (A)TOTAL OPD	Total number of study participants that sort care from the CHWS
REG (B) TOTAL SUSPECTED	Total number of study participants that sort care from the CHWs and were suspected of having malaria
REG (C) UNDER 5 RDT +VE	Total number of study participants that sort care from CHWs under the age of five and had a positive rapid diagnostic test result
REG (D) UNDER 5 RDT -VE	Total number of study participants that sort care from CHWs under the age of five and had a negative rapid diagnostic test result
REG (E) OVER 5 RDT +VE	Total number of study participants that sort care from CHWs over the age of five and had a positive rapid diagnostic test result
REG (F) OVER 5 RDT -VE	Total number of study participants that sort care from CHWs over the age of five and had a negative rapid diagnostic test result
REG (L) FEVER UNDER 5	Total number of study participants that sort care from CHWs under the age of five who were febrile
REG (M) FEVER OVER 5	Total number of study participants that sort care from CHWs over the age of five who were febrile
SCR (A) TOTAL SCREENED	Total number of study participants that consented to actively being tested for malaria by CHWs
SCR (B) UNDER 5 +VE	Total number of study participants that consented to actively being tested for malaria by CHWs under the age of five and had a positive rapid diagnostic test result
SCR (C) UNDER 5 RDT -VE	Total number of study participants that consented to actively being tested for malaria by CHWs under the age of five and had a negative rapid diagnostic test result
SCR (D) OVER 5 RDT +VE	Total number of study participants that consented to actively being tested for malaria by CHWs over the age of five and had a positive rapid diagnostic test result
SCR (E) OVER 5 RDT -VE	Total number of study participants that consented to actively being tested for malaria by CHWs over the age of five and had a negative rapid diagnostic test result
SCR (K) FEVER UNDER 5	Total number of study participants that consented to actively being tested for

	malaria by CHWs under the age of five who were febrile
SCR (L) FEVER OVER 5	Total number of study participants that consented to actively being tested for malaria by CHWs over the age of five who were febrile
(A) 6S PRESCRIBED	Total number of AL 6 packs prescribed and provided to study participants
(B) INSTOCK	Total available AL 6 packs
(C) 12S PRESCRIBED	Total number of AL 12 packs prescribed and provided to study participants
(D) INSTOCK	Total available AL 12 packs
(E) 18S PRESCRIBED	Total number of AL 18 packs prescribed and provided to study participants
(F) INSTOCK	Total available AL 18 packs
(G) 24S PRESCRIBED	Total number of AL 24 packs prescribed and provided to study participants
(H) INSTOCK	Total available AL 24 packs
(J) RDT AVAILABLE	Total available individual rapid diagnostic tests
(K) RDT USED	Total used individual rapid diagnostic tests

Once each data message was received by the NMCC, an acknowledgement message was manually sent back to the sender by a data entry associate. The data was reviewed by the study team and the CHWs were contacted by voice via mobile phone for verification or correction of the data if any clarifications were required. Once the verification process was complete, the data were then compiled into a Microsoft Excel® spreadsheet in database format. The Mobile phones were also used for communication among the CHWs and staff at their supervising health facility. In addition to provision of a basic mobile phone at a cost of ZMW 50 (approximately US$9.47 based on the 2012 average annual exchange rate of ZMW 5.23 to US$1 (BOZ 2012)) for each CHW, a monthly air time top-up worth ZMW20 (US$3.32) was electronically sent to each CHW in order to facilitate transmission of the SMS reports and any other communication requirements.

3.4 Concordance evaluation

The explicit, unaggregated, line-listed, manually double-entered reference data from the participant registers were independently aggregated by the NMCC into weekly estimates of total numbers malaria infections seen by each CHW and the corresponding diagnostic positivity (DP) of tests associated with those participant contacts. The weekly DP was estimated as outlined in the formula below:

$$DP = \frac{Total\ weekly\ number\ of\ confirmed\ RDT\ detected\ m\ malaria\ infections}{Total\ weekly\ number\ of\ study\ participants\ tested\ by\ RDT}$$

The strength of association of the overall number of malaria infections recorded in the reference register data with those reported via SMS was expressed in terms of the correlation coefficient (*r*) between the two data capture methods, which were also disaggregated by type of patient contact. Even through the relationship between the reporting systems was expected to be linear, the study took into consideration the fundamental limitation of pure correlation analysis that does adjust the value of *r* even when a linear transformation of either variables compared occurs, implying a focus on the degree of association and not agreement, as it is possible to obtain high correlation even when the absolute values of the variables are different (Taylor and Taylor 1990; Bland and Altman 2003). Thus in order to estimate the degree of agreement of these data sets, the numbers of infections detected by each CHW each week were subjected to Bland-Altman statistical analysis, which is a graphical analysis method whereby the difference between the weekly total number of study participant contacts from each reporting system was plotted against the mean across the two reporting systems to allow for measurement of the magnitude of the errors, as well as the trends in magnitude of the errors, relative to the magnitude of the total number of malaria infections recorded in the patient registers and reported through the SMS reporting system (Bland and Altman 1995).Time trend analysis was also applied to summarised reference register data compared to the SMS-reported total number of malaria infections data and DP in order to further assess the correlation between these datasets as a function of time lag. The total number of malaria infections time series data were further analysed by cluster using a bivariate local polynomial regression, specifically the locally-weighted scatter-plot smoother (LOESS) to provide a visually

smoothened fit of time series from each of the two reporting systems by identification of, and minimization of influence, of possible outlier data (Cleveland 1979; Jacoby 2000). Additionally this analysis also provided a qualitative comparison of the mean relative difference, both overall and disaggregated by type of participant contact, as well as time trends and seasonality of data provided from the reference paper-based records *versus* those reported in the SMS system.

3.3 Ethical approval

Ethical approval was obtained from the University of Zambia, Biomedical Research Ethics Committee (Reference 004-05-09) and the Research Ethics Committee of the Liverpool School of Tropical Medicine (Approval 09.60). Authority to conduct the study was also obtained from the Ministry of Health in Lusaka, Zambia.

3.4 Results

The reference paper-based register recorded an overall total of 129 weeks of data, while the SMS reporting system reported only 120 weeks of data, due to delayed initiation and earlier termination of the latter when funding ran out towards the end of the study. Out of an expected 5040 SMS weekly summaries, only 2934 (58.2%) were submitted by the CHWs. This may have been due to the lack of power sources for charging the mobile phones because most of the study sites were not connected to the national power grid. This resulted in the CHWs travelling long distances to charge their phones. Additionally this may also have been due to unstable mobile phone network coverage particularly in the most rural of the study sites. Of the 2934 SMS weekly summaries submitted by the CHWs, 81% (2375/2934) were submitted according to the timeliness targets at the outset (weekly) and

the remainder (19% (559/2934)) within one month. These delays in reporting a minority of weekly summaries may have been due a lack of access to mobile phone network coverage, particularly in the most remote parts of the study area. However, CHWs were encouraged to transmit any missing reports whenever they had access to mobile phone coverage. In some limited situations, this lead to a few CHWs choosing to transmit all their data at the end of the month when they travelled to areas of good mobile phone coverage, such as when they visited the HFs to collect supplies and submit the reference paper-based register records. All analyses of concordance reported here were based only on comparisons of data summaries from the reference paper-based register records and SMS reports where both were available for the same CHW in the same week which ranged from 40 to 100 weeks. All detected malaria infections considered in the analyses of these data refer only to infections which were detected parasitologically by RDT, rather than by clinical symptoms alone.

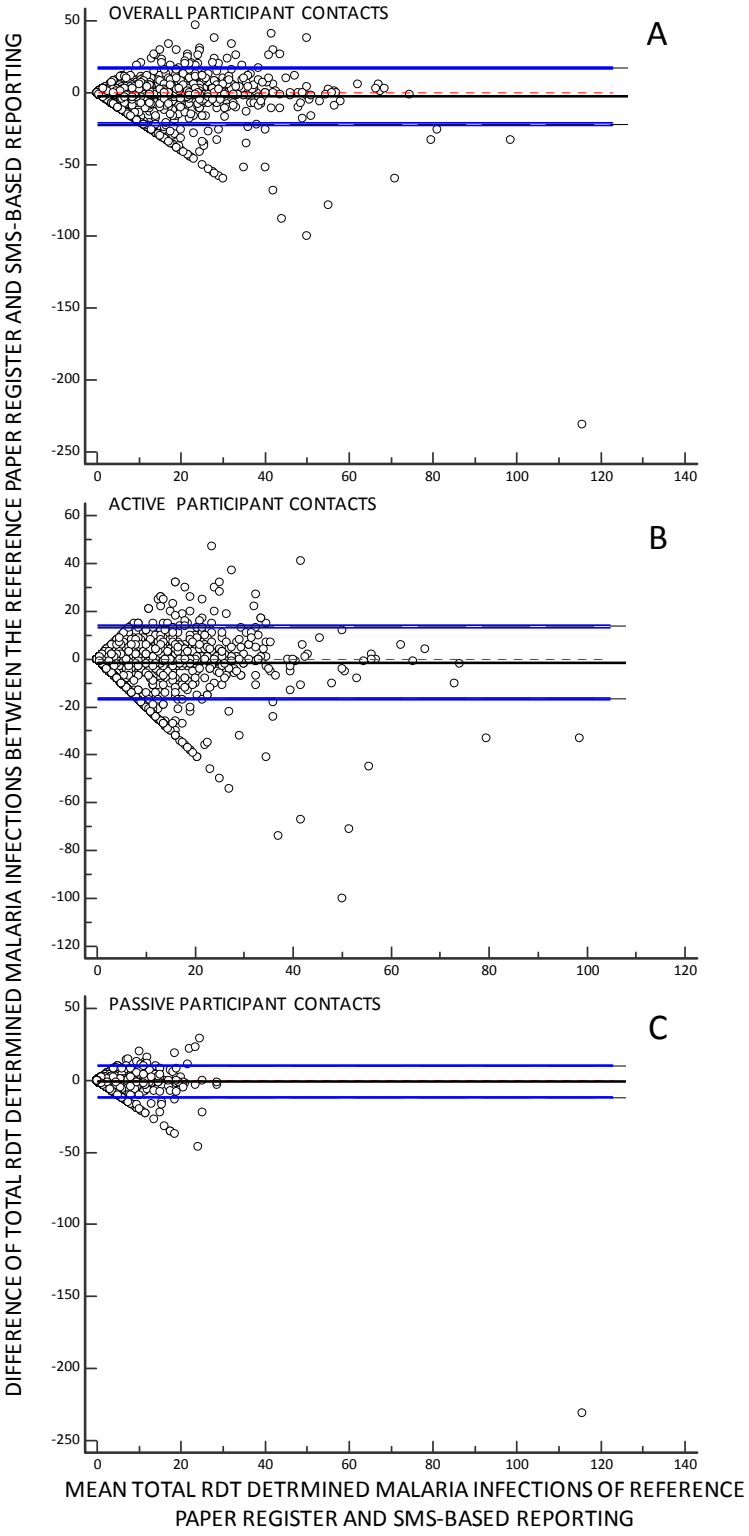
An overall total of 31,665 and 25,583 detected malaria infections were recorded through the paper-based participant register and reported through the SMS platform, respectively. The mean weekly number of RDT-detected malaria infections recorded by each CHW in the reference paper-based register was 7.1 (1.1 and 6.0 from the passive and active participant contacts, respectively), while that reported in the SMS systems was 9.0 (2.3 and 6.7 from passive and active contacts, respectively).

There was a clear association between the overall number of RDT-detected malaria infections recorded by the reference paper-based register and reported by SMS platform (r [95%CI] = 0.64 [0.61, 0.66], $p<0.001$). This association was also clear when the data was disaggregated by participant contact type, with a stronger correlation for the active contacts (r [95%CI] = 0.68 [0.65, 0.69], $p<0.001$) than the passive contacts (r [95%CI] = 0.36 [0.33,

0.39], p<0.001), as evidenced by the higher correlation coefficient and the lack of overlap between the confidence intervals for the two participant contact types.

The overall mean [95% limits of agreement (LOA)] difference in the number of RDT-detected malaria infections per week per CHW between the reference paper-based register and SMS reporting platform, as estimated by the Bland-Altman method, was -2.3 ([-21.9, 17.2]) infections per CHW per week (Figure 21). Bland-Altman estimates for the mean [95% LOA] difference in the number of register-recorded versus SMS-reported malaria infections in active participant contacts was -1.4 ([-16.5, 13.7]) infections per CHW per week and less for passive participant contacts at -0.9 ([-12.0 to 10.1]) infections per CHW per week (Figure 21).

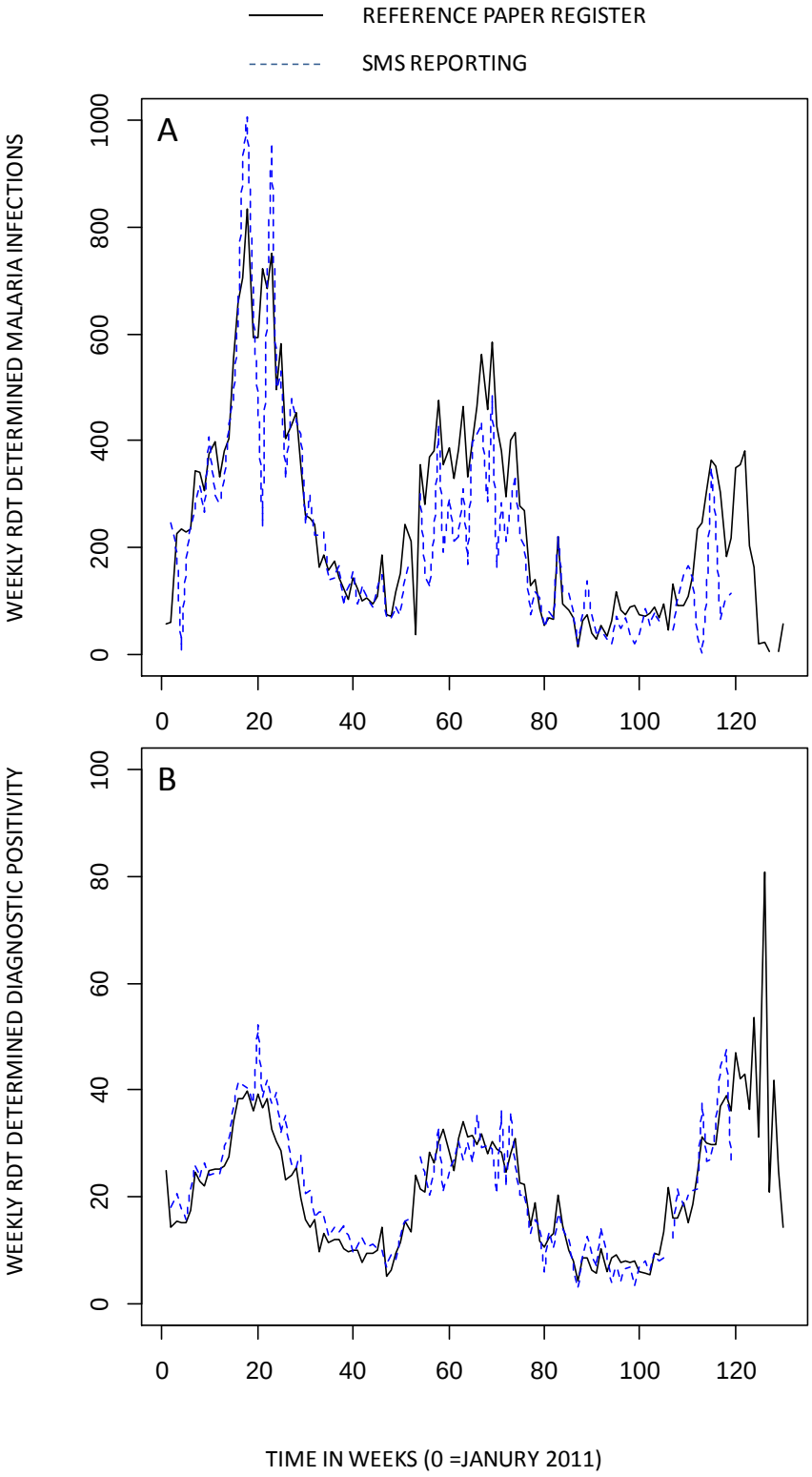
Figure 21.Bland-Altman plots of the difference between weekly numbers of rapid diagnostic test detected malaria infections recorded in the reference paper register and reported by SMS against their mean, overall (A), or disaggregated into active (B) and passive (C) participant contacts.



Thus, observations on the overall, active and passive malaria infections per CHW per week suggest that the reference paper-based register records and SMS reports are approximately similar but should be interpreted with caution due to the considerable data imprecision. While the DP of participants tested has proven a robust indicator of malaria risk as recorded in the reference patient registers (Hamainza, Moonga et al. 2014), the binomial distribution that presumably stabilizes this proportional outcome to variations in participant contact rates also precludes the use of Bland-Altman analysis. However, it was possible to compare trends in the weekly aggregate values for RDT-detected malaria infections and DP outcomes descriptively and using LOESS regression.

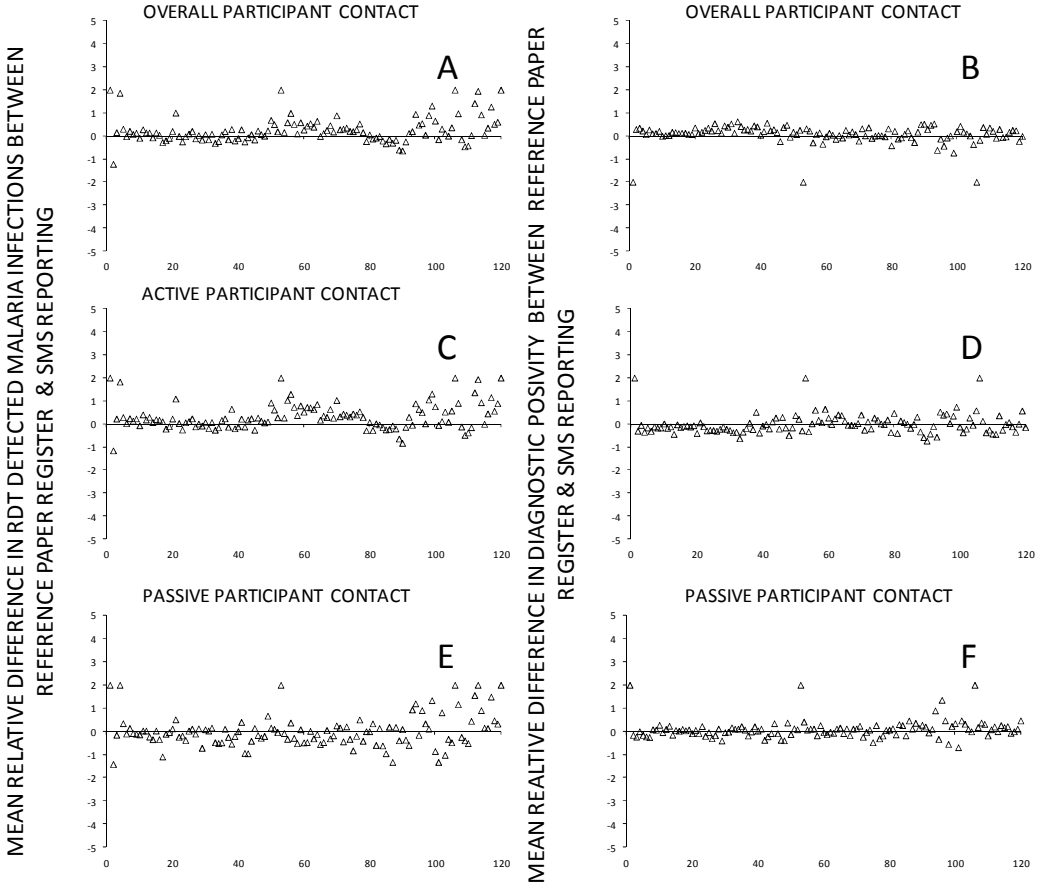
Trends over time for the overall number of malaria infections (Figure 22A) and, even more so, overall DP (Figure 22B) aggregated by week across all CHWs suggests a close positive association between the paper-based reference register and SMS reporting system. On average, the two datasets closely matched each other in absolute terms and exhibited very similar temporal variations that captured comparable seasonal trends, with both maxima and minima consistently occurring at approximately the same times.

Figure 22. Time series analysis plot of weekly means, aggregated across all community health workers, for numbers rapid diagnostic test detected malaria infections (A) and associated diagnostic positivity (B) as recorded by the reference paper-based registers and reported by SMS



This observation was confirmed when these two malaria infection burden indicators were compared in terms of their mean relative difference overall (Figure 23A and B), and when disaggregated into active (Figure 23C and D) and passive (Figure 23E and F) participant contacts. Overall, mean relative differences in the SMS reports and reference register data were more consistent with each other for DP than for absolute numbers of RDT-detected infections, presumably because this indicator is robust to variations in patient reporting rates by location, weather, season and calendar event because these are included in both the nominator and denominator.

Figure 23.Difference plots for numbers of detected malaria infections (A**,** C**,** E**) and diagnostic positivity (**B**,** D**,** F**) as recorded by the reference paper register and reported by SMS overall (**A **and** B**), or disaggregated by active (**C **and** D**) and passive (**E **and** F**) participant contacts**



Cluster-disaggregated time series for both the reference paper registers and SMS reports also exhibited similar time trends and seasonal variation in the number of RDT-detected malaria infections (Figure 24) and especially DP (Figure 25). Visual inspection of the results of LOESS regression, using identical bandwidths within each cluster, indicate that the SMS reporting system captures similar temporal and geographic variation to that recorded in the reference data detailed in the participant register.

Figure 24.Locally weighted scatter-plot smoother (LOESS) plots of weekly cluster-disaggregated mean numbers of detected malaria infections recorded by the reference paper-based registers and reported in the SMS system against time in weeks of reporting

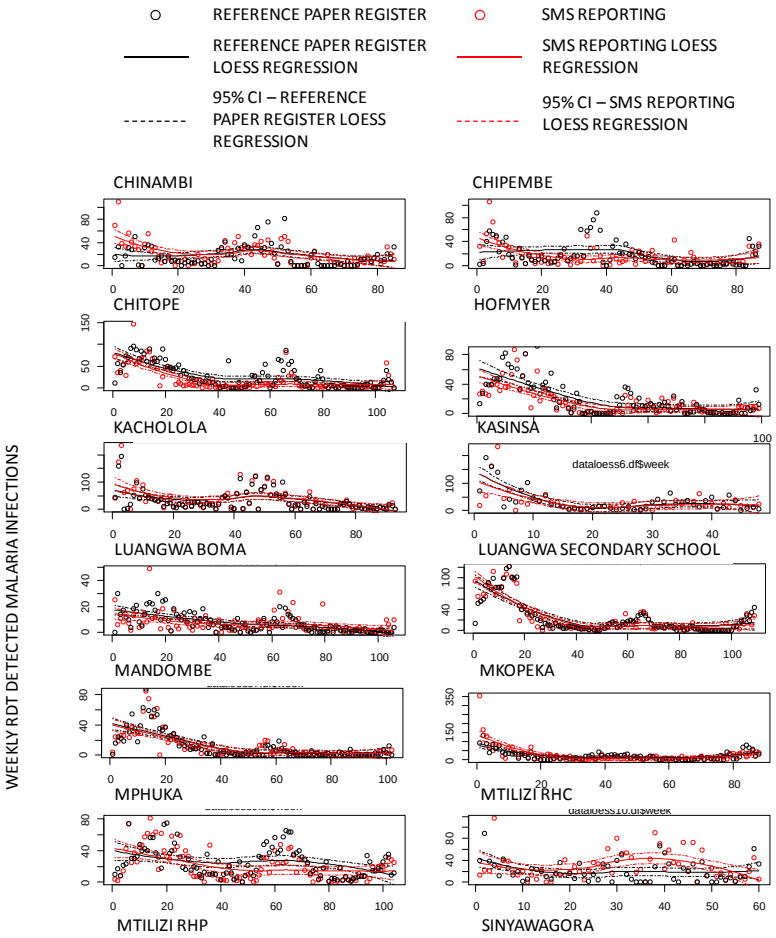
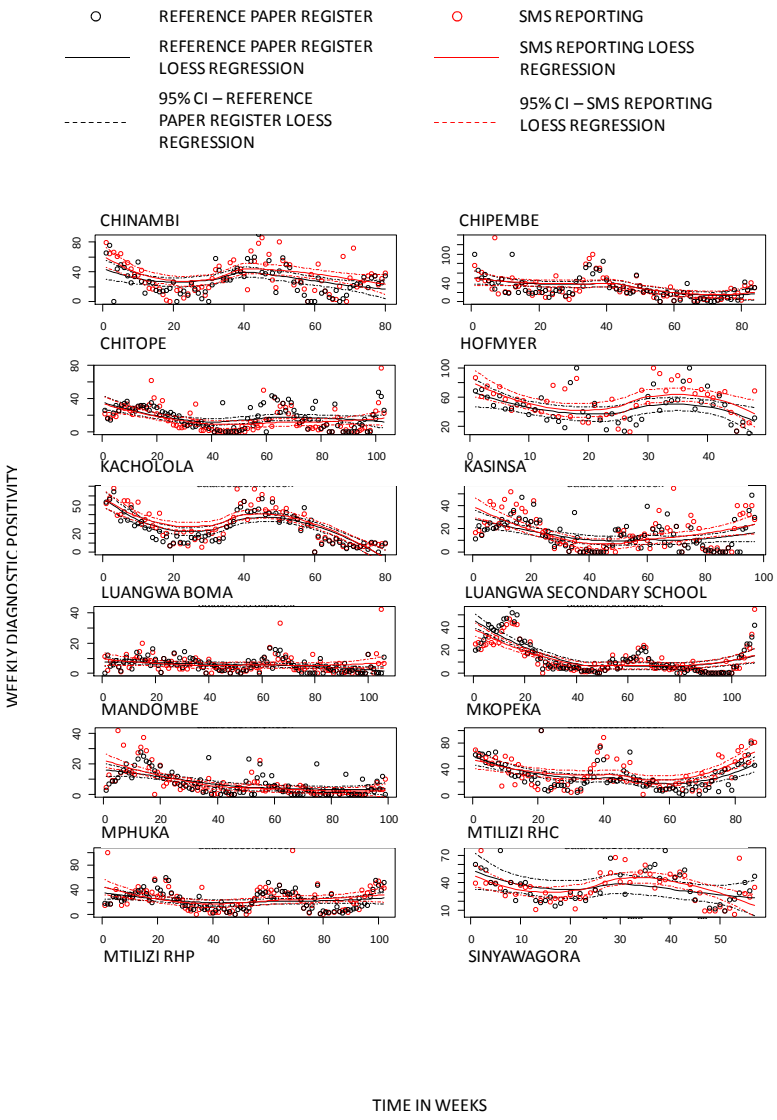


Figure 25.Locally weighted scatter-plot smoother (LOESS) plots of weekly cluster-disaggregated diagnostic positivity recorded by the reference paper-based registers and reported in the SMS system against time in weeks of reporting



A comprehensive cost-effectiveness analysis of the full running costs of CBSS, exclusive of SMS reporting system and costs, is described in detail elsewhere (Hamainza, Moonga et al. 2014). However at a total annual provider fee running cost of only US$39.84 and a single expenditure of US$9.47 for the mobile phone per CHW at start up, this SMS reporting system may be considered an affordable, inexpensive operational supplement to this CBSS

that may dramatically enhance the *de facto* value of its data recording and reporting mechanisms.

3.5 Discussion

The CBSS routinely captured large volumes data on population characteristics (age, sex and residence), symptomology, morbidity, diagnostic testing and treatment for malaria, as well as LLINs and IRS which is not routinely captured by the national HMIS (MOH 2008; Hamainza, Moonga et al. 2014). The study also further demonstrated that CHWs are able to reliably record, summarise and transmit this data through an electronic reporting platform, in this case a mobile phone. The study was conducted in a framework of evaluating the adequacy and effectiveness of the CBSS SMS reporting system rather than its efficacy in programmatic conditions (Habicht, Victora et al. 1999; Glasgow, Lichtenstein et al. 2003; Flay, Biglan et al. 2005). This allowed for an opportunity to learn lessons about its potential for scale up, generalizability and indeed relevance to real world program implementation (Habicht, Victora et al. 1999; Flay, Biglan et al. 2005).

The comparison of the reference paper-based, quality-controlled register records with the SMS weekly summaries indicates some modest incompleteness of the latter, with the paper based system recording a higher overall number of malaria infections. This may have been due to under-reporting in the SMS reporting system resulting from challenges in accurately summarising the reference registers data and non-submission of the SMS reports by some of the CHWs when they were required to. Additionally, the manual entry of the SMS-transmitted data into an excel spreadsheet by the study team may have resulted in some reports being lost in the process and not entered into the database because, unlike the paper-based register records, there was no double entry verification of this data.

Timeliness, a key performance measure of any public health reporting system (Jajosky and Groseclose 2004; Mawudeku, Ruben et al. 2007) was achieved by the SMS based platform in most instances, as these reports were received as expected within a week in the vast majority of cases, and within a reasonably adequate timeline of one month the rest of the time. Monthly quality control visits to CHWs were primarily provided to support correct recording of data in the reference data on paper, in the form of a detailed review of the data collected when the registers were handed over to the supervision team. During these visits the SMS reporting records were also reviewed but in most cases the CHWs had already submitted at least some of their SMS reports prior to the visit.

In the reference paper-based register, the high level of detail that included individual-level data over the entire implementation period provided an invaluable resource in the form of a rigorous epidemiological database for critical analysis of malaria transmission dynamics and risk factors (Hamainza, Moonga et al. 2014). While such comprehensive, detailed data entry and analysis is not realistically feasible beyond the unusually well-resourced context of such a research study, summaries of these indicators could nevertheless be included in scalable electronic reporting systems similar to the one described here. These data could support planning processes at district, provincial and national levels, and also assess independent reports from these levels on needs, forecasts, access and use of various malaria interventions. Additionally, these data may be used for targeting interventions as malaria transmission drops as these SMS based reports demonstrate adequacy in reflecting epidemiological trends in a population on a timely basis. Although, not explicitly explored in this study the findings suggest a role for SMS based reporting as a potential management tool to monitor performance of frontline health workers and in this case, the CHWs. This was exemplified in this study through the monitoring of CHWs performance as to whether

they achieved the targeted number of active household visits per week and tracking the availability and use of AL and RDTs.

An obvious limitation of the study was that the SMS platform would have benefited from an automated central server system of data capture when SMS reports were submitted, which would have reduced human error in the form of missing or duplicated data and transcription errors.

3.6 Conclusion

This SMS reporting system captured malaria transmission trends, as recorded in the reference paper based registers, with adequate accuracy, suggesting potential use in population-wide, continuous and longitudinal monitoring of temporal and geographic trends in disease incidence. Additionally, the summary reports through the SMS platform enabled reasonable timely collation, access and dissemination of the paper-based surveillance data which was shared among the study team and district health management teams in the study districts and could be used for programmatic decision making. Given the recent dramatic growth in mobile phone networks globally, and in Africa particularly, such CBSS may be highly effective if mobile technology platforms are harnessed for reporting directly from CHWs to central servers to allow for rapid data access, use and quality assurance by stakeholders at the district, provincial and national levels.

CHAPTER FOUR

INCREMENTAL IMPACT UPON MALARIA TRANSMISSION OF SUPPLEMENTING PYRETHROID-IMPREGNATED LONG LASTING INSECTICIDAL NETS WITH INDOOR RESIDUAL SPRAYING USING PYRETHROIDS OR THE ORGANOPHOSPHATE PIRIMIPHOSMETHYL

This paper is in preparation for submission

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4.0 Abstract

Background

LLINs and IRS are the most widely accepted and applied malaria vector control methods. However, evidence that incremental impact is achieved when they are combined remains limited and inconsistent.

Methodology

Fourteen population clusters of approximately 1000 residents in Zambia's Luangwa and Nyimba districts, which had high pre-existing usage rates (81.7%) of pyrethroid-impregnated long-lasting insecticidal nets (LLINs) were quasi-randomly assigned to receive IRS with either of two pyrethroids, namely Deltamethrin (Wettable granules (WG)) and Lambdacyhalothrin (Capsule suspension (CS)), with an emulsifiable concentrate (EC) or CS formulation of the organophosphate pirimiphosmethyl, or with no supplementary vector control measure. Diagnostic positivity of patients tested for malaria by community health workers in these clusters was surveyed longitudinally over pre and post-treatment periods spanning 29 months over which the treatments were allocated and re-allocated in advance of 3 sequential rainy seasons.

Results

Supplementation of LLINs with pirimiphosmethyl CS offered the greatest initial level of protection against malaria in the first 3 months of application (incremental protective efficacy (IPE) [95% confidence interval (CI)] = 0.63 [CI 0.57,0.69], P<0.001), followed by lambdacyhalothrin (IPE [95%CI] = 0.31 [0.10,0.47], P=0.006) and pirimiphosmethyl EC (IPE, 0.23 [CI 0.15,0.31], P<0.001) and then by deltamethrin (IPE [95%CI] = 0.19 [-0.01,0.35],

P=0.064). Neither pyrethroid formulation provided protection beyond 3 months after spraying, but the protection provided by both pirimiphosmethyl formulations persisted undiminished for longer periods: 6 months for CS and 12 months for EC. The CS formulation of PM provided greater protection than the combined pyrethroid IRS formulations throughout its effective life IPE [95%CI] = 0.79 [0.75, 0.83] over 6 months. The EC formulation of PM provided incremental protection for the first three months (IPE [95%CI] = 0.23 [0.15, 0.31]) that was approximately equivalent to the two pyrethroid formulations (lambdacyhalothrin, IPE [95%CI] = 0.31 [0.10, 0.47] and deltamethrin, IPE [95%CI] = 0.19 [-0.01, 0.35]) but the additional protection provided by the former, apparently lasted an entire year.

Conclusion

Where universal coverage targets for LLINs utilization has been achieved, supplementing LLINs with IRS using pyrethroids may reduce malaria transmission below levels achieved by LLIN use alone, even in settings where pyrethroid resistance occurs in the vector population. However, far greater reduction of transmission can be achieved under such conditions by supplementing LLINs with IRS using non-pyrethroid insecticide classes such as organophosphates, so this is a viable approach to mitigating and managing pyrethroid resistance.

4.1 Background

Long-lasting insecticidal nets (LLINs) and indoor residual spraying (IRS) are the two first-choice malaria vector control methods available globally (WHO 2013) because they can achieve massive community-wide impact upon malaria transmission, even at partial coverage (Killeen, Smith et al. 2007). This is possible because many of the world's most potent vector species prefer people as a source of blood and must feed several times upon humans inside houses before they are old enough for infectious sporozoite-stage malaria parasites to have fully developed within them (Koella, Sorensen et al. 1998).While IRS and LLINs decrease exposure of directly protected humans to infected vectors and *vice versa*, through contact irritancy or spatial repellency, most of the impact of LLINs and IRS upon human transmission exposure and parasitaemia, results from community-level suppression of vector population density and infection prevalence, achieved by reducing their longevity through lethal exposure to their toxic active ingredients.(Smith and Webley 1968; Lines, Myamba et al. 1987). The success of these modes of action are influenced by the choice, dosage and formulation of insecticide utilized, as well as its coverage and mode of application, combined with the behavioural and physiological susceptibility of the targeted vector species (Dezulueta, Cullen et al. 1963; Grieco, Achee et al. 2007; White, Conteh et al. 2011).

Compared to IRS, LLINs coverage is much higher in most endemic countries (Lengeler 2004; Pluess, Tanser et al. 2010) due to their flexibility of delivery mechanism and cheaper costs of implementation (Bhatia, Fox-Rushby et al. 2004). Also, while most African vector populations predominantly feed indoors, at night (Huho, Killeen et al. 2012), they may not rest on the walls after a blood meal or rest for a period insufficient to pick up a lethal dose

of the active insecticide (Pates and Curtis 2005). However, for LLINs to be fully effective they require deliberate active participation of individuals to use them consistently and appropriately, in addition to them being regularly replaced and kept in good repair (Rehman, Kleinschmidt et al. 2011; Mejía, Teklehaimanot et al. 2013). In contrast, IRS requires only initial consent by the community to have their houses sprayed and compliance with not painting or plastering over the sprayed walls for the expected duration of efficacy of the insecticide used. Additionally, a major advantage of IRS over LLINs is simply that the treated surfaces are rarely in direct contact with occupants of protected houses so the safety requirements for active ingredients that may be used are far less stringent and a much wider variety of active ingredients can therefore be used (Rehman, Kleinschmidt et al. 2011). The evidence on the effects of combining IRS and LLINs varies, with some studies suggesting an incremental benefit of using both interventions (Kleinschmidt, Schwabe et al. 2009; Fullman, Burstein et al. 2013), while others suggest that IRS adds no incremental impact relative to LLINs alone and/or vice versa (Curtis, Maxwell et al. 1998; Corbel, Akogbeto et al. 2012), that LLINs alone have greater impact than IRS (Curtis 1999; Mnzava, Dlamini et al. 1999) and others again indicate that the contrary is true (Misra, Webber et al. 1999; Mabaso, Sharp et al. 2004). These diverse comparisons between IRS and LLINs are based on a variety of outcome measures which include impacts on vector densities or entomological inoculation rates, as well as prevalence, incidence or diagnostic positivity of parasitaemia among humans, as well as the relevant costs of providing such protection (Curtis, Maxwell et al. 1998 ; Curtis 1999; Misra, Webber et al. 1999; Conteh, Sharp et al. 2004; Mabaso, Sharp et al. 2004).

Currently there are four classes of insecticides approved for use in IRS formats: organochlorines, organophosphates, carbamates and pyrethroids (Najera and Zaim 2001),

but only the latter are considered safe enough for use in LLINs. The wide-scale deployment of pyrethroids in both LLIN and IRS formats has undoubtedly exerted considerable selection pressure upon vector populations, resulting in the rapid and widespread emergence of physiological resistance to these active ingredients which may negatively influence the efficacy of LLINs in particular (WHO 1976; WHO 1992; Hargreaves, Koekemoer et al. 2000; Curtis, Jana-Kara et al. 2003). As a consequence, the WHO recommends a reduction in use of pyrethroids for IRS, particularly in areas where LLIN deployment has been scaled up to reach high coverage (WHO 1976; WHO 1992). Furthermore, IRS application of multiple insecticides from different classes, ideally with complementary modes of action and non-overlapping resistance mechanisms, in rotations or mosaics is recommended as the optimal means of insecticide resistance management in the short-to-medium term (WHO 2012). Unfortunately, the utilization of organochlorines for IRS, particularly DDT, has been discouraged and scaled down due to concerns about potentially negative environmental effects associated with their use (Eskenazi, Chevrier et al. 2009). The remaining recommended formulations of organophosphates and carbamates have not been extensively used in IRS programs due to their comparatively high cost and relatively short residual periods of approximately 2 to 6 months (Najera and Zaim 2001), which necessitates spraying more than once in areas with protracted transmission seasons or perennial transmission. Fortunately, new formulations of the organophosphate pirimiphosmethyl (PM) have recently reformulated been brought to market for public health use that appear to offer increased and prolonged efficacy, notably against pyrethroid-resistant vectors (Rowland, Boko et al. 2013; Tangena, Adiamoh et al. 2013).

Given the substantial additional cost of supplementing LLINs with IRS, especially with such expensive new insecticides, and the persisting controversy about whether incremental

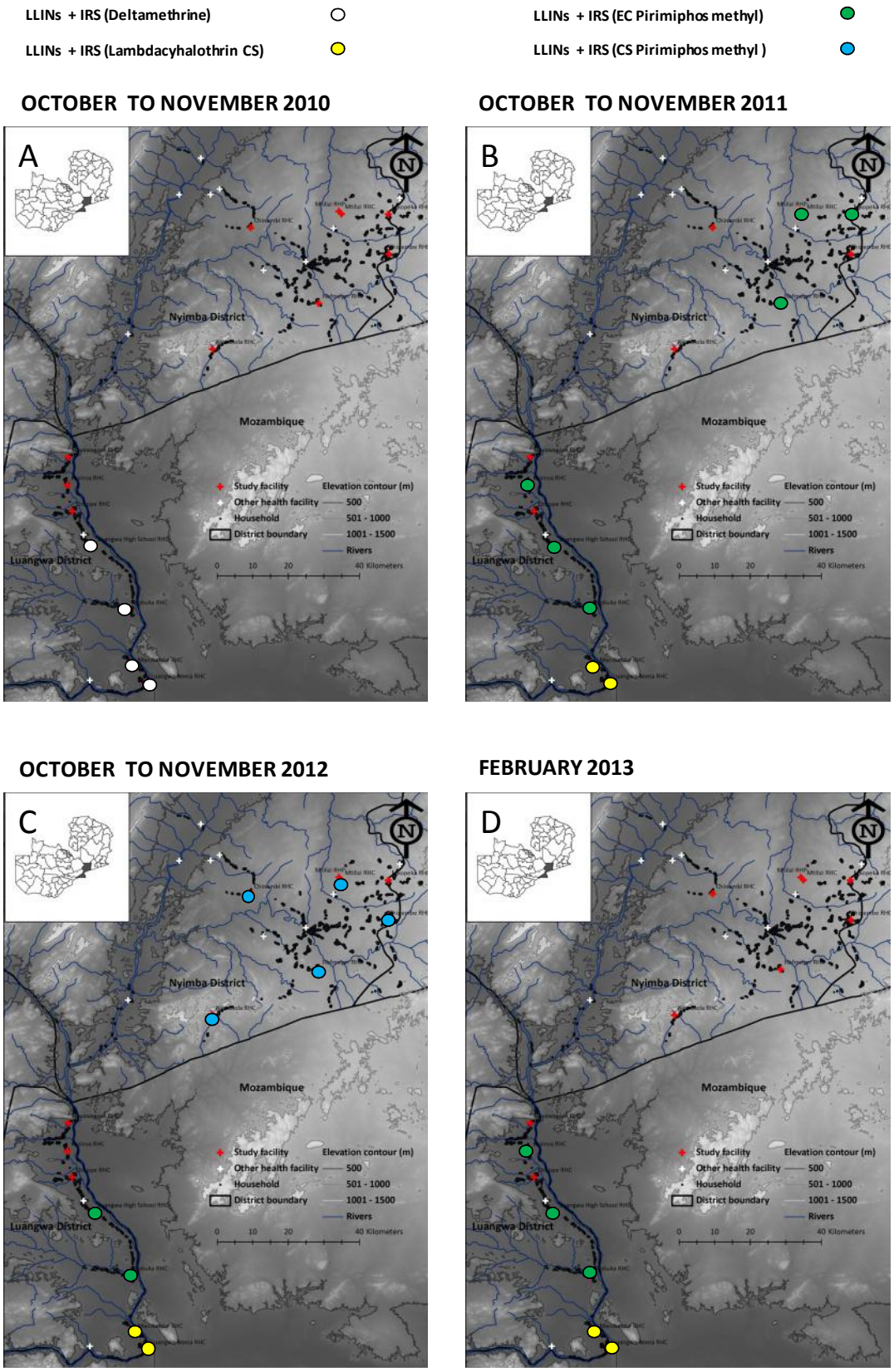
protection against malaria is accrued, it is important to directly evaluate such combinations at community-level with epidemiological primary outcomes and explanatory entomological secondary outcomes in representative malaria-endemic settings. Thus, the overall aim of the study was therefore to evaluate the incremental impact of supplementary vector control with IRS upon malaria transmission by the widespread and highly efficient African vector *An. funestus* in a study area with relatively high usage rates of pyrethroid-impregnated LLINs, using either, one of two different formulations of pyrethroids, or one of two different formulations of the new PM organophosphate.

4.2 Methods

4.2.1 Study area

The study was conducted in the predominantly rural districts of Luangwa and Nyimba, located in Lusaka and Eastern provinces, respectively, of the Republic of Zambia (Figure 26). These districts have perennial transmission of *Plasmodium falciparum*, with the overwhelmingly predominant vector being *Anopheles funestus* Giles, which mediates a mean entomological inoculation rate (EIR) for non-users of LLINs of approximately 70 infectious bites per unprotected person per year (Sikaala, Chinula et al. 2014).The district of Luangwa (3,468 km^2) is located 350-500 meters above sea level, 325 km south-east of Lusaka the capital city of Zambia. It has a population of approximately 27,560 residents, with an annual growth rate of 2.9% (CSO 2011). The main economic activities in the district are fishing and agriculture. Nyimba is a larger district (10,943 km^2), with an approximate population of 108,637 inhabitants and an annual growth rate of 3.4 % (CSO 2011). The district is located 400-1200 meters above sea level, 350 kilometres east of Lusaka. Agriculture is the predominant economic activity in Nyimba district.

Figure 26.Map indicating location of health facilities and associated catchment populations enrolled in the study, with allocation of IRS treatments per cluster and year



4.2.2 Study design

In each district, 7 clusters of approximately 165 households were selected and enrolled in the study to participate in longitudinal parasite surveys (Hamainza, Moonga et al. 2014). Of these, 15 households in each cluster were selected and enrolled at the discretion of the CHW, so that they were geographically distributed across the cluster, for participation in monthly entomological observations, with the exception of Luangwa High School where 30 household were enrolled. Both parasitological and entomological assessments were conducted continuously from January 2011 to March 2013 in Luangwa and from April 2011 to March 2013 in Nyimba district in all clusters.

The pyrethroid Deltamethrin (Wetable granule (WG) formulation) was sprayed in all consenting households at the four southernmost clusters in Luangwa in October 2010, immediately before that year's rainy season and initiation of this study. During the study period, 3 other selected IRS insecticide treatments (capsule suspension (CS) formulation of the pyrethroid lambdacyhalothrin, as well as the emulsifiable concentrate (EC) and CS formulations of the organophosphate pirimiphosmethyl) were randomly allocated to clusters in advance of each rainy season. In practice this randomized allocation was not strictly adhered to by the implementation agencies in the two districts (District Medical Office (DMO) in Luangwa and Abt Associates under the supervision of the DMO in Nyimba), thus resulting in a quasi-randomised study design, described cartographically in Figure 26. The parasitological and entomological surveys were conducted by paid community health workers (CHWs) as previously described in detail (Hamainza, Moonga et al. 2014; Sikaala, Chinula et al. 2014) and summarized below. In the south of Luangwa district, between October and November 2010, clusters 4, 5, 6 and 7 received pyrethroid-based IRS with deltamethrin (K-Othrine WG® 250, Bayer Environmental Science, South Africa) as described

in Figure 26A. Subsequently, the organophosphate pirimiphosmethyl was introduced as an alternative insecticide for IRS in a response to detection of resistance to pyrethroids in the primary vector, *An. funestus* in Luangwa district (Chanda, Hemingway et al. 2011; Seyoum, Sikaala et al. 2012; Sikaala, Killeen et al. 2013). The only formulation of pirimiphosmethyl that was available at the time was the relatively short-lived (Rowland, Boko et al. 2013; Oxborough, Kitau et al. 2014) emulsifiable concentrate (EC) formulation (Actellic® EC, Syngenta Crop Protection AG, Switzerland). This formulation was sprayed during the months of October and November 2011, in clusters 2, 4, 5 in Luangwa and 9, 11, 13 in Nyimba, while IRS with pyrethroid lambdacyhalothrin (Icon® 10 Capsule Suspension (CS) formulation, Syngenta Crop Protection AG, Switzerland) was applied in only two of the four clusters in the south of Luangwa district which had been sprayed with deltamethrin the previous year, specifically in clusters 6 and 7 (Figure 26B). The following year, in November 2012, the longer-lasting microencapsulated formulation of pirimiphosmethyl (Actellic® 300CS, Syngenta Crop Protection AG, South Africa) was applied in clusters 8, 9, 10, 12 and 14, all of which were in Nyimba district (Figure 26C). In February 2013, IRS in Luangwa district was implemented with PM-EC in clusters 2, 4 and 5 while clusters 6 and 7 received the CS formulation of lambdacyhalothrin (Figure 26D).

4.2.3 Parasitological surveys of human infection

Active monthly parasitological surveys were coupled with questionnaires recording clinical symptoms of illness, as well as access and utilization of preventive measures such as LLINs, IRS and intermittent preventive therapy (IPT), between January 2011 and May 2013, spanning a period of approximately 29 months, as described in detail previously (Hamainza, Moonga et al. 2014). These surveys were conducted by paid CHWs who made active monthly visits to households that consented to participate in the study. In between active

visits, study participants who developed symptoms were encouraged to seek care through passively offered diagnosis and treatment services, either from the CHWs at their place of residence or at the nearest health facility. The rapid diagnostic test used in the study was manufactured by ICT Diagnostics to detect circulating *P. falciparum* histidine-rich protein-2 antigen (ICT Malaria P.f. cassette test). All participants that were found positive for antigenemia, which was presumed equivalent to infection, were treated with Artemether-Lumefantrine as per National Malaria Diagnosis and Treatment Policy (NMCC 2010). In both the active and passive visits, all participants found to be negative for malaria infection but febrile or had any other complaints were referred to the nearest HF.

4.2.4 Mosquito densities, species identification

The monthly mosquito collections were conducted by paid Community Health Workers (CHWs) using Centres for Disease Control and Prevention light traps (LT) and Ifakara Tent Traps (ITT) between January 2010 and April 2013, spanning a period of approximately 28 months, as described in detail previously (Sikaala, Chinula et al. 2014). In each consenting household, the LTs were placed at the foot end of an occupied sleeping space covered with an LLIN, hanging approximately 1.5m above the floor. An ITT was placed immediately outside, approximately 5 meters away from the house where the LT was installed and was occupied by an adult male volunteer from the same household. All the mosquito traps were set up in the evenings and collection of the captured mosquitoes was done in the early morning by aspiration. All the collected mosquitoes were initially sorted in the field to genus level, by the CHWs, based on crude taxonomic features and then stored over silica until they were collected on a monthly basis and transported to a central laboratory at the NMCC for further detailed examination. Additional morphological identification of *Anopheles* to species group or complex (Gillies and Coetzee 1987) was conducted at the central

laboratory of the NMCC in Lusaka. Polymerase Chain Reaction (PCR) for the identification of species within the *An. funestus* group (Koekemoer, Lochouarn et al. 1999) or *An. gambiae* complex (Scott, Brogdon et al. 1993) were conducted on selected samples in the NMCC laboratory.

4.2.5 Vector susceptibility to different classes of insecticides

A team of trained entomological technicians from the NMCC periodically collected samples from the study sites to ascertain the susceptibility of the mosquitoes to different classes of insecticides, as background descriptive data to support appropriate interpretation of apparent impacts of various supplementary IRS treatments upon the vector population. In Luangwa district, mosquitoes were collected from cluster 2 from 2010 to 2013. However, in Nyimba district, collections were done over three years in different clusters (Cluster 14 in 2011, cluster 9 in 2012 and cluster 13 in 2013). Adult mosquitoes were either collected while attacking humans by human landing catch (HLC) or by using pack aspirators for the indoors wall resting mosquitoes. These were collected in cups covered with a netting material and placed in cooler box for transportation to the NMCC insectary where individual female *Anopheles funestus* mosquitoes where allowed to feed on mouse blood so they could lay eggs that were then reared into F1 generation mosquitoes. Standard World Health Organization (WHO) susceptibility tests using insecticide-impregnated papers with discriminatory dosages of two pyrethroids (Deltamethrin 0.05%, and Lambdacyhalothrin 0.05%), a carbamate (Bendiocarb 0.1%), an organophosphate (Malathion 0.4%) and an organochlorine (DDT 4%) were carried out on 1 to 3 day old F1 *An. funestus* mosquitoes. Control papers were impregnated with oil as directed by the WHO protocol (WHO 2013). Knock down and mortality rates after 1 hour and 24 hour post exposure periods were recorded.

4.2.6 Indoor-outdoor distribution of human exposure to *An. funestus* bites

To estimate proportions of human exposure to *An. funestus* bites and malaria transmission that occurs indoors and outdoors, HLCs were conducted both indoors and outdoors by a team of trained entomological technicians from the NMCC in Lusaka and these were complemented by cross-sectional questionnaire surveys of when residents went indoors for the night, went to sleep, awoke in the morning and left the house in the morning, as previous described in detail elsewhere (Seyoum, Sikaala et al. 2012), again as background descriptive data to support appropriate interpretation of apparent impacts of various supplementary IRS treatments upon the vector population and malaria transmission. Trained CHWs conducted HLC from 6pm to 6am the next morning, with the exception of the previously described 2010 studies where the starting time was 7pm and finishing at 7am. The 2010 and 2011 HLC surveys were conducted in cluster 4 (Chisobe and Nyamumba villages of Luangwa district) as part of a trap effectiveness study (Sikaala, Killeen et al. 2013), while those conducted in 2012 and 2013 where part of the quality assurance surveys conducted in 13 clusters as part of a subsequent effectiveness assessment for a community-based trapping scheme (Sikaala, Chinula et al. 2014). Mosquitoes were collected for 45 minutes per hour to allow a 15 minutes break for rest and refreshment to the collectors. Each hourly collection were labelled and kept for identifications to genus and species as described above. The proportion of time residents spent whilst outdoors and indoors, as well as asleep in bed, was estimated directly from answers to questionnaires during a cross-sectional household survey in April 2010 in Luangwa district, in which people indicated the time they usually went indoors and when they went to the bed as well as when they arose in the morning and when they left their houses in (Seyoum, Sikaala et al. 2012)

4.2.7 Data management and statistical analysis

The CHW malaria register data describing RDT results associated questionnaire responses were double entered into Excel®, verified, reconciled and then cleaned following descriptive frequency analysis of the distributions of values for each variable. All entomological data were single entered, verified and cleaned prior to analysis. All statistical analyses were accomplished using SPSS version 20 (IBM) and R version 2.14.1, augmented with the lattice, Matrix and LME4 packages.

4.2.7.1 Incremental protection of humans against malaria infection risk by IRS treatments

Previous analyses of these data collected by CHWs have demonstrated that diagnostic positivity (DP) for malaria infection, expressed as the proportion of RDT-tested individuals who were found to be positive, was a extremely powerful indicator of malaria risk that allowed numerous important epidemiological phenomena to be clearly illustrated (Hamainza, Moonga et al. 2014). It also proved to be a more consistent and robust indicator of geographic and temporal variation than absolute numbers of malaria infections detected, presumably because variations in CHW service utilization rates, as well as RDT and ACT availability, occur in both the nominator and denominator of DP (Hamainza, Killeen et al. 2014) and was therefore treated as the primary epidemiological outcome used for statistical analysis of the effects of various IRS treatments, rather than incidence in terms of detected events per number of participants per unit time.

Four sequential time period categories, based on the integer number of months since the most recent spray round was completed were created for all the IRS treatments: 1 to 3 months, 4 to 6 months and 7 to 12 months since beginning of the last spray round started, as well as a fifth category combining areas that had not yet received spraying during the

study period and those for which the last spray round began more than 12 months ago, which was treated as the reference value. Generalized linear mixed models (GLMMs) were fitted to evaluate the association between observed malaria infection risk among human residents and the various IRS treatments applied. Malaria infection status was treated as the binary dependent, with use of an LLIN, having slept in a house that had been treated with IRS in the previous 6 months and the categorised cluster-wide IRS treatments as the independent variables of primary interest. Age category (<1, 1 to 4, 5 to 10, 11 to 14, 15 to 24, 25 to 44 and >45 years of age), sex, season (hot and wet from December to April, cool and dry from May to August, and hot and dry from September to November), number of previous RDT tests conducted per individual and geographical location (cluster) were also included as independent variables of secondary interest (all categorical except for number of RDT tests) while random effects to capture variance associated with nuisance variables of no direct interest were also included in the model (the individual identity number nested within the CHW catchment nested within the study cluster, as well as date of participant contact). Further, in order to test for and quantify incremental impact of PM IRS as a supplement to LLINs, relative to LLINs supplemented with pyrethroid-based IRS, both pyrethroid formulations were represented by a single treatment variable, coding the same periods of months since before spraying. Similarly, in order to test for and quantify the incremental impact of the CS formulation of PM, relative to the EC formulation of the same active ingredient, as well as the two pyrethroid formulations, an additional variable was created which combined any previous treatment with any of the latter three formulations in the reference group. In all cases, incremental protective efficacy (IPE) was calculated as the complement of the odds ratio (OR) estimated directly by these GLMMs (IPE = 1-OR).

4.2.7.2 Incremental protection of humans against human exposure to mosquito bites and malaria parasite inoculation by IRS treatments

The effect of different IRS treatment regimens on densities of *An. funestus* species were estimated by fitting GLMMs where *An. funestus* densities were treated as a dependent variable with a Poisson distribution. In order to account for variance in mosquito densities by location, identities for households where nested within those villages and then nested within clusters as random effects. Similarly, nightly temporal variance in vector density was accounted for by including date as an additional random effect. While the presence or absence of open eaves is a simple binary independent variable with only two levels, it was nevertheless treated as a random effect because it is not of direct interest to this analysis so that the other independent variables represent values for the mean of all houses with and without open eaves, rather than the mean for a reference condition, presumably absence of eaves. The different IRS treatment regimens were coded in terms of time period since the last round of IRS application began, exactly as described above for the epidemiological primary outcomes, so that these treatments could be included as categorical independent variables with which to detect and quantify impact upon these entomological secondary outcomes. In all the relative rate (RR) at which mosquitoes were captured, was calculated as estimated directly by these GLMMs. Unfortunately, efforts to develop laboratory capacity for determining sporozoite infection status by enzyme-linked immunosorbent assay at NMCC were unsuccessful so neither sporozoite prevalence nor entomological inoculation rate could be assessed as additional entomological secondary outcomes.

4.2.8 Physiological resistance to insecticides

Insecticide susceptibility assays were conducted on 2 – 5 day old F1 generation *An. funestus* as described by the WHO standard protocol (WHO 1998) using papers impregnated with

Deltamethrin (0.05%), Lambdacyhalothrin (0.05%), Bendiocarb (0.1%), Malathion (0.1%) or DDT (4%).

In order to test for time trends in physiological resistance of *An. funestus* to pyrethroids and carbamates over time, survival status of mosquitoes exposed to these insecticides in standard WHO protocols (WHO 2013) was treated as the binary outcome variable in GLMMs with, year as a continuous covariate, and a unique identification code for each experimental replicate as a random effect. The data were stratified into subsets on the basis of the insecticide class with separate models fitted for the carbamate (Bendiocarb), and the combined pyrethroids (Deltamethrin and Lambdacyhalothrin). The model of resistance time trends for the two pyrethroids, the identities of these two insecticides within this class were included as a categorical independent variable. No such model was fitted for either the organochlorine (DDT) or the organophosphate (Malathion) because no resistance to either insecticide was apparent.

4.2.9 Proportions of human exposure to *An. funestus* bites occurring indoors and outdoors

The distribution of human exposure to *An. funestus* bites, and presumably malaria transmission, across different times of the night and across indoor and outdoor compartments of their living environment was calculated by weighting HLC measurements of indoor and outdoor biting rates for each hour of the night by the estimated proportion of humans indoors and outdoors during that time period, exactly as previously described (Seyoum, Sikaala et al. 2012). These estimates of human exposure distribution across indoor and outdoor environments were calculated and presented graphically for both users and non-users of LLINs, so that the proportions of human exposure that occur indoors in the presence ($\pi_{i,n}$) and absence (π_i) of a protective LLIN could be quantified and visualized.

4.2.10 Protection of human participants and ethical approval

Prior to the study, community sensitization was conducted and permission obtained from the local community leadership. Informed consent was obtained from all study participants during all surveys and spraying activities. The study team ensured that all treatment and diagnostic protocols were adhered to and that patients requiring malaria treatment received it promptly or were referred to the nearest health facility. All standard safety protocols were adhered to during the process of IRS as per national guidelines. Ethical approval was obtained from the University of Zambia, Biomedical Research Ethics Committee (Reference 004-05-09) and the Research Ethics Committee of the Liverpool School of Tropical Medicine (Approval 09.60). Authority to conduct and publish the study was also obtained from the Ministry of Health in Lusaka, Zambia.

4.3 Results

4.3.1 Characteristics of study participants and survey clusters

A total population of 25,354 people centred around HFs in the 14 clusters participated in the study and were followed up for a period of 29 months in Luangwa and 26 months in Nyimba, starting from January 2011 and April 2011, respectively. Out of these participants, 29% (7412) were children under the age of 5 but DP peaked in older children between the age of 5 and 10. The overall cluster populations ranged from 1,158 to 3,429. A total of 31,974 malaria infections (21.7% DP) were identified, which translates into an incidence of 9 infections per 100 person years. The study population reported a relatively high average rate of LLIN utilization of 81.7% of questionnaire responses over the course of the study indicating that the respondent had slept under an LLIN the previous night, while 39.2% of participant questionnaire responses indicated that the respondent's house had been

treated by IRS in the last six months. During same overall study period mean DP by cluster across all age groups and other potential stratification criteria ranged from 6.4% to 41.9% (mean = 24.5%), with the lowest being in the southern urban cluster and the highest in the northern rural cluster (Table 15). The potential confounding effect of LLIN ownership was excluded from the model because it had no significant effect (P>0.05) on diagnostic positivity (Table 15).

Table 15.Association of malaria infection status with age, sex, LLINs, IRS, number of tests conducted per participant, geographical location, season and IRS insecticide used [x xi]

Category	DP%[a]	n/N[b](I)	OR[95%CI][c]	P[d]
Overall	21.7	31974 /147257(25354)	0.13 [0.08,0.21]	<0.001
Age				
<1	14.2	501 /3535 (1735)	1.26[1.09,1.45]	0.001
1-4	24.0	6127 /25505 (5677)	2.75[2.54,2.98]	<0.001
5-10	27.4	10066 /36779 (7608)	3.62[3.35,3.91]	<0.001
11-14	26.0	4892 /18840 (4746)	3.36[3.09,3.65]	<0.001
15-24	20.3	4491 /22077 (5685)	2.04[1.88,2.22]	<0.001
25-44	14.9	4028 /27044 (5807)	1.24[1.14,1.34]	<0.001
≥45	13.8	1796 /13027 (2903)	1[NA]	NA
Sex				
Male	23.3	16068 /79208 (12008)	1[NA]	NA
Female	20.3	15750 /67567 (13228)	0.86[0.83,0.90]	<0.001
Interventions				
LLINs	20.0	20613/103149 (20706)	0.89[0.85,0.93]	<0.001
IRS	17.4	7568 /43560 (9926)	0.87[0.82,0.93]	<0.001
Number of tests conducted per participant	21.7[e]	31974 /147257(25354)	0.97[0.97,0.98]	<0.001
Type of visit				
Passive	43.4	6416 /14785 (8922)	1[NA]	NA
Active	19.2	25281 /131359 (22055)	0.29[0.28,0.31]	<0.001
Clusters				
Luangwa district				
Sinyawagora RHC	19.7	1314 /6655 (1959)	2.86[1.65,4.97]	<0.001
Kasinsa RHC	16.7	2232 /13402 (3429)	4.67[2.78,7.84]	<0.001
Chitope RHC	19.6	3419 /17463 (1215)	2.92[2.04,4.17]	<0.001
Luangwa High School RHC	16.5	2854 /17320 (1158)	7.37[4.31,12.61]	<0.001
Mphuka RHC	24.9	2981 /11957 (2147)	7.37[4.31,12.61]	<0.001
Mandombe RHC	10.3	1386 /13508 (1805)	1.54[0.89,2.66]	0.119
Luangwa Boma RHC	6.4	839 /13161 (2033)	1[NA]	NA
Nyimba district				
Kacholola RHC	26.7	3108 /11654 (1166)	7.50[4.75,11.84]	<0.001
Hofmeyer RHC	41.9	2601 /6214 (2120)	15.81[10.20,24.52]	<0.001
Mtilizi RHC	37.6	2238 /5949 (2024)	12.35[7.72,19.76]	<0.001
Mtilizi RHP	25.3	2478 /9788 (3379)	13.49[8.45,21.56]	<0.001
Chinambi RHC	31.9	1740 /5463 (1741)	9.16[5.79,14.48]	<0.001
Mkopeka RHC	32.8	2761 /8413 (1311)	14.22[8.55,23.63]	<0.001
Chipembe RHC	32.1	2023 /6310 (1916)	13.54[8.03,22.84]	<0.001

Season				
Hot & wet (Dec – April)	25.3	18283 /72217 (20243)	4.20[3.67,4.81]	<0.001
Cool & dry (May - Aug)	23.9	11216 /46860 (16513)	3.25[2.80,3.76]	<0.001
Hot & dry (Sept – Nov)	8.7	2444/ 27983 (12590)	1[NA]	NA
Insecticide applied for IRS				
Deltamethrine				
1-3 months since last spray	13.3	322 /2419 (2166)	0.81[0.65,1.01]	0.064
4-6 months since last spray	23.8	2411 /10150 (4231)	1.07[0.94,1.23]	0.295
7-12 months since last spray	13.6	2128/15640 (4434)	1.16[1.03,1.30]	0.013
Never sprayed and >13 months since last spray	22.8	27083/118899 (23233)	1[NA]	NA
Lambdacyhalothrin				
1-3 months since last spray	4.7	145/3102 (1526)	0.69[0.53,0.90]	0.006
4-6 months since last spray	9.4	207/2199 (1264)	1.26[1.01,1.57]	0.042
7-12 months since last spray	4.5	157/3508 (1469)	0.94[0.74,1.21]	0.653
Never sprayed and >13 months since last spray	22.7	31435/138299 (24931)	1[NA]	NA
Primiphosmethyl EC				
1-3 months since last spray	18.9	1922/10194 (5527)	0.77[0.69,0.85]	<0.001
4-6 months since last spray	28.8	2666/9259 (5926)	0.64[0.58,0.71]	<0.001
7-12 months since last spray	16.0	1793/11184 (5760)	0.63[0.56,0.71]	<0.001
Never sprayed and >13 months since last spray	21.9	25563/116471 (22311)	1[NA]	NA
Primiphosmethyl CS				
1-3 months since last spray	13.0	468/3590 (2675)	0.37[0.31,0.43]	<0.001
4-6 months since last spray	30.6	1386/4536 (3349)	0.24[0.21,0.27]	<0.001
7-12 months since last spray	49.5	95/192 (191)	1.35[0.85,2.15]	0.204
Never sprayed and >13 months since last spray	21.6	29995/138790 (24588)	1[NA]	NA

[x] a – Diagnostic positivity, b – (n – Number RDT positive, N – Total number), I – number of individuals that participated, c – odds ratio with 95% confidence intervals, d – p-value, e - RDT determined diagnostic positivity at first, NA –Not applicable /reference group

[xi] The association of malaria infection with age, sex, use of LLINs, use of IRS, geographical location (cluster), number of tests conducted per participant, season and insecticide used in IRS was determined using GLMM; with observed malaria RDT determined status as a binary dependent outcome with the independent categories of age, sex, access and use of LLINs or IRS, insecticide used in IRS, number of tested conducted per participant and seasons. The models included date and participant nested within CHW catchment nested within geographical location (cluster) as random effects except for one in which cluster was treated as a categorical variable to determine the effects of each cluster. The final model consisted of age, sex, access and use of LLINs or IRS, insecticide used in IRS, season, number of tests conducted per participant and geographical location as the determinants of malaria infection.

Table 16: IRS coverage, LLIN utilization and Diagnostic Positivity Snapshot 1 to 6 months post IRS implementation by cluster.

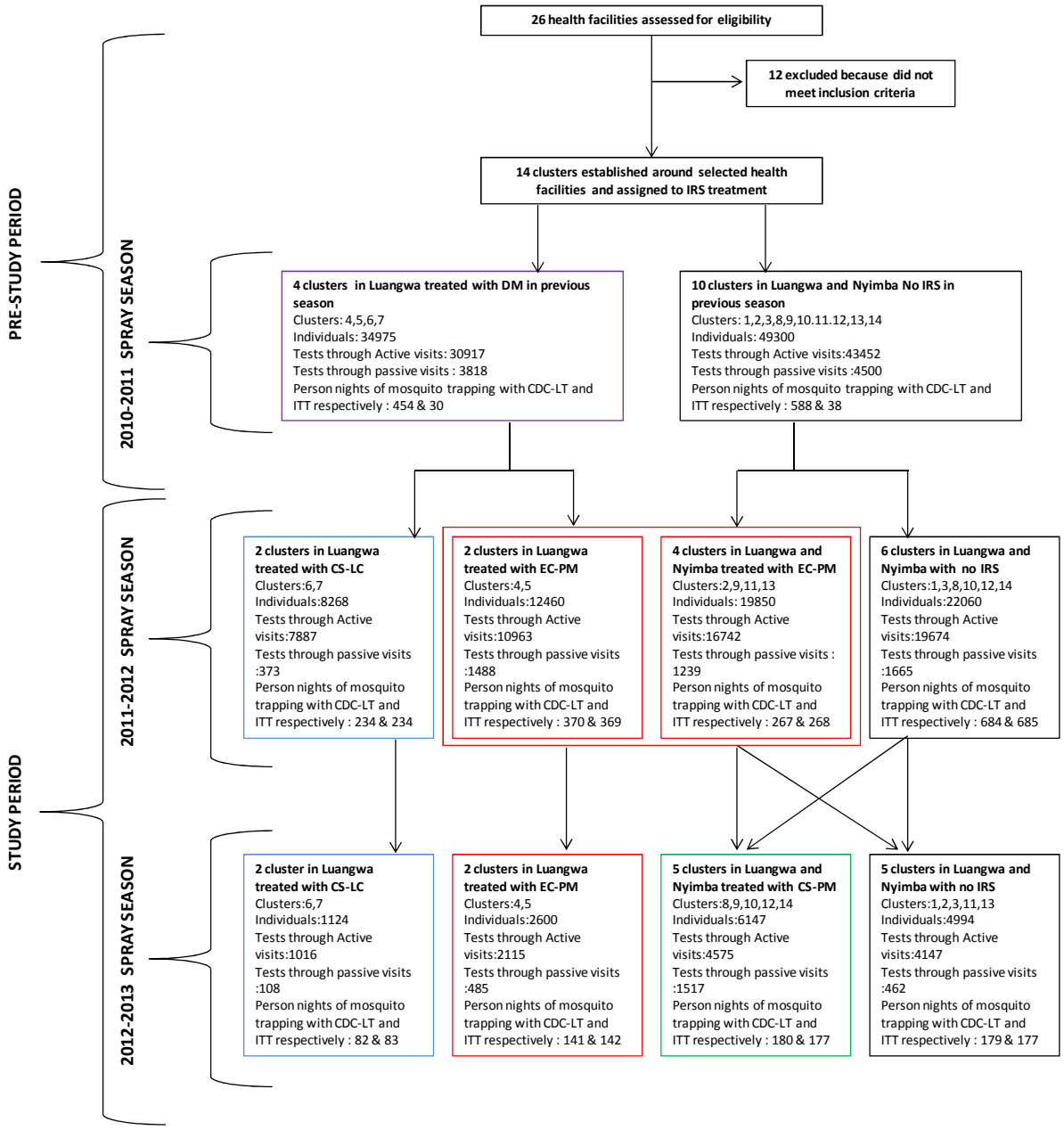
Period	Cluster	IRS Status	IRS Coverage % (n/N)	LLINs use % (n/N)	Diagnostic positivity % (n/N)
October 2010 to March 2011	1	None	0.0 (0/1577)	65.1 (1025/1575)	24.7 (372/1508)
	2	None	0.0 (0/827)	98.0 (2485/2537)	20.9 (559/2676)
	3	None	0.0 (0/2182)	49.0 (1492/3046)	26.9 (809/3006)
	4	Deltamethrin WG	72.0 (1920/2667)	82.3 (2194/2667)	33.2 (825/2489)
	5	Deltamethrin WG	92.4 (1362/1474)	95.8 (1412/1474)	27.5 (396/1439)
	6	Deltamethrin WG	95.3 (2750/2886)	95.4 (2744/2876)	11.9 (338/2845)
	7	Deltamethrin WG	98.5 (2358/2395)	99.7 (2467/2474)	6.0 (144/2415)
	8	None	0.0 (0/424)	6.6 (28/426)	55.7 (202/363)
	9	None	0.0 (0/7)	36.4 (4/11)	36.4 (4/11)
	10	None	0.0 (0/343)	39.7 (136/343)	50.7 (172/339)
	11	None	0.0 (0/134)	32.1 (43/134)	51.3 (60/117)
	12	None	0.0 (0/49)	79.6 (39/49)	61.9 (26/42)
	13	None	0.0 (0/221)	30.8 (68/221)	60.0 (120/200)
	14	None	0.0 (0/77)	41.6 (32/77)	52.4 (33/63)
October 2011 to March 2012	1	None	0.0 (0/1105)	87.4 (1063/1217)	9.5 (95/998)
	2	Pirimiphosmethyl EC	10.3 (245/2380)	75.8 (2235/2948)	8.5 (280/3292)
	3	None	0.0 (0/2593)	33.1 (1253/3790)	10.8 (436/4033)
	4	Pirimiphosmethyl EC	57.9 (2194/3788)	92.9 (3541/3811)	5.9 (217/3708)
	5	Pirimiphosmethyl EC	57.6 (1952/3392)	97.7 (3388/3469)	18.2 (624/3436)
	6	Lambdacyhalothrin CS	80.7 (1128/1398)	80.2 (1139/1421)	5.2 (76/1457)
	7	Lambdacyhalothrin CS	81.8 (2317/2833)	99.9 (3107/3109)	4.2 (130/3111)
	8	None	0.0 (0/3918)	75.4 (2952/2917)	29.9 (974/3261)
	9	Pirimiphosmethyl EC	33.5 (541/1613)	51.95 (838/1613)	46.6 (684/1467)
	10	None	0.0 (0/1466)	55.4 (812/1466)	35.4 (444/1254)
	11	Pirimiphosmethyl EC	1.8 (74/4117)	60.1 (2474/4117)	30.2 (941/3112)
	12	None	0.0 (0/1546)	59.7 (925/1549)	33.9 (514/1517)
	13	Pirimiphosmethyl EC	16.8 (407/2422)	45.9 (1111/2422)	27 (5.033/1974)
	14	None	0.0 (0/1964)	52.1 (1023/1964)	41.3 (786/1904)
October 2012 to March 2013	1	None	0.0 (0/464)	100.0 (464/464)	14.4 (150/1039)
	2	None	0.0 (0/1076)	99.8 (1074/1076)	11.9 (126/1061)
	3	None	0.0 (0/1915)	99.1 (1897/1915)	14.1 (282/2004)
	4	Pirimiphosmethyl EC	100 (1170/1170)	100.0 (2632/2632)	10.8 (314/2908)
	5	Pirimiphosmethyl EC	100 (1387/1387)	99.7 (1725/1730)	27.3 (456/1673)
	6	Lambdacyhalothrin CS	100 (348/348)	100.0 (1386/1386)	3.8 (57/1505)
	7	Lambdacyhalothrin CS	88.8 (645/726)	100.0 (1123/1123)	2.99 (33/1105)
	8	Pirimiphosmethyl CS	0.7 (18/2439)	28.5(696/2439)	9.0 (209/2321)
	9	Pirimiphosmethyl CS	73.7 (1386/1881)	48.5 (913/1881)	23.5 (366/1561)

10	Pirimiphosmethyl CS	0.7 (10/1379)	85.1 (1174/1379)	27.1 (363/1341)
11	None	0.0 (0/3440)	16.5 (566/3440)	11.9 (300/2531)
12	Pirimiphosmethyl CS	30.3 (346/1143)	82.2 (940/1143)	21.7 (246/1132)
13	None	0.0 (0/2433)	46.4 (1128/2433)	30.6 (666/2180)
14	Pirimiphosmethyl CS	40.4 (574/1421)	38.1 (541/1421)	16.99 (221/1301)

The close associations of DP for *P. falciparum* malaria infection and *An. funestus* density, clinical symptoms of illness, and a variety of other factors of this setting are described in detail elsewhere based on the first year of data collection (Hamainza, Moonga et al. 2014; Sikaala, Chinula et al. 2014). The detailed profile of the study participants, and their survey contacts over the course of the entire study, are summarized in the context of the study design in Figure 27.

A descriptive comparison of summarized data restricted to the period 1 to 6 months post spraying demonstrates variability among study clusters in not only in IRS coverage (Range = 0% to 100%, mean= 29.4%) but also LLIN use (Range = 6.6% to 100%, mean= 68.2%) and diagnostic positivity (Range = 2.99% to 61.9%, mean= 25.4%) (Table 16). Further analysis using Pearson's correlation, revealed a positive but weak association (R^2=0.31) between IRS coverage and LLIN use, suggesting that as IRS coverage increases, so does LLIN use. However, this does not necessarily imply any causal relationship and factors which affect delivery (e.g. accessibility) and acceptance (e.g. attitudes towards malaria or mosquitoes) may well be similar for both of these vector control measures. However, there was no obvious and clear-cut effect of any particular IRS treatment in this crude descriptive comparison (Table 16) so detailed regression modelling analysis was required to detect and estimate the separate impacts of these four different formulations (Figures 28 to 30).

Figure 27. Study profile indicating treatments provided to each cluster with associated timelines, population surveyed and persons nights of mosquito trapping

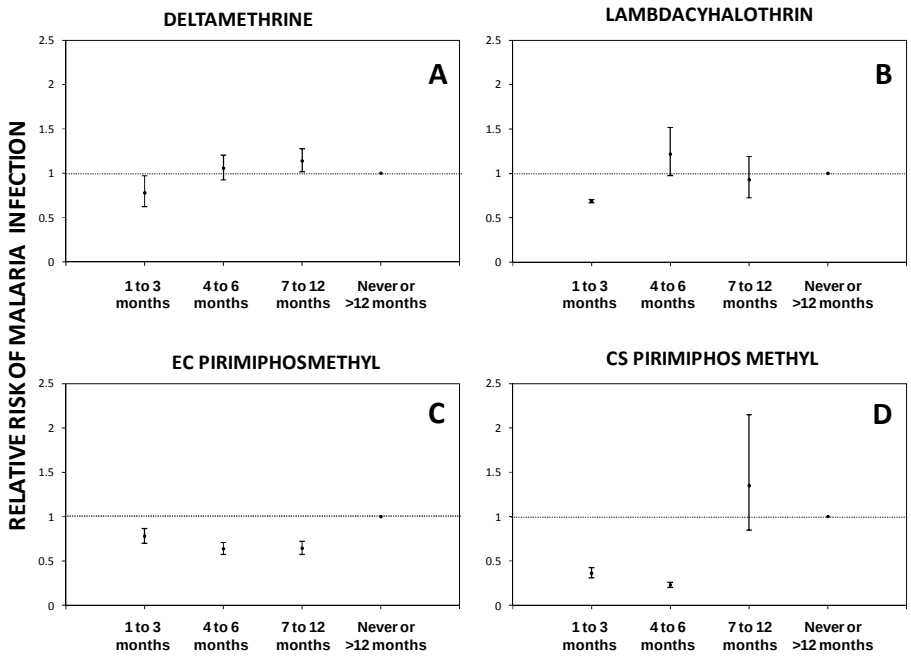


4.3.2 Magnitude and duration of incremental impact of IRS treatments as supplements to LLINs upon human risk of infection with malaria

Reported coverage of deltamethrin WG, lambdacyhalothrin CS, pirimiphosmethyl EC, and pirimiphosmethyl CS, by respondents within the first 3 months after their application in clusters to which they were assigned were 82% (2132/2599),61% (2068/3384), 53%

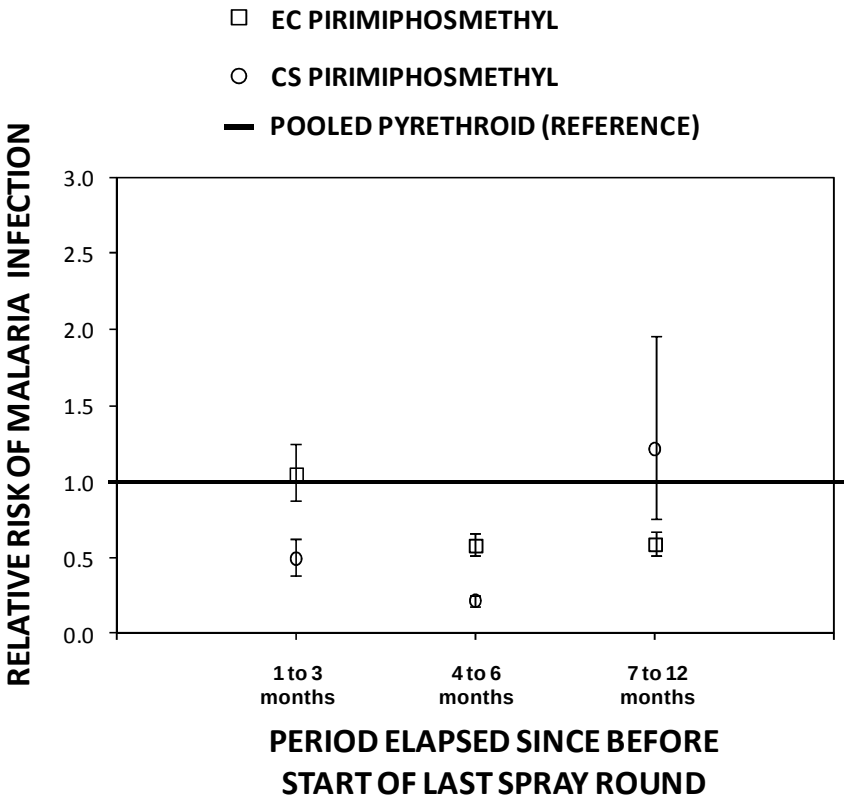
(5909/11078) and 69% (2716/3913), respectively. As illustrated in Figure 28, Primiphosmethyl CS conferred the strongest initial incremental protection in the first 3 months after application (Incremental protective efficacy (IPE) [95% Confidence interval (CI)= 0.63 [0.57, 0.69], P<0.001), relative to LLINs alone, followed by the CS formulation of Lambdacyhalothrin (IPE [95%CI] = 0.31 [10, 47], P=0.006), the EC formulation of primiphosmethyl (IPE [95%CI] = 0.23 [0.15, 0.31], P<0.001) and the WP formulation of Deltamethrin (IPE [95%CI] = 0.19 [-0.01, 0.35], P=0.064). However, neither pyrethroid formulation provided any incremental protection beyond 3 months post-application, while the incremental protection provided by CS and EC formulations of Pirimiphosmethyl persisted undiminished for 6 and 12 months respectively (Figure 28).

Figure 28. The incremental protective efficacy of each of the four IRS treatments on diagnostic positivity for *Plasmodium falciparum* malaria infection over several time periods since the last spray round began, relative to clusters that has either never been sprayed or had last been sprayed >12 months ago (reference group).



In the first three months after spraying, IRS with the CS formulation of pirimiphosmethyl offered greater protection against malaria infection than IRS with pyrethroids IPE [95%CI] = 0.51 [0.38, 0.62], P<0.001 for LLINs+IRS with PM-CS compared to LLINs+IRS in all clusters treated with either DM-WG or LC-CS but not PM-EC (P<0.001). The incremental protection against malaria infection by IRS with both PM formulations outlasted both pyrethroid formulations so that they both offered greater protection from 4 to 6 months post-application IPE [95%CI] = 0.79 [0.75, 0.83], P<0.001 for LLINs+IRS with PM-CS and IPE [95%CI] = 0.42 [0.33, 0.48], P<0.001 for LLINs+IRS with PM-EC, compared to LLINs+IRS with either DM-WG or LC-CS) (Figure 29).

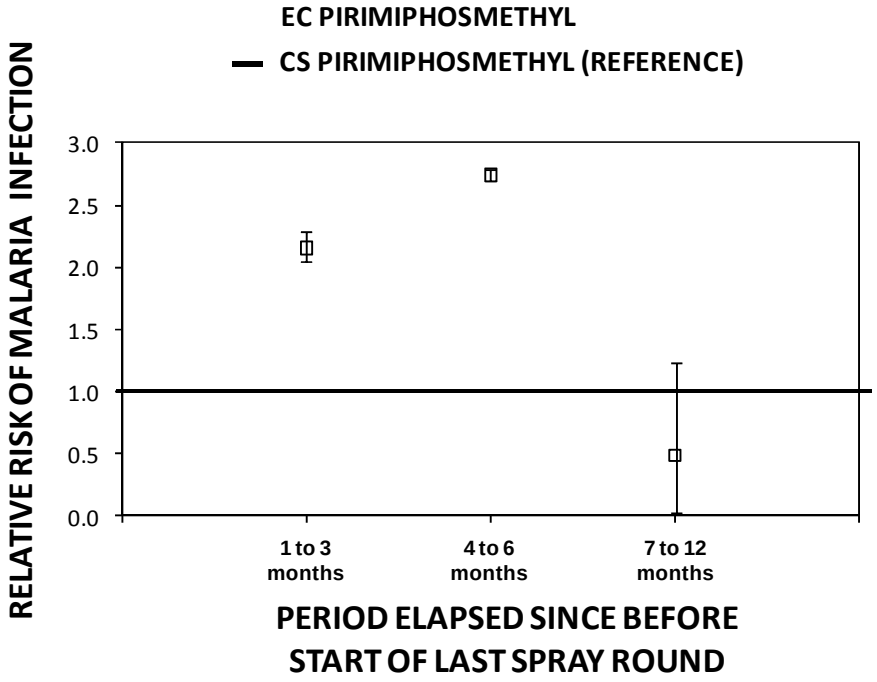
Figure 29. The incremental protective efficacy of primiphosmethyl EC and CS IRS treatments on diagnostic positivity for *Plasmodium falciparum* malaria infection over several time periods since the last spray round began, relative to clusters that have been sprayed with either deltamethrin and /or lambdacyhalothrin (reference group)



Beyond 6 months post-application, LLINs plus IRS with PM-CS provided no apparent incremental protection relative to LLINs alone (P=0.204), much less LLINs+IRS with pyrethroids (P=0.432). However, LLINs+PM-EC continued to provide incremental protection relative to not only LLINs alone (Figure 28), but also relative to all other LLIN+IRS treatments (IPE [95%CI] = 0.41 [0.34, 0.48], P<0.001). When the duration of efficacy of PM-EC was examined in further detail by breaking down the third post-spray time period into two halves, it was clear that it lasted approximately a full year because similar levels of incremental protection was confirmed for both the 7 to 9 month post-spray period (IPE [95%CI] = 0.32 [0.22, 0.40], P<0.001) and the 10 to 12 month post-spray period (IPE [95%CI] = 0.42 [0.31, 0.52], P<0.001).

Comparing these two IRS formulations of PM with each other as supplements to LLINs, the CS formulation confers greater protection than the EC formulation, (IPE [95%CI] = 53.6 [0.43, 0.66] %, P<0.001 from 1 to 3 months post-application and 0.64 [0.57, 0.69], P<0.001 from 4 to 6 months post-application for the contrast between LLINs+PM-CS *versus* the LLIN+PM-EC as the reference group) (Figure 30). However, once the incremental benefit of supplementing LLINs with IRS using PM-CS waned after 6 months, IRS using PM-EC proved statistically superior to all other IRS formulations as supplements to LLINs for a further 6 months, including the CS formulation of the same active ingredient (IPE [95%CI] =0.52 [0.21, 0.70], P<0.001 for the contrast between LLINs+PM-EC *versus* LLIN+PM-CS as a reference group between 7 and 12 months post application).

Figure 30. The incremental protective efficacy of pirimiphosmethyl EC IRS treatment on diagnostic positivity for *Plasmodium falciparum* malaria infection over several time periods since the last spray round began, relative to clusters that have been sprayed with pirimiphosmethyl CS (reference group)



4.3.3 Magnitude and duration of incremental impact of IRS treatments as supplements to LLINs upon human risk of exposure to bites of *An. funestus.*

Detailed description of the local mosquito fauna in the study area (Sikaala, Chinula et al. 2014) showed that 34.5% of all mosquitoes caught over the course of the study were identified morphologically as members of the *An*. *funestus* group, of which 96.5% (575/596) of those which were successfully amplified by PCR were confirmed to be *An. funestus* Giles. Densities of the *An. funestus* group*,* as determined by routine morphological classification can therefore be considered quite reliable as of *An. funestus,* the abundance of which is

consistent with previous studies in this area (Seyoum, Sikaala et al. 2012; Sikaala, Killeen et al. 2013) indicating it as the overwhelmingly dominant vector of malaria in these two districts of Zambia. Therefore, subsequently in this report we refer to all mosquitoes caught from the *An. funestus* group as the nominate species in the strict sense.

The relative rates and the mean catches of *Anopheles funestus* per IRS treatment are presented in Table 17. Relative to the times and places that had never been sprayed, or sprayed or had been sprayed >12 months ago, there were no obvious differences in the densities of *An. funestus* during the first three months post-spraying for both pyrethroid formulations (DM-WG (IPE[95%CI]=0.01[-0.56,0.37],P=0.103) and LC-CS (IPE[95%CI]=-0.03[-0.88,0.44],P=0.195) and PM-EC (IPE[95%CI]=-0.04[-0.30,0.17],P=0.103) (Figure 31, Table 17). However, where PM-CS was applied, mosquito densities were dramatically reduced during the same period of three months immediately after spraying (IPE [95%CI] =0.93[0.87, 0.97], P<0.001). Between the fourth and the sixth month after spraying with DM-WG, there was an apparent, but presumably spurious, three-fold increase in *An. funestus* densities while LC-CS, PM-EC and PM-CS achieved 5, 3 and 71-fold reductions, respectively (Table 17). However, from the seventh to twelfth months after spraying, DM-WG and PM-EC had no obvious effect on the *An. funestus* densities while insufficient data was available to examine the incremental impact of LC-CS or PM-CS.

Figure 31.The incremental protective efficacy of each of the four IRS treatments against *An. funestus* bites over several time periods since the last spray round began, relative to clusters that has either never been sprayed or had last been sprayed >12 months ago (reference group)

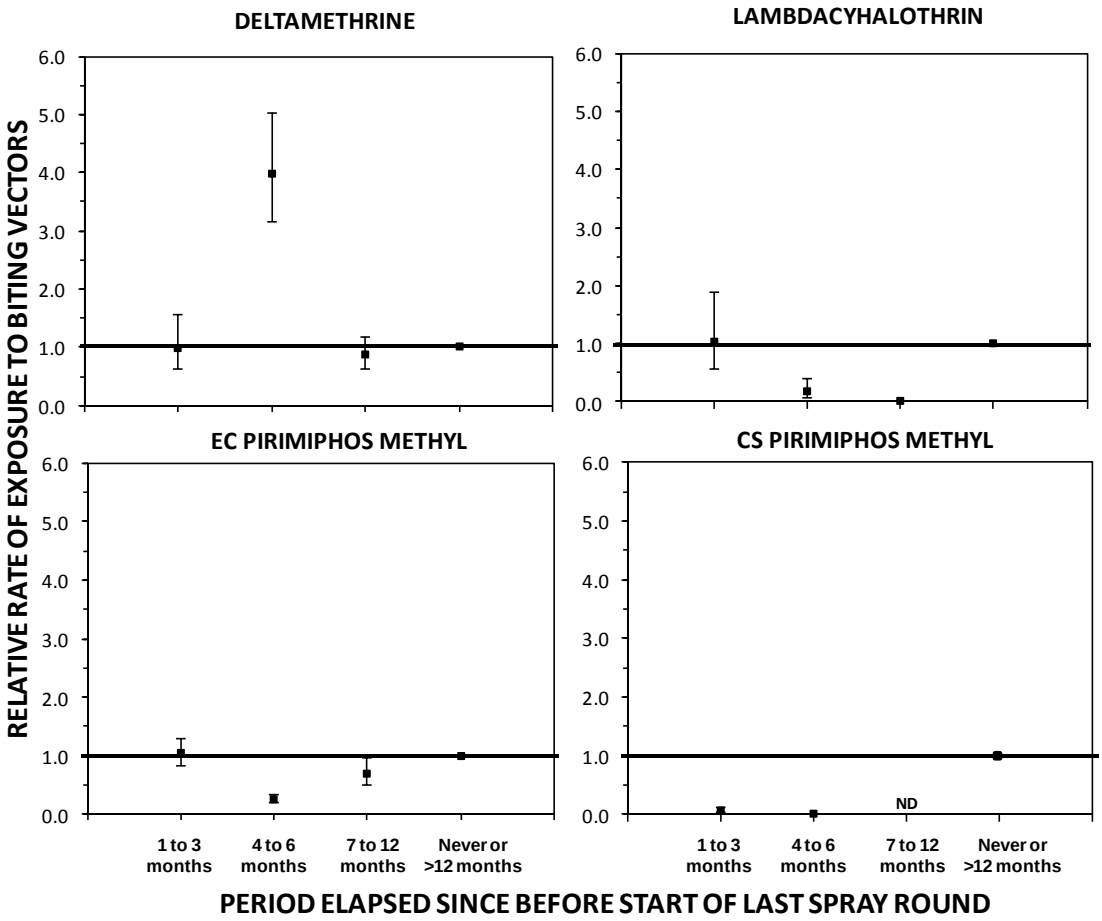


Table 17.Association of *Anopheles funestus* densities with different IRS insecticides supplementing LLINs upon months before, during and when not spraying

Indoor residual spraying Insecticide treatment regimen applied	Absolute numbers caught	Mean catches[a] [95% Confidence Interval (CI)]	Relative rates of *An. funestus* densities (RR)[b] [95% CI]	P-value
Deltamethrin WG)				
1-3 months since last spray	73	0.112[0.641, 0.371]	0.99 [0.63, 1.56]	0.897
4-6 months since last spray	1229	0.641[0.371, 1.109]	3.98 [3.15, 5.04]	<0.001
7-12 months since last spray	134	0.111[0.062, 0.199]	0.86 [0.64, 1.17]	0.067
>12 months since last spray or never	1186	0.189[0.113, 0.317]	1[NA][c]	NA
Lambdacyhalothrin CS				
1-3 months since last spray	20	0.191[0.090, 0.405]	1.03[0.56, 1.88]	0.805
4-6 months since last spray	6	0.055[0.022, 0.141]	0.17[0.08, 0.39]	<0.001
7-12 months since last spray	0	NE[e]	NE	0.972
>12 months since last spray or never	182	0.198[0.121]	1[NA][c]	NA
Pirimiphosmethyl EC				
1-3 months since last spray	478	0.234[0.131, 0.417]	1.04[0.83, 1.30]	0.786
4-6 months since last spray	346	0.055[0.030, 0.098]	0.25[0.20, 0.33]	<0.001
7-12 months since last spray	160	0.159[0.086, 0.293]	0.69[0.50, 0.95]	0.151
>12 months since last spray or never	2823	0.234[0.131, 0.417]	1[NA][c]	NA
Pirimiphosmethyl CS				
1-3 months since last spray	14	0.021[0.009, 0.047]	0.07[0.04, 0.13]	<0.001
4-6 months since last spray	70	0.004[0.002, 0.008]	0.02[0.01, 0.02]	<0.001
7-12 months since last spray	NA[d]	NA[d]	NA[d]	NA[d]
>12 months since last spray or never	2087	0.253[0.152, 0.422]	1[NA][c]	NA

[a & b] The effect of different IRS treatment regimens on the mean catches of An. funestus species where estimated by fitting generalized linear mixed models (GLMMs) with An. funestus catches treated as dependent variables. The households where nested within villages which were also nested within the clusters, these together with date were treated as random effects, while the different IRS treatment regimens were categorized as independent variables. A Poisson distribution with no intercept was used to estimate the mean catches while an intercept was included in estimating the RR.

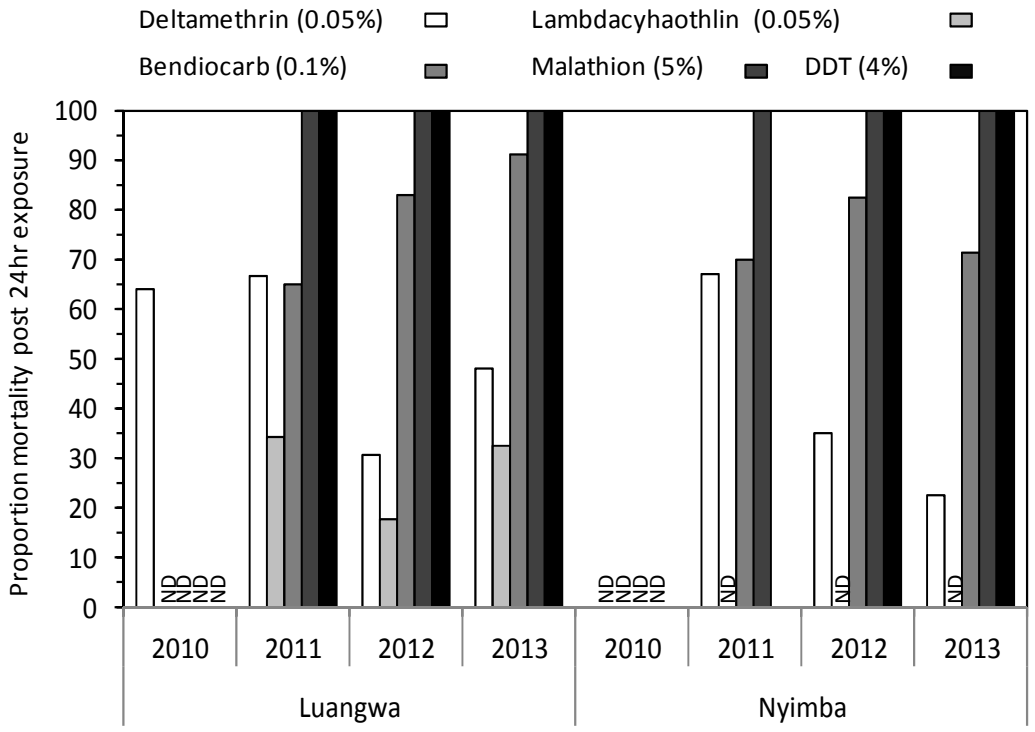
[C] Treated as the reference group

[d] No spraying was conducted therefore and no data available for estimates

4.3.4 Background observations of insecticide resistance and human exposure profiles for local Anopheles funestus populations

From the outset of the study, *An. funestus* exhibited high levels of resistance to both pyrethroids against which they were tested, and resistance level generally increased over the course of the study (P<0.001). Alarming rates of resistance to the carbamate bendiocarb were also observed but these did not increase over the course of the study (P=0.565). During this same period, there was no evidence of Malathion or DDT resistance detected in the mosquito populations (Figure 32).

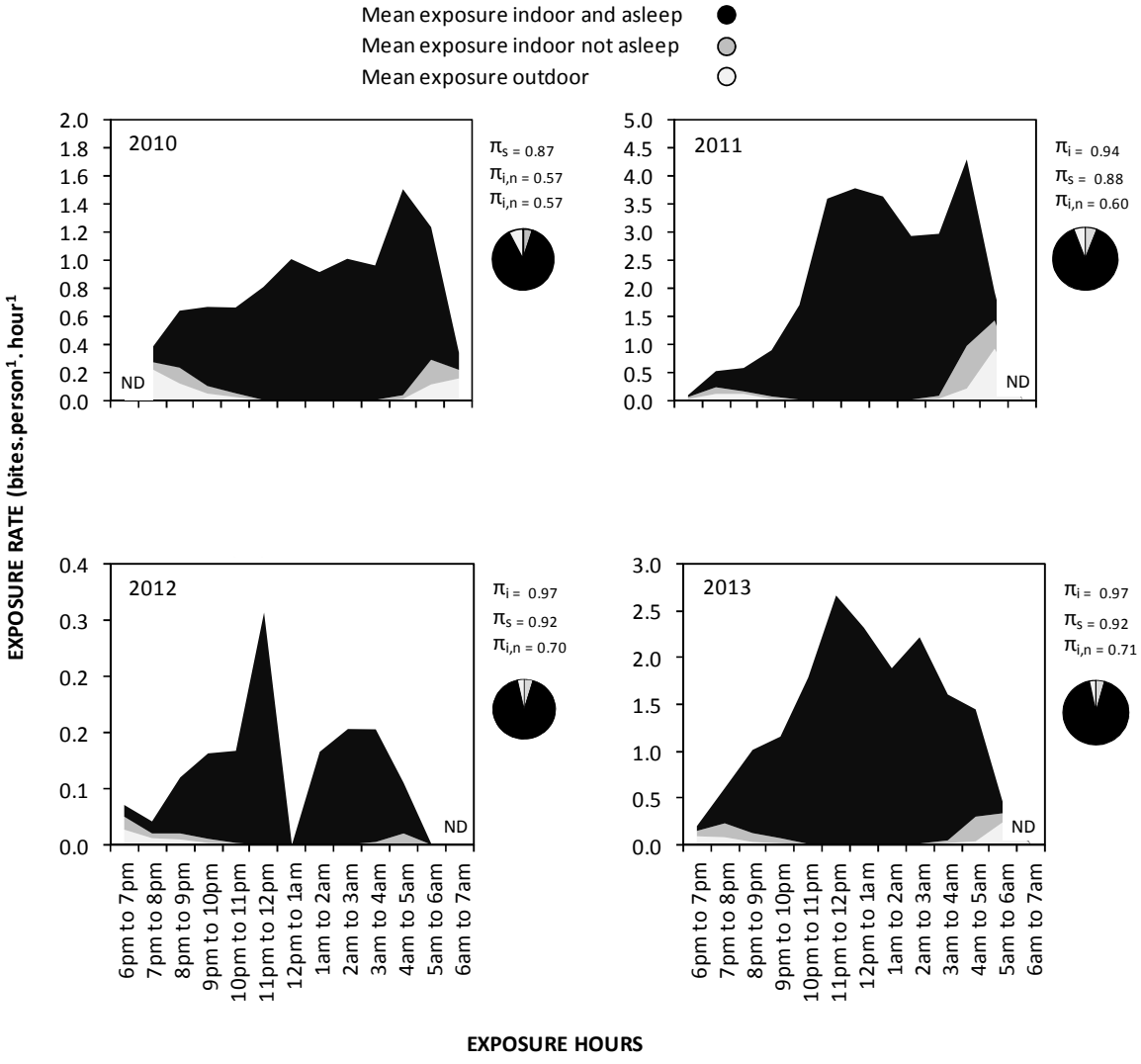
Figure 32. Insecticide resistance profile of *Anopheles funestus* in the study site from 2010 to 2013



Throughout the study period, humans lacking LLINs were exposed to far more bites by *An. funestus* indoors during the late hours of the night up to the early morning hours (Figure 33), consistent with the known behaviour of *An. funestus* across the continent (Gillies and

Coetzee 1987; Huho, Briet et al. 2013). The vast majority potential exposure to bites by this dominant vector occurred indoors at times when most individuals are asleep (Figure 33). Even for those using an LLIN to prevent most indoor transmission, most residual human exposure to *An. funestus* bites, and presumably malaria transmission, occurred indoors, increased gradually from 57% in 2010 to 71% by 2013 (Figure 33).

Figure 33.Mean exposure of humans to Anopheles funestus bites when they are indoors or outdoors, where π_i is the average proportion of human exposure to bites of Anopheles funestus' population which occurs indoors in the absence of any protective measure, π_s is the average proportion of human exposure to bites of *Anopheles funestus'* population which occurs indoors when individuals are asleep in the absence of any protective measure and $\pi_{i,n}$ is the average proportion of residual human exposure for users of net which occurs indoors



4.4 Discussion

In this setting of high LLIN unitization (>80%), even the modest (0-100%, mean=29.4%) coverage achieved with supplementary IRS conferred an incremental protection against malaria parasite infection through reduced vector population density, human exposure to bites and, presumably, to sporozoite inoculations. Overall supplementing of LLINs with IRS using PM-CS gave the greatest apparent protection against malaria risk, which lasted for a full 6 months, while IRS with PM-EC conferred less dramatic protection that was comparable with pyrethroids but apparently lasted for one full year. Neither of the two pyrethroid formulations exhibited any incremental protective effect for more than 3 months but it is notable that LC-CS conferred an apparently greater protective effect than DM-WG. These observations that quasi-randomly assigned IRS treatments conferred additional protection when provided as a supplement to LLIN utilization are consistent with a variety of other observational studies (Kleinschmidt, Schwabe et al. 2009; Kim, Fedak et al. 2012), as well as more recent randomized controlled studies (West, Protopopoff et al. 2014). The high level of incremental impacts observed, despite sometimes mediocre coverage with IRS, are actually consistent with the predictions of process-explicit models used to support the policy switch to universal coverage for both LLINs and IRS (WHO 2006), especially for a very anthropophagic mosquito like *Anopheles funestus* (Killeen, Smith et al. 2007), which is even more anthropophagic than the *An. gambiae* species (Killeen, McKenzie et al. 2001) used as an example mosquito in that simulation paper.

The high protective effect of PM-CS is also evident in the low densities of *An. funestus* caught in that group. The modest and short-lived protective effect of the two pyrethroid formulations, DM-WG and LC-CS most probably a result of the emergence of resistance to pyrethroids in the *An. funestus* population present in this study area, consistent with

evidence from Benin in west Africa that the protective effect of these insecticide formulations can be dramatically reduced to as little as a month by physiological resistance, even where these specific formulations have a residual activity against susceptible insectary-reared mosquitoes for up to 6 months (Rowland, Boko et al. 2013). While this rapid loss of incremental protection towards malaria elimination with pyrethroid-based supplementary IRS is of obvious and very direct concern (Wondji, Coleman et al. 2012; Haji, Khatib et al. 2013; Wang, Xia et al. 2013).The encouraging results obtained with IRS using PM, the CS formulation in particular, provide further evidence that pyrethroid resistance may be mitigated and managed in areas of high LLIN coverage using IRS (Corbel, Akogbeto et al. 2012; West, Protopopoff et al. 2014), or alternatively impregnating wall linings (Djènontin, Chandre et al. 2010; Ngufor, Tchicaya et al. 2014), with non-pyrethroids selected on the basis of standard WHO susceptibility assays. These observations are therefore consistent with similar recent reports from several distinct settings across Africa (Hunt, Edwardes et al. 2010; N'Guessan, Boko et al. 2010; Akogbeto, Padonou et al. 2011; West, Protopopoff et al. 2014) and can be readily rationalized on the basis of the combined observations of strong resistance to pyrethroids, complete susceptibility to organophosphates, and strong tendency to feed, and presumably rest indoors among the local *An. funestus* population.

It was expected that PM-CS would be the most persistent because this microencapsulated formulation is known to confer residual longevity for 6 months (Rowland, Boko et al. 2013; Oxborough, Kitau et al. 2014) as confirmed here. However, it was surprising that PM-EC had the longest longevity on these surfaces, apparently lasting 12 months after spraying, contrary to other studies suggesting that PM-EC is ineffective on mud surfaces (Oxborough, Kitau et al. 2014) and WHO estimates of a residual effect of only 3 months (WHO 2013) but is consistent with one other recent study (Fuseini, Ebsworth et al. 2011). While it is possible

to speculate that the persistence of PM-EC may have resulted from an initial absorption into the porous mud walls in most of the houses in the study areas, followed by slow subsequent release, it is also possible that this is simply the result of a spurious model fit to data from such a limited number of treated clusters with considerable inter-cluster variation in malaria risk level and seasonality, as presumably occurred for DM-WG which is highly unlikely to have really increased malaria transmission (Figure 28, Table 15) or vector density (Figure 31, Table 17). The observation that impact of both PM formulations and LC-CS upon vector density was greatest between 4 and 6 months after spraying suggests that maximum impact upon the vector population required sustained impact upon several generations of mosquitoes, well into the peak rainy season when they would be expected to grow exponentially and improve in reproductive fitness as the availability of larval habitat rapidly increases (Briet 2002; Russell, Lwetoijera et al. 2013).

Of course, there are several substantive limitations to this study. While the community-based nature of both the parasitological and entomological surveys, with only modest supervision and quality assurance, does leave some uncertainties about the data quality, recent detailed analyses of these primary (Hamainza, Killeen et al. 2014; Hamainza, Moonga et al. 2014) and secondary outcomes (Sikaala, Chinula et al. 2014) provide reassuring confirmation of their epidemiological relevance and discriminative power. While this study did not explicitly or comprehensively track the distinct costs of IRS and LLINs, these costs may be assumed to incurred largely independently of each other because of their distinct delivery methods, and have already been evaluated in detail across a variety of settings by other authors (Goodman, Mnzava et al. 2001; Guyatt, Kinnear et al. 2002; Bhatia, Fox-Rushby et al. 2004; Conteh, Sharp et al. 2004).

However, the most obvious limitation of this study is that it was not conducted as a rigorous randomized control trial and that deviations from the original randomization plan resulted in only a quasi-randomised design in practice, with known selection biases. This was also coupled with a lack of a statically estimated sample size. An additional considerable limitation arising from dependency on delivery of supplementary IRS through routine programmatic implementation mechanisms was the lack of consistent availability of a single, optimal formulation of a single pyrethroid or a single formulation of PM, so the study was unfortunately fragmented into more treatment arms with smaller numbers of assigned clusters per spray round than originally planned. Also, delays and limitations in the availability of PM formulations in the final year of the study resulted in a mismatch in the timing of application of PM-CS in Nyimba (November 2012), PM-EC, and LC-CS (Both February 2013).

So, in summary this study was not fully randomized because the implementation contractors did not fully adhere to the study design stipulated to them by the NMCC. This study may therefore be described as a quasi-randomized experimental evaluation to generate plausible evidence that the IRS treatments provide effective incremental impact beyond that already provided by high coverage with LLINs under near-programmatic conditions. The biggest inherent limitation of observational studies is their vulnerability to selection bias and confounding (Carlson and Morrison 2009; Boffetta 2011). The study largely adhered to its original randomization plan, and some of the most confounding variables were taken into consideration during planning and implementation (stratification of clusters into those that had previously been sprayed with deltamethrin and those that had not) and analysis (additional variables in regression models) phases. However, the deviations from the randomization plan by the implementing agencies were specifically

necessitated by product stock availability and motivated by efforts to target IRS to areas where they felt it was needed most, so treatment allocation was clearly systematically biased *a priori* in these cases. It is therefore prudent to interpret the level of evidence generated conservatively, and to classify this study as being essentially observational in nature. To generate probable evidence of efficacy under more precisely controlled (if somewhat less programmatically relevant(Habicht, Victora et al. 1999; Glasgow, Lichtenstein et al. 2003)) conditions would have entailed a rigorous, fully-randomised trial with a registered protocol including sample size estimates, data quality assurance and oversight committees (Habicht, Victora et al. 1999; Moher, Schulz et al. 2001; Glasgow, Lichtenstein et al. 2003; Flay, Biglan et al. 2005).

While the shortcomings of this study must be accepted, there is no obvious specific reason to suggest that they are inaccurate, and they do contribute to a relatively limited evidence base regarding the incremental impact of IRS formulations as supplements to LLINs(Pinder, Jawara et al. 2014). Despite these study design limitations, the evidence generated remains useful for guiding programmatic selection of IRS treatments. Perhaps just as important, it represents the first effort of the NMCC itself, rather than its specialist research and academic partner institutions, to conduct a cluster-randomized experimental evaluation of malaria transmission control measures. It therefore represents an invaluable experience through which the capacity of the NMCC has grown and can hopefully build upon.

4.5 Conclusion

Despite these study limitations, the results presented here do provide substantial evidence that (1) supplementing pyrethroid-based LLINs with pyrethroid-based IRS confers some, albeit short-lived, incremental protection against malaria infection relative to LLINs alone,

and (2) Replacing pyrethroids with an alternative insecticide class, in this case a long-lasting CS formulation of the organophosphate PM, as the active ingredient for supplementary IRS confers considerably enhanced protection, relative to IRS with pyrethroids. Supplementing LLINs with IRS using non-pyrethroids therefore appears to be efficacious for mitigating the immediate epidemiological consequences of vector population resistance to pyrethroids, and the observed impact on *An. funestus* densities suggest it may also be a valuable option for managing such resistance traits, ideally by using mosaics, rotations or combinations of complementary active ingredients (WHO 2012). Of course the primary limitation to the realization of such insecticide resistance management and mitigation plans in practice are (1) the availability of more efficacious, affordable and diverse insecticide formulations (Vontas, Moore et al. 2014), (2) increased financing for malaria vector control generally (WHO 2014), and (3) more cost-effective methods for targeting insecticides to vector populations so that both the biological resource coverage (Kiware, Chitnis et al. 2012; Killeen, Seyoum et al. 2013) and mortality rates arising from exposure to their active ingredients are maximized (Elliott 1972; Kitau, Oxborough et al. 2012; Okumu, Kiware et al. 2013; Okumu, Mbeyela et al. 2013; Killeen and Chitnis 2014).

CHAPTER FIVE

GENERAL DISCUSSION AND CONCLUSION

Further to the work presented in the previous chapters, here is presented a summary of how each objective was addressed, the immediate implications of the results, and remaining knowledge gaps which should be prioritized for future research are as follows:

5.1 The role Community health workers as a resource for delivery of health care and monitoring and evaluation

The roles of CHWs in provision of diverse health services has been well documented (Ajayi, Falade et al. 2008; Yeboah-Antwi, Pilingana et al. 2010; Counihan, Harvey et al. 2012; Ratsimbasoa, Ravony et al. 2012; Rutta, Francis et al. 2012) and have been promoted since the 1970s, when the World Health Organization's intensified the global drive for strengthened primary health care services particularly in middle and low income countries (WHO 1978; Cueto 2004) . The CHWs also provide a link between the community and peripheral health facilities through their services which increase the reach of the healthcare system within the communities they reside and serve (WHO 1989; Winch, Gilroy et al. 2005) resulting in appreciable declines in morbidity and mortality particularly in maternal and child health (Kumar, Mohanty et al. 2008; Thea and Qazi 2008; Lassi, Haider et al. 2010; Yeboah-Antwi, Pilingana et al. 2010; Chanda, Hamainza et al. 2011).

The findings described in Chapters 2, 3 and 4 are all derived from the activities of CHWs, whose primary goal was provision of malaria infection diagnostic testing, treatment of those found with infection and referral of patients with a negative test result who are febrile or have some other symptomatic compliant, thus further adding to the body of knowledge by re-affirming the role of CHWs in primary health care with regard to diagnosis and treatment of malaria. Chapter 2 further demonstrates that CHWs are able to follow provided

guidelines and adhere to treatment protocols more than trained health workers as demonstrated in other studies (Chanda, Hamainza et al. 2011; Chinbuah, Abbey et al. 2013).

In chapters 2, 3 and 4, in addition to the treatment functions, CHWs demonstrated their ability to contribute to the overall monitoring and evaluation system through the surveillance tools they were provided in the form of paper registers and mobile phones. Globally, most CHW driven monitoring and evaluation activities are conducted as disease specific vertical un-integrated programs resulting in information not being shared across disease areas to benefit users at different levels with regard to efficient data use, program implementation and resource allocation. However, there has been renewed interest in integration of services provided by CHWs. Refinements made to algorithms under integrated management of child illness (IMCI), has lead to the increased uptake of the concept of integrated community case management (iCCM) primarily targeted at children in most African countries (WHO 2004; WHO and UNICEF 2004). The iCCM concept increases access to treatment of common ailments such as diarrhoea, pneumonia, and malaria by training CHWs to assess / diagnose, treat and refer patients in their communities (WHO 2004; WHO and UNICEF 2004; Yeboah-Antwi, Pilingana et al. 2010; Hamer, Brooks et al. 2012). However, such concepts have not been effectively implemented in tandem with a robust integrated monitoring and evaluation systems and thus for all intensive purposes remain disease specific vertical programs. Integration has an advantage in that it fosters coordination and communication between different disease groups. In this regard for CHW operationalised monitoring and evaluation activities to achieve sustainability, they must be integrated across disease areas that CHWs have the mandate to provide diagnosis and treatment. Additionally these should be developed in a manner that can be integrated with national health information systems through harmonization of indicators. An integrated

system would facilitate pooling of resources from the different groups in order to sufficiently support implementation and other investment requirements (Ruebush and Godoy 1992; Ruebush, Zeissig et al. 1992; WHO 2012; WHO 2012).

5.2 Measuring malaria risk using Community Health Worker driven community based surveillance

In general malaria surveillance is guided by analysis of aggregated morbidity and /or mortality data which guides a programmatic population level response (WHO 2012; WHO 2012). Community Health Worker (CHW) implemented surveillance may be considered an intervention (Mueller, Slutsker et al. 2011; The malEra Consultative Group on Monitoring and Surveillance 2011; Chaki, Mlacha et al. 2012; WHO 2012), as it presumably increases the number of individuals tested, treated, recorded and subsequently reported allowing for more accurate risk mapping due to their larger geographical coverage (Alba, Hetzel et al. 2011; Liu, Sullivan et al. 2011; WHO 2012; WHO 2012; Tulenko, Mogedal et al. 2013). Decentralised monitoring and evaluation activities that are focussed on identifying malaria risk in communities are integral in evidence based planning and implementation of malaria control interventions and resource allocation (WHO 2012; WHO 2012). As the burden of malaria decreases there is need to strengthen monitoring and evaluation activities to the smallest reporting unit possible to allow for focal responses (Chaki, Mlacha et al. 2012; Smith, Cohen et al. 2013; Sikaala, Chinula et al. 2014). In most endemic countries CHWs serve the smallest catchment population in the health system and thus can potentially serve as such a reporting unit (Ruebush and Godoy 1992; Ruebush, Zeissig et al. 1992). Further, this allows for simplified identification, selection and follow-up of these populations at a reduced cost. These community based surveillance systems (CBSS) may additionally be

utilised to not only estimate access and use of interventions but also evaluate their impact longitudinally (Ruebush and Godoy 1992; Ruebush, Zeissig et al. 1992; Chaki, Govella et al. 2009; Alba, Hetzel et al. 2011; O'Sullivan, Kenilorea et al. 2011; WHO 2012; WHO 2012; Sikaala, Chinula et al. 2014).

As demonstrated in chapter 2, CBSS can be added to the well documented various diagnosis and treatment roles of CHWs (Ruebush and Godoy 1992; Ruebush, Zeissig et al. 1992; Counihan, Harvey et al. 2012; Ratsimbasoa, Ravony et al. 2012) . This longitudinal study was implemented through CHWs providing passive and monthly active malaria testing and treatment of infected participants. This allowed for the establishment of symptomatic criteria associated with malaria parasite infection to distinguish these from other causes of symptomology such as fever in the entire population under study (Roucher, Rogier et al. 2012).The coupling of these activities with a questionnaire at participant contact further allowed for in-depth assessment of various indicators with regard to symptoms, access and use of malaria interventions and selected population demographics. Thus the use of both passive and active testing and treatment in the CBSS allowed for an integrated and comprehensive analysis of changes within the communities under study. This chapter demonstrates that in areas of intense transmission, despite reasonably high LLIN and IRS coverage monthly active testing and treatment activities did not eliminate the human reservoir of malaria infection at the testing frequency experienced during the study (Okell, Drakeley et al. 2008; Shah, Tyagi et al. 2013; Trape, Tall et al. 2014). However, there was a notable decline in disease burden by participants that allowed to be tested and treated for infection more regularly (Okell, Drakeley et al. 2008; Shah, Tyagi et al. 2013; Trape, Tall et al. 2014). Additionally, this chapter demonstrated the strong association of symptoms with malaria infection, suggesting inexactness of the term “asymptomatic infection” with regard

to chronic malaria infections (Bisoffi, Gobbi et al. 2012; Cucunuba, Guerra et al. 2013; Lindblade, Steinhardt et al. 2013). A key programmatic emphasis of this chapter was to capture and report all malaria infections in the study population as a practical indicator of programmatic relevance. However this was not achieved due to being dependent on study participants consent to participate thus affecting the overall sensitivity of the intervention. This suggests that the community engagement approach used by the CHWs may have not been sufficient to impact community participation in repeated testing and treatment activities whether they had a positive or negative RDT result (Chen 1991; Espino, Koops et al. 2004; Beier, Keating et al. 2008). This is of relevance to malaria control, particularly in an elimination drive where such test and treatment activities play an integral part, particularly in identification of mildly symptomatic cryptic/chronic infections which are able to drive transmission and risk mapping for targeted intervention deployment (Bousema, Drakeley et al. 2010; Bousema, Griffin et al. 2012; Okell, Bousema et al. 2012; WHO 2012; Sturrock, Hsiang et al. 2013). Thus, a diminishing malaria burden scenario coupled with mild symptom manifestation requires a well co-ordinated and comprehensive community engagement approach for uptake of community based active test and treat programs among others (Chen 1991; Espino, Koops et al. 2004; Chaki, Dongus et al. 2011) .

As further demonstrated in chapter 3, CHWs were not only able to collect epidemiological data on the catchment populations they served (Baker and Ross 1996), but were able to transmit this information onward using both paper and electronic media via mobile phones. Information is critical to any effective malaria control program as it allows for data driven decisions on intervention options and investments required (Barclay, Smith et al. 2012; Chaki, Mlacha et al. 2012; Morse 2012; WHO 2012; WHO 2012; Free, Phillips et al. 2013; Sikaala, Chinula et al. 2014). The evidence globally on the ability for CHWs to routinely

transmit data through mobile phones or indeed any other electronic device remains limited. However, chapter 3 demonstrated that weekly summaries of malaria infections and estimates of diagnostic positivity transmitted via SMS were an adequate representation of the paper registers which were the primary data capture mechanism for the CHWs. The SMS reports also adequately reflected the epidemiological transmission patterns of the population under observation on a timely basis. Suggesting, a role for electronic devices for routine surveillance at a community level operationalised by CHWs or other health workers to allow for timely collation, access and dissemination for evidence based decision making (Kamanga, Moono et al. 2010; Zurovac, Talisuna et al. 2012; Free, Phillips et al. 2013; Githinji, Kigen et al. 2014). This technology thus, has the potential to improve service delivery by bridging the communication gap between program managers and frontline health workers including CHWs through provision of real time epidemiological spatial and temporal data (Kamanga, Moono et al. 2010; Zurovac, Talisuna et al. 2012; Free, Phillips et al. 2013; Githinji, Kigen et al. 2014).

Although, not explicitly explored the findings in chapter 3 further suggest a role for SMS reporting as a potential management tool to monitor performance of frontline health workers (Free, Phillips et al. 2013; Githinji, Kigen et al. 2014; Yukich, Butts et al. 2014) and in this case, the CHWs, as exemplified particularly with regard to whether they achieved the targeted number of active household visits per week and tracking the availability and use of AL and RDTs. The SMS reporting system however would have benefited from server based automation to reduce human error resulting from manual single entry of the received reports and also if there was consideration to bring such a platform to scale (Barrington, Wereko-Brobby et al. 2010; Meankaew, Kaewkungwal et al. 2010; NMCC 2011; Free, Phillips et al. 2013; Githinji, Kigen et al. 2013; Githinji, Kigen et al. 2014).

It is obvious that for the SMS platform to function there is need for mobile phone coverage, which though currently increasing in Africa (ITU 2010) has not yet been adequately made available in the remotest parts which are also in most cases the areas of highest burden. Thus a limitation to scale of such a platform may be the availability of mobile phone coverage which essentially is not in the hands of the health sector but a business decision by the mobile phone companies.

Chapter 4 evaluated the impact of four insecticides used in IRS on malaria risk under the backdrop of high LLIN utilization. This quasi-randomised study utilised data collected by the CHWs from their parasitological (passive and active) and entomological surveys. Even though active surveillance detects more malaria infections, it has been argued that it may not be the best approach for intervention evaluation because early detection and cure of infections by repeated testing and treatment may reduce prevalence by itself, and therefore limit the level of impact achieved by the intervention, as well as by preventing re-infection if drugs are used that have some prophylactic effects. Also, the most symptomatic acute phases occur relatively early in the course of infection (Jeffery and Eyles 1954; Collins and Jeffery 1999) so passive surveillance is considered a better measure of impact upon recently acquired infections (Tiono, Kangoye et al. 2014). However this approach limits the findings to only the population that seeks care and may not be representative of the entire population, which requires larger population numbers (Tiono, Kangoye et al. 2014). However, few studies have been conducted to directly compare active and passive surveillance for intervention evaluation and thus the evidence still remains limited and ambiguous (Tiono, Kangoye et al. 2014). This chapter demonstrated enhanced incremental protection against malaria infection through reduced vector population density, human exposure to bites and, presumably, to sporozoite inoculations using long lasting

organophosphate formulations in IRS than the pyrethroids in an area of high LLIN utilization and demonstrated pyrethroid resistance (Kleinschmidt, Schwabe et al. 2009; Kim, Fedak et al. 2012).

As expected all the insecticides evaluated decay with regard to incremental protective effect overtime at different rates, suggesting that even when IRS is applied, LLINs should continue to be used in order to maintain some level of protection whenever the IRS insecticidal effects diminish (Okumu, Chipwaza et al. 2012). These findings have programmatic implications as preventive interventions such as LLINs and IRS constitute a substantial cost of approximately 70% of the overall cost of malaria control (WHO 2009) and the debate of their optimal use singularly or in combination at programmatic level is yet to be exhausted (Curtis, Maxwell et al. 1998; Mnzava, Dlamini et al. 1999; Mabaso, Sharp et al. 2004; Kleinschmidt, Schwabe et al. 2009; Corbel, Akogbeto et al. 2012; Fullman, Burstein et al. 2013). Through the CBSS, it was possible to routinely monitor these vector control intervention utilization/coverage and their effect on both vector abundance and community malaria prevalence as indicators of program progress (Breman and Holloway 2007; Fillinger, Kannady et al. 2008; Chaki, Mlacha et al. 2012; Sikaala, Chinula et al. 2014). The demonstration that CHWs are able to not only provide curative services but also with minimum training provide complementary entomological data may provide an opportunity for integrated longitudinal epidemiological and entomological monitoring of intervention programs as part of a scalable national surveillance system (Breman and Holloway 2007; Fillinger, Kannady et al. 2008; Chaki, Dongus et al. 2011; Chaki, Mlacha et al. 2012; Sikaala, Chinula et al. 2014).

The uptake of complementary community based surveillance systems may mitigate a fundamental limitation of health facility based only surveillance which may be biased in its reporting based on only those patients that have access to the facilities, further biasing any analysis of epidemiological trends which may in fact not be a true reflection of the general community trends in the catchment population of the health facility. This may lead to incorrect application of interventions and resources. Additionally, health facility surveillance does not routinely incorporate access and use of preventive interventions such as LLINs and IRS (MOH 2008; WHO 2012; WHO 2012). Nationally representative surveys may also be used to estimate from a community perspective these short comings from routine health facility based surveillance. However these surveys are point estimates conducted at a very high cost and provide a general picture of community trends which may be not adequate to address specific focal community needs as can be done by a routine surveillance system that reflects both health facility and community trends. Furthermore, as malaria prevalence drops these surveys are not sensitive enough to capture changes in transmission without increasing the sample size which has rather huge cost implications which may be a challenge to met for most malaria programs (Hay, Smith et al. 2008).This further strengthens the suggestion of a complementary role of CBSS which if implemented across disease areas may be cost effective and sustainable by most endemic countries.

5.3 Limitations and future considerations for community based service provision and surveillance systems

The successes of CHW driven community based health interventions have been well documented and they range from health education, diagnosis and treatment, distribution of preventive measures such as LLINs, implementation of IRS and community mobilization/

sensitization among others (WHO 1989; Ruebush and Godoy 1992; Ruebush, Zeissig et al. 1992; Sikaala, Chinula et al. 2014) . For CHW driven activities to be successful they require constant logistical, management, supervisory and monitoring support (Ruebush and Godoy 1992; Ruebush, Zeissig et al. 1992; Chaki, Govella et al. 2009; Sikaala, Chinula et al. 2014) . An ever current, updated report on epidemiological indicators is vital to ensuring that CHWs meet the desired objectives whether preventive, curative or otherwise. However, there are some challenges that require urgent redress if CHW service provision is be successful.

A key challenge is the vertical approach to CHW program implementation (Ruebush and Godoy 1992; Ruebush, Zeissig et al. 1992; Chanda, Hamainza et al. 2011; Counihan, Harvey et al. 2012; Kalyango, Rutebemberwa et al. 2012; Keating, Hutchinson et al. 2012; Kisia, Nelima et al. 2012; Sikaala, Chinula et al. 2014). In most endemic countries the CHW approach is essentially threefold, where the service delivery needs of a community are divided among available CHWs with each managing one service delivery aspect alone or where CHWs manage several service delivery needs in a given community or a scenario where these two approaches are implemented in a single community (WHO 2012; WHO 2012; WHO 2012). This is driven by among other things availability of potential CHWs within a given community and the retention of these CHWs once selected. In all scenarios what still remains common is the linkage with the health facility which is not only the supervisory entity but also acts as commodity supply point and receives reports from the CHWs on their activities. The reports received particularly on any commodity consumption such as RDTs and treatments are incorporated as part of the health facility report, thus suppressing any knowledge with regard to CHW contributions to service provision particularly higher up in the reporting chain (Odhiambo-Otieno 2005; Mutemwa 2006; MOH 2008; Maokola, Willey et al. 2011). This is mainly due to the fact that, a vast majority of CHWs globally do not have

access to a surveillance reporting system that encompasses all their reporting requirements. Such a platform would not only provide community based surveillance data for decision making, but would also inadvertently be an advocacy tool for pushing for appropriate compensation for CHW efforts. Such a platform would be expected to be a result of an integrated training curriculum for CHWs across disease areas which would encompass an integrated package of deliverables at the community level which would in turn foster sustainability of the CHW activities though pooled infrastructure and resources both human and financial (Chaki, Govella et al. 2009; WHO 2012; WHO 2012).

As most CHWs provide services on a voluntary basis and thus are unpaid, they seek other activities that provide an income for them and their household. This may lead to attrition if their CHWs duties demand more of their time than they can allocate away from their income generating activities (Olang'o, Nyamongo et al. ; Bhattacharyya, Winch et al. 2001; Alam, Tasneem et al. 2011; Greenspan, McMahon et al. 2013). Though not a key study objective, of the 42 paid CHWs retained for the duration of the study, only 2 CHWs relinquished their roles, 1 due to illness and the other due to relocation. In addition the CHWs were incentivised by provision of a mobile phone and a weekly top for them to submit their reports and for any other communication official or not. Additionally, frequent supervisory visits and ensuring availability of commodities and any other requirements for the CHWs to function optimally may also serve as an incentive. A clear recommendation from chapters 2, 3 and 4 for malaria control is to formalise the establishment of CHWs into the health system. Although efforts towards this have been made in some countries (Medhanyie, Spigt et al. 2012; Kok and Muula 2013; Zulu, Kinsman et al. 2013), most endemic countries are yet to formalise the roles of CHWs and develop sustainable mechanisms to compensate them for their efforts which may even include formal

employment and associated job security and monetary benefits (Kok and Muula 2013; Zulu, Kinsman et al. 2013).

In addition to surveillance data providing raw numbers of disease trends, there is need to have process indicators for monitoring and evaluation to measure progress of interventions deployed, including the CHW activities themselves. As demonstrated in chapters 2,3 and 4 a robust and stable indicator such as diagnostic or test positivity may be considered to provide trends in spatial and temporal malaria transmission as estimates of this parameter are robust to variations in reporting rates, diagnostic practices and health utilization because these determinants equally affect the nominator and denominator (WHO 2012). Such an indicator can be automatically calculated, graphed and if required mapped in real time (Barrington, Wereko-Brobby et al. 2010; NMCC 2011; Free, Phillips et al. 2013; Githinji, Kigen et al. 2014) to allow for improved local planning, response and managerial follow-up to program implementation.

A general challenge for malaria control is the possibility of reduced acceptance of malaria control interventions as malaria transmission drops and is perceived to be less important in communities. Towards elimination, the CHW role on the curative side will greatly encompass programs that will actively seek out infection in communities. As described in chapter 2, communities may not be easily compliant to testing particularly when they perceive a low risk of infection. Although, compliance to treatment regimes is acceptably high in the communities, these assessments have only been done in populations that have been diagnosed and found with infection. As part of the “end game” for malaria elimination mass drug administration will become increasingly important and will most probably be in-part driven by CHWs. Currently there is no effective single dose anti-malarial available on

the market which could be used a “directly observed treatment” approach to MDA. Thus, there is need for locally tailored community engagement strategies to improve uptake of such interventions which could include in addition to MDA, mass or focal active test and treatment activities.

Community based surveillance for monitoring and evaluation to track progress towards set targets is an integral part of the overall malaria control and eventual elimination program and can be operationalised by non expert cadres such as CHWs.

5.4 Conclusions and recommendations

The global strategy for malaria control and elimination is based on 4 key elements: (1) Provision of prompt and effective treatment; (2) Implementation of sustainable preventive measures, including vector control; (3) Early detection, prevention and containment of epidemics, and; (4) Strengthening local capacities to generate evidence through research to guide program implementation (WHO 1999). Successful implementation of all these elements requires effective surveillance. One of the most important purposes of surveillance is to guide decentralized, fine-scale, local program implementation at the level were the data is collected. Local use of surveillance data, by the health system staff responsible for collecting it also has the advantage of engaging those staff as motivated stakeholders in maximizing the quality, quantity and relevance of the data(Wetterhall, Pappaioanou et al. 1992; WHO 2013; Yukich, Butts et al. 2014). All three chapters of this study provide evidence for the effectiveness, limitations and potential of a novel CB system for not only providing universal, regular malaria testing and treatment services, but also functions as a fine-scale system for surveillance of infection burden among the population. The findings of this study suggest that repeated testing and treatment of malaria parasite

infected populations may reduce the burden of disease and associated symptoms but only if much higher rates of frequent participation are achieved. Also, data generated as an added-value surveillance function, these activities can clearly be useful for tracking impact of program interventions. Additionally, the adequacy of electronic platforms for near-real-time reporting was also demonstrated. Important lessons learned and questions raised by this study, as well as derived recommendations for future investigation are summarized as follows:

1. The key to successful suppression of the infectious parasite reservoir in humans lies in improved community participation in frequent testing and treatment, especially when they are not acutely ill but may be carrying chronic, mildly symptomatic infections.

2. Decentralized, community-based health care and surveillance can be managed programmatically and sustainably through existing, facility-based supervision and support systems

3. Although many systems for engaging CHWs rely on volunteerism, it is important to examine and optimize systems for recruiting, remunerating, incentiving and retaining of such CB personnel.

4. Implementation of an effective CBSS beyond this pilot scale will require substantive capacity development at district, provincial and national levels to support collation, archiving, analysis and interpretation of the data generated to guide program decision making.

5. Mobile phone technology can improve routine reporting by CHWs to enable for fine-scale risk-mapping in near-real-time and enhance program targeting and implementation.

6. Further studies to establish and evaluate feedback mechanisms through which data from such a CBSS influence routine programmatic decisions and actions at district, HF and CHW levels, so that the effectiveness of these systems are optimized through a sense of local ownership and stewardship.

7. CBSS reports of commodity use and availability should be integrated with logistics management systems, to mutually strengthen both platforms and ensure timely availability of all CHW supply requirements.

8. In order to ensure maximum cost-effectiveness and affordability, CBSSs should integrate all major disease entities which CHWs can diagnose and treat.

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APPENDICES

Appendix 1. Community health worker paper register

Patient Register (National Malaria Control Centre / Malaria Transmission Consortium Impact Study)

(+ve = positive, -ve = negative, ND = Not Done, IRS = Indoor Residual Spraying, IPT = Intermittent Presumptive Treatment)

S/N	Date	Study ID Number	Name	Sex		Age	Where did you see patient?			Is the patient from your catchment area?		Patients symptoms /complaints (tick all that apply)										Intervention used (tick all that apply)			Temperature (°C)	RDT Results			Slide Results			Treatment					Patient Outcome				Any additional comments
				Male	Female		At patients home	At CHWs home	Health Facility	Yes	No	fever	History of fever (past 14 days)	headache	cough	Diarrhoea/ watery stool	Vomiting	Problems breathing	Chest pain	No symptoms	Other symptoms	ITNS	IRS	IPT		+ve	-ve	ND	+ve	-ve	ND	Coartem	Fansidar	Quinine Tab	Quinine Injection	Other treatment given	Referred	well	died	Other	

Appendix 2. SMS reporting summary form (Completed prior to sending report via SMS)

SMS Reporting Summary Form

Health Facility / CHW: ……Date Compiled:……Date sent: ……

Patient register (Passive)	Total OPD	Total suspected	RDT +ve	RDT –ve	Microscopy +ve	Microscopy - ve	Under 5 +ve	Over 5 +ve	Patient register (active)	Total screened	RDT +ve	RDT -ve	Under 5 +ve	RDT –ve
REG									SCR					

Example sms: REG xxxx-xxx-xx-xx-xx-xx-xx-xx-SCR-xx-xx-xx-xx-xx (REG 2000-500-50-450-0-0-10-40-SCR-50-0-0-0-0)

	6s prescribed	In stock Y/N	12s prescribed	In stock Y/N	18s prescribed	In stock Y/N	24s prescribed	In stock Y/N		Available	Used
ACT									RDT		

Example sms: ACT xx-y-xx-n-xx-y-xx-n-RDT-xx-xx (ACT 20-y-20-n-30-y-0-y-RDT-500-500)

SMS Reporting Summary Form

Health Facility / CHW: ……Date Compiled:……Date sent: ……

Patient register (Passive)	Total OPD	Total suspected	RDT +ve	RDT –ve	Microscopy +ve	Microscopy - ve	Under 5 +ve	Over 5 +ve	Patient register (active)	Total screened	RDT +ve	RDT -ve	Under 5 +ve	RDT –ve
REG									SCR					

Example sms: REG xxxx-xxx-xx-xx-xx-xx-xx-xx-SCR-xx-xx-xx-xx-xx (REG 2000-500-50-450-0-0-10-40-SCR-50-0-0-0-0)

	6s prescribed	In stock Y/N	12s prescribed	In stock Y/N	18s prescribed	In stock Y/N	24s prescribed	In stock Y/N		Available	Used
ACT									RDT		

Example sms: ACT xx-y-xx-n-xx-y-xx-n-RDT-xx-xx (ACT 20-y-20-n-30-y-0-y-RDT-500-500)

Appendix 3. Community health worker monthly summary report

NMCC/ MTC MONTHLY SUMMARY REPORT

DATE: _____ MONTH: _____ SUBMITTED BY _____ SIGN_____

HEALTH CENTER / COMMUNITY HEALTH WORKER: _____ CATCHMENT AREAS: _____

	ACTIVE	PASSIVE	TOTAL
TOTAL			
TOTAL SUSPECTED			
UNDER 5 RDT +VE			
UNDER 5 RDT –VE			
OVER 5 RDT –VE			
OVER 5 RDT +VE			
UNDER 5 MICROSCOPY +VE			
UNDER 5 MICROSCOPY -VE			
OVER 5 MICROSCOPY +VE			
OVER 5 MICROSCOPY –VE			
FEVER UNDER 5			
FEVER OVER 5			
UNDER 5 WITH HISTORY OF FEVER IN LAST MONTH			
OVER 5 WITH HISTORY OF FEVER IN LAST MONTH			
# WITH ITNS			
# SLEPT UNDER ITN PREVIOUS NIGHT			
# WITH IRS SPRAYED IN LAST 6 MONTHS			
# ON IPT			
NUMBER NOT TESTED - NOT AVAILABLE			
NUMBER NOT TESTED - REFUSED			
# OF RDTS IN STOCK			
# OF SLIDES IN STOCK			