## **Abstract Book**



## PAMCA 2018 5th PAMCA Annual Conference and Exhibition Victoria Falls ~ Zimbabwe

24th - 26th September

Building the Bridge Between Research and Implementation: A Necessity for Vector Borne Disease Elimination in Africa

<image><image>





## The 5th Pan-African Mosquito Control Association Conference

"Building the Bridge Between Research and Implementation: A Necessity for Vector Borne Disease Elimination in Africa"



Monday 24th - Wednesday 26th September, 2018 Elephant Hills Resort Hotel, Victoria Falls, Zimbabwe

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### **PAMCA Committees**

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### The 5th Pan African Mosquito Control Association (PAMCA) Conference

The 5th Pan African Mosquito Control Association (PAMCA) Conference themed, "Building the Bridge Between Research and Implementation: A Necessity for Vector Borne Disease Elimination in Africa" is being organized in Victoria Falls, Zimbabwe, from the 24th - 26th September 2018.

#### **Conference Sub-Themes:**

- Challenges in malaria vector surveillance and control: researcher and implementer perspectives
- Progress in Sterile Insect Technique and Gene Drive for the control of vector-borne diseases
- Mosquitoes: host-pathogens and bio-control
- Innovations and technologies in vector-borne disease surveillance and control
- Integrated mosquito vector abatement programs in Africa

The conference will feature oral presentations and poster sessions. The outcome of the conference will be summarized in a report that will be distributed to all participants and key stakeholders. The media is also invited to cover the event.

## Welcome Address

Prof. Charles Mbogo | PAMCA President

## Dr. David Zinyengere | Chair, Local Organizing Committee

## Abstracts

# **Plenary Sessions**

### Improvement of transgenic strains of *aedes aegypti* for the control of arbovirus transmission in brazil

#### Margareth Lara Capurro, Universidade de São Paulo

The most common strategy employed to combat target mosquito species is the Integrated Vector Management (IVM), which comprises the use of multiple activities and various approaches to preventing the spread of a vector in infested areas. IVM programs are becoming ineffective; and the global scenario is threatening, requiring new interventions for vector control and surveillance. Since 1940, the vector control has changed very little, showing that the integrated vector management (IVM) has an important role and these new techniques should be incorporated in order to increase the chances to avoid transmission. The possibility of using transgenic mosquitoes to fight against those diseases has been discussed over the last two decades and this use of transgenic lines to suppress populations or to replace them is still under investigation through field and laboratory trials. As an alternative, the available transgenic strategies could be improved by coupling suppression and substitution strategies. The idea is to first release a suppression line to significantly reduce the wild population, and once the first objective is reached a second release using a substitution line could be then performed. This hypothesis is based on the use of transgenic lines of Ae. aegypti, to be adding methodology of dengue control already used. The results using RIDL technology show that this strategy is viable to reduce mosquitoes population. However, it is unique and can be improved. Our strategy is based on the creation and use of three strains of genetically modified Ae. aegypti. The first act in suppressing populations (male sterile) in association with the second strain that is genetically modified to produce only males (female sex reversion - GSS) and the third acting in introducing gene (Virus-lethal) In terms of innovation and improvement in population suppression, our strain have two points: the first is that for the production of male-sterile, tetracycline not be used in mass production (only in the colonies), thereby reducing the cost of production thus as the treatment of produced water. In addition to obtaining the GSS strain (males only) would not need to physically separate the males from the females and 100% of the production will be released, greatly improving the efficiency of the process.

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# **Oral Abstracts**

### Entomological factors sustaining Residual malaria transmission in the coastal Kenya

**Authors:** Joseph Mwangangi<sup>1,2,3</sup>, Martin Rono<sup>1</sup>, Antonnio-Nkondjio Christophe<sup>4</sup>, Delenasaw Yewhalaw<sup>5</sup>, Simon Muriu<sup>3</sup>, Prophet Mulega<sup>1, 3,</sup> Janet Midega<sup>1</sup>, Norbert Peshu<sup>1</sup> and Charles Mbogo<sup>1,2</sup>

<sup>1</sup>Kenya Medical Research Institute, Center for Geographic Medicine Research Coast, Kilifi, <sup>2</sup>Public Health Department, KEMRI-Wellcome Trust Research Program, Nairobi, Kenya <sup>3</sup>Pwani University, Department of Biological Sciences, Kilifi, <sup>4</sup>Organisation de Coordination pour la lutte contre les Endémies en Afrique centrale (OCEAC), 5Tropical and Infectious Diseases Research Centre (TIDRC), Jimma University

**Background:** With the scale up of mosquito control strategies such as use of Long lasting insecticide treated bednets and indoor residual spraying, malaria transmission in some areas have reduced significantly. However, transmission still persists and contributing still to disease burden. This study assessed the entomological factors that sustain malaria transmission in the coastal region of Kenya.

**Methodology:** Indoor and outdoor entomological sampling was done 12 selected villages in Kilifi and Taveta subcounties using CDC light traps and Human Landing Catches (HLC). All Anopheles mosquitoes were identified morphologically and sibling species of An. gambiae complex further identified using rDNA Polymerase Chain Reaction (PCR) technique. All the Anopheles collected were tested for sporozoite infectivity using P. falciparum sporozoite infectivity and blood meal sources using enzyme linked immunosorbent assay (ELISA) technique. Phenotypic insecticide susceptibility testing was done using standard WHO test kit.

**Results:** Overall, a total of 2,042 Anopheles mosquitoes were collected in Kilifi (n = 415) and Taveta (n = 1,627) sub-counties. In Kilifi the vectorial system consisted of mainly Anopheles funestus s.l. (75%) as dominant species, followed by An. arabiensis, An. gambiae s.s., and An. pretoriensis. Plasmodium falciparum sporozoite infectivity showed an overall rate of 2.65 %. In Kilifi, most infectious bites was contributed by An. funestus s.l. transmitting outdoors. In Taveta, Anopheles mosquitoes consisted mainly An. arabiensis (78.05), and other species were An. funestus, An. coustani, An. pretoriensis, An. pharoensis and An. squamosus. Mosquitoes collected resting indoors had the highest sporozoite infection rates in Taveta (3.06% for An. gambiae and 2.99% for both An. gambiae and An. funestus). For mosquitoes collected outdoor, An. coustani had the highest sporozoite infection rate (3.66%) followed by An. funestus (1.18%) and An. gambiae (1.13%). Insecticide resistance to Permethrin was 93 % (0.88-0.98, 95 % C.I) while Deltamethrin - 85 % (0.90-1.01, 95 % C.I).

**Conclusion:** In this study, malaria transmission is happening both indoors and outdoors. *Anopheles gambiae* is main vector indoors, while An. funestus, An. arabiensis and An. coustani contributed most malaria transmissions outdoors. Further, this study has shown that there is an increase in these areas minor species like An. coustani which are transmitting malaria outdoors.

From this study, there is need to target outdoor malaria transmission and initiate an insecticide resistance management strategy to sustain the gains made in malaria control.

Key words: Residual transmission, insecticide resistance, Anopheles funestus, Vector control, Strategies

### Entomological surveillance as a cornerstone of malaria elimination: a critical appraisal

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Abstract: Global capacity for developing new insecticides and vector control products, as well as mathematical models to evaluate their likely impact upon malaria transmission has greatly improved in recent years. Given that a range of new vector control products now emerging that target a greater diversity of adult mosquito behaviours, it should soon be feasible to effectively tackle a broader range of mosquito species and settings. However, the primary obstacles to further progress towards more effective malaria vector control are now paucities of routine programmatic entomological surveillance, and capacity for data processing, analysis and interpretation in endemic countries. Well-established entomological methods need to be more widely utilized for routine programmatic surveillance of vector behaviours and insecticide susceptibility, the effectiveness of vector control products and processes, and their impacts on mosquito populations. Such programmatic data may also be useful for simulation analyses of mosquito life histories, to identify opportunities for pre-emptively intervening early in the life cycle of mosquitoes, rather than targeting transmission events occurring when they are older. Current obstacles to more effective utilization, archiving and sharing of entomological data largely centre around global inequities of analytical capacity. These prohibitive and unfair imbalances can be addressed by reorienting funding schemes to emphasize south-centred collaborations focused on malaria-endemic countries.

Keywords: Malaria, entomological surveillance, mathematical modelling, capacity strengthening, vector control

Building capacity for implementation of integrated vector management: Insights from a pilot project in Kenya and Ethiopia

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<sup>1</sup>International Centre of Insect Physiology and Ecology (ICIPE); <sup>2</sup>Kenya Medical Research Institute (KEMRI); <sup>3</sup>UP Institute for Sustainable Malaria Control & MRC Collaborating Centre for Malaria Research, School of Health Systems and Public Health, University of Pretoria

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**Background:** Integrated vector management (IVM) is a practical strategy for sustainable control of vectorborne diseases. However, few examples exist of coordinated and systematic implementation of the strategy's key elements including social mobilization, generation and utilization of research evidence and multi-sectoral collaboration. The objectives of the study were: to demonstrate IVM for malaria control at community level and assess its health and socio-economic impacts; develop and promote eco-friendly biopesticides for vector control; strengthen advocacy for implementation of IVM policies at national level.

**Methods:** The field research was conducted in Malindi and Nyabondo in Kenya and Tolay in Ethiopia from 2013-2015. A randomized controlled study design was used to assess the effects of the following four intervention options on mosquitoes and malaria prevalence: LLINs (control); LLINs+CEM; LLINs+Bti; and, LLINs+CEM+Bti (i.e. the relatively comprehensive IVM package). Potential plant-based biopesticides were evaluated for efficacy in the laboratory and under field conditions while door to door IVM awareness activities and multisectoral workshops were conducted at community and policy levels respectively.

**Results:** In Tolay and Malindi, malaria prevalence was generally low, but 50-70% lower in villages with LLINs+CEM+Bti option when compared to those with only LLINs. Similarly, anopheline mosquitoes were fewer in villages with integrated interventions at the two project sites. Nyabondo site had no clear pattern of reduction or increase of mosquitoes and malaria. An estimated 12,000 community members in Ethiopia, 81,000 in Kenya and 180 professional staff in both countries were reached with IVM information and education. One biopesticide was fully developed in Kenya while IVM inter-agency coordinating committees were formed in both countries. Furthermore, the project contributed to development of a new IVM project focused on six southern African countries.

**Conclusions:** IVM is feasible at community and policy levels in Kenya and Ethiopia. However, sustaining it will require long-term support from relevant government bodies, development partners and research institutes.

Key Words: Integrated vector management, vector borne diseases, malaria

### Integrated Vector Control: Proven Methods or Miasmatic Vapors?

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**Abstract:** Dr. Ronald Ross confirmed the transmission of malaria by anopheles mosquitoes during the last few years of the 19th Century. This earth-shattering accomplishment earned him the Nobel Price. A few years later, after decades of losses to malaria, yellow fever and other mosquito-borne diseases and two failed attempts by the French, environmental sanitation and larval source management (LSM) campaigns eliminated the mosquito from parts of Panama, saving countless lives and paving the way for the opening of the Panama Canal. In 1925, Israel Kligler eradicated malaria from Palestine using LSM. In the mid 1930s, Fred Soper eradicated the malaria vector from an area the size of Togo, West Africa using LSM and stopped an outbreak of malaria that had claimed a good portion of the population in northeastern Brazil. The Continental United States and Puerto Rico were declared malaria free during the first few years of the 1950s after implementing comprehensive LSM campaigns as part of an Integrated Vector Management program. Regrettably, by the end of the Twentieth Century, the names of those who eradicated malaria vectors from over one hundred countries and liberated countless citizens from the yolk of many mosquito-borne diseases have fallen off the pages of public health history books. More tragically, the methods they used are today considered valid only when mosquito breeding sites are "few, fixed and findable" and integrated mosquito control continues to be relegated behind better diagnostic and treatment methods.

Shortly after accepting the Nobel Price, Dr. Ross voiced his concern about malaria control by saying: "Malaria will continue to affect the world until the mosquito is taken seriously." Today, well into the Twenty-first Century, mosquitoes remain unchallenged, mosquito-borne diseases continue to afflict half the world's population and continue to have serious adverse effects on the global economy

### Emergent Multiple Anopheles Spp Surpassing Major Malaria Vectors In Zimbabwe

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**Background:** Malaria transmission has declined in most of Zimbabwe in the wake of scaled-up prevention, control and elimination interventions. However, this successful decline is made complex by being heterogeneous, with some areas experiencing resurgence, including invasion of erstwhile malaria-free highlands. This transitioning epidemiology complicates control efforts and demands systematic tracking of changing vector, composition and behaviour. Here we present findings from nationwide sentinel-based malaria vector surveillance in Zimbabwe.

**Methods:** Wild mosquitoes were captured indoors and outdoors from 18 national sentinel surveillance sites during the 2017 malaria season. The specimens were analyzed by Anopheles sibling species differentiation PCR and Plasmodium falciparum circumsporozoite ELISA.

**Results:** Despite continued malaria transmission, the expected main vectors of Zimbabwe, Anopheles arabiensis, An. gambiae ss and An. funestus were strikingly absent, appearing only sparingly in 5, 1 and 5 of the 18 sentinel sites, with overall mean abundances of 8.8%, 0.6% and 2.2%, and sporozoite rates of 0%, 0% and 7.4%, respectively. During the apparent malaria with missing vectors, there was marked abundance of hitherto secondary vectors (An. rufipes (8.8%), An. leesoni (9.6%), An. rivolurum (1.0%), An. parensis (4.1%) and another as yet unidentified species (An. unknown (17.9%)). The overall abundance of these emergent vectors surpassed the expected main vectors (M-W U = 61, p < 0.001, n = 2556). The An. rufipes, An. leesoni, An. rivolurum, An. parensis and An. (unknown), were found at 10, 6, 4, 6 and 9 sentinel sites, with sporozoite rates of 1.2%, 1.0%, 1.8%, 1.0% and 1.7%, respectively. The emergent vectors exhibited human blood indices [mean 0.65; maximum, 0.79], which were comparable to those for the main vectors [mean, 0.67; maximum, 0.71; P = 1.000, n = 107].

**Conclusions:** Scaled-up interventions against the major malaria vectors of Zimbabwe are proving successful. However, new vectors are being identified that may demand new targeting and surveillance strategies for malaria elimination to be successful.

### Genome wide selection scans discover novel putative insecticide resistance loci in An. gambiae

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Malaria vectors are subject to a variety of intense selective pressures. We performed a genome-wide scan for signatures of recent selection in Anopheles gambiae populations sampled from 8 African countries, using WGS data from the Anopheles gambiae 1000 genomes (Ag1000G) project.

We used several established statistical methods designed to detect selection. We then applied a novel regression analysis to identify genes putatively under selection and distinguish them from false positives.

Genes known to be involved in insecticide resistance were among the strongest signals, including the target-site resistance genes Vgsc, Rdl, Ace1, and the metabolic resistance loci Gste, Cyp6p and Cyp9k1. Additionally, we identified strong signals of selection at 7 novel loci. We describe these novel hits in detail, presenting the evidence for selection at each locus, and discussing previous work to better understand the biological reason for selection.

Four of these novel signals coincided with genes with a plausible biochemical role in insecticide resistance. One signal encompassed carboxylesterase genes, that have been associated with resistance to organophosphates and carbamates in several insect species. Another signal was present at an ABC transporter gene on chr2R, this class of membrane pump has been implicated in insecticide resistance via selective efflux of toxic compounds. A signal was found at the diacylglycerol kinase gene Dgk1 on chrX, which is an upstream regulator of AChE and thus could modulate resistance to insecticides that target AChE. Fourth, the KEAP1 gene, involved in the regulation of the transcription factor MAF-S. Three further signals with a similar degree of evidence did not coincide with any gene with a known link to insecticide resistance.

Many further signals were present at lower levels of significance, and all data generated by this study will be released openly as a resource for further investigation of recent selection in malaria vector populations.

Microsatellite relatedness analysis reveals relationships between larval abundance and oviposition behavior within and among Anopheles arabiensis habitats

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**Background:** Strategies to control malaria vectors at the larval stage become increasingly important as we reach the limitations of primary interventions targeting only adults biting and resting indoors. Larviciding strategies would benefit from knowledge on habitat productivity and the proportions of female mosquitoes that oviposit at multiple breeding sites to allow improved targeting. To investigate these questions, we applied genetic markers to Anopheles arabiensis larvae from artificial and natural ponds to investigate relatedness within and among habitats.

**Methods:** In a controlled experiment, 18 artificial ponds were left uncovered for 4 days to allow oviposition by wild females, then covered for a further 6 days to prevent further egg laying; early instar larvae were sampled daily from day 5. Additionally, Anopheles natural habitats were identified, and larvae sampled. The larvae were PCR-identified to species and An. arabiensis samples genotyped using microsatellites. We used BAPS and COLONY softwares, to provide methodologically-independent reconstructions of sibling groups.

**Results:** In the ponds, COLONY identified more families than BAPS but results were well correlated (r=0.7). In both cases the number of families strongly predicted the total number of larvae (r2 $\approx$ 0.5), supporting our hypotheses that more larvae result from more females laying rather than simply better survival of an equivalent number of families. From BAPS results, 64% of females had deposited larvae in multiple ponds and from COLONY, 45%, suggesting a high frequency of skip oviposition behaviour. In the natural habitats, the number of families from both software strongly correlated (r=0.85) and positively predicted larval abundance (r2 $\approx$ 0.8). The overall average number of larvae per family was 2.4±0.84 larvae.

**Conclusion:** The sharing of habitat by multiple females and high skip-oviposition rate shown here suggest that autodissemination approaches for larval control may be successful.

### The MalariaGEN Vector Observatory: a platform for genomic surveillance of malaria vector populations

Authors: Alistair Miles<sup>1,2,</sup> Dominic Kwiatkowski<sup>2,1</sup>, Mara Lawniczak<sup>2</sup>, Martin Donnelly<sup>3</sup>, Sonia Goncalves<sup>2</sup>, Victoria Cornelius<sup>1</sup>, Bernard Abong'o<sup>4,</sup> Jean Akiana<sup>5</sup>, El Hadji Amadou Niang<sup>6</sup>, Lucas Amenga-Etego<sup>7</sup>, Cristina Ariani<sup>2</sup>, Victor Asoala<sup>7</sup>, Diego Ayala<sup>8,</sup> Nora Besansky<sup>9</sup>, Philip Bejon<sup>10</sup>, Austin Burt<sup>11,</sup> Beniamino Caputo<sup>12,</sup> Edi Constant<sup>13,</sup> Mamadou Coulibaly<sup>14</sup>, Samuel Dadzie<sup>15</sup>, Adama Dao<sup>14,</sup> Alessandra Della Torre<sup>12</sup>, Dziedzom de Souza<sup>15</sup>, Abdoulaye Diabaté<sup>16,</sup> Luc Djogbenou<sup>17,</sup> Alexander Egyir Yawson<sup>18</sup>, John Essandoh<sup>3,</sup> Ousmane Faye<sup>19,</sup> Michael Fontaine<sup>20</sup>, Nicodem Govella<sup>31,</sup> Maite Guardiola<sup>21</sup>, Jeremy Herren<sup>22</sup>, Jeffrey Hii<sup>23,</sup> Helen Irving3, Bilali Kabula<sup>24</sup>, Emmanual Kaindoa<sup>31</sup>, Jonathan Kayondo<sup>25,</sup> Brigid Kemei<sup>26,</sup> Lassana Konaté6, Tovi Lehmann<sup>27</sup>, Neil Longo Pendy<sup>28,</sup> Eric Lucas<sup>3,</sup> Mara Maquina<sup>32,</sup> Janet Midega<sup>10</sup>, Sandrine Nsango<sup>29</sup>, Eric Ochomo<sup>30</sup>, Fredros Okumu<sup>31</sup>, Samantha O'Loughlin<sup>11</sup>, Krijn Paaijmans<sup>32</sup>, Christophe Paupy8, Badara Samb<sup>6,</sup> Claire Sangba-Kembi<sup>33,</sup> Brandy St Laurent<sup>34</sup>, Brian Tarimo<sup>31</sup>, David Weetman<sup>3</sup>, Charles Wondji<sup>3</sup>, Alpha Yaro<sup>35</sup>

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**Background:** The use of genome sequence to study malaria vector populations by the Anopheles gambiae 1000 Genomes Project has led to a richness of new discoveries, particularly regarding the evolution and spread of insecticide resistance. A platform is now needed to scale-up genome sequencing, in order to catalyse further research and to enable genomic data to be integrated into entomological monitoring by vector control programmes.

**Methods:** We have established the MalariaGEN Vector Observatory, an open, collaborative platform, leveraging genome sequencing technologies to enable coordinated research and surveillance of malaria vector populations. Building on insights, technologies and best practices developed in the Anopheles gambiae 1000 Genomes Project, the Vector Observatory has been operating in pilot phase since 2017, and has established lab, data processing and analytical pipelines for whole genome sequencing of 10,000 mosquitoes per year, as well as developing new sequencing technologies that can scale to larger sample sizes and be deployed to regional facilities.

**Results:** We give an overview of current projects and partnerships with the Vector Observatory, the datasets generated so far, the model for participation, and the roadmap for future data generation and technology development. We illustrate the observatory with case studies of longitudinal surveillance of An. gambiae complex populations in West Africa, and with a broad geographical survey of An. funestus populations across Africa.

**Conclusions:** Genomic surveillance can generate new insights for insecticide resistance management (IRM), and provide feedback on the impact of IRM implementations on vector populations. It can also accelerate applied research and development of new vector control tools, particularly new insecticides, synergists, and gene drive systems, and provide rich data to investigate the many unknowns regarding the ecology and dynamics of vector populations. By providing an open platform, the Vector Observatory enables these activities to be led by researchers and control programmes in malaria endemic countries.

Evidence of multiple mechanisms providing carbamate and organophosphate resistance in field An. gambiae population from Atacora in Benin : a threat for malaria vector control.

Author: Rock AÏKPON\*, Martin AKOGBETO

**Background:** Insecticide resistance in Anopheles gambiae population is a major concern to malaria vector control. In West Africa, resistance is mainly due to target-site insensitivity arising from a single point mutation. Metabolic-based resistance mechanisms have also been implicated and are currently being investigated. This study aims to better understand the origins of carbamate and organophosphate resistance in An. gambiae population from Atacora in Benin.

**Methods:** Anopheles mosquitoes were reared from larvae collected in two districts (Kouandé and Tanguiéta) in Atacora department. Mosquitoes were then exposed to WHO impregnated papers. Four impregnated papers were used: carbamates (0.1% bendiocarb, 0.1% propoxur) and organophosphates (0.25% pirimiphos methyl, 1% fenitrothion). PCR assays were run to determine the members of the An. gambiae complex, as well as phenotypes for insensitive acetylcholinesterase (AChE1). Biochemical assays were also carried out to detect any increase in the activity of enzyme typically involved in insecticide metabolism (oxidase, esterase and glutathion-S-transferase).

**Results:** Bioassays showed resistance to bendiocarb (78.57% to 80.17%), propoxur (77.21% to 89.77%), and fenitrothion (89.74% to 92.02%). However, the same populations of An. gambiae showed high susceptibility to pirimiphos methyl with recorded mortality of 99.02% and 100% respectively. The low rate of Ace-1R allele frequency (3.75% among survivors and 0.48% among dead) added to the high proportion of homozygous specimens which survived, suggest that the Ace-1R mutation could not entirely explain Anopheles gambiae resistance to carbamate and organophosphate. Biochemical assays suggest that resistance in this population is mediated by metabolic resistance with elevated level of GST, MFO and NSE compared to a susceptible strain An. gambiae Kisumu.

**Conclusions:** Anopheles gambiae populations resistance from Atacora is multifactorial and includes target-site mutation and metabolic mechanism. The co-implication of both resistance mechanisms in An. gambiae s.l may be a serious obstacle for the future success of IRS programme that is curuntly using carbamate in Atacora region.

Keywords: Evidence, Multiple resistance mechanisms, Carbamate, Organophosphate, Anopheles gambiae, Benin

### Phenotypic insecticide resistance, correlation of 2La chromosomal inversion and sporozoite infection rates in Anopheles gambiae complex species (Diptera: Culicidae) in Teso, Western Kenya

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**Background:** Malaria vector control depends on the use of insecticide treated nets (ITNs) and indoor residual spray (IRS). Insecticides recommended for use on ITNs and IRS are mainly pyrethroids. Resistance to these commonly used pyrethroids has become a threat to efforts made towards malaria control. Mosquito chromosomal inversion (2La) is trait that is thought to influence resting behavior of mosquitoes. It is also thought to cause Anopheles mosquitoes resting indoors to withstand prolonged exposure to insecticide without being "knocked-down". These traits to withstand exposure to insecticide can make mosquitoes to become effective in malaria transmission.

**Methods:** Adult mosquitoes were collected from both indoors and outdoors using CDC light traps and aspiration. Larvae were also collected from water sources using standard plastic dippers. Adult Anopheles mosquitoes were morphologically identified and female Anopheles were sorted according to their physiological status as fed, gravid and unfed. Polymerase Chain Reaction (PCR) analysis was performed to differentiate between Anopheles member species and knock down resistance genotyping was done using real Time PCR by TaqMan assay. Head and thoraxes were analyzed for Plasmodium falciparum sporozoite infection and fed abdomens were screened for blood meal analysis through Enzyme Linked Immunosorbent Assays (ELISA). Susceptibility test was done on 3-5 day old emerged adult Anopheles following WHO 2013 guidelines.

**Results:** Anopheles gambiae constituted 45.47% and Anopheles arabiensis 54.53%. Overall phenotypic resistance observed was 11.29 %, in deltamethrin and 14.81% in permethrin respectively. Anopheles arabiensis were more resistant to deltamethrin 24.64% and permethrin 30.37% as compared to Anopheles gambiae 18.62% and 26.36% respectively. Overall sporozoite infection rate was recorded at 8.54%, and infection distribution per species was 3.96% Anopheles gambiae and 4.58% Anopheles arabiensis. Knock down resistance East allele mutation (L1014S) was recorded at 20% with no detection of Kdr West. The correlation of sporozoite infection and knock down resistance in Anopheles gambiae species was significant with Pearson's correlation coefficient (R=0.87, P=0.001). Chromosomal inversion was higher in homozygous 2La inversion arrangements (2La/a) with 23.33%. Blood meal results showed a higher prevalence in human and cat feeding with 33.87% and 20.65% respectfully, the rest of the hosts dog, goat, chicken and bovine recorded less than 10% prevalence.

**Conclusions:** Resistance to commonly used pyrethroids in the study area has been confirmed. Resistant mosquitoes to the currently used pyrethroids (deltamethrin and permethrin) are more likely to influence malaria transmission in the study area.

### A comparison of Irradiation dose response between a representative wild type Anopheles arabiensis strain against its modified genetic sexing strain

Authors: Eunice Jamesboy<sup>1,2,</sup> Leanne Lobb<sup>1,2</sup> and Lizette L. Koekemoer<sup>1,2</sup> and Givemore Munhenga<sup>1,2</sup>

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 <sup>2</sup> Vector Reference Laboratory, Centre for Emerging Zoonotic and Parasitic Diseases, National Institute for Communicable Diseases, National Health Laboratory Services, Johannesburg, South Africa.

**Background:** The success of any SIT programme is dependent on induction of sterility in laboratory-reared males targeted for release into wild populations. This is mainly through ionizing radiation using gamma rays. This study investigated the effect of irradiation on a wild-type strain of Anopheles arabiensis strain in comparison to the corresponding genetic sexing strain (GSS).

**Method:** Batches of male pupae were irradiated in a gamma irradiator. The results of the effect of irradiation on emergence, survivorship and reproductive fitness were evaluated and compared against GSS.

**Results:** Adult emergency rates in treatment and control cohorts ranged between 70 - 90%. Emergence rates in irradiated pupae decreased with increasing irradiation doses with no statistical difference between the treatments (Chi-square =7.39, DF =5, P >0.05). The mean survival time of emerged males, varied from 14d to 20 days with a significant difference in survivorship within the treatments and between treatments and control (Chi-square = 11.828, DF= 5, P = 0.037). Effect of gamma radiation on proxies of reproductive fitness revealed that the number of eggs laid by females inseminated with irradiated males ranged between 27.5 - 89.5% with significant different in number of eggs from the control cohorts (one-way ANOVA, F = 9.32; P = 0.04). Results on fertility showed significate difference in percentage of egg hatched between the treatment and control (One way ANOVA: F = 92.6; P < 0.05).

**Discussion/Conclusion:** Pre-exposure of the wild strain pupae to ionizing resulted in reduction to both adult emergence and their survivorship. Furthermore, fertility and fecundity of females mated with irradiated males was affected in comparison to previous findings in dose response in GSS, which were more pronounced at higher doses. This confirms that higher irradiation doses are not suitable to induce sterility in both strains due to somatic cells damage that ultimately compromises mating competitiveness

### New Chemistry For Irs : The Future Looks Bright

Authors: Lucas John Richard

**Background:** The heavy reliance on a handful of modes of action chemistry for IRS has resulted in widespread resistance to many of the classes used, especially to pyrethroids, with organophosphates often being the only effective IRS tool remaining. There are however worrying signs that resistance is now growing to organophosphates (WHO, 2015) threatening the viability of IRS. New chemistry with different modes of action are desperately needed to allow rotation and help preserve insecticide susceptibility.

SumiShield 50WG, containing the neonicotinoid clothianidin, was prequalified by WHO in October 2017 - making this the first WHO recommended new mode of action chemistry for IRS in 40 years. Programs now have a choice of effective chemistries which is critical for the long term future of IRS, as this allows the implementation of rotational strategies. SumiShield is now being deployed extensively throughout Africa.

**Methods:** This presentation will share recent lab and field efficacy data where SumiShield has been evaluated alongside other IRS products under a range of conditions. Results of resistance testing and operational and pilot trial experiences of using the product will also be discussed.

**Results:** Studies indicate excellent and long residual activity of SumiShield and widespread field susceptibility against a range of Anopheles species. The product was also well received in pilot tests.

**Conclusions:** While results are very encouraging for the continued future of IRS, we need to ensure IRS products with different modes of action are rotated to help preserve their effectiveness. Plans for operational monitoring of entomological efficacy, resistance and evaluation of epidemiological impact will be discussed.

## A question of logic: Should PBO combination LNs be used in areas where pirimiphos-methyl IRS (Actellic 300CS) control programmes are employed?

### Author: Aurélie Baillet

Piperonyl butoxide, PBO, is an inhibitor of mixed function oxidase (MFO) enzymes, also known as cytochrome P-450, and to a lesser extent PBO also inhibits the function of esterase enzymes. MFOs and esterases are among the main metabolic routes for the degradation of xenobiotic compounds, such as insecticides, in insects, and indeed all organisms. As such, inhibition of MFO can lead to the prolonged exposure of an insecticide's target site within an insect to that insecticide, which may synergise its activity, especially in insects that have developed elevated MFO resistance mechanisms.

With the widespread development of resistance to pyrethroid insecticides in the anopheline vectors of malaria, Long lasting insecticide treated Nets (LNs) have been developed that contain both a pyrethroid and PBO. The rational being, that the PBO will inhibit the metabolic resistance mechanisms, restoring a degree of susceptibility to the pyrethroid in the exposed mosquitoes. A PBO pyrethroid combination LN should therefore offer greater personal protection to the user. The addition of PBO does not however reduce the selection pressure for pyrethroid resistance. PBO combination LNs should therefore only be deployed as part of a carefully managed Insecticide Resistance Management (IRM) programme that includes non-pyrethroid interventions to mitigate against the potential to further impact the efficacy of the pyrethroid only nets currently in almost universal use. Actellic 300CS is a formulation of pirimiphos-methyl that has been specifically developed for vector control. Pirimiphos-methyl is a thiophosphate pro-insecticide that requires metabolic activation to the insecticidal organophosphate. This oxidative desulphuration of pirimiphos-methyl is mediated by MFOs, as such, inhibition of MFO activity can slow the activation of the thiophosphate to the organophosphate. In laboratory studies, the exposure of mosquitoes to PBO prior to exposure to pirimiphos-methyl does indeed slow down the onset of intoxication. However, this delay has no impact on the final mosquito mortality.

Should PBO combination LNs be used in areas where Actellic 300CS control programmes are in place? The WHO GMP suggests not in their 'Recommendations on the use of LLINs treated with a pyrethroid and a synergist: An update Malaria Policy Advisory Committee, Geneva, Switzerland, 16 March 2016'. Ultimately the question is one of logic. For a mosquito to pick up a sufficient dose of PBO from a combination LN to impact the speed of knockdown when it subsequently encounters an Actellic 300CS IRS deposit, it must have survived the exposure to the LN. If the mosquito population can survive exposure to synergised pyrethroid, then deployment of further pyrethroid based interventions should be avoided, and a non-pyrethroid intervention employed, as prescribed by good IRM practice, but more importantly, to protect the population from anopheline mosquitoes.

### Evidence of perennial malaria transmismission under arid conditions and dry season refugia for anopheline larvae: a case study at Kandi in northeastern Benin, West Africa?

#### Authors: Renaud Govoetchan\*, Martin Akogbéto

In arid settings, droughts usually lead to periods of very low or no malaria transmission. However, in rural Kandi (Sonsoro) in northeastern Benin, several malaria cases are often diagnosed during dry seasons. The underlying factors accounting for this phenomenon remain unknown. For our investigations, the entomo-parasitological profile of the location (Sonsoro) was compared to a site in urban Kandi (Gansosso), meticulously focusing on what happens in the dry season. A GIS approach was used to access in 1-year period the spatial and seasonal distribution of mosquito larval habitats and identify their drought-refugia. Conjointly, adult Anopheles vector collections were monthly performed using Human Landing Catches (HLC). Elisa assays for P. falciparum circumsporozoite protein were conducted on vector specimens and the entomological inoculation rates (EIR) were determined per season. To investigate drought-malaria prevalence, Rapid Diagnostic Tests (RDTs) were conducted in children < 10 years in both sites. Overall 187 mosquito larval habitats were identified of which 56 were recorded during the dry season (73% in rural site against 27% in the urban). The drought-refugia for mosquito breeding were all of domestic nature mainly canaries, jars, and flower pots. HLC provided 966 mosquitoes belonging to 12 species and Anopheles gambiae is the main species sampled (69 %). It represented respectively 94% (628/668) and 13 % (39/298) of the collections in rural and urban Kandi. From wet to dry season, we observed a drastic 96% reduction of EIR in rural Kandi (23 infected bites/man/month to 1 infected/ man/month). Same trend was observed in urban Kandi (1 infected bite/man/month to 0 with the drought). RTDs data on 400 children showed that P. falciparum infections were significantly higher in rural Kandi (41%) than in urban (7.5%). Our results suggest that a suitable domestic management of larval habitats in droughts would provide a huge impact on malaria transmission.

Key words: drought, malaria, Anopheles gambiae, refugia

### The genetic architecture of target-site resistance to pyrethroid insecticides in the African malaria vectors Anopheles gambiae and Anopheles coluzzii

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Resistance to pyrethroid insecticides is a major concern for malaria vector control because these are approved for use in insecticide-treated bed-nets and have also been widely used for indoor residual spraying, both important vector control methods. Pyrethroids target the voltage-gated sodium channel (VGSC), an essential component of the mosquito nervous system. Substitutions in the amino acid sequence can disrupt the activity of these insecticides, inducing a resistance phenotype. Here we discuss the use of Illumina whole-genome sequence data from phase 1 of the Anopheles gambiae 1000 Genomes Project (Ag1000G) to provide a comprehensive account of genetic variation in the Vgsc gene in mosquito populations from eight African countries.

In addition to the three known An. gambiae Vgsc 'kdr' resistance alleles (L995F/S, N1570Y), we describe 20 non-synonymous nucleotide substitutions at appreciable frequency in one or more populations that were previously unknown in Anopheles mosquitoes. Thirteen of these novel alleles were found to occur almost exclusively on haplotypes carrying the L995F resistance allele, and may enhance or compensate for the L995F phenotype. A novel substitution was also found in tight linkage with a substitution known to be associated with pyrethroid resistance in several other insect species.

By analysing the genetic backgrounds on which these non-synonymous alleles are found we discovered twelve distinct groups of similar haplotypes with evidence of recent positive selection, five of which carry the known L995F resistance allele, five which carry the known L995S resistance allele, and two others carrying novel substitutions. The geographical location of these groups provides new information about the distribution and spread of resistance.

We also illustrate how these data can be used to design high-throughput, low-cost genetic assays for improved surveillance of pyrethroid resistance in the field. Our results demonstrate that the molecular basis of pyrethroid resistance in malaria vectors is more complex than previously appreciated, and provide a foundation for the development of new genetic tools to track the spread insecticide resistance and inform vector control programmes.

### Impact of indoor residual spraying operational coverage on malaria transmission in South Africa

## Authors: Eunice Misiani<sup>1,2</sup> Patrick Moonasar<sup>1,2</sup>, Bheki Qwabe<sup>3</sup>, Eric Mabunda<sup>3</sup>, Aaron Mabuza<sup>3</sup>, Philip Kruger<sup>3</sup>, Achia Thomas<sup>4</sup>, and Andy Beke<sup>1</sup>.

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**Background:** Timely and correct delivery of vector control operations is crucial for the achievement of effective and high vector control coverage. The impact of indoor residual spray coverage and type of insecticide on malaria transmission, was carried out as part of a vector control needs assessment for endemic provinces in South Africa.

**Methods:** Retrospective data on targeted structures sprayed, insecticides used, municipality population, and local malaria cases in Limpopo, KwaZulu-Natal and Mpumalanga provinces, was reviewed for the 2010/11 to 2016/17 malaria seasons. A multilevel Poisson regression model was used (STATA statistical software version 14) to assess the impact of operational coverage, type of insecticide and malaria season on local malaria incidence.

**Results:** The unadjusted relative risk suggests that increased operational coverage is associated with a decline in malaria risk [RR=0.22, 95% CI (0.08-0.59]. Malaria risk was significantly higher in Limpopo and Mpumalanga compared to KZN [RR=28.69, 95% CI =13.98-58.86] and RR =13.86, 95% CI = 7.02-27.4] respectively. The amount of DDT used declined over the years whereas the use of pyrethroids increased within the same period. Malaria risk was significantly higher when deltamethrin was used compared to DDT [ARR=1.24,95% CI=0.99-1.55]. A non-linear relationship between malaria risk and different malaria seasons was noted.

**Conclusion:** Timely application of indoor residual spraying with effective insecticides reduces the risk of malaria transmission. DDT was more effective in reducing the risk of transmission but may not be appropriate in all settings. It is recommended that other insecticides/insecticide classes with a longer residual effect than deltamethrin should be utilised in areas where DDT cannot be sprayed. The effect of climatic and health systems challenges should be included in future models. These results can guide effective vector control as South Africa aims for elimination of local malaria transmission.

Key words: indoor residual spraying, insecticides, operational coverage, elimination

### Larvicidal Properties Of Pseudomonas Fluorescens Isolates Against Malaria Vectors In Kenya

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**Introduction:** Bio-control agents targeting mosquito larval stages offer renewed hope in mosquito control. Most biological agents act as stomach poison with high level of initial mosquito larval mortality but very little residual activity necessitating regular application increasing the cost of operation. This necessitates search for innovative mosquito control tools and enhance existing microbial agents. Amarathus plant species contain Pseudomonas fluorescens bacteria whose metabolites have insecticidal properties. The objective of this study was to evaluate the larvicidal properties of P. fluorescens isolates against Anopheles larvae.

**Methodology:** Pseudomonas bacterial was isolated from field collected Amaranthus hybridus (locally known as Mchicha) in Kilifi, Coastal Kenya. Pseudomonas bacteria were isolated under sterile laboratory conditions and then cultured on King B media. Bacterial culture supernatant was prepared into three concentrations ( $30\mu g/ml$ ,  $60\mu g/ml$  and  $90\mu g/ml$ ), introduced bioassay basins with 20 Anopheles gambaie mosquito larvae. Negative and positive control included untreated water and Bacillus thuriengiensis var. israelensis (Bti) respectively. Each treatment had five replicates and the experiment was repeated thrice. Larva survival rate was scored on a 12-hour interval. Residual activity was evaluated by reintroduction of mosquito larvae at varied intervals.

**Results:** Within 72-hours, 46.3% larval reduction was observed in 90 $\mu$ g/ml treatment followed by 29.8% in 60 $\mu$ g/ml and 24.6% in 30  $\mu$ g/ml concentrates. Total (100%) larval mortality was achieved 5-days post-treatment in 90 $\mu$ g/ml treatments while 60 $\mu$ g/ml and 30  $\mu$ g/ml treatments complete larval mortality was achieved 6-days post-treatment. All larvae reintroduced in 90 $\mu$ g/ml treatments 7-days post-treatment died within 48-hours while those in 60 $\mu$ g/ml and 30  $\mu$ g/ml concentrates recorded at 93% and 71% respectively within same period.

**Conclusions:** Pseudomonas fluorescens metabolites from Amaranthus plants are efficacious against Anopheles mosquitoes providing a promising tool in malaria vector control.

Key words: Pseudomonas fluorescens, Anopheles, malaria, biocontrol.

### Comparative efficacy of SumiShield – a Clothianadin based Indoor Residual Spray measured through a parallel experimental hut and cluster randomized entomological trial

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**Abstract:** Given the limited epidemiological data for IRS (indoor residual spray) linking entomological surrogates to epidemiological impact, the current standard is to rely on data from experimental hut studies with a study design that is properly powered and randomized with adequate replication. An experimental hut and cluster-randomised trial of entomological outcomes were conducted in parallel over eight months to compare the methods for evaluating the efficacy of SumiShield\* 50WG a clothianidin based IRS in southern Tanzania.

The hut study was single-blinded, partially randomized, Latin square evaluation with five treatment arms (four IRS products, one negative control) in 10 huts. Outcomes were immediate and delayed vector mortality (up to 168 hours), blood feeding inhibition, insecticide residual activity, deterrence and induced exophily. The community study was single-blinded with two IRS treatment arms and five clusters per arm of around 200 households/cluster with clusters 2 km apart. The treatment arms were SumiShield and Actellic<sup>®</sup> 300CS. Outcomes were residual and biological efficacy of insecticide, vector density, vector mortality and infectivity rate (sporozoite rate). Primary vector Anopheles funestus and secondary vector An. arabiensis are resistant to all classes of pyrethroids and susceptible to pirimiphos methyl.

Vector mortality (caught by indoor and outdoor resting catches and held for 168 hours) was greater in the SumiShield arm than the Actellic arm of the village trial for both An. funestus and An. arabiensis. Odds ratio (excess mortality) for all 8 months of observations was 4.30 (95% CI 1.70-10.30) p=0.0002 for An. funestus and 1.60 (95% CI 1.03-2.48) p=0.036 for An. arabiensis. This agreed well with data from experimental huts that demonstrated Odds ratio (excess mortality) for all 8 months of observations was 2.01 (95% CI 0.93-4.33) p=0.075 for An. funestus and 1.28 (95% CI 1.13-1.46) p<0.0001 for An. arabiensis.

Data demonstrate that experimental hut studies are a useful substitute for cluster randomised village scale trials for entomological studies of new IRS compounds.

The Use Of Bayesian Spatio-Temporal Modelling To Determine The Impact Of Ulv Applications On Us Mosquito Populations: Lessons For African Mosquito Control

Authors: Daniel McDermott, Michelle Stanton, Mark Latham & David Malone

**Background:** While much of malaria transmission occurs indoors, the need to halt transmission outdoors is vital for achieving elimination goals. Outdoor ULV spraying for Anopheles spp. has been used previously by malaria control programmes in Haiti, India, Turkey and Sri Lanka. However, the current view on the impact of ULV spraying on malaria transmission differs widely within the research community and there is a lack of peer-reviewed evidence for or against this intervention. A recent study in Burkina Faso has shown that the spraying of outdoor male mating swarms can have a drastic impact on the size and the age-structure of the population. Due to the absence of well-designed observational studies on this topic, a collaboration was established with a US mosquito control programme that actively employs aerial ULV spraying as an intervention tool to review their ongoing use of this intervention and its impact on adult mosquito count data.

**Methods:** Seasonal adult mosquito count data (2012-2016) from Manatee County mosquito control district, Florida, was used to carry out a spatio-temporal Bayesian analysis (R-INLA). The models were created using weekly count data, environmental variables and the programmes intervention history to examine the association that their ongoing aerial spraying programme had on the adult population over time.

Results: This analysis is ongoing and is scheduled to be completed by July 2018.

**Preliminary Outcomes:** This approach allows us to correlate changes in the mosquito populations over space and time to the intervention that is being employed. Aerial ULV can produce a rapid reduction of the mosquito population in the short term when successfully targeted to the mosquito's bionomics. Although the setting and the vector populations may differ, we argue that there are aspects of US mosquito control programmes that can be effectively transferred to a malaria elimination setting.

### Impact of hole size, hole location, insecticide content and mosquito pyrethroid-resistant on the protective efficacy of long-lasting insecticidal nets.

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**Background:** Physical integrity and insecticide concentration are the keys parameters that ensure useful effective life of long lasting insecticidal nets (LLINs). Durability studies collect data on measures of physical integrity, attrition, bio-efficacy and insecticidal content. However, there is limited information on the impact of physical integrity (in particular, hole size and location) and its interaction with insecticide concentration on the personal and community protection in the field.

**Methods.** Nets that were artificially holed (total hole surface area 0–5800cm2) and treated with several different deltamethrin insecticide concentrations (0, 5, 15, 25 and 55mg/m2) were tested overnight in the newly developed Ifakara Ambient Chamber Test (I-ACT) to determine their efficacy against either released Anopheles gambiae s.s. susceptible to pyrethroids, and low (80% mortality with deltamethrin in standard WHO susceptibility tests) and high resistant An. arabiensis.

**Results.** "Too torn" insecticidal nets give 16 times more protection than equivalent "too torn" untreated nets even with moderately pyrethroid resistant mosquitoes. Results from 287 sampled nets revealed that tucking a net increased the protective efficacy by reducing mosquito blood feeding success to 2.22% (95% C.I. 1.5–2.9) as compared to over 20 percent when untucked against fully pyrethroid susceptible mosquitoes.

**Conclusions.** Both physical integrity and insecticidal content are important LLIN properties for personal and community level protection from malaria. Behavioral change communication is of major importance to the durability of nets since tucking nets dramatically improves their performance and even insecticidal holed nets will continue to provide personal and community level protection.

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The mosquitoes are preparing to attack us: knowledge and perceptions of Anopheles mosquito swarms in south-eastern Tanzania

Authors: Marceline F. Finda, Emmanuel W. Kaindoa, Anna P. Nyoni, Fredros O. Okumu

**Background:** Targeting swarms of male mosquitoes with aerosol spraying could potentially suppress malaria vector populations and parasite transmission. Unfortunately, research on Anopheles swarms is limited, particularly in East Africa where only a handful of studies have been done. New evidence has recently emerged that such swarms are common even in Tanzania, where they could be readily identified by community-based volunteers, characterized and potentially targeted for control. However, understanding public knowledge and perceptions will be crucial for successful uptake of any interventions targeting swarms.

**Methods:** We used an explanatory sequential mixed methods approach to assess knowledge and perceptions regarding Anopheles swarms among community members and volunteers participating in mosquito research programs in Ulanga and Kilombero districts, Tanzania. A survey questionnaire was administered to 307 respondents, after which focus group discussions (FGDs) were conducted to clarify responses regarding mosquito swarms and malaria transmission. Findings from both strands were used to make inferences.

**Results:** Most community members (83%) and the volunteers (93%) had previously seen mosquito swarms, predominantly in farms, long grasses or bushes, ponds and on roofs of houses and pit-latrines. However, there was little evidence that community members could distinguish between swarms of malaria mosquitoes and those of other insects or other mosquito species. Neither were they aware that swarms consisted mostly of male mosquitoes. The research volunteers however had higher awareness of mosquito swarms than other community members. Swarming was associated with mosquitoes preparing to attack people, foraging for food, playing or resting, but hardly with mosquito mating. Nearly all community members were willing to accept interventions targeting mosquito swarms; and approximately three quarters would pay for such interventions, between 0.9-2.3 USD/year.

**Conclusion:** Majority of the community members recognized presence of mosquito swarms in their communities but did not associate these swarms with mosquito mating. Instead, swarming was associated with mosquitoes seeking food or planning to attack people, a belief that increased risk perception, and subsequently acceptance of interventions against swarming mosquitoes. Volunteers previously trained to support swarm searching and spraying operations had higher knowledge on the swarms, confirming they could be relied upon to support interventions targeting swarms.

Piloting of an entomological surveillance planning tool to improve entomological intelligence for evidence-based vector control decision-making towards malaria elimination in Mozambique, Namibia, and Panama

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Abstract: To accelerate progress towards malaria elimination, the World Health Organization (WHO) Global Technical Strategy 2016-2030 calls for maximizing the impact of vector control by strengthening entomological surveillance and capacity and managing insecticide resistance and residual transmission. In response to these efforts, we have developed an Entomological Surveillance Planning Tool (ESPT) to distil WHO guidance into an operational, decision-support tool for national malaria programs to support cost-effective, locally tailored, evidence-based vector control. The ESPT aims to support countries in generating entomological intelligence that guides vector control intervention selection, deployment in time and space, and provides a platform to evaluate complementary strategies and tools. To this end, the ESPT consists of a series of decision trees to help guide countries in the collection of the priority entomological indicators needed to make decisions through: 1) baseline surveys, 2) routine sentinel surveys, 3) focus investigations, and 4) entomological surveys based on priority programmatic questions. These decision trees link to the priority minimum indicators, a trapping methodology matrix to guide collections, data collection forms, and guidance on selecting sites for entomological investigations. In collaboration with national malaria programs and local partners, pilots of the ESPT have begun in three countries: Mozambique, Namibia, and Panama. Parallel evaluation activities are underway and include: 1) qualitative assessments to measure the ESPT's feasibility, acceptability, utility, and impact on vector control program decision-making; 2) costing of entomological surveillance activities and the cost per indicator; and 3) tracking decision-making on vector control strategy as related to implementation of the ESPT. Preliminary data from the pilot evaluations will be available by mid-2018 and will inform future iterations of the ESPT for malaria programs.

Changes in insecticide resistance in laboratory-reared Anopheles gambiae s.l. mosquitoes from Accra, Ghana

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**Background and Objective:** Vector control remains one of the most effective malaria prevention strategies. Research laboratories require mosquito colonies for studies on vector biology, insecticide susceptibility, and testing of innovative vector control tools. Insecticide susceptible An. gambiae s.s. Kisumu is the standard testing strain. However, with the spread of insecticide resistance across Africa, testing with wild, resistant mosquitoes is increasingly required. Ideally, for consistency of experiments, a laboratory colony should maintain stable physiological and behavioral characteristics including level of insecticide resistance.

**Materials and methods:** Between February to October 2017, mosquito larvae were collected from Opeibea, a vegetable growing area in Accra, Ghana, and reared and maintained in the lab. WHO susceptibility tube tests were used to monitor insecticide resistance status of mosquitoes from F0 to F13 generation. Vgsc-1014F and Ace-1 frequencies were determined by PCR.

**Results:** Mosquitoes at the F0 were resistant to all four classes of public health insecticides. Mortality against deltamethrin and permethrin was 2.5% and 0%, respectively. By the F5 generation mortality against deltamethrin had increased to 59.1 and 62.5%, respectively, and continued to increase with successive generations. Resistance to carbamates and organophosphates was less variable across generations; for instance, mortality against bendiocarb ranged between 0 and 4.7%. Vgsc-1014F and Ace-1 mutation frequencies were 1.0 and 0.8 at F0, and 0.42 and 0.68 at F8, respectively.

**Discussion and conclusion:** Wild mosquitoes reared in laboratory conditions were observed to lose their resistance to insecticides over time, likely due to reduced selection pressure. This is a challenge for the maintenance of resistance mosquito colonies in the lab. This study is still ongoing and will aim to explore appropriate selection procedures that can help maintain insecticide resistance levels of resistant An. gambiae colonies.

### An assessment of mosquito collection techniques for xenomonitoring of anopheline-transmitted Lymphatic Filariasis in Ghana

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**Abstract:** Monitoring vectors is relevant to ascertain transmission of lymphatic filariasis (LF). This may require the best sampling method that can capture high numbers of specific species to give indication of transmission. Gravid anophelines are good indicators for assessing transmission due to close contact with humans through blood meals. This study compared the efficiency of an Anopheles gravid trap (AGT) with other mosquito collection methods including the box and the Centres for Disease Control and Prevention gravid, light, exit and BioGent-sentinel traps, indoor resting collection (IRC) and pyrethrum spray catches across two endemic regions of Ghana. The AGT showed high trapping efficiency by collecting the highest mean number of anophelines per night in the Western (4.6) and Northern (7.3) regions compared with the outdoor collection methods. Additionally, IRC was similarly efficient in the Northern region (8.9) where vectors exhibit a high degree of endophily. AGT also showed good trapping potential for collecting Anopheles melas which is usually difficult to catch with existing methods. Screening of mosquitoes for infection showed a 0.80–3.01% Wuchereria bancrofti and 2.15–3.27% Plasmodium spp. in Anopheles gambiae. The AGT has shown to be appropriate for surveying Anopheles populations and can be useful for xenomonitoring for both LF and malaria.

Putative impact of Asaia aff. bogorensis GD01 on Plasmodium falciparum sporogonic development in Senegal

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**Background:** The declining of malaria creates the opportunity to accelerate its elimination. However, the Elimination program is threatened by the spread of insecticide resistance among Anopheles mosquitoes, stressing the urgent need for new vector control strategies. Recent evidence of stable natural infection of anopheles' symbionts, that could affect the development of Plasmodium parasites within mosquitoes, has paved the way for the use of such organisms to target and control malaria vector populations.

**Methods:** Asaia GD01 strain was isolated using a standard protocol from wild specimen of An. gambiae collected form Dielmo (Senegal). Asaia infection was screened using a costume qPCR method on wild populations of An. funestus and An. gambiae s.l. collected in two Senegalese regions, and on an insectary colony of An. coluzzii. Further phylogenetic analysis based on the 16S rDNA was used to infer taxonomic relationships of A. aff. bogorensis with existing isolates. The putative negative correlation between A. aff. bogorensis and Plasmodium was assessed and compared between species.

**Results:** We have isolated a new strain of Asaia from wild population of An. gambiae s.l. and, for the first time, from natural population of An. funestus, and insectary colony of An. coluzzii. The new anopheline strain has been named Asaia aff. bogorensis GD01 because of its close relationship with A. aff. bogorensis isolated from plant flowers. We highlighted an antagonistic effect of A. aff. bogorensis GD01 on the sporogonic development of P. falciparum within the two major malaria vectors (*An. gambiae and An. funestus*) in Senegal.

The putative impact of A. aff. bogorensis GD01 infection on the *Plasmodium falciparum* sporogonic development offers an opportunity for new malaria vector control approaches that can be added to the limited arsenal required to fulfil the WHO recommendations for searching new tools to achieve malaria elimination goal.

**Conclusions:** Although this is an innovative and promising malaria control tool, more investigations are required to better characterize Asaia vs Plasmodium interactions, before any Asaia-based intervention. This is required to ensure their safe use as an alternative or complementary vector control strategy to achieve malaria elimination goal.

Assessing the susceptability of Anopheles arabiensis to Plasmodium falciparum infections in western Kenya using an optimized membrane feeding protocol

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**Background:** Anopheles arabiensis, the more opportunistic sibling of Anopheles gambiae s.s is a major malaria vector in Kenya, in addition to Anopheles funestus. An. arabiensis might have a lower vectorial capacity than An. gambiae s.s since it is a more exophilic vector with more ready acceptance of animal blood meals but the question remains if it is as easily infected with gametocyte infected blood and how this might affect its fitness. This is the first report on the use of the membrane feeding approach to assess the fitness of An. arabiensis to P. falciparum infection in relation to An. gambiae s.s.

**Methods:** The study was conducted in Mbita, a semi-arid area on the shores of Lake Victoria in western Kenya. Screenhouse reared An. arabiensis and An. gambiae s.s insectary mosquitoes were concurrently challenged with gametocyte infected P. falciparum blood from the same carrier using a glass-parafilm membrane feeding system. We recorded the feeding rate, mortality rate and sporozoite rates for surviving mosquitoes at day 11.

**Results:** In six paired experiments, 52% consisting 847 An. arabiensis and 715 An. gambiae s.s mosquitoes were fully fed upon exposure. The average feeding rate was 56.1% (range 47.5-63.7%) for An. arabiensis and 48.1% for An. gambiae s.s (range 24.0-74.3%). We observed comparable sporozoite rates for both species, although sporozoite rates were significantly affected by gametocyte density (p<0.001) for An. gambiae s.s. Overall, there was high mortality rate in An. gambiae s.s compared to An. arabiensis supporting the fitness premise of An. arabiensis.

**Conclusions:** Our results show that An. arabiensis is as efficient a vector as An. gambiae s.s. in relation to Plasmodium falciparum malaria transmission. Vector control methods need to target the outdoor An. arabiensis since they are equally susceptible to infection and have a higher chance of survival compared to An. gambiae s.s.

Impact of glutathione S-transferase mediated metabolic resistance to insecticides on the mating competitiveness of field resistant Anopheles funestus

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**Background:** The extent to which metabolic resistance affects the reproduction of malaria vectors such as An. funestus remains under-investigated because of the absence of DNA-based resistance marker. To investigate how insecticide resistance impacts the mating male competiveness of malaria vectors, we compared the mating ability of homozygote resistant mosquitoes to that of the homozygote susceptible and heterozygote mosquitoes from the mating swarms of Anopheles funestus s.s collected in Cameroon.

**Methods:** Anopheles funestus swarms were surveyed in Tibati, Cameroon between February and March 2018. WHO bioassays were used to characterise the susceptibility profile of the female populations, randomly collected indoor to insecticides. Mosquitoes collected were molecularly confirmed as Anopheles funestus s.s using a cocktail PCR and a new AS-PCR performed to genotype the L119F-GSTe2 mutation. Association between the GSTe2 mutations and mating success was assessed by calculating the odds ratio of mating between homozygous resistant, heterozygotes and homozygous susceptible mosquitoes in mated males compared to unmated group with statistical significance based on the Fisher's exact probability test

**Results:** Genotyping of L119F-GSTe2 mutation between mated and unmated males from the swarms revealed that the heterozygote genotype (RS) showed a significantly lower chance of being mated than homozygote resistant and homozygote susceptible mosquitoes indicating a heterozygote disadvantage effect. Despite the very low sample size, homozygote susceptible males were more competitive than homozygote resistant suggesting a negative impact of L119F-GSTe2 on An. funestus mating ability. In addition, An. funestus and An. gambiae s.l collected indoor showed high Plasmodium infection rate and multiple resistance to major public health insecticide classes tested.

**Conclusion**: For the first time, we observed in field that metabolic resistance mechanisms exerts a fitness cost on mating competiveness. Such impact can allow an implementation of future insecticide resistance management strategies which could help to manage the resistance in malaria vectors.

## High mosquito burden and pyrethroids resistance in Culex quinquefasciatus populations in the city of Yaoundé, Cameroon

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**Background:** Culex species are important source of nuisance for populations in sub-Saharan Africa major cities and vectors of diseases such as filariasis and arboviruses yet there is not enough information on this genus. We report about entomological studies conducted in the city of Yaoundé, Cameroon on Culex distribution and susceptibility to pyrethroids.

**Methods:** Mosquitoes were collected using CDC-Light Trap (CDC-LT) and Human Landing Catch (HLC) in human dwellings once every two months from march 2017 to march 2018. Bioassay were conducted with 2-5 days old females to assess their susceptibility to permethrin 0.75% and deltamethrin 0.05%. Mosquito were identified using morphological identification keys.

**Results:** A total of 226,127 mosquitoes were collected, and Culex species were the most abundant with 95.96% (N=216,999). Seven different Culex species were recorded. Culex quinquefasciatus emerged as the most abundant with 85.21%. Others mosquito genera identified were Anopheles (3.39%; N=7,673), Mansonia (0.51%; N=1,162); Aedes (0.12%; N=279), and Coquillettidia (0.006%; N=14). The Culex biting rate varied from 79.86 b/p/n indoor to 85 b/p/n outdoor. High seasonal variability in Culex densities were recorded using both CDC light trap and Human landing collections with high densities collected during the rainy season. High variability in Culex species distribution was also recorded. Culex quinquefasciatus as well as the other Culex species displayed high resistance to both permethrin and deltamethrin with levels of mortality rate varying from 11.5% for deltamethrin to 31.5% for permethrin.

**Conclusion:** The present study suggests high distribution of Culex species and perfect adaptation of this species to the urban environment and stresses the need for more studies in order to determine the epidemiological role of these species in the urban environment.

Key words: mosquito nuisance, pryrethroids resistance, Culex quinquefasciatus, Yaoundé

Urbanisation as a driver for Aedes aegypti density indices, risk of yellow fever and dengue virus transmission and their socio-ecological factors in Côte d'Ivoire

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**Background:** Our ability to deal with Aedes mosquito-borne viral epidemics is limited. We explored the impact of urbanisation on the risk of yellow fever (YF) and dengue (DEN) virus transmission assessing entomological indices in Ae. aegypti and socio-ecological factors across an urban-to-rural gradient within disease foci in Côte d'Ivoire.

**Methods:** Aedes eggs, larvae and pupae, household and water-container socio-ecological data were collected in urban, suburban and rural areas using standard procedures. Surveys were conducted during long dry (LDS), long rainy (LRS), short dry (SDS) and short rainy (SRS) seasons from January 2015 to December 2016. Ae. aegypti positive ovitrap index (POI), Stegomyia indices (house index: HI, container index: CI and Breteau index: BI) and pupal indices (pupae/container: PC, pupae/house: PH and pupae/person: PP) were compared.

**Results:** Overall, the risk of YF and DEN virus transmission was above the conventional thresholds in urban and suburban areas. POI was higher in urban (53.5%) compared to suburban (43.1%) and rural (29.7%). Stegomyia indices were highest in urban followed by suburban and rural areas, with respective HI of 67.2, 51.4 and 27.8, CI of 57.1, 43.7 and 19.7, and BI of 37.2, 18.9 and 4.8. PC, PH and PP were higher in urban (3.3, 2.4 and 1.9) than suburban (1.3, 0.6 and 1.1) and rural (1.3, 0.9 and 0.5) areas, respectively. All entomological risk indices were higher during LRS in all areas, with stronger magnitude in urban areas. They were associated with human activities and behaviours including water storage practices and solid waste management. Tires and discarded containers in all areas and water storage receptacles in urban areas were the most Ae. aegypti productive breeding sites.

**Conclusions:** In Côte d'Ivoire, Aedes aegypti density indices increased under socio-ecological determinants along urbanisation gradient and resulted in higher YF and DEN virus transmission risk above the conventional epidemic thresholds in urban and suburban, with greater magnitude in urban areas. Integrated community-based vector control programs should be applied.

Prevalence of Plasmodium falciparum transmission reducing immunity among primary school children in a malaria moderate transmission region in Zimbabwe

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Abstract: Malaria continues to cause alarming morbidity and mortality in more than 100 countries worldwide. Antigens in the various life cycle stages of malaria parasites are presented to the immune system during natural infection and it is widely recognized that after repeated malaria exposure, adults develop partially protective immunity. Specific antigens of natural immunity represent among the most important targets for the development of malaria vaccines. Immunity against the transmission stages of the malaria parasite represents an important approach to reduce malaria transmission and is believed to become an important tool for gradual elimination of malaria. Development of immunity against Plasmodium falciparum sexual stages was evaluated in primary school children aged 616 years in Makoni district of Zimbabwe, an area of low to modest malaria transmission. Malaria infection was screened by microscopy, rapid diagnostic tests and finally using nested PCR. Plasma samples were tested for antibodies against recombinant Pfs48/45 and Pfs47 by ELISA. Corresponding serum samples were used to test for P. falciparum transmission reducing activity in Anopheles stephensi and An. gambiae mosquitoes using the membrane feeding assay. The prevalence of malaria diagnosed by rapid diagnostic test kit (Paracheck)<sup>™</sup> was 1.7%. However, of the randomly tested blood samples, 66% were positive by nested PCR. ELISA revealed prevalence (64% positivity at 1:500 dilution, in randomly selected 66 plasma samples) of antibodies against recombinant Pfs48/45 (mean A405 nm = 0.53, CI = 0 .46 to 0.60) and Pfs47 (mean A405nm = 0.91, CI = 0.80to 1.02) antigens specific to the sexual stages. The mosquito membrane feeding assay demonstrated measurable transmission reducing ability of the samples that were positive for Pfs48/45 antibodies by ELISA. Interestingly, 3 plasma samples revealed enhancement of infectivity of P. falciparum in An. stephensi mosquitoes. These studies revealed the presence of antibodies with transmission reducing immunity in school age children from a moderate transmission area of malaria, and provide further support to exploit target antigens such as Pfs48/45 for further development of a malaria transmission blocking vaccine.

## Efficacy of Bacillus thuringiensis serovar israelensis and Temphos on Larvea of Anopheles arabiensis under selected temperature regimes

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**Background:** Anopheles arabiensis is the major vector of malaria in Africa. Targeting mosquitoes with insecticide residual spraying (IRS), insecticide treated nets (ITN) and long lasting insecticide treated nets (LLINs) has become the most effective way of preventing malaria transmission. The development of insecticide resistance to all the four classes of insecticides approved by WHO for IRS threatens the global malaria control program and climate change may worsen this. Larviciding has complemented these two methods. Among the larvicides agents available to control this specie, are biolarvicides based on Bacillus thuringiensis serovar israelensis (Bti) and the conventional insecticides like temephos (organophosphate). It is well known that temperature influences insecticide sensitivity but the extent to which this happens naturally in the context of climate change is not known.

**Methods:** Anopheles gambiae colony was established under insectarium conditions. Larval susceptibility to different concentration of temephos (2.5mg/L, 0.25mg/L, 0.025mg/L & 0.0025mg/L) and Bti (1mg/L, 0.1mg/L, 0.01mg/L & 0.001mg/L) was evaluated under different temperature regimes (15°C, 20°C, 25°C 30°C, 35°C, 40°C) through bioassays. Univariate analysis was used to assess mortality rates among different temperature categories. Association between temperature and mortality rates were measured using regression analysis. Proportional analysis was used to compare mortality rates between the two insecticides.

**Results:** Effect of temperature on mosquito larvae mortality rates was not clear in higher concentrations. There was a positive association (R2 > 0.69) between temperature and mortality rates in lower insecticide concentrations for both Bti and temphos. Higher temperatures were needed to kill 50% and 95% of mosquito larvae in Bti than in temphos in lower insecticide concentrations.

**Conclusions:** Data from this study show that temperature clearly influences insecticide toxicity in lower insecticide concentrations. Thus climate change may not change the burden of malaria if temperature increases as predicted.

Keywords: Vector control, Bti, Susceptibility, Temephos, insecticide toxicity, temperature, climate change

### Ifakara Ambient Chamber Test (I-ACT) - a high throughput means of measuring bednet efficacy

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While collecting data on net durability in the field can provide data on damage and attrition, this information is only truly useful if we can understand when nets stop providing adequate malaria protection to inform decision makers for net replenishment campaigns. This information needs to be simple to collect, cost effective and reproducible for use in multiple countries.

New bioassays have been developed to measure personal protection and mosquito mortality (community protection) with high throughput and data power. A WHO bednet evaluation to be conducted using the I-ACT in just 20 days with no risk of disease for human participants. Two experiments to demonstrate the utility of I-ACT are presented.

Whole Long lasting insecticidal nets (LLINs) are tested in an ambient chamber test with a human volunteer sleeping beneath to measure the protective efficacy under user conditions. Mosquitoes reared in the laboratory are released at 9pm and collected in the morning using aspiration and the mosquitoes are held for 24 hour mortality. The design of the chambers allows 100% recovery of released mosquitoes that gives remarkable data power and experiments can be conducted year round.

Mosquitoes of varying resistance status were released on the same night by marking strains with fluorescent dye. Data demonstrated the importance of mosquito resistance to pyrethroids on their entry through net holes and feeding success (personal protection) but not their mortality (community protection) when tested against LLINS with different levels of damage. Comparison of nets tested in experimental huts in three WHO sponsored studies run in parallel to a comparison with I-ACT demonstrated agreement between IACT and experimental hut studies from Cote d'Ivoire and Tanzania with alpha-cypermethrin + PBO (incorporated into polyethylene) LN and alpha-cypermethrin coated LNs. We therefore propose the I-ACT as an effective alternative or supplement to experimental hut studies and WHO tunnel tests for non-inferiority LLIN evaluations and longitudinal durability studies.

### Spatiotemporal Characterization of the Malaria Outbreak in Cape Verde in 2017.

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Cape Verde a country in pre-elimination stage of malaria continues reporting cases. Santiago the biggest island houses more than 50% of resident national population and where Praia, the capital is located. Praia registered in 2017 an outbreak of malaria, situation not verified in the last tens of years. This study aims to examine the epidemiologic and spatiotemporal profile of malaria in Cape Verde and to analyse the risk factors using the 2017 data of malaria. Local and space clusters of malaria in Praia were detected by applying the Spatial Autocorrelation and a Pearson correlation coefficient used between malaria cases and meteorological variables to identify underlying risk factors.

There was a total of 446 cases of malaria registered in Cabo Verde in 2017, used in the research, being 423 indigenous by Plasmodium falciparum all with origin in Praia. Were registered 23 imported cases of Plasmodium falciparum from African countries and one case of Plasmodium vivax from Brazil. We founded high-high cluster of malaria cases in the centre of the city with a correlation coefficient r=0.38 between breeding sites and malaria cases. The correlation (R2=0.699) show that 69,9% cases are due the environmental condition. The temperature has a strong positive correlation malaria cases (79.66%) and relative humidity a positive correlation (39.48%). A moderate positive correlation (28.61%) with the total pluviometry (mm) and the wind Speed had a strong negative correlation -54.4% with case.

The high numbers of cases in 2017 demonstrates the fragility and challenges that the country has face the elimination of indigenous cases and management of imported cases. The good capacity in cases management resulted in the low death. Politics and strategies needed to reinforce the individual and disease prevention, specially the environmental management, should be more stringent in Praia, to control malaria and achieve the elimination.

Prediction of malaria mosquito species and population age structure through mid-infrared spectroscopy and machine learning

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**Background and Objective:** Determining mosquito species distribution and age structures is essential to implement vector control, as these influence the intensity of disease transmission. Nevertheless, current methods to measure these traits are not practical for large scale entomological surveillance as they are expensive, time consuming and/or not accurate. Here we will present our work in the development of a rapid and cost effective tool based on mid-infrared spectroscopy (MIRS) and machine learning analysis to simultaneously determine species and age in malaria vectors.

**Materials and Method:** We have measured the amount of light absorbed by the mosquito cuticle through MIRS, which provides information on its biochemical composition. As cuticular composition varies between species and changes during mosquito ageing, we used MIRS information to predict these traits. To disentangle the complexity of the MIR spectra, we used several machine learning algorithms to predict these traits. We measured three morphological identical mosquito species in the Anopheles gambiae s.l. complex (An. gambiae, An. coluzzii and An. arabiensis) over the first 2 weeks upon emergence into female adults. Different physiological conditions have been included to incorporate biological variation.

**Results and Discussion**: Here we will present the age and species prediction model comprising mosquito spectra from laboratory and field sites. Initial analysis conducted on MIRS spectra from laboratory reared mosquitoes at University of Glasgow suggest that we can obtain more than 80% accuracy in species prediction and that we can accurately reconstruct age structures pre- and post- a control intervention. Increased sample sizes and more sophisticated machine learning analysis will be able to further increase the accuracy of this approach.

**Conclusion and Recommendation**: These results suggest that MIRS is a robust method for the simultaneous prediction of cuticular associated traits such as species and age; further work will include additional characteristics such as malaria infection status and cuticular insecticide resistance. This approach is easy-to-use, cost-effective and high-throughput. We envision that this technology can revolutionize vector surveillance programmes. In addition, this approach can be upgraded for other medical important vectors, including other mosquitoes, sand flies, and other vectors.

Preliminary Test For An Association Between Dieldrin Resistance And 2la Inversion Polymorphism In Anopheles Coluzzii From Lagos, Nigeria

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**Background:** Insecticide based interventions remain the cornerstone of malaria vector control in Nigeria. Resistance to these insecticides has been established in local populations of major malaria vector species of the Anopheles gambiae s.l. complex across the country. A major adaptive paracentric inversion - 2La – has previously been shown to assort with resistance to dieldrin in several laboratory colonies of Anopheles gambiae s.s. including one that originates from Southern Nigeria (Ian P20, 1979). This association is postulated to stabilize the resistance phenotype in the absence of selection by linkage to a strongly positive heterotic system. The aim of this project was therefore to test the association of 2La assortment with dieldrin resistance in a field population of Anopheles coluzzii from an area under intense malaria vector control in Nigeria.

**Methodology:** Immature stages of Anopheles mosquitoes were collected from Kosofe Local Government in Nigeria and reared to adults. Emergent adults were subsequently identified to species using morphological parameters and PCR. Samples of three to five day old adult females were exposed to 4% dieldrin insecticide papers according to the WHO insecticide susceptibility test. Knockdown was recorded at 10 min intervals for 1 hour and final mortality after 24 hours. Inversion 2La PCR was then conducted on the survivors and dead mosquitoes. KDT50 and KDT95 were derived using SPSS version 23 while the inversion karyotype results were subjected to Hardy Weinberg at P=0.05.

**Results:** The adult female mosquito population was highly resistant to 4% dieldrin (17.3% knock down and 23% final mortality) with KDT50 and KDT95 calculated as 170 and 1 514 minutes respectively, suggestive of high resistance intensity. Polymorphic inversion 2La frequencies in the survivors showed an especially high proportion of 2La/2La homokaryotypes and very low numbers of 2La+/2La+ homokaryotypes. Alternatively, the cohort of dead (susceptible) mosquitoes carried a greater proportion of 2La heterozygotes and a high proportion of 2La+/2La+ homokaryotypes. These 2La inversion frequencies occurred within Hardy-Weinberg proportions in the survivors and dead mosquitoes (2=1.32, p=0.8 for resistant mosquitoes i.e survivors, n=39, and 2=2.54, p=0.5 for susceptible mosquitoes i.e. survivors, n=113). Significant variation in inversion frequencies between the resistant and susceptible cohorts (2=54.64, DF=2, p<0.05) suggests that the 2La inversion associates with resistance to dieldrin in this population and that other factors may also be significant in construction of the resistance phenotype.

**Conclusion**: Variation in the assortment of 2La inversion arrangements between resistant and susceptible cohorts of this An. coluzzii population suggests that dieldrin resistance is at least partially linked to inversion 2La which may explain the persistence of dieldrin resistance in this population despite an absence of selection for resistance to this insecticide.

Biting Times of Plasmodium falciparum-Infected Mosquitoes and Transmission Intensities Following Five Years of Insecticide-Treated Bed Nets Use in Kamuli District, Uganda: Implications for Malaria Control

George Kabbale

#### Chlorfenapyr: Vector Control with a novel Mode of Action for Malaria Intervention

Authors: Austin James, Stutz and Weinmueller S. and E.

Abstract: The utility of bed nets and IRS to combat the scourge of malaria has been undeniably successful. However, a negative consequence of the widespread use of long-lasting insecticidal nets or LNs and IRS applications, has been the accompaniment of insecticide resistance issues. Interceptor® G2, a combinational bed net comprised of two distinct active ingredients, alpha-cypermethrin and chlorfenapyr, complementarily work together with each insecticide possessing unique modes of action. Interceptor® G2 has received interim recommendation by WHOPES. Interceptor® G2 is the only LN with two different classes of insecticide on a single LN, making it unique, and provides the ability to control OP, carbamate, DDT and pyrethroid resistant mosquitoes. Interceptor® G2 possesses both an excito-repellent component (alpha-cypermethrin) and a physiological insecticide (chlorfenapyr) that work in a concerted way to provide improved protection to LN end-users. Field testing results from Benin, Burkina Faso, Ivory Coast and Tanzania have unequivocally demonstrated significantly higher efficacy to resistant mosquito strains (40-60 percent higher mortality to resistant strains) compared with a LN containing alphacypermethrin only. This net, manufactured by BASF was developed through a funded partnership with IVCC and holds great promise as a LN that can complement areawide efforts to prevent malaria transmission unlike any other LN currently in the market in both areas with and without pyrethroid resistance. Similarly, evaluations of chlorfenapyr (240g ai/L SC formulation and branded as Sylando®), as an IRS tool show great promise. Hut trials conducted in Benin, Burkina Faso, Nigeria and Tanzania demonstrated its ability to kill resistant mosquitoes with IRS spray applications to walls over several months. The non-repellent action of chlorfenapyr allows blood-fed mosquitoes to rest on treated walls while acquiring a dose to kill them. This new tool can be complementary to LN use or as a solo IRS intervention where insecticide resistance issues plague communities and existing tools are not effective. The utility of chlorfenapyr, whether on an LN or as an IRS provide the first new insecticide to vector control in over 30 years, and can be an important additional tool in the fight against malaria.

# Geographic distribution of Aedes aegypti and Aedes albopictus (Diptera: Culicidae) and genetic diversity of invading population of A. albopictus in the Republic of Congo.

Authors: Basile Kamgang<sup>1\*</sup>, Theodel A. Wilson-Bahun<sup>1, 2</sup>, Helen Irving<sup>3</sup>, Arsene Lenga<sup>2</sup> and Charles S. Wondji<sup>1,3</sup>

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**Background:** The arbovirus vector, Aedes albopictus, originating from Asia, has recently invaded African countries including the Republic of the Congo where it was associated with a chikungunya outbreak. Up until now, little was known about its distribution in relation to the native Aedes aegypti and how the invasion will modify the epidemiology of arboviral diseases. Here, we assessed the current distribution of Ae. albopictus and Ae. aegypti in the Republic of the Congo and explored the genetic diversity of the invading species, Ae. albopictus.

**Methods:** Immature stages of Aedes were collected in nine locations in the Republic of the Congo in 2017 following a north-south transect and reared to adult stage. Adults were morphologically identified, counted and grouped according to species and location. Genetic diversity of Ae. albopictus was assessed by analyzing the cytochrome oxidase I (COI) gene.

**Results:** *Ae. albopictus* and *Ae. aegypti* were found together across the country in all the locations investigated. The invasive species is predominant over the native species in all the locations except in Brazzaville, suggesting that Ae. albopictus is displacing Ae. aegypti across Congo. When comparing the species distributions across the two largest cities: Brazzaville and Pointe Noire, Ae. albopictus was found to be more prevalent than Ae. aegypti in the suburbs whereas the opposite situation was reported in the city centre. Mitochondrial DNA analysis revealed very low genetic diversity of Ae. albopictus with only three haplotypes recorded across the country supporting the recent introduction of this species in Congo. Phylogenetic tree analysis revealed that Ae. albopictus from Congo originated from other tropical Asian countries such as China, likely as a result of increasing trade links.

**Conclusion:** These findings are important for the implementation of vector control strategies and can serve as a foundation for further research on these vectors in the country.

Keywords: Aedes albopictus, Aedes aegypti, ecological distribution, arbovirus vectors, genetic diversity, Republic of the Congo

Differential Regulatory Mechanism: Does Accessibility and Genetic Changes in Regulatory Elements Drive Overexpression of Metabolic Genes in Insecticide Resistant Anopheles gambiae population

Authors: Harun Njoroge, Arjen Van't Hof, Martin Donnelly.

**Background:** Insecticide resistance is a complex trait involving interplay of multiple genes and regulatory mechanism. Although some progress has been made in development of markers and characterizing resistance arising from target site insensitivity, other mechanisms such as metabolic, reduced cuticular penetration or behavioral avoidance which involves multiple components are still underexplored. In addition, very little is known on the regulatory mechanism driving overexpression of metabolic genes in insecticide resistant population. Our study aims to map regulatory elements and identify polymorphisms in these elements that are associated with differential gene expression in insecticide resistant populations. Molecular variants of identified components will further be explored as diagnostic markers for metabolic driven insecticide resistance.

**Methods**: Kisumu, Tiassale and Baharidjan laboratory strains were phenotyped for pyrethroid resistance by exposing them to deltamethrin concentrations coinciding to their LC50s determined through dose response assay. RNA sequencing and formaldehyde assisted isolation of regulatory elements coupled with sequencing will done on survivors of each strain to characterize their gene expression profile and regulatory elements respectively. Using both data sets, regulatory elements associated with overexpression of genes in resistant populations will be determined. Polymorphism in regulatory elements that are significantly associated with overexpression of genes will be screened in mosquitoes collected in West and East Africa.

Results: Study ongoing. We expect to have preliminary results by September 2018.

Outside surveillance of wild mosquito populations using Resting Bucket Trap around the containment insectary in Mali

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**Background:** Within Target Malaria, a not-for-profit research consortium, the Malaria Research and Training Center in Mali is preparing to import genetically modified Anopheles coluzzii mosquitoes for studies in containment, pending approval. The insectary is located on a campus with research institutions and schools of Medicine and Pharmacy and complies with national biosafety regulations. Surveillance of wild mosquitoes outside the facility is an additional measure that informs the likelihood of establishing laboratory mosquitoes in wild populations in the unlikely event of accidental escape from containment. The goal of outside trapping is to provide evidence of general trends in mosquito abundance and species composition for the location over dry and rainy seasons.

**Materials and method**: Four Resting Bucket Traps (RBT) were set up around the facility to collect mosquitoes overnight once a week from May 2016. Mosquitoes were morphologically identified and counted.

**Results and discussion:** During 103 weeks through April 2018, a total of 2873 mosquitoes were collected: 97.8% were Culex spp. (n=2810); 0.3% were An. gambiae s.l. (n=8); 0.6% were Aedes spp. (n=17); 1.3% were unidentified Culicidae (n=38). The majority of Culex mosquitoes were collected in dry seasons (64.4%; n=1810); 35.6% in rainy seasons (n=1000). An. gambiae s.l. were rarely trapped, only eight specimens were collected during the entire period: five in rainy seasons and three in dry seasons. Seven Aedes were collected in rainy seasons and ten in dry seasons. Given the very small number of An. gambiae collected and the strict biosafety measures in place, accidental interaction between laboratory and wild mosquitoes outside the containment facility is extremely unlikely.

**Conclusion:** Immediate and localised mosquito control in the event of an escape would eliminate the already low possibility of establishment of laboratory mosquitoes in the environment.

Keywords: mosquitoes, containment, outside surveillance, biosafety, transgenic

# A PiggyBac-like transposon insertion enhances transcription of Gste2, an insecticide resistance gene in Anopheles funestus

Authors: Leon. M. J. Mugenzi, Weedall G. D., Jacob. M. Riveron and Charles S. Wondji

**Background:** Variation in gene expression contributes greatly to adaptive changes in insects and is thought to play a major role in insecticide resistance. Mutations in cis-regulatory sequences can drive increased gene expression. A combination of overexpression and allelic variation in the glutathione S-transferase gene, GSTe2, can confer resistance to DDT and cross-resistance to pyrethroids in Anopheles gambiae and An. funestus. However, little is known about the factors driving overexpression.

**Methods:** whole genome sequencing (WGS) of pools of An. funestus mosquitoes from different African regions was used to identify genetic diversity upstream of GSTe2 and Luciferase reporter assays used to investigate the potential role of cis-regulatory mutations to the increased expression GSTe2 in resistant mosquitoes.

**Results:** WGS identified a 700bp PiggyBac-like transposable element upstream of GSTe2 (within the 3'UTR of GSTe1), in a population from Kpome, Benin in which the GSTe2-L119F mutation in the coding region of GSTe2 is at or near fixation. Elsewhere in West and Central Africa, where GSTe2 overexpression contributes to insecticide resistance, genotyping of the transposon showed that it was absent from Mibelong and Obout in Cameroon and showed a contrasting pattern in two populations from Ghana: In Obuasi, where GSTe2 expression is higher than Adawukwa (t(4) =7.62, P =0.0016), the transposon was present at 48% frequency while absent in Adawukwa. The Luciferase reporter assay was used to compare the activity of promoter region a GSTe2 from the resistant (with the transposon and without) and susceptible mosquitoes. A promoter from resistant mosquitoes showed higher activity than one from a susceptible strain, 3.7 fold higher with transposon (p<0.0001) and 2.3 fold higher for the promoter without transposon (p=0.0084). Presence of the transposon significantly increased promoter activity (1.6 fold higher luciferase activity; p=0.01).

Conclusion: this study showed that transposition in the promoter sequences of GSTe2 enhances gene expression.

### Pupal productivity of larval habitats of Aedes aegypti in Msambweni, Kwale County Kenya

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**Background:** Dengue is endemic in Kenya with recent studies suggesting continuous low transmission year round. Frequent outbreaks occur, especially on the coast. Ae. aegypti, the primary dengue vector in Kenya, breeds predominantly in water storage containers and discarded tires.

**Methods:** We estimated habitat-specific pupal productivity of Ae. aegypti in a south coastal Kenyan village. A total of 78 different water storage containers and discarded tires were inspected daily for Ae. aegypti larvae and pupae for 30 days. Containers were categorized as drums, tires, pots, small domestic containers (SDC) or 'others'. Small domestic containers (SDC) included small plastic food containers, tins, bottles, plates, cans, cooking pots (sufuria) and jars. 'Others' included; buckets, jerrycans, polythene bags, fallen leaves, coconut shells, hoof prints, drains, gutters, septic tanks, shoes, cisterns, sinks and animal feeding containers (AFC). Water management behavior including container covering, uses, filling and emptying, and habitat characteristics including size and movability were recorded.

**Results:** Overall, drums and tires were the most productive habitats accounting for 83% of all pupae collected. A poisson regression analysis showed that compared to SDC, the risk of finding pupae was 18% higher in drums, but comparable for the other habitats types. Pupae were more abundant in containers that were larger (15%), were stable (17%), and those that contained water with no particular purpose (60%).

**Conclusions:** In the absence of an effective antiviral and vaccine, control of the immature mosquitoes remains the most viable prevention method. Vector control interventions targeting primarily drums and tires in the study region could likely produce a positive impact on local dengue transmission.

# High Insecticide Resistance Intensity Of Anopheles Gambiae S.L. And Low Efficacy Of Pyrethroid Llins In Accra, Ghana

Authors: Rebecca Pwalia, Joannitta Joannides, Alidu Iddrisu, Charlotte Addae, Dominic Acquah-Baidoo, Dorothy Obuobi, Godwin Amlalo, Samuel Akporh, Sampson Gbagba, Melinda P. Hadi, Joseph Chabi, Samuel K. Dadzie

**Background & Objective:** Insecticide resistance of Anopheles gambiae s.l. against public health insecticides is increasingly reported in Ghana. Where resistance is confirmed, WHO recommends investigating the intensity of resistance. The aim of the study was to determine the status of insecticide resistance intensity of An. gambiae s.l. mosquitoes from Opeibea in Accra, Ghana and the effect against some commonly used long lasting insecticidal nets (LLINs).

**Materials & Methods:** Mosquito larvae were collected from Opeibea, a vegetable growing area and reared to adults. Three to five day old adults were assayed using WHO and CDC discriminating dose and intensity bioassays against different insecticides. Vgsc-1014F and ace-1 mutations within the population were characterized using PCR methods. Cone bioassay were conducted on commonly available LLINs in Ghana including DawaPlus 2.0, Olyset, PermaNet 2.0, PermaNet 3.0, and Yorkool.

**Results and Discussion:** An. gambiae s.l. from Opeibea were resistant to deltamethrin, permethrin, DDT, malathion and bendiocarb insecticides tested using WHO susceptibility tube tests. Vgsc-1014F and ace-1 frequency of the population tested were 0.99 and 0.76, respectively. Intensity assays using both CDC and WHO intensity bioassays showed high resistance intensity to pyrethroids and carbamates with mosquitoes surviving at 10x diagnostic dose. Only pirimiphos methyl recorded a low resistance intensity with 100% mortality at 5x diagnostic dose. Efficacy of pyrethroid LLINs ranged from 2.2 to 16.2% mortality at 24 hours. Bioefficacy against a PBO LLIN, PermaNet 3.0, was 75.7%.

**Conclusion & Recommendation:** WHO and CDC intensity bioassays confirmed high resistance intensity of An. gambiae s.l. in Accra, Ghana and reduced efficacy of the LLINs tested. Efforts must be put in place to mitigate the operational impact on the efficacy of vector control programs

# Integrated Approach In Malaria Vector Control

# Author: Dr. Tirthankar Basue

Main text: IVM is "a rational decision-making process for the optimal use of resources for vector control". The integrated vector management is not a tool in itself, and it is rather a policy framework and an umbrella in which proven intervention tools are utilized synergistically and cost-effectively through inter-sectoral action of health to impact on the reduction of disease transmission. Integrated vector management therefore, provides an adaptive management approach ensuring a permanent optimal level of effectiveness of vector control interventions in local settings.

**Background:** The scope of IVM is comprehensive. It promotes the appropriate management of insecticides, including their judicious use and the effective handling of stocks. It also allows a pro-active approach to vectorborne disease prevention by the incorporation of environmental management measures in water resources development. For these and all other components, the establishment or strengthening of intersectoral links is the most crucial of the various enabling conditions for IVM.

# Materials and methods: (PPT-Presentation)

- Major Challenges in Vector control
- · Elements of an integrated vector management strategy
- · Epidemiological and entomological assessment
- Assessment of challenges and constraints
- Problem-solving in IVM
- Role of IVM Team
- Intersectoral Collaboration
- Community participation
- Vector control need assessment
- Decision Making process in IVM
- Moto of Vector control intervention.
- Available methods used to control vector-borne diseases
- Criteria and procedures for selection of intervention tools
- List of intervention tools & their application in a judicious manner

**Results and Discussion:** The regional framework outlines the roles and expectations of countries as well as that of WHO in ensuring that countries develop and implement IVM in a sustainable and cost-effective manner.

**Conclusion:** A strategic plan for the implementation of IVM should be developed. To achieve these goals and create strong and effective advocacy for IVM, strengthen Intersectoral colaboration, Capacity building, Community mobilization, evidence based decision making process.

Complexity of the malaria vectorial system in Africa: update on two majors malaria vectors in the equatorial forest region: An. nili and An. moucheti

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**Background**: Mosquitoes of the An. moucheti and An. nili groups are important malaria vectors in the equatorial forest region yet their bionomic, and genetic structure is still not well known the present study provides updated data on the bionomic and genetic structure of these mosquitoes.

**Methods:** Mosquitoes collected all across Africa were used for both genetic and bionomic analysis. Microsatellite data and rDNA sequences were used to estimate the level of genetic diversity and differentiation between populations. Semi-gravid female ovaries were prepared for the description of chromosomes maps. Bionomic studies consisting in collection of mosquito using different sampling technics, analysis of blood meal and infection status through dissections and ELISA analysis were used to determine mosquito epidemiological role.

**Main findings:** Bionomic and genetic studies on An. nili and An. moucheti groups, permitted to confirm the high genetic heterogeneity of the two groups. An. moucheti group compose of An. m. moucheti, An. m. bervoetsi and An. m. nigeriensis saw An. bervoetsi race to the rank of full species on the basis of genetic differences. A new diagnostic PCR was described for the rapid identification of members of this group. Genetic structure analyses confirm isolation by distance as the main force driving the differentiation between An. m. moucheti populations.

Studies on An. nili demonstrated high genetic differences between members of the group (An. nili, An. ovengensis, An. carnevalei, An. somalicus). A new diagnostic PCR was described for the rapid identification of members of this group. Chromosomal maps for species of the group were described. No significant genetic difference was detected between An. nili ss populations distributed in Cameroon, Nigeria, Burkina Faso, Cote d'Ivoire, and Senegal.

Bionomic studies on these species across their distribution range showed a high implication of An. moucheti and An. nili in malaria transmission and their high susceptibility to insecticides used for vector control in Cameroon.

**Conclusion:** The study confirms high complexity of the vectorial system in Africa and stresses the need for more studies on these species.

Key words: An. moucheti, An. nili, genetic, bionomic, malaria, Africa

The influence of socioeconomic factors and breeding sites on the coverage and utilization of bed nets and House modification (screens and closed eaves) in Tanzania

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**Background**: Efforts to achieve universal coverage of insecticidal nets and advocacy for house improvement to prevent malaria are improving and scaling-up, after socioeconomic factors have been shown to be one of the major factors hindering such efforts in Tanzania. Therefore, the study investigates the influence of socioeconomic and proximity to breeding sites on the combination of nets and house improvement to understand the consequences of the efforts.

**Methods:** A cross-sectional survey using a structured questionnaire was administered to 7435 and 1223 households in Bagamoyo and Ulanga district, respectively. The association between socioeconomic factors and bednets coverage and use, with house improvement were analyzed using Regression models in STATA. While, the association between breeding sites and study forms of protection was analyzed using nearness proximity ArcGIS.

**Results:** Wealthiest households were half as less likely to use window screens and combination of bednets with screens than the poorest households (OR 0.18 (0.12 - 0.28) p<0.0001) and (OR 0.30 (0.21 - 0.44) p<0.0001), in Bagamoyo, while the use of window screens and a combination of nets with screens were more than fourfold more likely in the richest quintile than the lowest in Ulanga. Moreover, the demographic and breeding sites influenced population access to nets and house improvement.

**Conclusion:** The study suggests that population access to bednets and house modification were low in the population, therefore need improvement. The influence of breeding sites on use of nets and house modification tools cannot be ignored.

Keywords: Malaria, Bednets, House improvement, Breeding sites, Socioeconomic, Tanzania

Socio-demographic factors associated with Insecticide Treated Net use among under-five children in Nigeria.

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**Introduction**: The use of Insecticide-treated nets (ITN) is the most cost-effective malaria preventive measure. However, malaria remains a big killer of children in Nigeria, accounting for about 30% of the under-fives (U5) deaths. Despite massive distribution campaigns nationwide, ITN use among (U5s) still needs to be enhanced. This study assessed factors influencing ITN use among U5s across households in Nigeria.

**Methods**: We analyzed the 2015 Nigerian Malaria Indicator Survey with specific focus on ITN use in children. A total of 5,397 U5s who slept in households with at least one ITN the night prior to the survey were studied. We conducted binary logistic regression analysis to identify factors influencing ITN use by U5s in (1) households that have at least one ITN and (2) households with at least one ITN for every two people (universal coverage). Data analysis was conducted using STATA (version 14.0).

**Results:** In households with at least one ITN, 56.6 % of U5s slept under the ITN the night before the survey, whereas among households with universal coverage, only 33% of U5s slept under the ITN a night prior to the survey. Significantly, children less than three years old, those who reside in other regions of the country except the South West had higher odds of using ITN the night before the survey. Also, children from households with less than 7 members and those from the first, second and third wealth quintiles had significantly higher odds of using ITN. On the second regression model among households with universal coverage, U5s who reside in urban areas, those from other regions of the country except the South-south and South-west and those from the lowest three wealth quintiles still had significantly higher odds of sleeping under ITN the night before the survey.

**Conclusion and recommendation:** Our findings have programmatic implications for malaria control among U5s in Nigeria. Interventions to improve ITN use should be intensified among rural households with children older than 2 years, larger households, and residents of south western Nigeria

Study of the Efficacy of Bacillus Thuringiensis Israelensis for the Control of Anopheles Gambiae Sl Resistant To Pyrethroid.

#### Author: Roland ALIA

**Background:** Vector control is the WHO recommended strategy for fighting vector-borne diseases. In this study, the efficacy of Bacillus thuringiensis used today to replace chemical insecticides to circumvent vector resistance, was evaluated.

**Methods:**To determine the optimal dose applicable under natural conditions, three different doses  $(1g / m^2 - 1.5g / m^2 and 2g / m^2)$  of the commercial Vectobac GR 3.33% AI (200 ITU / mg) formulation of Bti were separately tested in laboratory on stage II larvae of resistant Anopheles gambiae sl population to measure adult emergence inhibition rates. The persistence of the product was then determined under natural conditions through the measurement of larval density reduction after application of the optimal dose.

**Results:** The linear regression model used considering the  $1g / m^2$  dose as a reference showed that the  $2g / m^2$  dose inhibited 1 / 76.71 = 0.01 - relative risk = 76.71 [69.41; 84,77] times the emergence of adults than doses  $1g / m^2$  and  $1.5 g / m^2$ . Under natural conditions, the larval density reduction observed on day 1 with evolved larvae stage (L3-4) was greater (99.8%) than that was observed (73.5%) with young larvae stage (L1-2). According to the mixed linear regression model, the persistence of the product was about 3 days on evolved larvae stage.

**Conclusion:** The formulation of Bti used in this study is not significantly effective to be considered at indicated doses in perspective of large scale control.

# The efficacy of Fludora® Fusion under South African field conditions

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2. Bayer (Proprietary) Ltd, SA

**Background:** The impact of vector control is declining, arguably due to the development of insecticide resistance to the classes of insecticides recommended for Public health until now and a reduction in coverage due to higher costs of using non-pyrethroid insecticides. Bayer has developed a new insecticide mixture, Fludora Fusion, as a tool for malaria vector control. Fludora Fusion is a combination of clothianidin and deltamethrin and needs to be evaluated for indoor residual spraying. This study was initiated to determine the long-term efficacy of Fludora Fusion under field conditions.

**Methods**: This study, conducted in Othobothini, northern KwaZulu-Natal, South Africa, utilised the WHO cone bioassay technique to test the assumption that Fludora Fusion kills malaria vectors and that the insecticide persists for at least 6 months. The insecticide was applied to mud, cement and painted surfaces using a calibrated Hudson X-Pert spray pump. DDT was used as a positive control. Bioassays were carried out at monthly intervals on all three surface types. For each monthly evaluation, knock-down (30 and 60 minute) and mortality assessments were carried out with An. Arabiensis female mosquitoes, after exposure to the sprayed surface. Mortality was recorded every 24 hours for 7 days.

**Results:** Fludora Fusion performed as well as the positive control, DDT, throughout the 12 month duration of the trial. At month 12 the test insecticide was continuing to achieve 100 % mortality within 72 hours on cement and painted surfaces and within 96 hours on mud surfaces.

According to WHO criteria, Fludora® Fusion is an effective insecticide for use in IRS programmes.

**Conclusions:** Fludora<sup>®</sup> Fusion meets the WHO standards of >80% mortality and has comparable residual efficacy in relation to DDT, the currently used insecticide in the South Africa malaria control programme.

### Mosquito Repellency Activities Of Selected Plants Used In Ethnomedicine In Embu County, Kenya

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#### Mount Kenya University

Malaria is still the world number one killer in Sub-Saharan Africa. 76% of the Kenyan, population risk contracting the disease annually. In Embu County, it accounts for 42% of all outpatient visits annually. In this County, the disease is alleged to be the most treatable by herbalists. However, in the ethnobotanical knowledge relevant to prevention of the disease has not been documented. Consequently, repellency activity of the plants used to repel/prevent mosquito bites are yet to be scientifically validated. Data was collected using semi-structured questionnaires. Essential oils of Premna resinosa and Osyris lanceolata leaves were extracted through hydrodistillation using a Clevenger-type apparatus and their repellency activity tested against Anopheles arabiensis mosquitoes. Forty eight herbalists were interviewed. A total of eight plants were reported as being used to prevent mosquito bite. The most commonly used plants were Tagetes minuta, Allium sativum, Ocimum gratissimum, Osyris lanceolata and Premna resinosa in descending order. P. resinosa and O. lanceolata leaves demonstrated repellency activity against A. arabiensis species and thus confirming their use in ethnomedicine. Based on reviewed literature, there is no data listing the use of the two plants as mosquito repellent and/or confirming their mosquito repellency activity.

Key words: Malaria, Repellancy, Mosquito

Evolution of malaria vector bionomics and transmission patterns in association with treated bed net use in the equatorial forest region of Cameroon

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**Background:** Increased use of long lasting insecticidal nets (LLINs) over the last decade considerably improved the control of malaria in sub-Saharan Africa. However, there is still paucity of data on the influence of LLINs use and other factors on mosquito bionomic in different epidemiological foci. The objective of this study was to provide updated data on the evolution of vector bionomic and malaria transmission pattern in association with LLINs use in the equatorial forest region of Cameroon.

**Methods:** Mosquitoes were sampled in Olama and Nyabessan in 2000-2001 and 2016-2017 using both human landing catches and indoor sprays, and were identified using morphological and molecular tools. Susceptibility level to both permethrin and deltamethrine were assess on An gambiae. Mosquitoes were screened for Plasmodium infection and blood feeding preference using the ELISA technique. Parasitological surveys in the population were conducted using malaria rapid diagnostic tests.

**Results:** A change in the dynamic of sampled mosquitoes was recorded between the 2000-2001 collections and those of 2016-2017. A drop in the density of local primary vectors in the forest region An. nili and An. moucheti was recorded whereas on the other side an increase in the density of An. gambiae sl, An. marshallii, An. ziemannii and An. paludis was recorded. A change in the biting behaviour from indoor to outdoor was recorded in Olama. A change in the night biting cycle was recorded with mosquitoes displaying a shift from night biting to late evening/ early in the night. Several mosquitoes were found positive for Plasmodium infection in both sites. A high level in resistance to pyrethroids was detected for An. gambiae in both sites. High parasite prevalence was recorded in both sites and LLINs were protective against malaria infection.

**Conclusions:** The study shows important changes in the bionomic of vector populations and malaria transmission patterns. The changes call for more concerted efforts to address challenges such as insecticide resistance or behavioural changes affecting the performance of current control measures.

Key word: Malaria, transmission, Equatorial forest region, Cameroon, LLINs, Anopheles

# Building the trust between local communities and researcher, a guarantor for the success of new technologies development in malaria elimination: GM mosquitoes as a case study in Bana, Burkina Faso

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**Introduction:** To definitively defeat malaria, WHO strongly recommends the contribution of basic research. In this context, new technologies focused on genetic engineering are being developed in some malaria-endemic countries, including Burkina Faso. In this country, Target Malaria, a not-for-profit research project in progress since 2012, seeks to develop GM mosquitoes as a complementary control tool to existing ones. The success of the research activities going on at the local level by this project depends on the support and engagement of all the actors, in particular the nearby local communities the trials are being experimented in. Therefore, this study seeks to understand the nature of interactions between researchers and local communities in Bana in regard to this project.

**Materials and method**: Data was collected using qualitative approaches from the actors, team research and communities, involved in the implementation of the project activities in Bana. 60 in-depth interviews were conducted. The data was recorded, transcribed and analyzed on the basis of thematic contents.

**Results and discussion:** The interactions between research teams and localcommunities in the implementation of Target Malaria project activities are taking place in a user-friendly environment. Most respondents say they have a collaborative and friendly relationship inregard to this project thanks to the transparency and inclusivity of the researchers. In addition, it appears that the permanent information, sensitization and dialogue activities conducted by the research teams promote better understanding of the project for the communities and at the same time strengthen trust relationships between researcher and communities.

**Conclusion:** The trust between the actors constitutes a bearing on which Target Malaria bases the success of its research activities. In this sense, the team in charge of the stakeholders'engagement plays a key role by building and sustaining this trust.

Key words: Target Malaria, experimental trials, interaction, GM mosquitoes, social sciences, trust building, Bana, Burkina Faso.

Common activities that put people at an increased risk of malaria transmission

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**Background:** Malaria has significantly decreased over the past decade, but the rate of this success seems to stagnate, leading to a low but persistent transmission. Several factors related to both the mosquito and human behaviors are attributable to the residual transmission. In this study we set to identify human behaviors that keep them at an increased risk of malaria transmission in East and West Africa.

**Method:** Communities from randomly selected households were observed in villages in Burkina Faso and Tanzania between 6PM and 7AM both at homes and away from home, in places where people gather. The location of people and activities they were doing were recorded on half-hour basis.

**Results:** Majority of the household members spent most of their early night hours outdoors in Tanzania and Burkina settings. Activities that kept people outdoors were similar between the two countries; these included cooking, story-telling and eating. Higher bed net usage was observed in Tanzania than in Burkina Faso. However, higher outdoor sleeping was observed in Burkina Faso.

**Conclusion:** It is necessary to target these human behaviors when designing new tools or improving the current tools for preventing malaria transmission.

Key words: residual malaria, community behavior, Burkina Faso, Tanzania

Survival, preference and use rates of 8 types of LLINs after 6, 12 and 24 months of a mass distribution campaign in Senegal.

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**Background:** Long Lasting Insecticide Nets (LLINs) are one of the core components of global malaria prevention and control strategies currently in use. Despite this, the effective lifespan of nets and their durability under field conditions, remain unclear. This study evaluated the survival, preference and use rates of 8 types of LLINs after 6, 12 and 24 months of a mass distribution campaign in Senegal.

**Methods:** In November 2014, 12,608 LLINs were distributed in 5 stratified rural / urban areas. As part of the longitudinal follow-up, 2230, 1669 and 1136 nets were randomly sampled, respectively at 6, 12 and 24 months post-distribution. Structured questionnaire responses from household surveys and the assessment of physical integrity allowed us to detect, for 8 types of LLINs, the loss due to the displacements of bed nets or physical failure, preference and use rates. All data were analyzed and compared using STATA\*11.2 software.

**Results:** The proportion of LLINs remaining with in surveyed households was significantly higher for circular net types of Interceptor<sup>®</sup> (OR 2.2; 95% CI 1.6-3; p< 0.05) and PermaNet<sup>®</sup> 2.0 (OR 2.1; 95% CI 1.6-2.9; p< 0.05). The nets survival was significantly higher in rural setting than in urban area (OR 2.2; 95% CI 1.9-2.6; p< 0.05). Despite this good retention, the usage rate was relatively low (OR 0.6; 95% CI 0.4-0.7; p< 0.05). Loss due to physical failure was higher for Interceptor<sup>®</sup> (OR 2.3; 95% CI 1.7-3; p< 0.05), Yorkool<sup>®</sup> (OR 2.2; 95% CI 1.6-3; p< 0.05) and Olyset<sup>®</sup> Net (OR 2; 95% CI 1.5-2.7; p< 0.05). The two types of circular shape (Interceptor<sup>®</sup> and PermaNet<sup>®</sup> 2.0 C) are most preferred with preference rates of 99.24% and 99.28%, respectively. The less commonly used types of LLINs were the MAGNet<sup>™</sup> (OR 0.4; 95% CI 0.2-0.6; p< 0.05) and the Netprotect<sup>®</sup> (OR 0.6; 95% CI 0.4-0.9; p< 0.05).

**Conclusions:** Our study showed that the retention and usage rates of LLINs in households are positively associated to the preference. Beyond the difference between LLINs brands and shapes, the area has an important influence on the two above parameters.

Keywords: LLINs, Survival rates, Preference, Use.

Persistent community Plasmodium prevalence after control of indoor biting Anopheles: a threat for malaria upsurge after halting indoor residual spraying in Tororo District, Eastern Uganda

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**Background:** Since 2014, use of long lasting insecticide treated nets (LLINs) and indoor residual spraying (IRS) have controlled indoor resting Anopheles, but Plasmodium transmission continues. We assessed the trends of malaria, indoor resting vector density and determined community Plasmodium prevalence after four rounds of IRS.

Methods: We analyzed Health Management Information System (HMIS) malaria surveillance data of 2,727,850 patient records in 61 health facilities from 2012 to 2015 to estimate monthly malaria incidence for the entire population and also separately for <5- and ≥5-year-olds before and after introduction of vector control interventions and determined laboratory test positivity rates. We tested 100 residents per village(n=20) for malaria using Rapid Diagnostic Tests (RDT) and microscopy. We classified malaria positive individuals as symptomatic or asymptomatic. We asked people about mosquito bites before sleeping under LIINs. Indoor resting Anopheles density was monitored monthly. Chi square and logistic regression were used for trend analysis.

**Results:** Mean monthly malaria incidence fell from 95 in 2013 to 76 cases per 1000 in 2014 (OR=0.99, 95% CI=0.96-1.01, P=0.37). Among children <5 years, the malaria incidence reduced from 100 cases/1000 in 2014 to 45/1000 in 2015 when IRS was introduced (OR=0.94, 95% CI=0.91-0.996, P<0.0001). Malaria RDT positivity rate reduced from 57% in 2013 to 30% (Chi =15, P<0.0001) in 2015. Slide positivity rate reduced from 45% in 2013 to 21% in 2015 (P=0.004) while RDT positivity declined from 69% to 40%. Asymptomatic community malaria prevalence after four rounds of IRS was 14%; higher among children <5 years (21%) than adults (7.0%). Indoor Anopheles resting density reduced to zero. 90% of people stay exposed to malaria vectors before sleeping under LLINs.

**Conclusion:** Moderate Plasmodium prevalence and changing biting behavior of Anopheles present a threat for malaria upsurge after stopping IRS. Active malaria testing and treatment should be considered in communities to remove the parasite source.

Keywords: Malaria, Prevalence, threat, vector control

# Plasmodium sporozoite infection rates among primary and secondary malaria vectors in Rwanda

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**Background:** Anopheles arabiensis, Anopheles gambiae s.s. and Anopheles funestus are considered primary malaria vectors in Rwanda; however, the role of secondary vectors in malaria transmission has not been evaluated quantitatively. Plasmodium sporozoite infection rates of malaria vectors were evaluated from 19 sites across Rwanda.

**Methods:** Mosquitoes collected monthly July 2016–June 2017 by Human Landing Catch (HLC) and Pyrethrum Spray Catch (PSC) methods were morphologically identified. An. gambiae s.l. complex was determined for a random subsample by PCR. Infective rates of Anopheles mosquitoes were determined by ELISA for P. falciparum sporozoites in mosquito head and thorax.

**Results:** Of 10,048 female Anopheles mosquitoes collected, 85.4% were by HLC, and 12 species were represented: An. gambiae s.l. complex (79.6%), An. funestus (6.9%), An. pharoensis (4.1%), An. ziemanni (4.1%), An. rufipes (0.04%) and other Anopheles spp. (5.4%). An. gambiae complex by PCR (n=504) were An. arabiensis (68.6%) and An. gambiae s.s. (31.4%). For HLC, proportion collected outdoors for An. gambiae s.l., An. funestus and other Anopheles spp. was 53.6%, 53.9%, and 62.5%, respectively. The overall Plasmodium sporozoite infection rate (SIR) was 1.4% (n=10,048), with SIR 0.9% (n=1,469) for PSC and 1.5% (n=8,579) for HLC. SIR was 1.4% (n=3,853) among indoor catches and 1.7% (n=4,573) outdoors. The SIR for each Anopheles species tested was 1.2% (n=8,000) for An. gambiae s.l., 1.3% (n=684) for An. funestus, 2% (n=410) for An. ziemanni, 6.4% (n=407) for An. pharoensis, and 25% (n=4) for An. rufipes.

**Conclusions:** High Plasmodium sporozoite infection rates among mosquitoes considered as secondary malaria vectors suggests that secondary vectors might be contributing to malaria transmission in Rwanda. Given the diversity of the An. coustani group of mosquitoes (containing An. ziemanni), future work will include molecular identification of species including sequencing. Future surveillance and vector control intervention efforts should include these vectors focusing on outdoor transmission.

Keywords: Secondary vectors, Plasmodium, malaria transmission, Rwanda

# Development of novel mosquito sampling methods under low malaria transmission settings in South Africa

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**Abstract**: Diagnosis of malaria infections and vector incrimination plays a central role in areas being considered for malaria elimination. However, current vector surveillance tools are not fully addressing surveillance requirements needed under low malaria transmission settings. Against this background there is an urgent need to develop affordable robust sampling tools to monitor vector populations during malaria pre-elimination and elimination phase. Our group is developing a low-cost mosquito surveillance system that can easily be used by malaria control programmes. Studies on a mosquito surveillance system under development in Mamfene, northern KwaZulu-Natal, South Africa using traditional clay pots and modified buckets will be presented. Preliminary data over a 2-yr sampling period shows the potential of clay pots for collecting male and female anophelines both outdoors and indoors. The first records of Plasmodium falciparum naturally infected populations of Anopheles vaneedeni and Anopheles arabiensis collected from outdoor-placed clay pots, will be discussed in the context of their possible role in residual malaria transmission in South Africa.

# Monitoring of functional mating swarms of Anopheles coluzzii in Mosquito Ecology Research Facility (MERF) in Burkina Faso

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**Background:** To fight against malaria, vector control based on the approach of released sterile or transgenic males is conceivable in Africa. The success of these promising technologies depends in part on the capacity of released males to mate with the wild females and, requires a better understanding of the reproductive system of malaria vector species. Previous studies have shown that swarming is the main reproductive behaviour of An. coluzzii and, swarm formation relies on environmental factors; sunlight variation and visual ground markers are thought to be important. The main objective of this study was to establish functional swarms of An. coluzzii above artificial visual markers in the MERF built in Burkina Faso.

**Methods:** Wild gravid females belonging to Anopheles genus were collected in VK5 using mouth aspirators. Mosquitoes were transported and maintained under MERF insectary conditions. Females were placed individually in cup for egg-laying. The females that laid eggs were identified by PCR. Newly hatched larvae were reared accordingly to their mothers have been identified as An. coluzzii. Pupae were removed, placed in sex-specific cages and, emerging adults were provided with 5% glucose solution. Three hundred males and one hundred females (3:1 sex ratio) of 3-5 days aged were released into each of 6 Rooms where a black sheet and a wood pile were used as visual markers to stimulate swarm formation. To monitor environmental conditions, Light intensity, Temperature and Relative Humidity were recorded by the MSR145<sup>R</sup> throughout the experiment. Swarm activities were monitored, by recording duration, height, number of swarming males, number of mating pairs and female insemination rate and, comparing between the two visual markers.

**Results:** In total 7200 males and 1200 females of An. coluzzii were released into 6 Rooms in 4 days. A total of 2421 swarming males representing 33.6% of released males were collected. The percentage of males collected above the black sheet (73.3%; 1799/2421) was higher than males above the wood pile (26.7%; 622/2421). In total 106 mating pairs were captured with a large majority on black sheet (77.5%; 83/106). The female insemination rate was 74.5% (73/98).

**Conclusions:** Our results have shown significant swarming success of An. coluzzii above the artificial ground markers in the MERF in Burkina Faso. The results would facilitate further studies on the reproductive system between different mosquito strains to guid vector control programs based on the Sterile Insect Technic and Gene Drives.

Keywords: Anopheles coluzzii, mating swarms, MERF, malaria

# Investigating on culturable bacteria associated with wild Anopheles gambiae s.l. in Burkina Faso for biocontrol prospect of malaria

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**Introduction:** The study of vector mosquito microbiota has recently gathered increased interest because of its potential influence on vector competence. However, the wild mosquito microbiota remains poorly studied, especially culturable bacteria. This work was primarily designed to isolate potential candidates for use in a paratransgenesis approach. Secondly, to isolate bacteria that could be used directly in biocontrol of mosquitoes. Finally, to give a picture of the microbiota of wild-caught Anopheles gambiae s.l. from the western part corner of Burkina Faso, which has the one of the highest malaria endemicity in the country.

**Methods:** Adults mosquitoes from inhabited and abandoned houses and swarms and different larvae stages from breeding sites were plated out onto 2 types of culture media (Chocolate + polyvitek agar and Bromocresol purple agar). Organs of females (cuticle, midgut, salivary glands and ovaries) and testes of males were dissected and plated also onto media. Then, bacterial colonies and species were isolates and were morphologically and biochemically identified and with the VITEK2 system as well.

**Resultats:** The prevalence of bacteria colonies was higher in Soumousso (an agriculture area) than Bama (a rice field area). The cuticle was most colonized with different colonies compared to the midgut, ovaries and salivary glands (46 and 51 respectively colonies). An average 40 different bacteria colonies were isolated in testes of males. The number of colonies with larvae was not important across stages (an average ~38). However, at the nymphae stage, this number decreased to ~24 different bacterial colonies. Promising culturable bacteria such as Lactobacillus sp, Serratia sp and have been identified. We also isolate mosquidocidal bacteria as Chromobacterium violaceum.

**Conclusion:** This study focused on culturable bacteria associated to mosquitoes. Supplementary studies such as sequencing of bacteria genomes should improve our knowledge of the microbiota and its interactions with host the host mosquito.

Keywords: Microbiota, Anopheles gambiae s.l, malaria, Bio-Control and Burkina Faso.

# Quantifying the intensity of permethrin insecticide resistance in Anopheles mosquitoes in western Kenya

Author: Seline Omondi

**Background:** The development and spread of resistance among local vectors to major classes of insecticides used in Long-Lasting Insecticidal Nets (LLINs) and Indoor Residual Spraying (IRS) poses a major challenge to malaria vector control programs worldwide. The main methods of evaluating insecticide resistance in malaria vectors are the WHO tube bioassay and CDC bottle assays, with their weakness being determination of resistance at a fixed dose for variable populations. The CDC bottle assay using different insecticide dosages has proved applicable in ascertaining the intensity of resistance.

**Methods:** We determined the status and intensity of permethrin resistance and investigated the efficacy of commonly used LLINs (PermaNet<sup>®</sup> 2.0, PermaNet<sup>®</sup> 3.0 and Olyset<sup>®</sup>) against 3–5 day-old adult female Anopheles mosquitoes from four sub-counties; Teso, Bondo, Rachuonyo and Nyando in western Kenya. Knockdown was assessed to 4 doses of permethrin;  $1 \times (21.5 \ \mu g/ml)$ ,  $2 \times (43 \ \mu g/ml)$ ,  $5 \times (107.5 \ \mu g/ml)$  and  $10 \times (215 \ \mu g/ml)$  using CDC bottle assays.

**Results:** Mortality for 0.75% permethrin ranged from 23.5% to 96.1% in the WHO tube assay. Intensity of permethrin resistance was highest in Barkanyango Bondo, with 84% knockdown at the 30 minutes diagnostic time when exposed to the 10× dose. When exposed to the LLINs, mortality ranged between 0–39% for Olyset<sup>\*</sup>, 12–88% for PermaNet<sup>®</sup> 2.0 and 26–89% for PermaNet<sup>®</sup> 3.0. The efficacy of nets was reduced in Bondo and Teso.

**Conclusions:** There was a reduced efficacy of nets in areas with high resistance portraying that at certain intensities of resistance, vector control using LLINs may be compromised. It is necessary to incorporate intensity assays in order to determine the extent of threat that resistance poses to malaria control.

Keywords: Insecticide resistance, Intensity, Permethrin, Anopheles gambiae.

Implementation Of Integrated Vector Management In A Large Zimbabwean Private Sugar Plantation: A Review Of Malaria Cases From 2013 To 2017.

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**Background:** Tongaat Hulett is an agro-based business situated in Chiredzi and specialising in irrigated sugarcane. The company employs approximately 9 000 workers and is home for close to 40 000 people. Chiredzi is one of the highest malaria burdened districts in Zimbabwe. The development and management of water resources for agriculture in tropical and sub-tropical climate zones has been linked to increased malaria transmission. It is estimated that malaria slows economic growth, in Africa, by about 1.3%. Each bout of malaria costs 12 productive labour days per infected person per year. The Estates established malaria control programmes in the 1950s with the aim of increasing productivity by protecting their employees and dependants.

**Programme Description:** This malaria control programme utilises the integrated vector management approach comprising of environmental manipulation, larviciding and adultciding using residual spraying (IRS) twice per year. The IRS programme involves the mosaic approach based on organophosphates and pyrethroids. Long lasting insecticidal nets (LLINs) and screening of some houses complement IRS.

**Lessons learned:** Over the 5 year period under review, 2 532 malaria cases were recorded in Triangle. These ranged from 123 to 1 035 cases per year with a crude average incidence of 12.7/1000 (ranging from 3.1 to 25.9/1000 per year). Total local transmission constituted 1.5% (39 cases ranging from 0 to 17 cases per year) translating to a local average annual incidence of 0.2 per 1000 (ranging from 0.0 to 0.4/1000 per year). Forty percent (18) of the local cases were employees.

**Conclusions:** The bulk (98.5%) of malaria cases in Triangle was imported. We noted that an increase in district malaria incidence rate did not affect malaria transmission in Triangle due to implementation of IVM in the sugar estates. Going forward, IVM has the potential of eliminating malaria in any setting.

# Innovative Communication Strategies For Enhancing Effective Community Participation In Integrated Vector Management In Malindi, Kenya .

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**Background:** Vector-borne diseases account for more than 17% of all infectious diseases, causing more than 1 million deaths annually. A key element in integrated vector management is that of community empowerment, social mobilization and inter sectoral collaboration. Integrated vector management involves making rational decisions on use of vector tools and disease control in relation to local conditions, community support, awareness and action.

**Methodology:** A community based integrated vector management was carried out in Malindi along the Kenyan coast since 2002. An easy to follow curriculum was developed which included components such as basic entomological skills and communication skills. Lay persons known as 'mosquito scouts' were trained for 3 weeks and supportive supervision provided during field work. Each mosquito scout was assigned an area of 1km2. Their activities included searching for mosquito breeding areas, communicating the information to the community and community based groups and stakeholders, holding awareness campaigns such as Mosquito Field Events, Neighborhood campaigns, door to door campaigns in mosquito control to mobilize and sensitize the community for resources and action.

**Results and Discussion.:** A total of 62 mosquito scouts and 11 community groups affiliated to an umbrella body known as PUMMA (Punguza Mbu Malindi) were trained and involved in mosquito control activities, 12 Annual mosquito field events held, and weekly neighborhood campaigns held. The various communication strategies with messages on mosquito life cycle, diseases transmitted by mosquitoes and control and prevention measures motivated and enabled communities to understand a lot about mosquitoes and voluntarily took part in vector management activities and to reduce several environmental health risks. The mosquito scouts gained recognition and honor at the community level and are referred to as "mosquito doctors". This made it easy to disseminate and communicate to the household owners on mosquito breeding areas and measures to be taken to control the larval habitats. Malaria reduction has been reported in the area and residents reported reduced mosquito biting nuisance. There is need to evaluate the approach to expound on benefits accrued and challenges faced for its sustainability.

Key words: integrated vector management, community, mosquito scouts

# Community's Knowledge, Attitude and Practice towards Yellow Fever in Outbreak Recently Re-appeared Area, Southern Ethiopia

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**Background:** Yellow fever (Yf) is one of the most re-emerging mosquito-borne viral diseases in many African countries despite the availability of an effective vaccine. Outbreak was recently reported in South Omo of Southern Ethiopia. This area was also highly affected by Yf outbreak in the 1960s. However, there is no reliable information on the level of community knowledge attitudes and practices about the disease in the area. The objective of the current study was to assess level of community knowledge, attitudes and practices about Yf.

**Methods:** Between March and May 2017, a community-based cross-sectional survey was conducted in two districts of the South Omo area. During the survey, 612 randomly selected adults were interviewed about Yf using structured questionnaire.

**Results:** Out of the 612 study participants, 508 (83.0%) reported that they heard about Yf which is locally known as "a disease that causes vomiting blood". Most (90.4%) of the study participants also said that Yf is different from malaria. Two hundred thirteen (41.9%) participants said that Yf can be transmitted from a patient to another person, while only 80 (37.6%) mentioned that the disease is transmitted through mosquitoes bite. Out of 333 (65.7%) study participants who believed that Yf is a preventable disease, 280 (84.1%) mentioned vaccine as a preventive method. The majority believed that the disease is a killer (97.2%) and a newly emerging (69.4%). Among the total of 612 study participants, 221(36.1%) were considered as having a high level of overall knowledge of Yf. Having educational level above 7th grade (AOR= 3.25, 95% CI: 1.39, 7.57) and being resident of Bena-Tsemay district (AOR= 1.77 95% CI, 1.12, 2.78) were significantly associated with having a high level of verall knowledge of Yf. Agro-pastoralism as an occupation compared to farming was associated with having a low level of overall knowledge of Yf (AOR= 0.51, 95% CI, 0.33, 0.79).

**Conclusion:** The findings indicate that most of the study community members had a low level of overall knowledge of Yf, especially about its cause, mode of transmission and preventive methods. Thus, there is a need to increase people's knowledge and practices regarding the cause, mode of transmission and preventive methods like avoiding mosquitoe breeding sites beside vaccination through various strategies like disseminating information through community health extension workers and community leaders in the study area.

Key Words: Yellow-fever, mosquito-borne, knowledge, attitudes, practices, Ethiopia

Community Engagement Campaign toward a small-scale pilot mosquito Sterile Insect Technique release programme in Jozini KwaZulu-Natal, South Africa

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**Introduction:** As South Africa moves towards malaria elimination there is a need for complementary vector control strategies. One of the methods under consideration is the use of the sterile insect technique (SIT). Key to the successful implementation of new health intervention such as the SIT technology is community participation and addressing of negative social perceptions often associated with release of laboratory reared mosquitoes. This work describes the engagement of a community targeted for a small scale pilot sterile mosquito release programme with the objective of increasing awareness and creating more positive attitudes towards the use of SIT.

**Methods:** Knowledge and information regarding malaria transmission and control strategies inclusive of the potential of using the sterile insect technique was disseminated through radio interviews distribution of reading materials, road shows and lectures in IsiZulu. The awareness campaign was done in collaboration with various stakeholders including malaria control programme teams, Environmental Health Practitioners, Information Education and Communication (IEC) teams among others. The activities targeted areas in which people had already congregated such as markets, schools, clinics and farms. Concurrently, consenting participants were tested for malaria using falciparum-specific rapid diagnostic tests

**Results:** Approximately 165 000 listeners were engaged during two 30 minutes radio interviews at Maputaland Community Radio Station. Two hundred and fifty farm employees from Sitilo farm factory were given education on malaria transmission and control strategies. Questions on how the Sterile Insect Technique works featured among the questions asked during the farm outreach programme. Three primary health care facilities (Makhathini, Gedleza and Ndumo Clinics) with an average daily consultation of approximately 50 - 200 people were visited during the engagement exercise. In total about 1 500 children, grades 8 to 12 from Gugulesizwe High School participated during a health education exercise which included lectures on the SIT. Two road shows, one in areas around Mamfene community and a second at KwaPhuza Market were conducted. In total 447 tests were conducted with 20 testing positive and immediately treatment was initiated.

**Conclusions:** The campaigns showed that majority of the people are informed on malaria transmission and control. However, there is lack of understanding regarding the SIT. A more extensive public awareness on the use of SIT as a vector control strategy is recommended to prepare the community of Mamfene, Jozini for any future small pilot field releases of sterile males.

# Hut trial evaluation of long-lasting nets coated with Deltamethrin and mixture of Deltamethrin and PBO against free flying of Anopheles gambiae s.l pyrethroid resistant populations in Burkina Faso.

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**Background & Objective:** Despite the fact that malaria still remains a serious public health, there is a significant reduction in malaria-associated morbidity and mortality across sub-Saharan Africa. This is partly due to the large scale implementation of long lasting insecticide treated nets (LLINs) and indoor residual spraying (IRS). But Pyrethroid-resistant to malaria vectors became a serious threat for malaria control. Bed nets that can protect againt resistance mosquitoes are urgently needed. Here, we evaluated the effectiveness of DawaPlus 3.0 and DawaPlus 4.0 in experimental huts against natural populations of Anopheles gambiae s.l..

**Materials & Method:** The experiments were conducted at Vallée du Kou (an irrigated rice field area) in experimental huts. Unwashed and 20 times washed DawaPlus 3.0 and 4.0 LNs were evaluated in experimental huts for their effects on free-flying wild Anopheles gambiae s.l. mosquitoes in terms of mortality, blood-feeding inhibition, deterrence, exit rate. A polyester net coated with deltamethrin (DawaPlus 2.0) unwashed and 20 times washed was used as a positive control.

**Results & Discussion:** An average of 1627 female mosquitoes per treatment were collected over 63 nights of the trial. Unwashed DawaPlus 4.0 scored the highest mortality and high blood feeding inhibition rate through the trial then unwashed DawaPlus 3.0 (P < 0.05) respectively 43% and 22% for mortality versus 47% and 33% for blood feeding inhibition. Significant different was obtained between unwashed DawaPlus4.0 and positive control unwashed DawaPlus 2.0 (P = 0.0067). In this area, the vectors species An. gambiae s.l. has developed high level resistance to Pyrethroid through a combination of L1014F kdr and CYP6P3P450 mechanisms and other metabolic enzymes

**Conclusion:** Pyretroid resistance can be considered as the main threat to the continued control of many mosquito vectors of disease. PBO LLINs could confer significant additional protection in areas of moderate to high resistance where there are oxidase based mechanisms only and where the malaria prevalence is high.

Key words: Malaria, An. gambiae, Piperonyl Butoxide (PBO), insecticide resistance, bed net.

Current status of insecticide resistance in Malaria vectors in Tanzania: A wake up call for an urgent action to sustain malaria control.

Author: Theresia Nkya

**Background:** Long lasting insecticidal treated nets and indoor residual spraying with insecticides are the main malaria vector control tools in Tanzania. However, reports of insecticide resistance have been recorded over the past decade. This calls for investigation on the dynamics of malaria vector population and their resistance to insecticides. This study was a part of ongoing annual insecticide resistance monitoring on malaria vectors from Tanzania mainland.

**Methods:** Mosquito larvae were collected in 20 sites of Tanzania in 2017. Insecticide resistance levels were measured in adults exposed in bioassays to discriminating dosages of permethrin, deltamethrin, pirimiphosmethyl and bendiocarb. Synergy tests using PBO were conducted. An. gambiae sibling species and pyrethroid target-site mutations (kdr) were investigated. Insecticide resistance intensity assays were conducted for permethrin and bendiocarb in five districts.

**Results:** An. gambiae s.l. were resistant to pyrethroids, and only one site had the vectors demonstrating resistance to pirimiphos methyl. An. arabiensis was the predominant vector species in the country, with highest kdr west gene, while kdr east mutations had allelic frequency of up to 50% in all surviving mosquitoes. An. gambiae s.s. represented 31.3% of the sampled mosquitoes, with kdr west allelic frequency of 30% and kdr east allelic frequencies of up to 92%. An increased mean recovery of susceptibility to pyrethroids was observed when mosquitoes were pre-exposed to PBO. Presence of kdr east mutation in surviving mosquitoes was associated with occurrence of resistance phenotypes in deltamethrin. The intensity of permethrin resistance was high, with An. gambiae s.l. surviving as far as 5 times the normal diagnostic concentration.

**Conclusion:** High strength of pyrethroids resistance coupled with co-occurrence of kdr east and west mutations were recorded with high frequency in all sites with pyrethroid resistance. This is a wakeup call for urgent action to sustain malaria control.

Analysis of aggregated data from residual efficacy tests on mud and cement surfaces across 9 countries, against Anopheles mosquitoes for Fludora Fusion - an IRS combination product containing clothianidin and deltamethrin.

Authors: Justin McBeath, Andre Laas, Sebastian Horstmann, Frederic Schmitt Bayer.

**Background:** The residual life of IRS insecticides influences the ongoing economic viability of implementing IRS.

Variables such as the quality of application; characteristics of the insecticide product; resistance profile of the mosquito population and the type of surface to which the insecticide is applied, impact effective life of an IRS application. There is no substitute for locally generated data but analysis of residual performance across a wide range of surface types, using multiple different spray applicators and with a range of different mosquito strains can be useful in developing a predictive profile of an IRS insecticide.

**Methods:** Cone-bioassay mortality results from nine separate, small-scale trials (run by separate institutions) were used for this analysis. This included results from either experimental hut or small-scale village trials in Benin, Ghana, Madagascar, Rwanda, Senegal, Tanzania, South Africa, Zambia and Zimbabwe. Number of months above the WHO Threshold of 80% mortality was used as the measure of residual efficacy. Mortality assessments were made up to 72 hrs after exposure where necessary to take into account potential delayed mortality effects of clothianidin (consistent with recent WHO evaluations of this compound). For the purposes of this exercise, spray deposition accuracy was ignored as this was assumed to be representative of 'real-life' variability which could be experienced within an operational program.

**Results** : Fludora Fusion exhibited an effective lifespan on mud surfaces of between 4 and 12 months (mean of 9 months); in six of the nine trials the lifespan exceeded 6 months. On cement surfaces the effective lifespan was in the range of 5 to 12 months (mean of 10 months).

**Conclusion:** This data set illustrates the variability which is inherent across trials carried out in Sub-Saharan Africa. The breadth of this data set allows for a reasonable conclusion that Fludora Fusion exhibits strong residual activity on mud and cement surfaces.

Meaningfull engagement prior to community decision making – the case of engagement for contained laboratory work of Target Malaria Mali

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**Background**: Target Malaria is a not-for-profit research consortium working on a genetic approach to reduce the mosquito vector of malaria.

**Materials and methods:** After identifying key stakeholders the Target Malaria Mali stakeholder engagement team, carried out aseries of engagements using different tools such as visuals describing the technology and activities, focus group meetings, as well as insectary visits.

**Results and discussion:** The project step-by-step to achieve its goal. At each step the project wants to ensure that local communities are adequately engaged. The first step of the project in Mali would be to work in laboratory containment with a genetically modified mosquito strain in which males are sterile. Before making an application to the National Biosafety Committee , the Malian regulatory authority, the project wanted to ensure that the community had a sufficient understanding about this mosquito strain and about the concept of contained laboratory work, so that they could make an informed decision as well.

**Conclusion and recommendation:** This paper will show the importance of some key principles for engagement such as: early and progressive engagement, openness and co-development. The use of visuals demonstrated the importance of adapting the information to the audience to ensure that knowledge blocks were in place before moving to the next knowledge. Visits to the insectary have further informed the stakeholders about the research process.

Keywords: Target Malaria - Communication — Engagement – modified mosquitoes

This engagement aims to project activities so that stakeholders can make and informed decision.

# Posters

LaHoSA: A Proposed Simple Method for estimating the functional effectiveness of LLINs under operational conditions

Authors: Dennis J Massue, Olivier J.T Briet, Lena M Lorenz, Jason D Moore, Hans J Overgaard, Zawadi M. Mboma, John Bradley, William N Kisinza and Sarah J Moore

**Background:** High use of Long-lasting Insecticidal Nets (LLINs) in most malaria endemic countries has been associated with decline in malaria related mortality.

Information on the physical condition of long-lasting insecticidal nets (LLINs) under field conditions can only be true if correct measurements of their functional effectiveness are done. Current guidelines by the World Health Organization Pesticide Evaluation Scheme (WHOPES) recommended assessing physical integrity through counting of holes in four hole size categories and summarize them in a standard proportionate Hole Index (pHI) to provide a single measure of condition of each net tested. However, little is known on the influence of location of holes on the LLINs integrity, which could result into under/over estimation of the overall effectiveness of nets for personal protection. In this study we are proposing a simple method of assessing LLIN durability under operational condition called LaHoSA (Location Adjusted Hole Surface Area).

**Methods:** Three net brands, Olyset<sup>®</sup> net, PermaNet<sup>®</sup> 2.0 and Netprotect<sup>®</sup> were randomly collected from eight districts in Tanzania as part of a three-year prospective study, sub-sampled, measure their standard pHI and compare with the LaHoSA.

**Results:** Main findings shown that; 1) location of net damage is not uniform with bottom side having most damage than other sides and mosquitoes enters the nets though holes of different sizes and in different locations with unequal probability; 2) Roof side of the net was the high risky area for mosquito entry; 3) Due to the above findings, the probability of mosquito feeding, dying and surviving feeding at 24-hours vary by hole size and location.

**Conclusion:** The standard method for assessing LLIN durability using pHI is good. However with the risky weighted based on the findings above i.e. the location adjusted hole surface area, the overall pHI can be slightly refined more accurate and more simple to better approximate the net condition (based on the overall damaged surface area) under operational conditions.

# Abundance ¬ And Diversity Of Culex Mosquitoes In Four Habitats Types In Talangaye Rainforest, South West Region Of Cameroon

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**Introduction:** Deforestation is of major concern not only because of loss of species but also because of its impact on the future climate of our planet and its contribution to the emergence of diseases. However, little data exists on how deforestation in real time affects the overall species diversity in mosquito communities. This is an important problem considering the number of pathogens transmitted by mosquitoes, coupled with high rates of deforestation worldwide. We proposed to monitor the abundance and diversity of Culex mosquitoes in four different habitats with different levels of fragmentation and degradation.

**Methods:** The collection survey of mosquitoes was conducted in 2017 in a primary forest, disturbed forest, secondary forest and a degraded forest using net traps, sweep net, resting trap and larval dipping from breeding sites. The collected mosquitoes were identified to species with the aid of a stereomicroscope and morphological identification keys.

**Results:** A total of 2756 mosquitoes were collected (527 in the primary forest, 1077 in the disturbed forest, 675 in the secondary forest and 477 in the degraded forest with a significant difference (p<0.001) between the sites. A total of 24 species were collected in primary forest, 34 in disturbed forest, 24 in secondary forest and 22 in degraded forest.

**Conclusion:** With the highest mosquito abundance and diversity, a disturbed forest appears to be a convenient habitat for the development of mosquitoes. Disturbing a forest and allow it is therefore a dangerous activity that can favor vector-borne diseases.

Key words: deforestation, Culex, mosquito abundance, mosquito diversity

Setting up a Reference Lab for programmatic entomological surveillance

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**Background:** Entomological investigations are important for selecting appropriate vector control and measuring its impact. These studies provide information on malaria transmission, spatio-temporal patterns of vector species, and their bionomics and susceptibility to insecticides. In Zambia few institutions have the resources, infrastructure or staffing to analyze mosquitoes to provide these data. In response to this need, Macha Research Trust (MRT) established a reference laboratory to analyze mosquitoes for species identification, blood meal analysis, and infectivity using both morphological and molecular methods.

**Materials and Methods:** Initially determination of the market was undertaken. During meetings with malaria progamme officers and partners, a need was identified for downstream processing of field caught mosquitoes. Visits from stakeholders allowed viewing of MRT's capabilities. During these consultative meetings, a list of standard assays was drawn up and protocols established to include mosquito species identification, PCR determination of blood meal host, and ELISAs to detect sporozoites in mosquitoes. A review of equipment needed was undertaken, and of those available at MRT, specific machines were assigned to the reference lab. A bench fee was included in costings to account for space, equipment use and maintenance costs. A work flow was established to avoid contamination. Staff solely dedicated to the reference lab were recruited and trained. Vendors, able to source and ship all supplies to Lusaka were identified and quotes compared. From this a cost per sample was calculated.

**Results and discussion:** Challenges identified during set up included balancing client demand and timeliness of analyses, delays in finalizing contracts, processing damaged samples and staff availability. These challenges have now been addressed and MRT actively supports entomological surveillance in Zambia at an affordable cost while ensuring client data is protected.

**Conclusion.:** Leveraging existing infrastructure and capacity, client engagement and laboratory standards, MRT successfully setup a reference laboratory for programmatic entomological studies.

# Comparison Of The Mosquito-Electrocuting Trap And Human Landing Catch Techniques As A Measure Of The Human Landing Rate Of Host-Seeking Aedes Aegypti In The Semi-Field System

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**Background:** Human landing catches (HLC) whereby a human collect host-seek mosquito over a set of time is a standard method used to measure human landing rate. However, there are ethical concerns associated with exposing human to potentially infectious mosquitoes. Thus, new or improved methods of mosquito collection that are equivalent to HLC are needed. Recently, the Mosquitoes Electrocuting Trap (MET) was evaluated against anopheles mosquitoes and found to catch the same number of mosquitoes as HLC. We conducted a semi field experiment to compare the efficacy of the MET and HLC to catches laboratory reared Aedes aegypti mosquitoes.

**Methods:** A Latin square design was used. This study was conducted for the period of sixteen days in the semi field system located in Bagamoyo, Tanzania. Treatments were randomly assigned to the separate compartment of the semi field system and rotated after every two days. Four volunteer were recruited and were rotated daily. One hundred laboratory reared Aedes aegypti mosquitoes were released in each compartment at 09.00hrs. Mosquitoes were collected for three consecutive hours in both compartments. Data were analyzed by generalized linear mixed model (GLMM) using a binomial distribution with logit link for proportional data and Bland and Altman plots.

**Results:** Results showed that the proportion of the caught mosquitoes by HLC were 62% (95% CI 58-60) compared to 43% (95% CI 34-69) caught by MET. Analysis of the GLMM showed that when a person is collecting mosquitoes using MET, the proportion of the collected mosquitoes was reduced by 54%(OR=0.46 (0.37-0.56), p=0.001). The bland and Altman analysis showed that the observed different is systematically and the MET results can be used to predict the proportion caught by HLC.

**Conclusions:** We concluded that the MET could be used as an alternative to HLC for catching Aedes aegypti mosquitoes in the semi-field condition.

Re-evaluating the coverage indicator for Indoor Residual Spraying (IRS): Who are we truely protecting in malaria control and elimination?

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The great achievements seen in malaria transmission reduction since 2000 have been largely attributed to the increase in coverage with indoor residual spraying (IRS) and long lasting insecticide treated nets (LLINs) alongside treatment and improved diagnosis. The Global Technical Strategy 2016-2030 for malaria elimination has set out ambitious goals to reduce malaria incidence and mortality by 90 % by 2030 globally through increased coverage and access to malaria interventions including vector control. While malaria vector control with IRS and LLINs remains the backbone on the road to elimination, their effectiveness relies upon but not limited to factors such as insecticide active ingredients selected, longevity of the tool, vector behaviors and insecticide susceptibility, but also high acceptance and use rates by the users. While the latter – human behavior – is well studied when it comes to LLINs, IRS is performed without tracking adherence over time. In this paper, we highlight some of the aspects of how human behaviour can change IRS coverage following a successful IRS campaign and how such changes can undermine the effectiveness of IRS and hence malaria transmission intensity consequently attenuating malaria elimination goal.

# Persistent community Plasmodium prevalence after control of indoor biting Anopheles: a threat for malaria upsurge after halting indoor residual spraying in Tororo District, Eastern Uganda

Authors: David W. Oguttu<sup>1\*</sup>, David C. Okumu<sup>2</sup>, Alex R. Ario<sup>5</sup>, Charles Ntege<sup>3</sup> and Michael Okia<sup>41</sup>

**Background:** Since 2014, use of long lasting insecticide treated nets (LLINs) and indoor residual spraying (IRS) have controlled indoor resting Anopheles, but Plasmodium transmission continues. We assessed the trends of malaria, indoor resting vector density and determined community Plasmodium prevalence after four rounds of IRS.

**Methods:** We analyzed Health Management Information System (HMIS) malaria surveillance data of 2,727,850 patient records in 61 health facilities from 2012 to 2015 to estimate monthly malaria incidence for the entire population and also separately for <5- and  $\geq$ 5-year-olds before and after introduction of vector control interventions and determined laboratory test positivity rates. We tested 100 residents per village(n=20) for malaria using Rapid Diagnostic Tests (RDT) and microscopy. We classified malaria positive individuals as symptomatic or asymptomatic. We asked people about mosquito bites before sleeping under LIINs. Indoor resting Anopheles density was monitored monthly. Chi square and logistic regression were used for trend analysis.

**Results:** Mean monthly malaria incidence fell from 95 in 2013 to 76 cases per 1000 in 2014 (OR=0.99, 95% CI=0.96-1.01, P=0.37). Among children <5 years, the malaria incidence reduced from 100 cases/1000 in 2014 to 45/1000 in 2015 when IRS was introduced (OR=0.94, 95% CI=0.91-0.996, P<0.0001). Malaria RDT positivity rate reduced from 57% in 2013 to 30% (Chi =15, P<0.0001) in 2015. Slide positivity rate reduced from 45% in 2013 to 21% in 2015 (P=0.004) while RDT positivity declined from 69% to 40%. Asymptomatic community malaria prevalence after four rounds of IRS was 14%; higher among children <5 years (21%) than adults (7.0%). Indoor Anopheles resting density reduced to zero. 90% of people stay exposed to malaria vectors before sleeping under LLINs.

**Conclusion:** Moderate Plasmodium prevalence and changing biting behavior of Anopheles present a threat for malaria upsurge after stopping IRS. Active malaria testing and treatment should be considered in communities to remove the parasite source.

Keywords: Malaria, Prevalence, threat, vector control

# Evaluation Of Visual Blood Meal Detection Among Malaria Vector Anopheles Species During Routine Entomological Surveillance In Manicaland, Zimbabwe

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**Background**: Accurate identification of Anopheline malaria vectors and determination of their host feeding behavior are critical steps in establishing effective vector control interventions. Most studies on vector behavior conduct mammalian host blood meal preference assays only on mosquito samples that are visibly blood fed. However, some blood meals may be missed by eye that can give valuable vector feeding behaviour data. The current study assessed the reliability of identifying blood-fed mosquitoes visually using blood meal identification PCR as reference.

**Methods:** A total of 150 mosquito samples from Chakohwa and 64 samples from Burma Valley were analyzed. Mosquitoes that were collected using pyrethrum spray catches and CDC light traps were visually rated for blood feeding status by experienced field entomologists. After dissection to separate head and thorax from abdominal sections, the latter were subjected to DNA extraction using the Marriott approach followed by host mammalian blood meal detection PCR on all the extracts.

**Results**: The prevalence of blood-fed mosquito field specimens identified visually (27%) was substantially lower than detected by PCR (59%), which had 3.9X higher odds of detecting a positive blood meal than the "eye-ball" approach (OR [95% CI]: 3.89 [2.579 – 5.867]; 2 = 43.742, df = 1, p< 0.001, N = 418). Visual blood meal detection showed poor sensitivity (42%) compared to PCR, which was tantamount to a false negative rate of 58% [95% CI: 49.3 – 66.4%].

**Conclusions:** Visual identification of blood meals on mosquito specimens has a high false negative rate that may lead to bias or loss of blood feeding surveillance information as majority of specimens are classified as unfed by eye. It is recommended that PCR analysis of vector blood meal preferences be based on all field collected adult mosquitoes regardless of their visual blood meal appearance.

Influence of house characteristics, usage of treated nets on mosquito distribution and malaria transmission in the city of Yaoundé, Cameroun

Author: Ngadjeu CARMENE SANDRA

**Background:** Malaria elimination required additional tools to complement mosquito net and indoor residual spraying. House characteristics could improve protection against malaria vectors and malaria transmission. A study was conducted to assess the influence of house characteristics on mosquitoes' abundance and malaria transmission.

**Method:** The study was carried out from March to June 2018 in the city of Yaoundé. We performed mosquito collection in 105 houses using CDC Light Traps during three consecutive nights. Data on house characteristics were recorded after interviewing the head of household. Mosquitoes collected were identified morphologically down to the genus and species level using morphological identification keys. All anopheline collected were analysed through ELISA or Taqman PCR to detect their infectivity to Plasmodium.

**Results:** A total of 8,883 mosquitoes were collected, with higher densities indoor (n=7,237) than outdoor (n=1,646). Among the Anopheline, An. gambiae s.l. was the predominant species (79.21%). 154 An. gambiae s.l were tested, 3 were found to be infected. Houses with a high number of people ([6-10]) registered high densities of An. gambiae s.l. (mean= 0.45; 3 infected) than those with less than 6 peoples (mean= 0.37). Mosquito densities were high in presence of LLINs (mean= 0.42; 2 infected) than in their absence (mean=0.32). Densities of An. gambiae s.l. varied with houses characteristics. An. gambiae was highly prevalent in houses made of mud (mean= 0.9) than in houses made of wood (mean= 0.53) and cement (mean= 0.32). Houses with open eaves and open ceilings recorded a high number of An. gambiae s.l. (mean= 0.53; 2 infected) than those with closed eaves and ceilings (mean= 0.38). An. gambiae s.l. densities also varied with the location of house.

**Conclusion:** The study highlight that houses conditions could influence exposition to malaria transmission risk in our study sites.

Keywords: Malaria, transmission, Anopheles gambiae s.l., houses characteristics, Yaoundé.

Heterogeneity in residual transmission and the consequence for the efficacy of net and spray interventions in areas with pyrethroid resistant mosquitoes

Authors: Ellie Sherrard-Smith, Tom S Churcher

**Background:** The anti-malarial efficacy of key vector control interventions – long-lasting insecticidal nets (LLINs) and indoor residual spraying (IRS) - are reliant on technologies that interrupt mosquito bites on people by exploiting the night time, indoor feeding patterns of primary African vectors. Residual transmission is the remaining transmission that happens beyond the reach of IRS and LLINs when used optimally. This can be the consequence of mosquitoes feeding outdoors or outside the time of day when nets and sprays can protect people. There is also a potential loss of effectiveness due to increasing physiological resistance; the ability of mosquitoes to overcome toxic insecticidal effects.

**Methods:** A systematic review of mosquito and human behaviour is used to inform an established transmission model for malaria that is parameterised to model physiological resistance to pyrethroid insecticides. The public health impact of residual transmission across Africa was estimated and explored in the context of pyrethroid resistance.

**Results:** Data indicate spatiotemporally heterogeneous outdoor biting for key malaria-transmitting mosquitoes and this can identify places where indoor interventions like LLINs and IRS may perform less well. The model predicted an additional 44 million clinical cases in Africa across the 3 years following universal implementation of nets and sprays at 80% coverage, if mosquito bites happening indoors were to decrease from c.97% to just c.92%.

**Conclusions:** We characterised the entomological efficacy of the most widely used IRS compounds and show how spray chemistries with alternative mechanisms of action to those of pyrethroids can mitigate, to an extent, the lost efficacy of LLINs in places with behavioural and pyrethroid resistance. Behavioural characteristics of both people and mosquitoes affect the efficacy of indoor interventions and determine the local challenge of residual transmission.

Challenges to malaria elimination: Community compliance, strategies of prevention, and therapeutic itineraries for febrile illness (sumaya) in the Comoé Province, Burkina Faso.

Author: F.Guglielmo, H.Ranson, C.Jones

**Background.** In Burkina Faso, high rates of bednets coverage and increased usage have not prevented as expected the overall rising of malaria infections. Vector insecticide resistance and behavioural changes, together with patterns of human exposure to bites are among the reasons behind this increase. The persistence of a human reservoir, however, is fostered by delays in treatment, which can only be addressed by understanding communities' preventive strategies, adherence to biomedical treatment, and their rationale behind therapeutic choices.

**Methods.** Ethnographic fieldwork was conducted for 13 months, between 2017 and 2018, in three rural communities in the province of Banfora. A mixed-method approach was used to collect data on human exposure and on preventive measures against mosquito bites. Individuals' therapeutic choices, perception of risk and efficacy of treatments, were explored through interviews and focus group discussions with heads of households and caregivers.

**Results and Discussion.** Access to health centres (proximity, costs), perceived efficacy of treatment with relation to vulnerability of the ill, and intrahousehold power relations affected therapeutic choices for febrile illness (sumaya). At the beginning of the rainy and peak transmission season (May-Aug), we recorded higher reticence to seek biomedical care due to farming and lack of financial means, employed during the dry season (Feb-May) for the celebrations of marriage and funerary rituals. Age was an additional factor. Younger individuals were perceived as more vulnerable and biomedical assistance more appropriate.

**Conclusions.** Interventions aimed at reducing the human reservoir of malaria currently rely on passive community participation, such as acceptance of domestic tools of prevention, neglecting the micro-dimensions in which therapeutic choices are made. These dimensions, however, are the framework in which interventions should be developed in order to be both efficient and sustainable.

#### Catch Composition And Abundance Of Aedes Species In Adamawa State, Nigeria

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#### Abstract

Aedes mosquito species are responsible for transmission of several important viral infections. Therefore, this study focused on the occurrence, distribution and abundance of Aedes species in communities displaced by lingering upheaval in the north-eastern region of Nigeria. Aedes species were sampled from three locations namely; Mubi, Yola and Numan towns from July to October, 2017. They were collected using human bait methods and identified to species level using microscopy and morphological identification keys of Hopkins. The abundance of each species of Aedes identified was expressed in percentages while diversity was revealed using Shannon-Wiener diversity index. A total of 768 mosquitoes were examined, higher abundance of Ae. aegypti species were observed in Numan (53.3%;176/330), while lower species occurrence was recorded in Mubi (12.7%; 42/330). The abundance of Ae. africanus was highest and lowest in Yola (66.4%;146/220) and Numan (12:7%; 28/220) respectively. Aedes hensili showed higher abundance in Yola (43.1%; 88/204) while lower number (16.7%; 34/204) was recorded in Mubi. Also, higher occurrence of Ae. triseriatus (77.8%:7/9) was seeing in Numan whereas it was absent in Mubi. There was higher occurrence of Ae. albopictus (40%:2/5) in Mubi and Yola, whereas lower species abundance was recorded in Numan (20%:1/5). However, Shannon index showed higher diversity and evenness (H'= 0.06; e^H/S = 0.96) of Ae. albopictus, followed by Ae. hensili (H'=1.03; e^H/S=0.93) and Ae. aegypti (H'=0.96; e^H/S=0.87) while lower abundance of Ae. triseriatus (H'= 0.53) was observed in the three locations. This study indicates that Ae. aegypti, Ae. africanus Ae. hensili, Ae. triseriatus and Ae. albopictus are abundant and well diverse. The large distribution and evenness of Ae. albopictus observed may result from the increase in international travel and urbanization. This posed a potential danger of spreading infections like dengue, yellow fever, chikungunya and zika viruses.

Key words: Abundance, diversity, dengue, chikungunya, Aedes, Nigeria.

# Repellent-Treated Eave Wraps As A Tool In The Arsenal Against Residual Malaria Transmission

Authors: Margaret M. Njoroge<sup>1, 2,</sup> Alex Hiscox<sup>2</sup>, Adam Saddler<sup>3, 4,</sup> Ulrike Fillinger<sup>1</sup>

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Tools that have largely been used in mosquito control; namely long lasting insecticide-treated nets and indoor residue spraying have contributed to the reduction in malaria transmission over the last 15 years. While tremendous gains have been achieved, increase in insecticide-resistant mosquitoes threatens further progress with the need for alternative control methods becoming more urgent. Exploring tools that are not reliant on pyrethroids, which target outdoor populations of mosquitoes, could supplement the existing tools which are mainly used indoors as well as address the rising concern of residual malaria transmission. Para menthane-3, 8-diol (PMD) was tested as a possible spatial repellent for the protection of humans from outdoor-biting mosquitoes.

PMD was microencapsulated in cotton fabric at 55g/m2. Experiments were implemented in large semi-field systems at the International Centre of Insect Physiology and Ecology, Mbita, western Kenya. Treated fabric was wrapped around the eaves of experimental huts built inside the semi-field system whilst untreated fabric was wrapped around hut eaves in a second system serving as control. Insectary-reared female Anopheles arabiensis were released in the screen houses. Human landing catches were conducted in the early evening during peak mosquito-biting hours by volunteers seated 2.5m from the experimental huts. Pyrethroid susceptibility of the insectary-reared mosquitoes was measured.

More than 80% of all released mosquitoes were caught within the first two hours in both the treated and control set up. Human landing rates in the screen house with the PMD treated huts were not significantly different to those in the control set-up (p=0.9952). There were no significant differences in the catches depending on point of release of mosquitoes. Insecticide susceptibility tests showed 92.5% susceptibility to 0.05% deltamethrin which is categorized by WHO as suggestive of the existence of resistance.

Initial studies on the possibility of using PMD as eave wraps for protection against outdoor-biting mosquitoes showed no protection. Studies are ongoing to further investigate this method with alternative repellents and to explore whether PMD could be effective at different concentrations.

Key words: Residual malaria transmission, spatial repellent, Anopheles arabiensis, pyrethroid-resistance

Genomic and Molecular Characterization of the Bacterial Populations observed in the water of breeding sites and in larvae of Anopheles coluzzii and Anopheles gambiae in Nanguilabougou and Kouroubabougou.

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**Abstract:** The microbiota composition of mosquitoes is influenced by their aquatic breeding environment. The exact factors that define the structure of the mosquitos' microbiota including malaria vectors are currently unknown.

From June 2014 to November 2015 we conducted genomic and molecular characterization study of the bacterial populations observed in the breeding sites water and larvae of An. gambiae and An. coluzzii the two majors vectors of malaria in Kouroubabougou and Nanguilabougou. The samples were collected from two breeding sites with different ecological settings. The inventory of the bacterial populations, was conducted to better understand their frequencies distribution in larvae and in their respective breeding sites water. Frequencies of the identified bacteria were associated with the allelic polymorphism of the TEP-1 gene in An. coluzzii and An. gambiae.

We collected 149 samples in which we identified 11 species, 49 genera and 82 strains. The bacterial species populations were composed of Bacillus sp. The species Bacillus anthracis and Bacillus thuringiensis were present only in An. coluzzii in Nanguilabougou and Kouroubabougou. Bacillus cereus, Bacillus amyloliquefacins and Bacillus subtilus were observed only in Nanguilabougou and in An. coluzzii. Enterobacter cloacae and Enterobacter ludwigii were present in An. gambiae with the S1/S1 TEP1 genoptype in Nanguilabougou. Bacillus cereus, Bacillus amyloliquefaciens and Bacillus subtilis were more associated with the TEP1 genotype of S1/S1 in An. gambiae in the village of Kouroubabougou.

In this study we used the classical method of bacteria culture and the associated molecular, genomics and bioinformatics tools to identify and characterize the bacterial populations. An optimization of the method for future studies would involve the metagenomics analysis approaches to increase the maximum size of identified bacteria species and strains. This will help to determine, the factors that might promote a specific microbiome associated with the population-dynamics of An. coluzzii and An. gambiae in Mali.

# A semi-field trial test on the efficacy of transgenic antimalarial biocontrol fungi in Burkina Faso

Authors: Etienne Bilgo<sup>1\*</sup>, Brian Lovett<sup>2</sup>, Issiaka Saré<sup>1,</sup> Roch K. Dabire<sup>1</sup>, Raymond St Leger <sup>2</sup> and Abdoulaye Diabaté <sup>1</sup>

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**Background:** An estimated 2 billion people live in areas where mosquito-borne diseases including malaria, filariasis, viral encephalitides, dengue and yellow fever are endemic. It was recently established that the insect pathogenic fungus Metarhizium has the potential to control adult mosquitoes in a field setting, but would work much better if its potency were increased. Here we report on a semi-field trial testing the efficacy of a mosquitocidal Metarhizium strain (Met-Hybrid) engineered to express an insect-specific spider neurotoxin (Hybrid) and GFP.

**Methods:** The experiments were conducted in a multi-chambered MosquitoSphere (a contained, near-natural environment) constructed for this purpose in a region of Burkina Faso, Soumousso where malaria is endemic. We used the MosquitoSphere to test a variety of low technology treatment protocols that could be used routinely by householders and found that suspending Metarhizium in locally produced sesame oil and spreading that on netting or black sheets achieves a long-term effect in the sphere.

**Results:** Compared to a strain of wild-type virulence expressing RFP (Met-RFP), Met-Hybrid kills anopheline mosquitoes in half the time and at much lower spore doses, which increases the efficacy and the effective persistence of the fungus. We also demonstrate that Met-Hybrid had important pre-lethal effects that include reduced blood feeding and flight capacity by infected mosquitoes. We also demonstrated that when the transgenic fungus is used in combination with pyrethroid insecticides, it synergistically improved the susceptibility of insecticide-resistant mosquitoes.

**Conclusion:** Transgenic fungi show considerable promise as a vector control technique. We aim to soon deploy these transgenic fungi by anticipating social, political and scientific hurdles to their approval and acceptance. Our results have broad implications for any project proposing to scale up new, complex, and potentially controversial technologies for malaria eradication.

# A genomic study of diverse outdoor-biting Southeast Asian malaria vectors

Authors: Brandyce St. Laurent<sup>1</sup>, Chris Clarkson<sup>1</sup>, Nick Harding<sup>2</sup>, Michael Fontaine<sup>3</sup>, Jorge E. Amaya Romero<sup>3</sup>, Robert Waterhouse<sup>4</sup>, Sonia Goncalves<sup>1</sup>, Jim Stalker<sup>1</sup>, Mara Lawniczak<sup>1,</sup> Dominic Kwiatkowski<sup>1,2</sup>, Alistair Miles<sup>2,1</sup>

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**Background:** Understanding the roll of outdoor-biting and secondary vector species in complex malaria transmission environments is crucial to reducing and eliminating malaria. Genome sequencing could provide important insights, but as yet no population genomic studies have been performed on Southeast Asian anophelines, or outdoor biting vector species.

**Methods:** We describe results from a pilot study using Illumina deep whole-genome sequencing of 128 wildcaught individual Anopheles mosquitoes representing 27 different species, collected from Cambodia. We investigated the suitability of using available reference genomes, even for specimens without a close reference available. To investigate the taxonomic relationships between these individuals, we used three approaches: 1) de novo assembly of mitochondrial genomes; 2) alignment of reads to a selection of sequences highly conserved across Anopheles; and 3) reference-free analysis using k-mer composition. We also investigated the accessibility of specific genes of interest, such as insecticide-resistance markers.

**Results:** Using multiple whole genome analyses, we have produced a revised phylogeny of these diverse vector species and alignments of known insecticide resistance markers. Whole genome sequence analyses of non-traditional vector species will help to lay the technical foundations for investigating population structure and gene flow of a multitude of malaria vectors worldwide.

**Conclusions:** These data will be publicly available as part of the MalariaGEN Vector Observatory, an open access resource of genome sequence data. Genomic analysis of diverse species of outdoor-biting and secondary vectors can inform control programs to best target residual malaria transmission across the tropics.

Influence of Physico-chemical and biological parameters on the distribution of anopheline larvae in the city of Yaounde.s

# Author: Nelly Armanda

**Background & objectives:** Malaria remains a major public health problem in Cameroon and the city of Yaounde is not saved. The main difficulty of vector control remains their adaptability in diverse environmental conditions. We undertook a study with main objective to determine the influence of bioecological parameters affecting the distribution of anopheline larvae in that city.

**Methods:** Prospections of anopheline larvae breeding sites were done during four seasons (from May to December 2017), in eight neighbourhoods of the city of Yaounde. Breeding sites were geo-referenced using a GPS (Garmin) and were physically characterised. For each cluster per collection period, ten breeding sites (five positive and five negative) were retained for the measure of physico-chemical parameters using a Wagtech (9100) portable kit. Those parameters were then correlated to the presence and the density of anopheline larvae.

**Results:** The obtained results revealed that, Nkolbisson, Ekounou-palais and Tsinga 8e were points with high density of anopheline larvae. The presence of anopheline larvae in a breeding site was positively and significantly correlated with temperature, turbidity, pH and distance to the nearest house while it was negatively correlated with the presence of predators, the size and the depth of the breeding site. However, pH, conductivity, temperature, turbidity and Calcium was significantly correlated with larval density contrarily to the presence of predators, the surface area and presence of vegetation around the breeding sites negatively impacted on larval density. Two sibling species of Anopheles gambiae s.l were identified using SINE PCR: Anopheles gambiae ss and Anopheles coluzzii. Cases of sympatry were observed between the two sibling species in about 22 breeding sites.

**Conclusion:** Parameters as seasons, clusters, temperature, turbidity, pH, conductivity, Calcium, distances to the nearest should be used in larvae control program.

Key words: influence, Physico-chemicals, biological, distribution, anopheline larvae, Yaounde

# Insects to feed insects: Larval diets based on insects for mass-rearing major Aedes vectors of Zika and Dengue

Authors: Nanwintoum Séverin Bimbilé Somda, Hamidou Maiga, Wadaka Mamai, Hanano Yamada, , Antoine Sanon, Abdoulaye Diabaté, Kounbobr Roch Dabiré, Jérémie Gilles, Jeremy Bouyer,

**Background & objectives:** Malaria remains a major public health problem in Cameroon and the city of Yaounde is not saved. The main difficulty of vector control remains their adaptability in diverse environmental conditions. We undertook a study with main objective to determine the influence of bioecological parameters affecting the distribution of anopheline larvae in that city.

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**Conclusion:** Parameters as seasons, clusters, temperature, turbidity, pH, conductivity, Calcium, distances to the nearest should be used in larvae control program.

Key words: influence, Physico-chemicals, biological, distribution, anopheline larvae, Yaounde

Polymorphic Inversion 21a Frequencies Associate With Ecotypes But Indicate Little Genetic Differentiation Between Populations Of Anopheles Coluzzii In Southwest Nigeria

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**Background:** Chromosomal paracentric inversions play crucial role in local adaptation of plants and animals. In disease transmission like malaria, it plays a key role in the ecological plasticity of the vector, Anopheles, enhanced by high amount of polymorphic inversions. Of importance is the polymorphic inversion 2La in Anopheles gambiae, combination of which suggests adaptive potential for inversions. In Nigeria, however, data on the role of this inversion in the selection of local Anopheles populations has not been available for over three decades. We therefore examine the pattern of inversion frequencies of 2La across ecotypes in Southwest Nigeria.

**Methods:** A minimum sample of 100 female Anopheles mosquitoes from each area were collected from eleven localities across three ecotypes (Mangroove, Rain forest and the Savannah regions) in Southwest Nigeria. The mosquitoes were first identified morphologically and then with PCR and restriction digest. Inversion 2La PCR was conducted on a minimum of 30 identified Anopheles coluzzii of the mosquito populations from each locality. Observed and expected frequencies of the paracentric inversion 2La for each locality was subjected to Hardy-Weinberg.

**Results:** All the mosquito samples collected were identified as Anopheles gambie s.l.. Further identification with PCR showed varying percentages of Anopheles coluzzii (31-100%) in the mosquito populations from each locality. A total of 333 Anopheles coluzzii were successfully scored for inversion 2La karyotypes. The frequencies shows an excess of heterokaryotypes across all localities, which indicate positive heterosis within the populations. The frequency of the standard arrangement 2La increased progressively from the mangrove/forest region to forest/savannah while that of the inverted arrangement 2La+ decreased. This pattern suggests an association with ecotypes. Inversion frequencies from all the populations were within Hardy-Weinberg estimates and this reaffirms the association with ecotypes.

**Conclusions:** This study therefore reports a low level of genetic differentiation between populations of Anopheles coluzzii in Southwest Nigeria which is primarily driven by adaptation to different ecotypes.

Keywords: Anopheles Coluzzii, Genetic Differentiation, Heterokaryotypes, Inversion 2la

# Results from a comparison-control trial examining different targeting strategies for IRS, Zambia 2017

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Indoor residual spraying (IRS) is a powerful intervention in the fight against malaria, but high cost prohibits most national malaria control programs from achieving universal IRS coverage. Most countries rely primarily on universal coverage of insecticide-treated mosquito nets (ITN) for malaria control and target IRS campaigns to specific prioritized areas. However, there is limited information about how IRS should be delivered to maximize impact and prevent the most malaria cases and deaths with the limited resources available. In collaboration with the National Malaria Elimination Centre in Zambia we conducted a comparison-control trial in 2017 and 2018 to evaluate different prioritization strategies for IRS. Six districts were divided into three groups (2 districts per group), with each receiving a different IRS targeting methodology for their IRS operations (pirimiphos-methyl) in 2017. Group A received a geographic concentration strategy wherein all structures within a geographic area were targeted for IRS. This method (blanket spraying) aims for heavy saturation of IRS without spatial gaps. Group B used health facility incidence data from the health management information system (HMIS) to estimate malaria burden in each area and prioritize houses according to incidence at the nearest facility. Group C used a strategy of ecological targeting to allocate IRS based on predicted probability of Anopheles funestus as developed by the Malaria Atlas Project. IRS was implemented in late 2017, and all data will be analyzed by July 2018. The final IRS coverage for the 2017 spray campaign reached 91% in the targeted areas in the six study districts. The primary outcome is the incidence of uncomplicated malaria as measured through the HMIS which will be analyzed using a difference-in-differences approach. The entomological inoculation rate (EIR) of local primary vector species is being measured through five routine entomological collections in 150 houses across the different arms and will serve as a secondary outcome. EIR will be assessed using a post-only comparison between trial arms as well as between sprayed and unsprayed areas. Analyses will be finalized by September 2018. This presentation will discuss the results of this study and its implications for future targeting methodologies in Zambia and other countries using IRS.

# Implication of Anopheles. funestus in malaria transmission in the city of Yaoundé Cameroon

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# Abstract

Despite its status as a major malaria vector in Africa, Anopheles funestus contribution to malaria transmission in the urban environment is still not well documented. The present study assesses the implication of An. funestus in malaria transmission in the city of Yaoundé.

The study was conducted in both urban and periurban districts of the city of Yaoundé. Adult mosquitoes were collected using CDC Light Traps and human landing collections from April 2017 to March 2018. After morphological and molecular species identification, the heads and thoraces of Anopheline mosquitoes were analyzed to determine their infection status using Taqman method or ELISA. Bioassays were conducted with 2-5 days old females to determine their susceptibility status to permethrin, deltamethrin and DDT.

A total of 7,136 mosquitoes including anopheline and culicine were collected. High species diversity was recorded in the periurban district of Mendong with six species collected An. gambiae, An. coluzzii, An. funestus, An. leesoni, An. ziemannii and An. marshallii whereas only four were recorded in the urban district of Nsam (An. gambiae, An. coluzzii, An. funestus, An. leesoni). Anopheles gambiae sl emerged as the most abundant anopheline species in Nsam (88.3% of the total) whereas, An. funestus was the most abundant in Mendong (55.3% of the total). An. funestus was found to be more prevalent at the onset of the small rainy season and was recorded biting frequently indoor whereas An. gambiae was found to bite more frequently outdoor. An. funestus infection rate varied from 2.94% to 4.7% in both sites. An. gambiae display infection rates of 1.9% in Nsam and 5.73% in Mendong. An. funestus was responsible for up to 41% of malaria transmission estimated at 89.3 infected bites/man/year at Mendong and of 3% of the annual entomological inoculation rate at Nsam (78.4 infected bites/man/year). An. funestus was found to be resistant to DDT, permethrin and deltamethrin.

The study provides evidences supporting the implication of An. funestus in malaria transmission in the city of Yaoundé. These findings highlight the need to tackle this species to maximize control strategies in this city.

Keywords: Malaria, An. funestus, An. gambiae, urban, peri-urban, Yaoundé, Cameroon

# Urbanization and dynamics of Anopheles. gambiae sl larvae in the city of Yaoundé (Cameroon)

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The rapid urbanization coupled with poor environmental management in sub-Saharan Africa has led to a high proliferation of Anopheline mosquitoes in the urban environment. The present study aim to assess the spatio-temporal distribution of anopheline breeding sites and the influence of physico-chemical parameters on mosquito distribution in the city of Yaoundé.

Anopheline larvae breeding sites were surveyed and water samples analyzed from March to August 2017 in 32 districts of Yaoundé. Water collections characteristics such as the size, distance to the nearest house, presence of vegetation alongside physico-chemical parameters such as turbidity, pH, temperature, conductivity, sulfates, orthophosphates, hydroxide peroxide, conductivity, iron and calcium were recorded and analyzed according to anopheline larvae presence or absence. All breeding sites were geo-referenced during each field collection to assess the productivity and sites distribution using GIS techniques.

A total of 9,673 aquatic habitats were sampled, 4,191 habitats were found with either culicine and/or anopheline larvae. Only 1,883 sites (19.47% of sites) contained anopheline larvae. Most of the breeding sites sampled had first or second instar larvae 76.9% (n=1,448). A high number of sites ( $\geq$ 30%) presented both early and late instar larvae. Correlation analysis suggested that the presence of vegetation was positively correlated to the presence of anopheline larvae. Comparison of physico-chemical parameters between sites with or without anopheline larvae did not demonstrated any significant difference. Standing water pools were the most common breeding sites in the city. Both Anopheles coluzzii and An. gambiae were recorded. Geographic characterization of breeding sites using GIS techniques demonstrated that from one season to the other, lowland areas were the places contributing most to anopheline presence and the most productive sites were always located in the same places.

The data confirms high abundance of anopheline larvae in the city of Yaoundé and a high capacity of adaptation to different types of habitats. The study highlights the need for an integrated approach in order to fight efficiently against malaria transmission in the city of Yaoundé.

Key words: Larval habitats, Anopheles, malaria, GIS, dynamic, Yaoundé

Description of immunity and detoxification related genes in Anopheles funestus and Anopheles gambiae s.s. sialotranscriptome using RNA-seq analysis.

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**Background and objective:** The genetically determined preference for blood meals on human, exposes Anopheles mosquitoes to two important challenges during their lifespan: surviving to plasmodium infection and resisting to antivectorial strategies. Some of the mechanisms that help malaria vectors to face these challenges are based on the expression of a set of immunity genes that can affect the plasmodium development and detoxification genes hydrolysing insecticides used for vectors control. The present study aimed to describe immunity and detoxification related genes present in the salivary gland of An. funestus and An. gambiae s.s, two African major malaria vectors.

**Material and methods:** Fed An. funestus and An. gambiae s.s female mosquitoes were collected in the field in the locality of Obout in Cameroon. Illumina sequencing technique was performed to generate and analyse the transcriptome profile of salivary gland extracted from salivary glands from F1 mosquitoes aged from 3 to 5 days.

**Results and discussion:** A total of 40.728.724 and 37.832.568 clean reads were obtained from the sialotranscriptome of An. funestus and An. gambiae respectively. Assembly and annotation led to 5.233 annotated unigenes for An. funestus and 7.165 for An. funestus. Among the 167 and 257 immunty related genes detected in An. funestus and An. gambiae s.s respectively, the most abundantly families identified were Clip- domain serine protease, leucine-rich immune protein and C-Type Lectin. Regarding detoxification genes, among a total of 95 and 117 in An. funestus in An. gambiae, only cytochromes P450s and Gluthation-S-transferase superfamilies were identified whereas, no carboxylesterase was identified.

**Conclusion and recommendations:** This study described immune and detoxification related genes identified in the salivary glands of An. funestus and An.gambiae mosquitoes. Our findings give important information for further studies to understand how malaria parasite invade salivary gland in the context of insecticide resistance.

Keywords: Anopheles; Sialotranscriptome; Immunity genes; Detoxification genes; RNA-seq

Competence of the secondary vectors An. coustani, An. squamosus and An. rufipes for Plasmodium falciparum as measured by direct membrane feeding assays.

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**Background:** In West Africa, An. rufipes, An. squamosus and An. coustani have been reported to be possible secondary malaria vectors but their intrinsic competence for P. falciparum has not been experimentally examined.

**Methods:** We infected F1 females of wild-caught An. rufipes, An. squamosus and An. coustani from South Western Burkina Faso with two sympatric field isolates of Plasmodium falciparum, using direct membrane feeding assays. Individuals from a laboratory colony of Anopheles coluzzii were used as controls. From day ten post-infection, the head and thorax of dead mosquitoes were individually stored at -20°C and sporozoite dissemination was assessed using qPCR.

**Results:** An. coluzzii and An. rufipes showed similar competence for P. falciparum sporozoites ( $69 \pm 14\%$  and  $62.5 \pm 23.7\%$ , respectively). However, An. coustani and An. squamosus were significantly less permissive for the development of P. falciparum sporozoites ( $11 \pm 14\%$  and  $35 \pm 22\%$ , respectively). Finally, mosquito longevity in laboratory conditions significantly varied among vector species, with best survivorship observed in An. coluzzii (mean longevity:  $26 \pm 1.4$  days), followed by An. rufipes ( $20.5 \pm 2.15$  days), An. squamosus ( $16.2 \pm 0.5$  days) and An. coustani ( $16 \pm 1.1$  days).

**Conclusions:** An. rufipes, An. squamosus and An. coustani were efficiently infected with P. falciparum gametocytes in laboratory conditions. The mechanisms leading to lower competence in An. squamosus and An. coustani are not yet known, but mosquito immune system response or the quantity of the blood meal could be involved. Because An. rufipes displayed relatively long lifespan and high competence for P. falciparum, it has the potential to ensure robust transmission, provided that it can feed on humans in natural conditions.

# Effects of cohabitation between Anopheles gambiae and Culex quinquefasciatus aquatic stages on their life history traits

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#### Abstract

The interactions between the aquatic stages of the malaria vector Anopheles gambiae (Diptera: Culidae) in cohabitation with those of filariasis Culex quinquefasciatus can affect the density of the future imagos and consequently influence the epidemiology of these diseases. Thus, the effective fight against malaria and filariasis vectors can be achieved by targeting on the regulating mechanisms (like biotic interactions with conspecifc and heterospecific larvae) of the fitness of their aquatic stages. This study explored the effects of cohabitation between An. gambiae and Culex quinquefasciatus on their life history traits under different densities and constant food supply under laboratory conditions. Anopheles and Culex larvae of different ages (L1 and L3) were reared together at varying densities in non-chlorinated water following the larval combinations as follows: An. gambiae (control treatment), An. gambiae and Cx. quinquefasciatus (test treatment) and Cx. quinquefasciatus (control treatment). For each cohabitation condition, developmental time and survival rate were evaluated. It appears that the cohabitation between An. gambiae and Cx. quinquefasciatus at stage 1 affected negatively and significantly Anopheles survival. However, it favor Culex's development capabilities. In addition, increased predation of Culex advanced stages (L3) on younger Anopheles (L1) was also noted. However, when Anopheles were older (L3), their development was better due to their predatory behavior on young Culex (L1). These results demonstrate that co-habitation between An. gambiae and Culex quinquefasciatus mainly affect the fitness of Anopheles. From there, several field studies are needed to evaluate the consequences of this cohabitation on the epidemiology of malaria and filariasis.

Key words: Anopheles gambiae, malaria, Culex quinquefascitus, filariasis, interactions, cohabitation.

Physiological and morphological responses prompted during the transition period between the rainy and dry seasons in preparation for survival during the subsequent severe dry season in Anopheles gambiae s.l. mosquitoes from Burkina Faso, West Africa

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**Background:** In tropical savannahs of West Africa, malaria vectors survive extended periods of aridity and drought (6-9 months) characterized by limited nutritional resources and severe water dehydration that occur either through the desiccation of breeding sites and/or the low ambient relative humidity. The underlying survival mechanisms remain poorly understood. However, there is a hypothesis that certain survival mechanisms are prompted by a switch between the rainy and dry season conditions to overcome the upcoming dry season. This study aimed to elucidate morpho-physiological responses that occur prior to the severe dry season in three members of the Anopheles gambiae complex (An. coluzzi, An. gambiae and An. arabiensis) that co-exist in Burkina Faso.

**Methods:** We used a comparative approach between two localities (with temporary or permanent breeding sites) and three members of the Anopheles gambiae s.l. Immature stages were collected during two different seasonal conditions (rainy vs transitional period between the rainy and dry season) and reared to adults using transplantation cages. The following parameters were analyzed: (1) ovarian development, (2) cuticular fat deposit, (3) morphometric traits, and (4) energy reserves of the emerged adult females.

Results: Cuticular fat deposit and body size were significantly higher among the three species during the transitional period with species-specific differences. An. coluzzii which was found in both localities exhibited higher energy reserves and with those from temporary breeding sites exhibiting significant differences in ovarian development at onset dry season.

**Conclusion:** Different morpho-physiological responses are prompted prior to the dry season in An. gambiae s.l. mosquitoes. This likely reflect divergent coping strategies and different trade-offs between resources acquisition, allocation and utilization. This knowledge improved our understanding of the dry season survival strategies in this important biological vector and may provide novel targets for disease control, targeting vector populations when they are most amenable to control.

**Key words:** Malaria vectors, Anopheles gambiae species complex, dry season, morpho-physiological responses, Burkina Faso

# Investigating the impact of L119F-Gste2 mutation on Anopheles funestus blood size

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**Background and objective:** Malaria vectors such as, Anopheles funestus have adopted haematophagy to ensure egg development and high reproductive rate. The volume of blood ingested from the host could impact on the number of eggs produced and consequently on the reproductive rate of the female. It was reported that blood meal size can be influenced by the age and the weight of the mosquito. We investigated the impact of insecticide resistance mechanisms on the blood meal size of An. funestus field mosquitoes.

**Material and Methods:** F1 mosquitoes from indoor blood fed females collected in Mibellon were grouped according to their weight and allowed to feed. Abdomen of fed mosquitoes were used to estimate the blood meal size by quantifying the human heamoglobin using the Drabkin's reagent protocol. The carcasses of fed and the whole body of unfed mosquitoes were used to genotype the L119F-GSTe2 mutation associated with DDT/ pyrethroid resistance.

**Results and discussion**: 38.088% (457/1200) mosquitoes successfully took the blood meal. Genotyping of L119F-GSTe2 mutation within fed mosquitoes revealed that 24 were homozygous resistant, 103 heterozygote and 233 were homozygous susceptible. Out of 150 unfed mosquitoes genotyped 8 were homozygous resistant, 48 heterozygote and 94 homozygous susceptible. No significant association was observed in the ability to feed between different genotypes. The median volume of blood meal size was not significantly different between homozygote resistant (2.516  $\mu$ l), heterozygote (2.743  $\mu$ l) and susceptible (2.745 $\mu$ l) mosquitoes suggesting that the L119F-GSTe2 has no impact on the blood meal size of An. funestus mosquito.

**Conclusion and recommendations:** Our finding showed that the L119F-GSTe2 mutation doesn't impact An. funestus blood meal size. However further studies are needed to assess the fitness cost of this mutation in this malaria vector.

Laboratory Evaluation Of Effects Of Cupressus Lusitanica, Ocimum Basilicum And Petroselinum Crispum On The Development Of The Malaria Vector Species Anopheles Coluzzii Coetzee & Wilkerson Sp. 2013 (Diptera: Culicidae)

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Abstract: Since malaria remains a plague in most Cameroonian and African communities, and numerous children still die from this disease, the current strategy for case management and prevention of malaria may not be the unique solution to achieve the elimination by 2030 as expected. We have therefore designed an experimental research protocol to assess the effects of three local medicinal plants (Cupressus lusitanica, Ocimum basilicum and Petroselinum crispum) on the development of the local malaria vector An. coluzzii. The research was carried out in OCEAC laboratory in 2016. Plants used for the study were collected locally from Santa in the North West region of Cameroon. WHO biological tests were performed with larvae from laboratory and field in the city of Yaoundé. For the potential effect of the 03 plants on aquatic development, batches of 100 larvae were reared in various concentrations of plant powder, methanolic extract and essential oil. The mortality was monitored each 24 hours until adulthood. The following results were obtained using 3rd larval instars. Mortality in powder at 0.1% conc. was 67%, 62% and 59% in cypress, parsley and basil respectively. Mortality in methanolic extract at 5% conc. was 100%, 88% and 82% in parsley, cypress and basil respectively. Mortality using 0.3% essential oil was 100%, 98% and 75% in parsley cypress and basil respectively. Knockdown 3rd instars were tested for 1hr and mortality for 24hrs. The following results were obtained using essential oil of parsley, cypress and basil at 0.3% concentration; 100%, 88%, 73% for laboratory and 100%, 64%, 53% respectively for field colonies). For the adult mosquito susceptibility to essential oils, CDC bottles were used to record knock down times after 1 hour of exposure and the mortality 24 hours post-exposure. Mortality at 0.3% conc. was 100%, 76% and 61% in parsley, cypress and basil respectively. The results of this study could be used in encouraging communities on the use of such plants as mosquito repellent and larval control. Industries can also use the results in the production mosquito nets especially parsley oil.

Key words: An. coluzzii, C. lusitanica, P. crispum, O. basilicum, biopesticides, essential oil

# Identification of two members of the Anopheles funestus (Diptera: Culicidae) group: first evidence of the implication of Anopheles leesoni in Plasmodium falciparum transmission in Cameroon.

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**Background:** To effectively fight malaria, knowledge of the vector role of each species of anopheles is essential to help national programmers to implement more effective and sustainable malaria control strategies in endemic countries. This study reported the probable implication of Anopheles leesoni in malaria transmission in Cameroon, Centre Africa.

**Methods:** Anopheles funestus s.l populations were collected using electric aspirators or CDC light trap from May 2015 to December 2017 in three sites selected on the basis of the environment(rural savannah, urban and rural forested). Anopheles funestus mosquitoes were identified to species using morphological criteria and PCR techniques. Mosquito infection status to Plasmodium falciparum was determined by ELISA. AS-PCR assay was carried out to determine the L119F-GSTe2 mutation frequencies in An. funestus s.s.

**Results:** A total of 12203 mosquitoes were collected during the study. Anopheles funestus was the major species and the main malaria vector in rural savannah and rural forested study site followed by An. gambiae and whereas in urban areas, it is rather An. gambiae which was the majority species followed by An. funestus. Cocktail PCR of 1396 An. funestus showed respectively 93.62% of An. funestus s.s and 6.38% of An. leesoni. Plasmodium falciparum infection rate determined by ELISA was 21.04% in An. funestus s.s. For the first time, An. leesoni was found positive for P. falciparum in Cameroon. A high frequency of 119F GSTe2 allele was showed in An. funestus s.s from Cameroon.

**Conclusion:** Our study showed the presence of two An. funestus species in Cameroon. The P. falciparum was first identified in wild population of An. leesoni hence confirming the role of this species in malaria transmission in Cameroon.

# Contribution of Anopheles nili in malaria transmission of North Benin

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**Abstract:** Malaria is one of the most serious vector-borne diseases, affecting half of the world's population of 7.4 billion people. As malaria transmission reduces, heterogeneity of infection often increases within the population, with considerable clustering seen at the household and neighbourhood level. Here, this study shows the first evidence of Anopheles nili as malaria vectors and their part of contribution in malaria transmission.

Mosquitoes were collected indoors and outdoors between October and December 2017 in Bambaba and Wodara, 2 differents village respectively in center and peripheral of Kerou, using human landing catches and pyrethrum spray catch methods. All the anopheline mosquitoes collected were assessed for species identify by using morphological characteristics, and molecular diagnostic tools for major vector species complexes. The Plasmodium falciparum circumsporozoite indices were measured by enzyme linked immunosorbent assay (ELISA) in all vectors.

The results showed the presence of An. gambiae s.l, An. funestus and An. nili, three vector species in the study area. A high significant human biting rate was recorded in An. nili compared to An. funestus (0.656 bites/ man/night) in Kérou district where an inhabitant received 5 bites of An. nili per night. On the other hand, this rate was significantly lower than those of An. gambiae s.l (26.19 bites/man/night). During this same time, the entomological inoculation rate (EIR) was 1,875 infected bites/man/month in An. nili against 13,05 infected bites/man/month in An. funestus. We also noted that An. nili was the only specie of the An. nili complex recorded after PCR in the study area.

This study provides useful informations on the contribution of Anopheles nili as secondary vectors to malaria transmission in Northern Benin and suggests further study to find out at what level other districts are involved and other species as well for better malaria vector control interventions.

Key words: Malaria, transmission, Anopheles nili, contribution, Benin

Increase of Ace-1 resistance allele in Anopheles gambiae population following indoor residual spraying (IRS) implementation using bendiocarb in Atacora region in Benin : a threat for malaria vector control.

Authors: Rock AÏKPON and Martin AKOGBETO

Abstract: The aim of this study was to evaluate the susceptibility of Anopheles gambiae mosquitoes to bendiocarb, before (2010) and after five years (2015) of IRS implementation and to report the evolution of Ace-1R mutation frequency in Atacora region. Indoor collection was carried out through Morning 7 to 9 a. m in five districts (Kouandé, Natitingou, Matéri, Tanguiéta and Copargo) of the Atacora-Donga region before and after IRS. Anopheles larvae were also reared in each district before and after IRS and emerging adults were exposed to bendiocarb (0.1%) in susceptibility tests. Polymerase chain reaction (PCR) assays were run to determine the members of the An. gambiae complex, as well for insensitive acetylcholinesterase (AChE1) due to Ace-1R mutation. This study showed that the mean Ace-1 mutation frequency had increased significantly from 2010 to 2015 after five years of IRS. Mortality data indicated that mosquitoes were susceptible in 2010 to bendiocarb 0.1%. By 2015, there was a drastic decline in the An. gambiae susceptibility to bendiocarb in intervention districts. An. gambiae s.s. and Anopheles coluzzi were the two members of An. gambiae complex that were found in sympatry in the study area. An. gambiae was predominant in tested samples (92.50%). The Ace-1R mutation was found in both An. gambiae s.s. and An. coluzzi with frequency of 7.33 and 7.35%, respectively. The high proportion of homozygous susceptible specimens that survived from the WHO bioassays suggests the implication of biochemical resistance mechanisms. These results are of prime importance in the effort to document multiple impacts of operational control programmes on mosquito vectors, and to conceive sustainable control strategies for future malaria control programmes.

Key words: Anopheles gambiae Ace-1R, increasing, indoor residual spraying, Benin.

Susceptibility Of Anopheles Gambiae S.L. To Pyrethroids And Organophosphate Insecticides In An Urban And Semi-Urban Area In Bauchi State

Author: Suwaiba Hussaini

#### Abstract

**Background:** Long Lasting Insecticide Nets (LLINs) and Indoor Residual Spray (IRS) are the major malaria vector control interventions in Nigeria. These interventions are currently threatened by the development of insecticide resistance in the local mosquito vectors, Anopheles. There are pointers that urbanization may have effect on the development of insecticide resistance in these local malaria populations. We therefore determine the resistance status of Anopheles gambiae s.l. from and urban and semi-urban area in Bauchi State.

**Method:** Larval samples of Anopheles mosquitoes were collected from an urban area (Bauchi metropolis) and a semi-urban area (Dass local government) in Baichi state. Three to five day old female adult emergence were exposed to 0.05% Deltamethrin, 0.05% Lamdbacyhalothrin and 0.25% Primiphos methyl according to WHO standards (25 non blood fed female mosquitoes in four replicates with two control replicates of 25 mosquitoes per tube). Mosquitoes exposed were morphologically identified using standard morphological keys. The results of the test was analyzed using SPSS version 23.0.

**Results:** All the Anopheles mosquitoes used for the test were identified as Anopheles gambiae s.l.. The results indicate that the mosquitoes were more resistant to pyrethroids, especially deltamethrin in the urban area as compared to the semi-urban area (64%, 92% for Deltemethrin and Lambdacyhalothrin respectively for the urban settlement and 82%, 96% for Deltamethrin and Lambdacyhalothrin respectively for the semi-urban settlement). This suggests an effect of increase use of pyrethroid based insecticides for vector control associated with urban settlements. However, all the mosquito populations from both the urban and semi-urban settlements were susceptible to Pirimiphosmethyl insecticides (100% mortality). With this, it suggests that the insecticide pyrimiphos methyl may be a good candidate for vector control interventions in both the urban and semi-urban settlement in Bauchi state.

**Conclusion:** Mosquitoes from urban settlements are more resistant to pyrethroids than those from semi-urban areas and urbanization plays a role in the selection of resistant species of Anopheles gambiae s.l. in Bauchi state. Such data could assist in planning vector control activities for the State.

Update the geographical distribution of Aedes aegypti and Aedes albopictus (Diptera: Culicidae) and genetic diversity of invading species A. albopictus in Cameroon

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**Background:** Aedes albopictus, and Ae. aegypti are the most important arbovirus vectors worldwide. Ae. albopictus was first recorded in Cameroon in 1999. Previous studies in Cameroon 10 years back had showed that the invasive species Ae. albopictus have a geographical distribution limited to south under 6°N while the native species Ae. aegypti was present across the country. To update our knowledge, this study aimed to assess the current geographical distribution of both species in Cameroon and their prevalence in sympatric area, and the genetic diversity of invasive species using Cytochrome oxidase subunit 1 gene.

**Methods:** Immature stages of Aedes were sampled between March and August 2017 in 29 localities across Cameroon according to North-South and East-West transects. In each location, larvae and pupae were collected in several containers, transported in the insectary, reared to adult and identified. Aedes albopictus from 16 locations were analysed using COI gene.

**Results:** In total, 30,452 immature stages of Aedes were identified across the country comprising Ae. albopictus (69.3%) and Ae. aegypti (30.7%). Analysis revealed that Ae. aegypti is still present across the country whereas Ae. albopictus is limited to south around 6°5'N. In south, Ae. albopictus was the most prevalent species excepted in Douala locality in which Ae. aegypti was dominant. This suggest that climate, vegetation and building density impact the distribution of both species in Cameroon. Mitochondrial DNA analysis revealed a low genetic diversity of Ae. albopictus with five haplotypes resulting in low haplotype diversity (Hd=0.32) and nucleotide diversity ( $\pi$ =0.00075) indexes. This low genetic polymorphism is consistent with recent introduction of Ae. albopictus in Cameroon

**Conclusion:** These findings updating the current geographical distribution of Ae. albopictus and Ae. aegypti and could serve to plan vector control programme against arbovirus vectors in Cameroon.

Keywords: Aedes albopictus, Aedes aegypti, geographical distribution, genetic diversity, Cameroon.

### Susceptibility of Field-collected Aedes aegypti (L.) to conventional insecticides in coastal Kenya.

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**Abstract:** Aedes aegypti are the vectors for many important arboviruses in Kenya, and control options are limited. We evaluated the susceptibility of field populations of nonblood-fed Aedes aegypti mosquitoes to four conventional insecticides: permethrin, 0.75%; deltamethrin, 0.05%; bendiocarb, 1%; and fenitrothion, 1%; according to WHO standard procedures. We also determined the efficacy and residual activity of a water dispersible formulation of the biolarvicide Bacillus thuringiensis var israelensis (Bti) (VectoBac<sup>®</sup> WG) under semi-field conditions during the dry and wet seasons. Three concentrations of larvicide in 2-L plastic trays were used in this study: 1×, 10× and 20× the manufacturer's recommended dosage (8mg/L, 80mg/L and 160mg/L respectively). Bioassays of adults showed full susceptibility to permethrin and fenitrothion. A low level of resistance was observed for deltamethrin and bendiocarb with 86 and 84% mortality, respectively. Knockdown rate for the first 20 min was notably higher (>90%) for both deltamethrin and permethrin than for bendiocarb and fenitrothion (<10%). Bti was effective under semi-field conditions for an average of 16 days at 8 and 80mg/L and 160 mg/L lasted for 22 days. The results of this study suggest a need to monitor for developing resistance along the Kenya coast in order to recommend suitable adulticides and dosages for Aedes aegypti control, especially given the recent outbreaks of dengue and chikungunya.

Comparison of Community implementation of Human Landing and Non-human landing collection methods of vectors of Wuchereria bancrofti and Onchocerca volvulus for entomological monitoring of transmission.

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**Abstract:** In the context of malaria and vector-borne NDT elimination and since human landing catch is becoming less and less accepted, expensive and requiring well-trained personnel, it is crucial to validate a sampling method able to obtain a large number of vectors for entomological surveillance. For the past decade, several traps and collections methods had been experimented, even used locally by communities and validated as alternative to HLC. However, as vector dynamics and local communities behavior differ from one region to another, it is important to test a panel of tools in each context to see the efficiency of such tools compared to HLC and the vector density patterns.

This study sought to address these issues in Burkina Faso by testing the efficiency of four mosquitoes collecting tools that included HLC, CDC light trap, Net trap (a modified-Ifakara trap design) and Exit trap in areas with high (Bapla), medium (Ouessa) and relatively low mosquito densities.

The results showed that HLC remained the most efficient collection method whatever the village followed by Net traps that was superior to but sometimes similar to CDC traps. It is also interesting to report that the Net trap except of area of high anopheline density like Bapla, always collected more An. gambiae s.l. than CDC traps. Furthermore for black flies collections no alternative than adults collection by HLC or larvae collection by prospecting larva source were available. Encouraging results from the study was that only local people in each community managed the collections even in the face of the more difficulties for larva collection.

In conclusion if considered that CDC trap is already validated and adopted for entomological surveys and that the Net traps results were observed to be similar of those of CDC traps, this Net trap can be recommended for communities to use for LF vector surveillance program.

Keywords: Mosquito, Black flies, Malaria, Vector-borne NTD, Community, Burkina Faso

Meaningfull engagement prior to community decision making – the case of engagement for contained laboratory work of Target Malaria Mali

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**Background:** Target Malaria is a not-for-profit research consortium working on a genetic approach to reduce the mosquito vector of malaria.

**Materials and methods:** After identifying key stakeholders the Target Malaria Mali stakeholder engagement team, carried out aseries of engagements using different tools such as visuals describing the technology and activities, focus group meetings, as well as insectary visits.

**Results and discussion:** The project step-by-step to achieve its goal. At each step the project wants to ensure that local communities are adequately engaged. The first step of the project in Mali would be to work in laboratory containment with a genetically modified mosquito strain in which males are sterile. Before making an application to the National Biosafety Committee , the Malian regulatory authority, the project wanted to ensure that the community had a sufficient understanding about this mosquito strain and about the concept of contained laboratory work, so that they could make an informed decision as well.

**Conclusion and recommendation:** This paper will show the importance of some key principles for engagement such as: early and progressive engagement, openness and co-development. The use of visuals demonstrated the importance of adapting the information to the audience to ensure that knowledge blocks were in place before moving to the next knowledge. Visits to the insectary have further informed the stakeholders about the research process.

Keywords: Target Malaria - Communication - Engagement - modified mosquitoes

# Occurrence Of Several Species Of Mosquitoe In Makurdi And Its Possible Role In Disease Transmission In Humans And Animals

Authors: Oke P.O, Adamu M. Mbaave J & Oke-Egbodo B.E

A survey was conducted to determine the species of mosquitoes in Makurdi using larval identification, between May and October, 2017. Six (6) areas comprising eighty-one (81) sites, was randomly selected across the metropolis and samples were collected using pipettes and a scoop. The habitats sampled were, abandoned tyres, domestic containers, ground pools, drainages, and tree trunks. Field identification of larvae was carried out based on their posture in water to classify them into anophelines and culicines. The larvae were killed in tepid water and preserved in 75% alcohol. The third and fourth larval instars were subsequently separated from the entire collections and identified to species level based on their morphological characteristic features. The results of this study identified several species of mosquitoes in the following five genera; Aedes (44.27%), Anopheles (6.25%), Culex (35.16%), Toxorhynchites (3.65%) and Wyeomyia (1.82%) and an unidentified genus (8.85%). This study reports the presence of larvae of a predaceous mosquitoe species actively ingesting larvae of other genera. An incidental finding of a fungal infested larvae of Anopheles species and Culex salinarius is also reported. The importance of this mosquitospecies and their epidemiological importance in the transmission of diseases in humans and animals will be discussed. The use of molecular tools to identify the unidentified genus may give further details on the mosquitoe species in the study area.

House screening - a potential tool for integrated mosquito vector abatement program in Kenya

Authors: Peter Njoroge Ng'ang'a\*, Mutunga, J., Oliech, G., Mutero C.M.

**Background:** House screening as an environmental tool for malaria control has been evaluated in several parts of Africa and is increasingly being recommended as an effective and practical method against malaria transmission. The objective of the study was to assess community knowledge and perceptions on malaria prevention and house screening in Western Kenya.

**Methods:** A cross-sectional household survey was conducted between December 2016 and January 2017. A total of 80 households were randomly selected to participate in the study. Structured questionnaires, key informant interviews and focus group discussions were used to gather data from members of the households and other participants.

**Results:** A total of 80 respondents participated in the survey, 67.5% of whom were females and 32.5% were males. Most of the houses had un-cemented earthen floors (76.3%) with almost all windows and eaves unscreened (85% and 97.5% respectively). Ninety one percent (91.3%) of the respondents had previously seen or heard malaria control messages and 57.5% acknowledged not to have previously heard any information or knowledge on house screening. Use of treated mosquito nets was by far the most reported known (97.4%) and applied (97.6%) personal protective method while only 15.6% respondents were aware of house screening. Major reason given for screening doors, windows and eaves was to prevent entry of mosquito and other insects (>85%). There was no association between reason given for screening doors, windows and eaves and the sociodemographic characteristics of respondent like the level of education, occupation, religion, gender and village of origin. 57.5% of the respondents did not have any prior information on house screening while economic factors and lack of screening knowhow were the major reasons given for not screening houses in the area. Grey was the most preferred colour for screen materials (48%), followed by Blue (13.6%). Major reasons given for the colour preference were that it matched the colour of the walls (21%) and it did not show or get dirty quickly (16.3%).

**Conclusion:** House screening was not a common intervention for self-protection against malaria vectors in the area. There is need for advocacy, mobilization, and social promotion of house screening as part of integrated vector control interventions.

Keywords: Knowledge, Perception, Screening, Malaria, IVM.

Tracking of Malaria Vectors Using Stable Isotopes to Measure the Contribution of Aestivation to Dry Season Persistence

Authors: Alpha Seydou Yaro

#### Efficacy & Residual Activity Of Clothianidin + Deltamethrin Wp-Sb, A Combination Formulation For Irs

#### Author: Alpha Seydou Yaro

#### Introduction

Indoor Residual Spraying (IRS) is a primary intervention for malaria control and eradication. The application of IRS consistently over time in large areas has had a massive impact on vector populations and subsequently the epidemiological pattern (reduced burden of morbidity and mortality) of malaria in many sub-Saharan African countries. IRS is now a mainstay tool of the PMI with over 16M people protected by IRS to-date. Nevertheless, intensive vector control campaigns are not without drawbacks and the spread of resistance mechanisms in Anopheles mosquitoes is arguably the most significant. There is now a dire need to supplement or replace pyrethroids, carbamates and OPs with new classes of insecticide for IRS against which there is no resistance in mosquitoes.

The IRS formulation Fludora<sup>®</sup>Fusion (Clothianidin [CTD]+ Deltamethrin [DLT] WP 56.25-SB, Bayer CS) was developed to fill this gap. It is the first insecticide combination developed for use in IRS programs and is intended for use in areas where there is established or emerging resistance to currently available IRS insecticides. It contains clothianidin, a neonicotinoid that acts as a nicotinic acetylcholine receptor competitive modulator with immediate and delayed mortality effects against insects expressing target site and/or metabolic resistance, and the pyrethroid deltamethrin that will kill any susceptible mosquitoes in the target population. The residual activity and efficacy of this insecticide was assessed over a 10-month period in lower Moshi, Northern Tanzania.

**Materials and Methods:** Fludora<sup>®</sup>Fusion was evaluated on three substrates – mud, concrete and thatch matting. The insecticide was applied at 200mg/m2 CTD + 25mg/m2 DLT, and compared with a WHOPES benchmarking product, Ficam<sup>®</sup> (Bendiocarb WP 80), sprayed at 400mg/m2. In addition, the two active ingredients [AI] of Fludora<sup>®</sup>Fusion were sprayed as single AIs at the same concentrations in two other huts. A fifth unsprayed hut was used as a negative control. The experimental huts were of the East Africa design with concrete walls and concrete ceilings, At several time points up to 6 months post-spraying, free-flying wild Anopheles arabiensis were recovered and scored as dead or alive, bloodfed or unfed, then observed for delayed mortality up to 72h post-exposure. Mortality, bloodfeeding inhibition and induced exiting were the main outcome measures. In five simple huts, the same treatments were applied to mud walls and thatch matting panels. To assess duration of effective action, cone assays of 30-minute exposure were conducted monthly, up to 10 months, on the treated surfaces in all experimental huts and simple huts. Insectary-reared susceptible An. gambiae s.s. Kisumu and pyrethroid-resistant An. gambiae s.s. Muleba-kis (kdr L1014S and MFO metabolic resistance mechanisms) were exposed in the cones.

The application of the insecticide was assessed by chemical analysis of filter papers attached to the hut walls, and resistance status of all test systems used was confirmed by qPCR techniques and standardized bottle bioassays.

**Results:** Free-flying Anopheles arabiensis were recovered from the experimental huts during two periods: between 27th Sept – 23rd Oct 2016 (month 1 post-spraying) and between 15th January - 13th March 2017 (months 5&6 post-spraying). In month 1 post-spraying Fludora<sup>®</sup> Fusion killed 100% of An. arabiensis that entered the experimental huts, and though the efficacy declined over time, it was still killing 74% (65-83) at 6 months. In contrast Bendiocarb was not efficacious even in month 1 (though numbers caught were very low), and by month 6 was only killing 33% (22-44). Fludora<sup>®</sup> Fusion outperformed bendiocarb and deltamethrin at

all time points. Clothianidin alone also performed significantly better than bendiocarb and was not statistically inferior to Fludora<sup>®</sup> Fusion.

In the in situ cone assays on concrete Fludora<sup>®</sup> Fusion was still killing 100% of An. gambiae s.s. susceptible and resistant strains after 9 months. In contrast, for An. gambiae s.s. Kisumu, Bendiocarb fell below the WHO 80% threshold after 3 months, deltamethrin failed at 4 months and clothianidin alone at 8 months. For the pyrethroid-resistant strain bendiocarb never induced mortality above 50% and deltamethrin never above 30%. On mud surfaces, Fludora<sup>®</sup> Fusion was still inflicting mortality in the Kisumu strain above 80% at 9 months post-spraying, dropping below the threshold only after 10 months. The failure point for the other insecticides was the same as reported for the concrete substrate. Most significantly, Fludora<sup>®</sup> Fusion, deltamethrin and clothianidin were all killing above 80% of Kisumu at 9 months. In contrast, bendiocarb efficacy dropped to 25% at 5 months post-spraying.

**Discussion and conclusions**: The efficacy and duration of effective action of the test item Fludora<sup>®</sup> Fusion was clearly demonstrated in this phase II experimental hut study. Its durability on mud, concrete and thatch surfaces compares favourably with insecticides already approved by WHOPES for IRS application, and its efficacy against pyrethroid-resistant mosquitoes should help to promote its widespread use in Sub-Saharan Africa for IRS. The planning of a community randomised controlled trial to assess efficacy, operational safety and acceptability in a realistic setting has started in Muheza, N.E. Tanzania. The design and early findings of this phase III study will also be described in the presentation.

Efficacy and residual performance of Fludora<sup>®</sup> Fusion a next generation combination insecticide (Clothianidin + Deltamethrin) developed for Indoor Residual Spraying: Twelve months of WHO cone bioassay evaluations in Zambia

#### Author: Muleba Mbanga

**Introduction:** Insecticide-based vector control remains fundamental to control or eliminate malaria. The global strategy shifted from malaria eradication to control and now elimination. Residual insecticides applied indoors have been key to the implementation of the global strategy. The successes scored in malaria management based on vector control are due to just a few classes of insecticides. Sadly however, insecticide resistance against these few insecticide classes has escalated. This threatens to negate the achievements scored against malaria in all endemic countries. For Zambia currently the organophosphates are the only class of insecticides against which resistance is yet to be reported. With control programs now emphasizing elimination there is a growing need for more efficacious and cost-effective residual insecticides. This study evaluated the efficacy and residual performance of Fludora Fusion in northern Zambia. Fludora Fusion is a new generation insecticide, and the first ever combination product for indoor residual spraying which includes a new mode of action based on the neonicotinoid.

**Methods:** The insecticide was applied to both Cement and Mud surfaces at the recommended dose rate. The WHO cone bioassays were used to evaluate the efficacy and residual activity of Fludora Fusion in comparison to Actellic<sup>®</sup> 300 CS that is currently used in control program.

**Results:** On cement surfaces Fludora<sup>®</sup> Fusion was effective for the entire twelve months at 24-hours postexposure. On mud surfaces Fludora<sup>®</sup> Fusion was effective for 12 twelve months when evaluated t72-hr post exposure; up to 8 months at 48-hr and up to 4 months at 24-hr post exposure. The efficacy, residual activity and knockdown effects of Fludora<sup>®</sup> Fusion was far superior compared to the benchmarking product, Actellic<sup>®</sup> 300CS.

**Conclusion:** Fludora<sup>®</sup> Fusion showed good efficacy and residual performance as an IRS product. The product should be considered for inclusion in IRS programs upon confirming susceptibility of local vectors by either CDC or WHO standard assays.

Efficacy of chlofernapyr and clothianidine insecticides against Anopheles malaria vector populations of western Kenya.

Authors: Agumba Silas, Eric Ochomo, Lillian Ogonda, Maurice Ombok, Bernard Guyah, and John Gimnig

**Background:** Increase of malaria cases as recently reported by world malaria report is a clear indication of backdrop in the fight against malaria. Malaria vector control is highly dependent on chemical with pyrethroids as the only insecticide recommended for treatment of ITNs. However, global emergence and spread of insecticide resistance in the major malaria vectors may compromise the gains made in the fight against malaria, it is therefore important to evaluate other safe alternative insecticides for mosquito control. The aim of this study was to evaluate efficacy of chlorfenapyr (pyrolle) and chlothianidin (neonicotinoid) insecticides on Anopheles malaria vectors of western Kenya.

**Methodology:** CDC-Bottle assay was used to determine the diagnostic doses of chlorfenapyr and clothianidine insecticdes against susceptible laboratory strain, An. gambiae sensu stricto, Kisumu strain. Insecticides efficacy were evaluated against field collected female anopheles malaria vectors sampled from Nyando, Bumula and Ndhiwa sub Counties in western Kenya following WHOPES phase II guidelines. The LC50 and LC100 was determined using probit analysis where Chi-square goodness-of-fit test was performed in SPSS v20.0 software. The Mortality rates were calculated as a percentage of individual mosquitoes that died within 24 hours 48 hours and 72 hours following WHO guidelines on insecticide susceptibility. Anopheles gambiae and An. funestus complexes were identified using PCR.

**Result:** Diagnostic concentration of Chlorfenapyr insecticide for Kisumu strain and wild mosquitoes from field was at 5% while Clothianidine insecticide had a diagnostic dose of 15% for both susceptible Kisumu strain and wild mosquitoes. Anopheles malaria vector populations in western Kenya were highly susceptible to both Chlorfenapyr and Clothianidine insecticides.

Conclusions: These findings provide initial starting points for alternative insecticides for malaria vector control.

Experimental huts performance of Veeralin against natural populations of Anopheles funestus s.s. in Muheza, Tanzania

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**Background:** The success of long-lasting insecticidal nets (LLIN) as the primary method for preventing malaria is threatened by pyrethroid resistance in Anopheline vectors. New generation long-lasting nets incorporating PBO synergist (piperonyl butoxide) with pyrethroid are designed to control insecticide resistant mosquitoes. The efficacy of Veeralin PBO LLIN was evaluated in experimental hut against wild free flying pyrethroid resistance Anopheles funestus.

**Methods:** The East African experimental huts were used to assess wild mosquito population responses. Also WHO method was also used to detect resistance in wild anopheles mosquitoes exposed to 0.75% permethrin. Mosquito mortality, blood feeding inhibition and personal protection were compared between untreated nets, standard LLINs and PBO/pyrethroids combination nets.

**Results:** Blood feeding rates recorded by 20 times washed Veeralin was statistically similar to that of the 20 times washed WHOPES approved PBO/pyrethroid and a positive control in this trial PermaNet 3.0 LN, this provide evidence that Veeralin LN provides similar blood feeding inhibition to the standard approved LN and thus meeting WHOPES criteria for blood feeding inhibition.

**Conclusions**: Results show partial improvement of Veeralin PBO LLINs efficacy against pyrethroid resistant Anopheles funestus compared to standard pyrethroid only LLIN. This study's eta- analysis found that PBO LNs are significantly more effective against pyrethroid resistant mosquitoes than standard pyrethroid only LNs.

Keywords: Resistance, PBO LN, Exophily, Mortality, Knockdown

#### Effects Of Medicinal Plant Extracts On Mosquito Vectors In Muheza Tanzania

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**Abstract:** Increase in insecticides resistance among mosquito population presents a great challenge to chemicals that are used for mosquitoes control in sub-Saharan African. Currently, there is no approved alternative insecticide to pyrethroid which is used in impregnated bed nets. This study aimed to determine effects of Chenopodium ambrosioides essential oils against malaria and arboviruses mosquito vectors. Insecticide susceptibility bioassays were performed according to the World Health Organization guidelines on 2-5 days old human biting mosquitoes in Kwabada village, Muheza district. Each of the four species of mosquitoes were exposed to C. ambrosioides essential oils (10%) and two classes of insecticides commonly used for malaria vector control. Mosquito mortality rates (%) were determined after 24 hours post insecticide exposure. All mosquito species tested were susceptible to C. ambrosioides essential oils (10%). An. gambiae s.l. showed possible resistance to Permethrin (0.75%) and Deltamethrin (0.05%) with 93% and 92% mortality rates (%) 24 hours post exposure to insecticides. With Lambdacyhalothrin (0.05%), An. gambiae s.l. was resistance with mortality rate of 57%. An. funestus was susceptible to all insecticides tested. Aedes aegypti formosus was susceptible to all insecticides tested except DDT (4%) which showed possible resistance. However, all insecticides tested were able to knock down 50% of all mosquito species (KDT50) exposed within 1 hour. C. ambrosioides essential oils (10%) took the shortest mean time to knock down 50% of all mosquitoes tested. The mean time ranged from 11.4 to 13.1 min. The mean time taken to knock down 95% of all mosquito species ranged from 17.8 to 148.3 min, from C. ambrosioides essential oils (10%) and Lambdacyhalothrin (0.05%) in An. gambiae s.l., respectively. This study has revealed that, C. ambrosioides essential oils have demonstrated appreciably higher strong insecticidal effects on malaria and arboviruses mosquito vectors. Further studies are needed to determine the long-lasting insecticidal efficacy of essential oils extracts from C. ambrosioides for development of novel methods of controlling mosquito vectors.

Semi-field evaluation of Fludora<sup>®</sup> Fusion (Bayer), a new IRS combination containing clothianidin and deltamethrin, for the control of Anopheles gambiae s.l. in experimental hut trials, Rwanda

**Authors:** Emmanuel Hakizimana<sup>1,</sup> Fredrick Kateera<sup>2</sup>, Dunia Munyakanage<sup>1</sup>, Beatus Cyubahiro<sup>1</sup>, Xavier Misago1, Andre Laas<sup>3</sup>, Aimable Mbituyumuremyi<sup>1</sup>

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**Background:** The spreading of insecticide resistance in African malaria countries currently denotes a major threat on the gains achieved on malaria control. Thus, there is an urgent need for new insecticide formulations to supplement the existing ones in the framework of insecticide resistance management. The evaluation of Fludora<sup>®</sup> Fusion, (Bayer's new combination product for Indoor Residual Spray (IRS) is fitted into the above context.

**Methods**: The trial was carried in experimental huts (Western Africa model) established in South Eastern of Rwanda for 12 consecutive months. The WHO cone bioassay standard protocol (2006) was used for assessments of insecticide and residual efficacy post spraying using Anopheles gambiae s.s., Kisumu strain. The tests were conducted on indoor cement and mud walls from which one hut per type of wall was respectively treated with Fludora<sup>®</sup> Fusion 56.25 WP, Pirimiphos- methyl 300 CS and distilled water for the control huts. The knockdown (KD) was counted up to 60 minutes and mortality after a holding period of 24 hours or extended up to 96 hours if mortality dropped below 90%.

**Results:** After 12 months of bio-efficacy tests, Fludora Fusion, assessed at 24h post exposure, showed a mortality of 100% both on cement and mud wall with respective knock down of 96% and 98%. The Actellic 300 CS displayed 88% mortality and 14% of KD on cement wall while on mud wall; the mortality was 94% with a KD of 34%.

**Conclusion:** These findings showed that Fludora Fusion 56.25 WP has the highest mortality and KD effects against susceptible Anopheles gambiae s.s. over a period of 12 months. It appeared to be one of the best alternative insecticides for IRS and should be applied in a single annual spraying in Rwanda. Also, it appears as a worthy vector control tool for insecticide resistance management.

Key words: Fludora® Fusion, Anopheles gambiae s.l., experimental huts.

### A Comparison Of House Eave Aspiration To Mosquito Swarm Sweep Net Survey As A Tool For Male Mosquito Collection

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**Background and Objectives:** Many Traditional Malaria Vector Sampling Methods Such As Human Landing Catch, Indoor Pyrethroid Spray Catch And C02-Baited Traps Bias Collections Towards Female Mosquitoes. Balanced Vector Sampling Of Both Males And Females Is Necessary For Comprehensive Understanding Of Vector Dynamics. Swarm Sampling (Swn), Is Thought To Be The Most Efficient Method For The Collection Of Male Mosquitoes. However, This Method Requires A Large Amount Of Labour, And Resources To Train And Remunerate Collectors. Swm Also Depends Largely On Mosquito Mating Behaviour, Environment And Other Competing Insect Swarms. We Evaluated The Efficiency Of Asp-Eav When Compared To Swn In An Effort To Ascertain If Asp-Eav Would Be A Viable Supplementary Or Replacement Method For Swm.

**Materials and Methods:** Mosquitoes Were Collected In Three Villages Located In Mukono And Kayunga Districts Of Uganda. Three Collections Were Made Per Season (Wet And Dry) In Each Village During Which All Available Swarms Were Collected Using Sweep Nets And Ten Household Eaves Were Aspirated Using Battery Powered Aspirators Over A Period Of Two Days Per Trip. The Data Collected Was Analysed For Seasonal Variation Between Male Mosquito Catches And Yield Per Man Hour By Method.

**Results and Discussion:** During This Study, 1011 Male Mosquitoes Were Collected By Swn And 893 By Asp-Eav. We Found That Asp-Eav Was A Comparable Male Mosquito Collection Method To Swn In Both Seasons. While Swn Had Higher Numbers Captured, The Yield Per Man Hour Was Still Lower Than Asp-Eav. The Smaller Number Of Collectors Required To Carry Out Asp-Eav Also Meant Reduced Chances Of Human Error Affecting Catches Than With Swn.

**Conclusion and Recommendation:** We Concluded That Asp-Eav Was A Viable Male Mosquito Collection Method At Our Study Sites And Could Also Be An Effective Substitute For Swn When Not Specifically Studying Mosquito Swarming Behaviour.

#### Evaluating the efficacy of mini double net trap (MDN) for sampling host seeking mosquitoes

Authors: Alex J. Limwagu, Hamis Kifungo, Anna Nyoni, Gasper Shubi, Marceline Finda, and Fredros Okumu.

**Background and Objectives:** Effective and a more diverse host seeking sampling tool is essential for monitoring malaria transmission. Human landing catch is considered as a gold standard but it exposed human being to both malaria and non-malaria pathogens. We have introduced a mini-double net trap as a replacement of HLC. Mini double net (MDN) with different sizes of the outer layer were tested to observe which height is suitable for collecting more mosquito species.

**Material and Methods:** A study was done in one village in the South eastern part of Tanzania running from 6pm to 6am. Mini Double net trap was design in different outer layers which measures varieties of height from the ground. The trap was designed with different sizes of the outer layer, i.e. 20cm, 50cm, 80cm from the ground and one with the holes on the side. A Latin square design was used to evaluate the MDN trap, each trap were located 100m apart to avoid competition between traps. The trap was design to offer a full protection for a person collecting mosquitoes.

**Results and Discussion:** A total of 2,004 mosquitoes were collected using a MDN trap. The trap with holes on the side caught 468 (23.3%), MDN-20cm caught 505 (25.1%), MDN-50cm caught 535 (26.7%) and MDN-80cm caught 496 (24.7%). The MDN trap with the length of 50cm had significantly higher number of Anopheles arabiensis than the standard height MDN-20cm [Relative rate, RR=1.37, 95%CI (1.02 - 1.85), P<0.05]. MDN-80cm had fewer catches of An. arabiensis compared to MDN-20cm, [RR=0.88, 95%CI (0.68 - 1.14), P=0.337]. There is not significantly differences in the number An. arabiensis capture by MDN with side holes compared to MDN-20cm [RR=1.05, 95%CI (0.767 - 1.44), P=0.759]. The number An. funestus captured by MDN-50cm, MDN-80cm and MDN with side holes were not significantly different with the MDN-20cm [RR=0.33, 95% CI (0.06 - 1.69), P=0.184), [RR=0.82, 95%CI (0.24 - 2.82), P=0.754) and [RR=0.98, 95%CI (0.30 - 3.20), P=0.979) respectively. We can therefore conclude that, MDN trap with the outer layer measuring 50cm from the ground can be considered as the most suitable options for collecting host-seeking mosquito species especially the most abundance vectors, An. arabiensis.

#### The Incidence Of Malaria Among Human Landing Catch Volunteer Participants

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**Background & Objectives:** Human Landing Catch (Hlc) Has Long Been Considered A Gold Standard For Collecting Information On Human-Mosquito Contact. Hlc Samples The Mosquito In The Act Of Host-Seeking, And Thus Integrates Information On Behaviour And Abundance Of Vector Species. Hlc Is, However, Labour-Intensive, May Expose Collectors To Infectious Mosquito Bites And Is Subject To Collector Bias. This Study Estimates The Malaria Incidence Of Volunteer Study Participants (Collectors) Relative To Background Population Levels.

Materials & Methods: With Regular Supervision, A Collector Is Seated Either Indoors Or Outdoors With One Lower Leg Exposed. Collections Take Place From Dusk Until Dawn; Collectors Are Regularly Rotated And Work In Shifts. The Collector Sits With A Flash Light And Mouth Aspirator And Captures Any Mosquitoes Landing On Their Exposed Leg. The Mosquitoes, Organized Into Time-Slot Collections, Are Later Identified For Further Analysis. Collectors Are Encouraged To Report Any Incidence Of Malaria Associated With The Hlc Activity And Are Offered Treatment.

**Results & Discussion:** A Total 2,756 Female Anopheles Gambiae S.L Were Caught, 42.7% Were Collected Indoor And 57.3% Outdoor. Over The Ten Trap Nights In These Three Villages 96 Volunteers Participated In The Study. Of These, 1% Reported Malaria Within 14 Days Of Participating And All Accepted The Treatment Offered. It Is Possible That There Is Under-Reporting Of Malaria Episodes And Active Stake-Holder Engagement Is Used To Reduce This Likelihood.

**Conclusion & Recommendation**: Our Findings Indicate That When Properly Used, Collectors Do Not Have An Increased Or Have Lower Incidence Of Malaria Relative To The Background Population Levels. This Provides Some Evidence That The Use Of Hlc Does Not Increase Malaria Risk To Volunteer Study Participants And Supports Its Use As A Method To Improve Understanding Of Malaria Vectors In Uganda.

Climatic variation and abundance of the vectors of the malaria, in two villages of the district of Kati (Region of Koulikoro, Mali)

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**Background:** Meteorological and seasonal variations affect the abundance of malaria vectors. The relationship between climatic parameters and vector populations is important to understand seasonal patterns of malaria transmission. This study estimates the effect of three climatic variables on the abundance of mosquitoes.

**Methods:** Mosquito abundance data was collected during a longitudinal study in two villages, Tieneguebougou and Ouassarola from January 2016 to December 2017. In both, Pyrethroid Spray Catch was used monthly in 25 rooms (15 fixed / 10 randomly selected). There are meteorological stations in these villages.

**Results:** In both villages, the highest monthly mean temperature (33°C) was in April of both years. The highest mean annual relative humidity in Ouassorola was 97% in September 2016 and in 83% in August 2017. In Tieneguebougou, relative humidity peaked earlier in the year in August 2016 (87%) and in June 2017 (56%). The most rain usually fell in August (204-303mm). The exception was Tieneguebougou in 2017 where July was the rainiest month.

*Anopheles gambiae s.l.* was the only malaria vector identified in the catch and the two villages had a similar numbers of mosquitoes each month. In separate models testing the relative effect size of climatic parameters on mosquito abundance, temperature was not significant, and explained only 4% of the monthly variation; in contrast, rainfall and relative humidity were highly significant accounting for 47 % and 56 % of the variation respectively.

**Conclusions:** Relative humidity may be a better proxy for available water, and thus breeding sites, in the environment than rainfall itself and it may also account for a lag between rain falling and mosquito production. These results suggest that densities of malaria vectors could potentially be predicted via satellite date of surface relative humidity (e.g. MODIS), paving the way for improved tools for vector control and monitoring.

#### Challenges In Malaria Vector Control In Mandera County-Kenya

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Key word: Mosquitoes, Mathenge, Malaria

**Background**: Anopheles gambiae is the malaria vector that predominantly carries the most dangerous malaria parasite Plasmodium falciparum. The mosquito bites at night and breads in stagnant water, open water reservoirs, waste plastics bottles with water, bushes and shrubs. Mandera County is dry hot area with average annual rainfall of 244mm. The area has less forest cover with Prosopis juliflora commonly known as Mathenge tree taking up the whole County. Mathenge tree traps waste plastics making it hard to clean and clear the waste which formed breading grounds for mosquitoes and cutting down the trees will cause afforestation. The aim of this study was to discuss the challenges of malaria vector control in Mandera County-Kenya.

**Methodology:** A cross-sectional study was conducted in which integrated disease surveillance and response (IDSR) data was utilized from January 2016 to December 2017. In addition to that structured questionnaires were given to 500 patients who visited seven sub-county hospitals from January 2016 to December 2017. Age, Malaria cases, Malaria tests done and Malaria mortality were the variables. Statistical package for social sciences (SPSS) was used to analyze the data under strict ethical considerations to ensure clients' confidentiality was safeguarded.

**Results:** Out of the 500 patients 30% used mosquito net in 2016 and 95% in 2017, 2% used mosquito repellant 2016 and 46% in 2017 while 15% had opened water tank in 2016 and 86% in 2017.

	2016		2017	
	< 5 years	≥ 5 years	< 5 years	≥ 5 years
Cases	174	354	449	597
Deaths	0	0	5	28

Malaria cases and mortality 2016 and 2017

**Conclusion**: Cases and mortality has increased in 2017 regardless of the fact that mosquito net and repellant usage increase. This clearly shows that the increase in open water storage system increases mosquitoes yield and growth doubled by the increase in the Mathenge cover that captures plastic waste which is major breeding ground for mosquitoes. To control Malaria in Mandera County requires clearing of Mathenge around the residential areas and asking residents to cover their water storage tanks in addition to treating them.

The Knowledge, Attitudes and Practices Study of the Cape Verdean population in the Context of the Malaria Elimination

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Malaria is a major public health problem in over 90 countries, especially in Africa, among pregnant women and children under 5 years. With the progress made in reducing disease morbidity and mortality and elimination in some countries, the target is to reduce the incidence by 90% and the elimination at least 35 countries by 2030, with Cape Verde eligible to eliminate the disease in 2020.

This study about the Knowledge, Attitudes and Practices (KAP) analyses in the population about the malaria in Cape Verde.

The results show that 88% of respondents knew the form of transmission, 96% knew the entire population was vulnerable and main symptoms were identified. Regarding attitudes, 58% seek a health structure towards the symptoms and 64% do it in the first 24 hours. More than 97% heard about mosquito nets and 19% used it. In practice, 53% use mosquito nets / fume, 45% sprays the house and 43% benefited from indoor spraying. About 90% received information and 7.3% had no information. A total of 54% have the complete knowledge of the disease, related, especially with the educational level.

The level of knowledge about transmission, symptoms and preventive measures Is high when analysed separately in comparison with the level of complete knowledge. There are some gaps, misunderstandings and lack of information, as well as, little participation in fighting actions, which demonstrates the need to improve strategies. Antibodies in the Cape Verdean population lead one to suppose that they are effectively induced and maintained, unlike in the native people of endemic countries. These results encourage more specific studies to complement CAP, as well as the characterization, quantification and durability of the antibodies in the population. With these results, we hope to contribute to the improvement of policies and interventions on the elimination of malaria in the country.

### Factors that Contribute to Dropout of entomological activities at Kibbuye, Katuuso and Kayonjo field sites in Mukono and Kayunga districts; Uganda

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Malaria is a major public health problem in the world today. Over 200million people are infected with malaria every year. In Uganda, over 12 million cases were reported in 2016, of which an estimated 13,000 death were reported (WHO 2017: 124). Uganda's strategies to fight malaria, like mass distribution and use of insecticide treated nets throughout the country, have contributed to progress in reduction of malaria. Unfortunately the available malaria control methods are faced with various challenges such as insecticide resistance in the malaria vectors, high costs and misuse of available control methods. Hence, the need to develop innovative and complementary malaria control interventions, for successful malaria eradication. Target Malaria Uganda does mosquito collection studies from mainland sites; of Kibbuye, Katuuso and Kayonjo villages in Mukono and Kayunga districts, which are relatively isolated with large tracks of farmland and/or swamp for one year and a half now to acquire entomological baseline vector population characteristics around the country. Ten (10) participants withdrew from activities, which affects the analysis of mosquito trends, pace and progress of the research since the same households are supposed to be sampled during mosquito collections. Qualitative methods of Key informant interviews and observation were used to find out the factors that contribute to participant's dropout of entomological activities. Household members shifting, demolished households, no tangible benefits, allergic to insecticide used for spraying, unfavourable timing for mosquito collection, seasonal workers and tiredness are some of the reasons for withdrawal. Through regular engagement, use of different sampling methods, reduction in mosquito collection frequency and rotation during collections using pyrethrum spray catch method helped to decrease the dropouts. Continuously addressing the above barriers will contribute to stable progress towards the goal of a malaria free world.

Overcoming the translation challenge: Development of a glossary for stakeholder engagement on genetic approaches to malaria control

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Malaria still remains a burden in sub Saharan Africa hence the need for development of innovative and complementary malaria control tools. Target Malaria project aims to develop and share genetic modification approaches to reduce the population of malaria transmitting mosquitoes as a control tool.

Target Malaria considers stakeholder engagement a key pillar (alongside scientific research and regulatory compliance) for the successful development of its technology. Thus, there are dedicated stakeholder engagement teams to dialogue with stakeholders at the various levels to ensure that the technology developed will be accepted. However, at community level, all engagements are better delivered in the local languages across the partner sites. Building knowledge blocks on malaria, mosquitoes; and gene drive technology is a relatively new field and terminologies are very scientific and not easy to be communicated by Stakeholder engagement teams or comprehended by the local populations. This is a challenge to the value of co-development – hence the development of the glossary. The objectives for developing the glossary were to create consistence in communicating scientific terminologies; improve stakeholders' understanding about project activities and ensure that the consent (individual level) and acceptance (community level) are informed.

The development of translated glossary required hiring professional translators to translate the scientific terminologies to local language. Our team identified and defined the scientific terminology, which were later translated to the local language. This was followed by a pretest of the translated glossary in the local communities to assess and reconcile words to achieve the objectives. A final copy of the glossary was then formulated and staff trained on its usage. The glossary will annually be reviewed.

The glossary has facilitated uniformity of communication by the stakeholder engagement team and has enhanced better understanding in communities on the science and the project thereby receiving acceptance for project activities.

### Knock Down Resistance (Kdr) Of Anopheles Gambiae Complex (Diptera: Culicidae), An Eye Opener On Resistance Surveillance In Zimbabwe

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**Background**: Establishing the extent, geographical distribution and mechanisms of insecticide resistance in malaria vectors is a prerequisite for effective resistance management. The current monitoring of insecticide resistance in Zimbabwe is often performed reactively or dependent upon on local projects being conducted. Use of adult Anopheles mosquitoes raised from larvae for insecticide resistance monitoring raises the risk of inclusion in the test of high numbers of non-vectors. Basing on a combination of different sampling methods, we report findings on the distribution of kdr in the major malaria vector, An. gambiae sl across Zimbabwe.

**Methods**: Adult mosquito samples were collected by pyrethrum spray catches, CDC light night-landing proxy catches, CDC light catches (indoor and outdoor) and raising of adults from larval collections. These samples were collected from end of year 2016 to end of year 2017 from 16 sentinel sites nationwide. Vector identification and the detection of kdr resistance alleles were determined by PCR.

**Results**: There was evidence of emerging kdr resistance alleles, ranging from 1.2 to 19.2 %, among 2282 samples of Anopheles gambiae complex sibling species submitted by sentinel sites for species identification after collection of adults in and around living structures and adults raised from wild collected larval stages. Also demonstrated was the existence of high numbers of, An. quadriannulatus within the An. gambiae complex sibling species collected as adults and when raised to adults from larvae which can significantly bias results of resistance tests conducted without confirmatory identification by PCR.

**Conclusions**: There are emerging kdr mutants among vectors collected from sentinel sites. Continued monitoring is recommended at least biennially to enable timely interventions before escalation to levels that can trigger programme failure. Confirmatory PCR identification should be mandatory for vector resistance tests as high abundance of non-vector sibling species can substantially bias the findings.

#### Mapping Resistance Intensity To Inform On The Risk Of Insecticide Vector Control Failure In Anopheles Species

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**Background:** Widespread insecticide resistance in Anopheles species threatens the efficacy of insecticidal malaria vector control tools. In 2016, WHO updated insecticide resistance monitoring test procedures in Anopheles species to include resistance intensity bioassays with 5x and 10x the discriminating concertation. In addition to visualizing discriminating dose assay data, intensity assay data can provide additional spatiotemporal information to inform on the likelihood of operational failure. IR Mapper is being updated to include data on insecticide resistance intensity in malaria vectors.

**Methods:** The Anopheles map on IR mapper (www.irmapper.com) was adapted to include filters and legends for 5x and 10x intensity assay data. A systematic literature search was conducted for publications on insecticide resistance intensity bioassay in Anopheles species from Google Scholar and PubMed from 2000 to 2016. Data were extracted, verified and uploaded to the IR Mapper platform.

**Results:** As of June 2018, peer reviewed, published insecticide resistance intensity data was available from Burkina Faso, Cameroon, Ethiopia, Kenya, Mali, Nigeria, Uganda, Zambia and Peru. All countries, with the exception of Peru, tested for pyrethroids. High intensity resistance for pyrethroids was confirmed in all African countries except Cameroon. Mali reported the greatest number of localities with surviving mosquitoes at 10x diagnostic dose. Between 2010 and 2017, 970 localities in Africa reported confirmed resistance to pyrethroids in Anopheles species on IR Mapper; only 46 localities have data on resistance intensity.

**Conclusions:** High intensity resistance was reported in the majority of countries that conducted testing. Operational failure of pyrethroid vector control tools may be likely. It is important to monitor resistance intensity where there is confirmed insecticide resistance. IR Mapper contributes to the decision making process for deployment of insecticidal tools by collating and interactively displaying resistance data on a free mapping platform.

Efficacy of Permanet<sup>®</sup> 2.0 nets against pyrethroids resistant Anopheles gambiae sl from forested area of south Cameroon

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**Background:** The rapid expansion of insecticide resistance is considered as a major threat to the use of insecticide based interventions for malaria vector control. A study was conducted to assess the influence of insecticide resistance on the efficacy of pyrethroids treated bed nets.

**Methods**: Field mosquito susceptibility to permethrin and deltamethrin was screened using the World Health Organization bioassay tube tests. Bioefficacy tests to assess bednets efficacy were conducted with the Kisumu, Ngousso and an An. gambiae resistant field colony. An experimental huts trial was conducted to compare the efficacy of Permanet 2.0. nets vs untreated nets against a resistant field strains of An. gambiae. A sub sample of An. gambiae females was analysed for species identification and to check for the presence of knockdown mutation (kdr alleles).

**Results**: A total of 2764 mosquitoes including An. gambiae, An. moucheti, An. paludis, An. nili and An. marshallii were exposed to permethrin and or deltamethrin impregnated papers. An. gambiae display high resistance to both permethrin and deltamethrin in both Olama and Nyabessan. Increase tolerance to pyrethroids was recorded for the other species. Bioefficacy tests with permanet 2.0 nets indicated low efficacy on resistant field An. gambiae colony. Experimental huts trial with the resistant colony indicated that permanet 2.0 nets induced an exophily rate of 22.6% a reduction of the blood feeding rate of 65% and a mortality rate of 23.2%. Only the west kdr allele was recorded in resistant mosquitoes. Resistant field mosquito populations included both An. gambiae and An. coluzzii.

The study suggested negative influence of insecticide resistance on the efficacy of permanet 2.0 nets. This information need to be considered when designing new strategies for the control of malaria vectors in Africa.

Key words: An gambiae sl, Pyrethroid resistance, susceptibility, LLINs, experimental hut, bioassay, Cameroon

Insecticide resistance status and high frequencies of the L1014S and L1014F mutations in Anopheles gambiae populations in Senegal

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**Background and Objective:** Like several other sub-Saharan African countries, Senegal has reduced the burden of malaria through the implementation of effective prevention and control strategies, including core vector control tools such LLINs and IRS. Despite the significant progress, the sustainability of the current vector control tools is threatened by the spread of insecticide resistance in main vectors species. A study was carried out in 2016 to characterize insecticide resistance mechanism and the target site kdr mutations in various natural populations of *Anopheles gambiae s.l.* across the four eco-epidemiological areas of Senegal: sahelian, sudano-sahelian, sudano-guinean.

**Materials and Method:** Bioassays were performed using WHO tube and CDC bottles tests on 3-5 days-old wild adult females of An. gambiae s.l.. The molecular identification of the An. gambiae complex species and the characterisation of the kdr mutations were carried out by PCR methods described by Wilkins et al. (2006) and Huynh et al. (2007), respectively.

**Results and Discussion:** Molecular identification of *An. gambiae s.l.* revealed the predominance or exclusive presence of An. arabiensis in all the study sites, except in Kedougou (sudano-guinean area) where An. gambiae and An. coluzzii were the most frequent species of the complex.

WHO impregnated paper test revealed full susceptibility to organophosphates in all the studied populations. While for the bendiocarb, the populations tested were either resistant in the urban area of Dakar or suspected to be resistant in Kedougou (sudano-guinean area), and in Niayes and Diourbel in the sudano-sahelian area. Notably, the resistance was more marked for pyrethroids in almost all the surveyed sites, except in Colobane (deltamethrin), Niakhar (deltamethrin and lambdacyhalothrin) and Koungheul (permethrin) which are also in the sudano-sahelian area. Similar trend was observed with the CDC bottles assays with less pronounced resistances levels.

The L1014F mutation (Kdr-w) was found in all the 20 health districts monitored. Overall, its allelic frequencies varied spatially, being relatively lower in the Ferlo (sahelian area), especially in the health district of Ranerou (9%). In the Senegal River Valley (sahelian area), the allelic frequencies of the Kdr-w mutation ranged from 21% (Kanel) to 35% (Podor). While in Kedougou it was almost fixed with a frequency of 95.45%. Note worthily, the east kdr mutation (L1014S) was found frequently in association with the west kdr mutation (L1014F). Its allelic frequencies varied spatially with 40.83% in the Niayes, 64.44% in Pikine, 73.53% in Rufisque, 47.73% in Koungheul(sudano-sahelian area) and 46.15% in Richard-Toll (sahelian area).

**Conclusion:** This study, in addition to updating resistance data, provides a nationwide map of the kdr resistances in Senegal to support targeted and evidence-based vector control intervention to drive toward the malaria control and elimination as aimed by the Senegalese authorities. This work is also a solid support for insecticide resistance management to keep the current core vector control tools as long as possible.

Evidence that Agricultural use of Pesticides selects Pyrethroid Resistance within Anopheles gambiae s.l. populations from Cotton growing areas in Burkina Faso, West Africa.

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**Background and objectives:** Many studies have shown the role of agriculture in the selection and spread of resistance of Anopheles gambiae s.l. to insecticides. However, no study has directly demonstrated the presence of insecticides in breeding sources as a source of selection for this resistance. It is in this context that we investigated the presence of pesticide residues in breeding habitats and their formal involvement in vector resistance to insecticides in areas of West Africa with intensive farming.

**Materials and Method:** This study was carried out from June to November 2013 in Dano, southwest Burkina Faso in areas of conventional (CC) and biological cotton (BC) growing. Water and sediment samples collected from breeding sites located near BC and CC fields were submitted for chromatographic analysis to research and titrate the residual insecticide content found there. Larvae were also collected in these breeding sites and used in toxicity tests to compare their mortality to those of the susceptible strain, Anopheles gambiae Kisumu. All tested mosquitoes (living and dead) were analyzed by PCR for species identification and characterization of resistance genes.

**Results and Discussion:** The toxicity analysis of water from breeding sites showed significantly lower mortality rates in breeding site water from biological cotton (WBC) growing sites compared to that from conventional cotton (WCC) sites respective to both An. gambiae Kisumu (WBC: 80.75% vs WCC: 92.75%) and a wild-type strain (49.75% vs 66.5%). The allele frequencies L1014F, L1014S kdr, and G116S ace -1R mutations conferring resistance, respectively, to pyrethroids and carbamates / organophosphates were 0.95, 0.4 and 0.12. Deltamethrin and lambda-cyhalothrin were identified in the water samples taken in October/November from mosquitoes breeding in the CC growing area. The concentrations obtained were respectively 0.0147ug/L and 1.49 ug/L to deltamethrin and lambdacyhalothrin. Conclusion and recommendation: Our results provided evidence by direct analysis (biological and chromatographic tests) of the role of agriculture as a source of selection pressure on vectors to insecticides used in growing areas.

Keys words: Anopheles gambiae s.l, resistance, insecticide, conventional cotton, biological cotton, Burkina Faso.

#### Impact of kdr and Ace-1 R mutations on Anopheles gambiae blood meal intake and parasite infection success.

#### Author: Moussiliou Azizath

Anopheles gambiae is a major malaria vector in Africa. Its vector importance involves the emergence of insecticide resistance mechanisms within its population. This includes kdr and Ace-1 R mutations which are associated with resistance to pyrethroid and to organophosphates and carbamates respectively. The selection of these mutations leads to an increase in vector survival, population size, fitness cost and competence. However, the impact of these resistance mechanisms on mosquito's blood meal intake and parasite development remains unclear. The present study aimed to determine the relationships between the mutations, Ace-1 R and kdr, and the mosquito's susceptibility to take a blood meal and to develop the parasite.

Four strains of An. gambiae, sharing a common genetic background, were used: one homozygous for the ace-1R mutation one for the kdr, one with both Ace-1 R and Kdr mutations and one without any of both mutations. Experimental infections of the four strains were conducted using 3D7 strain of P. falciparum gametocytes culture by direct membrane feeding assays.

The proportions of mosquitoes that took blood were 90%, 80%, 30% and 4% within the kdr, Ace1Kdr, Ace-1 R, and the susceptible strains respectively. The proportions of infected mosquitoes follow the same trend. The presence of kdr allele likely favors to the blood meal intake and the parasite infection success.

These results highlight concerns about the vector importance of mosquitoes carrying the kdr and Ace-1 R mutations. However, they are great of interest since the kdr strain could be used to better understand the parasite-vector interactions.

Role of Entomological factors in sustenance of residual malaria parasite transmission in Kilifi, Kenyan coast

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**Introduction:** The persistence in local malaria transmission has not been fully comprehended. The current study assessed the entomological factors sustaining residual malaria transmission in selected loci along the Kenyan coast in Kilifi County.

**Methodology:** Adult mosquito vectors were collected indoor and outdoor stations from six (6) selected villages in Kilifi using CDC light traps and Human Landing Catches (HLC). All the Anopheles mosquitoes collected were morphologically identified and then tested for sporozoite infection and blood meal sources using ELISA. Phenotypic insecticide susceptibility testing was done using standard WHO test kit. Sibling species of An. gambiae s.l. were separated by PCR.

**Findings:** A total of 415 Anopheles mosquitoes were collected; 57 % outdoors and 43 % indoors in the 6 villages (fisher's exact, p < 0.00001). Majority were collected by HLC (76.4 %) fisher's exact, p = 0740. Anopheles funestus s.l. was the dominant species (75 %) followed by An. arabiensis (16 %), An. gambiae s.s. (4 %) and An. pretoriensis (1 %). Plasmodium falciparum circumsporozoite protein test showed an overall sporozoite infectivity rate of 2.65 %. The total overall the indoor annual EIR for the study area (Kilifi) was estimated to be 62.05ib/p/year. Insecticide susceptibility levels to Permethrin was 96 % (0.92-0.99, 95 % C.I) and Deltamethrin - 94 % (0.89-0.99, 95 % C.I).

**Conclusion:** Malaria transmission in the study area is maintained by modestly zoophagic vectors An. funestus s.l. and An. arabiensis mostly happening outdoors. This study has shown evidence of resistance to the common pyrethroids used in LLINs. This validates the need to expand monitoring for insecticide resistance and implement an insecticide resistance management strategy to sustain the gains made in malaria control.

Attract to a habitat – kill with a larvicide: Evaluation of a potential new attract and kill strategy for the control of mosquitoes

#### Authors: Oscar Mbare

**Background:** Larvicides that persist in water over extended time periods can only efficiently control mosquitoes if their application does not produce repellent effect on gravid females. Furthermore, addition of an attractive oviposition semiochemical to a larvicide might turn an aquatic habitat into a 'reproductive sink' for mosquitoes. Here, we explored the (1) oviposition response of gravid Anopheles gambiae sensu stricto to water treated with the insect growth regulator Sumilarv<sup>®</sup>0.5G or the silicone-based surface film Aquatain<sup>®</sup> Mosquito Formulation (AMF) in semi-field systems and (2) potential of combining these larvicides with a recently discovered oviposition attractant (cedrol) for the development of a novel 'attract and kill' strategy.

**Method:** Electrocuting nets powered by a 12V battery via a spark box and surrounding an artificial pond were used to evaluate the orientation of gravid An. gambiae s.s. towards test or control pond in a semi-field system. First, the orientation of gravid females towards test pond treated with Sumilarv®0.5G or AMF was compared to control untreated water. The attractiveness of ponds treated with 20ppm cedrol was compared to untreated pond. 'Attract and kill' strategy was assessed by evaluating the orientation of females to test pond containing water treated with Sumilarv®0.5G or AMF plus cedrol compared to untreated water.

**Results:** Equal proportions of gravid *An. gambiae s.s.* approached untreated and larvicide-treated ponds indicating that neither attractive nor repellent cues from the larvicides were received by females from a distance. Neither 20ppm cedrol-treated ponds nor ponds treated with both cedrol and a larvicide increased response of gravid females.

**Conclusion:** This study did not confirm the possibility of attracting gravid females to an aquatic habitat and then killing them with a larvicide applied to the habitat. For development of this strategy, more work is required to develop improved mechanisms to release cedrol and other oviposition attractants from water

### Addressing the shortage of vector control researchers through the Partnership for Increasing the Impact of Vector Control (PIIVeC)

#### Authors: Thomsen Edward Kendall

Many of our current tools to control vector-borne diseases are inadequate in twenty-first century urban environments or failing due to insecticide resistance, and the evidence base for newer interventions remains weak. Selecting the most cost-effective and acceptable tools requires an integrated approach involving multiple disciplines. There is a particularly acute shortage of vector biologists but also an urgent need to attract leading scientists from other disciplines to tackle the persistent burden of vector-borne disease. The Partnership for Increasing the Impact of Vector Control (PIIVeC) brings together leading research institutes and control programmes from three African countries, Burkina Faso, Cameroon and Malawi, to develop evidence-based solutions for integrated vector control. One of the workstreams of this partnership is a professional development scheme aimed at post-doctoral researchers from multiple disciplines working on vector-borne disease. The scheme emphasizes three key avenues of support: mentoring packages aiming to provide fellows with career guidance and advice, secondments aiming to improve relationships with key organizations relevant to a fellows' area of expertise, and bespoke professional development plans aiming to allow fellows to acquire the training and experience they need to succeed. We will monitor the effectiveness of the scheme by tracking progress indicators on a quarterly basis, and through annual focus group discussions with the fellows. The results from these focus groups will be fed back into the scheme to improve its implementation. At the time of writing, 11 fellows have been recruited across the three partner countries. Over the next three months, their mentoring team will be established, they will complete professional development needs analyses, and they will develop their bespoke professional development plans. It is the vision of PIIVeC that through this fellowship scheme, we will contribute to the increased influence of nationally generated evidence and African researchers in global vectorborne disease policy-making.

Profil trophique des femelles d'Anopheles arabiensis, vecteur de Plasmodium humains à Ndioukhane (THIES, Senegal

**Authors:** Ndéye Diango Faye<sup>1</sup>; Oumou Kalsou Guèye<sup>2;</sup> Assane Yade Ndiaye<sup>1</sup>; Papa Mahary Fall<sup>1</sup>; Badara Samb<sup>1</sup>; Lassana Konaté<sup>1</sup> Ousmane Faye<sup>1</sup>

#### Résumé :

La détermination de l'origine des repas sanguins des femelles d'Anopheles arabiensis a été effectuée à Ndioukkane, village situé en zone soudano sahélienne, à environ 10 Kilomètres de la ville de Thiès.

De septembre à décembre 2013, des collectes mensuelles au pyrèthre de femelles endophiles ont été effectuées dans les habitations humaines où la présence ou non de moustiquaires a été notée. La composition spécifique des espèces du complexe Anopheles gambiae s.l. a été déterminée par PCR (Polymerase Chain Reaction) selon le protocole de Wilkins et al., (2006). La source du repas de sang des femelles gorgées a été déterminée par la technique de l'ELISA (Enzyme-Linked Immunosorbent Assay) directe selon la méthode de Beier et al., (1988).

Anopheles arabiensis a été la seule espèce du complexe Gambiae identifiée à Ndioukhane (171 spécimens). Des 171 repas sanguins analysés, 164 ont été identifiés, soit une proportion de 4,1% de repas n'ayant réagi avec aucun des anticorps testés.

Le repas de sang était simple (source unique) dans 71% (homme:16,5% ; cheval: 50,6% ; boeuf: 2,4% ; mouton: 1,2%) contre 29% de repas mixtes, provenant de deux à trois hôtes différents (homme, cheval, mouton, bœuf et poule).

Le taux relativement élevé de repas pris sur animal montre une déviation animale, notamment vers le cheval fréquent dans les habitations. Le recours aux moustiquaires imprégnées d'insecticide également fréquent dans le village, explique la non accessibilité des populations humaines pour les femelles de vecteurs. Par ailleurs, l'importance des repas mixtes (48/164) indique la diversification et la proximité du bétail avec l'homme.

Mots clés : Anopheles arabiensis ; plasmodium

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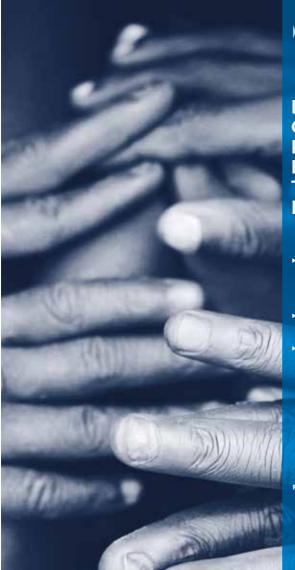
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### NOTES

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