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# Impact of entomological interventions on malaria vector bionomics in low transmission settings in Zambia

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Entomological interventions for malaria control are being scaled up in the context of the integrated vector management strategy in Zambia. This paper reports the continuous entomological monitoring of the operational impact of indoor residual insecticide spraying (IRS) and distribution of about 6 million insecticide-impregnated bed-nets (ITN) over two peak malaria transmission seasons. Mosquitoes were captured daily using exit window traps at monitoring sentinel sites and analyzed for species identification, densities, and sporozoite rates to assess the efficacy of the vector control tools. All the three major malaria vectors; *Anopheles gambiae* sensu stricto (s.s.), *Anopheles arabiensis* and *Anopheles funestus* were collected and identified. The intervention effect of IRS and ITNs was more pronounced on *A. gambiae* s.s. and *A. funestus* than *A. arabiensis* ( $\chi^2 = 0.003$ , df = 1, P = 0.956), indicating that *A. gambiae* s.s. and *A. funestus* are amenable to control by IRS and ITNs. None of these vectors tested positive for *Plasmodium falciparum* sporozoites, thus, signifying their lack of transmission potential. This study demonstrates that entomological monitoring and evaluation is an indispensable underpinning for rational insecticide based malaria vector control. It provides compelling evidence for the need to integrate entomological parameters into routine surveillance systems, and also strongly substantiates the deployment of the integrated vector management strategy.

Key words: Zambia, malaria, impact, indoor residual spraying, insecticide treated nets, transmission.

# INTRODUCTION

In sub-Saharan Africa, high malaria transmission rates are attributable to the strong vectorial capacity of *Anopheles gambiae* sensu stricto (s.s.), *Anopheles Arabiensis*, and *Anopheles funestus* (Gillies and Coetzee, 1987; Gillies and De Meillon, 1968). However, effective malaria control efforts, including vector control and case management (Bhattarai et al., 2007; Fegan et al., 2007; Sharp et al., 2007) has resulted in decreased malaria transmission in many areas (Guerra et al., 2007; Okiro et al., 2007; Rodrigues et al., 2008; Ceesay et al., 2008; O'Meara et al., 2008). In order to reduce disease transmission more rapidly, combinations of vector control

tools have been deployed in the same malaria risk areas (Beier et al., 2008; Kleinschmidt et al., 2009).

The frontline malaria vector control interventions being harnessed for reducing vector daily survival rates in endemic countries are indoor residual spraying (IRS) and insecticide treated nets (ITNs) (Beier et al., 2008). Determining the spatial and temporal vector distribution, including monitoring of entomological risk factors and evaluating the impact of interventions on malaria transmission is essential for effective malaria control program policy development and management (Okara et al., 2010. To objectively evaluate options for malaria control, it is critical to have a thorough understanding of the ecological and epidemiological aspects of malaria and accurate estimates of malaria transmission intensity (Smith et al., 2007), as well as options for study designs to either strengthen the plausibility of findings, or establishing cause and effect.

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Figure 1. Map showing the location and spatial distribution of sentinel sites in Zambia (Chanda et al., 2011).

Available evidence indicates that malaria prevalence, incidence, morbidity, and mortality increase with transmission intensity (Molineaux, 1997; Lengeler et al., 2007; Beier et al., 1999). As such, they have frequently been used as indicators for impact of control interventions. However, measurable impacts of specific interventions on the vector population, sporozoite rates, and insecticide resistance have been observed in the field (Macdonald, 1957; Molineaux, 1997; Killeen et al., 2000; Protopopoff et al., 2007; Sharp et al., 2007).

In the past, malaria was broadly endemic across Zambia (MoH, 2000). However, significant scale-up in coverage rates of malaria control, including vector control using IRS and ITNs over the last ten years has culminated in a dramatic shift in the epidemiology of malaria (MoH, 2010). Presently, Zambia can be stratified into three malaria epidemiological zones: very low transmission areas with <1% parasite prevalence; low transmission with 10% prevalence in young children at peak transmission; and persistent high transmission with parasite prevalence of >20% at peak transmission season (MoH, 2006, 2008, 2010). This entails that malaria vector species composition; densities and infectivity are unlikely to have remained constant.

Herein, we report on the monitoring of the relative index for transmission through species abundance and infectivity over two malaria peak transmission seasons in Zambia.

# MATERIALS AND METHODS

#### Study sites and interventions

Zambia is situated in the Southern African region between 8 and 18° south latitude and between 20 and 35° east longitude with a population of approximately 13 million (CSO, 2000). Topographically, the country consists largely of a highland plateau with elevations ranging from 915 to 1,520 m above sea level. There are three distinct seasons: a cool and dry season from April to August, a hot and dry season from August to November and a warm and rainy season from November to April. The average temperatures range from 16 to 27°C in the cool dry season and from 27 to 38°C in the rainy and hot season, and vary as a function of altitude. Rainfall decreases from north to south with an average annual rainfall from 600 mm in the south to 1400 mm in the north per year. Malaria is endemic across the entire country with transmission peaks coinciding with the rainy season. The intervention consists of vector control deployment in a low malaria transmission area (Figure 1). A detailed description of interventions was presented elsewhere (Chanda et al., 2011).

#### Mosquito species identification

Mosquitoes were collected by the window exit trap method from

April 2008 to May 2010 in both IRS and ITN operational areas. *Anopheles* mosquitoes were identified morphologically as *A. gambiae* complex and *A. funestus* group (Gillies and De Meillon, 1968; Gillies and Coetzee, 1987). Sibling species were identified using polymerase chain reaction (PCR) (Koekemoer et al., 2002; Scott et al., 1993).

#### Mosquito species abundance, infectivity, and transmission

The numbers of malaria transmitting anopheline mosquitoes caught were compared over time with respect to species abundance, infection rates, transmission index, and available vector control interventions. During the collections, the number of culicines caught was recorded to ensure that in the absence of anopheline catches, the traps were being successfully operated.

#### Data management and statistical analysis

Data was collected and entered in 2007 Excel spread sheets (Microsoft Corporation<sup>®</sup>) and statistically analyzed by employing the Statistical Package for the Social Sciences (SPSS) software version 17.0. Chi-square test was used to determine the reduction in vector abundance.

#### **Ethics consideration**

Ethical clearance for this study was sought from the University of Zambia Biomedical Research Ethics Committee (Assurance No. FWA00000338, IRB00001131 of IOR G0000774 reference code 002-07-07). Written informed consent was obtained from all householders who participated in this study.

# RESULTS

# Mosquito species identification

During the period of April 2008 to May 2010, mosquitoes were trapped for 85,320 nights from 18 sentinel sites (Figure 1). *A. gambiae* s.s. was detected in two ITN sites (Chipepo and Nyamankalo) and one IRS area (Manueli). *A. arabiensis* was detected at thirteen sites; ten ITN sites (Chiawa, Chikankata, Chibombo, Chobana, Chipepo, Manueli, Mulungushi, Munenga, Nyamankalo, and Rufunsa) and three IRS sites (Kabulongo, Mukobeko, and Shyamunyimba). *A. funestus* s.s. was predominantly detected at four ITN sites (Chiawa, Chibombo, than those with IRS (Kabulongo and Mukobeko) (Figure 1). Chanda et al. (2011) reported the details of the numbers of *A. gambiae* sensu lato (s.l.) and *A. funestus* s.l. collected and identified to species.

# Mosquito species abundance, infectivity, and transmission

In this study, the relative abundance of house exiting *A. gambiae* s.s., *A. Arabiensis*, and *A. funestus* s.s. during the peak malaria transmission season showed marked heterogeneity (Table 1). The intervention effect over the

main malaria transmission season of October to April, was stronger on A. gambiae s.s. and A. funestus, as compared to A. arabiensis ( $\chi^2 = 0.003$ , df = 1, P = 0.956). There was insignificant reduction in the number of A. arabiensis from 2.14 to 0.91 ( $\chi^2$  = 0.496, df = 1, P = 0.481) with no *A. gambiae* s.s. collected in this time period. The ITNs reduced the calculated number of A. arabiensis caught per window trap per 100 nights from 2.11 to 0.18 ( $\chi^2$  = 0.579, df = 1, P = 0.447) than *A.* funestus s.s. from 0.16 to 0.05 ( $\chi^2$  = 0.058, df = 1, P = 0.810) (Table 1 and Figure 2). In the IRS areas, there was a small increase of A. arabiensis from 0.03 to 0.10  $(\chi^2 = 0.038, df = 1, P = 0.846)$  during the same periods (Table 1 and Figure 3). No A. funestus were trapped during the peak transmission season in IRS sites. Overall, there was no significant change in the numbers of vectors caught between the ITN and IRS areas ( $\chi^2$  = 0.147, df = 1, P = 0.701) in both transmission periods. The ITNs reduced the calculated number of A. arabiensis to a minimum, but IRS brought them to below detectable levels (Figures 2 and 3). No Plasmodium falciparum sporozoites were detected in A. gambiae s.s., A. arabiensis or A. funestus. Thus, no transmission index could be calculated for the three major malaria vectors during this peak transmission season (Table 1). The culicine numbers varied between sentinel sites, with densities ranging from <1 to 255.9 and from <1 to 56.0 per trap per 100 nights in 2008 and 2010, respectively. The culicines indicated that traps were being successfully operated.

# DISCUSSION

Major malaria vectors co-exist much in sub-Saharan Africa with marked variations in their malaria transmission potential (Gillies and Coetzee, 1987; Bruce-Chwatt, 1985; Coluzzi, 1984; Fontenille and Simard, 2004). Sound knowledge of their distribution and bionomics is critical in guiding and monitoring vector control efforts (Okara et al., 2010). Pioneering entomological work in Zambia implicated A. gambiae s.s., A. Arabiensis, and A. funestus s.s. as the principle vectors of malaria (DeMeillon, 1937; Adams, 1940; Watson, 1953; Pielou, 1947; Paterson, 1963; Shelly, 1973; Bransby-Williams, 1979). The present findings corroborate these studies as all the three major malaria vectors were detected. However, additional Afro tropical vectors of malaria, A. funestus-like, Anopheles rivulorum, and Anopheles nili have recently been described in the country. This necessitates assessment of their transmission potential in Zambia (Chanda et al., 2011).

To effectively manage malaria vector populations and prevent, reduce or eliminate transmission, Zambia implements an Integrated Vector Management (IVM) strategy for vector control using IRS and ITNs as main thrust interventions supplemented with larval source management in areas with amenable eco-epidemiological **Table 1.** Vector abundance, infectivity, and transmission index by period of time and intervention.

X	October to April (All sites)		October to A	pril (ITN sites)	October to April (IRS sites)	
rear	10/08 - 4/09	10/09 - 4/10	10/08 - 4/09	10/09 - 4/10	10/08 - 4/09	10/09 - 4/10
A. gambiae s.l.						
No. caught	187	38	186	31	1	7
No. analyzed for species id	187	38	186	31	1	7
A. arabiensis propn (%)	43.9	92.1	43.6	100	100	57.1
A. gambiae s.s propn (%)	0	0	0	0	0	0
A. gambiae s.s.						
No. Estimated	0	0	0	0	0	0
No per trap per 100 nights	0	0	0	0	0	0
Sporozoite rate	0 (n = 0)	0 (n = 0)	0 (n = 0)	0 (n = 0)	0 (n = 0)	0 (n = 0)
Transmission index*	0	0	0	0	0	0
Transmission index $\degree$	1	0	1	0	1	0
A. arabiensis						
No. Estimated	82	35	81	31	1	4
No per trap per 100 nights	2.14	0.91	2.11	0.81	0.03	0.10
Sporozoite rate	0 (n = 82)	0 (n = 35)	0 (n = 81)	0 (n = 31)	0 (n = 1)	0 (n = 4)
Transmission index*	0	0	0	0	0	0
Transmission index $\degree$	1	0	1	0	1	0
A. funestus s.l.						
No. caught	74	38	69	38	5	0
No. analyzed for species id	74	38	69	38	5	0
No. An. funestus s.s.	6	2	6	2	0	0
A. funestus s.s. propn (%)	8.11	5.26	8.70	5.26	0.00	0.00
A. funestus s.s.						
No. Estimated	6	2	6	2	0	0
No per trap per 100 nights	0.16	0.05	0.16	0.05	0.00	0.00
Sporozoite rate	0 (n = 6)	0 (n = 2)	0 (n = 6)	0 (n = 2)	0 (n = 0)	0 (n = 0)
Transmission index*	0	0	0	0	0	0
Transmission index $^{\circ}$	1	0	1	0	1	0



Figure 2. Average number of *A. gambiae* s.s., *A. arabiensis* and *A. funestus* s.s. per window trap per 100 nights collected, all ITN sites combined.



Figure 3. Average number of *A. arabiensis* and *A. funestus* s.s. per window trap per 100 nights collected, all IRS sites combined.

attributes. This has resulted in a marked drop in malaria morbidity and mortality (MoH, 2006, 2008, 2010). An IVM-based approach should be cost-effective, have indicators for monitoring efficacy with respect to impact on vector populations and disease transmission (WHO, 2004). Several studies on comparative operational impact of IRS and ITNs upon malaria transmission have been conducted (Neville et al., 1996; Lengeler and Sharp, 2003; Maharaj et al., 2005). Nevertheless, the potential of routine entomological surveillance data, that is, vector abundance, infectivity, and insecticide resistance have not been fully exploited in evaluation studies (WHO, 2009). Rigorous impact evaluation of the IVM is pivotal despite the limited resources and minimal time allocation. This has invariably resulted in the utilization of less rigorous study designs for establishing causal inference.

Year round tracking of entomological indicators is crucial for accurate monitoring and evaluation of ITN and

IRS impact on malaria transmission. In this study, two malaria peak seasons were compared in a low transmission operational setting. This facilitated for comparison between surveys conducted in different seasons with less bias. Deployment of IRS and ITNs during high transmission season is expected to significantly reduce the densities of malaria vectors. However, community sensitization through enhanced Information, Education and Communication/Behavior Communication Change (IEC/BCC) to scale-up acceptance of IRS and ITN utilization and adherence is critical for maintaining the efficacy of ITNs and IRS.

In Zambia, the end of the rainy season coincides with the peak in abundance of the three major vectors (Rogers et al., 2002; Gillies and De Meillon, 1968; Smith et al., 1993). The estimated numbers of *A. arabiensis* also peaked during this period. However, the relative abundance of malaria vectors was significantly reduced Table 2. Indoor resting malaria vector abundance and sporozoite rates.

Reference	Site	Ecotype	Abundance of indoor resting malaria vectors			Sporozoite rates of indoor resting malaria vectors			
			A. gambiae s.s.	A. arabiensis	A. funestus	A. gambiae s. l	A. arabiensis	A. gambiae s.s.	A. funestus
Paterson (1963)	Chirundu	Hot riverine valleys	-	-	-	2.3	-	-	-
Zahar (1985)	Chirundu	Hot riverine valleys	-	-	-	3	-	-	0
	Ndola	Savanna plateaus	-	-	-	1.6	-	-	1.6
	Livingstone	Hot riverine valleys	-	-	-	2.4	0.18	-	-
Shelly (1973)	Chirundu	Hot riverine valleys	-	-	-	1.2	-	-	-
Bransby-Williams (1979)	Chipata	Savanna plateaus	-	981	-	-	1.1	-	-
	Lusaka	Savanna plateaus	-	-	-	-	0	-	-
Chimumbwa (2000)	Lukwesa	Luapula river valley	271	29	648	-	0	5.9	4.4
	Kapululila	Hot riverine valleys	21	119	167	-	5.6	0	0
Siachinji et al. (2001)	Chibombo	Savanna plateaus	29	115	13	-	-	-	-
	Ndola	Savanna plateaus	127	5	23	-	-	-	-
	Chingola	Savanna plateaus	20	0	0	-	-	-	-
Siachinji et al. (2002)	Macha	Savanna plateaus	-	-	-	-	4.23	-	-
Kent et al. (2007)	Chidakwa	Savanna plateaus	-	-	-	-	1.6	-	-
	Lupata	Savanna plateaus	-	-	-	-	18.3	-	-

in IRS areas relative to ITN areas. This reduction concurs with findings from earlier studies that ITNs and IRS suppress the density of malaria vector populations (Neville et al., 1996; Lengeler and Sharp, 2003; Maharaj et al., 2005). Earlier data on malaria vector abundance and infectivity collected in the country exhibit markedly diverse results (Table 2). However, the lack of infectivity and thus transmission potential for *A. gambiae* s.s., *A. Arabiensis*, and *A. funestus* observed in this study could be ascribed to the low numbers of mosquitoes caught and a change in the population structure of the vectors, particularly in relative densities of *A. arabiensis*, coupled to the effective case management using artemisininbased combination therapy (ACTs) and the improved health care seeking behaviour of residents.

There is mounting evidence that combining IRS and ITNs affords enhanced protection to exposed populations compared to using one method alone (Kleinschmidt et al., 2009). However, it remains unclear whether the use of these interventions can reduce transmission intensity and result in malaria elimination. To achieve this goal, these core interventions can be supplemented in specific locations, by larval source management (LSM) strategies and maximize their impact (Utzinger et al., 2001; Killeen et al., 2002; Utzinger et al., 2002; Keiser et al., 2005; Townson et al., 2005). Nevertheless, the implementation of IVM approaches and evaluation of their impact does not only require a large financial investment in commodities and implementation, but an investment in human resources for planning, targeting, monitoring, and evaluating the various control interventions (Beier et al., 2008).

The present results validate the findings of Lengeler and Sharp (2003) that A. gambiae s.s. and A. funestus are characteristically more amenable to control by IRS and ITNs than A. arabiensis. However, the predominance of A. arabiensis that followed in the wake of effective interventions may be as a result of its exophilic nature and its catholic feeding behavior, thus, rendering it evasive to the effects of indoor targeted control interventions. These findings further substantiate the premise that vector control potentially results in a shift in species composition, as reported previously (Shelly, 1973; Bransby-Williams, 1979; Lindsay et al., 1998). The study also explains the low transmission levels of malaria in these areas, as well as authenticating the assumption that IRS has a more prompt and powerful impact than ITNs.

Earlier studies established that A. arabiensis, a vector associated with unstable malaria transmission, was the one driving transmission in the country (Shelly, 1973; Bransby-Williams, 1979; Zahar, 1985). The observed predominance of this species implies that it may be the one still perpetuating malaria transmission in Zambia. This necessitates scaled up implementation of LSM strategies, including environmental management and larviciding to facilitate the complete control of this behaviourally facultative malaria vector. The continued presence of both A. arabiensis and A. funestus in intervention areas may have implications of possible failure of the malaria control programme. It may also indicate that insecticide resistance could have been selected within the populations of these vectors, thus, making resistance surveillance imperative for the malaria control programme.

While this study has shown that entomological monitoring and evaluation is an indispensable tool for rational large scale malaria vector control using IRS and ITNs, the low numbers of malaria vectors collected may indicate a compromise in the progress and efficiency of window exit traps in low transmission zones by non-compliance of householders. Therefore, monitoring of indoor vector densities could be streamlined by replacing or complimenting the window exit traps with a more robust collection tool like the Centers for Disease Control (CDC) light trap coupled with the involvement of dedicated technical staff for close monitoring of their operations.

The recent shift in strategic emphasis from malaria control to elimination and eradication has highlighted major gaps in knowledge that need to be addressed before such achievement is contemplated (Feachem and Sabot, 2008; Feachem et al., 2009; Mendis et al., 2009). This study was conducted in low transmission settings achieved primarily by successful malaria vector control. The fact that transmission index is below 1 (Table 1), means that the disease will keep reducing. However, any strategy that targets reduction of transmission down to the level where elimination is within reach will need to strengthen its surveillance systems through very effective decision support with respect to evaluation of current vector control programmes. Furthermore, very different rigorous study designs are needed, and multiple indicators used to either establish cause and effect, or assess the strength of plausible causality.

# Conclusion

Though basic knowledge in vector bionomics is well appreciated, the demonstrated impact of IRS and ITNs provides compelling evidence for the need to integrate entomological parameters into routine surveillance systems, and strongly substantiates the deployment of an integrated vector management strategy.

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