RESEARCH ARTICLE

REVISED Implementing a community vector collection strategy for monitoring vector-borne diseases in Ghana [version 2; peer review: 1 approved, 1 approved with reservations]

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v2 **First published:** 06 Mar 2019, **3**:722
https://doi.org/10.12688/gatesopenres.12933.1
Latest published: 29 May 2019, **3**:722
https://doi.org/10.12688/gatesopenres.12933.2

Abstract

Background: Monitoring vector-borne diseases requires sampling of very large numbers of disease vectors in order to corroborate infections in the human population. This can be challenging, as current vector collection tools are either inefficient, or expensive to implement from a public health perspective. To circumvent this challenge, this study compared a community vector collection strategy using a double-netted mosquito collection method (a tent trap (TT)) to the traditional human landing collection (HLC) method in three communities in lymphatic filariasis-endemic districts in Ghana.
Methods: Following community entry and sensitization, community volunteers appointed by the community leaders were trained in the mosquito collection and storage methods and provided with supplies for mosquito collection over a 7-month period. They were visited occasionally by the study team to retrieve the mosquito samples for identification. The collectors were also assessed to evaluate their perspectives on using community vector collectors for monitoring vector-borne diseases.
Results: The results of the study indicated that the TT method collected significantly more mosquitoes (63%) over the collection period than HLC (37%). Thus, the TTs were observed to be performing relatively better than the HLC (P<0.001). The collectors knew the importance of mosquitoes in transmitting diseases, could identify the main diseases that were locally transmitted within their communities. They appreciated the involvement of the community in the collection as this enhanced community ownership of the programme as well as providing some financial incentives to those directly involved in the collection.

Open Peer Review

Approval Status ? ✓

	1	2
version 2 (revision) 29 May 2019		✓ view
version 1 06 Mar 2019	? view	? view

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Any reports and responses or comments on the article can be found at the end of the article.

Conclusions: The study revealed that use of community volunteers for the collection of mosquitoes for xenomonitoring purposes can be a viable strategy in the monitoring of vector-borne diseases. However, further development of the strategies and assessments of the costs involved will be required to make this a sustainable approach to monitoring vector-borne disease interventions and enhance community ownership of the programmes.

Keywords
Xenomonitoring, community vector collectors, lymphatic filariasis, Ghana.

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Competing interests: No competing interests were disclosed.

Grant information: This work received financial support from the Coalition for Operational Research on Neglected Tropical Diseases (COR-NTD), which is funded at The Task Force for Global Health primarily by the Bill & Melinda Gates Foundation (OPP1053230), by the United Kingdom Department for International Development, and by the United States Agency for International Development through its Neglected Tropical Diseases Program.
The funders had no role in study design, data collection and analysis, decision to publish, or preparation of the manuscript.

How to cite this article: Boakye DA, Frempong KK, Ogoussan KT *et al.* **Implementing a community vector collection strategy for monitoring vector-borne diseases in Ghana [version 2; peer review: 1 approved, 1 approved with reservations]** Gates Open Research 2019, **3**:722 https://doi.org/10.12688/gatesopenres.12933.2

First published: 06 Mar 2019, **3**:722 https://doi.org/10.12688/gatesopenres.12933.1

REVISED **Amendments from Version 1**

In this revised version of the paper, we addressed the comments by the reviewers.

Several minor modifications were done to the text to reflect changes recommended by the reviewers.

One of the reviewers felt that the paper was more oriented towards lymphatic filariasis, than vector-borne diseases in general. We have addressed this. We have also provided new references [Ndebele *et al.*, 2012 and Jamrozik *et al.*, 2015] in relation to the ethical questions around the HLC.

In the methods section, a new figure (Figure 1) has been created, identifying the study locations in Ghana.

The methodology section has been expanded to provide detailed information on the human landing collection and the tent trap.

In the results section, the caption of Figure 2 has also been modified to reflect the "average number of mosquitoes collected per month using the HLC and TT".

Table 2 has also been updated to present the communities collectors came from.

In the discussion, we attempted to give reasons why collectors preferred the HLC and perceived it as collecting more mosquitoes than the TT.

In the acknowledgment, we recognized Harold Nyarko Osei, for the help in producing the map of the study areas.

All the Figures have been renumbered (Figure 1 – Figure 6) due to the study map requested by reviewer 1.

See referee reports

Introduction

Vector-borne diseases remain an important threat to the health of the human population. These are diseases caused by pathogens in humans, and transmitted by mosquitoes and other arthropod organisms. It is estimated that more than one billion people get infected with vector-borne diseases every year, with more than one million deaths[1], and many more suffering from permanent disability and morbidity. Among the most important vector-borne diseases in the world are malaria, onchocerciasis, lymphatic filariasis, dengue, yellow fever, leishmaniasis, chikungunya, West Nile virus, Chagas and more recently emerging infections such as Zika virus[2].

Of all arthropods that transmit diseases, mosquitoes remain the best-known vectors, transmitting diseases such as malaria and lymphatic filariasis, two of the best-known diseases in Africa. Malaria for instance leads to an estimated 210 million cases and 430,000 deaths annually[3], while lymphatic filariasis is responsible for an estimated 36.45 million infections, 19.43 million hydrocele cases and 16.68 million lymphedema cases[4]. While significant efforts and achievements have been made in the control of such diseases, through vector control such as indoor residual sprays (IRS), use of long-lasting insecticide nets (LLINs)[5,6] and treatment of endemic populations[7,8], there is a need to assess the effectiveness of these interventions.

The most direct timely measure of transmission (or the lack thereof) is through the examination of vectors for the presence of infective stages of the parasites responsible for the infection. To this effect, xenomonitoring (the examination of disease vectors for pathogens) serves as an important assessment tool[9,10]. However, in order to document that transmission has been interrupted, it is necessary to screen large numbers of insect vectors to assess their infection and infectivity rates. While pool-screen PCR methods have been developed that can efficiently screen large numbers of vectors[11], the challenge of collecting large numbers of vectors remains. Several factors such as the placement of traps, use of baits, and the type of trap may influence the number of mosquitoes collected. The gold standard of collecting vector mosquitoes is the human landing method (HLC), which is ethically questionable due to the exposure of the collectors to infections[12,13]. As a result, modifications of this approach to involve a double-netted human baiting method have been designed.

A further challenge to the collection of large number of mosquitoes is the use of trained entomologists, which significantly increases the cost of the surveys[14]. To circumvent this challenge, this study opted for the use of community volunteers in the collection of the mosquitoes[14–16]. Operation of the traps by community members themselves has many advantages. First, this would be much less expensive than if trained individuals were required to operate them, and second, this would permit the traps to be more widely distributed and to be operated for longer periods in each community than would be possible if trap operation were confined to traveling teams of trained individuals. Such a community-based operation of the traps has the potential of providing a much more comprehensive estimate of the intensity of transmission, both over time and space, than can be obtained using teams of trained individuals. The usefulness of community vector collection approaches has been demonstrated in Mexico[17], and Togo[18].

This study therefore sought to evaluate a community-based vector collection approach and to assess the performance of the double-netted human baiting method, the tent traps (TT) for the *Anopheles* vectors of lymphatic filariasis in Ghana.

Methods

Selection and sensitization of communities

The study communities were Akonu, Asemko and Aketenkyi in the Nzema East, Ellembele and Ahanta West Districts, respectively (Figure 1). All study communities are located in the Western region of Ghana, and the selected districts are lymphatic filariasis endemic. The Districts are approximately 300 km west of Accra, the capital of Ghana. The chiefs, opinion leaders and communities were informed on the purpose of the study, using the information document developed for the study[19].

Recruitment and training of community vector collectors

To enhance community ownership, the chiefs and elders were asked to identify individuals within the community who will serve as vector collectors. Thus, no strict inclusion or exclusion criteria were applied, so long as the collectors were deemed capable by the community. The vector collectors were either male or female, with formal or informal education. No prior knowledge

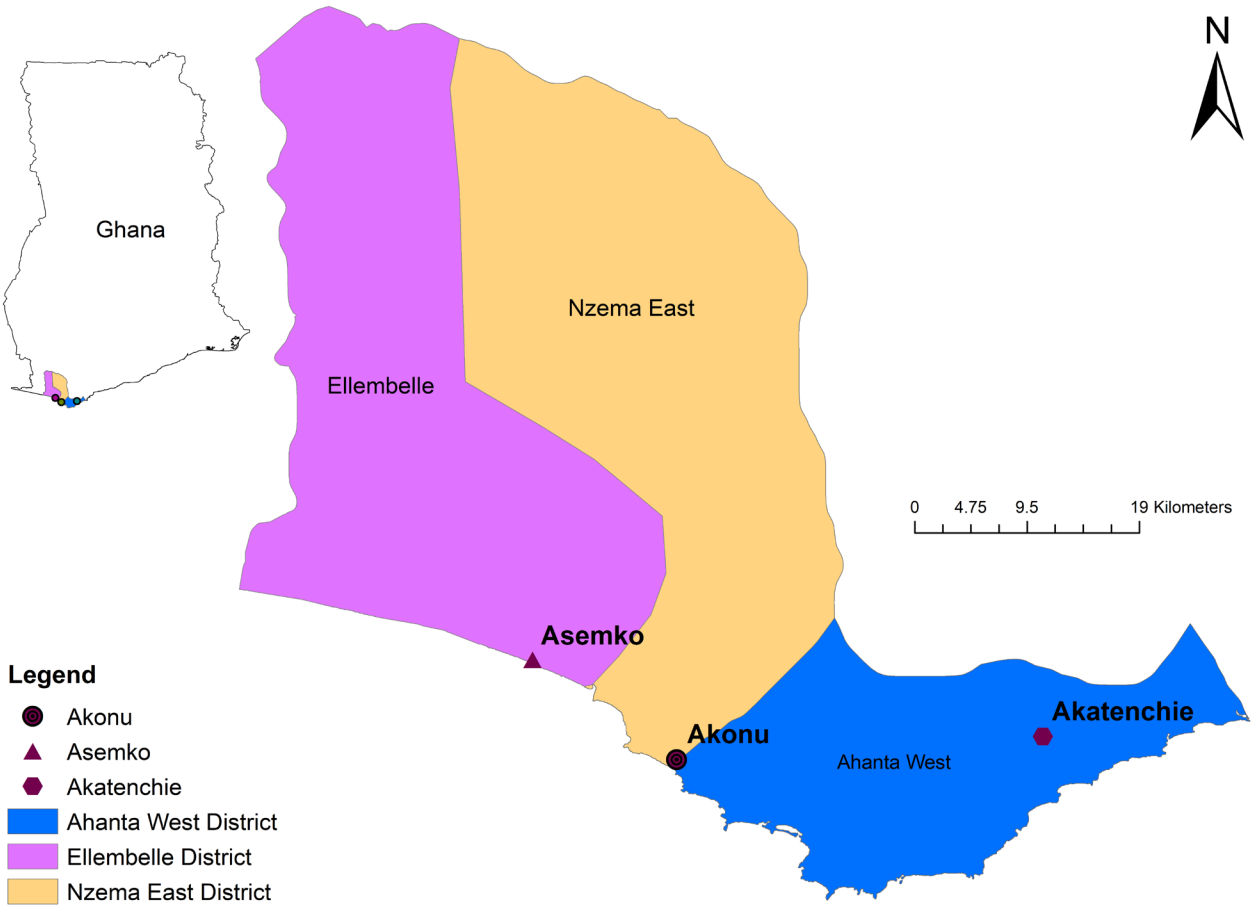

Figure 1. Map of Ghana showing the three study sites and their districts.

in mosquitoes or mosquito collection was required from the collectors. The vector collectors were trained on setting up the TT (Figure 2), and collecting the mosquitoes using aspirators. Two traps were provided for each community, with two vector collectors per trap. They were also trained on mosquito collection using the HLC method.

Mosquito collection

The mosquitoes were collected by the volunteers, without supervision from the study team using human landing catches (HLC) and tent traps (TT). The collectors were advised not to use any repellants, perfumes or fragrances, alcohol or smoking during collection. The collectors were also rotated between collection methods. i.e. HLC collectors for the first collection slept in the TT for the second collection and vice versa. The procedures for the collections were as described below. For the HLC, two collectors were involved. The HLC collections were done outdoors under a shed or a porch. Collections were done from 20:00 till 03:00 hours. At each location, the two collectors sat close to each other and exposed their legs to the knee level to attract the mosquitoes. Each collector captured mosquitoes that landed on their exposed body surface using a test tube with the help of a torch light. The captured mosquito was transferred into a labelled paper cup. Hourly collections were done and different labelled paper cups were used for each hour of collection. After each hour the two collectors switched positions and continued with the collection.

The TT collections were also done outdoors in different locations from the HLC. Two locations of the TT were set, 20–30 meters apart in the same compound. For each, one volunteer slept on a mattress in the inner part of the TT and closed up the inner net. The outer net was raised slightly to enable mosquitoes fly in. This was also done from 20:00 till 03:00 hours. At dawn, the HLC collectors quickly assisted with collection at the TT when they are done. The outer net was released to prevent trapped mosquitoes from escaping. The HLC collectors entered through the outer net and collected trapped mosquitoes using aspirators into labelled paper cups. On days when the collection was prevented by rains, both HLC and TT collections were cancelled, and the collections rescheduled.

Collections were done from June to December 2017. Two TT and two HLC collections were carried out every month

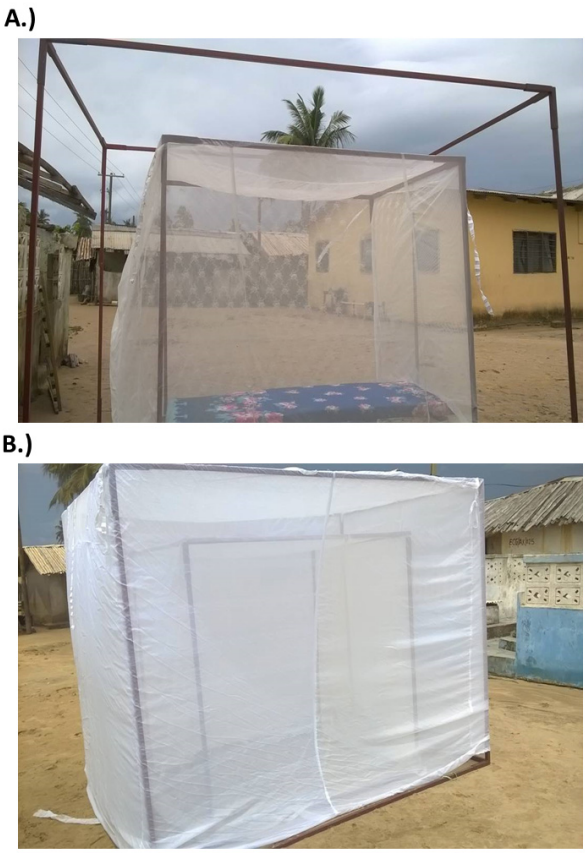

Figure 2. Setting the tent traps. (**A**) The inner sleeping tent. (**B**) The outer tent.

at a time suitable to the collectors, but at two weeks interval. On each collection day, the mosquitoes collected for each method were stored in a 50-ml falcon tube, provided to the collectors and labelled with all the information including the collection method and month of collection.

Once every two months, a member of the study team visited the communities to collect the mosquitoes to the laboratory at the Noguchi Memorial Institute for Medical Research, Accra where they were identified, counted and stored.

Evaluation of community xenomonitoring study: perspectives of collectors

The study also assessed the views of the community vector collectors, using a simple questionnaire designed for this purpose[19]. This was done to enhance future operational activities.

Ethical considerations

The study was approved (CPN 062/16-17) by the NMIMR IRB with Federal Wide Assurance Registration (FWA 00001824). Following community sensitization, verbal approval was obtained from the chiefs and elders of the communities. Written consent was also obtained from the vector collectors. The collectors were given prophylaxis as part of malaria prevention measures.

Statistical analysis

The number of mosquitoes collected was represented through graphs drawn in Microsoft Excel. The difference between the number of mosquitoes collected using the TT and HLC methods was assessed using the chi square test in IBM SPSS Statistics 20.0. Statistical significance was assessed at a p value ≤0.05.

Results

Results of mosquito collection

A total of 3363 mosquitoes (Table 1) were collected from June to December 2017 comprising; 1581 (47%) *An. gambiae s.l.* 1266 (37.6%) *Culex* spp., 6 (0.2%) *Aedes* sp. and 510 (15. 2%) *Mansonia* sp. *An. gambiae s.l.* was the only *Anopheles* species identified. A significantly ($P<0.001$) higher percentage of mosquitoes were collected using the TT (63%) compared to the HLC (37%). However, the number of Anopheles collected by the TT and the HLC was not statistically significant ($P = 0.213$). The average monthly collections also revealed higher collections by the TT compared to the HCL, with significant differences observed during the dry season months of August, September, November and December (Figure 3). Generally, the variation (range) in mosquito numbers collected each month was relatively smaller for the HLC (apart from June and October) compared with the TT (Figure 3).

The general trend in the total number of mosquitoes collected reflects the influence of seasons on the behavior and the population of mosquito species, with the Anophelines and Culicines revealing opposing trends. The main collection peak for *An. gambiae* was from June to August, which coincides with the major rains and the beginning of the dry season. On the other hand, the main peak for Culex was during the minor rains and dry season months of October to December (Figure 4). A similar trend was observed in mosquitoes collected using the TT and HLC (Figure 5).

Among the three communities where collection was done, the highest number of mosquitoes was collected from Aketenkyi and the least from Akonu (Figure 6). In Akonu though the number of mosquitoes collected from TT was generally higher than those from the HLC between June and September, the reverse was the case between October and December. Asemko showed higher numbers in TT than HLC in almost all the months apart from June and July, whiles Aketenkyi showed similar pattern except from June where HLC numbers were higher.

Evaluation of community xenomonitoring study: perspectives of collectors

A summary of the demographic information of mosquito collectors and responses to questionnaire is shown in Table 2. There were 12 collectors involved in the study, three of whom were females. The age of the collectors ranged from 16 to 58 years with a median age of 34 years. Seven of the collectors had received primary/JHS education, 3 secondary education and the remaining

Table 1. Mosquitoes collected as part of the trapping study in Ghana.

			Akonu				Asemko				Aketenkyi				
Method	**Season**	**Month**	***An. gambiae***	***Culex***	***Aedes***	***Mansonia***	***An. gambiae***	***Culex***	***Aedes***	***Mansonia***	***An. gambiae***	***Culex***	***Aedes***	***Mansonia***	**Total**
Tent trap	**Rain**	June	49	1	1	0	9	31	0	0	65	0	0	0	**156**
		July	46	6	0	0	57	38	0	0	138	8	3	0	**296**
		August	47	22	0	0	24	79	0	27	64	0	0	35	**298**
		September	17	39	0	0	15	63	0	1	48	14	0	25	**222**
		Sub-total	***159***	***68***	***1***	***0***	***105***	***211***	***0***	***28***	***315***	***22***	***3***	***60***	***972***
	Dry	October	30	84	0	0	35	59	0	0	6	0	0	163	**377**
		November	7	113	0	0	81	75	0	0	19	86	0	0	**381**
		December	18	104	0	0	88	61	0	0	65	0	0	55	**391**
		Sub-total	***55***	***301***	***0***	***0***	***204***	***195***	***0***	***0***	***90***	***86***	***0***	***218***	***1149***
Human landing collection	**Rain**	June	25	5	0	0	12	18	0	0	78	6	0	0	**144**
		July	33	1	0	0	83	42	0	0	105	4	0	0	**268**
		August	28	0	0	0	3	39	0	19	38	1	0	10	**138**
		September	0	33	1	0	7	32	1	4	35	3	0	8	**124**
		Sub-total	***86***	***39***	***1***	***0***	***105***	***131***	***1***	***23***	***256***	***14***	***0***	***18***	***674***
	Dry	October	55	66	0	0	20	18	0	0	2	0	0	88	**249**
		November	17	58	0	0	15	17	0	0	18	0	0	50	**175**
		December	31	14	0	0	21	26	0	0	27	0	0	25	**144**
		Sub-total	***103***	***138***	***0***	***0***	***56***	***61***	***0***	***0***	***47***	***0***	***0***	***163***	***568***
		Total	**403**	**546**	**2**	**0**	**470**	**598**	**1**	**51**	**708**	**122**	**3**	**459**	**3363**

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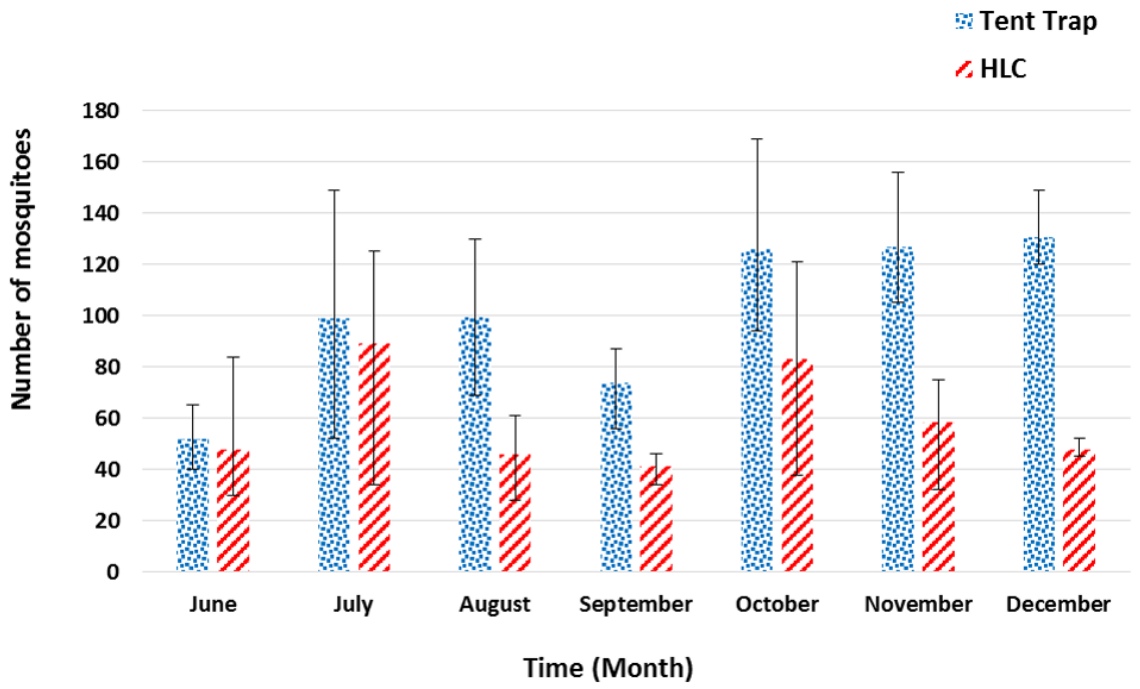

Figure 3. Average number of mosquitoes collected per month using HLC and Tent Trap. Error bars represent the minimum and maximum number of mosquitoes collected. HLC, human landing collection.

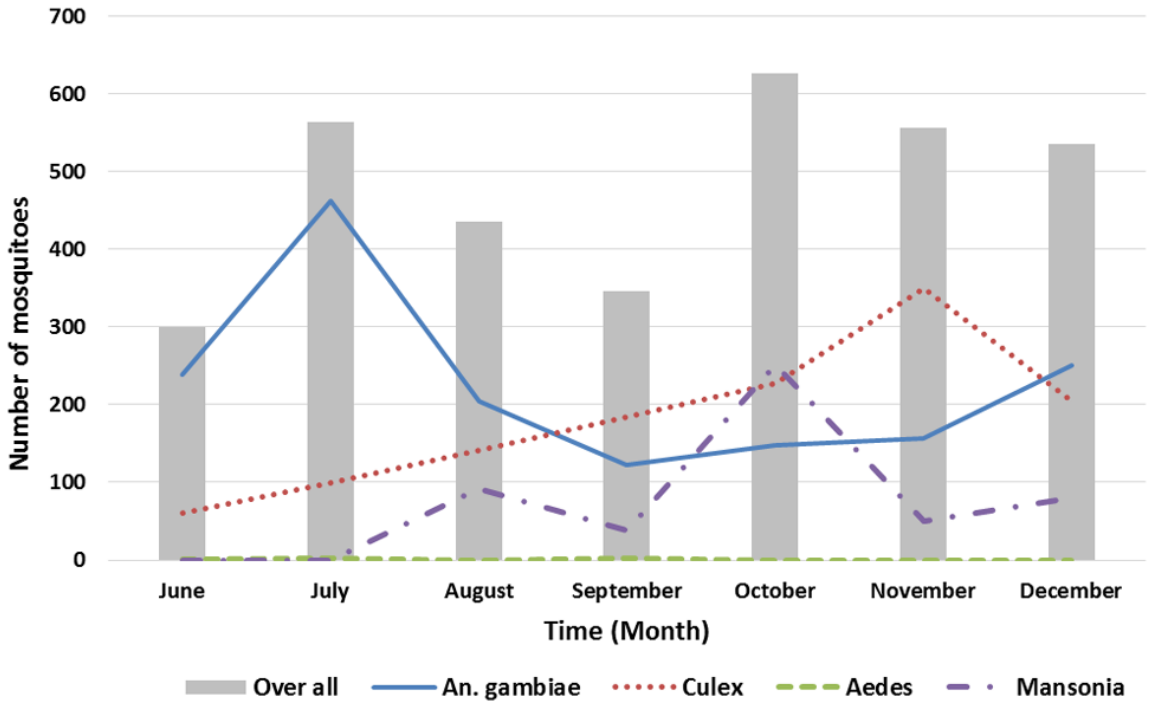

Figure 4. Total number of mosquitoes and the species abundance per month irrespective of collection method.

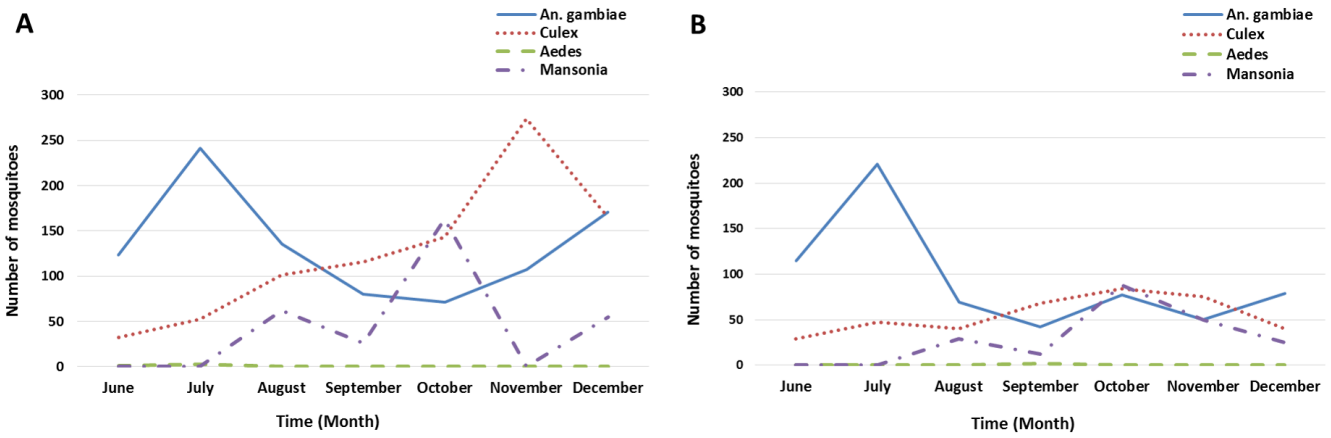

Figure 5. Trends showing species composition of mosquitoes collected using (**A**) tent trap and (**B**) human landing collection.

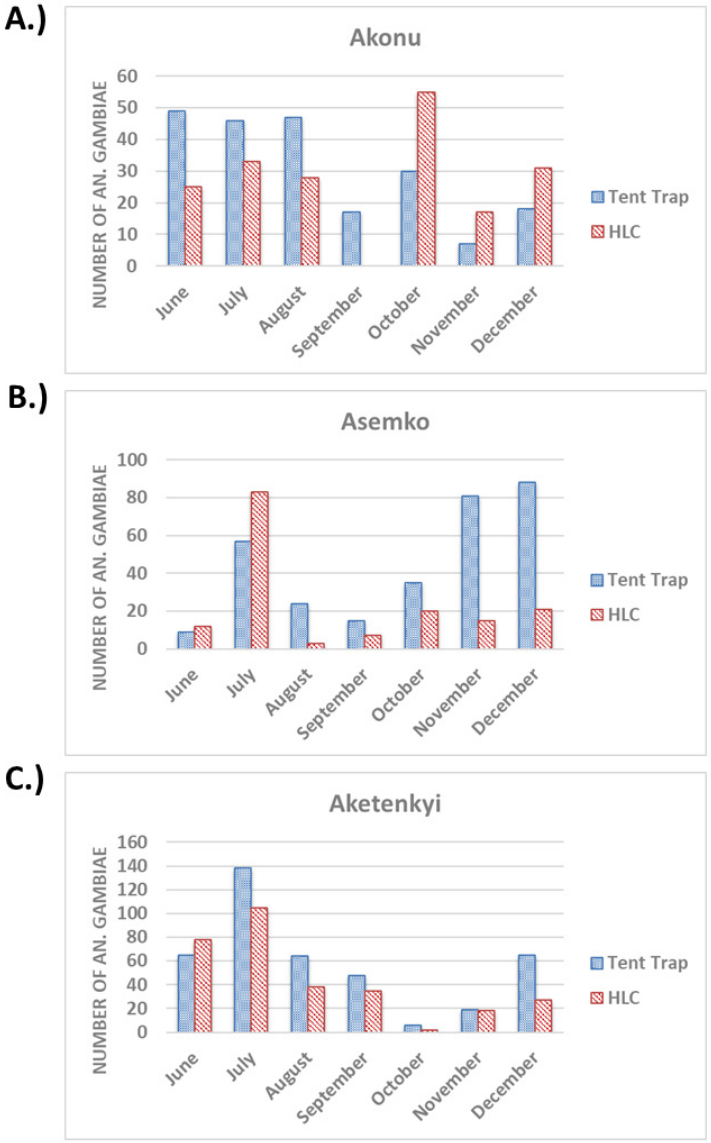

Figure 6. Frequency distribution of *Anopheles gambiae* s.l. mosquitoes collected from (**A**) Akonu (**B**) Asemko and (**C**) Aketenkyi, using HLC and Tent Trap. HLC, human landing collection.

Table 2. Details of the demographic information of mosquito collectors and their responses to questionnaire administered.

						Individual responses to questionnaire						
#	**Sex**	**Age**	**Educational level**	**Occupation**	**Community**	**Importance of mosquito study**	**Mosquito-borne disease at study area**	**Importance of community involvement**	**Preference of collection method**	**Willingness to continue mosquito collection**	**Person to bear cost of mosquito collection**	**To volunteer without pay**
1	M	32	Secondary	Teacher	Aketenchie	Transmit elephantiasis & malaria	Malaria	Save time and cost	HLC	Yes	Research team	Yes
2	M	58	Primary/JHS	Farmer	Aketenchie	Transmit elephantiasis & malaria	Malaria	Save time and cost	HLC	Yes	GHS	Yes
3	M	38	Post-Secondary	Not employed	Aketenchie	Investigation of disease transmission	Malaria & elephantiasis	Motivation to be part of fighting disease	Tent trap	Yes	Government	Yes
4	M	40	Primary/JHS	Farmer	Aketenchie	Transmit elephantiasis & malaria	Malaria	To have more knowledge on mosquitoes	HLC	Yes	Community can pay	Yes
5	M	16	Primary/JHS	Not employed	Akonu	Transmit diseases	Malaria & cholera	Motivation to be part of fighting disease	HLC	Yes	GHS	Yes
6	F	25	Primary/JHS	Seamstress	Akonu	Transmit elephantiasis & malaria	Malaria & elephantiasis	Save time and cost	HLC	Yes	Government	Yes
7	F	21	Primary/JHS	Not employed	Akonu	Transmit elephantiasis & malaria	Malaria & elephantiasis	Save time and cost	HLC	Yes	Government	Yes
8	M	37	Primary/JHS	Farmer	Akonu	Transmit elephantiasis & malaria	Malaria & elephantiasis	Motivation to be part of fighting disease	HLC	Yes	Government	Yes
9	M	17	Secondary	Student	Asemko	Transmit malaria and TB	Malaria	To have more knowledge on mosquitoes	HLC	Yes	Research team	No
10	F	36	Primary/JHS	Seamstress	Asemko	Transmit elephantiasis & malaria	Malaria	Motivation to be part of fighting disease	Tent trap	Yes	Government	Yes
11	M	42	Post-Secondary	Farmer	Asemko	Transmit elephantiasis & malaria	Malaria & elephantiasis	Helps to accept research findings and get some income	HLC	Yes	GHS	Yes
12	M	19	Secondary	Student	Asemko	Transmit malaria and TB	Malaria & TB	Save time and cost	HLC	Yes	Government	No

F, female; M, male; JHS, junior high school; HLC, human landing catches; TB, tuberculosis; GHS, Ghana Health Services.

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2 post-secondary education. Four of the collectors were farmers, 3 unemployed, 2 seamstresses, 2 students and 1 teacher.

The assessment showed that the mosquito collectors had a fairly good idea about the importance of mosquitoes in disease transmission especially malaria and lymphatic filariasis. This is exemplified by some of the responses below:

"They bite and bring diseases like malaria & elephantiasis, kill pregnant women and children cause us to spend a lot"

"To investigate the disease they transmit"

"They suck blood and contain parasites in them to give diseases"

"They are strange animals that bite and cause diseases such as high fever, rashes & elephantiasis"

Generally, the volunteers were aware of the main diseases transmitted by mosquitoes in their community. They all identified malaria as a disease transmitted by mosquitoes. In addition, others mentioned elephantiasis, with two individuals mentioning cholera and tuberculosis. On the importance/benefits of using community members for mosquito collection, the main views expressed were with the speed of collection, the willingness of the community and individuals to be involved in the study and feel a part of the disease elimination process, and the opportunity costs to the funders and collectors. These are reflected in the views below:

"When we are involved, we believe the results, it also helps to get some income and we feel part of eliminating the disease"

"We will help the GHS reduce cost of travelling to community and also get some income"

"We are able to collect fast. The team from Accra sometimes come late and we cannot do much…. The people also know us so they allow us into their houses"

"So, we can help ourselves to drive away mosquitoes to prevent diseases"

"Saves time and cost, no tension in sample collection, shows commitment level of community"

When asked which of the mosquito collection methods they preferred, 10/12 volunteers stated that they preferred the HLC. The reasons given for their preference were; the number of mosquitoes collected using the HLC, and the speed and ease of collection. The preference to the TT was due to the absence of mosquito bites. All the volunteers responded in the affirmative to the question of using the community to collect mosquitoes for the GHS as part of disease monitoring activities. However, 11/12 volunteers indicated that payment for the collection must come from the government or the Ghana Health Service. When asked whether they will consider collecting mosquitoes for the community without payment, 10/12 volunteers said "yes". The main reasons given were to help protect the community, themselves and their families from infection. However, two of the collectors said they will not collect the mosquitoes without payment. Below are some of the views expressed:

"Disease is destroying our community & I need to help; I have been doing volunteer work already for NGOs"

"To help the community eliminate the disease and protect my children"

"I could get the disease and the cost will be more to me, also I will receive blessings"

"It is volunteer work and that is my contribution to the community"

"Already involved in a lot of volunteer work without pay"

The volunteers mentioned the need for more logistics such as consumables and better torch lights, malaria prevention using anti-malaria prophylaxis, improved methods for mosquito collection and enhance cohesion between the collection teams, as some of the ways of improving the mosquito collection and the work of the collectors. Complete answers to the questionnaire are available on OSF[19].

Discussion

The surveillance of vector-borne diseases requires an assessment of the infection in the human population, whiles at the same time undertaking entomological surveillance to detect the infection in the vectors of the disease. Such is the plan for the post-intervention surveillance phase of lymphatic filariasis[20]. However, conducting entomological surveys especially in the context of elimination activities is considered expensive due to the limited resources[21] and the high number of samples that may be required to assess transmission of the disease after interventions as well as the cost involved in the collection and processing of the mosquitoes[14,16]. Thus, there is the need for more cost-effective strategies for entomological assessments of infections within vectors of diseases.

In this study, the TTs were observed to be performing relatively better than the HLC ($P<0.001$). This study showed that the TTs collected higher number of mosquitoes compared to HLC and may be a better tool for the collection of a larger number of mosquitoes for xenomonitoring surveys, even though a majority of the volunteers thought the HLC was better. The preference of collectors for the HLC due to the number of mosquitoes collected could be linked to the high human-mosquito contact, and thus the perception of high mosquito abundance. Nonetheless in a largely unsupervised setting, the TT offers the advantage of collecting similar or higher numbers of mosquitoes, while helping reduce the likelihood of infection presented by the HLC.

Mosquitoes are often observed to be more abundant during the rainy season compared to the dry season[22]. This is due to the presence of more breeding sites created during the rainy season. In this study this was not the case from the collections as there was no significant differences in the numbers collected in both

seasons. On the contrary, the data showed more mosquitoes collected during the dry season. This observation may be attributed to disruptions during rainy days. In fact, the collectors complained about their inability to set up the traps for long periods during the rainy season.

The seasonal changes observed in the distribution pattern of the mosquito species can be explained by their habitat preferences[23]. The numbers of *An. gambiae* during the rainy season was higher than the other species but low during the dry season. This is not surprising because *An. gambiae* prefer breeding in shallow and fresh water which are often abundant during the raining season[24,25]. *Culex* spp. on the other hand was higher during the dry season when fresh water is less abundant (Figure 6). This is could be due to its ability to breed in polluted breeding sources[26,27], which are prevalent in Akonu and Asemko.

The assessment of the volunteers' perspectives served to assess the level of understanding and importance of the task given to them as well as the feasibility of using such approach for future national programme implementation activities. The assessment revealed that generally, the collectors knew the importance of mosquitoes in transmitting diseases and were able to identify the main diseases that were locally transmitted within their communities. They appreciated the involvement of the community in the activities as this enhanced community ownership of the programme as well as providing some financial incentives to those directly involved in the collection (Table 2). For example, the community selection of ivermectin drug distributors as part of the Community Directed Treatment with ivermectin, improved access to the drugs and enhanced the community ownership of the activities[28,29]. The use of the community volunteers also provides opportunity costs to both the programme and the communities involved. To the national programmes, there are fewer expenses on the use of trained entomologists for vector collection and subsequent reduction in transportation and logistics costs. To the community volunteers, this provides some opportunities for them to earn some allowances, even though the work is entirely voluntary. However, the motivation of community volunteers is an important factor that determines the success of programme activities[30,31]. Recent use of community volunteers for large-scale monitoring activities have suggested that it is a useful strategy for programme implementation[18,32]. However, this needs to be tailored to country-specific situations.

In conclusion, the study revealed that use of community volunteers for the collection of mosquitoes as part of xenomonitoring purposes, can be an important strategy in the undertaking of monitoring of vector-borne diseases. However, further development of the strategies and assessments of the costs involved will be required in order to make this a public health approach to monitoring vector-borne disease interventions, and enhance community ownership and sustainability of the programmes. Though the use of community members for mosquito collection is promising, there is the need for further community education and training of community volunteers for xenomonitoring purposes.

Data availability

Underlying data

Open Science Framework. A community vector collection strategy for monitoring vector-borne diseases in Ghana. https://doi.org/10.17605/OSF.IO/C6BHP[19].

Questionnaire data.xlsx contains the complete responses to each question of the questionnaire.

Extended data

Open Science Framework. A community vector collection strategy for monitoring vector-borne diseases in Ghana. https://doi.org/10.17605/OSF.IO/C6BHP[19].

The project contains the following extended data:

- Collectors' questionnaire.docx (the questionnaire administered to the mosquito collectors).
- Information document for mosquito collection using the tent trap.docx.

Grant information

This work received financial support from the Coalition for Operational Research on Neglected Tropical Diseases (COR-NTD), which is funded at The Task Force for Global Health primarily by the Bill & Melinda Gates Foundation (OPP1053230), by the United Kingdom Department for International Development, and by the United States Agency for International Development through its Neglected Tropical Diseases Program.

The funders had no role in study design, data collection and analysis, decision to publish, or preparation of the manuscript.

Acknowledgements

We are grateful to the study communities and volunteers for their role in the study. We are also grateful to Harold Nyarko Osei for the map of the study communities.

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Open Peer Review

Current Peer Review Status: ? ✓

Version 2

Reviewer Report 10 July 2019

https://doi.org/10.21956/gatesopenres.14144.r27266

✓ **Seth R. Irish**
Entomology Branch, Centers for Disease Control and Prevention, Atlanta, GA, USA

There were two comments of mine that were not addressed. In the second paragraph of the results, Culex should be italicized. Also, in table 1, the second to last row is the subtotal for HLC in the dry season, and "Subtotal" should be bolded to make it consistent with the rest of the table.

Competing Interests: No competing interests were disclosed.

Reviewer Expertise: Medical entomology

I confirm that I have read this submission and believe that I have an appropriate level of expertise to confirm that it is of an acceptable scientific standard.

Version 1

Reviewer Report 29 April 2019

https://doi.org/10.21956/gatesopenres.14035.r26975

? **Seth R. Irish**
Entomology Branch, Centers for Disease Control and Prevention, Atlanta, GA, USA

This paper describes a community-based mosquito collection method tested in Ghana. In addition to evaluating the entomological collections using the different methods tested, the response of

the community is recorded, an important factor in community-based surveillance.

Introduction:

- The authors speak about vector-borne diseases in general, but it seems they are thinking mostly about lymphatic filariasis. For example, the statement about a prevalence of the pathogen which "has reached a relatively low threshold, below which the parasite population cannot be sustained", is hard to imagine for a disease such as malaria.

- The authors state that the human landing catch is ethically questionable and I think there are lots of arguments for this position, however, to support this statement, they cite a study that shows significantly less malaria detected in HLC collectors than in the general population. I would think there would be better references to cite unless I have missed the intention of the authors.

Methods:

- The mosquito collection methods are inadequately described, making replication of this work impossible. What were the starting and ending times of the collections? Were they done indoors or outdoors? Were the HLC collections made in the same locations as the tent trap collections? Did the same collectors conduct both HLC and TT?

Results:

- In the "Results of mosquito collection" section, second sentence: "proportion" is mentioned, but then percentages are given. Please be consistent between the text and the values provided.

- In the following paragraph, please italicize Culex.

- Table 1: I think the last "subtotal" column should be bolded. Also, the numbers collected by the two methods don't look sufficient for the collection of large numbers needed for xenomonitoring. Finally, was *Anopheles gambiae* the only Anopheles species collected? If not, please note that the other Anopheles collected are not presented.

- Figure 3: Please include in the figure legend what collection method was used.

- When people stated their preference for HLC, one of the reasons was "the number of mosquitoes collected using the HLC" - can you explain this further? Did people prefer collecting fewer mosquitoes?

Discussion:

- The authors are discussing mosquitoes in general, but disease transmission depends on the species collected. For me, one of the big questions I would have for this paper is whether the two methods were significantly different in the number of *An. gambiae* collected.

- The rain was discussed, but it is not clear how the rain delays were dealt with. Was trapping cancelled every time there was rain, or was there a certain amount of delay that was acceptable?

- The authors make a statement about the ability of all Culex species to breed in polluted

breeding sources. I assume they are really talking about *Culex quinquefasciatus*, and if this is the case, they should state it clearly.

Is the work clearly and accurately presented and does it cite the current literature?
Yes

Is the study design appropriate and is the work technically sound?
Yes

Are sufficient details of methods and analysis provided to allow replication by others?
No

If applicable, is the statistical analysis and its interpretation appropriate?
Partly

Are all the source data underlying the results available to ensure full reproducibility?
Yes

Are the conclusions drawn adequately supported by the results?
Yes

Competing Interests: No competing interests were disclosed.

Reviewer Expertise: Medical entomology

I confirm that I have read this submission and believe that I have an appropriate level of expertise to confirm that it is of an acceptable scientific standard, however I have significant reservations, as outlined above.

Author Response 21 May 2019

Dziedzom K. de Souza, Noguchi Memorial Institute for Medical Research, College of Health Sciences, University of Ghana, Accra, Ghana

We thank the reviewer for the valuable comments to the paper. We have addressed the comments in the point-by-point response below.

.....

This paper describes a community-based mosquito collection method tested in Ghana. In addition to evaluating the entomological collections using the different methods tested, the response of the community is recorded, an important factor in community-based surveillance.

Introduction:
Comment: The authors speak about vector-borne diseases in general, but it seems they are

thinking mostly about lymphatic filariasis. For example, the statement about a prevalence of the pathogen which "has reached a relatively low threshold, below which the parasite population cannot be sustained", is hard to imagine for a disease such as malaria.
Response: We agree with the reviewer on this point. To capture malaria and the rest of vector-borne diseases, the sentence has been modified to read “However, in order to document that transmission has been interrupted, it is necessary to screen large numbers of insect vectors to assess their infection and infectivity rates.” See lines 73-74.

Comment: The authors state that the human landing catch is ethically questionable and I think there are lots of arguments for this position, however, to support this statement, they cite a study that shows significantly less malaria detected in HLC collectors than in the general population. I would think there would be better references to cite unless I have missed the intention of the authors.
Response: The ethical questions relating to the HLC specifically in this study, relate to its use by community members without supervision, and the associated risks of infection. We do agree that the references supporting the statement “human landing catch is ethically questionable” does not fit this context. They have been replaced by references that support the context in which the statement was used. See line 79.

Methods:
Comment: The mosquito collection methods are inadequately described, making replication of this work impossible. What were the starting and ending times of the collections? Were they done indoors or outdoors? Were the HLC collections made in the same locations as the tent trap collections? Did the same collectors conduct both HLC and TT?
Response: We have provided further description of the HLC and TT collection methods. See lines 117-138.

Results:
Comment: In the "Results of mosquito collection" section, second sentence: "proportion" is mentioned, but then percentages are given. Please be consistent between the text and the values provided.
Response: This has been corrected to “percentage”. See line 169.

Comment: In the following paragraph, please italicize Culex.
Response: This has been addressed accordingly.

Comment: Table 1: I think the last "subtotal" column should be bolded. Also, the numbers collected by the two methods don't look sufficient for the collection of large numbers needed for xenomonitoring. Finally, was *Anopheles gambiae* the only Anopheles species collected? If not, please note that the other Anopheles collected are not presented.
Response: The last “subtotal” column has been bolded. This study looks at whether communities can implement vector collection properly by themselves, and without supervision. Given that only two collectors were used, at each collection time for each method, we consider the numbers collected large enough. Thus, if the number of collectors and frequency of collection was increased, much more mosquitoes could be collected for xenomonitoring purposes, based on the same costs that would otherwise be used in paying

experienced entomologists.
*An. gambiae s.l.*was the only *Anopheles* species identified in this study. This has been stated clearly in the results. See line 168.

Comment: Figure 3: Please include in the figure legend what collection method was used.
Response: Figure 3: This figure was to show the number of mosquito species collected out of the total per month. Including the collection method may be a bit challenging using the bar chart and line graph combination we have in the figure. As mentioned, this figure was to show the number of mosquito species collected out of the total per month. The Figure title has been changed to reflect this. The breakdown of captures by collection method is shown in Figure 5. Additionally, Figures 2 and 5 all give information about the collection method. Figure 3 has now been renumbered as Figure 4 in the new version.

Comment: When people stated their preference for HLC, one of the reasons was "the number of mosquitoes collected using the HLC" - can you explain this further? Did people prefer collecting fewer mosquitoes?
Response: When people stated their preference for HLC, one of the reasons was "the number of mosquitoes collected using the HLC". This could be linked to the high human-mosquito contact, and thus the perception of high mosquito abundance. However, the results show that the TT collected more mosquitoes than the HLC. We have addressed this in the discussion.

Discussion:
Comment: The authors are discussing mosquitoes in general, but disease transmission depends on the species collected. For me, one of the big questions I would have for this paper is whether the two methods were significantly different in the number of *An. gambiae* collected.
Response: The number of *An. gambiae* collected from TT was higher than HLC but this was not statistically significant (P= 0.213). However, this study shows that the TT can be an alternative to the HLC, while helping reduce the likelihood of infection presented using the HLC. We have added this point to the text.

Comment: The rain was discussed, but it is not clear how the rain delays were dealt with. Was trapping cancelled every time there was rain, or was there a certain amount of delay that was acceptable?
Response: Trapping for both HLC and TT were cancelled anytime there was rain. These were not recorded too many times during the study. Therefore numbers obtained from the two methods can be compared. This has been addressed in the methods.

Comment: The authors make a statement about the ability of all Culex species to breed in polluted breeding sources. I assume they are really talking about *Culex quinquefasciatus*, and if this is the case, they should state it clearly.
Response: *Culex* was not identified to the species level. Nonetheless, previous studies indicate *Culex quinquefasciatus*as the major species known to be found in Ghana. We have edited the sentence to reflect that the presence of Culex “could be” due to polluted breeding sites.

Competing Interests: No competing interests were disclosed.

Reviewer Report 09 April 2019

https://doi.org/10.21956/gatesopenres.14035.r26974

?

Chadwick Sikaala
[1] Southern Africa Malaria Elimination Eight Secretariat, Windhoek, Namibia
[2] National Malaria Elimination Center, Lusaka, Zambia

Overall the article addresses a critical area of entomological surveillance within the context of cost effectiveness and community ownership. The authors did cite the relevant articles and are scientifically sound in this regard.

Below are a few critical areas that the authors need to address prior to the indexing of the article:

1. Introduction:
   - While the introduction is well structured and the justification for the study stipulated, in the third paragraph the authors might need to state that HLC is the gold standard method as opposed to common method. Within the same chapter, the sentence mentioning the challenge of collecting large numbers of mosquitoes, may need to be rephrased as there is evidence to the fact that different methods collect large numbers of mosquitoes depending on several factors.

2. Methodology:
   - The authors could consider putting a map of the areas of study for the readers to appreciate the distribution pattern of mosquito collections as stated in the result section. While most readers are familiar with HLC, the authors may consider elaborating more on how the 2 methods function (HLC & TT) for the sake of readers who may not be familiar with the methods. Under the mosquito collection part, explaining the time of collections is vital. This also can explain the differences in the number of mosquitoes collected per method. Was it done between 6pm to 6am? While the TT appears to be placed outdoors, the authors need to show whether HLC was conducted outdoors or indoors - as this may have repercussions on data interpretation. The authors may need to include this. While in the discussion section it is mentioned that volunteers participated in prophylaxis, this might need to be mentioned under the ethical considerations section as well.

3. Results:
   - On Figure 2, the authors suggest apart from June and October, HLC collected less mosquitoes; this does not appear to be the case on the graph. The title of the figure needs

to include TT since it is not only HLC represented. This includes graph/Figure 5. With Figure 5 the authors may consider to elaborate as it appears only Anophelines are represented.

4. Discussion:
   - The authors could have considered indicating the communities from which the different volunteers in terms of occupation came from. While not significant, it would have highlighted which occupation was collecting more mosquitoes. A good example is that these results though not statistically significant, students are likely to participate if an incentive is offered.

Is the work clearly and accurately presented and does it cite the current literature?
Yes

Is the study design appropriate and is the work technically sound?
Partly

Are sufficient details of methods and analysis provided to allow replication by others?
Partly

If applicable, is the statistical analysis and its interpretation appropriate?
Partly

Are all the source data underlying the results available to ensure full reproducibility?
Yes

Are the conclusions drawn adequately supported by the results?
Partly

Competing Interests: No competing interests were disclosed.

Reviewer Expertise: Vectors of medical importance

I confirm that I have read this submission and believe that I have an appropriate level of expertise to confirm that it is of an acceptable scientific standard, however I have significant reservations, as outlined above.

Author Response 21 May 2019

Dziedzom K. de Souza, Noguchi Memorial Institute for Medical Research, College of Health Sciences, University of Ghana, Accra, Ghana

We thank the reviewer for the valuable comments to the paper, and have addressed them in the point-by-point response below.

.....

Overall the article addresses a critical area of entomological surveillance within the context of cost effectiveness and community ownership. The authors did cite the relevant articles and are scientifically sound in this regard.

Below are a few critical areas that the authors need to address prior to the indexing of the article:

1. Introduction:
Comment: While the introduction is well structured and the justification for the study stipulated, in the third paragraph the authors might need to state that HLC is the gold standard method as opposed to common method. Within the same chapter, the sentence mentioning the challenge of collecting large numbers of mosquitoes, may need to be rephrased as there is evidence to the fact that different methods collect large numbers of mosquitoes depending on several factors.
Response: The statement has been modified to read "The gold standard of collecting vector mosquitoes is the human landing method (HLC), which is ethically questionable due to the exposure of the collectors to infections". See lines 78-80.

2. Methodology:
Comment: The authors could consider putting a map of the areas of study for the readers to appreciate the distribution pattern of mosquito collections as stated in the result section. While most readers are familiar with HLC, the authors may consider elaborating more on how the 2 methods function (HLC & TT) for the sake of readers who may not be familiar with the methods. Under the mosquito collection part, explaining the time of collections is vital. This also can explain the differences in the number of mosquitoes collected per method. Was it done between 6pm to 6am? While the TT appears to be placed outdoors, the authors need to show whether HLC was conducted outdoors or indoors - as this may have repercussions on data interpretation. The authors may need to include this. While in the discussion section it is mentioned that volunteers participated in prophylaxis, this might need to be mentioned under the ethical considerations section as well.
Response: A map showing the study sites has been drawn and added to the new version of the manuscript as Figure 1. The other figures have been renumbered appropriately.

Additional information on HLC and TT have been added under Mosquito collection in lines 117-138 of new version as follows:
The mosquitoes were collected by the volunteers, without supervision from the study team using human landing catches (HLC) and tent traps (TT). Collectors were advised not to use any repellants, perfumes or fragrances, alcohol or smoking during collection. The procedures for the collections were as follows:
For the HLC, two collectors were involved. The HLC collections were done outdoors under a shed or a porch. Collections were done from 20:00 till 03:00 hours. At each location, the two collectors sat close to each other and exposed their legs to the knee level to attract the mosquitoes. Each collector captured mosquitoes that landed on their exposed body surface using a test tube with the help of a torch light. The captured mosquito was transferred into a labelled paper cup. Hourly collections were done and different labelled paper cups were used for each hour of collection. After each hour the two collectors switched positions and

continued with the collection.

The TT collections were also done outdoors in different locations from the HLC. Two locations of the TT were set, 20-30 meters apart in the same compound. For each, one volunteer slept on a mattress in the inner part of the TT and closed up the inner net. The outer net was raised slightly to enable mosquitoes fly in. This was also done from 20:00 till 03:00 hours. At dawn, the HLC collectors quickly assisted with collection at the TT when they are done. The outer net was released to prevent trapped mosquitoes from escaping. The HLC collectors entered through the outer net and collected trapped mosquitoes using aspirators into labelled paper cups. On days when the collection was prevented by rains, both HLC and TT collections were cancelled, and the collections rescheduled.

3. Results:
Comment: On Figure 2, the authors suggest apart from June and October, HLC collected less mosquitoes; this does not appear to be the case on the graph. The title of the figure needs to include TT since it is not only HLC represented. This includes graph/Figure 5. With Figure 5 the authors may consider to elaborate as it appears only Anophelines are represented.
Response: The statement “apart from June and October” was in reference to the range (Minimum – Maximum) based on the error bars and not the number of mosquitoes collected. We observed that this may be a bit confusing to our readers. This has therefore been modified to read “Generally, the variation (range) in mosquito numbers collected each month was relatively smaller for the HLC compared with the TT (Figure 2)”. The statement “apart from June and October” has been deleted.
The caption of Figure 2 which is now Figure 3 has been modified to read “**Figure 3. Average number of mosquitoes collected per month using HLC and TT.** Error bars represent the minimum and maximum number of mosquitoes collected. HLC, human landing collection and TT, tent trap”. See lines of new version.
The comment on Figure 5 (now Figure 6) has been addressed as requested.

4. Discussion:
Comment: The authors could have considered indicating the communities from which the different volunteers in terms of occupation came from. While not significant, it would have highlighted which occupation was collecting more mosquitoes. A good example is that these results though not statistically significant, students are likely to participate if an incentive is offered.
Response: The communities of the volunteers/collectors have been inserted in Table 2. Unfortunately, it is difficult to state if a certain occupation collected more mosquitoes. The study team had no influence on the pairing of collectors. Also, the collectors were rotated between the collection methods. For each community, the mosquitoes were pooled together irrespective of who collected them.

Competing Interests: No competing interests were disclosed.