Indoor environmental factors and house structures affect vaporization of active ingredient from spatial repellent devices placed in typical rural houses in south eastern Malawi

Hitoshi Kawada, Shusuke Nakazawa, Kazunori Ohashi, Eggrey Aisha Kambewa, and Dylo Foster Pemba

Received: August 10, 2021. Accepted: October 19, 2021. Published online: November 30, 2021. DOI:10.7883/yoken.JJID.2021.584
Indoor environmental factors and house structures affect vaporization of active ingredient from spatial repellent devices placed in typical rural houses in south eastern Malawi

Hitoshi Kawada¹*, Shusuke Nakazawa¹, Kazunori Ohashi², Eggrey Aisha Kambewa³, Dylo Foster Pemba³

¹ Department of Vector Ecology and Environment, Institute of Tropical Medicine, Nagasaki University, Sakamoto 1-12-4, Nagasaki, Nagasaki, 852-8523 JAPAN

² Health and Crop Sciences Research Laboratory, Sumitomo Chemical Co., Ltd., Takatsukasa 4-2-1, Takarazuka, Hyogo 665-8555 JAPAN

³ Vector Borne Disease Laboratory, Chancellor College, University of Malawi, P.O.Box 280, Zomba, Republic of Malawi

*Corresponding author: Hitoshi Kawada
Department of Vector Ecology and Environment, Institute of Tropical Medicine, Nagasaki University, Sakamoto 1-12-4, Nagasaki, Nagasaki, 852-8523 Japan.
Tel: +81-95-819-7811, Fax: +81-95-819-7812
E-mail: vergiss@nagasaki-u.ac.jp

Running title: Factors for vaporization of spatial repellent
**Key-words:** *Anopheles*, House structure, Malawi, metofluthrin, temperature
**ABSTRACT:** The use of a metofluthrin-impregnated spatial repellent device (MSRD) is a new, practically effective method for preventing mosquito blood feeding. Indoor environmental factors such as room temperature and ventilation rate are thought to be important for MSRD activity. Measurements of room temperature and vaporization of metofluthrin from MSRD in typical rural metal-roof houses and thatched-roof houses in southeastern Malawi were conducted. The relationship between house structure and number of collected Anopheline mosquitoes with and without MSRD treatment was also investigated. The difference between daytime room temperature and nighttime room temperature was significantly higher in metal-roof houses than thatched-roof houses. The vaporization of metofluthrin from the MSRD was not accelerated by the high room temperature, but by the high indoor air flow by ventilation. The number of mosquito collections was significantly higher in thatched-roof houses than in metal-roof houses. MSRD-treated thatched-roof houses have a higher probability of mosquito infestation, but the vaporization of metofluthrin is also higher due to indoor air flow, resulting in a reduction in mosquito numbers. Metal-roof houses with closed eaves reduce the probability of mosquito invasion, and a longer predicted effectiveness occurs with MSRD because of the controlled release of metofluthrin through lower indoor air flow.

**INTRODUCTION**

Insecticide-treated nets (ITNs) and long-lasting insecticidal nets (LLINs) are effective only when the mosquito vectors are endophagous and their blood-feeding time corresponds to the human sleeping period (1). Iwashita et al. reported that children under 15 years of age had lower rates of bed net use than those in other age groups in houses along Lake Victoria in
western Kenya (2). The use of bed nets was limited to parents and their infants sleeping in the bedroom, while the rest of the family (especially children) often slept in the living room without bed nets, leading to a high malaria transmission rate among this generation (2). The effective use of bed nets is strongly influenced by the ease with which they can be installed in the house, and this is especially important for children above infancy who usually sleep in rooms away from their parents where bed nets are often not straightforward to install. Therefore, simplifying the installation of bed nets in places where they are generally difficult to position, or preventing mosquito attacks by other methods could protect this generation from malaria and thus could reduce the overall morbidity of the community.

Metofluthrin (SumiOne®), 2,3,5,6-tetrafluoro-4-methoxymethylbenzyl (EZ) (1RS, 3RS;1RS,3SR)-2,2-dimethyl-3-(prop-1-enyl) cyclopropanecarboxylate, belongs to a new type of pyrethroid group that has a high vapor pressure and high insecticidal and spatial repellent activity against mosquitoes (3). Metofluthrin-impregnated spatial repellent devices (MSRD) have been suggested to be practically effective in preventing mosquito blood feeding in several regions with different conditions (4–9). MSRD has also been reported to be highly effective in repelling pyrethroid-resistant malaria vector mosquitoes in Kenya (10) and Malawi (11). We examined the effect of the combinational use of the MSRD and the long lasting insecticidal net, LLIN (Olyset® Plus) on malaria prevalence and vector mosquitoes in malaria endemic villages in south-eastern Malawi. The intervention reduced the infection rate in children as well as the number of pyrethroid-resistant vector mosquitoes invading in houses. We concluded in the above study that continued intervention using MSRD at 3-month intervals with 2 strips per 10 m² to reduce the density of malaria mosquitoes was recommended in order to effective malaria control (11).
The effective duration of the MSRD appears to be correlated to the vaporization over time, which can be regulated by formulation factors, such as polymer type and density which determine the bleeding rate of an active ingredient. A high vaporization speed might lead to rapid effectiveness, but the effective duration may be lessened. In contrast, a low vaporization rate might result in long-lasting efficacy, but there could be a minimum vaporization over time under which no efficacy is expected. Indoor environmental factors, such as room temperature and ventilation rate, are thought to be important for residual activity as well as formulation factors. In the present study, we measured the room temperature in typical rural houses in southeastern Malawi. The vaporization of metofluthrin from the MSRD in the houses was also detected by chemical analysis, and the relationships among house structures, indoor environmental factors such as room temperature and air flow, and the vaporization of metofluthrin from MSRD are discussed.

**Materials and Methods**

**Study area and houses:** The study was performed in two adjacent villages, Chiliko and Chilore, in Likangala, in the eastern area of the Zomba district (15°23'S, 35°20'E), in a south-eastern region of Malawi. The Zomba district is located in a malaria-endemic area with a high malaria prevalence (12). Malaria is one of the leading causes of death in the district (Zomba District Health Office, 2009). *Anopheles arabiensis* Patton and *Anopheles funestus* s.s. (Giles) are the main malaria vectors in the study area, and both possess high metabolic resistance to pyrethroids and DDT, while not a single point mutation at the voltage-gated sodium channel (L1014F or L1014S) was detected in *An. arabiensis* and *An. funestus* s.s. in the study site (11). Most of the houses in the study area were built of mud bricks, some of
which were fired, and the rest were dried. The houses were broadly categorized into two types: metal-roofed and thatched-roofed (Fig. 1).

**Relationship between room temperature and the vaporization of metofluthrin from the MSRD**

**Selection of the houses and intervention of the MSRD:** The study was performed between January and May 2015, which is the rainy season of the study area (http://www.worldweatheronline.com/Zomba-weather-averages/Zomba/MW.aspx). Nineteen metal-roof houses and 21 thatched-roof houses in Chiliko village were selected for the study (Table 1). The MSRD (polyethylene net materials of 8 × 15 cm impregnated with metofluthrin 10 % w/w per plastic strip, Sumitomo Chemical Co., Ltd., Tokyo, Japan) was hung in the rooms of the 20 houses at two different dose regimens, approximately two strips per 10 m² for 9 houses, and three strips per 10 m² for the other 11 houses (Fig. 1 A, B). The house residents were informed of the study, and their written consents were obtained before conducting the study. The MSRD intervention was done on January 22 and 23, 2015.

**Measurement of room temperature:** The room temperature of the 40 houses was recorded at 2-h intervals with a micro data logger (Thermochron® Type G, KN Laboratories Inc., Osaka, Japan). The data loggers were hung from ceilings at the same height as the MSRD (Fig. 1 E).

**Analysis of the amount of metofluthrin in the MSRD:** Four months after the hanging of the MSRD (on May 18, 2015), all strips were collected and the remaining quantities of metofluthrin in the MSRDs were analyzed using gas chromatography.
Relationship between house structure and number of anopheline mosquitoes

**Selection of houses and collection of anopheline mosquitoes:** The study was performed between December 2015 and May 2016. Seven metal-roof houses and 13 thatched-roof houses in Chiliko village and six metal-roof houses and 13 thatched-roof houses in Chilore village were selected for the study (Table 2). Prior to the MSRD intervention, Olyset® Plus (permethrin 2% and PBO 1% w/w, Sumitomo Chemical Co., Ltd., Tokyo, Japan) were delivered to the above 39 houses after removing the old bed nets for making the indoor mosquito repelling measure except for MSRD uniform among the houses. Two interventions of MSRD (2 strips /10 m²) were implemented for the houses in Chilore village in January and April 2016, whereas no intervention of the MSRD was done in Chiliko village. Second intervention of MSRD in Chilore village was done after removal of all MSRDs placed in the 1st intervention. The house residents were informed of the experiment, and their written consents were obtained before conducting the study. Pyrethrum spray collections (13) for the houses were performed in the morning (07:00–10:00) by three to four staff members. Collections were done on December 3–4, 2015 before the intervention of MSRD, and February 12, March 8–9, April 8–13, and May 9–11, 2016 after the intervention of MSRD.

**Identification of mosquitoes:** Collected mosquitoes were identified as *An. gambiae* s.l. and *An. funestus* s.l. microscopically based on the identification keys (14). DNA of the mosquito sample was extracted using the RedExtract-N-Amp™ Tissue PCR kit (Sigma-Aldrich Japan, Tokyo, Japan) according to the manufacturer’s instructions, and the multiplex
PCR method (15, 16) was used for species identification.

**Data analysis:** The average daytime (06:00-18:00), nighttime (20:00-04:00), and overall room temperature was calculated for four months for each house. Wilcoxon rank sum test and one-way analysis of variance for the room temperature and the number of mosquito collections, and linear regression analysis between the average temperature and the average amount of remaining metofluthrin in the MSRD were performed using JMP Pro 15.0.0 (SAS Institute Japan Inc. Tokyo, Japan).

**Ethics:** The protocol for the study was reviewed and approved by the University of Malawi, Chancellor College Research and Ethics Committee and the Ethical Committee of Institute of Tropical Medicine, Nagasaki University (Approve No. 140605123-2).

**Results**

**Relationship between room temperature and the vaporization of metofluthrin from the MSRD:** Changes in the room temperature in the metal-roof houses and thatched-roof houses are shown in Fig 2-A and B. Average room temperature calculated for each house during the day (Dt, 06:00-18:00) and night (Nt, 20:00-04:00) in the metal-roof houses were 28.7 °C (standard deviation, 0.93) and 25.5 °C (SD, 0.67), while those in the thatched-roof houses were 26.0 °C (SD, 0.61) and 25.1 °C (SD, 0.82), respectively (Fig. 2-C). The Dt was significantly higher than the Nt in both the metal-roof houses ($\chi^2 = 27.8$, df = 1, $P < 0.0001$) and thatched-roof houses ($\chi^2 = 12.2$, df = 1, $P = 0.0005$). Although the differences in
the room temperature between metal-roof houses and thatched-roof houses were significant in both Dt \((\chi^2 = 28.2, \text{df} = 1, P < 0.0001)\) and Nt \((\chi^2 = 4.21, \text{df} = 1, P = 0.040)\), the deviation between Dt and Nt \((\text{Dt} - \text{Nt})\) was significantly larger in the metal-roof houses than in the roofed houses \((\chi^2 = 26.2, \text{df} = 1, P < 0.0001)\).

Chemical analysis of the amount of metofluthrin remaining in the MSRD was performed for the 20 MSRD-treated houses (8 metal-roof and 12 thatched-roof houses) (Fig 3-A). A total of 124 MSRD (62 MSRD each for the metal- and thatched-roof houses,) was analyzed. The average remaining percentage (w/w) of metofluthrin in the metal-roof houses and thatched-roof houses were 3.81 % (SD, 2.1) and 1.68 % (SD, 0.98), respectively. The actual percentage of metofluthrin in the MSRD obtained by measurement before intervention was 10.6 % (W/W). The remaining amount was significantly higher in the metal-roof houses than in the thatched-roof houses \((\chi^2 = 33.8, \text{df} = 1, P < 0.0001)\). Figure 3-B shows the relationships between the average room temperature and the average amount of metofluthrin remaining in the MSRD per a house at four months after treatment. No correlation was observed between the remaining amount and the average room temperature in the thatched-roof dwellings \((R^2 = 0.0002)\). The correlation was also low in the metal-roof houses, while it showed a slight positive slope \((R^2 = 0.2097)\), indicating that the higher the room temperature, the lower the vaporization of metofluthrin.

**Relationship between house structure and number of anopheline mosquitoes:** A total of 221 female *An. arabiensis* and 65 female *An. funestus s.s.* were collected over the five periods from December 4, 2015, to May 9, 2016, in Chiliko village (without MSRD intervention) and the first collection (December 3, 2015, before the intervention of the MSRD) in Chilore village. The number of female *An. gambiae s.s.* was low (one for Chiliko...
village and two for Chilore village). The number of anopheline mosquitoes (*An. arabiensis*, *An. gambiae* s.s., and *An. funestus* s.s.) in the metal-roof and thatched-roof houses per collection in Chiliko village without MSRD intervention and Chilore village before MSRD intervention are plotted in Fig 4-A. The average number of mosquito collection in the metal-roof houses and the thatched-roof houses were 1.67 (Standard Error, 0.83) and 2.99 (SE, 0.77), respectively. The number of mosquitoes collected was significantly higher in thatched-roof than in metal-roof houses ($\chi^2 = 6.94$, df = 1, P = 0.0084). Anopheline mosquitoes in the metal-roof and thatched-roof houses in Chilore village in the three collections, from March to May 2016 (two, three and four months after the intervention of the MSRD), are plotted in Fig 4-B. The average number of mosquito collections in the metal-roof houses and thatched-roof houses were 0.95 (SE, 0.53) and 0.67 (SE, 0.17), respectively. The difference in the number of mosquito collections between metal-roof houses and thatched-roof houses was not significant ($\chi^2 = 0.64$, df = 1, P = 0.424).

**Discussion**

The present study showed higher daytime room temperature in metal-roof houses than in thatched-roof houses, mainly likely due to the differing effect of solar radiation on the two roof materials. The difference in ventilation rates between the two roof types could also have contributed to the temperature differences, because most thatched-roof houses have open "eaves" between the roof and the wall, as shown in Fig. 1-D, which are not present in the majority of metal-roof structures (Fig. 1-C). All the thatched-roof houses (21 houses) investigated in the present study had open eaves, whereas this feature was only present in one house among 19 metal-roof houses. A recent study reported a large difference in the
ventilation rate between houses with thatched-roof and open eaves and those with metal roofs and closed eaves (17). The openness (open area/volume of house) in African thatched-roof houses (in Tanzania) was reported to be twice as high as that in concrete houses in southern Vietnam (9).

The present results were unexpected, in that the vaporization of metofluthrin from the MSRD did not increase in correlation with room temperature, but the amount of metofluthrin remained in MSRD showed a slight positive correlation with room temperature, indicating that the higher the room temperature, the lower the vaporization of metofluthrin. This indicates that the major vaporization factor for metofluthrin bleed on the plastic surface of the MSRD was not temperature, but another microclimate factor, possibly the air flow by ventilation due to the difference in the structure of the houses, as explained above. The more closed indoor condition, the lower the ventilation and the higher the room temperature. The mathematical model developed for predicting the release of metofluthrin from a cellulose-based paper revealed the dependence of vaporization as an Arrhenius function of temperature, and the mass transfer coefficient in the air was linearly dependent on the air velocity (18). The experimental data for designing the above model show that a 10 °C difference in temperature caused a $< 10^{-4}$ mol difference in the vaporization of metofluthrin, whereas only a 0.2 m/sec increase in the wind velocity caused $10^{-4}$ mol or more vaporization, indicating that wind velocity has the greatest influence on the vaporization of metofluthrin (18). The loss of additives in low-density polyethylene (LDPE) is controlled by the desorption of the additive from the surface of LDPE and depends on the migration speed of the additive through the polymer bulk to reach the plastic surface. An increase in temperature led to an increase in the release of the additive in an LDPE film (19). However, the temperature range by which a substantial increase in release percentage of additive was noted was 35–115 °C (19), and the increase in release percentage was very low in the range 25–
35 °C. Therefore, the release of the additive (metofluthrin in the MSRD in the present case) on the surface of the MSRD occurring only from the increase in the temperature range in the present study (20–30 °C) (Fig 3-B) was suggested to be very low. Thus, it can be concluded that most of the vaporization of metofluthrin is accelerated by the air flow caused by ventilation.

The openness of houses affects the ability of mosquitoes to enter and invade. The eave gap between roof and wall is thought to be the major route of entry for mosquitoes, and closing the eaves resulted in a 66 % reduction in the number of *An. gambiae* s.l. in houses (20). The number of indoor resting anopheline mosquitoes, among which *An. arabiensis* was the most prevalent, was higher in houses with open eaves than in those with closed eaves (21), and when the comparison analysis was performed only for thatched-roof houses, *An. gambiae* s.l. abundance between the open and closed eaves was still statistically significant (22). In the present study, the number of female anopheline mosquitoes collected in thatched-roof houses was almost twice that of their metal-roof counterparts under the conditions that MSRDs were not treated. The intervention of the MSRD caused a decrease in mosquito invasion both in the metal-roof and thatched-roof houses, and the difference in the number of invasions was not significant between the two roof types (Fig 4-B).

MSRD-treated thatched-roof houses have a higher probability of mosquito infestation, but the vaporization of metofluthrin is also higher due to indoor air flow, resulting in greater mortality or spatial repellency to mosquitoes. Metal-roof houses with closed eaves reduce the probability of mosquito invasion, and a longer effectiveness with the MSRD might be expected because of the controlled release of metofluthrin by lower indoor air flow. Moreover, high temperatures in metal-roof houses may shorten the life span of mosquitoes as Lindsay et al. reported that the metal roof caused an increase in room temperature as compared to the thatched roof, causing the mosquitoes to have a short life span and
negatively affecting the development of *Plasmodium* parasites (23). We have shown that the combinational use of LLIN (Olyset® Plus) and the MSRD with two strips per 10 m² at three-month intervals reduced the vector mosquito density in the field test results performed in the present trial sites in Malawi, where more than half of the houses were rural African-type thatched-roof houses (11). Thatched-roof houses which encompass the majority of houses in the rural African area, have a higher probability of mosquito infestation, but the vaporization of metofluthrin from the MSRD is also higher in such a house structure due to indoor air flow, resulting in a reduction in mosquito numbers. Longer durations of effectiveness could be expected in urban areas where house structures are more closed.

**Acknowledgments** We thank Bosco Rusuwa, Malawi University-Chancellor College, Zomba, Malawi for assisting with this study. We also thank Takashi Suzuki and Kozue Shimabukuro, Kobe-Tokiwa University, Kobe, Japan, for assisting and providing technical support.

**Financial Support** This study was supported by the Japan Society for the Promotion of Science (JSPS) KAKENHI Grant Number JP26305012 and funded by the joint research between Nagasaki University and Sumitomo Chemical Co. Ltd for the creation of field and semi-field bioassay systems for insecticide delivery technology.

**Conflict of interest** None to declare
References

1. Kawada H, Ohashi K, Dida GO, et al. Preventive effect of permethrin-impregnated long-lasting insecticidal nets on the blood feeding of three major pyrethroid-resistant malaria vectors in western Kenya. Parasit Vectors. 2014;7:383.

2. Iwashita H, Dida G, Futami K, et al. Sleeping arrangement and house structure affect bed net use in villages along Lake Victoria. Mal J. 2010;9:176.

3. Matsuo N, Ujihara K, Shono Y, et al. Discovery and development of a novel pyrethroid insecticide 'Metofluthrin (SumiOne®, Eminence®)'. Sumitomo Kagaku. 2005;2005-II:1–15. (https://www.sumitomo-chem.co.jp/english/rd/report/files/docs/20050200imi.pdf)

4. Kawada H., Maekawa Y, Tsuda Y, et al. Trial of spatial repellency of metofluthrin-impregnated paper strip against Anopheles and Culex in shelters without walls in Lombok, Indonesia. J Am Mosq Control Assoc. 2004;20:434–437.

5. Kawada H, Maekawa Y, Tsuda Y, et al. Laboratory and field evaluation of spatial repellency with metofluthrin-impregnated paper strip against mosquitoes in Lombok Island, Indonesia. J Am Mosq Control Assoc. 2004;20:292–298.

6. Kawada H, Maekawa Y, Takagi M. Field trial of the spatial repellency of metofluthrin-impregnated plastic strip against mosquitoes in shelters without walls (Beruga) in Lombok, Indonesia. J Vect Ecol. 2005;30:181–185.

7. Kawada H, Yen NT, Hoa NT, et al. Field evaluation of spatial repellency of metofluthrin impregnated plastic strips against mosquitoes in Hai Phong city, Vietnam. Am J
8. Kawada H, Iwasaki T, Loan LL, et al. Field evaluation of spatial repellency of metofluthrin-impregnated latticework plastic strips against *Aedes aegypti* (L.) and analysis of environmental factors affecting its efficacy in My Tho city, Tien Giang, Vietnam. Am J Trop Med Hyg. 2006;75: 1153–1157.

9. Kawada H, Temu EA, Minjas JN, et al. Field evaluation of spatial repellency of metofluthrin-impregnated latticework plastic strips against mosquitoes of *Anopheles gambiae* group in Bagamoyo, coastal Tanzania. J Am Mosq Control Assoc. 2008;24:404–409.

10. Kawada H. Potential control measures for pyrethroid-resistant malaria vectors. Acta Hortic. 2017;1169:59–72.

11. Kawada H, Nakazawa S, Shimabukuro K, et al. Effect of metofluthrin-impregnated spatial repellent devices combined with new long-lasting insecticidal nets (Olyset® Plus) on pyrethroid-resistant malaria vectors and malaria prevalence - Field trial in south-eastern Malawi. Jpn J Infect Dis. 2020;73:124–131.

12. Kleinschmidt I, Sharp B, Mueller I, et al. Rise in malaria incidence rates in South Africa: small area spatial analysis of variation in time trends. Am J Epidemiol. 2002;155:257–264.

13. WHO. Manual on practical entomology in malaria. Part II Methods and techniques. 1975, World Health Organization, Geneva.

14. Gillies MT, Coetzee, M. A supplement to the Anophelinae of Africa south of the Sahara (Afrotropical region). South African Institute for Medical Research. 1987; No. 55.
15. Koekemoer LL, Kamau L, Hunt RH, et al. A cocktail polymerase chain reaction (PCR) assay to identify members of the *Anopheles funestus* (Diptera: Culicidae) group. Am J Trop Med Hyg. 2002;66:804–811.

16. Scott JA, Brogdon WG, Collins FH. Identification of single specimens of the *Anopheles gambiae* complex by the polymerase chain reaction. Am J Trop Med Hyg. 1993;49:520–529.

17. Knudsen JB, Pinder M, Jatta E, et al. Measuring ventilation in different typologies of rural Gambian houses: a pilot experimental study. Mal J. 2020;19:273.

18. Bal V, Gayasena V, Bibals R, et al. Modeling and experiments on release of metofluthrin from a thin cellulosic-polymer film. Chem Eng Res Des. 2017;118:31–40.

19. Haider N, Karlsson S. Migration and release profile of Chimassorb 944 from low-density polyethylene film (LDPE) in simulated landfills. Polym Degrad Stabil. 1999;64:321–328.

20. Njie M, Dilger E, Lindsay SW, et al. Importance of eaves to house entry by Anopheline, but not Culicine, mosquitoes. J Med Entomol. 2009;46:505–510.

21. Animut A, Balkew M, Lindtjørn B. Impact of housing condition on indoor-biting and indoor-resting *Anopheles arabiensis* density in a highland area, central Ethiopia. Mal J. 2013;12:393.

22. Ondiba IM, Oyieke FA, Ong’amo GO, et al. Malaria vector abundance is associated with house structures in Baringo County, Kenya. PLoS One. 2018;13:e0198970.

23. Lindsay SW, Jawara M, Mwesigwa J, et al. Reduced mosquito survival in metal-roof
houses may contribute to a decline in malaria transmission in sub-Saharan Africa.
Sci Rep. 2019;9:7770.

**Figure legends**

**Fig. 1** Typical brick houses in the study sites (A, metal-roof house; B, thatched-roof house), MSRD hung from ceiling in the metal-roof house (C) and in the thatched-roof house (D), and a micro data logger (Thermochron® Type G) for measuring room temperature (E).

**Fig. 2** Changes in the room temperature in metal-roof houses (A) and in thatched-roof houses (B) between January and April, 2015 and average day time (06:00-18:00) and night time (20:00-04:00) room temperature in metal-roof houses and thatched-roof houses (C).

**Fig. 3** Amount of remaining metofluthrin in the MSRD at 4 months after treatment in the metal-roof and thatched-roof houses (A) and relationships between the average overall room temperature and amount of remaining metofluthrin in the MSRD per a house (B). The linear regression formula and correlation coefficient for each roof type are shown in the graph.

**Fig. 4** Number of anopheline mosquitoes (*An. arabiensis*, *An. gambiae* s.s., and *An. funestus* s.s.) collected by five pyrethrum spray collections from December 4, 2015 to May 9, 2016 in the metal-roof and thatched-roof houses in Chiliko village without MSRD intervention and on December 3, 2015 in Chilore village before MSRD intervention (A) and number of mosquitoes collected in the metal-roof houses and the thatched-roof houses in Chilore village after two, three, and four months of the MSRD intervention (B).
| House No. | No. of Persons | Floor Area (m²) | <= 10 year | > 10 year | No. of MSRD / house | No. of MSRD / 10 m² | Roof Type |
|-----------|----------------|-----------------|------------|-----------|---------------------|---------------------|-----------|
| A002      | 1  2           | 28.16           |            |           | 8                   | 2.84                | Thatched  |
| A004      | 1  2           | 21.84           |            |           | -                   | -                   | Thatched  |
| A006      | 2  4           | 33.39           |            |           | -                   | -                   | Metal     |
| A008      | 4  2           | 19.84           |            |           | -                   | -                   | Thatched  |
| A010      | 2  2           | 19.89           |            |           | -                   | -                   | Thatched  |
| A018      | 2  2           | 33.92           |            |           | -                   | -                   | Thatched  |
| A018      | 2  2           | 19.8            |            |           | -                   | -                   | Thatched  |
| A021      | 3  2           | 32.66           |            | 10        | 3.06                | Metal               |
| A022      | 3  3           | 35.38           |            |           | -                   | -                   | Metal     |
| A023      | 1  2           | 14.4            |            |           | -                   | -                   | Metal     |
| A025      | 2  3           | 38.86           |            |           | -                   | -                   | Metal     |
| A026      | 2  2           | 38.08           |            | 11        | 2.89                | Metal               |
| A027      | 2  2           | 15.75           |            | 3         | 1.90                | Metal               |
| A030      | 0  2           | 23.22           |            | 7         | 3.01                | Thatched            |
| A032      | 0  2           | 20.01           |            | 6         | 3.00                | Metal               |
| A033      | 2  2           | 30.72           |            |           | -                   | -                   | Metal     |
| A036      | 1  2           | 30.24           |            | 8         | 2.65                | Metal               |
| A042      | 2  3           | 14.35           |            | 3         | 2.09                | Thatched            |
| A044      | 2  5           | 30              |            |           | -                   | -                   | Metal     |
| A046-1    | 2  2           | 12              |            | 4         | 3.33                | Metal               |
| A046-2    | 2  4           | 29.76           |            |           | -                   | -                   | Metal     |
| A047      | 1  5           | 37.52           |            | 8         | 2.13                | Metal               |
| A049      | 3  2           | 44.16           |            |           | -                   | -                   | Metal     |
| A052      | 1  3           | 18.32           |            |           | -                   | -                   | Thatched  |
| A053      | 2  3           | 16.47           |            | 3         | 1.82                | Thatched            |
| A062      | 0  2           | 37.24           |            | 11        | 2.95                | Metal               |
| A064      | 0  5           | 34.76           |            |           | -                   | -                   | Thatched  |
| A067      | 1  2           | 25.2            |            |           | -                   | -                   | Metal     |
| A070      | 2  3           | 38.54           |            | 8         | 2.08                | Thatched            |
| A072      | 0  2           | 19.8            |            | 4         | 2.02                | Thatched            |
| A076      | 0  3           | 15.66           |            |           | -                   | -                   | Thatched  |
| A080      | 1  2           | 29.67           |            | 6         | 2.02                | Metal               |
| A090      | 3  3           | 22.95           |            | 5         | 2.18                | Thatched            |
| A091      | 3  4           | 20.4            |            |           | -                   | -                   | Thatched  |
| A092      | 0  3           | 15.66           |            | 3         | 1.92                | Thatched            |
| A099      | 2  2           | 32.76           |            | 10        | 3.05                | Thatched            |
| A100      | 0  3           | 36.4            |            |           | -                   | -                   | Metal     |
| H41       | 1  2           | 18              |            |           | -                   | -                   | Metal     |
| H42       | 0  6           | 26.46           |            | 8         | 3.02                | Thatched            |
| H43       | 1  3           | 19.04           |            | 6         | 3.15                | Thatched            |
Table 2 Number of persons dwelling, total floor area, and type of roofs in the houses in Chiliko and Chilore villages.

| House No. | Village | No. of Persons | Floor Area (m²) | MSRD | Roof Type |
|-----------|---------|----------------|-----------------|------|-----------|
|           |         | <= 10 year | > 10 year |                  |          |           |
| A236      | Chiliko | 0         | 2          | 15.64 | -         | Thatched  |
| A251      | Chiliko | 2         | 5          | 14.52 | -         | Thatched  |
| A255      | Chiliko | 2         | 3          | 17    | -         | Thatched  |
| A266      | Chiliko | 2         | 2          | 15    | -         | Thatched  |
| A269      | Chiliko | 2         | 2          | 14.4  | -         | Thatched  |
| A270      | Chiliko | 1         | 3          | 36.57 | -         | Metal     |
| A271      | Chiliko | 1         | 2          | 20.4  | -         | Thatched  |
| A279      | Chiliko | 0         | 5          | 42.35 | -         | Metal     |
| A282      | Chiliko | 0         | 2          | 13.32 | -         | Thatched  |
| B581      | Chiliko | 0         | 1          | 14.62 | -         | Thatched  |
| B582      | Chiliko | 0         | 4          | 33.39 | -         | Metal     |
| B584      | Chiliko | 2         | 2          | 34.16 | -         | Metal     |
| B594      | Chiliko | 3         | 1          | 18.7  | -         | Thatched  |
| B595      | Chiliko | 2         | 5          | 30    | -         | Metal     |
| B600      | Chiliko | 1         | 4          | 14    | -         | Thatched  |
| B601      | Chiliko | 1         | 2          | 15.4  | -         | Thatched  |
| B604      | Chiliko | 1         | 2          | 18.5  | -         | Thatched  |
| B622      | Chiliko | 2         | 3          | 38.86 | -         | Metal     |
| B628      | Chiliko | 3         | 1          | 9     | -         | Metal     |
| B635      | Chiliko | 0         | 1          | 10.92 | -         | Thatched  |
| A293      | Chilore | 2         | 5          | 35    | 2 strips / 10 m² | Metal  |
| A299      | Chilore | 2         | 2          | 23.22 | 2 strips / 10 m² | Metal  |
| A305      | Chilore | 2         | 3          | 21.15 | 2 strips / 10 m² | Thatched  |
| A310      | Chilore | 0         | 2          | 17.68 | 2 strips / 10 m² | Thatched  |
| A320      | Chilore | 1         | 3          | 16.32 | 2 strips / 10 m² | Thatched  |
| A341      | Chilore | 2         | 4          | 33.28 | 2 strips / 10 m² | Thatched  |
| A349      | Chilore | 4         | 4          | 36.96 | 2 strips / 10 m² | Metal  |
| A350      | Chilore | 2         | 2          | 15.84 | 2 strips / 10 m² | Thatched  |
| A351      | Chilore | 0         | 2          | 17.86 | 2 strips / 10 m² | Thatched  |
| C001      | Chilore | Not available | Not available | Not available | 2 strips / 10 m² | Thatched  |
| C002      | Chilore | Not available | Not available | Not available | 2 strips / 10 m² | Metal  |
| C003      | Chilore | Not available | Not available | Not available | 2 strips / 10 m² | Thatched  |
Fig 2

A

Room Temperature (°C)

Date

B

Room Temperature (°C)

Date

C

Room Temperature (°C)

Day Time

Night Time

Thatched-roof house

Metal-roof house
Fig 4

A

No. of Collection / House / Day

Metal

Thatched

B

No. of Collection / House / Day

Metal

Thatched

Roof Type