Outdoor biting behaviour and insecticide resistance in malaria vectors might challenge malaria elimination in Southern Province, Zambia

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**Background**
- In the last decade, malaria has been reduced in Africa by 33% as a result of the rapid roll-out of malaria control interventions (WHO, 2011).
- In Zambia, long-lasting insecticide-treated nets (LLINs) and indoor residual spraying (IRS) are the main malaria vector control interventions.
- The use of these interventions depends on high coverage, vector susceptibility status to insecticides, and the indoor biting and resting behaviour of malaria vectors.
- However, there is limited information on the behaviour of malaria vectors and their susceptibility to insecticides used for vector control in Zambia’s Southern Province.
- This study aimed to monitor vector behaviour, species vector composition, and vector susceptibility to insecticides used for malaria vector control in Southern Province to guide the malaria elimination agenda.

**Methods**
- Indoor, host-seeking mosquitoes were collected from 36 randomly selected houses from April to May 2015 and from November to December 2015 using Human Landing Catches, a method used to collect live mosquitoes attempting to bite human baits both inside and outside a house.
- Three pairs of human landing collectors were recruited and trained in 12 study areas on how to collect mosquitoes. Mosquito collection was conducted on an hourly basis starting from 18:00 and ending at 06:00 for 15 consecutive days. Individual mosquito collectors were given one tablet of deltamethrin once a week as a prophylactic drug.
- Insecticide susceptibility tests were conducted on 0.05% deltamethrin, 0.1% bendiocarb, 4% DDT, and 0.25% pirimiphos-methyl following the WHO standard protocol. Metabolic resistance was determined in populations of *A. gambiae* s.l and *A. funestus* by using a synergist piperonyl butoxide (PBO).
- Multiplex Polymerase Chain Reaction (PCR) was used to determine sibling species of *A. gambiae* s.l and *Anopheles funestus*.

**Results**
- A total of 5,509 adult *Anopheles* mosquitoes were collected from Apr–May 2015 and Nov–December 2015.
- 72% (n= 3966) constituted *A. gambiae* s.l, 18% (n= 991) constituted *A. funestus*, and 2% (n = 113) were other anopheline mosquitoes (summarized in Figure 2).
- PCR confirmed the presence of *A. arabiensis*, *A. quadrimanus*, and *A. funestus s.s* in the study sites.

**Results continued**
- *A. arabiensis* mainly bit humans outdoors (0.55) rather than indoors (0.45), (ANOVA: F=7.1294, df=11, P=0.0217). In contrast, *A. funestus* mainly bit humans indoors (0.55) rather than outdoors (0.45), ANOVA: F=6.55, P=0.03.

**Conclusions**
- Standard WHO bioassays indicate that populations of *A. arabiensis* are resistant to deltamethrin; mortality rates ranged from 78% to 95% (Figure 4).
- In *A. funestus*, high resistance was detected to deltamethrin (mortality rates ranged from 14% to 42%) and bendiocarb (mortality rates ranged from 41% to 56%).
- In all the study sites, *A. arabiensis* and *A. funestus* recorded 100% susceptibility to DDT and pirimiphos-methyl.
- Pre-exposure of *A. arabiensis* and *A. funestus* to piperonyl butoxide nullified both pyrethroid and carbamate resistance.

**Figure 1.** Map of Zambia showing study sites in Southern Province

**Figure 2.** Vector species composition in the study areas of Southern Province

**Figure 3.** Biting behaviour of *Anopheles arabiensis* and *funestus* in the study areas of Southern Province

**Figure 4.** WHO bioassays of malaria vectors in the study areas of Southern Province