Assessment of Novel Lehmann's Funnel Entry Trap Prototypes Performance to Control Malaria Mosquito Populations

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Abstract

Background

There is a global consensus that new intervention tools are needed to cross the last miles in malaria elimination/eradication. In a recent study in Burkina Faso, the Lehmann Funnel Entry Trap (LFET) have shown excellent promise in mosquito densities reducing even in area of high insecticide resistance up to 80%. It requires no chemicals and is self-operated. However, one of the issues of the LFET is the big size of the funnel occupying lot of space inside houses. Here we compared the performance of three new prototypes of LFET with reduced size that combine screening and killing effect on mosquitoes.

Methods

The study was carried out for three months during the rainy season both in low and high malaria vector density sites, Soumousso and Vallée du Kou respectively. The original LFET was modified and 3 new prototypes were produced locally and tested over 3 months (8 days/month) to evaluate their effectiveness in trapping and killing mosquitoes entering houses through the windows.

Results

In both sites, an overall of 78,435 culicine mosquitoes collected in both traps and houses and most of them were mainly *Anopheles gambiae* s.l. n= 76,558 (98%) and other species represented n = 1,877 (2%). Of the culicine caught in the trial, n= 55,256 (72%) were collected in traps. The 3 new LFET prototypes reduced the indoor density of mosquitoes collected in the houses by a range of 36 to 73% and 69 to 70% in low vector density setting, Soumousso and high vector density area, Vallée du Kou respectively. The prototype 1 caught a greater number of mosquitoes than the prototype 2 whereas no difference was observed between other prototypes in VK3. In Soumousso, the prototypes 1 and 2 collected significantly higher number of mosquitoes compared to the prototypes 3 and 4.

Conclusion

This study has shown that the new LFET prototypes are promising for malaria vector control and could enter in the malaria vector control toolbox in the coming years. Therefore, a large-scale study with one of the prototypes is needed on the practical ability and community acceptance of the LFET to control malaria vectors.

Background

Malaria has decreased dramatically over the last decade, the number of deaths has dropped to 405, 000 deaths reported in 2018 in the world [1] mainly thanks to a significant up scaling of vector-control interventions. Current vector control relies primarily on two main interventions, the use of Long-Lasting Insecticidal Nets (LLINs) and indoor residual spraying (IRS) [2]. However, the widespread of insecticide resistance among *Anopheles* mosquito species is threatening and undermining the global effort against malaria elimination/eradication [3, 4].

Thus, in order to manage the widespread of mosquito insecticide resistance, one important consideration is focusing on monitoring mosquito vector populations, a key element of vector management and assessment of mosquito borne disease [5]. Therefore, there is a consensus and urgent need in malaria control community for exploration of novel approaches and tools toward malaria control.

Malaria parasite transmission is mediated by females of *Anopheles* mosquitoes in Sub-Saharan of Africa. These female mosquitoes to have progeny and survive, need to seek for blood meal in order to lay eggs, and that blood meal takes place most of the time late and indoors on humans [6–8].

The anthropophilic and endophilic behaviours of malaria vectors force them to enter the house for having blood meal on humans [6, 7] whereas this renders them vulnerable and tacklable in many ways. So even malaria being mostly transmitted indoors, some studies in East Africa, have shown a malaria residual transmission occurring outdoor by mosquitoes feeding outside on humans [9–11]. Mosquito entry rates and hence transmission are affected by house construction [12] and it is widely acknowledged that poor quality housing is generally accepted to be an important contributor to ill health [13]. Recently, new approaches based on house ceilings, doors, windows and eaves were recently developed to block and reduce malaria vector densities entering into houses and thus transmitting diseases [12–14]. Furthermore, these approaches exploiting the behaviour and house entry ways of the vectors, mainly malaria mosquitoes have been explored as part of malaria vector control strategies [16, 17]. These aspects were therefore brought together to design the Lehmann's Funnel Entry Trap (LFET), a house screening trap, combining mosquito behaviour and entry route [18]. The potential of the original LFET to control mosquito densities has been successfully demonstrated and preliminary results showed a house entry reduction by 71% in Vallée du Kou, a high mosquito density in Burkina Faso [18].

However, the original LFET prototype was taking lot of space in the house due to its high volume, and people were complaining in the trial site. In response to this major issue, the original LFET was modified in several ways to produce smaller prototypes for malaria vector control.

Here we report the development and the evaluation of 3 novel and smaller LFET prototypes compared to the original LFET prototype [18] in low and high mosquito density and ecological areas. In addition, this study aimed at determining a promising prototype that could be proposed as such, as a vector control tool.

Methods

Study areas

The study was carried out in two ecological settings, Vallée du Kou and Soumousso, two villages close to Bobo-Dioulasso, Burkina Faso:

Vallée du Kou, a village located in the north-west of Bobo-Dioulasso. The site is characterized by wooded Savannah and covers 1,200 ha. Vallée du Kou (11°23' N, 4°24' W) is a rice area with high mosquito densities all the year. A high insecticide resistance level is then occurring in this area due to the large use of insecticide in rice culture and the cotton around. The village is composed of seven quarters and the trial has been conducted in one of these quarters, Vallée du Kou 3 (VK3). Given the presence of water all year round, mosquitoes are found accordingly with a peak density observed in August-September during the rainy season. *An. gambiae* and *An. coluzzii* are alternated during the year with a predominance of *An. coluzzii* throughout the year. Both species in this area are highly resistant to pyrethroids and DDT (kdr based mechanism, (0.8–0.95). A raise in ace-1 resistance frequency is also observed.

Soumousso, located in the southern of Bobo-Dioulasso, 11°04' N, 4°03' W), is a drier setting than VK3 with less water and where the most dominant species are *An. gambiae* and a mixture of *An. funestus*, *An. arabiensis* and other sibling species of *An. gambiae* s.l.

In this area, the mosquito density is lower compared to that of Vallée du Kou and the dynamics of the mosquitoes follow the rainy season scale.

Description Of The Traps

The original LFET prototype (Prototype 1) (P1) was described by Diabaté and collaborators in 2013 [18] and was made of a metal frame (Long = 69 cm, Large = 51 cm, Height = 165 cm) fitted from the bottom to the top with a regular mosquito net to prevent mosquitoes and other insects entering the trap to escape (Fig. 1). A funnel made of metal frame was inserted at the top of the trap in such a way that mosquitoes approaching the window go first through the big opening of the funnel and enter the trap passing through the small opening.

The big opening of the funnel is long of 70 cm and the diagonal is long of 54 cm while the small opening is long of 13.3 cm and large of 11.2 cm. The small opening of the funnel is distant of 10 cm from the backside of the trap. The funnel, the way and the place where it is inserted in the cage allow mosquitoes to enter easily the trap but prevent them from escaping. Once mosquitoes enter the trap, they have a big volume beneath the funnel where they can find refuge. For a mosquito to escape, it will have to fly all the way up towards the small opening of the funnel and be able to navigate through the 10 cm space separating the small opening of the funnel and the back side of the cage. Ultimately it will fly to exhaustion before it can find the way out. The principle of LFET is to lose the mosquito inside the volume of the trap until dehydration and finally death without finding the way out to exit. The trap is fitted with three sleeves on the side (one below, one in the middle and one on the top) through which mosquitoes were aspirated from the cage. The trap was secured to window using nails.

The Prototype 2 (P2) was built on the same model of the original LFET prototype but half reduced, with the same dimensions except the funnel height of 82.5 cm (Fig. 2). The whole trap is covered by a net, fitted with three sleeves for mosquito collection.

The Prototype 3 (P3) is made of a metal funnel frame measuring (Length = 81 cm, Height = 80 cm). Here the small funnel opening used as entry is a circular metal funnel which opening size is 16.5 cm of diameter, instead of small rectangle in the original prototype. Two circular openings on right and left (distance between both circular openings is 12.5 cm) were planned to fit the window position and the place where it should be fixed to the wall in the house. When one is used, the second is closed by the net. The volume of trap was made of a rectangular metal form (dimensions are, Length = 110 cm, width = 16 cm, height = 56 cm) covered by a net. A net is covering the trap on backside of the funnel. This trap fitted five sleeves allowing mouth aspiration of mosquito within the cage (Fig. 3).

The prototype 4 (P4) is similar to the prototype 3 with the same dimensions of funnel and volume, but the difference between both is a little circular funnel frame extending the funnel. This small circular funnel is 9 cm-long from the beginning to the end, like a shrinking canal giving access to the beneath volume. This last circular funnel (10.5 cm of diameter) is the funnel whereby mosquitoes must pass to access the trap volume. This last circular funnel is far from the back side of the trap of 10 cm. In addition, this trap was equipped with a mirror for internal uses within the house. This trap was also outfitted by five sleeves enabling the mosquito collection. Other difference between both prototypes 3 and 4, is the mirror added. Finding a useful trap for the occupants coupled to reducing mosquito density may help increasing the acceptability and friendly use of the trap (Fig. 4).

Study Design And Mosquito Collection

Three LFET prototypes (P 2, 3, 4) were tested in two ecological settings in comparison to the original LFET prototype (P1). The performance of the traps was assessed in terms of trapping and suppressing the indoor malaria vectors as well as other vectors entering the house through window. A total of 12 houses were chosen in each testing site. Only houses with single room, single window and single door were included in the study. To ensure that mosquitoes had no other alternative except the windows to enter in the house, all the small holes of the ceiling and the wall were closed with sponges or clothes and a curtain was placed at the entrance of each house (Fig. 5). The inhabitants were informed on the aim of the study and therefore free to use their doors as they will. The window with LFET was left continuously opened to allow the mosquitoes to access the inside of house. Trap installation and mosquito sampling were performed over 8 consecutive days per month, from September to November in VK3 and Soumousso.

The daily mosquito collection started at 07:00 am using a mouth aspirator and a pyrethrum spray catch [19] was performed in the trap and the corresponding house. All the mosquitoes caught were kept in single cup, then killed with chloroform and morphologically identified on site. The daily work on site consisted in mosquito separation, sorting, identification, sexing according to Gillies key [20], counting and scoring per genus, species, physiological status and the numbers recorded on a spreadsheet.

All traps set up each month were removed at the end of the trial. Along this study, the villagers involved in the trial, were asked to feel free to give their opinion on any inconvenience related to the new LFET prototypes.

Statistical analysis

Microsoft Excel 2007 (Microsoft®, New York, USA) was used to record data, and R-3.6.2 (package dplyr, questionr, and coin) to produce the graphs and the statistical analyses.

In the study, four main variables were analyzed:

1) Total number of *An. gambiae* s.l. caught calculated as (number in trap) + (number in corresponding room);

2) Proportion of mosquitoes caught in trap: calculated as (number caught in trap)/ (total number)

3) Proportion of entry reduction: calculated as the proportion of mosquitoes caught in trap/total of mosquitoes caught (trap + room) × 100.

4) A daily removal of mosquitoes was calculated as the number of all mosquitoes collected per site (trap + house) /24 days (8 consecutive days over 3 months)

All collected mosquitoes in the traps were sex-sorted. Female mosquitoes were sorted according to the gonotrophic status and the proportion of each status (gravid, blood fed and unfed) was estimated by dividing the number of females of the specific status by the total number of females caught in the traps.

The numbers of mosquitoes in the trap and the matching house did not follow a normal distribution. Therefore, a non-parametric test, Kruskal-wallis sum test, was used to assess the overall performance of the traps. The effects of monthly collection in mosquito density reduction were evaluated using a Levene's test for homogeneity of variance (center = median) with a Wilcoxon rank sum test with a p-value bonferroni adjustment method. A Tukey multiple comparison of means with 95% family-wise confident level test, was used for pair-wise comparison between LFET prototypes.

To assess whether the traps caught more mosquitoes than those entered in the matching houses, a comparison using Wilcoxon rank sum test with a holm p-value adjustment method was used.

Results

The LFET is simple and can be used friendly and easily by the populations, requiring no high technology to be produced, no chemical or attractant. It's easy to install and use in the house.

Performance Of The Lfet Prototypes For Mosquito Density Reduction

Overall 78,435 mosquitoes were collected in the 2 study sites and were composed of n = 76,558 (98%) of *An. gambiae* s.l. (table 1) and n = 1,877 (2%) of other species including *Culex* sp, *An. pharoensis, Mansonia* sp, *An. coustani, An. rufipes, An. funestus, An. flavicosta*, and *Aedes* sp (table 2). Out of these *An. gambiae* s.l. mosquitoes, 75,471 and 1,087 were caught in both traps and houses with 72% and 60% of these trapped in VK3 and Soumousso, respectively (table 1). In VK3, the original LFET prototype collected a daily average of mosquitoes ranging from 36 to 675 during the trial while the prototype 2 had 26 to 414 and the prototypes 3 and 4 collected between 17 to 635 and 19 to 490 respectively (Fig. 6).

In Soumousso, the daily average mosquito collection per trap ranged from 0 to 9 mosquitoes for the prototype 1, from 0 to 19 for the prototype 2, from 0 to 2 mosquitoes for the prototype 3 and from 0 to 4 mosquitoes for the prototype 4 (Fig. 6).

During the study in VK3, the prototype 1 showed a house entry reduction by 78%, the prototype 2 by 70%, and the prototypes 3 and 4 by 69% (Table 1).

When the performance of the traps was assessed in terms of trapping and suppressing the indoor malaria vectors, the monthly effect on the mosquito collection in traps showed a significant variation between traps according to the study site (Kruskal-wallis, χ^2 = 10.9, df = 3, p = 0.012 in VK3; and χ^2 = 40.7, df = 3, p < 0.0001 in Soumousso). The variation level in performance between traps was confirmed, by the Levene's test for homogeneity of variance (center = median) (group 284, df = 3, F value = 1.9, Pr (> F) = 0.13) in VK3) and (group 283, df = 3, F value = 6.56 Pr (> F) < 0.001 in Soumousso).

The prototype 1 caught a greater number of mosquitoes than the prototype 2 (p = 0.014) whereas no difference was observed between other prototypes in VK3 (Fig. 7a). In Soumousso, the prototypes 1 and 2 collected similar number of mosquitoes but significantly higher than that of the prototypes 3 and 4 (p < 0.0001) (Fig. 7b). The prototype 1 reduced the mosquito entry by 73% while the new prototypes (2, 3, and 4) reduced the house entry reduction by a range of 36 to 73% in Soumousso (table 1). A greater number of mosquitoes were caught in the traps compared to the matching houses (p [illegible] 0.0001) (Fig. 8).

The daily mosquito removal on average *An. gambiae* s.l. density was 262/house/night/trap in VK3 and 3.77/house/night/trap in Soumousso during the study period and the Fig. 9 summarised the dispersal numbers of mosquitoes collected per trap/month/site of collection per site.

Gonotrophic Status Of Collected Mosquitoes

Out of 76,558 *An. gambiae* s.l. mosquitoes caught in the traps, n = 58,439 (76%) were females and n = 18,119 (24%) were males. Out of the female mosquitoes (n = 58,439) collected in the traps, the unfed mosquitoes likely seeking for a blood meal represented n = 23,051 (39%), the blood fed n = 28,359 (49%), and the gravid females n = 7,029 (12%).

Discussion

The objective of this study was to assess the modified LFET prototypes performance in terms of suppressing malaria vectors and to determine the most promising prototype as a vector control tool. We found that the new LFET had a potential to reduce malaria mosquito densities in VK3. Conversely in Soumousso, the new prototypes, except the prototype 2, showed a relative low-density mosquito reduction compared to the original LFET prototype. In addition to *An. gambiae* mosquitoes, other mosquito species responsible of neglected tropical diseases such as lymphatic filariasis and dengue were collected.

Our results are consistent with the previous study where the original LFET [18] was able to reduce by 70 to 80% the number of mosquitoes in the houses in high mosquito density area meaning that the size of the new prototypes did not impact their performance. In addition to the smaller size (reducing the cost of the manufacture) of the modified LFET, the prototype 4 has a mirror for inhabitant free uses, making the trap design more beautiful and which would probably improve acceptability. This would increase the chance of using the new prototype in a larger scale evaluation if a deployment is envisioned. The newly developed prototypes were able to deny the house entry to malaria mosquitoes and other insects and preventing from harmful effect of mosquito bites. The design of the new prototype 2 was quite similar to the original prototype except the height of the volume. However, the most important part of the trap, which is the funnel, was the same and could explain why the prototype 2 performed as well as the original prototype. The circular funnel of the prototypes 3 and 4 makes them different from the prototypes 1 and 2 and this may explain why they did not catch significant numbers of mosquitoes. Moreover, the smaller, horizontal volume screen and reduced airflow inside these prototypes 3 and 4 may contribute and explain the less catches of mosquitoes compared to the prototypes 1 and 2. Several studies have demonstrated that some variables such as vertical or horizontal screens, air flow and direction, trap colour, screen mesh size, etc. could more directly affect the trap function [21, 22].

Since the establishment of a link between malaria and mosquito [11], house screening has been one of the first experiments used as part of malaria vector control management [23]. Recent studies using house screen have found a reduced number of mosquitoes entering the house and a consequent protection of the households against mosquito bites compared to houses without screen [18, 24, 25]. In addition to the house screening, other studies demonstrated that well-built house with fewer entry points for mosquitoes or specific modifications of house design can help reducing malaria transmission by lowering human exposure to infectious bites [26, 27].

Thereby, the use of the LFET could be an effective and relatively simple method of reducing indoor mosquito vector densities and consequently decreasing malaria transmission and harmful effect of other insects, at a population level when all people concomitantly use it in the same area. In this way, the trap could help removing not only young mosquitoes but also the old ones able to transmit malaria parasites, in addition to catching and killing any mosquito, whatever resistant or susceptible, male or female. For example, Vallée du Kou is an irrigated area where a lot of pesticides are used in rice and cotton fields surrounding the village, and significant proportion of mosquitoes exhibited high resistance level to pyrethroids [18, 28, 29].

Malaria control is undermined by the raise of a growing proportion of outdoor feeding mosquitoes, [9–11] due the use of insecticide in LLINs and IRS, that need also to be tackled. The LFET could help removing these outdoor fed mosquitoes looking for refuge areas inside the house compared to that of CDC light trap which sampled mosquitoes, and need to be close to the indoor sleepers [8]. This would also not be achieved with window screening as mosquitoes would be denied entering in a given house, but not trapped and so, they might find a suitable place to bite, transmit their pathogens, rest and reproduce.

In this study, *An. gambiae* was the main species collected in both sites. Female *Anopheles* mosquitoes regularly take blood meal every 3 days to mature their eggs and owing to this obligation and depending on the cues smelt, they need to enter to human habitats or animal housing [6–8]. During this study, LFET prevented and protected people from a significant proportion of unfed female mosquitoes likely searching a blood meal, 39% of total *An. gambiae* mosquitoes caught, and removed a greater proportion of the outdoor blood fed mosquitoes (49%) that were looking for resting sites to digest their blood and 12% of outdoor gravid females, that may be found in the house without the installation of LFET at the windows and curtain at the doors. When the trap is installed, any mosquito or insect coming through the window, male, female, resistant or susceptible, young or old without any distinction, is intercepted and killed, and consequently the numbers of indoor mosquitoes are reduced in the house. Given that malaria parasites are transmitted by old female mosquitoes, so trapping all mosquitoes and mainly the old ones, could reduce the malaria cases and burden in endemic areas. Furthermore, the traps caught harmful insects, 2% of overall mosquitoes collected in both study sites, as *Culex*, *Mansonia*, *Aedes* and other *Anopheles* species involved in vector borne diseases transmission such as lymphatic filariasis and dengue.

Conclusion

The new LFET prototypes developed and tested in high and low mosquito density settings, have shown a promising performance in mosquito density reduction suggesting their potentially useful supplement to existing vector control tools to drop down the malaria mosquito populations.

Importantly, when the trap is well used, it protects from the harmful insects such as malaria mosquitoes and other vector borne-diseases mosquitoes. Based on our findings, the most promising trap is the prototype 2.

Although an improved LFET has been developed, further studies at a village scale will be needed, to draw out its effectiveness in dropping down malaria vectors populations.

Declarations

Competing interests

The authors declare that they have no competing interests.

Availability of data and materials

All data generated and analysed during this study are available on Github: https://github.com/RogerSANOU/Lehmann-trap-dataset.git.

Authors' contributions

RS carried out the field work, participated in the analysis of the data, and wrote the original manuscript. HM and EB contributed to write and critically revised the paper. AO carried out the social field work and revised the manuscript. PSS, KB, AMGB, LP and RKD revised the manuscript. BDS carried out the statistical analyses. DA designed the trap and the study, analysed the data and supervised the entire work. All authors have read and approved the final manuscript.

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Figures

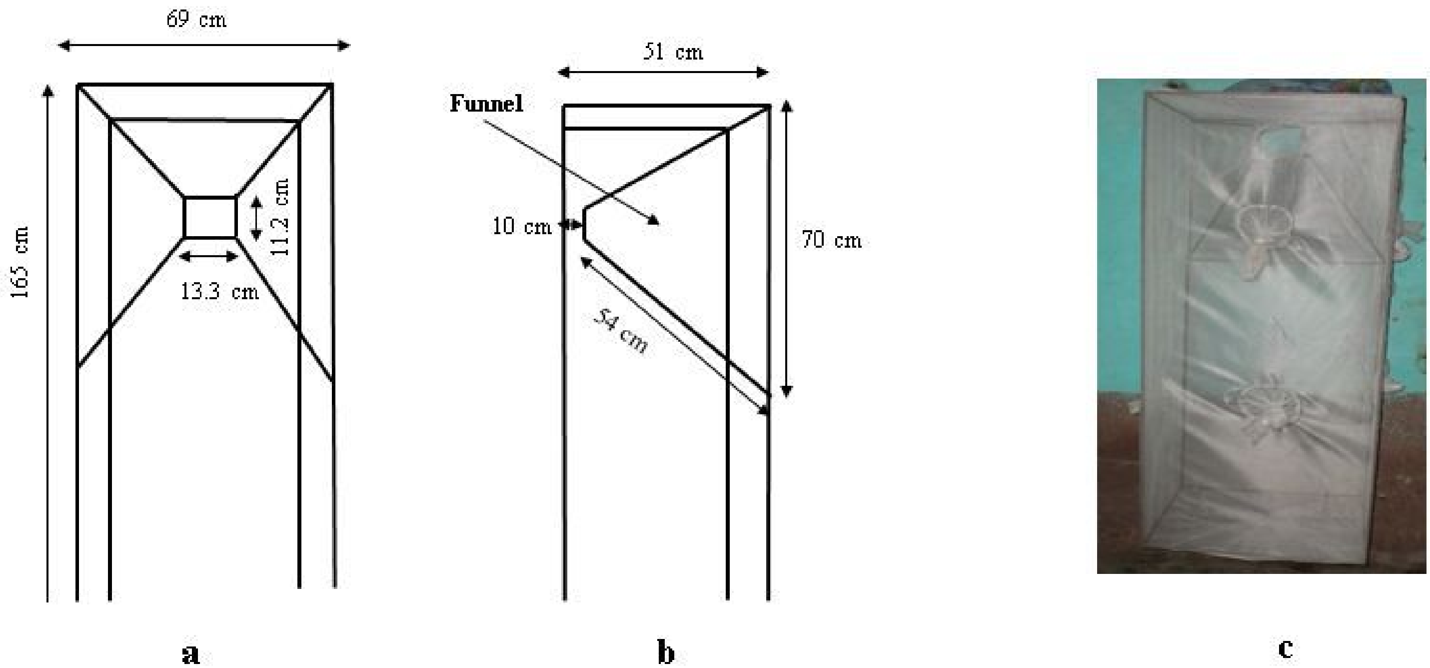


Figure 1

Dimensions of the trap, front and side view. (a) Inserts front view: 69 cm wide × 165 cm high; 13.3 cm long × 11.2 cm wide (small opening of the funnel). (b) Inserts side view: 51 cm depth of the trap, 70 cm long × 54 cm diagonal (large opening of the funnel); 10 cm distance of the small opening of the funnel from the backside of the trap (Diabaté et al. 2013); (c) original prototype 1 outside view inside a house

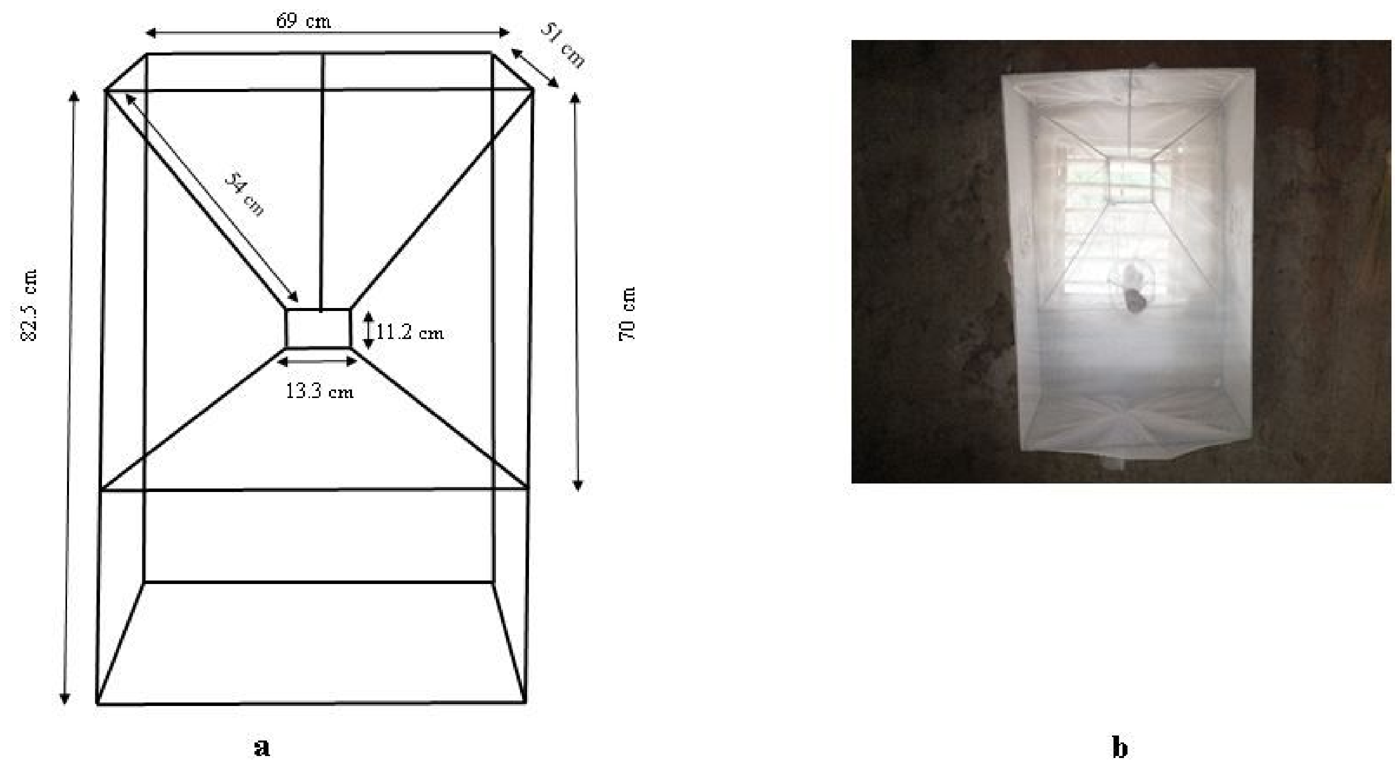
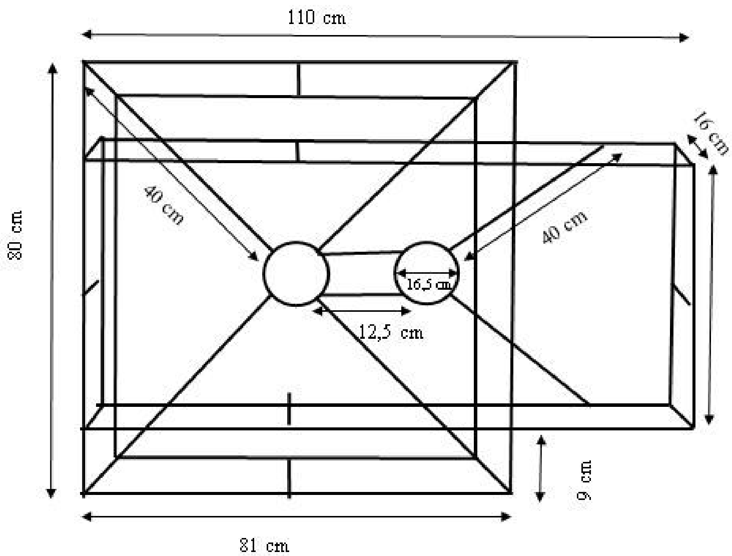


Figure 2

(a) Dimensions of the trap, front and side view. Inserts front view : 69 cm wide × 82.5 cm high; 13.3 cm long × 11.2 cm wide (small opening of the funnel). Inserts side view: 51 cm depth of the trap, 70 cm long × 54 cm diagonal (large opening of the funnel); 10 cm distance of the small opening of the funnel from the backside of the trap.; (b) Prototype 2 outside view inside a house

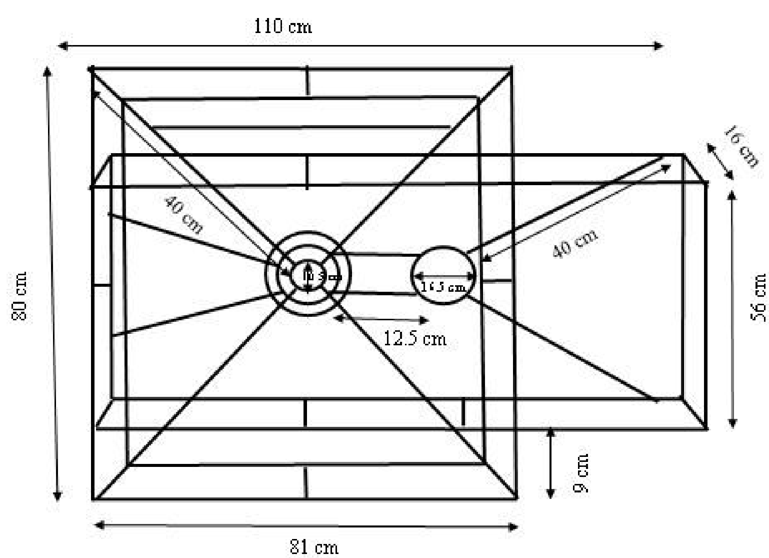


a

b

Figure 3

(a) Dimensions of the trap, inserts front view. 16 cm wide × 80 cm high; 16.5 cm diameter (circular opening of the funnel), 56 cm depth of the trap, 81 cm long × 40 cm diagonal (large opening of the funnel); 10 cm distance of the small opening of the funnel from the backside of the trap.; (b) Prototype 3 outside view inside a house, with circular funnel as entrance



a

b

Figure 4

(a) Dimensions of the trap, inserts front view. 16 cm wide × 80 cm high; 16.5 cm diameter (circular opening of the funnel), 56 cm depth of the trap, 81 cm long × 40 cm diagonal (large opening of the funnel); small circular funnel diameter 10.5 cm, distance from the beginning of the large circular to the end of small circular 9 cm, and 10 cm distance of the small opening of the funnel from the backside of the trap; (b) Prototype 4 outside view inside a house, with small circular funnel entrance

Figure 5

a curtain placed at the door of a study house

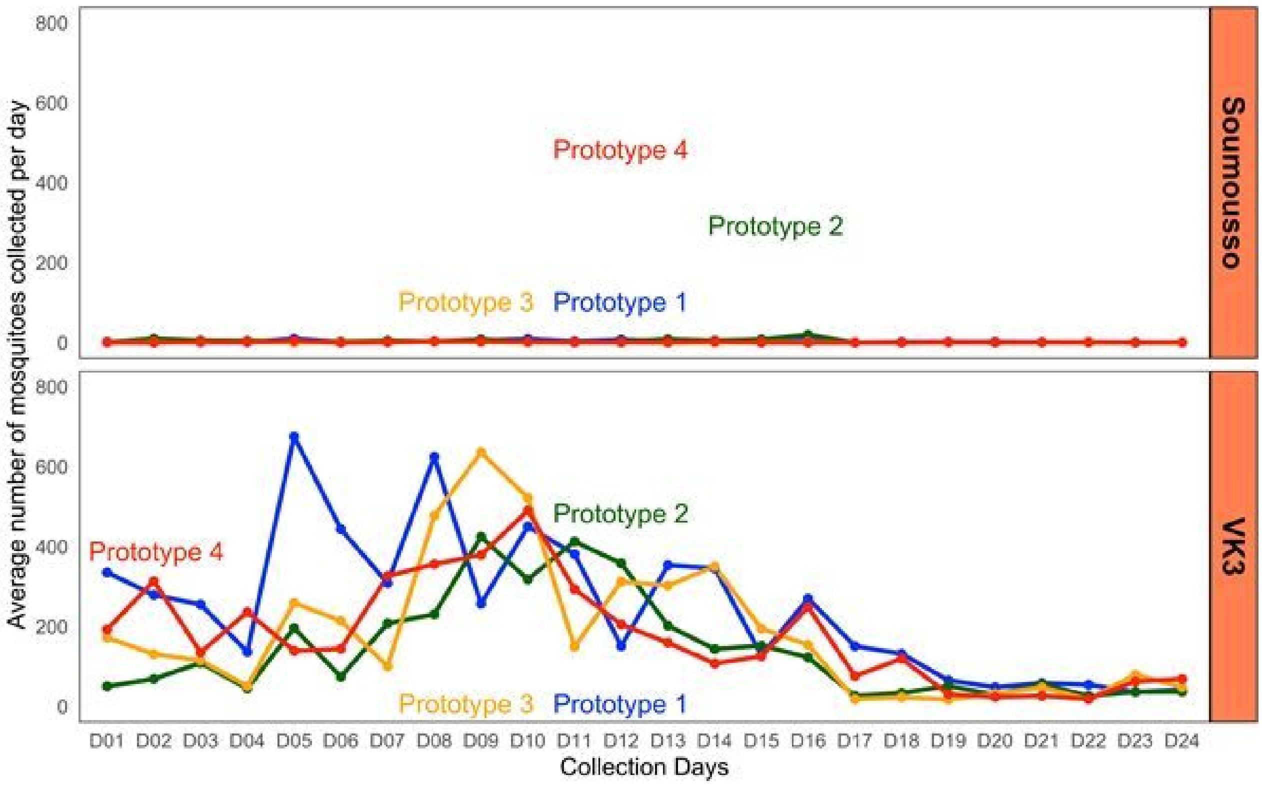


Figure 6

Mean number of mosquitoes collected per Lehmann's Funnel Entry Trap prototype/day/site

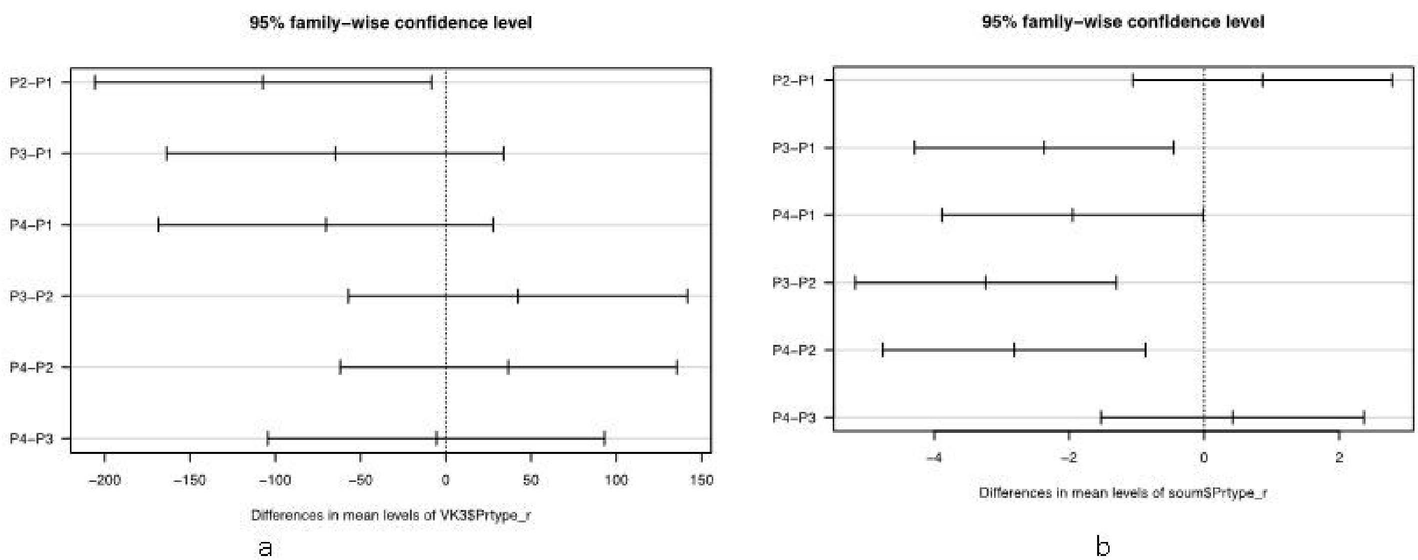


Figure 7

Tukey multiple comparisons of means 95% family-wise confident level (a) in VK3 and (b) in Soumousso

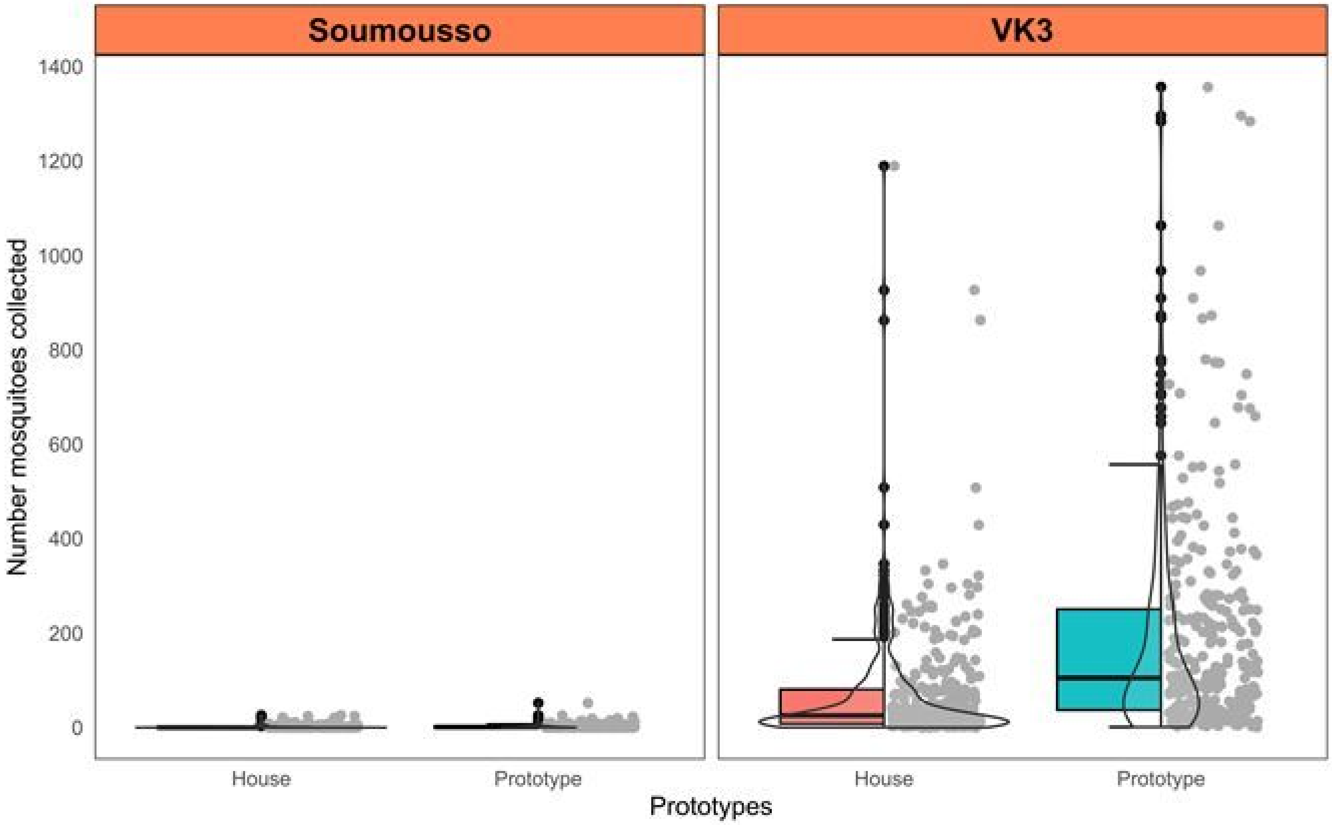


Figure 8

Number of mosquitoes collected per trap versus house per site during the study period

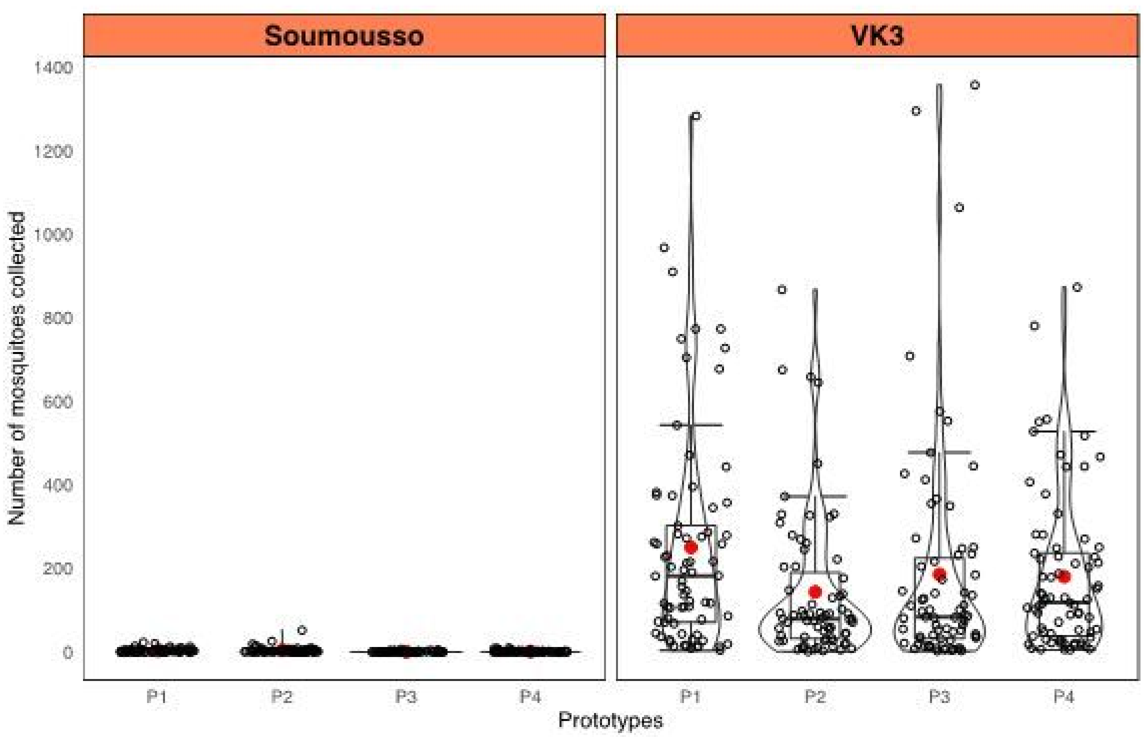


Figure 9

Number of mosquitoes collected per trap and per month

Supplementary Files

This is a list of supplementary files associated with this preprint. Click to download.

- supplement1.pdf