Influence of Indoor Residual Spay on An. Gambiae S.L. and An. Funestus Abundance, Resting Behavior and Community Perception on Irs Intervention In Kagera-Tanzania

Gordian Kikompolisi
  Department of Health, Missenyi District Council, Kagera, Tanzania

Basiliana Emidi
  National Institute for Medical Research Mwanza Research Centre  https://orcid.org/0000-0001-6856-541X

Billy Ngasala
  Department of Parasitology and Entomology, Muhimbili University of Health and Allied Science, Dar es Salaam, Tanzania

Research

Keywords: Indoor residual spray, An. gambiae s.l., An. funestus, resting behaviors, community perception, Pirimiphos-methyl, Kagera, Tanzania.

Posted Date: October 5th, 2021

DOI: https://doi.org/10.21203/rs.3.rs-948331/v1

License: ☺️ ️ This work is licensed under a Creative Commons Attribution 4.0 International License. Read Full License
INFLUENCE OF INDOOR RESIDUAL SPAY ON \textit{AN. GAMBIAE} S.L. AND \textit{AN. FUNESTUS} ABUNDANCE, RESTING BEHAVIOR AND COMMUNITY PERCEPTION ON IRS INTERVENTION IN KAGERA-TANZANIA

Gordian Kikompolisi\textsuperscript{1, 2}, Basiliana Emidi\textsuperscript{3, 4*}, Billy Ngasala\textsuperscript{2}

Affiliations

\textsuperscript{1}Department of Health, Missenyi District Council, P. O. Box 38, Kyaka, Kagera, Tanzania.

\textsuperscript{2}Department of Parasitology and Medical Entomology, Muhimbili University of Health and Allied Sciences, P. O. Box 65007, Dar es Salaam, Tanzania.

\textsuperscript{3}Department of Infectious Diseases, National Institute for Medical Research, Mwanza, P. O. Box 1462, Mwanza, Tanzania.

\textsuperscript{4}Liverpool School of Tropical Medicine, Pembroke Place, Liverpool L3 5QA, United Kingdom.

*Corresponding author: ebasi2002@gmail.com

Emails:

Billy Ngasala - bngasala70@yahoo.co.uk

Gordian Kikompolisi - gordc84@gmail.com

Abstract:

\textbf{Background:} There has been a persistence of malaria transmission in Kagera Region despite consecutive phases of indoor residual spraying (IRS) targeting malaria vectors. In addition to
that, there is limited information on the resting behaviors of An. gambiae s.l. and An. funestus and community perception on IRS in Kagera.

**Methods:** The study was conducted in two districts; Missenyi and Karagwe. Missenyi was IRS sprayed with Pirimiphos methyl insecticide and Karagwe was unsprayed. Mosquito collections were conducted using bucket traps and CDC (Centre for Disease Control) light traps. Mosquitoes were identified morphologically using taxonomical key. Sibling species were identified by polymerase chain reaction (PCR). Interviews and focused group discussions (FGDs) were conducted in order to obtain information on community perception with regard to IRS interventions.

**Results:** A total of 5,777 mosquitoes were collected indoors by CDC light traps. *An. gambiae* s.l. and *An. funestus* accounted for 13% (n=749) of all mosquitoes collected. Unexpectedly, large proportions of *An. funestus* were collected indoors in Missenyi district, despite the fact that, the area was sprayed. In Karagwe district, *An. gambiae* s.l. was collected in large proportion. Results have showed that, *An. arabiensis* was the dominant sibling species among the *An. gambiae* s.l. in Karagwe and Missenyi districts with 82.8% (n=24) and 98.8% (n=166), respectively. Study participants agreed that, IRS campaign is useful for control of malaria vectors. They also reported that, malaria transmission have declined by comparing before and after IRS. Challenges reported which the IRS campaign poses to them, including the emergence of other insects in their houses and increase of mosquitoes later after IRS. They also reported some mosquito resting places which are not a target during spray operations.

**Conclusion:** The present study has revealed the abundance of malaria vectors and community perception on IRS intervention efficacy and sustainability. Surprisingly, large proportions of *An. funestus* were collected indoors, despite the fact that, Missenyi district was sprayed. This situation calls for further studies on *An. funestus* behaviors and possible reasons for tolerance in
sprayed area. Community sensitization before, during and after IRS application needs to be
strengthen for getting intended results.

Keywords: Indoor residual spray, An. gambiae s.l., An. funestus, resting behaviors, community
perception, Pirimiphos-methyl, Kagera, Tanzania.

Introduction

Malaria is among the most burdening public health problem globally. It is the leading cause of
morbidity and mortality in many third world countries. Children are more at risk and they
accounted for more than two thirds of the global deaths [1]. Malaria remains a serious vector-
bone disease across sub-Saharan Africa where transmission is maintained by Anopheles species
such as; An. gambiae sensu stricto, An. coluzzii, An. arabiensis and An. funestus [2]. In Tanzania,
malaria transmission is still high especially around Lake Victoria regions whereby the main
vectors include the An. gambiae s. s., An. arabiensis and An. funestus s.s. The Ministry of Health
of Tanzania through the National Malaria Control Programme (NMCP) has been implementing a
number of interventions to control malaria. These interventions includes preventive vector
control interventions which involve the use of long lasting Insecticide Nets (LLINs), indoor
residual spraying (IRS), and larval source management (LSM) Also treatment of malaria case
with artemisinin- combination therapies (ACTs), diagnostic testing with malaria rapid diagnostic
tests (mRDTs), Intermittent preventive treatment during pregnancy (IPTp), behavioral change
communication (BCC) campaigns and surveillance, monitoring and evaluation activities [3].
Among the interventions implemented by NMCP, the IRS has been implemented in selected
districts in the Lake zone regions due to high malaria prevalence. In 2011, malaria prevalence in
children under five years of age was 32% in Geita region, which was the highest prevalence compared to other regions in Tanzania [4].

A number of factors interact to influence the transmission of malaria. These factors includes the quality of insecticides, human behaviors and practices such as fishing, agriculture and staying outside while getting dinner/sapper, effectiveness of IRS and LLINs, mosquito feeding behaviors and weather conditions of a specific area interact altogether to influence the resting behaviors of malaria vectors [5]. *Anopheles* species have different resting and feeding behaviors [6]. Chemicals used as insecticides such as Dichloro Diphenyl Trichloroethane (DDT) are irritant to malaria vectors; therefore force them to exit houses quickly after entering them. When mosquitoes cannot obtain blood meals indoors, they will obtain it outdoors in which they may either feed on people outside houses or on animals these mosquitoes will continue surviving and will account for residual malaria transmission [7]. Insecticides used for IRS and LLINs have been reported to influence the malaria vectors’ abundance, resting behaviors and species composition [8,9].

It was estimated that, 124 million people were protected by IRS, representing 4% of the global population at risk of malaria in 2013[10]. Indoor residual spraying (IRS) in mainland Tanzania began in 2006 [11]. In 2018, in Tanzania mainland and Zanzibar a total of 15 district councils were already implementing IRS. The districts were Ngara, Missenyi, Bukoba rural, Chato, Musoma rural, Butiama, Sengerema, Kwimba and in Pemba and Unguja, Central, North A, North B, South, West, Chakechake and Micheweni [11]. During the National IRS campaign in 2016, a total of 543,865 house structures were targeted out of these 515,217 house structures were sprayed equal to 94.7%. IRS was implemented in Missenyi in December, 2017, by using
Primiphos-methyl (Actellic 300CS) sprayed on indoor wall surfaces. Primiphos-methyl (Actellic 300CS) efficacy is up to 8 months post spray [12]. In Missenyi district, 45,344 structures were targeted and 44,111 sprayed whereby the coverage was 97.3% [11].

Previous studies conducted in Bioko island have reported that, indoor vector control interventions have led to changes in behavior of malaria vectors [13,14]. Assessment of IRS impact on primary malaria vectors in West, East and South Africa revealed a great increase of *An. arabiensis* with a decrease in *An. funestus, An. gambiae and An. coluzzi* [9]. This is also the case in lake zone regions in Tanzania [15]. The change in behavior among malaria vectors have reported to contribute to persistent malaria transmission [16]. Although the use of insecticides reduce mosquito density, it leads to the selection of resistant species that account for residual malaria transmission in intervention area [17]. There is limited information on resting behaviors of malaria vectors in Misenyi and Karagwe districts. Therefore, the present study focused on assessing the influence of indoor residual spraying on abundance, resting behaviors of malaria vectors and community perception on mosquito bite protection in Misenyi (indoor residual sprayed district) and Karagwe (non- indoor residual sprayed district) in Kagera-Tanzania.

### Materials and methods

#### Study area

A cross-sectional study with both quantitative and qualitative approach was conducted from June to July, 2018. It was conducted in two districts of Misenyi and Karagwe (Figure 1). Misenyi district covers a geographical area of 270,875 hectares. Misenyi district is located on an altitude...
between 31.45° east Greenwich line and 0.9° southern of equator, it has 2 rain zones, heavy rainfall zone: The rain falls to between 400 – 2000mm per annum. The district is comprised of Missenyi division and Kiziba division [18]. It has a population of 227,270 with growth rate of 1.1. Population per division, In Kiziba division, there are 98,754 people while in Missenyi division there are 128,516 people [19].

Karagwe district has a population of 332,020 [19]. Population distribution has a rural urban difference in which the rural population is higher than urban population of 307,573 and 24,447, respectively. The District is characterized by mountain ranges, which are separated by swampy valley bottoms and wet lands. The altitude ranges between 1500-1800 meters above sea level while valley bottoms and wet lands are 1150 meters to 1450 meters above sea level. Most of the District has a tropical highland climate. The annual average temperature is 26°C, rainfall distribution is bi-modal with peak rains from September to December and from March to May (Karagwe district profile, 2012). The main economic activity for both districts is agriculture.

Selection of districts for IRS implementation

In 2016, the NMCP in collaboration with President Malaria Initiative (PMI) and other key malaria vector control stakeholders selected the IRS intervention areas which among them, Missenyi district council was included. The criteria used were high malaria burden districts as indicated by the epidemiological data on malaria incidence in the DHIS2.

Site selection
In the present study, three villages were selected in each district, for Missenyi; Kassambya, Gabulanga and Mabuye villages while in Karagwe; Kihanga, Katanda and Kishoju villages were selected. In each village, eight households were obtained based on the presence of eaves in Karagwe and presence of eaves plus being sprayed during the December, 2017 in Misenyi for mosquito collection giving a total of twenty-four (24) households [20]. The procedure was the same in both study areas where a total of 48 households were obtained. Selection of villages in Missenyi district was based on having high number of confirmed malaria cases (DHIS2 and top ten diseases from the health facilities), villages with many houses having open eaves and having many breeding sites of mosquitoes as per the report from District Malaria Vector Surveillance Officer (DMVSO). The same criteria were used to obtain the three villages in Karagwe district.

**Mosquito collection**

Mosquitoes were collected from six selected villages using bucket traps (BT) for collection of mosquitoes both indoors and outdoors and CDC-light trap for indoors collection [21]. Mosquitoes were trapped for three consecutive nights per house until all 24 houses in each district were covered [15]. For each house structure, six bucket traps were used per house; three indoors and the other three outdoors. A single CDC-light trap was located per house at night in a room with no bucket trap. Daily mosquito collections were carried out from 6.00 pm to 6.00am[22]. All collected mosquitoes were stored to species level using morphological identification key [23]. Culicines were recorded and discarded. *An. gambiae* s.l. and *An. funestus* group were identified to sibling species by method developed by Scott [24] and Koekemoer and others, respectively [25].
Figure 1: Map of Kagera region showing Missenyi and Karagwe districts.

Data collection using questionnaires and interview guide
A total of 396 heads of households were randomly selected and interviewed using structured questionnaires. Four focused group discussions (FGDs) were conducted two in each district. Each group was composed of eight members aged 18 years and above.

**Data analysis**

The number of mosquitoes and variables on the questionnaire were entered in SPSS software version 20 and analyzed to obtain the proportions and frequencies. Statistical tests such as t-test, Anova and Chi-Squared tests were performed. Identification of Mosquitoes to sibling species was achieved by molecular technique (PCR) which was conducted at NIMR, Amani centre in Muheza, Tanzania.

**Results**

**Mosquito abundance in Missenyi and Karagwe districts**

A total of 5,777 mosquitoes were collected. Majority of mosquitoes (95.7%, n=5530) were collected in Missenyi (indoor residual sprayed district). A total of 247 mosquitoes were collected in Karagwe (non-indoor residual sprayed district). Among them, 5679 were collected indoors and 98 were collected outdoors by using bucket traps. Majority of mosquito collected were *Culex* species. Malaria vectors; *An. gambiae* s.l. and *An. funestus* accounted for 13% (n=749) of all mosquitoes collected. In Missenyi district, *An. funestus* were collected in large proportions while in Karagwe district, *An. gambiae* s.l. were collected in large proportion. A total of 98 mosquitoes were collected by bucket traps outdoors in Missenyi and Karagwe districts, majority of them (88.8%, n=87) were collected in Misenyi district. All *An. funestus* were collected by CDC light traps indoors in Missenyi district (Table 1).
Table 1: Proportion of mosquitoes collected using CDC-light traps and the bucket traps indoors and outdoors in Missenyi and Karagwe districts

| Species          | Missenyi district, n (%) | Karagwe district, n (%) |
|------------------|--------------------------|-------------------------|
|                  | CDC-light trap indoors   | Bucket trap outdoors    | Bucket trap indoors |
|                  | Bucket trap outdoors     | Bucket trap indoors     |
| An. gambiae s.l. | 3 (0.1)                  | 6 (60.0)                | 4 (40.0)             |
| An. funestus     | 683 (12.6)               | 0 (0.0)                 | 0 (0.0)              |
| An. coustani     | 0 (0.0)                  | 0 (0.0)                 | 0 (0.0)              |
| An. rufipes      | 4 (0.1)                  | 0 (0.0)                 | 0 (0.0)              |
| Culex species    | 4,277 (78.8)             | 81 (88.0)               | 11 (12.0)            |
| Mansonia species | 461 (8.5)                | 0 (0.0)                 | 0 (0.0)              |
| Total            | 5,428                    | 87                      | 15                    |

An. gambiae s.l. and An. funestus sibling species

A total of 211 female Anopheles mosquitoes; 40 from Karagwe district and 171 from Missenyi district were analysed by polymerase chain reaction (PCR) to reveal the composition of the sibling species. A total of 29 An. gambiae s.l. and 11 An. funestus were obtained from Karagwe district. In Missenyi district, An. gambiae s.l. and An. funestus were 168 and 3, respectively. The PCR results have showed that, An. arabiensis was the dominant sibling species among the An. gambiae s.l. in Karagwe and Missenyi districts with 82.8% (n=24) and 98.8% (n=166), respectively. Higher proportions of An. gambiae s.s. were from Karagwe district than in Missenyi district. The PCR results have revealed that, all An. funestus were An. funestus s.s. in both districts (Figure 2).
Figure 2: Sibling species of An. gambiae s. l. and An. funestus in Karagwe and Missenyi districts

Social demographic characteristics of study participants

A total of 396 study participants were interviewed from Missenyi and Karagwe districts. Missenyi contributed 50% (n=198) of the study participants. Majority of the study participants were males 59.6% (n=236). The age of respondents was categorized whereby age group 21-30 years made up the highest percentage (27.3%, n=108). Majority of the participants (73.7%, n=292) were married. The economic activity of most of the participants in both districts was farming (83.8%, n=332). When education level was assessed, many of the participants had a primary level of education (57.3%, n=227) (Table 2).
Table 2: Social demographic characteristics of study participants (n=396)

| Characteristics          | Missenyi district, n (%) | Karagwe district, n (%) | Total  |
|--------------------------|--------------------------|-------------------------|--------|
| **Age**                  |                          |                         |        |
| <20                      | 10 (5.1)                 | 12 (6.1)                | 22     |
| 21-30                    | 48 (24.2)                | 60 (30.3)               | 108    |
| 31-40                    | 39 (19.7)                | 54 (27.3)               | 93     |
| 41-50                    | 41 (20.7)                | 32 (16.2)               | 73     |
| 51-60                    | 23 (11.6)                | 18 (9.1)                | 41     |
| >60                      | 37 (18.7)                | 22 (11.1)               | 59     |
| **Sex**                  |                          |                         |        |
| Male                     | 118 (59.6)               | 118 (59.6)              | 236    |
| Female                   | 80 (40.4)                | 80 (40.4)               | 160    |
| **Education Level**      |                          |                         |        |
| Never Attended           | 0 (0.0)                  | 0 (0.0)                 | 0      |
| Primary                  | 137 (69.2)               | 90 (45.5)               | 227    |
| Secondary                | 28 (14.1)                | 55 (27.8)               | 83     |
| University               | 1 (0.5)                  | 24 (12.1)               | 25     |
| **Marital Status**       |                          |                         |        |
| Single                   | 16 (8.1)                 | 19 (9.6)                | 35     |
| Married                  | 131 (66.2)               | 161 (81.3)              | 292    |
| Divorced                 | 41 (20.7)                | 18 (9.1)                | 59     |
| Widow/Widower            | 10 (5.1)                 | 0 (0.0)                 | 10     |
| **Occupation**           |                          |                         |        |
| Fishing                  | 11 (5.6)                 | 0 (0.0)                 | 11     |
| Farming                  | 169 (45.4)               | 163 (82.3)              | 332    |
| Animal Keeping           | 42 (2.0)                 | 12 (6.1)                | 16     |
| Market(gulio)            | 5 (2.5)                  | 7 (3.5)                 | 12     |
| Business                 | 9 (4.5)                  | 16 (8.1)                | 25     |

Human behaviors in relation to Indoor residual spray protection

A total of 396 participants were interviewed. In Missenyi 2.1% (n=24) reported to carry out fishing activities from 08:00 am to 12:00 noon. Participants were also interviewed on malaria diagnosis in Missenyi and Karagwe those reported to be diagnosed positive for malaria were 36.9% (n=73) and 34.8% (n=69), respectively. There was no association between where they sit while having dinner and malaria diagnosis. Participants were also interviewed about the time...
they go to sleep. Majority (59.6%, n=118) in Missenyi reported going to bed from 8:00 to 10:00 pm which is earlier than in Karagwe where majority (45.5%, n=90) go to sleep from 10 p.m. to 12 midnight. In both Missenyi and Karagwe, majority 84.3% reported staying inside their houses after the evening meal and only 4% and 2% reported to have dinners outdoors in Missenyi and Karagwe, respectively.

Most of the interviewed participants 95.9% (n=187) in Missenyi reported to leave the walls of their rooms as they are after they were sprayed while 1.5% (n=3) reported to plaster them after they were sprayed. Majority of participants, 51.5% (n=102) from Karagwe reported to go out at night for a refreshments. Majority 84.8% (n=168) and 89.4% (n=177) of the respondents in Missenyi and Karagwe, respectively do not use any measure to protect themselves against mosquito bites while to go out at night for a refreshments. In Missenyi, only 13.6% (n=27) wear long sleeved shirts and trousers for protection and 1.5% (n=3) respondents reported using mosquito repellants (Table 3).

**Table 3: Showing frequency of participant responses to various variables regarding community behaviors in relation to IRS protection in Missenyi and Karagwe districts**

| Variables                | Frequency (n) | Percentage (%) |
|--------------------------|---------------|----------------|
|                          | Missenyi | Karagwe | Missenyi | Karagwe |
| Fishing time             |          |          |          |          |
| 4:00 a.m to 6:00 a.m    | 2       | 0       | 1.0      | 0       |
| 4:00 p.m to 6:00 p.m    | 5       | 4       | 2.5      | 2       |
| 06:00 p.m to 08:00 p.m  | 2       | 0       | 0.1      | 0       |
| 08:00 am to 12:00 noon  | 24      | 0       | 12.1     | 0       |
| Not applicable           | 165     | 194     | 83.3     | 98      |
| Hours spent when farming? | n =198 | n = 198 |
|--------------------------|--------|--------|
| Less than 2 hours        | 21     | 13     |
| Two to four hours        | 92     | 49     |
| Four to 6 hours          | 28     | 52     |
| Six to eight hours       | 3      | 17     |
| More than eight hours    | 0      | 14     |
| Not applicable           | 54     | 53     |

| Sitting place when having dinner or sapper n = 198 | n = 198 |
|---------------------------------------------------|--------|
| Outside the house                                 | 8      | 4      |
| In the kitchen                                     | 6      | 39     |
| Inside the house                                  | 184    | 155    |

| Sleeping time at night n= 198 | n = 198 |
|-------------------------------|--------|
| 8 p.m to 10 p.m               | 118    | 55     |
| 10 p.m to 12 midnight         | 69     | 90     |
| 12 midnights to 2 am          | 2      | 16     |
| 2 am to 4 a.m                 | 9      | 37     |

| Where do you stay after dinner before sleeping? n= 198 | n = 198 |
|-------------------------------------------------------|--------|
| Outside the house                                    | 16     | 26     |
| Go out for a drink                                   | 15     | 5      |
| Watch TV/radio inside                                | 167    | 167    |

| Do you go out during the night? n = 198 | n = 198 |
|----------------------------------------|--------|
| YES                                    | 56     | 102    |
| NO                                     | 142    | 92     |

| Hours spent out at night n = 198 | n = 198 |
|----------------------------------|--------|
| Less than 2 hours                | 29     | 61     |
| 3 hours to 5 hours               | 31     | 42     |
| Not applicable                   | 138    | 95     |

| Protective measures n = 198 | n = 198 |
|----------------------------|--------|
| Wear long sleeved shirts and trousers | 27     | 21     |
| Mosquito repellents         |        |        |
| I don’t use any measure     | 3      | 0      |

Qualitative analysis from Focused Group Discussion (FGD)

A total of 4 focused group discussions were conducted; two in both districts. Themes identified were; community members’ acceptance of IRS campaign as the useful control measure against malaria vectors though reported a number of challenges, targets for mosquito control...
interventions, risky community behaviors for mosquito bites, complementary mosquito control measures, and risk of outdoor mosquito bites and influence on the efficiency and effectiveness of IRS. Author’s note: F indicates female and M indicates male and numbers are identity given to members of the group.

Major themes discussed

Theme 1 Community perception on IRS campaign
Community members accept the IRS campaign as the useful control measure against malaria vectors though reported a number of challenges the campaign pose to them, including emergence of insects not formerly present in their houses and increase of mosquitoes later after IRS.

Negative perceptions: “You know after these chemicals are sprayed many insects not formerly present in ours houses come and are very troublesome to kill” (Member 03 F Karagwe).

“To be sincere, in the first three months after spraying, the mosquitoes are few but thereafter, the numbers of mosquitoes increase even more” (Member 01 F Missenyi).

“Last year after spraying, mosquitoes did not die” (Member 05 F Karagwe).

Both females and male reported that malaria transmission have declined by comparing before and after IRS.

“You know in the past every minute you go to hospital they tell you that you have malaria but nowadays it is not easy, or even if they say it is malaria, it is not severe as that of the past before these chemicals” (Member 07 F Missenyi).

Theme 2 Targets for mosquito control interventions
Members in all the groups were asked on where they find many mosquitoes inside and outside their houses, most of them reported to find mosquitoes outside the house in places with water, grasses, coffee trees, near animal sheds and in bathrooms. Inside the house, they reported to find mosquitoes behind doors, in bed rooms and in dark places like under the bed.

“The challenge with mosquitoes is that during the rainy season especially when there are planted beans and maize and presence of stagnant water usually are many” (Member 02 M Missenyi).

“Please this is how I see; many mosquitoes are hiding in the coffee trees, banana trees, and in grasses around our houses” (Member 02 M Karagwe).

“Inside the houses they like to hide behind doors, in bed rooms, and in dark places like under beds but they fear the kitchen because of smoke” (Member 08 M Missenyi).

Theme 3 Risky community behaviors for mosquito bites

Members in all the groups mentioned the following behaviors that expose them to mosquito bites; bathing children outside in the evening, not closing doors and windows early in the evening, not cleaning the environment like not pruning banana leaves, while going to the farm, while farming, while cooking outside the house if no kitchen.

“You know us people in Kagera we grow bananas, sometimes we leave our farms with grasses, weeds and banana leaves where mosquitoes hide” (Member 07 M Missenyi).

“While going to the farm early in the morning there are many mosquitoes and they are biting like bees” (Member 04 F Karagwe).

“While bathing children outside at night mosquitoes are many you know we fear going to the bath rooms” (Member 07 M Missenyi).
Theme 4 complementary mosquito control measures

Community members reported few of other chemicals apart from IRS and protective measures against mosquito bites, most of the members declared not using other chemicals while few reported using example Rungu® and Doom® (pyrethroid insecticide sprays) while few of the members reported to burn eucalyptus tree leaves to serve as mosquito repellents.

“I used Rungu® the time when mosquitoes were many but I stopped using them after spraying these chemicals” (Member 07 M Karagwe).

“When mosquitoes are too many we burn leaves from eucalyptus trees on the door so when mosquitoes smell the smoke they run away, challenge is mosquitoes go inside when smoke is over” (Member 03 M Missenyi).

Theme 5 Risk of outdoor mosquito bites

Most members of the groups reported being outdoors at night before going to sleep without protection which makes them prone to mosquito bites outdoors. However most of them reported not being aware of protection measures against mosquito bites.

“Now life is difficult, we stay out doing business” (Member 02 M Karagwe).

“It is time to prepare food and bath children and all these are done outside” (Member 07 F Missenyi).

“You know in our traditions men do not stay at home watching their wives cook, we normally go out at night making stories with our fellow men” (Member 08 M Karagwe).

“Me I usually catch them and kill them because I don’t see any means to avoid them” (Member 07 F Karagwe).
Theme 6: Influence on efficiency and effectiveness of IRS

Most of the community members reported on a number of factors that might influence the effectiveness and efficiency of IRS, which either emanates from the community itself.

“Some people do not want their houses to be sprayed” (Member number 04 M Missenyi).

“Sometimes it happen that, village leaders and the rich are not involved except us the poor who have few items in the house to take out during the spray exercise”. (Member number 5 F Karagwe).

Discussion

In many part of Africa, the main malaria vectors are the An. gambiae s. s., An. arabiensis and An. funestus [26], this is also the case in Tanzania [27–29]. This study has revealed the abundance of An. gambiae s.l. and An. funestus in Missenyi (indoor residual sprayed district) protection and Karagwe (non-indoor residual sprayed district) in Kagera, Tanzania. Missenyi district was sprayed with pirimiphos-methyl in 2016 and 2017 with 93.7% coverage [30]. Pirimiphos-methyl (Actellic® 300CS) has been reported to have a residual efficacy beyond 9 months on oil paint wall surfaces post-IRS [31]. Unexpectedly, large proportions of An. funestus were collected indoors, despite the fact that, Missenyi district was IRS sprayed. Our findings contrast with the previous finding conducted in the lake zone Tanzania by Kakilla and others [30] which reported very low An. funestus indoor density in most sites including Missenyi district in the as evidenced by monthly mosquito collection for a period of one year in 2017. The findings of the present study were also dissimilar to a study conducted in western Kenya whereby IRS using pirimiphos-methyl was associated with 88% reduction in An. funestus numbers in intervention areas.
compared to non-intervention areas [32]. Indoor residual spray with pirimiphos-methyl has reported to maintain sporozoite rates at low levels hence an important tool for malaria vector control in the area [33]. However, there is limited information on insecticides susceptibility status among *An. funestus* in Tanzania [34]. Missenyi district usually receive high amount of rainfall from March to May hence creates and maintain permanent and semi-permanent breeding sites which are favorable for *An. funestus*. The proportions of *An. funestus* collected indoors were lower in Karagwe district than in Missenyi district, although the area was not sprayed.

The density of adult mosquitoes is determined by the productivity of larval habitats and their proximity to human hosts where they can obtain blood meal [35]. In the present study, the proportion of *An. gambiae* s.l. collected indoors in Missenyi was very low compared to Karagwe district. These findings contrast with the previous study conducted in the lake zone, Tanzania which reported the high density of *An. gambiae* s.l. was collected in Missenyi district post-spray with pirimiphos-methyl [30]. This disparity could be due to rapid species shift as a result of LLINs and IRS interventions implementation [36,37].

A better understanding of mosquito behavioral patterns such as breeding, resting and feeding are essential in selection of the mosquito control interventions [38]. In the present study, large proportion of *An. gambiae* s.l. was collected resting outdoors in Missenyi district while in Karagwe district, majority of *An. gambiae* s.l. was collected resting indoors. Indoors vector control intervention such as LLINs and IRS may result to malaria vectors’ behavior change [14,39]. The malaria vectors that were known to feed and rest indoors may prefer to feed and rest
outdoors in order to avoid contacts with insecticides inside houses and repellent effect of IRS chemicals on the resting behaviors of malaria vectors [13,40]. No An. funestus were collected outdoors in Missenyi district. Our findings is comparable with a study conducted in Burkina Faso which reported the An. funestus prefer to feed indoors than outdoors [41].

In the present study, polymerase chain reaction (PCR) results have showed that, An. arabiensis was the dominant sibling species among the An. gambiae s.l. in Karagwe and Missenyi districts with 82.8% and 98.8%, respectively. This zoophilic species primarily feed and rest outdoors also play important role in malaria transmission [42]. This is in line with a previous study that reported a lesser effect of indoor-based insecticide control on An. arabiensis [9]. Higher proportions of An. gambiae s.s. were from Karagwe district which was unsprayed areas. This species primarily prefer to feed and rest indoors [42]. All An. funestus were An. funestus s. s. in both districts. These findings are in line with a previous study conducted in the northeastern Tanzania [28,43]. However, the reason for recent changes in malaria vectors sibling species composition is less documented.

Human behaviors in relation to IRS protection in Missenyi has revealed that, most of the residents in Missenyi and Karagwe are crop farmers and only a few are animal keepers though most households were observed to keep few animals like pigs, goats, chickens and cows at home. Presence of animals in the compound favors persistence of malaria vectors in the areas under IRS [7]. Most interviewed residents of both Missenyi and Karagwe reported, staying outdoors from 8:00 pm to 10:00 pm. For this case, the time between 8:00 to 10:00 pm, is the particular time when they are protected by IRS intervention. Additionally, majority of people go to sleep
from 10 p.m. to 12 midnight in Karagwe. According to a previous study conducted in Dar es Salaam this is a peak outdoor biting time for *An. arabiensis* [5]. However, in the present study, *An. arabiensis* is the dominant sibling species in *An. gambiae* complex in Missenyi. Despite the fact that, Missenyi district was sprayed, these findings suggest a lowered community protection against malaria vectors offered by IRS.

In this study, very few residents in both Missenyi and Karagwe districts reported using alternative protective measures such as use of mosquito repellents, wearing long sleeved shirts and insecticidal sprays for protecting themselves from mosquito bite while they are outside their houses at night. It is important to device vector control interventions which target outdoor and early biting behavior such as application of larvicides in mosquito breeding sites [44]. This is consistent with a study by [5], and is in contrast to findings in cities in Kenya that reported high usage of personal of protective measures which was reported to be due to their high economic status [45]. Few of the members reported to burn eucalyptus tree leaves to serve as mosquito repellents. A member from Missenyi district stated “When mosquitoes are too many we burn leaves from eucalyptus trees on the door so when mosquitoes smell the smoke they run away, challenge is mosquitoes go inside when smoke is over”. Our findings are supported by the previous studies with regard on the use eucalyptus extracts to repel mosquito vectors [46,47].

Residents in both Missenyi and Karagwe were also asked on where they sit while having evening meal; most of them reported having these meals indoors. This was reported to be influenced by their traditions that they are not allowed to get food outdoors as reported by the residents. The findings are opposite to those by [5] in Dar es Salaam. No any resident reported sleeping
outdoors in both Missenyi and Karagwe which might be due to the fact that these areas have
lower temperatures compared to Dar es Salaam so residents are forced to sleep indoors in search
for warmth. Those who reported resting indoors after dinner were 84.3%. In Missenyi contrast to
findings from studies in Dar es Salaam which showed only 47.8% rested indoors.

All the studied house structures were traditional being muddy with open eaves, no window
screens, unsealed window/door frames and no ceiling. A high number of mosquitoes were
collected from these houses. This is in line with a study by [5] in Dar es Salaam that reported a
reduced mosquito biting rates of malaria vectors under improved housing standard especially
mosquito-proof screening, closed eaves, ceiling and sealed window/door frames.

The present study has revealed the behaviors that predispose community member to mosquito
bites which included; bathing children outdoors in the evening, not closing doors and windows
early in the evening, not cleaning their environment like pruning banana leaves, coffee trees,
slashing of grasses and shrubs in the compound and removal of stagnant water. Poor
environment creates mosquito resting places and breeding sites [48]. They also acknowledged
being bitten by mosquitoes while going to the farm early in the morning also while in farms as a
man from Karagwe declared “Now life is difficult, we stay out doing business”. Also a woman
from Karagwe insisted that “While going to the farm early in the morning there are many
mosquitoes and they are biting like bees”. Our findings concur with a previous study which
reported the occurrence of early morning mosquito biting among malaria vectors [49].
Furthermore, women without kitchens reported being bitten by mosquitoes while cooking
outdoors as supported by a woman from Missenyi “It is time to prepare food and bath children
and all these are done outside”. In addition to that, most of the men reported staying outdoors
before going to sleep at night without any protection against mosquito bites. A member from Karagwe added that “You know in our traditions men do not stay at home watching their wives cook, we normally go out at night making stories with our fellow men”. This situation also reveals how traditional attributes lead people at risk of being bitten by mosquitoes [50].

In high malaria transmission like the study area, health education is deemed important for residents to adopt informed healthy choices. Complementary outdoor protective measures such as; wearing long sleeves, socks, use of mosquito repellents and environmental management should be considered during health promotion strategies. These will help in reducing outdoor malaria transmission.

In the present study the community members in Missenyi and Karagwe districts agreed that, IRS implementation is a useful control measure against malaria vectors. All participants reported that malaria transmission have declined by comparing before and after IRS. As a participant from Missenyi added that “You know in the past every minute you go to hospital they tell you that you have malaria but nowadays it is not easy, or even if they say it is malaria, it is not severe as that of the past before these chemicals”. Our findings are similar to previous studies conducted in Tanzania and elsewhere [50–53]. Although IRS and LLINs has contributed to the decline in malaria in the past decade [54], the continue community sensitization in paramount for sustainability of such interventions [50,55].
On the other hand, participants reported a number of challenges with regard to the IRS campaign posed to them, including emergence of insects such as fleas, bedbugs and lice which were not formerly present in their houses. A participant from Karagwe insisted that, “You know after these chemicals are sprayed many insects not formerly present in ours houses come and are very troublesome to kill”. Our findings are comparable to a previous study conducted in Tanzania by Kaufman and others [50]. The insecticides used for IRS may have irritated the hidden insects on the walls which lead them to increase mobility and become visible to house occupants [50,56]. Furthermore, our study contrast with a previous study which reported IRS reduce other insects and rodents [57].

The present study has revealed the perceived low efficacy of the insecticides used during IRS. The participants reported an increase of mosquitoes later after IRS campaign as pointed out by a woman from Missenyi “To be sincere, in the first three months after spraying, the mosquitoes are few but thereafter, the numbers of mosquitoes increase even more” and also a woman from Karagwe who added that “Last year after spraying, mosquitoes did not die”. These findings are unlike to the previous studies which reported that, IRS reduce mosquitoes and lower the cost of malaria prevention at household level [51,57]. The perceived benefit of IRS such as reduction in mosquito bites, incidental benefits were also reported [52].

In the present study, few participants also mentioned some behavioral factors such as washing walls, repainting, plastering and covering walls using newspapers after IRS which may hinder the efficacy of the intervention. The behaviors are influence by the smell of the insecticide used during IRS. Our findings concur with the previous study whereby modification of wall surfaces post-spraying potentially reduce the tangible IRS coverage, effectiveness and impact hence offer
Providing equate information before and during IRS implementation could raise the awareness about spraying, resulting in high acceptability and sustainability [50,51,58]. Participants also suggested that chemicals used for IRS should have less smell and less allergic to humans.

In this study, participants mentioned location/sites where they use to find mosquitoes. These location included outside houses in the areas with grasses, coffee trees, banana trees, water pools, near animal sheds and in bath rooms. These kinds of mosquito resting places are sometimes not a target for IRS. Indoors they reported finding mosquitoes behind doors, in bed rooms, and in darker areas like under beds. One member from Missenyi stated that, “Inside the houses they like to hide behind doors, in bed rooms, and in dark places like under beds but they fear the kitchen because of smoke”. These findings are in line with the previous finding conducted in south-eastern Tanzania [59]. Community awareness on these sites is a stepping stone towards influencing practices such as environmental cleaning like pruning of banana and coffee trees, slashing of grasses near houses, clearing of stagnant water in compounds especially during the rainy season when breeding sites are many. Sometime microclimate may favor malaria vector to rest in or outside the house [60]. In addition, in both Missenyi and Karagwe areas, residents reported using local herbs for instance burning eucalyptus tree leaves with intension of chasing mosquitoes from their houses using the smoke released. Others reported using insecticide sprays such as “Doom®” and “Rungu®”. The use of locally available herbs for repelling mosquitoes from entering their home have been reported in north-eastern Tanzania and elsewhere [61,62].
In the present study, most of the participants reported on a number of factors that might influence the effectiveness and efficiency of IRS, which are either due to the community itself or from spray operators. Participants reported cases of chemicals not being mixed in the required ratios, not spraying some houses, selling of chemicals to farmers to be used in gardens for killing pests. Our findings are similar to a previous study conducted in Tanzania and Mozambique with regard to quality and performance of the spray operators which affects their acceptability to the IRS intervention [50,55]. With regard to the community itself, few participants reported plastering sprayed walls to avoid bad smell. A man from Missenyi added that “Some people do not want their houses to be sprayed”. A member from Karagwe stated that, “Sometimes it happen that, village leaders and the rich are not involved except us the poor who have few items in the house to take out during the spray exercise”. Difficulties in moving items outside during spray and the lack of uniformity in the spray operation between the rich and the poor, unpleasant smell of the insecticide used were among the reason for them to perceive negatively on the intervention. These findings concur with the previous study conducted in Iran [63]. Community sensitization before and during IRS implementation has to strengthen [50,64]. In the present study, participants have agreed that, IRS intervention is important for malaria control. They also suggested that, close supervision should be in place in order to mitigate the mentioned behaviors that may compromise IRS efficiency and effectiveness.

Conclusion

The findings of the present study have revealed the abundance of malaria vectors in IRS and unsprayed districts. Unexpectedly, large proportions of *An. funestus* were collected indoors, despite the fact that, Missenyi district was sprayed. This situation calls for further studies on *An.*
funestus behaviors and possible reasons for tolerance in sprayed area. The findings have also revealed community perception on IRS intervention which may affects its efficacy and sustainability. Community sensitization before, during and after IRS application needs to be strengthen for getting intended results.

531  **List of abbreviations**

532  ACT: Artemisinin Combination Therapy, BCC: Behavioral Change Communication, CDC: Center for Disease Control, CI: confidence intervals; DDT: Dichloro Diphenyl Trichloroethane, DHIS: District Health Information System, DMVSO: District Malaria Vector Surveillance Officer, FGD: Focused Group Discussion, GEE: Generalized Estimating Equation, IRS: Indoor Residual Spraying, IPT: Intermittent Presumptive Treatment, LSM: Larval Source Management, LLIN: Long Lasting Insecticide Net, mRDT: Malaria Rapid Diagnostic Test, MoHCDGEC: Ministry of Health Community Development Gender Elderly and Children, MUHAS; Muhimbili University of Health and Allied Sciences, NIMR: National Institute for Medical Research, NMCP: National Malaria Control Program, NBS: National Bureau of Standards, PCR: Polymarase Chain Reaction, USAID: United States Agency for International Development, WHO: World Health Organization.

544  **Ethics approval**

545  The present study was approved by the Muhimbili University Health and Allied Science IRB, by the ethical clearance with reference number. DA.287/298/01.A/ dated 29th May, 2018.
Funding

This work was supported by the Ministry of Health Community Development Gender Elderly and Children (MoHCDGEC), Tanzania.

Availability of data materials

The data supporting the conclusion are included within the article.

Authors’ contributions

GK conceived the study, design and performed the study, participated in the field, contributed to interpretation of results and drafted the manuscript; BE contributed to overall study design and critically reviewed the manuscript; BN contributed to overall study design and critical review of the manuscript. All authors read and approved the final version of the manuscript.

Competing interests

The authors declare that they have no competing interests.

Consent for publication

Not applicable.

Acknowledgements

The authors acknowledge the financial support from the Ministry of Health Community Development Gender Elderly and Children (MoHCDGEC), Missenyi District Council through
its Malaria vector surveillance office. The National Institute for Medical Research Amani centre, Muheza for laboratory analysis of samples. Bernard Batenga, Abdallah Zakaria, Wajihu Katakweba, Frednand Johaness and Adolf Ngaiza, are acknowledged for their support during preparation of this work. We are grateful for the cooperation given to us by the Missenyi and Karagwe communities.

References

1. WMR. WHO World Malaria Report 2016. 2016.

2. Sougoufara S, Doucouré S, Sembéne PMB, Harry M, Sokhna C. Challenges for malaria vector control in sub-Saharan Africa: Resistance and behavioral adaptations in Anopheles populations. J. Vector Borne Dis. 2017.

3. MoHCDGEC. Malaria Surveillance Monitoring and Evaluation plan. 2017.

4. TACAIDS. Tanzania- 2011-12 HIV/AIDS and Malaria Indicator Survey 2011-12: Key Findings. Tanzania Comm AIDS (ZAC), Zanzibar AIDS Comm (NBS), Natl Bur Stat (OCGS), Off Chief Gov Stat ICF Int. 2013;16.

5. Geissbühler Y, Chaki P, Emidi B, Govella NJ, Shirima R, Mayagaya V, et al. Interdependence of domestic malaria prevention measures and mosquito-human interactions in urban Dar es Salaam, Tanzania. Malar J. 2007;6.

6. Drake JM, Beier JC. Ecological niche and potential distribution of Anopheles arabiensis in Africa in 2050. Ecological niche and potential distribution of Anopheles arabiensis in Africa in 2050. 2014;13:1–11.
7. Killeen GF, Marshall JM, Kiware SS, South AB, Tusting LS, Chaki PP, et al. Measuring, manipulating and exploiting behaviours of adult mosquitoes to optimise malaria vector control impact. BMJ Glob Heal. 2016;2:doi:10.1136.

8. Gatton ML, Chitnis N, Churcher T, Donnelly MJ, Ghani AC, Charles HJ, et al. The importance of Mosquito behavioral adaptations to malaria control in Africa. Soc Study Evol Evol. 2013;67:1218–30.

9. Sinka ME, Golding N, Massey NC, Wiebe A, Huang Z, Hay SI, et al. Modelling the relative abundance of the primary African vectors of malaria before and after the implementation of indoor, insecticide-based vector control. Malar J. 2016;15.

10. WHO. An operational manual for Indoor Residual Spraying (IRS) for malaria transmission control and elimination. 2015.

11. IRS P|A. TANZANIA Indoor Residual Spraying (IRS 2) Task Order Four END OF SPRAY REPORT. 2016.

12. Haji KA, Thawer NG, Khatib BO, Mcha JH, Rashid A, Ali AS, et al. Efficacy, persistence and vector susceptibility to pirimiphos-methyl (Actellic® 300CS) insecticide for indoor residual spraying in Zanzibar. Parasit Vectors [Internet]. Parasites & Vectors; 2015;8:628. Available from: http://www.parasitesandvectors.com/content/8/1/628

13. Bradley J, Lines J, Fuseini G, Schwabe C, Monti F, Slotman M, et al. Outdoor biting by Anopheles mosquitoes on Bioko Island does not currently impact on malaria control. Malar J. 2015;14.

14. Meyers JJ, Pathikonda S, Popkin-Hall ZR, Medeiros MC, Fuseini G, Matias A, et al. Increasing outdoor host-seeking in Anopheles gambiae over 6 years of vector control on Bioko
15. Mashauri FM, Manjurano A, Kinung S, Martine J, Lyimo E, Kishamawe C, et al. Indoor residual spraying with micro-300CS against malaria vectors in the Lake Victoria basin, Tanzania. 2017;1–21.

16. Degefa T, Yewhalaw D, Zhou G, Lee MC, Atieli H, Githeko AK, et al. Indoor and outdoor malaria vector surveillance in western Kenya: Implications for better understanding of residual transmission. Malar J. BioMed Central; 2017;16:1–13.

17. Sokhna MON and CR. The changes in mosquito vector behaviour and the emerging resistance to insecticides will challenge the decline of malaria. Clin Microbiol Infect. 2013;19.

18. Mwegoha. Environmental protection and conservation of Kagera river at Kyaka. 2014.

19. NBS. 2012 POPULATION AND HOUSING CENSUS Population Distribution by. 2013.

20. Wanzirah H, Tusting LS, Arinaitwe E, Katureebe A, Maxwell K, Rek J, et al. Mind the gap: House structure and the risk of malaria in Uganda. PLoS One. 2015;10.

21. Kreppel KS, Johnson PCD, Govella NJ, Pombi M, Maliti D, Ferguson HM. Comparative evaluation of the Sticky-Resting-Box-Trap, the standardised resting-bucket-trap and indoor aspiration for sampling malaria vectors. Parasites and Vectors. Parasites & Vectors; 2015;8:3–7.

22. Govella NJ, Chaki PP, Mpangile JM, Killeen GF. Monitoring mosquitoes in urban Dar es Salaam: evaluation of resting boxes, window exit traps, CDC light traps, Ifakara tent traps and human landing catches. Parasit Vectors [Internet]. BioMed Central Ltd; 2011 [cited 2014 Jan 23];4:40. Available from: http://www.pubmedcentral.nih.gov/articlerender.fcgi?artid=3069960&tool=pmcentrez&rendertype
23. Gillies & de Meillon. The Anophelinae of Africa South of the Sahara; Ethiopian zoogeographical region. 2nd ed. Institute for Medical Research, Johannesburg, South African; 1968.

24. Scott J. Species Complex Authentication by PCR, Anopheles gambiae Complex. Chapter 8 F Tech. 1993. p. 95–6.

25. Koekemoer LL, Kamau L, Hunt RH, Coetzee M. A cocktail polymerase chain reaction assay to identify members of the Anopheles funestus (Diptera: Culicidae) group. Am J Trop Med Hyg. 2002;66:804–11.

26. Sinka M, Bangs M, Manguin S, Ruyio-palis Y, Chareonviriyaphap T, Coetzee M, et al. A global map of dominant malaria vectors. Parasit Vectors. BioMed Central Ltd; 2012;5:1–11.

27. Derua Y, Alifrangis M, Magesa S, Kisinza W, Simonsen P. Sibling species of the Anopheles funestus group, and their infection with malaria and lymphatic filarial parasites, in archived and newly collected specimens from northeastern Tanzania. Malar J. 2015;14:1–8.

28. Derua YA, Alifrangis M, Hosea KM, Meyrowitsch DW, Magesa SM, Pedersen EM, et al. Change in composition of the Anopheles gambiae complex and its possible implications for the transmission of malaria and lymphatic filariasis in. Malar J [Internet]. Malaria Journal; 2012;11:1. Available from: Malaria Journal

29. Kabula B, Derua YA, Tungu P, Massue DJ, Sambu E, Stanley G. Malaria entomological profile in Tanzania from 1950 to 2010: a review of mosquito distribution, vectorial capacity and insecticide resistance. 2011;13:1–14.
30. Kakilla C, Manjurano A, Nelwin K, Martin J, Mashauri F, Kinung’hi SM, et al. Malaria vector species composition and entomological indices following indoor residual spraying in regions bordering Lake Victoria, Tanzania. Malar J [Internet]. BioMed Central; 2020;19:1–14. Available from: https://doi.org/10.1186/s12936-020-03452-w

31. Haji KA, Thawer NG, Khatib BO, Mcha JH, Rashid A, Ali AS, et al. Efficacy, persistence and vector susceptibility to pirimiphos-methyl (Actellic®; 300CS) insecticide for indoor residual spraying in Zanzibar. Parasites and Vectors [Internet]. Parasites & Vectors; 2015;8:1–7. Available from: http://dx.doi.org/10.1186/s13071-015-1239-x

32. Biology V, Unit C. Implementation of Indoor Residual Spraying of Insecticides for Malaria Control in the WHO African Region Report. 2007;

33. PMI. PMI | AfricaIRS (AIRS) Project Indoor Residual Spraying (IRS) Task Order six AIRS Tanzania Project Entomological Monitoring of 2016 IRS activities final report. Report. 2017;

34. Matiya DJ, Philbert AB, Kidima W, Matowo JJ. Dynamics and monitoring of insecticide resistance in malaria vectors across mainland Tanzania from 1997 to 2017: A systematic review. Malar. J. 2019.

35. Takken W, Verhulst NO. Host preferences of blood-feeding mosquitoes. Annu Rev Entomol. 2013;58:433–53.

36. Chinula D, Hamainza B, Chizema E, Kavishe DR, Sikaala CH, Killeen GF. Proportional decline of Anopheles quadriannulatus and increased contribution of An. arabiensis to the An. gambiae complex following introduction of indoor residual spraying with pirimiphos-methyl: An observational, retrospective secondary analysis of pre-. Parasites and Vectors. Parasites &
37. Musiime AK, Smith DL, Kilama M, Rek J, Arinaitwe E, Nankibirwa JJ, et al. Impact of vector control interventions on malaria transmission intensity, outdoor vector biting rates and Anopheles mosquito species composition in Tororo, Uganda. Malar J [Internet]. BioMed Central; 2019;18:1–9. Available from: https://doi.org/10.1186/s12936-019-3076-4

38. WHO. Manual on Practical Entomology in Malaria. Geneva; 1975.

39. Thomsen EK, Koimbu G, Pulford J, Jamea-Maisa S, Ura Y, Keven JB, et al. Mosquito behavior change after distribution of bednets results in decreased protection against malaria exposure. J Infect Dis. 2017;215:790–7.

40. Reddy M, Overgaard H, Abaga S, Reddy V, Caccone A, Kiszewski A&, et al. Outdoor host seeking behaviour of Anopheles gambiae mosquitoes following initiation of malaria vector control on Bioko Island, Equatorial Guinea. Malar J. BioMed Central Ltd; 2011;10:1–10.

41. Guelbeogo WM, Sagnon N, Liu F, Besansky NJ, Costantini C. Behavioural divergence of sympatric Anopheles funestus populations in Burkina Faso. Malar J. 2014;13:1–8.

42. Manguin S. Anopheles Mosquitoes: New Insights into malaria vectors. Croatia: InTech; 2013.

43. Derua YA, Kisinza WN, Simonsen PE. Differential effect of human ivermectin treatment on blood feeding Anopheles gambiae and Culex quinquefasciatus. Parasit Vectors. 2015;8:1–6.

44. Geissbühler Y, Kannady K, Chaki PP, Emidi B, Govella NJ, Mayagaya V, et al. Microbial larvicide application by a large-scale, community-based program reduces malaria infection prevalence in urban Dar Es Salaam, Tanzania. PLoS One. 2009;
45. Macintyre K, Keating J, Sosler S, Kibe L, Mbogo CM, Githeko AK, et al. Examining the determinants of mosquito-avoidance practices in two Kenyan cities. Malar J. 2012;10:1–10.

46. Trigg JK. Evaluation of a eucalyptus-based repellent against Anopheles spp. in Tanzania. J Am Mosq Control Assoc. 1996;12:243–6.

47. Maia MF, Moore SJ. Plant-based insect repellents: a review of their efficacy, development and testing PMD from lemon eucalyptus (Corymbia citriodora) extract. Malar J. 2011;10:1–15.

48. Sattler MA, Mtasiwa D, Kiama M, Premji Z, Tanner M, Killeen GF, et al. Habitat characterization and spatial distribution of Anopheles sp. mosquito larvae in Dar es Salaam (Tanzania) during an extended dry period. Malar J. 2005;4:4.

49. Moiroux N, Damien GB, Egrot M, Djenontin A, Chandre F, Corbel V, et al. Human exposure to early morning Anopheles funestus biting behavior and personal protection provided by long-lasting insecticidal nets. PLoS One. 2014;9.

50. Kaufman MR, Rweyemamu D, Koenker H, MacHa J. My children and I will no longer suffer from malaria: A qualitative study of the acceptance and rejection of indoor residual spraying to prevent malaria in Tanzania. Malar J. 2012;11:1–17.

51. Dimas HJ, Sambo NM, Ibrahim MS, Ajayi IOO, Nguku PM, Ajumobi OO. Coverage of indoor residual spraying for malaria control and factors associated with its acceptability in Nasarawa state, north-central Nigeria. Pan Afr Med J. 2019;33:1–9.

52. Suuron VM, Mwanri L, Tsourtos G, Owusu-Addo E. An exploratory study of the acceptability of indoor residual spraying for malaria control in upper western Ghana. BMC Public Health; 2020;1–11.
53. Munga S, Kimwetich Z, Atieli F, Vulule J, Kweka EJ. Knowledge and perceptions about indoor residual spray for malaria prevention in numberes division, Nandi county in central province of Kenya. Tanzan J Health Res. 2017;19:1–9.

54. Bhatt S, Weiss DJ, Cameron E, Bisanzio D, Mappin B, Dalrymple U, et al. The effect of malaria control on Plasmodium falciparum in Africa between 2000 and 2015. Nature. Nature Publishing Group; 2015;526:207–11.

55. Magaço A, Botão C, Nhassengo P, Saïde M, Ubisse A, Chicumbe S, et al. Community knowledge and acceptance of indoor residual spraying for malaria prevention in Mozambique: A qualitative study. Malar J [Internet]. BioMed Central; 2019;18:1–12. Available from: https://doi.org/10.1186/s12936-019-2653-x

56. Rafatjah H. Problem of resurgent bed-bug infestation in malaria eradication programmes. J Trop Med Hyg. 1971;74.

57. Ediau M, Babirye JN, Tumwesigye NM, Matovu JK, MacHingaidze S, Okui O, et al. Community knowledge and perceptions about indoor residual spraying for malaria prevention in Soroti district, Uganda: A cross-sectional study. Malar J. 2013;12:1–9.

58. Opiyo MA, Paaijmans KP. “We spray and walk away”: Wall modifications decrease the impact of indoor residual spray campaigns through reductions in post-spray coverage. Malar J [Internet]. BioMed Central; 2020;19:1–6. Available from: https://doi.org/10.1186/s12936-020-3102-6

59. Msugupakulya BJ, Kaindoa EW, Ngowo HS, Kihonda JM, Kahamba NF, Msaky DS, et al. Preferred resting surfaces of dominant malaria vectors inside different house types in rural south-eastern Tanzania. Malar J [Internet]. BioMed Central; 2020;19:1–15. Available from:
60. Paaijmans KP, Thomas MB. The influence of mosquito resting behaviour and associated microclimate for malaria risk. Malar J. 2011;10:1–7.

61. Kweka EJ, Mosha F, Lowassa A, Mahande AM, Kitau J, Matowo J, et al. Ethnobotanical study of some of mosquito repellent plants in north-eastern Tanzania. Malar J. 2008;7:1–9.

62. Shukla D, Wijayapala S, Vankar PS. Effective mosquito repellent from plant based formulation. Popul Dyn Aedes Mosq larvae from peridomestic water bodies. 2018;5:19–24.

63. Madani A, Soleimani-Ahmadi M, Davoodi SH, Sanei-Dehkordi A, Jaberhashemi SA, Zare M, et al. Household knowledge and practices concerning malaria and indoor residual spraying in an endemic area earmarked for malaria elimination in Iran. Parasites and Vectors. Parasites & Vectors; 2017;10:1–9.

64. Munguambe K, Pool R, Montgomery C, Bavo C, Nhacolo A, Fiosse L, et al. What drives community adherence to indoor residual spraying (IRS) against malaria in Manhiça district, rural Mozambique: A qualitative study. Malar J. 2011;10:1–13.