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EXECUTIVE SUMMARY

In 2019, Actellic® 300CS was sprayed in Migori County for the third consecutive year and for the second time in Homa Bay County. To monitor the impact of spraying on entomological indicators, mosquitoes were collected monthly in the two sprayed counties as well as four unsprayed counties—Kisumu, Siaya, Busia and Bungoma—using pyrethrum spray catches (PSCs), indoor CDC light traps, and window exit traps. Insecticide resistance testing was conducted using pyrethroids with and without PBO, pirimiphosmethyl, and the new insecticide clothianidin. Cone bioassays to determine the residual life of Actellic® 300CS and SumiShield® 50WG were also conducted, with SumiShield® 50WG being sprayed for the first time in Kenya in a pilot area in Homa Bay County.

Densities of *An. funestus* s.l. in Migori remained extremely low before the third year of spraying, indicating year-round suppression by IRS conducted with Actellic® 300CS in 2018. This was reinforced with the third spray round in 2019, as densities remained low. In Homa Bay County, which received the second round of IRS in February 2019, the pre-spray period consisted primarily of *An. funestus* (57.8%), while *An. gambiae* s.l. (mainly *An. arabiensis*) was the most commonly collected anopheline species (54.2%) after IRS.

The indoor host seeking and resting densities of both *An. funestus* and *An. gambiae* s.l. were significantly lower in the IRS sites in Migori and Homa Bay counties compared to unsprayed sites in Kisumu, Busia, Siaya apart from Bungoma County. In Siaya County, indoor host-seeking density of *An. funestus* was high throughout the collection period between October 2018–September 2019 peaking in April with a mean of >50 per light trap per night. The low densities of *An. funestus* in sprayed sites compared to unsprayed sites may be attributed to the impact of IRS. It might also indicate that IRS can be effective if implemented in other counties where there is no IRS, especially Siaya where the highest densities of *An. funestus* mosquitoes were observed.

Sporozoite rate in *An. funestus* was 0% in Migori while in Homa Bay, only two mosquitoes tested positive for sporozoites. In Homa Bay, low numbers of mosquitoes inflated the sporozoite rate; 1/23 (4.3%) for *An. arabiensis* before IRS and 1/18 (5.6%) for *An funestus* after IRS. In Siaya, the high number of *An funestus* mosquitoes improved the accuracy of the sporozoite rate, thus, 99/2,807 *An funestus* were positive between October 2018-February 2019, while the period beginning March–September 2019, 17/1,195 *An funestus* were positive. IRS resulted in lower malaria transmission in sprayed compared to unsprayed counties as estimated by the entomologic inoculation rate (EIR).

Pyrethroid resistance is widespread in Kenya. In Siaya, where *An. funestus* is the major vector, mosquitoes survived 10 times the diagnostic dose of deltamethrin. Similarly, in other sites, *An. gambiae* s.l. survived even when the diagnostic dose was increased up to 10 times. Partial susceptibility to deltamethrin/permethrin was restored when PBO was added to deltamethrin/permethrin indicating that PBO nets may potentially be useful in areas of pyrethroid resistance.

The successful involvement of community health volunteers (CHVs) in doing routine mosquito collection under a 'community surveillance program' indicate that it is possible to incorporate the community in routine surveillance activities but with caution. The strategy as implemented can be further strengthened by putting in place a more robust system to monitor the activity to ensure data quality.

In conclusion, in line with the recently launched Kenya Malaria Strategy, IRS with Actellic® 300CS is very effective in controlling malaria vectors in Kenya, and vectors continue to be susceptible to pirimiphosmethyl. *An. funestus* biting rates were particularly high in unsprayed Siaya County and expansion of IRS would be beneficial. It is necessary to continue monitoring the susceptibility of malaria vectors to all public health insecticides to help guide selection of effective malaria vector control interventions.

INTRODUCTION

Malaria vector control in Africa chiefly depends on the use of long-lasting insecticidal nets (LLINs) and the application of indoor residual spraying (IRS). The malaria endemic Lake Victoria basin in western Kenya has been identified as the highest burden area for malaria transmission nationally¹. Subsequently, application of IRS was reintroduced in one county in 2017 in addition to LLINs in an attempt to drive down malaria transmission through rapid reduction in vector densities. A second county was targeted for IRS beginning in 2018.

In Kenya, LLINs are mainly distributed through mass campaigns and routine distribution at antenatal clinics. The last mass campaign took place in 2017 where Dawa Plus was distributed in Homa Bay and Migori. The next campaign is scheduled for 2020, and PBO nets will be introduced in three counties of Busia, Bungoma, and Kakamega.

IRS was recently reintroduced in parts of western Kenya, with three rounds (2017, 2018, and 2019) of spraying with Actellic® 300CS (pirimiphos-methyl) in Migori County and two rounds in Homa Bay County (2018 and 2019). SumiShield® 50WG was also sprayed in two sites in Homa Bay County in 2019. The combination of IRS and use of LLINs is expected to provide greater protection to the populations at risk within the Lake Victoria malaria endemic regions of western Kenya.

Application of insecticide-based vector control methods are threatened by the rise and development of insecticide resistance in mosquito populations. While it is critical to use other classes of insecticide in IRS to preserve pyrethroids for LLINs, repeated application of the same insecticides should be avoided to prevent or delay the rise in insecticide resistance. The Kenya National Resistance Management Plan recommends rotation of insecticides with different modes of actions every two years2. Only pyrethroid and organophosphate insecticides were approved for IRS in Kenya by the Pest Control Products Board (PCPB) in 2019. With previous reports of widespread pyrethroid resistance, only organophosphates could be used for IRS. In February 2020, Fludora® Fusion (mixture of neonicotinoid and pyrethroid) was registered by the PCPB for IRS and this will make insecticide rotations viable in future. With repeat IRS in Migori County for three years with the same insecticide, it is critical to monitor susceptibility levels of the local vector populations to pirimiphos-methyl and clothianidin as well as other insecticides used in vector control to assess their efficacy against the local vector population. As pyrethroid LLINs are still the primary vector control strategy in most counties in western Kenya, it is critically important to monitor pyrethroid resistance levels and intensity.

In western Kenya, *An. funestus*, *An. gambiae s.s*. and *An. arabiensis* are the main malaria vectors3. Following the first application of IRS with pirimiphos-methyl in Migori County in 2017 and Homa Bay County in 2018, a remarkable reduction in populations of *An. funestus* were observed, but the impact on *An. arabiensis* was not as large. Application of IRS and use of LLINs have been reported to have a lesser impact on *An. arabiensis* as the vector is reported to rapidly exit houses after entry4.

The current report covers monthly entomological surveillance in Migori and Homa Bay counties which were sprayed with IRS and in unsprayed Kisumu, Siaya, Busia and Bungoma counties (all located in western Kenya). Additional seasonal data is presented for Turkana, Trans-Nzoia, and Kwale counties. Monthly community surveillance was piloted in epidemic highlands of Kakamega and Vihiga counties. Insecticide resistance testing with pyrethroids and pyrethroids plus PBO synergist was conducted in eleven and five counties, respectively.

The objectives were to: monitor malaria vector densities and behaviour in Migori and Homa Bay counties after IRS, in comparison to control sites; determine levels and mechanisms of insecticide resistance of local malaria vector populations; determine decay rates of insecticide on the walls following IRS; and pilot community-based surveillance. Insecticide resistance monitoring was also conducted to inform decision making for IRS and LLINs. Results from this monitoring are intended to guide decision making by the Division of National Malaria Program (DNMP) and other development partners in the fight against malaria.

1. METHODOLOGY

1.1 SURVEILLANCE SITES

Monthly entomological data was collected to monitor species composition, vector density, behavior and evaluate the impact of IRS on vector mosquitoes. Monitoring was conducted in six sites, four in Homa Bay and two in Migori (Table 1). The four comparison sites were Seme, Nyando, Muhoroni, and Nyakach sub-counties in neighbouring Kisumu County. Vector populations were also monitored in two sites each in Siaya, Bungoma, and Busia where pyrethroid LLINs are used. Seasonal trapping was conducted in Kwale, Trans-Nzoia, and Turkana counties. Monthly community-based surveillance was piloted in two sites each in Kakamega and Vihiga counties, which are highland epidemic sites. Insecticide resistance monitoring was conducted in one site per county in 11 counties (Table 1, Figure 1).

Table 1: Entomology Surveillance Sites with Details of Data Collected at Each Site

Figure 1: Map of Kenya Showing Location of Sentinel Sites

1.2 VECTOR DENSITY SURVEILLANCE

Pyrethrum spray collections (PSCs), indoor light traps, and window exit traps were used to monitor mosquito densities monthly. Outdoor trapping was introduced for monthly surveillance using the outdoor CDC light trap (CDC LT) and Furvela tent trap (FTT) Each month, from October 2018 to September 2019, 17 houses were sampled in each site, ten by indoor CDC-LT, seven by PSC, and five by exit traps (paired with five PSC houses) and one outdoor CDC-LT and one FTT. Different houses were sampled every month. All procedures were conducted according to VectorLink standard operating procedures.

1.3 PYRETHRUM SPRAY COLLECTIONS (PSC)

To monitor the number of indoor resting mosquitoes, PSCs were conducted early in the morning according to SOP 03/01 in seven houses per month in each site using 0.025% pyrethrum amplifiable concentrate mixed with 0.1% piperonyl butoxide in kerosene. The house was closed for 10 to15 minutes after which dead mosquitoes were collected from the sheets and transferred to the laboratory in a scintillation vial containing 70% ethanol.

1.4 INDOOR CDC LIGHT TRAP

CDC light traps were used to monitor densities of host seeking mosquitoes inside 10 houses per month per site with trapping conducted according to SOP 01/01. A single 12V CDC light trap was hung in each house in the sleeping area, approximately 1.5 meters from the ground, adjacent to an existing occupied pyrethroid bed net. The traps were run from 6:00 pm and mosquitoes were collected at 7:00 am the next morning.

1.5 WINDOW EXIT TRAPS

Window exit traps (WET) were used to monitor proportions of mosquitoes that exit houses before morning. They were installed in the same houses in the evening before PSC were conducted the following morning. In each house sampled, a single exit trap was installed on a window in the sleeping area. Trapped mosquitoes were removed from the trap in the morning using aspirators and were then placed into paper cups.

1.6 OUTDOOR CDC LIGHT TRAP AND FURVELA TENT TRAP

The CDC-LT was hung outdoors at 1.5 meters above the ground next to an occupied, untreated bed net. This was to sample outdoor biting mosquitoes. The basic principle of the FTT is that host odour and exhaled gases attract mosquitoes to the gap on the tent door. Close to the gap, a CDC trap (without the light, lid or grid) is placed horizontally outside the tent, two to three cm from the opening in the door. On approach to the opening, the insects are sucked into the trap and held in the standard CDC trap conical collection bag. The suction from the fan effectively prevents any mosquitoes from entering the tent, even at very high densities, so that the sleeper is not exposed.

1.7 INSECTICIDE RESISTANCE MONITORING

Mosquito collections for insecticide resistance monitoring were performed between January and September 2019. WHO susceptibility tests were conducted according to SOP 06/01 for permethrin (0.75%), deltamethrin (0.05%), alpha-cypermethrin (0.05%), pirimiphos-methyl (0.25%) and clothianidin (2%) 5. Synergist assays were conducted by pre-exposing mosquitoes to WHO papers treated with piperonyl butoxide (PBO) (4%) for one hour prior to exposure to pyrethroid treated paper for 60 minutes. Testing was conducted in one site per sub-county. All papers (except for clothianidin) were prepared by the WHO collaborating center, University Sains Malaysia. Clothianidin tests were conducted using filter papers prepared by VectorLink staff. Whatman® No.1 filter papers were treated with the diagnostic dose of clothianidin according to project standard operating procedures. After 60 minutes exposure to clothianidin treated papers, mosquitoes were transferred to a holding tube and mortality was monitored up to seven days post exposure. Treated papers were tested within 24 hours of preparation.

Larval stages of *An. gambiae* s.l. were collected in all sub-counties using larval dippers and sieves. The emergent adults were raised to two to five day old adults for insecticide resistance tests. *An. funestus* larval sites were difficult to locate, therefore adult mosquitoes of unknown age and mixed feeding status (mostly blood-fed and half-gravid) were collected by morning indoor resting catch using mouth aspirators and exposed the following day in bioassays.

At least 100 mosquitoes were exposed to each insecticide at a time in four replicates of 25 mosquitoes each. The samples were then transferred to a holding tube with cotton wool soaked in sugar solution. Mortality was scored 24 hours after exposure (and up to seven days for clothianidin).

1.8 QUALITY OF SPRAY AND DECAY RATE OF INSECTICIDE ON THE **WALL**

To determine the quality of spray in 2019 and residual duration of insecticide on the walls, wall bioassays were conducted within two weeks of IRS and monthly using an insectary-reared susceptible colony of *An. gambiae* s.s. Bioassays were conducted using the WHO cone bioassay in Migori and Homa Bay counties in 10 houses (making a total of 20 houses per month per county). Cone bioassays were conducted early in the morning according to SOP 09/01. The exposure time was 30 minutes with mosquitoes then monitored for mortality at 24 hours post exposure for houses sprayed with Actellic® 300 CS and for up to seven days for SumiShield®50WG. A parallel control exposure was run on an unsprayed surface (block board) close to each sprayed house. When mortality reduced, we investigated whether houses were replastered/re-smeared after IRS and if so, additional houses were selected for wall bioassays.

To check the air-borne fumigant effect of the insecticide, a small cage 20 cm x 20 cm x 20 cm containing 10 insectary-reared *An. gambiae* was placed one meter away from the sprayed wall (i.e., mosquitoes did not contact the sprayed walls). The mosquitoes were exposed for 30 minutes then transferred into paper cups and taken to an untreated holding room. Mortality was recorded 24 hours post exposure. Fumigant tests were done monthly in parallel with cone bioassay.

1.9 MOLECULAR ANALYSIS

All *Anopheles* mosquitoes collected (using all methods) were identified morphologically to species^{6,7}. The physiological status was determined by observation of the abdomen, and female mosquitoes were classified as either unfed, blood-fed, half gravid, or gravid. Female mosquitoes were divided into three parts for additional laboratory analyses: head and thorax were used for determination of sporozoite infection by enzyme linked immunosorbent assay (ELISA) techniques, ⁸ the abdomens of blood-fed and half-gravid females were kept for blood-meal host determination, and the legs and wings were used in polymerase chain reaction (PCR) assays to identify members of the *An. gambiae* s.l. complex and the *Anopheles funestus* s.l. group, and preserved for future genetic/molecular analysis. All mosquitoes morphologically identified as *An. gambiae* s.l. were analyzed by PCR for species identification while a random selection of 20% of *An. funestus* s.l. collected per month across all sites were initially analyzed for species identification in each month.

1.10 DATA MANAGEMENT

Field data was collected on tablets using open data kit software (ODK). Individual mosquitoes were labelled with pre-printed barcodes and linked to the field data by house code and study number. Sporozoite rate was estimated by calculating the ration of mosquitoes positive for sporozoite ELISA to total number of mosquitoes screened for sporozoites. Biting rate was estimated using CDC-LT as the number of mosquitoes collected divided by the total number of trap nights. To estimate the entomologic inoculation rate (EIR), the biting rate was multiplied by sporozoite rate. Monthly EIR was estimated by multiplying EIR by 30 days in a month. Seasonal EIR (pEIR) was computed by multiplying monthly EIR by number of months in each season (October 2018 to February 2019 and March to September 2019).

2.1 MALARIA VECTOR SPECIES COMPOSITION

A total of 17,550 *Anopheles* mosquitoes were collected by all trapping methods in all sites combined. Of these, 14,293 (81.4%) were *An. funestus* s.l., 2,716 (15.5%) *An. gambiae* s.l., 507 (2.9%) *An. Coustani,* and 34 (0.2%) *An. pharoensis*.

In the sprayed counties of Migori and Homa Bay, the total number of anopheline mosquitoes collected was relatively low and *An gambiae* s.l. was the predominant species after IRS (Figure 2), consistent with the previous 2018 annual report.

In unsprayed counties of Kisumu and Siaya, *An. funestus* was the predominant species during both collection periods (Figure 3). In the non-sprayed sites of Busia and Bungoma, species composition changed during the collection period between October 2018 and September 2019 in response to changes in seasonality (Figure 3). In Bungoma, *An. funestus* was the major malaria vector between October 2018 and February 2019 but from March to September 2019, *An gambiae* s.l. was the primary vector collected. In Busia, between October to February 2019, *An gambiae* s.l. was the predominant species but from March to September 2019 it was *An. funestus*.

A total of 1,259 *An. gambiae* s.l. were analysed by PCR for species identification: 826 (65.6%) were *An. arabiensis* and 433 (34.4%) were *An. gambiae* s.s*.* A total of 5,262 *An. funestus* were analysed by PCR and those that amplified were confirmed to be *An. funestus* s.s. A total of 504 (7%) *Anopheles* (morphologically identified as either *An gambiae* s.l or *An funestus* complex) did not amplify. In the next work plan, the number of *An funestus* to be identified by PCR will be reduced considering nearly all were *An. funestus* s.s.

In the two counties receiving IRS, *An. arabiensis* was the most common species compared to non-sprayed sites where *An funestus* and *An gambiae* s.l predominated. Detailed tables are presented in Annex A1 and A2 showing catch size by mosquito species, trapping method, and site. In Migori and Homa Bay, mosquito densities were generally far lower than non-sprayed sites (Annex A1).

Figure 2: Species Composition (Morphological ID) for Migori and Homa Bay Counties pre-2019 IRS (October 2018–February 2019) and post-2019 IRS (March–September 2019) Collected by CDC LT Indoors, PSC, and WET.

Figure 3: Species Composition (Morphological ID) for All Non-IRS sites from October 2018– February 2019 and March–September 2019, collected by CDC LT Indoors, PSC and WET.

2.1.1 MALARIA VECTOR SPECIES COMPOSITION IN SEASONAL AND **COMMUNITY SURVEILLANCE SITES**

In Turkana and Trans Nzoia, only one data point was collected, while in Kwale, mosquitoes were collected in November 2018, June 2019 and October 2019. *An funestus* (79.8%) was the major malaria vector followed by *An gambiae* s.l. (12.9%) and other species (7.3%) (Figure 4). In Trans Nzoia, collections were made in December 2018 and mosquito density was relatively low compared to the other two counties. In Trans Nzoia, *An funestus* comprised 54.2% of collected mosquitoes while the remaining 45.8% were *An gambiae* s.l. In Turkana, collections were done in May 2019 and the majority were *An gambiae* s.l. (82.5%) followed by *An funestus* (14.4 %) and other species 3.1% (Figure 4).

Figure 4: *Anopheles* **Species Composition Collected from Seasonal Surveillance Sites in Turkana, Kwale, and Trans Nzoia in 2018–2019.**

In Kakamega and Vihiga counties, where a community surveillance system was implemented for the first time, a total of 4,302 *Anopheles* were sampled. *An. gambiae* s.l. comprised 70.1% of mosquitoes collected followed by *An funestus* (29.6%), *An coustani* (0.3%), and *An pharoensis* (0.07%). The distribution of these species varied by collection site with the majority of mosquitoes coming from Buhili and Eshiakulo villages in Kakamega county (Figure 5, Annex A3).

2.2 MALARIA VECTOR SEASONALITY BY INDOOR CDC LIGHT TRAP, PYRETHRUM SPRAY CATCH, WINDOW EXIT TRAP, AND OUTDOOR TRAPS

In the unsprayed counties, the density of *An funestus* as sampled by CDC-LT was higher than that of *An gambiae* with Siaya and Kisumu counties having the highest density of malaria vectors (Figure 6, Annex A1-A2). Mean monthly biting densities estimated from indoor CDC-LT were low in the sprayed counties compared to unsprayed (Figure 6). Siaya County had the highest mean monthly biting density, particularly for *An funestus*. In Bungoma the biting density was highest from April, peaking in June. The CDC-LT captured the highest densities compared to other traps (Figures 7 and 8). A detailed table is presented in Annex A1 and A2 showing catch size by mosquito species, trapping method, and site.

Figure 6: Mean Monthly Density of *An. gambiae* **s.l. (top panel) and** *An. funestus* **(bottom panel) per Trap per Night in Sprayed and Unsprayed Counties.**

Figure 7: Mean Monthly Density per Hut per Day of *An. gambiae* **s.l. (left panel) and** *An. funestus* **(right panel) in Sprayed and Unsprayed Counties as Sampled by PSC (top panel) and WET (bottom panel).**

Figure 8: Mean Monthly Density of *An. gambiae* **s.l. (left panel) and** *An. funestus* **(right panel) per Trap per Night in Sprayed and Unsprayed Counties as Sampled by FTT (top panel) and Outdoor CDC LT (bottom panel).**

2.2.1 COMMUNITY SURVEILLANCE

A total of 4,302 mosquitoes were sampled in Kakamega (Buhili and Eshiakulo) and Vihiga (Busamo and Ebulakho) counties where community surveillance was carried out for the first time. The density of malaria vectors in Kakamega was higher than that of Vihiga for both *An. gambiae* s.l. and *An. funestus* (Annex A3 and 4). The mean monthly biting density was highest in Buhili and Eshiakulo both for *An gambiae* s.l. and *An funestus* (Figure 9). Mean biting density was highest in June for *An gambiae* s.l. and April and July for *An funestus* (Figure 9).

Figure 9: Mean Monthly Density of *An. gambiae* **s.l. (top) and** *An. funestus* **(bottom) per Trap per Night in Community Surveillance Sites in Kakamega and Vihiga as Estimated by Indoor CDC LT.**

2.3 MALARIA VECTOR SPOROZOITE RATES AND ENTOMOLOGIC INOCULATION RATE

In Migori County (sprayed sub-county), no sporozoite-infected mosquitoes were found during the oneyear sampling duration between October 2018 to September 2019, mainly due to the very low number of malaria vectors collected (Figure 10). In Homa Bay (sprayed sub-county), only two mosquitoes tested positive, one each in pre and post 2019 IRS periods (Figure 10). The unsprayed counties had the highest sporozoite rates at 3.5% (Siaya) to 9.4% (Bungoma) during October 2018 to February 2019. Sporozoite rates were slightly lower between March to September 2019 but were higher in unsprayed sites than sprayed sites (Figure 10). In Bungoma, Siaya, and Busia, both *An arabiensis* and *An funestus* are involved in malaria transmission (Figure 11). Detailed sporozoite rate by species by county is presented in Annex A1.

Labels show number positive / total tested.

Sporozoite rate varied by county and species when separated by malaria vector species and spray status for the period October 2018 to September 2019. In Homa Bay, low sample size inflated the sporozoite rate as only 1/23 *An arabiensis,* and 1/18 *An funestus* was positive for malaria sporozoite (Figure 11, Annex A5). In Siaya, the high number of *An funestus* mosquitoes allowed for a more accurate estimate of sporozoite rate. Between October 2018 to February 2019, 99/2,807 *An. funestus* were positive while the period beginning March to September 2019, 17/1,195 *An funestus* were positive. Table 2 shows sporozoite rates by species and county.

Figure 11: Sporozoite Rate by Vector Species and Sites, October 2018–September 2019.

Malaria transmission as estimated by entomologic inoculation rate (EIR) was slightly lower in the period following IRS in Homa Bay (Table 2). There was no infected mosquito sampled in Migori. For other counties, EIR was higher in the period March to September 2019 because the monthly EIR was multiplied by the number of months (Table 2).

Table 2: Malaria Transmission in Kenya as Estimated by Entomologic Inoculation Rate.

Period EIR = Monthly EIR multiplied by number of months of collection period (e.g., Oct 18–-Feb19—5 months)

2.4 INSECTICIDE RESISTANCE TESTING

Anopheles gambiae s.l. from different counties had varying levels of resistance to pyrethroid insecticides. There was widespread resistance to the diagnostic dose of permethrin and deltamethrin (Figure 12). The synergist PBO increased mortality of *An. gambiae* s.l. to deltamethrin to more than 90% in all sites except in Msambweni (Kwale) and Kakuma (Turkana). For permethrin, PBO increased mortality to more than 90% in two of five sites tested -Kinango and Lodwar (Figure 12). Results are also are shown as a table in Annex A6.

Pyrethroid resistance was also reported in *An. funestus* in Kisumu and Siaya (Figure 13). PBO only restored full susceptibility to deltamethrn from Kisumu but not Siaya. *An funestus* are also still fully susceptible to pirimiphos-methyl (Figure 13).

Different concentrations of permethrin and deltamethrin were used to investigate intensity of resistance. The number of mosquitoes dying increased with every increase in concentration of insecticides on bottle assays (Figures 14 and 15). In Siaya, *An gambiae* s.l. were resistant to all doses of deltamethrin and permethrin, indicating high intensity resistance. The intensity of insecticide resistance to pyrethroids in malaria vectors was generally moderate or high (Figures 14 and 15). Resistance intensity results are also are shown as a table in Annex A7.

Figure 15: Intensity of Insecticide Resistance of *An. gambiae* **sl to Permethrin in Kenya in 2019 using the CDC Bottle Bioassay for Intensity.**

There was resistance to alpha-cypermethrin in all counties where testing was conducted (Figures 16 and 17). *An. gambiae* s.l. were susceptible to pirimiphos-methyl in all locations tested, except from Loima in Turkana County (Figure 16). *An. gambiae* s.l. that were exposed to clothianidin were all killed within seven days of exposure, although 100% mortality was only achieved on day six for Homa Bay and day seven for Siaya.

Figure 16: Susceptibility of *An. gambiae* **sl to Alpha-cypermethrin and Pirimiphos-methyl in Kenya in 2019.**

Figure 17: Susceptibility of *An. gambiae* **s.l. to Clothianidin in Kenya in 2019.**

2.5 RESIDUAL DURATION OF ACTELLIC® 300 CS AND SUMISHIELD® 50WG INSECTICIDE FORMULATIONS

The susceptible insectary strain *An. gambiae* s.s. Kisumu was exposed to sprayed walls at different heights over six months post-IRS. Thirty houses sprayed with Actellic®300 CS were tested every month (7 mud and 3 cement per site). Within one week of IRS, 100% mortality was observed in all sub-counties on both mud and cement walls, but a gradual decrease occurred in most sites from month to month. Mortality stayed above the WHO threshold of 80% for only two to four months after spraying and was consistent in all sites on mud and cement (Figure 18).

Figure 18: Mean Monthly Mortality (24h) of Susceptible *An. gambiae* **Kisumu Following Cone Bioassay on Walls Sprayed with Actellic ®300 CS**

Twenty houses sprayed with SumiShield® 50WG were tested every month (7 mud and 3 cement per site). Mortality was recorded every 24 hours for up to seven days. Figure 19 presents 24 hours mortality, and Figure 20 shows mortality at 72 hours after exposure. Control mortality was generally greater than 20% four or five days after exposure, therefore results beyond 72h are not presented. When considering 24 hour mortality, the residual duration of SumiShield® 50WG was approximately three months (Figure 19), but this increased to at least seven months (the last month covered in this reporting period) when 72 hour mortality was considered (Figure 20).

Figure 20: Mean Monthly Mortality (72h) of Susceptible *An***.** *gambiae* **Kisumu Following Cone Bioassay on Walls Sprayed with SumiShield® 50WG**

3. DISCUSSION

Indoor residual spraying (IRS) with Actellic®300 CS in western Kenya continues to be highly effective against indoor *Anopheles* mosquitoes, consistent with previous reports (2017/2018). Analysis of mosquito density data collected from October 2018 to September 2019 show extremely low numbers of malaria vectors in the two counties of Homa Bay and Migori where spraying was undertaken. In unsprayed counties, malaria vector densities were higher than sprayed sites, but as expected, were not uniform across all villages and in Siaya County *An. funestus* densities were slightly skewed by one particularly productive site. Since IRS has reduced the density of indoor biting mosquitoes (*An. funestus* and *An. gambiae* s.s.), *An arabiensis* has become the dominant vector in the two sprayed counties, albeit at lower densities. Although we did not study the behaviour of *An arabiensis*, this species is known to feed outdoors and on other animals and is not directly targeted by IRS and LLINs.

Subsequently, the number of sporozoite positive mosquitoes have remained extremely low in sprayed counties compared to those not sprayed. In Migori County, there were no positive samples for malaria sporozoites. This implies that the chances of the two million residents in the two counties being bitten by infected mosquitoes have significantly reduced. Although the remit of VectorLink is to monitor impact of IRS on malaria vectors, monitoring epidemiological outcomes will help corroborate the entomological outcomes. In Homa Bay, there was reduced malaria transmission post IRS as estimated by EIR. There were no infected mosquitoes in Migori to allow estimation of EIR. Other counties had higher EIR in the two periods compared to the sprayed counties of Homa Bay and Migori indicating the impact of IRS in reducing malaria transmission. Since mosquito numbers vary by village, the overall EIR reported for each county may also vary by village. Overall, EIR was lower in IRS-sprayed counties than unsprayed.

The duration of Actellic®300 CS on walls only lasted three months in 2019 compared to 2017/2018 where it lasted about eight months. To confirm reduced mortality, VectorLink investigated how many houses were re-plastered/re-smeared. Even when additional houses were selected, mortality patterns remained the same. The reasons for this are undetermined, but analysis of insecticide by an independent laboratory in the UK confirmed no issues with the insecticide. Additionally, the project will investigate natural reasons for the fluctuation of the duration of longevity on the wall. VectorLink is investigating whether changes in weather patterns (temperature and humidity) could have resulted in reduced duration of insecticides on the walls.

The residual effect of SumiShield® 50WG was also similar to Actellic® 300CS where its effectiveness in killing 80% of mosquitoes 24 hours post exposure as required by WHO lasted about three months. However, when 72 hours mortality was considered, it was effective up to seven months (with bioassays ongoing).

Pyrethroid resistance is widespread. In Siaya, where *An. funestus* is the major vector, mosquitoes still survived when the insecticide dose was increased 10 times. Similarly in other sites, mosquitoes survived even when the diagnostic dose was increased, with moderate to high resistance intensity common. Partial susceptibility to deltamethrin and permethrin was restored when PBO was mixed with either of the insecticides. This indicates that nets treated with PBO or those with a dual active ingredient may improve control of pyrethroid resistant mosquitoes.

To counter the insecticide resistance problem, the Division of National Malaria Program has planned to distribute PBO nets in Busia, Kakamega, and Bungoma. It will be important to control malaria vectors in Siaya where there is also high malaria burden in the western region. Since this is the third year of IRS with pirimiphos-methyl, it will be important to continue monitoring the susceptibility of malaria vectors to it and introduce sub-national rotation with insecticide with a different mode of action to preserve the effectiveness.

The successful involvement of community health volunteers (CHVs) in doing routine mosquito collection under 'community surveillance program' indicate that it is possible to incorporate the

community in routine surveillance activities but with caution. The strategy as implemented needs to be strengthened by increasing the level of monitoring to ensure quality data is generated without falsification. Lessons can be learnt from Tanzania during the urban Malaria Control Program between 2007–2015 where the community was involved in monitoring malaria vectors⁹.

Limited outdoor longitudinal trapping was conducted using Furvela tent traps and outdoor CDC LT. However, the number of malaria vectors collected per trap was far lower than by indoor CDC. The time taken to set up an outdoor trap per site limited the number of traps per site to just one. While it is important to gather more information on outdoor biting risk, particularly for *An. arabiensis*, it may be better to conduct shorter more intensive studies with specific research questions rather than conducting longitudinal monitoring with a small number of outdoor traps. It appears that CDC-LT is an effective trapping method to sample indoor biting mosquitoes and collected far greater numbers than PSC or WET collections. WET can be useful for specific studies interested in exiting behaviour but is not providing useful information for longitudinal monitoring. Hence, the recommendation to stop routine monitoring with WET. PSC collections should continue as it can provide an indication of IRS performance.

In conclusion, in line with the recently launched Kenya Malaria Strategy, IRS is very effective in controlling malaria vectors, especially pyrethroid-resistant indoor mosquitoes in Kenya. *An. funestus* biting rates were particularly high in unsprayed Siaya County and expansion of IRS would be beneficial. In addition, it might be beneficial to deploy PBO nets or nets with dual active ingredients across the entire lake region where vectors are resistant to pyrethroids. It is also necessary to continue monitoring the susceptibility of malaria vectors to Actellic® 300CS to avert development of resistance to IRS insecticides. Additional tools to control the opportunistic *An. arabiensis* will be needed in Kenya as the country intensifies control toward malaria elimination.

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ANNEX

Table A1: Mean Number of *An. gambiae* **s.l. Collected Per Trap-Night By Different Collection Methods, Before (October 2018–February 2019) and After (March–September 2019) 2019 IRS.**

Table A2: Mean Number of *An. funestus* **Per Trap-Night By Different Collection Methods, Before (October 2018–February 2019) and After (March– September 2019) 2019 IRS.**

Table A3: Mean Number of *An. gambiae sl* **Per Trap-Night by CDC Light Trap Collected in 2019 in Kakamega and Vihiga**

Table A4: Mean Number of *An. funestus* **Per Trap-Night by CDC Light Trap Collected in 2019 in Kakamega and Vihiga**

Table A5: Sporozoite Rate by Vector Species and IRS Status, October 2018-September 2019.

Table A6: Percentage Mortality of *An gambiae* **s.l. to Deltamethrin or Permethrin Alone and Following Pre-Exposure to PBO in WHO Tube Tests in Kenya, 2019 (n≈100)**

Table A7: Percentage Mortality of *An gambiae* **s.l. to Deltamethrin or Permethrin at 1×, 2×, 5× and 10× the diagnostic concentration in CDC bottle bioassays in Kenya, 2019 (n≈50)**

