IMPLEMENTATION OF
INDOOR RESIDUAL
SPRAYING
TO ASSESS FEASIBILITY
IN SIERRA LEONE

FINAL REPORT OF A PILOT PROJECT
Implementation of indoor residual spraying to assess feasibility in Sierra Leone: Final report of a pilot project

1. Malaria – prevention and control
2. Insecticides – administration and dosage – supply and distribution
3. Mosquito Control – methods
4. Health Plan Implementation
5. Pilot Projects
6. Sierra Leone
7.

I. World Health Organization. Regional Office for Africa II. Title

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<td>Antenatal care</td>
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<td>BMP</td>
<td>Best management practices</td>
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<td>DHMP</td>
<td>District health management team</td>
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<td>EIA</td>
<td>Environmental impact assessment</td>
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<td>EWT</td>
<td>Exit window trap</td>
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<td>FAO</td>
<td>Food and Agriculture Organization</td>
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<td>GFATM</td>
<td>Global Fund against AIDS, Tuberculosis and Malaria</td>
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<tr>
<td>IMNCI</td>
<td>Integrated management of neonatal and childhood illnesses</td>
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<td>IRS</td>
<td>Indoor residual spraying</td>
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<td>ITN</td>
<td>Insecticide-treated bed net</td>
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<td>Long-lasting insecticide-treated net</td>
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<td>MIP</td>
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<td>MOHS</td>
<td>Ministry of Health and Sanitation</td>
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<td>NGO</td>
<td>Non-governmental organization</td>
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<td>NMCP</td>
<td>National Malaria Control Programme</td>
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<td>PEA</td>
<td>Programmatic environmental assessment</td>
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<td>PHU</td>
<td>Primary health unit</td>
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<td>PMI</td>
<td>President's Malaria Initiative</td>
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<td>PPE</td>
<td>Personal protective equipment</td>
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<td>PSC</td>
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<td>Training facilitator</td>
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EXECUTIVE SUMMARY

Malaria is holoendemic (perennial intense transmission). It is also one of the leading causes of morbidity and mortality in Sierra Leone, accounting for about 47% of outpatient morbidity. Prior to the inception of this study, malaria control efforts included treatment of cases, use of long-lasting insecticide-treated nets (LLINs), and malaria prevention advocacy programmes. The initial objective of this pilot project was to consider indoor residual spraying (IRS) as part of the malaria control scale-up programme in four pilot districts (Bo, Bombali, Kono and Western Area Rural). It aimed to cover an average minimum of 85% of the target population – hence to protect an estimated 851 000 people.

The project was carried out between 2010 and 2012 following a systematic process that began with feasibility studies in Bo, Bombali, Kono and Western Area Rural districts. These localities were chosen on the basis of high malaria transmission; they were therefore ideal as pilot areas for IRS. Levels of insecticide susceptibility of the vector were checked to determine the class of insecticide to be used for IRS. Both theoretical and practical training of national and district staff in IRS was conducted. The training covered proper application and handling of insecticides, as well as waste disposal and waste management. Malaria disease data were collected both in health centres and within the community. Community education and sensitization on IRS was conducted. Operational quality control and insecticide efficacy evaluation through mosquito bioassays were also conducted to ensure quality spraying.

The programme underwent two phases of spraying between December 2010 and June 2012. Various activities were undertaken to monitor and evaluate the outcome and impact of the pilot project. Qualitatively, the study included District health management team (DHMT) task group strategic evaluation meetings; there were also community focus group interviews and discussions. Technically and quantitatively, the study involved collection of vector, parasitological and primary health unit (PHU) epidemiological data.

Two vector species were identified in all the four districts: *Anopheles gambiae* s.l. and *An. funestus*. However, their densities were low, and none of them were infected. Consequently, determination of infectivity rate was not possible.

Insecticide susceptibility tests carried out in the four districts showed emergence of resistance to DDT. All mortality rates ranged from 90% to 97%, suggesting the need for confirmation of resistance. The knockdown rates were not significantly different in mosquitoes collected from all the four areas (p=0.8). The mortality rates were not significantly different among mosquitoes collected in all the four areas (p=1).

The implementation of the IRS programme using lambda-cyhalothrin insecticide was very successful, with a spraying coverage rate of about 98% in all pilot districts. The residual effect of lambda-cyhalothrin in the four districts was effective four months after spraying, although this could not be ascertained for up to six months due to financial constraints. The spraying remained effective for up to four months, with an average mortality rate of 96%.
There were more malaria cases reported in 2009 (50%) and 2010 (53.4%) in health centres situated in Bo, Bombali, Kono and the Western Area, compared to malaria cases reported in 2011 and 2012 after the launch of the IRS programme. Generally, there was no significant decline in the number of malaria cases during the IRS pilot project from 2010 to 2012. Children under the age of 5 years were the most affected with malaria in all the four districts in 2009 and 2010, but this declined slightly in 2011 and 2012.

In conclusion, the pilot IRS implementation programme was successful. It received overwhelming acceptance, and community participation was impressive. However, the monitoring and evaluation plan was poorly implemented as the key indicators, including entomological parameters (vector density, infection rates, etc.) and epidemiological indicators were inconsistently collected. It is therefore recommended that IRS be expanded to other districts. Capacity for developing resources (human, infrastructure, financial) should be strengthened at national and district levels. On the other hand, surveillance, monitoring, and evaluation should be an integral part of the IRS programme. Indeed, required capacity in this area should be created in line with IVM approaches.
1. INTRODUCTION

1.1 Country profile

Sierra Leone is a West African country bordered by Liberia to the east and south-east, Guinea to the north and northeast, and the North Atlantic Ocean to the west and south. Administratively, Sierra Leone is divided into four provinces. Each province is subdivided into districts, and each district is divided into chiefdoms. In all, there are 14 districts and 149 chiefdoms. The Western Province is divided into 69 wards. Among the 14 districts, there are five city councils and 14 district councils, including Freetown, the capital, for a total of 19 local councils (SSL, 2006). The country has eight main river systems: Great Scarcies, Little Scarcies, Rokel, Jong, Sewa, Wanjé, Moa, and Mano. The rivers typically flow from northeast to southwest, eventually emptying in the Atlantic Ocean.

The country has a tropical climate with temperatures ranging from 21 °C to 32 °C and a mean daily temperature of 25 °C. There are two major seasons: the dry season (November to April) and the wet season (May to October), with heavy rains in July and August. Sierra Leone has an average annual rainfall of approximately 3200 mm. Relative humidity is high, ranging from 60% to 90%. The country has a varied terrain that goes from coastal swamps, to inland swamps, to a rainforest, to one of the highest mountains in West Africa ─ the Bintumani which stands at 2200 m. The vegetation is mainly secondary palm bush interspersed with numerous swamps, which are mostly cultivated for rice. These swamps provide ideal breeding habitats for the anopheline vectors of malaria. Moreover, the coast line has several mangrove swamps, which provide the breeding sites for *Anopheles melas* mosquitoes. This species of mosquito is one of the major vectors of malaria, besides *An. gambiae* and *An. funestus*.

Sierra Leone has a population of 5,525,000, with children under five years comprising 17%. The annual population growth rate is 2.6% with a total fertility rate reaching 6.5 per woman.

1.2 Malaria burden

Malaria is endemic in Sierra Leone, with stable and perennial transmission in all parts of the country. The entire population is therefore at risk of developing the disease. Malaria accounts for about 50% of outpatient morbidity, and is presently the leading cause of morbidity and mortality among children under the age of 5, with a mortality attributed to malaria estimated to be 38% among this age group, and 25% for all ages (Outpatient morbidity statistics, MoHS, 2009, MIS 2010). It is estimated that about 2,240,000 annual outpatient visits are due to malaria, of which about 1,000,000 patients are under five years of age. Pregnant women and children under 5 constitute 4.4% and 17.7% respectively of the current total population that is most vulnerable.
Of the four species of human *Plasmodium* in Sierra Leone, *P. falciparum* and *P. malariae* are prevalent. *Plasmodium falciparum* which causes the severest form of the disease accounts for over 90% of all malaria infections. The major malaria vectors in Sierra Leone are members of the *Anopheles gambiae* complex and *An. funestus*.

### 1.3 Socioeconomic impact

Even though the socioeconomic impact of malaria has not been assessed, the cost of treatment borne by families and the cost of lost days of work can be considerably high. The effects of malaria on the community may include substantial financial losses due to payment for treatment or consultations, anti-malarial drugs and vector control measures at the household level. Sickness may cause further losses due to an inability to work or the need to look after other family members, thereby preventing attendance at work. Other impacts include absenteeism and general overburdening of the already over-stretched health service. Overall productivity for the country as a whole is significantly affected.

### 1.4 History of malaria control in Sierra Leone

The National Malaria Control Programme (NMCP), under the Directorate of Disease Prevention and Control (DPC), is mandated to manage all malaria control efforts in the country. The guiding document for malaria control is the National Malaria Control Programme Strategic Plan, 2011–2015. This strategy informs all interventions and sets national targets based on established indicators.

Malaria control efforts in Sierra Leone have focused on scaling up interventions, which include the ownership and use of long-lasting insecticide-treated nets (LLINs), providing prompt effective treatment with artesunate + amodiaquine (ASAQ) within 24 hours of onset of symptoms, and intermittent preventive treatment in pregnancy (IPTp) for pregnant women. Cross-cutting interventions such as behavior change communication have been critical for increasing knowledge of prevention and rapid case identification and management.

In Sierra Leone, net distribution is conducted routinely through public health facilities that target pregnant women and children under age 5. In 2010, a mass distribution of over 3.2 million LLINs was conducted to complement the country’s routine net distribution programme. Although the NMCP has not been using indoor residual spraying (IRS) as one of the main vector control interventions, in 2010, it identified the method as one of the control strategies to be used in the country in selected geographic areas. Pilot tests were carried out in selected chiefdoms in four districts with a view to documenting experiences and extending the programme to other chiefdoms. Financial and technical support came from WHO. The feasibility pilot study involved all the relevant stakeholders, including the Ministry of Health and Sanitation, WHO, the Ministry of Agriculture, and local communities at district level.
2. THE IRS PILOT PROJECT

IRS has been used for decades and has helped eliminate malaria from many parts of the world, particularly where the mosquitoes are indoors resting, and where malaria is seasonally transmitted. In the WHO African Region, IRS was implemented only in a few countries in southern and eastern Africa until 2005, protecting about 13 million people in epidemic-prone areas. Since 2005, however, the number of countries introducing IRS as part of their malaria control programme has significantly increased. Today, the method is applied in more than 30 countries, and protecting more than 76 million people, including those in perennial transmission areas of the Region. It is in line with this trend that the NMCP of Sierra Leone decided to introduce and scale up IRS. In 2010, Sierra Leone launched IRS in the four selected districts of Bo, Bombali, Kono, and Western Area Rural as a pilot vector control intervention. The main objective was to assess feasibility and community acceptability of IRS, and to generate the evidence base with which to scale up the intervention as a component of the IVM strategy.

2.1 The goal

The goal of the IRS pilot project was to accelerate malaria control for increased impact in order to achieve national, regional and global targets. Consequently, the IRS pilot was considered as part of the malaria control strategies scale-up programme. The pilot programme was initially introduced in four pilot districts and targeted a minimum coverage of 85% of the target population, the aim being to protect an estimated 851,000 people.

The specific objective of the project was to assess its feasibility and acceptability by communities, determine the effectiveness of IRS under local circumstances, and expand the intervention to other districts in the country. This specific objective also involved:

(a) measuring the impact of IRS on malaria prevalence levels in the four districts; and
(b) monitoring the reduction of vector densities and the impact of IRS on local vector populations.

2.2 Implementation of IRS

The pilot IRS programme was implemented in two phases: Phase 1 (December 2010) and Phase 2 (April 2012). Phase 1 focused on capacity building, planning, preparation and collection of baseline data and analysis, in addition to launching the first round of IRS. The information collected included malaria morbidity data from health facilities in the selected districts, vector susceptibility data, and community awareness and perception. All relevant stakeholders, including the Ministry of Health and Sanitation, WHO, the Ministry of Agriculture and local communities were involved. Phase 2 focused on micro planning, training of IRS operators and implementation of IRS, including monitoring and evaluation of the key indicators.
A pre-planning meeting was held in February 2012, followed by recruitment and training of spray operators in April 2012 before actual spraying started in May 2012.

3. MATERIALS AND METHODS

3.1 Phase 1: December 2010

The aim of this phase was to collect baseline information, support overall capacity building, plan for IRS operations, work out logistical aspects, and implement the IRS intervention. Phase 1 started in mid-2010 with a series of meetings to discuss planning, preparation and implementation. These meetings were followed by district selection, baseline data collection and analysis, including data on malaria morbidity, vector susceptibility to insecticides and community awareness and perception on malaria and vector control.

Selection of the districts to be studied

Four districts were selected for the implementation of the IRS pilot project: Bo, Bombali, Kono and Western Area Rural (see Figure 1). The following key criteria were considered in the selection of the districts: (1) high seasonal transmission of malaria; (2) good community setup; (3) availability of resources (financial, human, infrastructure), and economic activities in the area; and (4) availability of sentinel sites for vector surveillance. The selection of these districts was done in consultation with the NMCP, WHO, and the community, and in collaboration with other relevant stakeholders. The districts were subsequently visited to engage all the relevant stakeholders through district health management team (DHMT) task group meetings. What follows is a short description of each of the selected districts.

(a) Bo District is located in the Southern Province with a population of 561,524. The district’s main economic activity is trading, gold and diamond mining.

(b) Bombali District is located in the Northern Province. It has a population of 434,319. Its economic activities are mining, trading and agriculture.

(c) Kono, the diamond-rich district in the Eastern Province has a population of 352,328. Apart from diamond and gold mining, agricultural production of rice, coffee and cacao are the major economic activities in the district.

(d) Western Area Rural is located around the peninsula in the Western Area of the country. It has a population of 197,098. The district’s main economic activity is small-scale agriculture, trading and other commercial activities.
Baseline data collection
A pre-project evaluation and baseline data collection were undertaken to assess feasibility, acceptability by communities, and the effectiveness of IRS under local circumstances, the aim being to scale up the intervention and reduce the malaria burden in the country. The following key indicators were measured: community acceptance, malaria prevalence and vector densities. Bioassays were also carried out. Additionally, community surveys were conducted to ensure acceptance and to enlist the full participation of all stakeholders.

(a) Assessment of uncomplicated malaria at the health facilities
The evaluation of malaria morbidity among outpatients was done by passive case detection within the health facilities in each district for the years 2009 to 2012. Data were collected on the number of febrile cases among outpatients (fever defined as an auxiliary temperature of >37.5 °C) and the number of suspected “malaria” cases among outpatients. The data were categorized by age, and adult females were classified as pregnant or not pregnant. Complete monthly malaria morbidity and mortality returns were collected and entered into the project database.

(b) Entomological data collection
Adult mosquito numbers were monitored by monthly sampling during the baseline survey. In each district three to four sentinel villages or chiefdoms were monitored for entomological activity. In each sentinel village or chiefdom, simple random sampling was used to select houses or sites for adult mosquito collections. Sentinel villages
were selected based on a combination of geographic location, accessibility and proximity to health facilities.

As to mosquito collection methods:

(i) Exit window trap (EWT) collection was done. Two exit-window traps were used to collect mosquitoes exiting from houses in each sentinel site in the district per month. The window traps were placed over the windows on the outside of the house at 6.00 p.m. Trapped mosquitoes were collected the following morning before 9.00 a.m. using a mouth aspirator.

(ii) Pyrethrum spray collections (PSC) were also used. Pyrethrum spray collections were positioned inside houses between 6.00 a.m. and 8.00 a.m. Occupants of the house were asked to wait outside during the procedure. All food items and drinking utensils were removed from the house. White sheets were spread on the floor or placed over furniture. Two collectors, one inside the house and one outside, sprayed around the eaves with 0.025% pyrethrum emulsifiable concentrate mixed with 0.1% piperonyl butoxide in kerosene. The collector inside the house then sprayed the roof and walls. The house was closed for 10 minutes, after which dead mosquitoes were collected from the sheets and transferred to the laboratory on moist filter paper inside Petri dishes. During sampling, the number of children and adults who reportedly slept in the house the previous night was recorded, as was the presence of ITNs.

(iii) All mosquitoes caught by each of the above methods were identified by species morphologically (Gillies and DeMeillon, 1968; Gillies and Coetzee, 1987). Their physiological status was determined by observation, and sporozoite infections were determined by dissection of the salivary glands.

(c) Assessment of vector susceptibility to insecticides

In order to perform insecticide resistance susceptibility tests, batches of 20 non-blood-fed female anopheline mosquitoes, aged 2 to 3 days, were used for the assays as described in Annex 1. Briefly, susceptibility tests were done as per WHO standard guidelines (WHO 2013). Twenty to 25 female *Anopheles gambiae* s.l. mosquitoes aged about 2 to 3 days and non-fed females were exposed to the diagnostic dosages of standard WHO insecticide papers. The mosquitoes were exposed to a dosage of 4% DDT (organochloride) pyrethroids, 0.05% deltamethrin, 0.05% lambda cyhalothrin, 0.1% fenitrothion (organophosphate) and 0.1% bendiocarb (carbamate) using WHO test papers. Numbers of mosquitoes knocked down during exposure time were recorded at intervals of 10, 15, 20, 30, 40, 50, and 60 minutes of exposure. The mosquitoes knocked down were then transferred to the holding tubes where 10% glucose was provided. They were held here for 24 hours, after which mortality was recorded. This susceptibility test was conducted under 26 °C to 29 °C and a relative humidity of 70% to 80%. When mortality in control exceeded 20%, the experiment was repeated; and if the control mortality was between 5% and 20%, the Abbots formula was used to correct percentage mortality (WHO, 2013). The current WHO criteria for the
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Planning and preparation

During the planning and preparation phase, key deliverables, including procurement of insecticides and equipment, training of supervisors and spray operators, community mobilization and awareness, were agreed upon. A detailed log-frame was then developed and shared with all stakeholders (Annex 3). Each district, in consultation with the stakeholders, prepared a micro-planning schedule for the district IRS campaign. The schedule included reparation of a district map with all the district villages or towns plotted; production of an operational plan of movement; organization of logistical and other support from within and from other stakeholders (NGOs, etc.); preparation of social mobilization plans; production of communication and reporting tools; organization of transport and other logistics; and designing of monitoring and reporting strategies.

Relevant logistical planning was carried out and all materials required were formally purchased by WHO and delivered to NMCP. Equipment, insecticides and administrative and operational tools for the programme, including the various forms required for spraying operations, were designed and procured as necessary. In order to minimize delays in acquisition of the IRS materials, the procurement of equipment and insecticides was done by informal tender instead of following the formal tender procedure. This was approved by the procurement office to enhance administrative logistics.

(a) Training of trainers (TOT) and spray operators

Two levels of training were conducted in each district; training of trainers (TOTs) and training of spray operators. These courses were conducted in accordance with the WHO “Manual for indoor residual spraying” (WHO 2002). Trainers were trained as supervisors (and facilitators) at the national level while spray operators were trained as IRS operators at the district level. An intensive seven-day training-of-trainers (TOT) workshop was conducted, immediately following which the project provided guidance for the newly trained team leaders to conduct and lead an intensive five-day training course for spray operators, thereby putting their acquired skills and knowledge to the test. Meetings were held with the task groups in all four districts where the full IRS implementation programme was discussed, and issues of community mobilization and participation emphasized. The communities were given the responsibility of recruiting spray operators from their respective districts in time for the training. Relevant issues on information, education and communication, and on social mobilization were also outlined. A five-day intensive training course was then carried out concurrently in all the districts by the TOTs. All district training courses were monitored as they were being implemented, and all relevant operational issues and strategies were also developed.

(b) Community awareness and engagement

In all IRS operations, the imminent risk of residential exposure is always present and cannot be overlooked. With the plans to help guide operations, the project reached
out to communities to gain their support and acceptance. Team members visited all
the four districts and held stakeholder information meetings with key community
leaders and groups to introduce the planned pilot project. District authorities and
programme staff worked with relevant boards, committees, and non-governmental
organizations to carry out appropriate information and education campaigns to
sensitise residents on IRS activities, in accordance with relevant guidelines. The
campaigns focused on the elements of residential safety during the IRS operations,
and the necessary community support in providing mixing water and having their
households ready in time for the spray operators. Further, the campaign meetings
helped to prepare local residents for spraying, and gained their support by raising
awareness of IRS practices and benefits. Focus group discussions, questionnaires and
stakeholder meetings were used to assess community acceptance and perception of,
and compliance with, IRS programme implementation plans before and after the
completion of the spraying.

(c) Environmental mitigation and monitoring plan

An environmental assessment preview in relation to IRS was carried out for the
proposed pilot districts. Mitigation measures were designed and put in place.
Anticipated adverse events from exposure to the environment, or affecting human
and animal health were to be avoided or minimized. In order to ensure this,
appropriate correction and mitigation plans were put in place and strategies against
any cumulative adverse effects (USAID-PEA, 2007), both in the communities and
among the spray operators, were developed. The following measures and mechanisms
were used to mitigate any adverse impact of IRS in all the districts.

(i) Personal protective equipment (PPE) meeting recommended standards by WHO
for IRS activities was provided to all the spray teams;

(ii) Spray teams and drivers were trained on good spraying techniques and coached
on how to respond in cases of emergency;

(iii) Awareness among all residents of the targeted districts was raised; they were
further sensitised on what to do and what not to do before and after the
spraying to reduce exposure incidents;

(iv) Pregnancy testing for all female spray candidates was conducted, as was general
physical testing for all the spray teams;

(v) Surrounding health care facility staff were trained on emergency response to
acute pesticide poisoning;

(vi) All health facilities in the immediate area were supplied with the recommended
antidotes for pesticide poisoning;

(vii) Storage facilities were located in safe, secure and suitable sites;

(viii) Storage facilities were also checked to ensure that they were well built,
adequate and secure, to avoid pilferage, fire and any other negative incidents;

(iv) Spray pesticide residues, empty pesticide sachets, and unused pesticides were
properly disposed of or locked away securely following the recommendations of
WHO and the FAO on the safe use of pesticides.
Compliance with the measures described was monitored on a regular basis by NMCP, the Ministry of Agriculture, and the Environmental Protection Agency (through the DHMT task group). The plan ensured that the measures required to mitigate the undesirable impact of the IRS pilot programme on the environmental were maintained at all times.

(d) Household selection

All the structures and households in the selected areas of the four districts were targeted for the spray operations with the objective of achieving at least 85% minimum coverage. The selection carried out ensured that all the structures used for sleeping or relaxing at night were treated during the spraying operations. Prior to the spraying, the communities were informed in advance about the operations using different communication and education methods so that they were well prepared.

**Supervision, monitoring and evaluation — Phase 1**

Monitoring and supervision of IRS project activities was crucial to achieving expected outcomes. In order to ensure reliable and credible data, a monitoring and supervision plan was developed and utilized by monitoring and supervision staff in all pilot districts. While the exercise was ongoing, data were collected on a daily basis, compiled and forwarded to the national IRS focal person for analysis. The quantity of chemical (sachets) supplied to operators, the quantity used and the balance were tracked on a daily basis. The use and care of spray equipment were routinely supervised, including the quantity of water used to mix the chemicals and spraying of structures. Data collection and reporting took place between 20 December 2010 and 4 February 2011. The spray operators, district supervisors, monitoring and evaluation officers, and national supervisors were all involved in the daily collection of data on all aspects of the pilot: insecticides used, number of rooms sprayed, number locked, number of rooms to which entry was denied, number of ITNs and LLINs found during the spraying exercise.

### 3.2 Phase 2: April 2012

The second phase of the IRS programme commenced in February 2012 with pre-planning meetings and the participation of all partners (see Annex 5 for a detailed work plan). The main objectives of this phase were:

(a) To increase overall coverage of the IRS programme
(b) To secure community acceptance of the IRS programme
(c) To measure the impact of IRS on malaria prevalence levels in the four districts
(d) To monitor the reduction of vector densities and the response of mosquitoes to the insecticides used for IRS.

**Process of implementation**

Following a successful planning exercise, the recruitment and training of spray operators was carried out in April, culminating in the actual spraying from 21 May 2012 to 13 June
2012. At the end of the spray operations, debriefing meetings at both national and district levels were held to reconcile overall progress made. During the meetings, levels of IRS coverage and any challenges encountered in each district were discussed. Outstanding gaps requiring house structure treatment were identified in Kono District. These were subsequently treated with left-over insecticide from Western Area Rural District.

Supervision, monitoring and evaluation were carried out at both local district and national levels. Supervision at national level involved a checklist as outlined in Annex 7. Any areas found to be under-performing were appropriately addressed and corrective measures taken. The Roll Back Malaria (RBM) monitoring and evaluation framework was adopted with a few modifications. This consisted of the use of a quality checklist (see Annex 6), inventory control, and vector bionomic studies, while the evaluation focused on post-spray task group feedback meetings, community focus group discussions, vector bionomic studies and analysis and comparison of pre- and post-spray epidemiological data. To assist and enhance the quality of evaluation, eight sentinel sites were identified and established (two in each district – one in an IRS area and the other in a non-IRS area). The sentinel sites were also used as field insectary sites for breeding of bioassay mosquitoes and pre-sporezoite analysis. Bioassays were started one month after the spray, and every month thereafter for the duration of the study.

Assessment of malaria cases was conducted in health facilities from 21 chiefdoms within the four districts as described above (section 3.1.2 (a)), while entomological surveillance (vector densities and bioassays) was carried out twice a month using two standard mosquito collection methods as described in section 3.1.2 (b). Each study arm (district) had three to four sentinel villages or chiefdoms for entomological monitoring. In each sentinel village or chiefdom, simple random sampling was used to select the houses or sites for adult mosquito collections. Sentinel villages were selected based on a combination of geographic location, accessibility and proximity to health facilities. For the bioassays, the residual effects of insecticides were monitored through mosquito bioassays conducted monthly where mosquitoes were exposed to sprayed walls using WHO cones and their mortality observed after holding them for 24 hours.
4. RESULTS

4.1 Results of Phase 1

Launch of operations

The official launch of the indoor residual spraying operations was conducted at a special function in Western Area Rural on 16 December 2010 by the Minister of State in the Vice President’s Office, Dr Komba Kono. The launch was witnessed by the WHO Country Representative for Sierra Leone, Dr Wondimagegnehu Alemu and senior government and community officials. The launch was an important milestone in the history of malaria control in Sierra Leone (Figures 2 and 3).

Training of trainers

An intensive seven-day training-of-trainers (TOT) workshop focusing on malaria vectors and transmission, IRS operational strategy, insecticides, spraying techniques and environmental compliance was conducted in Makeni, Bombali District (Figure 4). A total of 17 participants (14 men and 3 women) from all the four pilot districts were trained. Since the TOTs did not
have knowledge and experience in IRS operations, intensive training was provided to them to adequately acquire the essential knowledge and ability to train team leaders and spray operators to the required standard for the operations.

**District level training of spray operators**  
Training for the spray operators was completed on 16 December 2010. One day was devoted to a mock spraying exercise with insecticide prior to actual full-scale operations commencing on 17 December 2010. A total of 130 recruited spray operators from all the four districts were trained (Figure 5). This workshop incorporated theory and practice modules, and concluded with a test on both theory and practice. Based on test results, spray operators (trainees) were categorized as spray operators or washers. Spray operators were also trained on the use of data collection tools. They were further trained on the “progressive rinse” best practice to ensure proper decontamination of pumps while minimizing environmental contamination with pesticide residues (Annex 4). Water used to rinse out sprayers at the end of each day was re-used at the beginning of the next day’s work both to save water and reduce the potential for pollution from contaminated rinse-water.

![Image](image_url)

**Coverage rate**  
Overall, the IRS programme operations were successful, with average coverage of 96.5%. Coverage in Bombali District was 94% and 99.5% in Kono District. Figure 6 shows the percentage coverage of IRS in the four districts. The IRS approach was acceptable in all chiefdoms and zones as evidenced by local community cooperation and support. Community participation was evidenced by the way they prepared their homes for spraying without delay, and by their embrace of safety measures as recommended. An average of 26 days (range: 24–28 days) were spent to cover the IRS programme in all the four districts.
Figure 6: Proportion of structures covered in target areas by indoor residual spraying in 2010

Table 1 summarises the baseline malaria prevalence rates in children, adolescents, and adults in health facilities in all the four districts in 2009 and 2010. Overall, the malaria prevalence rate averaged 50% (range: 47.3%–53%) in 2009; it was 53.4% (range: 40.7%–72.8%) in 2010. However, the highest malaria prevalence (72%) was recorded in 2010 in Bombali District. Children under the age of 5 years were the most affected by malaria in all the four districts compared to adolescents and adults.

Table 1: Summary of baseline malaria prevalence rates among children, adolescents and adults in the four districts in 2009 and 2010

<table>
<thead>
<tr>
<th>District</th>
<th>Year</th>
<th>Level</th>
<th>Total Number of malaria cases</th>
<th>Test positive rate (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td></td>
<td>Children (0-4 years)</td>
<td>Adolescents (5-14 years)</td>
</tr>
<tr>
<td>Western Area</td>
<td>2009</td>
<td>Bo</td>
<td>Health facility</td>
<td>86 211</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Community</td>
<td>36 575</td>
<td>15 703</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Subtotal</td>
<td>122 786</td>
<td>54 009</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Bombali</td>
<td>Health facility</td>
<td>31 954</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Community</td>
<td>34 202</td>
<td>20 754</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Subtotal</td>
<td>66 156</td>
<td>32 161</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Kono</td>
<td>Health facility</td>
<td>18 433</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Community</td>
<td>49 195</td>
<td>24 092</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Subtotal</td>
<td>62 628</td>
<td>30 939</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Western Area</td>
<td>Health facility</td>
<td>55 606</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Community</td>
<td>8539</td>
<td>6286</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Subtotal</td>
<td>64 145</td>
<td>24 961</td>
</tr>
<tr>
<td></td>
<td>2010</td>
<td>Bo</td>
<td>Health facility</td>
<td>121 423</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Community</td>
<td>41 809</td>
<td>20 966</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Sub-total</td>
<td>163 232</td>
<td>61 724</td>
</tr>
</tbody>
</table>
Six different insecticides recommended by WHOPES were evaluated for potential use in IRS operations. Results of the wall bioassays indicated that all the six different insecticides tested were still effective for use in the selected districts (Table 2). However, mortalities due to DDT showed some indication of emerging insecticide resistance as it was taking longer than 24 hrs to effect 100% mortality. The performance of permethrin is also slow compared to the other pyrethroids both at knockdown and 24-hour mortality. Pyrethroids, deltamethrin and lambda cyhalothrin were recommended as the first choice insecticides for
the pilot IRS programme. Even though both bendiocarb and malathion performed better, their documented residual periods were perceived to be short considering that the transmission season within the pilot programme districts was known to last almost nine months. Based on these results, lambda cyhalothrin was therefore recommended as the insecticide of choice for IRS.

4.2 Results of Phase 2

**IRS coverage**

A pyrethroid (lambda cyhalothrin) insecticide was sprayed in all the four pilot districts during the 2012 phase 2 IRS implementation plan, achieving a coverage rate of over 97% (Fig. 7) of about 76,000 targeted households, and protecting approximately 380,000 people in the spray round (Table 3). As in Phase 1, Bombali District recorded a low IRS coverage of 95% compared to the other districts. A total of 15,508 sachets (approx. 9,700 kg) of lambda cyhalothrin insecticide formulations were used for IRS in the four districts (Table 3). IRS has proven to be successful, as evidenced by persistently low vector populations since 2010, as well as drastic reductions in malaria cases in health facilities.

![Figure 7: Percentage of house structures covered by IRS in 2012](image-url)
Table 3: Summary of households or rooms sprayed, sachets used and percentage coverage in 2012

<table>
<thead>
<tr>
<th>District</th>
<th>Total rooms targeted</th>
<th>Coverage rate (%)</th>
<th>House hold population</th>
<th>Population protected</th>
<th>People using nets</th>
<th>No. of sachets used</th>
</tr>
</thead>
<tbody>
<tr>
<td>Western Area Rural</td>
<td>21 936</td>
<td>98.0</td>
<td>35 029</td>
<td>130 592</td>
<td>24 850</td>
<td>4779</td>
</tr>
<tr>
<td>Bombali</td>
<td>24 810</td>
<td>95.0</td>
<td>78 603</td>
<td>97078</td>
<td>69 059</td>
<td>5304</td>
</tr>
<tr>
<td>Kono</td>
<td>11 312</td>
<td>99.5</td>
<td>30 509</td>
<td>101 495</td>
<td>22 027</td>
<td>1817</td>
</tr>
<tr>
<td>Bo</td>
<td>18 335</td>
<td>99.0</td>
<td>45 004</td>
<td>51 661</td>
<td>38 229</td>
<td>3608</td>
</tr>
<tr>
<td>Overall</td>
<td>76 393</td>
<td>97.8</td>
<td>189 145</td>
<td>380 826</td>
<td>154 165</td>
<td>15 508</td>
</tr>
</tbody>
</table>

Community perception, acceptance and compliance
Several focus group meetings were held in all four IRS pilot districts. Community perception and acceptance was very positive and encouraging. Key observations noted were that the stakeholder task groups considered the IRS concept user-friendly and cost effective while the target communities considered the project as an umbrella malaria prevention strategy that covered all age groups unlike the LLINs. Furthermore, the communities were very pleased with the gender balance in the selection and recruitment of spray operators. Community awareness programmes were well accepted and appreciated, which resulted in better coverage in all districts. In addition to providing the expected benefit of malaria vector control, communities appreciated the broad-spectrum effect of the insecticide on other insects such as cockroaches. Finally, no incidents of chemical reaction to or poisoning involving operators were reported.

Although the communities participated and overwhelmingly accepted the IRS programme, there were concerns on the lack of complete coverage in the districts resulting from a few persons that were non-compliant. These issues were addressed within the chiefdoms and districts and resolved. Communities noted a significant reduction in disease burden and noted the economic benefits, such as more time for other social and economic activities.

Assessment of uncomplicated malaria at health facilities
Epidemiological data were collected from health facilities within the chiefdoms of each of the four districts. Health facility clinical malaria cases from 2009 to 2012 are shown in Figure 8. Generally, there was no significant decline in the number of malaria cases during the IRS programme from 2010 to 2012, indicating that a single round of spraying may not show an immediate reduction in malaria morbidity.
Summary data on the number of malaria cases for 2009 to 2012 is presented in Table 4. Data on malaria morbidity was passively collected from health facilities in 21 chiefdoms in the four districts. Malaria cases varied considerably within small geographical areas (chiefdoms) and districts, and within age-groups.

Table 4: Summary of morbidity data collected from IRS pilot districts, 2009–2012

<table>
<thead>
<tr>
<th>District</th>
<th>Chiefdom</th>
<th>0-4 years</th>
<th>5-14 years</th>
<th>&gt;15 years</th>
</tr>
</thead>
<tbody>
<tr>
<td>Western Area</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td>2012</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td>2012</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td>2012</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>
Figure 9: Mean number of malaria cases in the four districts, 2009–2012

A. Mean number of malaria cases; 0-4 year old

B. Mean number of malaria cases; 5-14 years

C. Mean number of malaria cases; >15 years
In children under five (0-4 years old), more malaria was recorded in all the districts (Fig 9A) compared to adolescents (5-14 years; Fig 9B) and adults (>15 years; Fig 9C). Generally, malaria morbidity patterns were similar in the four districts among children, adolescents and adults. However, more malaria cases were observed in Bombali from 2010 to 2012 in under-five-year olds compared to the other three districts (except the Western Area Rural) in 2010.

Figure 10: Mean monthly malaria morbidity in the four districts, 2011–2012

Figure 10 shows monthly malaria morbidity data for the four districts during the period 2011 to 2012. Generally, there was no change in the mean number of malaria cases both spatially and temporally over the two-year period. However, in the Western Area Rural, there was a slight decrease in the number of malaria cases. No epidemiological data was collected from Kono District in 2011. Monthly trend analyses show that the malaria morbidity pattern peaks in April to August, with a decline beginning in September. However, this is not consistent in the four districts. There were marked differences in patterns of malaria morbidity within the chiefdoms.

Entomological surveillance

Table 5 gives a summary of the 2012 susceptibility tests carried out following the introduction of IRS with lambda cyhalothrin in June 2012. Insecticide susceptibility tests carried out in the four districts showed that there was emergence of insecticide resistance in all the four districts (see Table 5 and Fig. 11). There is need to confirm this resistance as it ranged from 91.5% to 96.9%. Results on the monthly wall bioassays showed that lambda cyhalothrin was still effective up to four months after the spray. However, these bioassays
were carried out for only the first four months; funds ran out before the final two months of the programme were completed.

Table 5: Summary of 2012 susceptibility studies on Anopheles gambiae s.l. against insecticides in four districts in Sierra Leone

<table>
<thead>
<tr>
<th>District</th>
<th>No. exposed</th>
<th>30min</th>
<th>60min</th>
<th>120min</th>
<th>24hrs</th>
<th>KDt50 (95%CI)</th>
<th>KDt95 (95%CI)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Bo</td>
<td>164</td>
<td>110 (67.07)</td>
<td>137 (83.54)</td>
<td>137 (83.54)</td>
<td>157 (95.73)</td>
<td>24.73 (10.79-38.68)</td>
<td>61.06 (46.32-75.80)</td>
</tr>
<tr>
<td>Bombali</td>
<td>163</td>
<td>122 (74.85)</td>
<td>147 (90.18)</td>
<td>152 (93.25)</td>
<td>158 (96.93)</td>
<td>29.68 (11.65-47.70)</td>
<td>49.84 (38.80-60.88)</td>
</tr>
<tr>
<td>Kono</td>
<td>131</td>
<td>66 (50.38)</td>
<td>90 (68.70)</td>
<td>110 (83.97)</td>
<td>125 (95.42)</td>
<td>39.44 (28.23-50.65)</td>
<td>58.43 (36.86-79.99)</td>
</tr>
<tr>
<td>WAR</td>
<td>154</td>
<td>91 (59.09)</td>
<td>114 (74.03)</td>
<td>134 (87.01)</td>
<td>141 (91.56)</td>
<td>41.97 (31.70-52.24)</td>
<td>49.51 (34.35-64.67)</td>
</tr>
<tr>
<td>Total</td>
<td>612</td>
<td>389 (63.56)</td>
<td>488 (79.74)</td>
<td>533 (87.09)</td>
<td>581 (94.93)</td>
<td>38.21 (32.20-44.22)</td>
<td>50.99 (44.26-57.72)</td>
</tr>
</tbody>
</table>

Table 6: Summary of mosquito collections and sporozoite counts in IRS and non-IRS sites, July–August 2012

<table>
<thead>
<tr>
<th>District</th>
<th>Treatment</th>
<th>Sentinel sites/Chiefdom</th>
<th>Sampling method</th>
<th>#An. gambiae s.l. collected</th>
<th>#An. funestus collected</th>
<th># with Sporozoite Gambiae s.l. collected</th>
<th># An. funestus collected</th>
<th># with Sporozoite</th>
</tr>
</thead>
<tbody>
<tr>
<td>Kono</td>
<td>IRS</td>
<td>Yengema</td>
<td>PSC</td>
<td>19</td>
<td>0</td>
<td>6</td>
<td>2</td>
<td>0</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>Exit traps</td>
<td>23</td>
<td>0</td>
<td>4</td>
<td>14</td>
<td>0</td>
</tr>
<tr>
<td>Bo</td>
<td>IRS</td>
<td>Gbaima</td>
<td>PSC</td>
<td>8</td>
<td>0</td>
<td>2</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>Exit traps</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>Western Area</td>
<td></td>
<td>Waterloo</td>
<td>PSC</td>
<td>5</td>
<td>0</td>
<td>15</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>Exit traps</td>
<td>0</td>
<td>1</td>
<td>3</td>
<td>2</td>
<td>0</td>
</tr>
<tr>
<td>Bombali</td>
<td>IRS</td>
<td>Binkolo</td>
<td>PSC</td>
<td>0</td>
<td>0</td>
<td>4</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>Exit traps</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>Total</td>
<td>IRS</td>
<td></td>
<td></td>
<td>55</td>
<td>1</td>
<td>34</td>
<td>18</td>
<td></td>
</tr>
<tr>
<td>Kono</td>
<td>Non-IRS</td>
<td>Sewafe</td>
<td>PSC</td>
<td>3</td>
<td>5</td>
<td>0</td>
<td>13</td>
<td>13</td>
</tr>
<tr>
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<td>Exit traps</td>
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<td>0</td>
<td>0</td>
</tr>
<tr>
<td>Bo</td>
<td>Non-IRS</td>
<td>Koribondo</td>
<td>PSC</td>
<td>1</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
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<td></td>
<td></td>
<td>Exit traps</td>
<td>0</td>
<td>0</td>
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<td>0</td>
<td>0</td>
</tr>
<tr>
<td>Western Area</td>
<td></td>
<td>George Brooke</td>
<td>PSC</td>
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<td>0</td>
<td>4</td>
<td>4</td>
<td>0</td>
</tr>
<tr>
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<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>Bombali</td>
<td>Non-IRS</td>
<td>Kamabai</td>
<td>PSC</td>
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<td>0</td>
<td>0</td>
<td>0</td>
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<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>Total</td>
<td>Non-IRS</td>
<td></td>
<td></td>
<td>11</td>
<td>8</td>
<td>17</td>
<td>3</td>
<td>0</td>
</tr>
</tbody>
</table>

Cone test bioassays to determine spray quality were performed on lambda-cyhalothrin-sprayed walls and it was noted that the bioassay data showed 99% mortality for all mosquitoes exposed to the sprayed walls at all sentinel sites in the four districts after 24 hours (Fig. 11). As expected, very low mortalities were recorded in the non-IRS sentinel sites. Four months after spraying, it was noted that the lambda cyhalothrin insecticide continued to be effective on the sprayed walls. However, the number of An. gambiae s.l. and An. funestus collected over two months in IRS sentinel sites was almost similar to those recorded in non-IRS sentinel sites, indicating that not much change in vector density was observed two month after the spray period (Table 6).
Table 6 summarizes mosquito collections and sporozoite counts in IRS and non-IRS sites in the four districts two months after IRS implementation. Very few mosquitoes were collected using the PSC and EWT methods. A total of 89 *An. gambiae s.l.* and 19 *An. funestus* were collected over two months in the IRS sentinel sites, while only 11 *An. gambiae s.l.* and 25 *An. funestus* were collected from the non-IRS sentinel sites. However, of the few collected no sporozoites were detected. The study also confirmed the existence of the two main vector species: *An. gambiae s.l.* and *An. funestus*. A higher proportion of malaria vectors (43.5%) exited the sprayed houses compared to only 23.1% in the non-IRS areas, indicating that lambda cyhalothrin had some exit-repellency effect. However, six months after spraying, this exit-repellency effect decreased to almost zero as none of the malaria vectors were captured in exit traps (Table 7).

**Table 7: Mosquitoes collected for six months after the IRS programme, January 2013**

<table>
<thead>
<tr>
<th>District</th>
<th>Treatment</th>
<th>Sentinel sites/Chiefdom</th>
<th>Sampling method</th>
<th># <em>An. gambiae s.l.</em> collected</th>
<th># with Sporozoite</th>
</tr>
</thead>
<tbody>
<tr>
<td>Kono</td>
<td>IRS</td>
<td>Yengema</td>
<td>PSC</td>
<td>13</td>
<td>0</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>Exit traps</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>Bo</td>
<td>IRS</td>
<td>Gbaima</td>
<td>PSC</td>
<td>8</td>
<td>0</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>Exit traps</td>
<td>1</td>
<td>0</td>
</tr>
<tr>
<td>Western Area</td>
<td>IRS</td>
<td>Waterloo</td>
<td>PSC</td>
<td>8</td>
<td>0</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>Exit traps</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>Bombali</td>
<td>IRS</td>
<td>Binkolo</td>
<td>PSC</td>
<td>8</td>
<td>0</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>Exit traps</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Total</td>
<td></td>
<td>38</td>
<td>0</td>
</tr>
<tr>
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<td>PSC</td>
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<td>Exit traps</td>
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<td>Exit traps</td>
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<td>PSC</td>
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<td>Exit traps</td>
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<td>0</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Total</td>
<td></td>
<td>26</td>
<td>0</td>
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</tbody>
</table>
In January 2013, six months after the start of the IRS programme in 2012, a total of 64 *Anopheles gambiae s.l.* were collected by PSC and using exit window traps in both IRS and non-IRS sentinel sites (Table 7). Of these, 38 *An. gambiae s.l.* were collected by PSC in IRS sentinel sites and none from the exit window traps. Likewise, 26 *An. gambiae s.l.* were collected from non-IRS sentinel sites by PSC, but none from the exit window traps. No *An. funestus* was captured in either IRS or non-IRS sentinel sites, indicating that the insecticide used was very effective on *An. funestus*. None of the mosquitoes dissected were found positive for sporozoites.

### 5. OUTCOME OF THE IRS PILOT PROJECT

#### 5.1 Project evaluation

The RBM Monitoring and evaluation framework was adopted for evaluating the project. Figure 12 illustrates the strategy. The evaluation highlights and discusses the achievements of the pilot project. Both the outputs, outcomes and impacts achieved within the project implementation have been considered.

#### Figure 12: Malaria monitoring and evaluation framework

- Assessment and planning
  - Situation analysis
  - Response analysis
  - Stakeholder needs
  - Resource/logistics analysis
  - Collaboration
- Input
  - Funds
  - Staff
  - Materials
  - Facilities
  - Supplies
- Process
  - Trainings
  - Services
  - Education
  - Treatments
  - Interventions
  - Services delivered
    - No. of LLINs distributed
    - No. of HHs sprayed
    - No. of IPTs delivered
    - No. RDTs/slides taken
    - No. of antimalarial drugs delivered
- Output
  - Coverage
  - HH LLIN possession (%)
  - LLIN use (%)
  - IRS coverage
  - U5 treatment (%)
- Outcomes (Intermediate effects)
  - Malaria incidence/prevention
  - USMR
  - Malaria morbidity/mortality
  - Economic impact
- Impact (Long-term effects)

#### 5.2 General project outputs and outcomes

Based on the RBM framework, the project achieved most of its expected objectives at both the output and outcome levels.

(a) A high proportion of houses (>97%) in target areas (chiefdoms) were sprayed.

(b) The two phases of IRS built the skills of national and district officers to plan, implement, and supervise IRS operations according to the required standards.
The project created sufficient IRS infrastructure, equipment and materials that should be used for scaling up the project and expanding it to other districts in the country.

Most importantly, a positive perception towards IRS among beneficiaries as well as national and district level implementers was established. This was evidenced by the appropriate community focus group evaluation meetings carried out in the four districts.

Evaluation of vector bionomic activities was conducted as expected, with only 67% of activities on bioassays and vector surveillance completed (only four bioassays instead of 6); results were the same for vector surveillance and sporozoite determination. These could not be fully implemented due to lack of appropriate and sufficient resources.

Exit window traps were set and monitored concurrently with the bioassay activities.

Bioassays carried out confirmed four months of residual effectiveness of the insecticide used.

Data collection at PHUs was carried out in the first month but protocols of data collection systems then changed due to the demands of other competing programmes needing funding (performance-based financing); the challenges of these changes were adequately addressed.

Post-spraying environmental compliance was adequately carried out. It addressed the areas of waste disposal, equipment use and storage, and storage and inventory evaluation at all stages of the operations. Finally, post-spray district debriefing was done, and future plans discussed and compiled. Community acceptance continued to be very positive and high, and demand for service was overwhelming.

Target population: Some inconsistencies were noted both in Phase 1 and in Phase 2 regarding the expected target population and the actual population treated. However, it was expected that some of these and other problem areas and inconsistencies would be addressed during the 2013 Malaria Indicator Surveys (MIS).

6. CONCLUSION

The success of IRS implementation was clearly demonstrated by the massive high coverage rate of sprayed structures. Although a major impact on the reduction in malaria cases and positivity rates was not clearly evident, a slight decline in morbidity was noted. Substantial impact was recorded on the reduction in vector density for both An. gambiae and An. funestus in January 2013, six months after spraying. During this period none of the mosquitoes were infected with Plasmodium species, indicating that no transmission was taking place. Mosquito bioassay results also showed that lambda cyhalothrin was still effective against anopheles mosquito vectors four months after spraying.
7. RECOMMENDATIONS

(a) The pilot IRS implementation programme was successful for malaria control and was fully embraced by the community as evidenced by the positive compliance, acceptance and favourable perceptions; it is therefore recommended that the programme should be expanded to other districts. This sentiment was also strongly expressed by the various stakeholders within the pilot districts.

(b) Capacity building has been identified as a major challenge in establishing a successful IRS programme. In particular, the dearth of human resources, infrastructure, and financial resources needs to be addressed. The IRS programme draws heavily on the knowledge and skills of persons (TOTs) in functions at national, district and chiefdom level. These need to be fully developed. Future operations will need to ensure strategic fund management for monitoring and evaluation activities to avoid challenges at the end of operations.

(c) Progress in capacity building needs monitoring and evaluation to measure change, and to identify the focus for further attention. Epidemiological and entomological assessment in the IRS programme also needs to be strengthened. A comprehensive framework for monitoring and evaluation should be developed in line with integrated vector management (IVM) approaches. This would ensure that some of the key operational activities that were not fully explored are continuously implemented and expanded. Such a framework should include an evaluation of complimentary strategies so as to fully understand the effect and impact of all interventions.

(d) It is also strongly recommended that scale-up activities be carried out in a timely manner so as to enhance the effectiveness of the IRS programme. This will have the added advantage of retaining skilled human resources before they are engaged in other activities. Timely scale-up of activities will also reduce the cost of training while improving the quality of services.
REFERENCES


ANNEX 1: SUMMARY OF INSECTICIDE SUSCEPTIBILITY TESTS

Annex 1 is a summary of the laboratory tests carried out to evaluate the susceptibility of five insecticides for potential use in the IRS pilot programme in four districts of Sierra Leone: Bendiocarb 1%, malathion 5%, DDT 4%, permethrin 0.25%, deltamethrin 0.05%, lamda cyhalothrin 0.05%.

David T Zinyengere, HEDEC Consulting; Paul Conteh, MOHS Sierra Leone; Manfred Morovia, MOHS Sierra Leone; Alfred George Gbla. 21 June 2010 to 9 July 2010.

1. Background

Sierra Leone intends to introduce indoor residual spraying against malaria vectors as part of the recently launched integrated vector management (IVM) strategy. The predominant vector-borne diseases in Sierra Leone, namely, malaria schistosomiasis, onchocerciasis and lymphatic filariasis account for the bulk of its disease burden. However in view of the fact that IRS was last carried out at national level before 1980, there was a need to establish the susceptibility of potential insecticides to utilize for the pilot programme.

2. Materials and Method

Batches of 20 non-blood-fed female anopheline mosquitoes, aged 2–3 days, were introduced into a holding tube (marked with a green dot) and held for one hour at 25 °C ± 2 °C to acclimatize. They were then transferred by gentle blowing into the exposure tube (marked with a red dot), and the kit was held vertically for one hour under subdued light. At the end of the exposure time, mosquitoes were gently blown back into the holding tube, which was placed vertically in a dark place for 24 hours at 25 °C ± 2 °C. A piece of cotton wool soaked in sucrose solution was placed on a section of the wire mesh covering the top of the holding tube.

Knockdown mortality was recorded at 10, 30, 60, and 90 minutes. Dead mosquitoes were counted after 24 hours. A total of 60 mosquitoes (three replicates containing 20 mosquitoes each) were used for each test concentration for the four sites visited and for the control. Results were expressed as percentage mortality after 24 hours and corrected for any control mortality. Concentrations used were the maximum recommended by WHO.

It must be noted that the numbers selected were due to the constraints involved in appropriate laboratory facilities and breeding cages. Two laboratory set-ups were constructed at the Onchocerciasis Laboratories in Bombali District and the others at the WHO offices in Freetown. Mosquitoes used were field-reared from the collection of larvae from all the selected pilot districts. It must also be noted that relative humidity could not be measured as we lacked the relevant instruments.
3. Results and Conclusions

All the 6 different insecticides tested are still effective for use in the selected districts (see table below showing vector knockdown by insecticide and district). However, DDT seems to be taking longer than 24 hrs to effect a 100% mortality. The performance of permethrin is also slow compared to the other pyrethroids, both at knockdown and at mortality after 24 hours. The pyrethroids, deltamethrin and lambda cyhalothrin were recommended as the first choice insecticides for the pilot programme. Even though both bendiocarb and malathion performed better, their documented residual periods were perceived to be a negative factor considering that the transmission season within the pilot programme districts was known to last for almost nine months.

4. Classification of insecticide resistance levels

The indicator for vector susceptibility is the number of vectors killed on a discriminating dose of a given insecticide at a given time of exposure. According to WHO (2008), the standard criteria for determining resistance are as follows:

(i) <90% mortality of vectors after exposure: resistance confirmed;
(ii) 90-98% mortality of vectors after exposure: resistance needs to be confirmed with further testing;
(iii) 98-100% mortality of vectors after exposure: population fully susceptible.

If there is evidence of resistance (<90% mortality) carry out resistance mechanism investigations by molecular and biochemical methods.
Breakdown of vector knockdown by insecticide and district

<table>
<thead>
<tr>
<th>District</th>
<th>Insecticide</th>
<th>Total tested</th>
<th>10</th>
<th>30</th>
<th>60</th>
<th>90</th>
<th>% Mortality (24 Hr)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Bombali</td>
<td>DDT</td>
<td>60</td>
<td>61.7</td>
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<td></td>
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<td>93.3</td>
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<tr>
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<td>93.3</td>
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<td>86.7</td>
<td>93.3</td>
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</tbody>
</table>

Notes
Three replicates for each insecticide, each with 20 adults, were conducted for each of the four districts and the total added up as summarised in the tables. During the test period, temperatures ranged from 25 °C–29 °C despite the various measures taken to maintain them at average 25 °C–27 °C using wall mounted fans in the make-shift laboratory at WHO offices. Control mortalities averaged 1.1% (range 0–3.3%).
ANNEX 2: INSECTICIDE SELECTION CRITERIA

1. Pesticide selection:

The selection of a pesticide for use in IRS is the sole prerogative of the country. However, the process is guided by the following threshold criteria that must be met in making decisions on pesticides used in malaria vector control: that the pesticide must be approved by the World Health Organization pesticide evaluation scheme (WHOPES) and should be preferred based on their safety and environmental effects. In addition, the pesticide must:

(a) Be registered for use in the country for IRS by the ministry of agriculture, health or any other recognized entity;
(b) Be a pesticide accepted by the national malaria control programme (NMCP);
(c) Have a residual effect of more than 4 months (effective on the types of walls for a period longer than, or at least equal to, the average duration of the malaria transmission season in the area);
(d) Be vector-susceptible (for malaria vectors in the region) and demonstrate low toxicity to humans and external environment;
(e) Demonstrate low risk to the environment, livestock and agriculture in terms of toxicity;
(f) Be appropriate for use on the wall surfaces of the selected location;
(g) Be competitive in terms of cost (as against other pesticides).

Finally, the country must demonstrate its capacity to prevent pilferage.

The following insecticides were recommended in order of priority:

(a) Pyrethroids preferably deltamethrin or any other
(b) Organophosphates – only malathion was tested but research carried out elsewhere in the Region shows good residual results with chlorpyrifos methyl encapsulated formulation.
(c) Carbamates e.g., bendiocarb
(d) Organochlorines – DDT. It must be noted that DDT would be difficult to use at the moment considering that the country is not fully geared to meet the provisions of the Stockholm and Basel conventions.

As mentioned before all the above insecticides are WHOPES-approved, but they have different residual periods and more importantly the vectors in the area are susceptible to all the classes of pesticides as demonstrated by the entomological studies undertaken.
2. Selected insecticide

Pyrethroid and lamda cyhalothrin were selected for the IRS programme. The total amount of insecticide required was estimated to be 68 121 wettable powder (WP) sachets of 62.5g lamda cyhalothrin. The target application dosage was set at 0.03g per m². The approximate cost of the insecticide at US$ 2.31 per sachet was US$ 158 000.

3. Target area for the IRS programme

Four pilot districts were selected with a total targeted population of 851 513 and a total of 141 918 sprayable structures.

4. Target area for spraying

The approximate total area to be treated was estimated at 14 191 900 sq. metres (100 m² per structure). Each structure had an average of 3 to 5 sprayable rooms (for a total of 100 m² approx.).

The table below is a summary of the total population and number of structures in each of the four pilot districts used to calculate the amount of insecticide required for the IRS programme.

Population by district used to calculate the amount of insecticide required

<table>
<thead>
<tr>
<th>District</th>
<th>Total population</th>
<th>Total No. of structures under IRS</th>
<th>Av. No. of people per household</th>
<th>Proposed HH count</th>
</tr>
</thead>
<tbody>
<tr>
<td>Bo</td>
<td>206 107</td>
<td>34 351</td>
<td>5.2</td>
<td>39 636</td>
</tr>
<tr>
<td>Bombali</td>
<td>234 589</td>
<td>39 098</td>
<td>5.2</td>
<td>45 113</td>
</tr>
<tr>
<td>Kono</td>
<td>169 340</td>
<td>28 223</td>
<td>5.2</td>
<td>32 565</td>
</tr>
<tr>
<td>Western Rural</td>
<td>241 477</td>
<td>40 246</td>
<td>5.2</td>
<td>46 438</td>
</tr>
<tr>
<td>Total</td>
<td>851 513</td>
<td>141 918</td>
<td>20.8</td>
<td>163 752</td>
</tr>
</tbody>
</table>
# Annex 3: Operational Log Frame for Phase 1 (December 2010)

| Week/Activity                                                                 | 1 | 2 | 3 | 4 | 5 | 6 | 7 | 8 | 9 | 10 | 11 | 12 | 13 | 14 | 15 | 16 | 17 | 18 | 19 | 20 | 21 | 22 | 23 |
|--------------------------------------------------------------------------------|---|---|---|---|---|---|---|---|---|----|----|----|----|----|----|----|----|----|----|----|----|----|----|----|
| Administrative issues -WHO                                                      |   |   |   |   |   |   |   |   |   |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |
| Meeting with all stakeholders & review of activities                          |   |   |   |   |   |   |   |   |   |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |
| Analysis of objectives and strategies, situation analysis (environmental scan) |   |   |   |   |   |   |   |   |   |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |
| Stakeholders agree goals & objectives of programme                             |   |   |   |   |   |   |   |   |   |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |
| # Stakeholder analysis                                                         |   |   |   |   |   |   |   |   |   |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |
| # Strengths, weaknesses, opportunities and threats (SWOT)                      |   |   |   |   |   |   |   |   |   |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |
| # Driving and retraining forces                                                 |   |   |   |   |   |   |   |   |   |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |
| # Key success factors                                                          |   |   |   |   |   |   |   |   |   |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |
| Visit to proposed districts and evaluation capacity                           |   |   |   |   |   |   |   |   |   |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |
| Health facilities                                                              |   |   |   |   |   |   |   |   |   |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |
| Human resources                                                                |   |   |   |   |   |   |   |   |   |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |
| Equipment                                                                      |   |   |   |   |   |   |   |   |   |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |
| Research capacity                                                              |   |   |   |   |   |   |   |   |   |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |
| Target population                                                              |   |   |   |   |   |   |   |   |   |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |
| Evaluation of technical support capacity                                       |   |   |   |   |   |   |   |   |   |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |
| Vector surveillance and bioassays; susceptibility tests                        |   |   |   |   |   |   |   |   |   |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |
| Financial planning                                                             |   |   |   |   |   |   |   |   |   |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |
| Draft report consolidation and writing                                         |   |   |   |   |   |   |   |   |   |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |
| Procurement of insecticides and equipment                                      |   |   |   |   |   |   |   |   |   |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |
| Repair of infrastructure                                                       |   |   |   |   |   |   |   |   |   |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |
| Community mobilization and IEC finalization                                    |   |   |   |   |   |   |   |   |   |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |
| Recruitment of spray operators                                                 |   |   |   |   |   |   |   |   |   |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |
| Conduct of TOT                                                                  |   |   |   |   |   |   |   |   |   |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |
| Spray operator training                                                        |   |   |   |   |   |   |   |   |   |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |
| Spraying operation begin                                                        |   |   |   |   |   |   |   |   |   |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |
| Post-spray debriefing workshop                                                  |   |   |   |   |   |   |   |   |   |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |
| Monitoring and evaluation activities                                           |   |   |   |   |   |   |   |   |   |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |

*Implementation of indoor residual spraying to assess feasibility in Sierra Leone*
ANNEX 4: THE PROGRESSIVE RINSE METHOD

1. PREPARATION FOR PROGRESSIVE TRIPLE RINSE OF SPRAY EQUIPMENT

At the end of each day, spray pumps are washed following the Progressive triple rinse method. Drums are clearly marked as ‘Hazardous Waste’.

Water is used to thoroughly rinse drums #3, #5 and #7; the same water is used the next day for mixing the insecticide.

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(a) Place 7 open drums (200 litres) in a single line on a plastic sheet (or on the cement apron where an evaporation tank is being used) to contain any spillage.

(b) The first drum is empty, the second filled with clean water, the third drum empty, the fourth drum full, the fifth drum empty, the sixth drum full and the last drum empty.

(c) Where only three drums are used - the first is empty (to contain returned spray solution), the second filled with clean water, and the third empty; water from the second drum is used to wash spray equipment and wash water poured or sprayed into the third drum. This to be repeated three times.

(d) Enough buckets to be provided to dispense water.
2. Washing

(a) Spray operators returning from the field are to line up and follow the washing instruction steps, moving from one drum to the next.
(b) Supervisor to dispense clean water to spray operators and ensure the triple rinse wash regime is strictly followed.

3. Handling waste wash water the following day

(a) Supervisor to dispense from drum 1 (i.e., the solution returned from the previous day);
(b) This should be thoroughly mixed before dispensing;
(c) Spray cans to be filled to 10 litres and operator to use this, undiluted for the first spray fill;
(d) After all the solution from drum 1 has been dispensed, Supervisor to fill other spray cans with wash water from drums 3, 5 and 7;
(e) Operators to top these up in the field when mixing the first spray charge;
(f) In this way all the previous day’s solution and wash water will be re-cycled and not allowed to carry over;
(g) Should there be any carry over wash water, this should be consolidated into one or two drums and dispensed to spray operators the following day.
ANNEX 5: SENTINEL SITES — TERMS OF REFERENCE FOR IRS — PILOT DISTRICTS 2012

1. SELECTION CRITERIA FOR SENTINEL SITES

The following should be considered in establishing a sentinel site:

(a) There should be at least two sentinel sites per pilot district;
(b) One of these should be in a non-IRS chiefdom;
(c) They should be preferably at a PHU or in close proximity to one;
(d) There should be communication facilities available:
   (i) Internet service;
   (ii) Telephone;
   (iii) Easy access.
(e) Laboratory facilities should be available or within easy access, considering existing infrastructure;
(f) Morbidity and mortality data, including incidence statistics, should be available;
(g) There should be representative population age groups;
(h) Human resources should be available;
(i) Personnel should be sober and proactive.

2. ACTIVITIES TO BE CARRIED

2.1 PHU and district malaria focal person

(a) All suspected patients should be diagnosed with RDTs and only positives should be treated in the pilot districts;
(b) Data should be submitted monthly to the national malaria focal person or programme officer;
(c) District malaria focal persons should collect data using Form P1 and attach same weekly and monthly.

2.2 Field officers and vector control technicians

(a) Establish and sustain field insectaries;
(b) Maintain laboratory-reared vector colonies for bioassay exposures.
(c) Set up exit traps at relevant points in sentinel sites — with equal representation in treated and non-treated areas; carry out species identification and sporozoite evaluation twice a month.
(d) Carry out PSC, and subsequently identify vector species and carry out sporozoite evaluation twice a month; determine if this is by dissection or ELISA; if dissection,
determine if there is adequate capacity for that; also decide if it is really necessary to do so twice a month, particularly in high transmission areas; once a month may seem adequate and normal.

(e) Carry out cone bioassays once a month and report according to schedule.

2.3 NMCP — national malaria focal officer

(a) Monitor the reporting system;
(b) Compile all data and share with WHO accordingly;
(c) Provide and set up laboratory facilities for microscopic examinations;
(d) Provide logistics for vector bionomic studies.

3. OPERATIONAL PROCEDURES

3.1 Data collection

(a) Data collection, compilation and submission to be done at the relevant times;
(b) PHU or district malaria focal person to use Form IRS P1;
(c) Field officers and vector control technicians to use Forms IRS 5a, IRS 5b, IRS 6, IRS B1;
(d) National malaria focal person to provide detailed report and summary.

3.2 Insectaries

Adequate field insectaries should be established at all sentinel sites in each district. These should have the following:

(a) 3 Breeding cages — one large (1mx1mx1m) and 2 medium (0.5mx0.5mx0.5m);
(b) 2 Mosquito transportation jars or cages (approximately 3–5 litre volume);
(c) 3 medium size breeding dishes for larvae (from the field);
(d) Cotton wool;
(e) Surgical gloves;
(f) Room thermometer;
(g) Medium size chicken;
(h) Larvae breeding bowls.

3.3 Breeding of bioassay mosquitoes

(a) Local vector species should be used for bioassays and these should be established through sentinel site insectaries;
(b) Larvae should be collected from local breeding areas and kept in jars within the breeding cages until they emerge;
(c) A minimum of 130 adult female anopheline mosquitoes are required per bioassay exposure exercise;
(d) Prior to conducting the bioassays species identification should be carried out to establish the species within the sentinel site area;
(e) Emerging mosquitoes should be fed using live chickens or guinea pigs.

3.4 Bioassays

Bioassays must be carried out to:

(a) assess the potency of the insecticide deposit for adult mosquitoes at monthly intervals after indoor residual application;
(b) define the decline in the toxic effect of the deposit due to ageing, absorption or other factors;
(c) ascertain whether or not the spraying exercise was carried out satisfactorily.

However these bioassays should be complemented by other methods to effectively evaluate the impact of the vector control interventions in the pilot districts.

Test guidelines and limitations

Tests should commence a few days (5-7 days) after the spraying operations have finished. These tests must be conducted in both the sprayed and selected unsprayed areas (control). Local female anopheline vector species must be used for the bio-assays.

In sprayed areas the objective is to establish the potency of the insecticide deposits, quality of the spraying operation and the duration of the insecticide on the sprayed surfaces. The non-sprayed areas are evaluated for comparison purposes to provide passive baseline data in an endeavour to show the impact of the IRS intervention strategy. Bioassays should be carried out in three different households within each of the sentinel areas selected. Household selection should be done following an agreed, unbiased random selection method. The method selected must be applied to all sentinel sites. Tenants of households selected should be well educated on the importance of these tests. The conditions they must adhere to should be carefully explained to them prior to IRS operations.

Bioassay kits

Each sentinel site will require the following for its bioassay test kit;

(a) 9 WHO bioassay exposure cones;
(b) 4 hard glass aspirator tubes – each with at least 60 cm of flexible rubber or plastic tubing;
(c) 2 rolls of adhesive plastic sponge tape;
(d) 1 box of upholstery tacks with large heads;
(e) 2 rolls of adhesive plaster;
(f) Cotton wool;
(g) 14 transparent plastic holding jars or tumblers;
(h) 2 mosquito transporting jars (approx. 3–5 litre in volume).

Bioassay procedure
(a) The exposure bioassay cone or chamber is fastened to the selected spot on the surface to be tested, using the upholstery tack or any other appropriate harness that will hold the cone or chamber tight against the surface. Care should be taken not to slide the cone or chamber while it is being attached or removed. Allowing this to happen could compromise the tests since this has the effect of wiping out some of the insecticide deposits on the surfaces to be tested.

(b) At least 10 but not more than 15 mosquitoes are collected with a straight loading tube and introduced into the cone or chamber by blowing gently; great care should be taken to avoid the end of the tube touching the test surface. The cage or transportation jar containing the test insects must not be taken inside a house that has been sprayed with insecticide but should be left outside the house on an insecticide-free surface in a shade. Avoid contamination of the transportation jars at all costs.

(c) The cone or chamber must be left undisturbed for a standard exposure time of 30 minutes. If more time is needed, this must be given to all areas for appropriate comparison. But exposure time must not be more than one hour.

(d) At the end of the exposure period the mosquitoes are collected carefully using the bent transfer tube; without touching the treated test surface area immediately transfer the mosquitoes into the holding jars.

(e) The room temperature and relative humidity should be recorded at the beginning and at the end of each day of testing, and at hourly intervals during the work period.

(f) The recovery (holding) jars are kept for 24 hours in a secluded, shaded place, where the temperature does not exceed 30 °C if feasible. Maximum and minimum temperatures during the recovery should be recorded. The humidity must be kept high by use of damp towelling where necessary.

(g) The exposure cones or chambers and transfer tubes are carefully washed in detergent after each use, rinsed and allowed to drain dry.

Results and interpretation
(a) Observations should be carried out at intervals of 30 minutes, one hour, two hours, and 24 hours. At each observation time point the number of live and dead mosquitoes must be recorded on the appropriate form. After 24 hours the dead and live mosquitoes should be counted. Observed mortalities (%) must be recorded for each individual test. Where control mortalities exceed 20% the series of tests should be considered unsatisfactory and repeated if possible.
(b) Where control mortality is between 5% and 20% the average observed mortality should be corrected using Abbott’s formula:

\[
\frac{\text{% test mortality} - \text{% control mortality} \times 100}{100 - \text{% control mortality}}
\]

(c) Immediately after the average value, the lowest and highest values should be written in parenthesis, thus “68% (20%–90%)”, to indicate the range of variation in the result. Please note that a wide differential in mortality rates from one point to another may reflect either the unevenness of spraying or a differential in the rate of loss of potency, due to domestic fires, the microclimate or other localized variables. Even after considering all these variables there could still be inherent differences in susceptibility of the individual mosquitoes used in the tests.

4. OTHER EVALUATION TOOLS TO BE USED

4.1 Exit light window traps

Once a month exit light traps should be fixed at strategic points in all the bioassay selected households. Mosquitoes captured should be collected after 12 hours between 8 a.m. and 9 a.m. The number of mosquitoes captured should be counted and identified. All female anopheline mosquitoes found should be analysed for sporozoites as per the procedures outlined below.

4.2 Pyrethrum aerosol spray collections

4.3 A white sheet should be placed on the floor and the room sealed. An application of pyrethrum spray should be carried out and knocked down mosquitoes collected from the sheet 15–30 minutes after the application. The number collected should also be counted and identified. All female anopheline mosquitoes found should be analysed for sporozoites as per the procedures outlined below.

5. VECTOR ANALYSIS — PROCEDURES

5.1 Materials

Equipment
(a) Screened pint cups for holding mosquitoes
(b) Paper towels
(c) Filter paper
(d) Petri dishes of different sizes
(e) 1.8 mL, 1.5 mL, and 0.5 mL plastic tubes with snap-on caps
(f) Insulated cooler for transporting mosquitoes
(g) Ice packs
(h) Color-indicator desiccant
(i) Glass wool
(j) Dissecting and compound microscopes
(k) Field and laboratory processing data forms
(l) Glass cover slips
(m) Aspirators for handling mosquitoes
(n) Plastic disposable beakers of different sizes
(o) No. 5 forceps
(p) Hand-held motor with polypropylene pellet pestle
(q) 1.5 mL grinding tube
(r) Freezer and refrigerator
(s) Micropipettes of different sizes, ranging from 10 μL to 1 mL
(t) Vacuum dryer or a drying oven
(u) Thermocycler
(v) Agarose gel and gel apparatus
(w) An ultraviolet light source.

5.2. Reagents

(a) Carnoy’s solution: 3 parts absolute ethanol: 1 part glacial acetic acid;
(b) Physiological saline, phosphate-buffered saline (PBS), or M-199 media for dissecting mosquitoes;
(c) Mercurochrome solution;
(d) DNA extraction buffer (DEB): 0.5% SDS, 0.2 M NaCl, 25 m methylene diamine tetraacetic acid (EDTA), and 10 n M Tris-HCl, pH 8.0. Prepare fresh. To prepare 10 mL, combine 50 mg of sodium dodecyl sulfate (SDS), 0.4 mL of 5 M NaCl, 0.5 mL of 0.5 M EDTA, and mL of 1.0 M Tris-HCl, pH 8.0. Bring the total volume to 10 mL using ddH2O stored in an incubator at 37°C, and heat to 65°C for 10 min. Swirl until mixed thoroughly;
(e) RNase A and proteinase K (20 μg/mL);
(f) Phenol and chloroform;
(g) 70% Ethanol;
(h) ddH2O;
(i) Primers for mosquito species diagnosis;
(j) 1 M MgCl2;
(k) Taq polymerase;
(l) DNA markers such as 100 bp and/or pUC18;
(m) Ethidium bromide.
5.3 Methods

Handling mosquitoes in the field

(a) Live-caught mosquitoes from exit window trap catches are held alive in screened holding cups for observation over 24 hours. If they are to be transported to the field laboratory within hours of collection, then they should be held either indoors or in the shade, with minimal handling. If they are to be held in the field more than two hours, then precautions such as placing water-soaked paper towels over the cups need to be taken to ensure that the mosquitoes do not become too dry or too hot.

(b) Freshly killed mosquitoes from any type of collection method require special handling methods, and several options are available. Mosquitoes from pyrethrum spray collections can be held on filter paper in petri dishes containing water moistened cotton; this will allow the mosquitoes to be held for 3–6 hours before further processing either in the field or field laboratory. Alternatively, mosquitoes can be placed in labeled tubes and held in an insulated cooler containing ice packs; this is a standard method of transport when the field laboratory is within about two hours' drive from the field site.

(c) When it is not possible to transport mosquitoes from field sites to the laboratory on the same day, it is necessary to take more elaborate steps for mosquito processing. Sometimes it is feasible to identify and process mosquitoes directly in the field, by setting up a field laboratory containing microscopes and other essential supplies and equipment. Some investigators prefer to dry mosquitoes by placing individual specimens in individual 0.5 mL labeled tubes containing desiccant granules covered by glass wool.

(d) Freshly collected mosquitoes can also be preserved in Carnoy's solution. This is quite effective for a variety of purposes:
   (i) Mosquitoes from the same collections can be pooled in individual labeled tubes;
   (ii) Mosquito samples in Carnoy’s solution can be held at room temperature for 24 hours or more, after which they can either be refrigerated for several days or frozen indefinitely;
   (iii) Mosquitoes held in Carnoy's solution can be processed by standard taxonomic identification, cytogenetics, polymerase chain reaction (PCR) methods for species identification, blood meal and sporozoite enzyme-linked immunosorbent assay (ELISA), or other procedures, except routine salivary gland dissections for sporozoites where fresh specimens are required.

(e) Logistically, it is normal for field teams to complete standard field forms for each collection and to turn these forms over to the laboratory teams doing the subsequent processing. In such field studies, it is necessary to give unique identification numbers to each mosquito, either in the field at the time of collection, or in the laboratory during subsequent processing.
Assembly-line approaches for specimen processing

Mosquitoes captured in the field require processing using a variety of methods. For the sake of efficiency, it is standard practice to run an assembly-line mosquito processing operation using one or more qualified technicians at each station. Below is one possible strategy when mosquito dissections are required:

(a) Station No.1: Mosquito identification. For each screened cup or tube of mosquitoes from the field, identify individual mosquitoes taxonomically, using a dissecting microscope. Assign each identified mosquito a unique number and place it in a small petri dish labeled with the number.

(b) Station No. 2: Records. Record the identification number for each mosquito on field forms along with specific field data (e.g., study site, type of collection, station or house number, and date of collection) and the taxonomic identification. Normally, data are entered on standard field forms, but it may also be possible to enter the data directly into a computer.

(c) Station No. 3: Mosquito Dissection. Place each mosquito on a labeled glass slide and examine under a dissecting microscope.
   (i) Note the blood-feeding stages (see subheading 3.4.1);
   (ii) Remove the ovaries and note both parity (parous or nulliparous) and Christophers’ stages of ovarian development (see subheadings 3.4.2. and 3.4.3);
   (iii) Check for malaria oocysts, dissect the midgut in physiological saline, add a small drop of diluted mercuriochrome solution, and add a cover slip (1);
   (iv) Dissect the salivary glands to examine them for malaria sporozoites;
   (v) Record results on standard forms.

(d) Station No. 4: Examination of mosquito midguts and salivary glands. Using a compound microscope, inspect midguts for malaria oocysts (×100) and examine salivary glands for malaria sporozoites (×400). Record results on standard forms.

(e) Station No. 5: Additional processing. One or more technicians can be added to the assembly line to help prepare additional mosquito samples. For example, specific parts of the mosquito can be prepared for blood meal identification, sporozoite ELISA testing, determination of sugar-feeding (2), measurements of wing-length (3), PCR identification of species (see subheading 5.3.5), or any other type of processing needed for the scope of investigations being conducted.

(f) Check data forms to ensure they are complete and accurate. Also ensure that mosquito samples to be frozen are numbered correctly and that each vial number corresponds to the corresponding numbers on the data forms.

Taxonomic identification of mosquitoes

(a) Immobilize live wild-caught mosquitoes by aspirating them from cages, blowing them into disposable beakers containing 70% ethanol, and within 5 min, transferring them by forceps to other beakers containing either physiological saline, PBS, or M-199 media. This method immobilizes but does not normally kill the mosquitoes, and if the mosquitoes are allowed to dry on the slide, they will recover and fly away.
(b) Place mosquitoes individually on a glass slide and examine at ×10 under a dissection microscope.

(c) Identify the mosquito using standard taxonomic criteria. There are standard taxonomic keys available for each major geographic region and these need to be consulted if necessary. For the African region, one useful key is Gillies and DeMeillon.

5.4 Classification of blood-feeding stages and Christophers' stages of ovarian development

Figure 1A provides diagrams of mosquito blood-feeding stages, differences in ovaries between parous and nulliparous mosquitoes, and Christophers’ stages of ovarian development; further information is available in a WHO training manual (1). After the mosquito is identified, the following parameters can easily be determined.

**Blood-feeding stages**

(a) Examine the mosquito on a labelled glass slide, under a dissecting microscope at ×10.

(b) Classify blood-feeding stages as either empty (E), fed (F), half-gravid (HG), or gravid (G), according to the amount and condition of the blood in the mosquito midgut.

**Figure 1A: Mosquito blood-feeding stages**

A Blood-feeding stages

Empty  Fed  Half-gravid  Gravid

(c) Record results on the standard forms.

(d) For indoor-resting mosquitoes, data on the mosquito blood-feeding stages provide an account of the proportion of mosquitoes feeding on blood per day. The frequency of blood feeding defines the duration of gonotrophic cycles, an important parameter in calculating the survival rate based on determinations of mosquito parity rates.

**Parity status**

(a) Dissect ovaries from the same mosquito, as above, into a drop of physiological saline or PBS by gently pulling the last two abdominal segments with forceps while securing the mosquito thorax with another pair of forceps.

(b) Examine the ovaries to determine parity by observing them at about ×40. Nulliparous mosquitoes contain tightly coiled ovarioles while parous mosquitoes do not. (Fig 1B). Record results.
(c) If a mosquito is parous, determine the Christophers’ stage of ovarian development.
(d) The parity rate can be used to calculate the daily survival rate of mosquito populations.

**Christophers’ stages of ovarian development**
(a) For parous mosquitoes, gently tease apart one ovary to release the oocytes.
(b) Observe the oocytes at ×40 and record the Christophers’ stage of oocyte development as stage I, II, III, IV, or V (see Fig. 1C). Defining characteristics of each stage are as follows (1):“Stage I — egg follicle round, yolk granules absent; Stage II — egg follicle oval, yolk granules present; Stage II — early, a few fine granules of yolk around the nucleus of the ovum; Stage II — mid-yolk granules easily visible under low power; Stage II — late yolk granules very abundant, occupying about half the follicle; Stage III — yolk occupying about three-quarters of the follicle; Stage IV — egg follicle sausage-shaped; Stage V — ova fully formed with well-developed floats.” Record results on standard forms.

(c) For some *Anopheles* species, over at least part of their geographic ranges, a portion of mosquitoes feed on blood multiple times per gonotrophic cycle. Using combined data on blood-feeding stages and Christophers’ stages, those mosquitoes that are classified as F (i.e., fed) and contain stage IV or V oocysts are clearly refeeding before the completion of normal gonotrophic cycles.
Molecular methods for identifying species in anopheles species complexes

For *Anopheles* species complexes, it is standard practice to use either cytogenetic or molecular approaches for identifying species. Methods for cytogenetic identification: Fig. 1A: Mosquito blood-feeding stages. (B) Ovaries of mosquitoes showing differences between parous and nulliparous specimens. (C) Christophers’ stages of ovarian development.
ANNEX 6: VARIOUS FORMS FOR PHU DATA COLLECTION

Form P1

GOVERNMENT OF SIERRA LEONE
MINISTRY OF HEALTH AND SANITATION
DISTRICT HEALTH MANAGEMENT TEAM

MALARIASURVEILANCE AT SENTINEL SITES — INDOOR RESIDUAL SPRAYING

NAME OF PHU: ........................................... CHIEFDOM: .................................................................

SUSPECTED FEVER CASES

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Submitted by: ………………………………………………………………
Approved by: …………………………………………………………………
Date: …………………………………………………………………………
Date: …………………………………………………………………………
ANNEX 7: CHECKLIST FOR EVALUATING INDOOR RESIDUAL SPRAYING OPERATIONS

Daily IRS work plan — In-field spray operator

Monitoring..................................................

1. Spray Operator appearance or habits
   1.1 Use of protective clothing
   1.2 Procedure before starting to spray
   1.3 Mixing of insecticide
   1.4 Spraying technique
   1.5 Reporting procedures and record keeping
      1.5.1 Daily reporting format
      1.5.2 Weekly reporting format
      1.5.3 Monthly reporting format
      1.5.4 Completeness of all
   1.6 Feedback from community
   1.7 Community participation or involvement

2. Checklist for supervisors
   2.1 Completeness of the checklist
   2.2 Daily inspection and monitoring of use of insecticide by each spray operator (how do they do it?)

3. Evaluating sprayed structures
   3.1 Availability and completeness of IRS card in each household
   3.2 Completeness of spraying in a household
   3.3 Completeness of spraying in each structure

4. Equipment — spray pumps
   4.1 Check calibration of spray pumps
   4.2 Check condition of pump and maintenance

5. Daily inspection, maintenance and storage of sprayers
   5.1 Pre-spray checks
   5.2 Periodic checks
   5.3 Spares
   5.4 Nozzles
   5.5 Storage
6. Cleaning the spray pump in the field
   6.1 Triple rinse
   6.2 Progressive rinse method
   6.3 Essential field tools and spares

7. Storage and storage facilities for insecticides for indoor residual spraying
   7.1 Insecticide storage facility
   7.2 Precautionary equipment and materials
   7.3 Storage of insecticide and stock management
   7.4 Storage capacity
   7.5 Stock management

8. Facilities for spray operator
   8.1 Ablution or washing facilities
   8.2 Transport
   8.3 Camp

9. Efficacy of spray operation
   9.1 Bioassay schedule
   9.2 Solution strength

10. Sentinel Sites
    10.1 Situation in place and preparedness
        10.1.1 Availability of materials
        10.1.2 Capacity of staff

11. Capacity requirements and building

12. IRS campaign – IEC materials
    12.1 Communication and engagement of community leaders
    12.2 Awareness of communities on the IRS programme

13. Programme coordination with other interventions

14. Waste management
    14.1 Disposal of wash water
    14.2 Disposal of empty insecticide containers and sachets.