Evaluating indoor residual spray for reducing malaria infection prevalence in Eritrea: results from a community randomized control trial

Joseph Keating[1], Andrea Locatelli[2], Andemariam Gebremichael[3], Tewolde Ghebremeskel[4], Jacob Mufunda[3], Selam Mihreteab[4], Daniel Berhane[4], & Pedro Carneiro[2]

[1]*Department of International Health and Development, Tulane University School of Public Health and Tropical Medicine, 1440 Canal Street, Suite 2200, New Orleans, LA, 70112;*

[2]*Department of Economics, University College London, Gower Street, London WC1E 6BT;* [3]*Orotta School of Medicine and Dentistry, P.O. Box 10549, Asmara, Eritrea;* [4]*National Malaria Control Program, P.O. Box 212, Ministry of Health, Asmara, Eritrea;*

*Corresponding Author: Joseph Keating, Department of International Health and Development, Tulane University School of Public Health and Tropical Medicine, 1440 Canal Street, Suite 2200, New Orleans, LA, 70112; Tel: 504.988.1348; Fax: 504.988.3653; Email: jkeating@tulane.edu

Abstract

This paper examines the relationship between indoor residual spray (IRS) and malaria parasite infection in Gash Barka Zone, Eritrea, an area with near universal coverage of insecticide treated bednets (ITN) and already low malaria parasite prevalence. A community randomized control trial was conducted in 2009. Malaria parasite infection prevalence was 0.5% [95% confidence interval (CI): 0.37 – 0.78%], with no significant difference detected between treatment and control areas. ITN possession remains high, with over 70% of households reporting ITN ownership [95% CI: 68.4 – 72.9]. ITN use among individuals within ITN-owning households was just under half [46.7% (95% CI: 45.4 – 48.0)]. Slight differences in ITN possession and use were detected between treatment and control areas. There was no significant difference in malaria parasite infection prevalence among individuals in households with ≥1 ITN compared to those in households without ITNs, nor among individuals reporting ITN use. Among individuals in ITN-owning households, sleeping under an ITN offered no statistically significant protection from malaria parasite infection. Community participation in environmental and larval habitat management activities was low: 17.9% (95% CI: 16.0 – 19.7). It is likely that IRS, larval habitat management and ITN distribution alone may be insufficient to interrupt transmission without corresponding high ITN use, sustained IRS application in areas where infections are

clustered, and promptly seeking laboratory diagnosis and treatment of all fevers. Eritrea is ready for elimination, irrespective of inconclusive impact evaluation results.

1. Introduction

Eritrea has made great strides with respect to the control of malaria, having greatly reduced malaria infection prevalence using a combination of case management, larval habitat management (LHM), free insecticide treated net (ITN) distribution strategies, prompt epidemic response, and indoor residual spray (IRS) in epidemic prone areas. Despite considerable advances in the reduction of malaria transmission over the last 12 years, malaria elimination has not yet been achieved and malaria remains the leading cause of death among

children under 5 years old in Eritrea. Many National Malaria Control Programs (NMCP) in Sub-Saharan Africa (SSA) are developing a plan for further reducing infections in already low malaria transmission settings, with the goal of eventual malaria elimination.

Recent studies have shown IRS to be an effective strategy for preventing malaria infection and mortality across a range of settings (Musawenkosi *et al.* 2004, Sharp *et al*. 2007, Zhou *et al*. 2010, Kleinschmidt *et al*. 2009, N'Guessan *et al*. 2007, Pluess *et al.* 2010), although more tests are needed to assess the effect when used in combination with other interventions such as ITNS (Pluess *et al.* 2010). One recent review by Eisele and colleagues (2010) estimates the protective efficacy (PE) of ITNs and IRS on reducing malaria-attributable mortality among children aged 1–59 months to be 55% (Range = 49–61%), in areas where *P. falciparum* is the predominant parasite type. These data support the continued testing and scale-up of malaria prevention interventions, as the combination of proven intervention measures will likely reduce morbidity and mortality due to malaria. That being said, we know little about the added benefit of IRS, over and above existing control tools, in the context of already low transmission and near universal coverage of ITNs.

There has recently been renewed interest in the use of IRS for malaria prevention and control (WHO 2006), especially as countries scale up for eventual malaria elimination. As well, there are now standardized household surveys (e.g. Malaria Indicator Survey) that include questions for collecting data on houses sprayed and assessing IRS coverage at the population level. When collected in concert with blood samples, these surveys can be used for measuring program progress in a standardized way across time and space. However, to date there are relatively few data showing the relationship between IRS and malaria parasite infection prevalence under extremely low transmission conditions. Because of the positive externalities observed during trials, where IRS confers protective benefits to unprotected houses when high intervention coverage levels are achieved, relative reductions in malaria parasite prevalence associated with IRS under

low transmission conditions may be lower than achieved elsewhere. As malaria transmission continues to drop in many areas of SSA, it is increasingly critical that studies be conducted to assess ideal malaria control and prevention strategies under low-transmission conditions.

This paper examines the relationship between IRS and malaria parasite infection in Gash Barka Zone, Eritrea, an area with near universal coverage of ITNs and relatively low malaria parasite prevalence. The objectives of this paper are to: 1) test whether adding IRS to an existing package of interventions (i.e. LHM, case management, and ITNs) can further reduce malaria parasite infection prevalence; 2) determine whether IRS coverage is associated with reduced risk of malaria parasite infection among residents within this context, while controlling for potential confounding factors related to age, sex and ITN possession and use; and 3) report on the current state of ITN possession and use within Gash Barka. This paper lends insight into whether Eritrea is poised for malaria elimination, what additional steps may be necessary to further this goal, and important lessons related to study design and sample size in low-transmission settings. Ethical approval for this analysis was obtained from the Institutional Review Boards (IRB) of Tulane University and the NMCP in Eritrea.

2. Materials and Methods

2.1 Study site description

Eritrea has an estimated population of 3.6 million people and is divided into 6 administrative *zobas* (zones); this study was conducted in Gash Barka, a mostly rural/agricultural area with relatively higher malaria transmission than the other malarious zones in the country (Figure 1). The population of Gash Barka Zone is 30% of the total population in Eritrea. Altitudes range from below sea level to 3,000 meters above sea level country-wide, although altitudes in Gash Barka range from approximately 500 meters to 1,500 meters only. Temperatures vary widely across Eritrea; the western lowlands (Gash Barka) are generally associated with extremely hot and dry climatic

conditions with seasonal precipitation, concentrated in the summer months. The average precipitation in the western lowlands is approximately 200 mm per year (NMCP program data).

Anopheles arabiensis and *An. gambiae* s.s., both belonging to the *An. gambiae* complex, form the main vectorial system in Eritrea (Shililu *et al.* 2003b; Shililu *et al.* 2003a; Shililu *et al.* 2004). *Plasmodium falciparum* is the primary species of malaria found in Eritrea, although *P. vivax* has also been reported (Shililu 2004). Malaria transmission is seasonal, with peak transmission occurring during the months of September through November, just after the rainy season. A smaller malaria season is often observed during the months of March through April, depending on the timing and quantity of precipitation occurring. Estimates of entomological inoculation rates range from zero to 70.6 infective bites per person per year in Gash Barka Zone (Shililu *et al*. 2003b).

Health facility data in our study area suggest that malaria infections continue to fall (Figure 2). In 1998, over 110,000 cases of malaria were diagnosed in health facilities; by 2009 this number had fallen to just under 18,000. (Note: data on the normalized vegetation index (NDVI), a commonly used proxy indicator for vegetation cover and wetness in an area, shows that vegetation levels have increased over the past 10 years, so malaria reductions observed are likely not due to less favorable environmental conditions). Over 50% of all outpatient and inpatient diagnosed malaria in the country comes from Gash Barka Zone and over 60% of all malaria related deaths came from Gash Barka in 2007 and 2008. Rapid diagnostic tests (RDTs) have been used since 2005 throughout the public sector for malaria diagnosis, with Artemisinin-based combination therapy (ACT) (Artesunate + Amodiaquine) used as the first-line treatment for *P. falciparum* and *P. vivax* malaria infection.

2.2 Evaluation study design

A two-arm cluster-randomized community-controlled trial, post-test only design was used to evaluate the impact of IRS on malaria infection prevalence. This evaluation design measures the additive impact of IRS over current NMCP interventions. Effectiveness was measured as a single difference between treatment and control groups. The study was conducted in Gash Barka Zone only. We have chosen to conduct this evaluation in Gash Barka because of its high burden of malaria relative to other Zones in Eritrea and because the Eritrean NMCP is already rolling out ITNs, case management, and LHM in the area. Fifty-eight (58) villages within Gash Barka were randomly assigned to the treatment group and 58 villages in Gash Barka were randomly assigned to serve as the control group. A geographic buffer was used to insure that treatment and control villages were at least 5 km apart. The NMCP verified the distance between treatment and control villages; in two instances whereby a treatment village was too close (< 5 Km) to a control village, the closest village > 5 km was selected into the control group. As well, replacements were made where the originally chosen village had moved or could not be reached. Again, the closest eligible village was chosen as a replacement.

2.3 *Intervention*

The intervention involved the control of adult mosquito populations using IRS with the insecticide DDT, the insecticide recommended by the Eritrean NMCP. In each intervention village, dwellings were sprayed according to the manufacturer's recommended guidelines. The spraying targeted all households to ensure a minimum coverage of 80%, as per WHO recommendations. Villages in the intervention arm also benefitted from existing NMCP vector control interventions such as ITN and LHM, as well as continued case management. Spraying was done during the months of June-July in 2009. The control arm villages received ITN, LHM and case management only.

2.4 *Survey design*

As described elsewhere (Turner *et al.* 1996), a modified two-stage cluster design was used to generate a sample of households within the study area for treatment and control survey domains, separately. A list of eligible villages was obtained from the NMCP. STATA (*Sample* command) was used to randomly select 116 villages, and then randomize the villages into treatment and control groups at the first stage, thus ensuring that any differences in important village-level characteristics are due to chance. At the second stage, segmentation was used to divide the village into equal segments of approximately 200 houses. Houses were enumerated and simple random sampling was used to select 15 houses within each village to serve as ultimate sampling units, thereby obtaining a total of 870 (n = 15*58) houses per domain (*n*= 1,740 total).

Data were collected during October 6 – 15, 2009, which corresponds to the peak of the malaria transmission season. Medical students from the Orotta School of Medicine and Dentistry (OSMD) in Asmara served as data collectors, with supervision from staff at the NMCP and OSMD.

2.5 Outcomes

A modified malaria indicator survey (MIS) style questionnaire was administered to one resident adult at randomly selected houses in both the control and intervention villages at the completion of intervention. Household data were used to generate control variables related to age, gender, vector control, ITN possession and ITN use. ITNs were defined as any net that was treated at least once in last 11 months, or is a permanently treated net. Parasite infection and febrile illness data were collected from all household residents greater than one month old. For all residents present at the time of the survey (15.4% were away) and who did not refuse (11.8% of those present refused), a small sample of blood was taken from the finger (by standard finger-prick methods using a sterile lancet) for the rapid diagnosis of malaria using RDTs (*Carestart*®). For individuals testing positive, a second drop of blood was taken to make a thick blood film for confirming parasite

infection status. Blood slides were transported to Barentu Zonal Referral Hospital Laboratory and NMCP headquarters in Asmara for staining and microscopic evaluation. Two separate microscopists examined blood-slides (100 thick film fields x 1,000x), one in Gash Barka and a second in Asmara, to determine parasite infection status. Individuals testing positive for parasites were treated with the recommended first-line anti-malarial drugs used in Eritrea. The parent/guardian was responsible for administering the treatment and team members were present to observe the first dose of treatment. Malaria parasite infection prevalence was defined as the proportion of respondents with any detectable parasites out of all respondents that provided a blood sample.

To compare ITN-related behaviors in treatment versus control households, ITN possession and use among household residents were ascertained with a net roster and a registry of all household residents. ITN possession was defined in the following two ways: 1) the proportion of individuals living in a household with ≥1 ITN; and 2) the proportion of individuals living in households with at least 1 ITN per 2 residents (i.e. ITN-to-occupant ratio of 1:2). ITN use was defined in the following two ways: 1) the proportion of individuals in households reporting to have slept under an ITN the night before the survey, and 2) the proportion of individuals living in households where any ITN was used the night before the survey. ITN possession analyses were conducted among all individuals providing a blood sample (n = 5,502). ITN use analyses were conducted among individuals providing a blood sample living in households with ≥1 ITN to assess the effect of personal protection, independent of having access to an ITN in the household (n = 4,078).

Confirmed cases of malaria at health centers within Gash Barka Zone were collected and plotted to investigate trends in malaria infection over the past 10 years. While no analysis was done with these data, it does serve to elucidate the malaria transmission trend in the area at the health facility level. These data represent only those infected individuals seeking treatment, although it is plausible that a

significant proportion of infections will present at hospitals due to lowered protective immune response as a result of reduced exposure to parasites over time, thus serving as a proximate determinate of the overall malaria burden within the community.

2.6 Data analysis

Stata 10.1 (Stata Corporation, College Station, Texas) was used to perform all statistical analyses. Chi-square statistics and analysis of variance statistics were used to test for differences in ITN ownership between treatment and control houses. Chi-square statistics were used to test for differences in malaria parasite infection prevalence by treatment and control groups, and by ITN possession and use variables.

Logistic regressions were used to test whether IRS predicted malaria parasite infection prevalence, while controlling for age, sex, and ITN possession at the individual level. Secondly, we tested whether IRS predicted malaria parasite infection prevalence, while controlling for age, sex, and ITN use at the individual level. The age variable was dichotomized to equal 1 if the individual was under the age of 15 and 0 if the individual was 15 years old or older (Note: the variables capturing larval habitat management and environmental management at the household level were not used as a control variables in regressions, as no infections were detected among those practicing LHM or environmental management. Thus, the distribution of the data was such that performing meaningful multivariate regression analysis was not possible). Wald statistics were used to identify variable significance with the probability of committing a type-1 error (alpha) set at 0.05. Standard errors were adjusted to account for correlated data at the village level (*cluster*). Probability weights (*pweight* command in Stata), equal to the inverse of the probability of household selection within a village given the sampling design, were applied to point estimates.

3. Results

A total of 1,616 households in the sample completed a questionnaire, giving a household non-response rate of 7.1%. A total of 7,273 individuals resided in participating houses; 5,502 individuals were home and consented to testing for malaria parasite infection, resulting in an individual test refusal rate of 10.6%. Among individuals at home, test refusal rates differed between treatment (8.5%) and control (12.7%) groups (p < 0.05). Where individuals were either at work or school at the time of the survey and follow-up visits, no malaria parasite testing was performed. The distribution of individuals living in houses located in treatment and control villages was similar on sex, age, employment status of the respondent, and education level of respondent. As such, we are confident that our treatment and control groups were similar on characteristics (i.e. the randomization strategy worked) that may have influenced the overall risk of malaria parasite infection and use of malaria prevention and control interventions.

3.1 Indoor Residual Spray

Within treatment villages, 84.8% of respondents reported that the inside of the house walls was sprayed within the last 12 months. Ninety-five percent (95%) of those houses located in treatment villages and reporting spraying indicated that someone had sprayed their house within the past 4 months, suggesting that most spraying was done as part of this intervention. While some houses within control villages also reported that their houses had been sprayed within the last 12 months (9.4%), over 25% of these respondents reported that spraying had occurred greater than 8 months previously, suggesting that any spraying done was not part of this intervention. Program records indicate that no NMCP staff sprayed within houses located in control villages over the past 1 year; as personal spraying in rural and semi-rural setting is uncommon in Eritrea, it is not clear if these households engaged in personal spraying using commercially bought insect repellent to coat their walls, or if ambiguity in the question existed during the interview. That being said, DDT is not available on the local

market for purchase, so any spray activity likely involved a different insecticide, which has a much smaller effect than DDT and which becomes ineffective shortly after use.

3.2 Larval Habitat Management

Among household respondents completing the questionnaire (n = 1,616), 17.9% (95% CI: 16.0 – 19.7) reported participating in LHM activities. There was no difference in the proportion of respondents reporting LHM practices among houses located in treatment versus control villages (χ^2 = 1.35, p = 0.25) (Table 1). Significantly more females (23.8%) reported LHM activities than males (15.1%). Among those reporting environmental management activities (n = 292), 76.0% reported either filling in or draining water bodies. Approximately 12% reported removing standing water from around the house and 11.0% reported cutting tall grass around the house to eliminate mosquito habitat. Most respondents reported that the household spent less than or equal to 2 days per month participating in environmental management activities. There were no differences detected among respondents located in treatment versus control villages. A total of 25,151 water bodies were either filled or drained in the study area during 2009, and as many as 44,790 water bodies were treated with Temephos® within the study area during 2009.

3.3 Insecticide Treated Nets

Results from the household survey show that overall 70.6% [95% CI: 68.4 – 72.9] of households in our study area possessed ≥1 ITN. No difference in ownership of any ITN. was detected between houses located in treatment (72.1%: [95% CI: 69.0 – 75.3]) versus control villages (69.1%: [95% CI: 65.9 – 72.3]) (χ^2 = 1.79, p = 0.189). Over 81% (95% CI: 79.1 – 83.0%) of households owned ≥1 mosquito net whether treated or not. The majority of respondents with nets that were never treated or retreated reported not being aware of the need to

retreat as the main reason for not seeking retreatment. Absence at the time of community-level retreatment programs was the second most common reason cited for not seeking retreatment of nets.

Among households reporting ITN ownership, only 39.7% (95% CI: 36.8 – 42.5) reported having at least 1 ITN per two household occupants (i.e. ITN-to-occupant ratio of 1:2) ($n = 1,146$). No difference in the ITN-to-occupant ratio was detected between houses in treatment and control villages ($\chi^2 = 1.35$, p = 0.248). Over the entire sample, we detected an average of 1.35 ITNs per house (95% CI: 1.29 – 1.40); among houses owning an ITN ($n = 1,146$), the mean number of ITNs possessed was equal to 1.9 per house (95% CI: 1.85 – 1.96). A statistically significant difference was detected between the mean number of ITNs in houses located in treatment (1.97 [95% CI: 1.89 – 2.05]) versus houses located in control (1.82 [95% CI: 1.75 – 1.90]) villages; houses located in treatment villages had slightly more ITNs than houses located in control villages (F = 7.87, p < 0.01).

Among all individuals in the sample (n = 7,894), 73.5% (95% CI: 72.5 – 74.5) slept in a house possessing ≥1 ITN. Among individuals in the sample that provided a blood sample for microscopy diagnosis (n = 5,502), 74.0% (95% CI: 72.0 – 75.8) slept in a house with ≥1 ITN (Table 1). ITN household possession was slightly higher among individuals living in houses located in treatment villages as compared to houses in control villages ($\chi^2 = 11.2$, p < 0.01) (Table 1). There was also slightly lower proportions of older individuals living in houses with an ITN as compared to younger individuals in the sample who provided blood for parasite testing ($\chi^2 = 31.54$, p < 0.01). No significant differences in ITN household possession was detected between males and females ($\chi^2 = 1.51$, p = 0.22).

Among individuals living in ITN-owning households and providing a blood sample ($n=4,078$), just under one-third [28.9% (95% CI: 27.5 – 30.3)] slept in a house with ≥1 ITN per 2 occupants (Table 1). The proportion of individuals living in households with ≥1 ITN per 2 occupants was slightly higher among individuals living in houses in treatment villages (31.2% [95% CI: 29.2 – 33.1]) as compared to

control villages (26.3% [95% CI: 24.3 – 28.3]) (χ^2 = 14.6, p < 0.01). As well, older individuals were more likely to be living in an ITN-owning household with a ratio of at least 1 ITN per 2 people, as compared to younger individuals providing a blood sample.

ITN use was low within ITN-owning households, with just under half [46.7% (95% CI: 45.4 – 48.0)] sleeping under an ITN the previous night. Among individuals in ITN-owning households located in treatment villages, 48.8% (95% CI: 47.0 – 50.6%) slept under an ITN the night before the survey; only 44.4% (95% CI: 42.5 – 46.2) of individuals living in ITN-owning households located in control villages reported using an ITN the night before the survey (χ^2 = 11.5, p < 0.01). ITN use was significantly higher among younger individuals as compared to older individuals in the sample (χ^2 = 85.6, p < 0.01). Among ITN-owning households, significantly higher ITN use was observed among females (51.1% [95% CI: 49.2 – 52.9]) as compared to males (41.9 [95% CI: 40.1 – 43.8]) (χ^2 = 48.2, p < 0.01). Similar relationships were found when the data were restricted to only individuals consenting to provide a blood sample; these numbers are reported in table 1.

3.4 Malaria Parasite Infection Prevalence

A total of 30 infections were detected among individuals in our sample; all RDT positive tests were also positive via microscopy. Ten infections were detected in Haykota sub-zone; no infections were detected in Barentu, Dighe, or Tesseney sub-zones. Infections were distributed across 22 villages; a total of 24 houses within those villages contained at least one infection. One house in Hlet Jedida village contained 6 infections: this anecdotal observation suggests that at such low prevalence spatial clustering of infection remains an important consideration.

Malaria parasite infection prevalence by RDT was very low at 0.57% (95% CI 0.37 – 0.78) among all individuals living in selected houses and consenting to give blood (n = 5,502) in our study area. Bivariate analyses showed no difference in parasite infection prevalence

between those individuals in households in treatment versus control villages ($\chi^2 = 0.24$, p = 0.62), or between male and female individuals in selected houses ($\chi^2 = 2.54$, p = 0.12). A total of 26 RDT positive individuals (26/30) resided in treatment villages. However, there was a difference detected among those less than 15 years old (0.83% [95% CI: 0.5 – 1.2]) versus those greater than 15 years old (0.30% [95% CI: 0.12 – 0.58]) ($\chi^2 = 5.77$, p = 0.02), with 17 of 30 infections detected among those between 5 and 15 years old and 4 infections detected among those less than 5 years old.

Bivariate analyses also showed little difference among individuals living in houses with or without an ITN ($\chi^2 = 3.27$, p = 0.07), although it is worth noting that randomization was performed on IRS, not ITN distribution. Among all individuals providing a blood sample (i.e. within both ITN-owning and non-ITN owning houses), there was no difference in parasite infection prevalence between those who used an ITN or did not use an ITN the previous night ($\chi^2 = 0.18$, p = 0.67), nor between those in households where the ITN to occupant ratio was greater than 1 ITN per 2 people ($\chi^2 = 1.33$, p = 0.27). A total of 26 RDT positive individuals (26/30) reportedly slept in a house that owned at least 1 ITN.

Among those individuals within ITN-owning households, there was also no difference in parasite infection prevalence detected between those who used or did not use an ITN the previous night ($\chi^2 = 1.47$, p = 0.24). A total of 10 RDT positive individuals (10/30) reportedly slept under an ITN the previous night.

3.5 Multivariate Regression Analyses

Results from two logistic regressions, controlling for individual age, sex, ITN use, and treatment versus control in the study area in one regression, and controlling for sex, age, household-level ITN possession, and treatment versus control in a second regression suggest that neither IRS, ITN possession nor ITN use is significantly related to malaria parasite infection prevalence in this area (Tables 2 and 3). A

marginally significant positive relationship was however detected between age and infection; those under the age of 15 were over 2 times more likely to be infected as compared to those 15 years old and over across all models. No other control variables tested significant in these analyses.

Additional regressions were performed using the ITN-to-occupant ratio variable (e.g. proxy for ITN ownership), and whether anyone in the house slept under an ITN variable (e.g. proxy for ITN use) as control for ITN possession and use (results not shown). The results of these additional regressions were similar to the results we present in the above models, and also show that neither IRS, ITN possession, nor ITN use are related to malaria parasite infection prevalence in this context.

4. Discussion

The prevalence of malaria parasite infection in our study area was extremely low (0.5%) at the end of the peak malaria transmission season in 2009, and we showed no difference in prevalence between individuals in treatment versus control villages. This low malaria infection prevalence in our sample population could be due to the fact that the parasite or mosquito vector population had been sufficiently suppressed in this area of Eritrea prior to the survey, implying that the time to contemplate strategies for elimination is now. Given that the prevalence of malaria parasite infection in Gash Barka was twice as high among younger individuals in our sample, even with high ITN coverage and overall low malaria parasite infection prevalence in the population, children and young adults may be a good target for further reductions in parasite infection towards elimination.

Results from this evaluation also show that Gash Barka Zone, Eritrea, has achieved near universal coverage of ITNs with over 70% of households possessing ≥1 ITN. However, only a minority of individuals in the sample lived in households with adequate intra-household

access to an ITN, defined as ≥1 ITN for every 2 household occupants, and ITN use in ITN-owning households remains low with less than half of all individuals reporting to have slept under an ITN the previous night. This finding also provides a useful starting point for further efforts to scale up control towards elimination.

Further, within this context of intervention-suppressed transmission with high ITN coverage, there was no significant difference in malaria parasite infection prevalence among individuals living in households with or without an ITN. Our analyses also showed sleeping under an ITN the previous night to offer no statistically significant reduction in the odds of malaria parasite infection, among those within ITN-owning households. This observation could be related to low mosquito population densities at the time of the survey (perhaps as a result of IRS), or the level of precipitation in parts of the study area was insufficient to produce an abundance of malaria vectors in 2009; however NDVI in 2009 was not particularly low and the precipitation pattern in 2009 was very intermittent, which can be conducive to more malaria not less. Although an attempt was made to collect entomological data in support of this evaluation, insufficient numbers of mosquitoes were caught (and indeed those that were caught were collected after the collection of parasite data), which precluded a robust analysis.

Previous studies have shown reductions in *Anopheles* density and transmission within areas of very high IRS coverage (Zhou et al. 2010). It has also been suggested that concomitant protection against malaria may be conferred to control villages without IRS in proximity to intervention villages, although this should not be the case given that treatment and control villages were located at a minimum distance of 5km from each other. Given that this intervention achieved greater than 85% coverage of houses in treatment villages, protection may have been conferred to our control groups, which may help explain our null results. To some extent, we hypothesize that within this context individuals unprotected by IRS (i.e. in households that did not receive spray) may have benefited from an overall suppression of

transmission in this area due to near universal coverage of ITNs and control efforts ongoing for the past 12 years. In other words, repeated high coverage of ITNs and treatment efforts in the past for this area of Eritrea may still be working to suppress mosquito population, rendering any additive effect very difficult to detect. Although our data preclude definitive evidence of a community effect of IRS on decreasing malaria parasite infection prevalence, our results do suggest that at least partial protection has been conferred on the population in Gash Barka, as evidenced by similar prevalence of malaria parasite infection between treatment and control areas, irrespective of exposure to ITN household possession or use, or LHM.

This study had several limitations worth noting, which might also explain our null results and should serve as lessons learned for future impact evaluations. First, it is possible that our study design (post-test only) failed to capture pre-IRS information on prevalence within our study areas, thus determining how low prevalence was prior to the intervention was not possible. However, our analysis does show that there are few differences between treatment and control household characteristics (e.g. sex composition, size, age), which suggests that randomization worked, thus making our groups comparable. While there is anecdotal evidence that malaria transmission was once high within this area of Eritrea, we could identify no published data that quantified the level of malaria transmission prior to the IRS roll out in 2009, thus an assessment of the relative decline in malaria transmission associated with IRS intervention was only possible using non-representative health facility data.

Second, given that malaria parasite prevalence will likely continue to decrease with increasing ITN coverage, and sustained LHM and access to treatment it is possible that monitoring and evaluation of malaria control scale-up efforts over time using cross-sectional data collection strategies and post-test only study designs will not be feasible due to high costs associated with large sample sizes needed to detect small changes; other studies have also shown that large sample sizes are needed to assess changes at very low transmission levels

(Eisele *et al*. 2010). We therefore recommend that in low transmission settings, following a cohort of individuals through time to get at malaria incidence may produce more useful results for the monitoring and evaluation of malaria prevention and control activities, while maintaining reasonable costs and generalizability. Our study was indeed not powered to detect such small difference in malaria parasite prevalence between treatment and control areas at this very low transmission level, as it was assumed that pre-intervention prevalence was much higher than was most likely the case. Our sample size of 870 households in the treatment group and the same number in the control group was calculated to detect a reduction of 10.5% points at a 5% level of significance in malaria parasite infection prevalence from a baseline of about 15% with 80% power, and assuming a design effect of 2.0. We are under the assumption that baseline prevalence was much lower than 15%, although the exact figure remains unknown.

Third, the timing of the study was such that data were collected at the end of the peak malaria transmission season, possibly skewing the prevalence estimates downward, although RDTs can detect residual antigen from infection (e.g. current and previously treated infection) up to several weeks after treatment (Hopkins *et al*. 2007, Mayxay *et al*. 2001). It is also possible that the timing of the study was such that mosquito vector populations had already been suppressed in our study areas or the majority of cases had already been detected and treated, resulting in point estimates reflective of late transmission season only.

5. Conclusion

In conclusion, our results suggest that with near universal coverage of ITNs and continued efforts to manage larval habitats, providing access to prompt and effective treatment, and applying IRS in response to epidemics as in the past, malaria parasite infection prevalence is extremely low at 0.5% in Eritrea, which is therefore poised for elimination. Our results also show that even under such

intervention-suppressed transmission, age remains an important risk factor for malaria, making this group a likely target for elimination strategy development. ITN use was also low in this area in 2009 among those in households possessing them, so campaigns geared towards increasing ITN use could also confer additional protection. It is quite possible that Eritrea is ready for elimination, irrespective of our inclusive impact evaluation results. That being said, ITN possession and IRS alone may not be sufficient to interrupt transmission; it is therefore imperative that Eritrea, continue promoting prompt seeking of laboratory diagnosis and treatment of all fevers, ITN use, and continued IRS over multiple transmission seasons.

Acknowledgments

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Table 1: ITN ownership, use and environmental management among those tested for malaria parasites in Gash Barka, Eritrea 2009

	Individual lives in household owning ≥1 ITN (n=5,502)		Individual lives in household with ITN to occupant ratio ≥1:2 (n=4,078)[§]		Individual used ITN previous night (n=4,078)[§]		Household participated in environmental management in past month (n = 1,616)[Ŧ]	
	%	95% CI	%	95% CI	%	95% CI	%	95% CI
Age in years								
0-4	77.6	75.0 – 80.3	24.1	21.0 – 27.2	58.8	55.2 – 62.3	N/A	N/A
5 - 14	77.7	75.6 – 79.8	22.3	19.9 – 24.6	46.5	43.7 – 49.3	N/A	N/A
15 - 24	70.5	67.2 – 73.7	32.3	28.3 – 36.2	42.5	38.4 – 46.7	25.8	14.0 – 37.6
25 - 34	77.9	74.7 – 81.0	32.9	28.8 – 36.9	55.0	50.7 – 59.3	16.7	12.4 – 20.9
35 - 44	75.5	72.0 – 79.0	30.9	26.6 – 35.3	46.0	41.4 – 50.7	17.8	14.0 – 21.7
45 – 54	67.4	62.9 – 72.0	31.8	26.3 – 37.3	46.3	40.4 – 52.2	20.3	16.2 – 24.3
55 – 64	60.6	55.0 – 66.3	47.6	40.3 – 54.9	36.4	29.4 – 43.5	17.1	12.7 – 21.5
< 65	57.6**	51.0 – 64.3	49.7**	40.9 – 58.4	40.4**	31.8 – 49.0	13.9*	9.3 – 18.5
Sex								
Male	74.7	72.9 – 76.5	27.8	25.7 – 29.9	43.1	40.8 – 45.5	15.1	13.0 – 17.3
Female	73.2	71.7 – 74.8	30.0	28.1 – 31.9	52.5**	50.5 – 54.6	23.8**	20.1 – 27.5
Intervention								
Treatment	75.8**	74.2 – 77.4	31.2	29.2 – 33.1	50.7**	48.6 – 52.8	18.6	16.0 – 21.3
Control	72.0	70.2 – 73.7	26.3	24.3 – 28.3	46.2	43.9 - 48.6	17.1	14.5 – 19.8
Total	74.0	72.8 – 75.1	28.9	27.5 – 30.3	48.6	47.1 – 50.2	17.9	16.0 – 19.7

[§]Among those living in households owning an ITN
[Ŧ] Among household respondents
*P < 0.05; **P < 0.0 1
N/A = no household respondents were below the age of 15 years old

Table 2: Logistic regression predicting odds of parasite infection relative to ITN ownership and treatment with IRS, among those providing a blood sample

	Malaria parasite infection prevalence ($n=5,508$)	
	AOR	95% CI
Household possesses any ITN	2.38	0.75 – 7.52
Age in years (dichotomized)		
Less than 15 years old	2.23	0.91 – 5.43*
15 years and older (Reference)	1.00	
Sex		
Male	0.62	0.29 – 1.32
Female (Reference)	1.00	
Intervention with IRS		
Treatment	1.16	0.40 – 3.34
Control (Reference)	1.00	
Pseudo R^2	2.8%	

* $P < 0.07$
AOR: Adjusted odds ratio
CI: Confidence interval

Table 3: Logistic regression predicting odds of parasite infection relative to ITN use and IRS treatment, among those providing a blood sample and living within an ITN-owning house

	Malaria parasite prevalence ($n=4,042$)	
	AOR	95% CI
Individual used ITN night before the survey	0.61	0.19 – 2.00
Age in years (dichotomized)		
Less than 15 years old	2.47	0.94 – 6.49*
15 years and older (Reference)	1.00	
Sex		
Male	0.63	0.28 – 1.42
Female (Reference)	1.00	
Intervention with IRS		
Treatment	1.04	0.30 – 3.54
Control (Reference)	1.00	
Pseudo R^2	2.6%	

* $P < 0.07$
AOR: Adjusted odds ratio
CI: Confidence interval