

Jatta, Ebrima; Jawara, Musa; Bradley, John; Jeffries, David; Kandeh, Balla; Knudsen, Jakob B; Wilson, Anne L; Pinder, Margaret; D'Alessandro, Umberto; Lindsay, Steve W (2018) How house design affects malaria mosquito density, temperature, and relative humidity: an experimental study in rural Gambia. The Lancet. Planetary health, 2 (11). e498-e508. ISSN 2542-5196 DOI: https://doi.org/10.1016/s2542-5196(18)30234-1

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DOI: 10.1016/s2542-5196(18)30234-1

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Articles

How house design affects malaria mosquito density, temperature, and relative humidity: an experimental study in rural Gambia

Ebrima Jatta, Musa Jawara, John Bradley, David Jeffries, Balla Kandeh, Jakob B Knudsen, Anne L Wilson, Margaret Pinder, Umberto D'Alessandro, Steve W Lindsay

Summary

Introduction Unprecedented improvements in housing are occurring across much of rural sub-Saharan Africa, but the consequences of these changes on malaria transmission remain poorly explored. We examined how different typologies of rural housing affect mosquito house entry and indoor climate.

Methods Five typologies of mud-block houses were constructed in rural Gambia: four were traditional designs with poorly fitted doors and one was a novel design with gable windows to improve ventilation. In each house, one male volunteer slept under a bednet and mosquitoes were collected indoors with a light trap. Typologies were rotated between houses weekly. Indoor conditions were monitored with data loggers and the perceived comfort of sleepers recorded with questionnaires. We used pyschrometric modelling to quantify the comfort of the indoor climate using the logger data. Primary measurements were mean number of *Anopheles gambiae* and mean temperature for each house typology.

Findings In thatched-roofed houses, closing the eaves reduced *A gambiae* house entry by 94% (95% CI 89–97) but increased the temperature compared with thatched-roofed houses with open eaves. In houses with closed eaves, those with metal roofs had more *A gambiae*, were hotter $(1 \cdot 5^{\circ}C$ hotter [95% CI $1 \cdot 3 - 1 \cdot 7$]) between 2100h and 2300 h, and had 25% higher concentrations of carbon dioxide (211 $\cdot 1$ ppm higher [$117 \cdot 8 - 304 \cdot 6$]) than those with thatched roofs. In metal-roofed houses with closed eaves, mosquito house entry was reduced by 96% (91–98) by well fitted screened doors. Improved ventilation of metal-roofed houses made them as cool as thatched houses with open eaves. Metal-roofed houses with closed eaves were considered more uncomfortable than thatched ones with closed eaves. In metal-roofed houses, ventilated houses were more comfortable than unventilated houses before midnight, when people retired to bed.

Interpretation Closing the eaves reduced vector entry in thatched houses but increased entry in metal-roofed houses. Metal-roofed houses with closed eaves were, however, protected against malaria vectors by well fitted screened doors and were made comfortable by increasing ventilation. House designs that exclude mosquitoes and are comfortable to live in should be a priority in sub-Saharan Africa.

Funding Sir Halley Stewart Trust, Global Clinical Trials, and Global Challenges Research Fund.

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Introduction

The International Monetary Fund forecasts that Africa will have the world's second fastest growing economy by 2020.¹ Increased prosperity means families having more disposable income to spend on housing improvements. The improving economy has contributed to a quiet revolution in rural African housing, with traditional thatched-roofed houses gradually being replaced by more modern housing materials, particularly with metal roofs.² The scale of building new housing will be huge in Africa, where more than half of the anticipated growth in global population will occur between 2017 and 2050.³ Of the forecasted extra $2 \cdot 2$ billion people, $1 \cdot 3$ billion will be born in Africa.³ The implications of these unprecedented changes in rural housing on health have not been considered.

Since 80-100% of malaria transmission in sub-Saharan Africa occurs indoors.4 reduction of the entry of malaria vectors indoors should reduce the numbers of malaria cases. Indeed, evidence is growing that suggests modern housing provides greater protection against malaria vectors than traditional housing. A systematic review and meta-analysis² found that residents of modern houses were 47% less likely to contract malaria and had a 45-65% lower incidence of malaria than those living in traditional houses. However, a meta-analysis5 of malaria surveys carried out in 21 sub-Saharan Africa countries between 2008 and 2015 found that modern housing was associated with a 9-14% reduction in the odds of malaria infection compared with traditional housing. If modern housing is protective against malaria, which features of modern sub-Saharan African housing are protective?





Lancet Planet Health 2018; 2: e498–508

National Malaria Control Programme, Banjul, The Gambia (E Jatta BSc, B Kandeh MSc); Medical Research Council Unit The Gambia at the London School of Hygiene & Tropical Medicine, Banjul, The Gambia (M Jawara MSc, M Pinder PhD, D Jeffries PhD,

Prof U D'Alessandro MD): The Royal Danish Academy of Fine Arts, Schools of Architecture, Design and Conservation. Copenhagen, Denmark (J B Knudsen MAA); Department of Biosciences Durham University, Durham, UK; (A L Wilson PhD, M Pinder, Prof S W Lindsav PhD): and **Tropical Epidemiology Group** and Infectious Diseases Department, London School of Hygiene & Tropical Medicine. London, UK (J Bradley PhD, Prof U D'Alessandro. Prof S W Lindsay)

Correspondence to: Prof Steve W Lindsay, Department of Biosciences, Durham University, Durham DH1 3LE, UK s.w.lindsay@durham.ac.uk

Research in context

Evidence before this study

Despite 80-100% of malaria transmission in sub-Saharan Africa occurring indoors, few experimental studies have investigated how house design affects mosquito house entry and even fewer have considered how the design of rural houses in sub-Saharan Africa affects indoor climate. We searched Google Scholar using the terms "Anopheles gambiae" and ("architecture" or "house" or "indoors" or "indoor climate" or "comfort index") for articles published in any language between database inception and Dec 20, 2017. We also examined reference lists from review articles. Considerable evidence suggests that open eaves—ie, the gap between the top of the wall and the roof—are the major route by which A gambiae sensu lato, the major sub-Saharan African malaria vector, enters houses. Nowadays, however, modern houses are built with metal roofs with closed eaves and the traditional thatched-roofed houses with open eaves are less common. The consequences of these changes on house entry by malaria vector has not been quantified, and pilot studies on indoor climate in The Gambia and Tanzania suggest that modern housing, in which the eaves are closed and the roof is made of metal, is hotter and more uncomfortable than traditional housing.

Added value of this study

This experimental study is the first to use buildings of a size and structure similar to those constructed in rural parts of sub-Saharan Africa, which is important since the heating and cooling of houses is dependent on the mass and position of the different materials used for house construction. Ultimately, the typology of house shapes the way plumes of human odours are released from the house, which are used by mosquitoes to locate a human bloodmeal indoors, and makes a building comfortable or not for the human occupants. Rotating the typologies between houses each week allowed us to adjust for any variation in mosquito numbers that might arise from house position—eq, one house having high numbers of mosquitoes since it is positioned closer to aquatic habitats than the other houses. The replicated, Latin rectangle experimental design simultaneously guantified how four different building typologies common in rural Gambia and one novel house type, with a metal roof and increased ventilation, affected house entry by A gambiae sensu lato and indoor climate. Indoor climate is important for malaria control since although a perfectly sealed house might reduce mosquito house entry, it might be too hot for the occupants to use a bednet. We found that closing the eave gaps reduced mosquito house entry in thatched houses but increased entry in metal-roofed houses. This difference is important since most new houses in sub-Saharan Africa are constructed with metal roofs. We showed that screening the doors of metal-roofed houses reduced the number of vectors entering the house and improving ventilation, by inserting screened windows in the gable ends under the saddle roof, made the house cooler at night.

Implications of all the available evidence

Roll Back Malaria, UN Development Programme, and UN-Habitat's Housing and Malaria consensus statement asked what architectural features are protective against malaria. The available evidence shows clearly that closing the eaves of thatched-roofed houses will reduce malaria transmission, but that this modification is insufficient for metal-roofed houses because mosquitoes enter through gaps in poor quality doors. The use of well fitted screened doors, together with improved ventilation, should be encouraged. The unknown factor is the acceptability of the interventions and how local populations will use them. Building designs that include effective screening against mosquitoes and increase airflow in the house could help reduce malaria transmission and maintain areas free from malaria after elimination.

See Online for appendix

To date, little research has been done on this subject. Observational studies have found associations between reduced numbers of malaria vectors indoors and the presence of closed eaves, the gap between the top of the wall and the overhanging roof and ceilings,6 which has been supported by experimental studies.^{7,8} The findings show that open eaves are the major route by which Anopheles gambiae sensu lato enters houses and that closing them or installing a ceiling below the eaves prevents a substantial proportion of the vectors from entering the living space. A pilot study⁹ of modern houses in rural Tanzania showed that mosquito numbers indoors were 96% less in screened doublestorey buildings than outdoors, with corresponding values of 77% less in screened single-story buildings and 23% less in traditional houses. Thus, accumulating evidence suggests that mosquito house entry can be reduced appreciably by filling or screening entry points.

In rural Gambia, single-room houses have distinct typologies, which are also common in many parts of sub-Saharan Africa. Nowadays, most rural Gambian houses are constructed from sundried mud bricks, which became common during the mid-twentieth century. These traditional houses have two doors, front and back, one or two windows, or none at all, and have thatched roofs with open eaves (appendix). In the past 5-10 years, however, finding thatched-roofed houses with open eaves is increasingly more difficult in The Gambia since householders are closing them with mud, partly because of recommendations broadcast by the national malaria control programme to reduce the entry of mosquitoes and also because they think it prevents strong winds removing the roof. The most obvious change in housing stock, however, is the gradual replacement of thatched-roofed houses with metalroofed ones, most of which also have closed eaves.

Alongside the single-room houses are some multiroom line houses, which are also increasing in number and almost always have metal roofs. One problem with metal-roofed houses is that they are often hotter than thatched ones,10 and this increase in temperature might reduce the number of people sleeping under a longlasting insecticidal net (LLIN; Olyset, Sumitomo Chemical, Tokyo, Japan)1-one of the most effective malaria control tools currently available.¹² In this study, we assessed not only how local house designs affected mosquito ingress but also how mosquito screening could be used to protect occupants from mosquitoes and keep the house cooler. The costs of building alterations were also assessed. This study is the first to use an experimental design to quantify how different house typologies affect mosquito house entry and indoor climate. The conundrum to solve is one of two opposing forces, to maximise airflow in a house to keep the occupants cool and to keep mosquitoes out. Building homes that reduce malaria vector house entry might not only reduce malaria transmission but also, after elimination, when current interventions are scaled back, might help prevent malaria from returning.

Methods

Study design

This was an experimental study of five human-baited houses identical in size, shape, and walls. Each house was modified to represent different house typologies, which were rotated weekly between the different houses following a replicated Latin rectangle design (appendix). We did two experiments. The first experiment investigated the relative attractiveness of different housing typologies to mosquitoes and compared the indoor climates, and the second experiment re-examined the differences between metal-roofed and thatched-roofed houses, both with closed eaves and poorly fitted doors.

Study area

The study site was in Wellingara village (N 13° 33·365', W 14° 55·461') on the south bank of the River Gambia in Lower Fulladu West, Central River Region, The Gambia (appendix). This area is an open Sudanian savannah with extensive rice irrigation nearby. An intense rainy season runs from June to November, followed by a long dry season. High numbers of *A gambiae* sensu lato occur in the area during the rainy season,¹³ with *Anopheles coluzzii* predominating.¹⁴

Experimental houses

Five experimental houses positioned on a north–south axis were constructed 10 m apart on the western perimeter of the village close to the large irrigated Jahally-Pacharr rice fields to the west (appendix). Houses were the average size of a single-roomed house in rural Gambia, obtained from a survey of 400 randomly selected houses in the Upper River Region of The Gambia. The base of each house was $4 \cdot 20 \times 4 \cdot 20$ m and the $2 \cdot 20$ m high walls were constructed from sunbaked mud blocks (each 16 cm high $\times 20$ cm wide $\times 32$ cm long), with a front and back door on opposite sides, perpendicular to the line of houses, each 175 cm high and 75 cm wide. The only nontraditional building components were reinforced concrete ring-beams (20 cm high) on top of the wall, which were added to prevent the mud blocks cracking when the heavy roofs were moved between houses, and the metal profiles used to construct the roof frame.

The study was explained in the local language to male villagers and five healthy male volunteers aged older than 15 years who live in the local community provided signed-witnessed consent and were recruited to the study. When the volunteers were giving consent, emphasis was placed on the fact that if they fell ill during the study they would be treated at the local health clinic and treatment costs and transport would be reimbursed by the project. Each volunteer slept under an intact LLIN in each house from 2100 h to 0600 h, and a field assistant was posted throughout the night to ensure that the volunteers did not leave the huts except to use the bathroom. Mosquitoes were collected indoors with a US Centers for Disease Control and Prevention light trap, with the light bulb positioned 1 m above the mud floor next to the foot of the bed from 2100 h to 0600 h. The same man slept in the same house for the duration of the study, so the relative attractiveness of the person to mosquitoes was aliased with house position. Sleepers subjectively graded the night-time indoor climate as comfortable or uncomfortable. Mosquito collections were carried out for five nights each week for 5 weeks.

Experiment 1: differences between house typologies

Four house typologies common in The Gambia were included in the experiment, along with one novel design of a metal-roofed house, which was screened and ventilated. Houses with open eaves had a 3 cm gap between the top of the wall and the roof and those with closed eaves were blocked with a mixture of broken mud blocks and clay mortar. Traditional doors were constructed from a single panel of corrugate galvanised steel pinned to a wooden frame $(2 \times 2 \text{ cm})$. To simulate poorly fitted doors, which are common in villages, we made a 2 cm gap along the top and bottom of each door. Screened doors were made of 25 mm square steel profiles treated with anticorrosion paint. Two screened panels made from polyester netting $(2 \times 2 \text{ mm mesh}; \text{ appendix})$ were placed at the top and bottom of the door, each 75 cm wide and 60 cm high and fixed to the steel frame with flat bars and bolts. Each door was connected to the doorframe by a short bungee cord that made it self-closing, unlike the traditional doors. The ventilated metal-roofed house had screened doors and triangular screened windows (200 cm wide and 45 cm high at the apex), constructed with wooden frames $(2 \times 2 \text{ cm})$ and mosquito screening and were positioned in the gable ends of the building. Thatched roofs were pyramidal in

For more on the Sudanian savannah see https://vimeo.com/232447959 shape and 3.6 m high and metal roofs were saddle shaped and 3.1 m high. Thatched-roofed houses had a volume of 47.0 m^3 and metal-roofed houses had a 46.8 m^3 volume. Floors were beaten mud.

At the start of the experiment, we randomly allocated each typology using computer software to one of



Figure 1: House typologies

(A) Thatched roof, open eaves, and poorly fitted doors. (B) Thatched roof, closed eaves, and poorly fitted doors. (C) Thatched roof, closed eaves, and screened doors. (D) Metal roof, closed eaves, and poorly fitted doors. (E) Ventilated metal roof, closed eaves, and screened doors (designed by JBK).

	Permeability*, m ²	Cost†, US\$ 2016	Rationale for inclusion
Thatched roof; open eaves; traditional, poorly fitted doors	0.56	434-36	Traditional rural house (comparator)
Thatched roof; closed eaves; traditional, poorly fitted doors	0.06	460-06	Most common type of thatched house
Thatched roof; closed eaves; screened, well fitted doors	0.65	589.37	Traditional rural house with complete mosquito screening
Metal roof; closed eaves; traditional, poorly fitted doors	0.06	540.39	Most common type of house in rural Gambia
Metal roof with ventilation in gables; closed eaves; screened, well fitted doors	2.90	672.05	Modified modern house with mosquito screening and increased ventilation

*Permeability refers to the cross-sectional area open for airflow. Screening was assumed to reduce airflow by 71%. +Cost includes parts and labour.

Table 1: Typologies of houses

five identical partially-built houses, each consisting of a mud-walled building with no doors, windows, or roof. Each house was then completed according to the typology selected by randomisation for week one (appendix). The typologies rotated weekly between houses replicating a Latin rectangle design—ie, the houses were partially deconstructed and reconstructed at the end of each week. At the end of the 5-week study, each of the five typologies had been tested in each of the five experimental houses, which allowed us to measure the effect of the different typologies adjusting for geographical position of the house.

Experiment 2: differences between metal-roofed and thatched-roofed houses with closed eaves

In experiment 1 we were surprised to find more mosquitoes in closed-eave houses with metal roofs than thatched ones. Therefore, we repeated the experiment and examined whether the hotter temperatures in metalroofed houses increased carbon dioxide production, a major mosquito attractant.¹⁵ In experiment 2, we made three changes to experiment 1. First, we enrolled two human participants to sleep in each house, since most Gambian houses have more than one occupant. Second, we fitted two metal windows on the same sides as the doors, 1.6 m above the ground, measuring 30×30 cm, with 1 cm gaps at the top and bottom of each one. Third, we measured carbon dioxide concentrations indoors. The primary purpose of experiment 2 was to compare the concentration of carbon dioxide in both house typologies. To avoid repetition, we only describe the comparison between closed-eave houses with metal-roofed and thatched-roofed houses here.

Outcomes

The primary outcomes measured were mean number of A gambiae sensu lato in the light trap at night and mean temperature for each house typology. In both experiments, indoor temperature and relative humidity were measured every 30 min with data loggers (Tiny tag, TGU 4500) positioned 1 m from the floor in the centre of the room. Outdoor temperature, relative humidity, wind speed, and wind direction were recorded with an automatic weather station (MiniMet, Skye Instruments, Llandrindod Wells, UK) positioned next to the houses. Thermal images were captured with a thermal imaging camera (FLIR ONE). Carbon dioxide concentrations were measured indoors in experiment 2 with data loggers (Onset Hobo MX1102) at 15 min intervals throughout the night from 2100 h to 0600 h for 25 nights, with the two loggers being moved between houses on alternate nights to reduce recording bias because of one logger recording consistently higher or lower than the other. All environmental measurements were made when the men were resident in the houses at night. Each morning during experiment 1, the volunteers were asked to score whether they had been comfortable or uncomfortable during the night.

Statistical analysis

For analyses, apart from psychrometric analysis, we used IBM SPSS Statistics 20 and Stata version 14. We estimated the sample size via simulation based on data from an experimental hut trial done in the same area,⁷ where the mean number of *A gambiae* sensu lato collected indoors over a 25-night study was 10.8 mosquitoes per house per night (SD 8.7). From the simulation, a 5×5 Latin square was powered to detect an intervention that reduced the number of mosquitoes collected in light traps by at least 50% at the 5% level of significance and 80% power.

We based the cost of materials and construction of each house typology on actual payments made to local suppliers and builders in 2016, converted from Gambian Dalasi to US dollars (42.61 GMD=US\$1; Sept 1, 2016). The costs included materials and labour and excluded transportation costs. We calculated the permeability of each typology from the area of entry points in the building—ie, including gaps in the eaves, around the doors, and screening. We assumed that screening doors and windows would reduce airflow by 64%.¹⁶ In experiment 1, we assessed the effect of house typology on mosquito house entry and indoor climate using generalised linear modelling, using a negative binomial model with a log link function for count data and a normal distribution with identity link for continuous variables. In addition to house typology, we included house position and day in the model as fixed effects. For experiment 2, we used generalised linear modelling to compare mosquito numbers between typologies. We used polar plots to depict the direction and strength of the wind during the day and night. We used linear regression to explore the relationship between carbon dioxide concentration and covariates.

We analysed human comfort in two ways. First, we summed the number of nights people were comfortable and uncomfortable for each type of housing typology from their questionnaire answers. Second, we assessed human comfort using the software package LadyBug (LadyBug Products, Athol, ID, USA), which was used to estimate the proportion of time occupants of various house typologies spent in the so-called comfort zone.^{*v*}

	Anopheles gar	nbiae females pe	er night		Other female mosquitoes per night				
	Mean	Mean ratio	Effect estimate	p value	Mean	Mean ratio	Effect estimate	p value	
Thatched roof; open eaves; traditional, poorly fitted doors	10·4 (3·8 to 17·0)	Reference			47·2 (36·0 to 58·4)	Reference			
Thatched roof; closed eaves; traditional, poorly fitted doors	0·7 (0·0 to 1·4)	0·06 (0·03 to 0·11)	-94% (89 to 97)	<0.001	36·8 (22·8 to 50·9)	0·57 (0·42 to 0·77)	-43% (23 to 58)	<0.001	
Thatched roof; closed eaves; screened, well fitted doors	0·4 (0·1 to 0·7)	0·04 (0·02 to 0·09)	-96% (91 to 98)	<0.001	6·2 (3·6 to 8·9)	0·11 (0·07 to 0·16)	-89% (84 to 93)	<0.001	
Metal roof; closed eaves; traditional, poorly fitted doors	17·2 (11·0 to 23·3)	2·99 (1·96 to 4·57)	+199% (96 to 357)	<0.001	66·2 (50·4 to 82·1)	1·21 (0·90 to 1·64)	+21% (-10 to 64)	0.204	
Metal roof with ventilation in gables; closed eaves; screened, well fitted doors	0.8 (0.0 to 1.6)	0·06 (0·03 to 0·10)		<0.001	4·6 (2·1 to 7·1)	0.07 (0.04 to 0.10)		<0.001	
Data calculated with 95% CIs. General linearised modelling results, adjusted for house position and night.									

Table 2: Mosquito house entry in different typologies of housing

Maximum temperature			Average temperature 2100–2330 h			Average temperature 0000–0600 h		
Average	Difference from traditional house	p value	Average	Difference from traditional house	p value	Average	Difference from traditional house	p value
33·5°C (32·8 to 34·2)			27·2°C (26·6 to 27·7)			25·2°C (24·7 to 25·8)		
33·0°C (32·9 to 33·2)			32·0°C (31·8 to 32·1)			29·3°C (29·2 to 29·4)		<0.0001
33·3°C (33·2 to 33·5)	0·3 (0·1 to 0·5)	0.009	32·5°C (32·3 to 32·5)	0·5 (0·3 to 0·6)	<0.0001	29·7°C (29·6 to 29·9)	0·4 (0·2 to 0·6)	<0.0001
33·0°C (32·8 to 33·1)	0·0 (-0·3 to 0·2)	0.649	31∙5°C (31∙4 to 31∙7)	-0·4 (-0·6 to -0·24)	<0.0001	28·9°C (28·8 to 29·0)	-0·4 (-0·6 to -0·2)	<0.0001
34·7°C (34∙6 to 34∙8)	1·7 (1·5 to 1·9)	<0.0001	33·4°C (33·3 to 33·6)	1·5 (1·3 to 1·7)	<0.0001	30·2°C (30·1 to 30·4)	0·9 (0·7 to 1·1)	<0.0001
33·7℃ (33·6 to 33·9)	0·7 (0·5 to 0·9)	<0.0001	32·0°C (31·9 to 32·2)	0·1 (-0·1 to 0·30)	0.383	29·0°C (28·8 to 29·1)	-0·4 (-0·5 to -0·2)	0.0005
Data calculated with 95% CIs. General linearised modelling results, adjusted for house position and night.								
	Maximum tem Average 33.5°C (32.8 to 34.2) 33.0°C (32.9 to 33.2) 33.3°C (33.2 to 33.5) 33.0°C (32.8 to 33.1) 34.7°C (34.6 to 34.8) 33.7°C (33.6 to 33.9)	Maximum temperature Average Difference from traditional house 33.5°C (32.8 to 34.2) 33.0°C (32.9 to 33.2) 33.3°C 0.3 (33.2 to 33.5) (0.1 to 0.5) 33.0°C 0.0 (32.8 to 33.1) (-0.3 to 0.2) 34.7°C 1.7 (34.6 to 34.8) (1.5 to 1.9) 33.7°C 0.7 (33.6 to 33.9) (0.5 to 0.9)	Maximum temperature Average Difference from traditional house p value traditional house 33.5°C (32.8 to 34.2) 33.0°C (32.9 to 33.2) 33.3°C 0.3 0.009 (32.2 to 33.5) (0.1 to 0.5) 33.0°C 0.00 0.649 (32.8 to 33.1) (-0.3 to 0.2) 34.7°C 1.7 <0.0001	Maximum temperature Average temperature Average Difference from traditional house p value Average 33.5°C 27.2°C (32.8 to 34.2) 27.2°C 33.0°C 22.0°C (32.9 to 33.2) 32.0°C 33.3°C 0.3 0.009 32.5°C (32.2 to 33.5) (0.1 to 0.5) (32.3 to 32.5) (3.1 co 13.5) (0.1 to 0.5) (32.3 to 32.5) (3.2 to 33.5) (0.1 to 0.5) (31.4 to 31.7) (32.8 to 33.1) (-0.3 to 0.2) (31.4 to 31.7) (34.7°C 1.7 <0.0001	Maximum temperature Average temperature 2100-2330 Average Difference from traditional house p value Average Difference from traditional house 33.5°C 27.2°C 33.0°C 27.2°C 33.0°C 27.2°C 33.0°C 32.0°C 33.3°C 0.3 0.009 22.5°C 0.5 (32.2 to 33.5) (0.1 to 0.5) 32.9°C 0.5 (32.3 to 33.5) (0.1 to 0.5) 31.5°C -0.4 (32.8 to 33.1) (-0.3 to 0.2) 31.5°C -0.4 (32.8 to 33.1) (-0.3 to 0.2) 33.4°C 1.5 (34.4 to 31.4%) (1.5 to 1.9) 33.4°C 1.5 (34.6 to 34.8) (1.5 to 1.9) 33.4°C 0.1 33.7°C 0.7 <0.0001	Maximum temperature Average temperature 2100-2330 h Average Difference from traditional house p value Average Difference from traditional house p value 33.5°C 27.2°C 33.6°C 27.2°C 33.0°C 27.2°C 33.0°C 27.2°C 33.0°C (26.6 to 27.7) 33.3°C 0.3 0.009 32.0°C 33.3°C 0.3 0.009 32.5°C 0.5 <0.0001	Maximum temperature Average temperature 2100-2330 h Average temperature 2100 h Average temperature 2100 h Average temperature 2100 h Average 220 h Average temperature 2100 h Average temperature 2100 h Average 220 h <td>Maximum temperature Average temperature 2100-2330 h Average temperature 0000-0600 Average Difference from traditional house p value P val</td>	Maximum temperature Average temperature 2100-2330 h Average temperature 0000-0600 Average Difference from traditional house p value P val

The comfort zone is defined by the comfort polygon for temperature and relative humidity and provides an estimated proportion of people satisfied with the indoor climatic comfort. The human energy balance model used by the psychrometric chart is the predicted mean vote model developed by P O Fanger.¹⁸ The predicted mean vote model is a seven-point scale from cold (–3) to hot (+3) that is used in comfort surveys. Each integer value of the scale indicates cold, –3; cool, –2; slightly cool, –1; neutral, 0; slightly warm, +1; warm, +2; and hot, +3. The accepted range of comfort is a predicted mean vote between –1 and +1 and defines the area of the comfort polygon on the psychrometric chart. We assumed that from 2100 h to 0000 h men were seated and quiet and wore thin straight trousers, briefs, and



Figure 2: Mean diurnal temperature (A) and relative humidity (B) in different typologies of housing and outdoors

1, thatched roof, open eaves, and poorly fitted doors; 2, thatched roof, closed eaves, and poorly fitted doors; 3, thatched roof, closed eaves, and screened doors; 4, metal roof, closed eaves, and poorly fitted doors; 5, ventilated metal roof, closed eaves, and screened doors; 6, automatic weather station. T shirts.¹⁹ From 0000 h to 0600 h the men were reclining or sleeping. χ^2 tests were used for making comparisons between the proportions of time spent comfortable or uncomfortable, as predicted from the psychometric modelling. For each typology, we calculated the proportion of time the indoor climate was in the comfort zone for two periods: 2100–0000 h, when people retire to bed, and 0000–0600 h, when people are usually sleeping. In experiment 2, we fitted a mixed effects regression model to each response to allow for random effects (experiment) and fixed effects (position and data logger).

The study was approved by the Gambia Government and Medical Research Council's joint ethics committee (May 16, 2016, and March 16, 2017) and the Department of Biosciences ethics committee, Durham University, UK (May 13, 2016, and June 29, 2017).

Results

In experiment 1, from Sept 18, 2016, to Oct 21, 2016 (rainy season), a total of 4762 female mosquitoes were collected in the five house typologies (figure 1; table 1; appendix) over 25 nights (n=125 nights): 2857 (60.0%) were Culex spp, 1088 (22.8%) were Mansonia spp, 734 (15.4%) were A gambiae sensu lato, and 83 (1.7%) were other anophelines. Of the 728 A gambiae sensu lato that were identifiable by PCR, 460 (63 · 2%) were Anopheles arabiensis and 268 (36.8%) were A coluzzii. A breakdown of mosquito species by typology is shown in the appendix.

Closing the eaves of a thatched house with poorly fitting doors resulted in 94% (95% CI 89-97) fewer A gambiae sensu lato collected indoors, with a 43% (23-58) reduction in other mosquito species compared with a thatched house with open eaves (table 2). A similar reduction in A gambiae sensu lato was achieved when the doors were screened and the eaves closed, irrespective of whether the roof was thatched or made of metal. With screened doors, the number of other mosquito species collected indoors reduced by 89-93% compared with thatched-roofed houses with open eaves. The number of mosquitoes collected in metal-roofed houses with poorly fitting doors was three times greater than thatched-roofed houses with open eaves and poorly fitted doors (mean ratio 2.99 [95% CI 1.96-4.57]; p<0.001)—in other words, closing the eaves of metal-roofed houses increased mosquito house entry if the doors were not fitted.

Permeability strongly influences air flow in a building and varied markedly between housing typologies (table 1). The greatest permeability was in the ventilated metal-roof house, which was 85% greater than the permeability in the thatched-house with open eaves. Closing the eaves in houses with poorly fitted doors reduced the permeability by 89% compared with the traditional thatched-roofed house with open eaves, whereas fitting screened doors in houses with closed

Mean relative humidity			Average relative humidity 1900–2330 h			Average relative humidity 0000–0600 h		
Average	Difference from traditional house	p value	Average	Difference from traditional house	p value	Average	Difference from traditional house	p value
74·5% (71·7 to 77·4)			82·9% (77·8 to 88·1)			87·4% (82·1 to 92·7)		
86·1% (85·6 to 86·6)			76·7% (76·0 to 77·5)			80·2% (79·6 to 80·8)		
86·3% (85·8 to 86·8)	0·2 (-0·5 to 0·9)	0.552	77·3% (76·6 to 78·0)	0·6 (-0·5 to 1·6)	0.268	80∙9% (80∙3 to 81∙5)	0·7 (-0·2 to 1·5)	0.125
87·2% (86·7 to 87·7)	1·1 (0·4 to 1·8)	0.004	78·1% (77·4 to 78·8)	1·3 (0·3 to 2·4)	0.012	82·0% (81·4 to 82·6)	1·8 (1·0 to 2·7)	0.000
88·3% (87·8 to 88·8)	2·2 (1·5 to 3·0)	<0.0001	78·6% (77·8 to 79·3)	1·8 (0·8 to 2·9)	0.001	83·9% (83·3 to 84·5)	3·7 (2·9 to 4·6)	<0.000
87·8% (87·3 to 88·3)	1·7 (1·0 to 2·4)	<0.0001	77·8% (77·1 to 78·5)	1·1 (0·0 to 2·1)	0.044	82·9% (82·3 to 83·5)	2·7 (1·9 to 3·6)	<0.000
	Mean relative h Average 74-5% (71-7 to 77-4) 86-1% (85-6 to 86-6) 86-3% (85-8 to 86-8) 87-2% (86-7 to 87-7) 88-3% (87-8 to 88-8) 87-8% (87-3 to 88-3)	Mean relative humidity Average Difference from traditional house 74-5% (71-7 to 77-4) 86-1% (85-6 to 86-6) 86-3% 0-2 (85-8 to 86-8) (-0-5 to 0-9) 87-2% 1-1 (86-7 to 87-7) (0-4 to 1-8) 88-3% 2-2 (87-8 to 88-8) (1-5 to 3-0) 87-8% 1-7 (87-3 to 88-3) (1-0 to 2-4)	Mean relative humidity Average Difference from traditional house p value 74-5% 74-5% 74-5% 86-1% 86-1% 86-3% 0-2 0-552 86-3% (-0-5 to 0-9) 87-2% 87-2% 1.1 0.004 86-7 to 87-7) (0-4 to 1.8) 88-3% 88-3% 2-2 <0.0001	Mean relative humidity Average relative Average Difference from traditional house p value Average 74-5% p value Average 74-5% 82-9% (71-7 to 77-4) (77-8 to 88-1) (77-8 to 88-1) 86-1% 76-7% (85-6 to 86-6) (76-0 to 77-5) (76-6 to 78-0) 86-3% 0-2 0-552 77-3% (85-8 to 86-8) (-0-5 to 0-9) (76-6 to 78-0) (76-6 to 78-0) 87-2% 1-1 0-004 78-1% (86-7 to 87-7) (0-4 to 1-8) (77-4 to 78-8) 88-3% 2-2 <0-0001	Mean relative humidity Average relative humidity 1900-2 Average Difference from traditional house P value Average Difference from traditional house 74-5% 82-9% 74-5% 82-9% 74-5% 82-9% 74-5% 76-7% 86-1% 76-7% 86-3% 0-2 0-552 77-3% 0-6 (75-6 to 78-0) (-0-5 to 1-6) 78-64 1-3 86-7 to 87-7) (0-4 to 1-8) (77-4 to 78-8) (0-3 to 2-4) 88-3% 2-2 <0-0001	Mean relative humidity Average relative humidity 1900-2330 h Average Difference from traditional house p value 74-5% Average Difference from traditional house p value 74-5% 82-9% 74-5% 82-9% 86-1% 76-7% 86-1% 76-7% 86-3% 0-2 0-552 77-3% 0-6 0-268 (76-6 to 78-0) (-0-5 to 1-6) 76-78 87-2% 1-1 0-004 78-1% 1-3 0-012 (77-4 to 78-8) (0-3 to 2-4) 78-6% 1-8 0-001 88-3% 2-2 <0-0001	Mean relative humidity Average relative humidity 1900-2330 h traditional house Average relative humidity 1900-2330 h Average Average relative humidity 1900-2330 h traditional house Average relative Average Difference from p value traditional house Average Difference from p value traditional house Average Difference from p value Average Average <t< td=""><td>Mean relative humidity Average relative humidity 1900-2330 h Average relative humidity 0000-06 Average Difference from traditional house p value Average Difference from traditional house Iverage Difference from traditional house Iverage<!--</td--></td></t<>	Mean relative humidity Average relative humidity 1900-2330 h Average relative humidity 0000-06 Average Difference from traditional house p value Average Difference from traditional house Iverage Difference from traditional house Iverage </td

eaves only increased permeability by 150% relative to the thatched house with open eaves.

Closing the eaves of a thatched-roofed house increased the indoor temperature by 0.5°C (95% CI 0.3-0.6; p<0.0001) from 2100 h to 2330 h and 0.4°C (0.2-0.6; p<0.0001) from 0000 h to 0600 h compared with the traditional thatched-roofed house and open eaves (table 3). Temperatures could be made cooler than a house with open eaves by adding the screened doors to a thatchedhouse with closed eaves. Metal-roofed houses with closed eaves and doors with gaps were 1.5°C (95% CI 1.3-1.7; p<0.0001) hotter than traditional thatched-roofed houses from 2100 h to 2330 h but could be made cooler than the traditional houses by screening the doors and windows of metal-roofed houses. All indoor temperatures were at their lowest shortly after sunrise but increased as the sun began to heat the house and peaked at 1900 h, around sunset (figure 2A). During the night, indoor temperatures declined progressively in all house typologies. The daily pattern of indoor temperatures differed to those outdoors. First, from a minimum at dawn, indoor temperatures rose far more slowly than outdoor temperatures. Second, they peaked in the late evening, several hours later than outdoor temperatures. A thermal image of a metal-roofed house (appendix) taken in the evening shows the hot walls and cool roof, illustrating the different heat capacities of the two materials.

The mean relative humidity of thatched-roofed houses with open eaves was lower than that in the other typologies of housing (table 4). In all typologies, the daily pattern of indoor relative humidity was the opposite of that seen with temperature, with the relative humidity declining during the heat of the day and rising slowly after sunset through the night and until the first few hours after sunrise. During the day, outdoor relative humidity was considerably lower than that indoors, but at night relative humidity outdoors was higher than that indoors (figure 2B). The predominant wind direction in the study site came from the west, with wind strength dropping substantially at night (appendix). Thus, since the major aquatic habitats in the vicinity are to the west of the houses, most mosquitoes will travel with the wind and enter through the side of the house facing away from the village.

Metal-roofed houses with closed eaves were considered uncomfortable by volunteers on 14 (56%) occasions of 25 and were significantly more uncomfortable than thatched-roofed houses with closed eaves, which were deemed uncomfortable on two (8%) occasions (χ^2 =13·2; p<0·001). All other house typologies were considered comfortable by volunteers on every night of the study. Volunteers were uncomfortable 14 times at higher temperatures (34·3°C [95% CI 33·4–35·1] at entering the house at 2100 h) and were comfortable 96 times at lower temperatures (32·8°C [32·5–33·0] at entering the house at 2100 h; *t* test –3·769; temperature difference 1·5°C [0·7–2·3]; p<0·001).

Analysis of psychrometric data (figure 3) showed that all houses cooled during the night; therefore, after midnight sleepers might need a covering sheet to stay comfortable. Between 2100 h and 0000 h, when people are preparing to sleep, the metal-roofed houses with closed eaves were more uncomfortable, because of the higher temperatures, than the thatched-roofed houses with open eaves ($\chi^2=22.86$; p<0.001). Conversely, from 0000 h to 0600 h, when most people sleep, the metal-roofed houses were more comfortable since they were warmer than the thatched-houses with open eaves (χ^2 =19.03; p<0.001). In the early morning both screened houses tended to be cooler than the thatchedroofed house with open eaves. The ventilated metalroofed house was more comfortable than the unventilated metal-roofed house before midnight ($\chi^2 = 12.64$; p<0.001), but it was less comfortable later from 0000 h to 0600 h (χ^2 =39·13, p<0·001) because it was cooler during both periods.



Figure 3: Psychrometric charts from 2100 h to 0000 h (A) and 0000 to 0600 h (B) for different typologie of housing

1, thatched roof, open eaves, and poorly fitted doors; 2, thatched roof, closed eaves, and poorly fitted doors; 3, thatched roof, closed eaves, and screened doors; 4, metal roof, closed eaves, and poorly fitted doors; 5, ventilated metal roof, closed eaves, and screened doors. Points lying inside the polygon are deemed comfortable, whereas those outside are uncomfortable. The percentage represents the proportion of time each typology is comfortable. Clothing rate is the thermal resistance from the skin to the outer surface of the clothed body.¹⁶

> The cost of constructing a metal-roofed house was over US\$100 more than a traditional thatched-roofed house (table 1). A thatched-roofed house with open eaves cost \$210 in materials and \$224 for labour. Filling the eaves with mud cement and broken bricks cost an additional \$26 and the two metal-screened doors cost \$129. A metal roof costs \$80 more than a thatched roof. The cost of a ventilated metal-roof house is \$132 more expensive than an unventilated house due to the cost of the two screened doors and two screened windows. The cost of labour



Figure 4: Changes in carbon dioxide concentration in thatched-roofed (A) and metal-roofed (B) houses with closed eaves

Trend lines shown for the nights data were recorded. CO₂=carbon dioxide.

varied between 39% and 53% of building costs, thus the price of building could be reduced if the owners built the house themselves or used cheaper labour.

In experiment 2, from Sept 4, 2017, to Oct 8, 2017 (rainy season), a total of 3685 female mosquitoes were collected in two house typologies over 25 nights (n=50 nights): 2360 ($64 \cdot 0\%$) were *Mansonia* spp, 819 ($22 \cdot 2\%$) were *Culex* spp, 436 ($11 \cdot 8\%$) were *A gambiae* sensu lato, and 70 ($1 \cdot 9\%$) were other anophelines.

Greater numbers of female *A gambiae* sensu lato were collected in metal-roofed houses each night (mean number of mosquitoes per house per night was $11\cdot0$ [95% CI $7\cdot9-14\cdot1$]) than thatched-roofed houses ($6\cdot4$ [$3\cdot7-9\cdot1$]; odds ratio [OR] $2\cdot51$ [95% CI $1\cdot54-4\cdot10$]; p<0 $\cdot001$). The same was also true for other species of mosquito, with

more caught in metal-roofed houses $(75 \cdot 3 \ [53 \cdot 3 - 97 \cdot 3])$ than thatched ones $(54 \cdot 6 \ [35 \cdot 8 - 73 \cdot 4]; OR 1 \cdot 42 \ [1 \cdot 07 - 1 \cdot 90]; p=0 \cdot 016)$.

The shapes of the carbon dioxide concentration profiles over time were highly variable (figure 4). Despite the shape variability—ie, the gradient of concentration increase between 2100 h and 2300 h—the magnitude and time to peak concentration were consistent features and were selected as summary statistics for each profile (table 5). Evidence suggests that carbon dioxide accumulated more rapidly (1.9 ppm/min [95% CI 1.3-2.5]) and to higher concentrations in metal-roofed houses (211.1 ppm greater [117.8–304.6]) than in thatched-roofed houses (p<0.0001). The time taken to reach peak carbon dioxide concentration was 73.7 min (95% CI -1.9 to 149.3) slower in metal-roofed houses than thatched ones, although this difference was of borderline significance (p=0.056).

Discussion

The number of A gambiae sensu lato mosquitoes entering houses of different typologies varied greatly and thus the difference in the risk of malaria transmission could also be large. Importantly, this study highlights two ways of reducing the number of malaria mosquitoes entering houses with thatched or metal roofs. First, the simplest and cheapest method is to close the eaves with mud mortar; if large gaps are present, close with broken mud blocks and mortar. Thatched-roofed houses with closed eaves and poorly fitted doors had 94% fewer A gambiae sensu lato than those with open eaves and fewer than metal-roofed houses with closed eaves and poorly fitted doors. This difference is important since most modern houses in rural areas have metal roofs, closed eaves, and poorly fitted doors. Nonetheless, the consequence of closing the eaves of thatched houses is that indoor temperature will increase 0.4-0.5°C during the night, making them more uncomfortable than the traditional thatched houses with open eaves. Second, providing the eaves are closed, well fitted screened doors will reduce substantially the entry of A gambiae sensu lato by 94-96%, provided the doors are also kept closed during the evening and night. This outcome supports the results of an earlier randomised controlled trial²⁰ in The Gambia, showing that closing the eaves and screening the doors of thatched houses reduced the number of A gambiae sensu lato entering village houses by 59% and supports the results of a study in Ethiopia,²¹ showing that screening reduced indoor densities of A arabiensis by 40%. In practise, the best option is to combine closing the eaves with screened doors because this house design not only kept out mosquitoes but made the houses cooler at night than the traditional thatched houses with open eaves.

Although closing the eaves will protect occupants from malaria vectors, it is not so effective at reducing nuisance biting from other species of mosquito, as has been reported previously, since these mosquitoes are more

	House typology		Adjusted difference between typologies, ppm (95% CI)	p value	
	Thatched-roofed house	Metal-roofed house	-		
Median CO2 gradient from 2100 h to 2300 h, ppm/min	1.68 (0.74 to 2.30)	3·41 (2·26 to 4·25)	1·9 (1·3 to 2·5)	p<0.0001	
Median CO_2 concentration, ppm	847 (735 to 940)	1026 (888 to 1160)	211·1 (117·8 to 304·6)	p<0.0001	
Median time of maximum CO_{2} concentration, h	23·45 (22·45 to 02·40)	01·20 (23·48 to 04·44)	73·7* (-1·9 to 149·3)	0.056	

Data are median (IQR), unless otherwise specified. Adjusted differences are from a formal regression analysis, fitting a linear mixed model, separately for each of the summary statistics with experiment night as a random effect, which removed any influence of night to variation, such as temperature or humidity. The effect of roof type was adjusted for house position, data logger, and within-day differences. CO₂=carbon dioxide. *Min rather than ppm.

Table 5: Differences in CO₂ concentrations between thatched-roofed and metal-roofed houses

likely to enter through doors and windows.⁸ Closing the eaves will reduce the number of nuisance mosquitoes by only 43%, and since in experiment 1 over 84% of houseentering mosquitoes were nuisance biters, closing the eaves will not dramatically reduce overall indoor biting. Screened doors, however, will suppress the number of nuisance mosquitoes entering houses by 89–93%, marking a substantial reduction in house entry.

Of houses with closed eaves and poorly fitted doors, those with metal roofs had far more mosquitoes than those with thatched roofs, a finding from experiment 1 that was supported by experiment 2. At least two explanations are possible for this finding. First, we found that the maximum concentration of carbon dioxide in houses with metal roofs was 25% higher than in thatched-roof houses. Since carbon dioxide is a major mosquito attractant,15 the greater concentrations of carbon dioxide, and perhaps other host odours, emanating from the metal-roofed houses will attract host-seeking mosquitoes from further distances than thatched-roofed houses. Second, the metal-roofed houses are hotter than thatched-roofed houses, which will increase sweating rates of people indoors and might account for the faster rates of carbon dioxide accumulation observed when men enter the house and the raised concentrations of carbon dioxide found in metal-roofed houses. Sweating in naked adult men at rest commences at air temperatures of 26-33°C, depending on the part of the body, and rises sharply above 30°C.²² These temperatures are precisely what the volunteers experienced in the experimental houses before midnight. For example, the rate of sweating in a resting individual in humid environments will double from 0.4 L/h at 27°C to 0.8 L/h at 35°C.23 Increased sweating is likely to increase the production of volatiles that mosquitoes use for host location.24,25

All typologies of houses were several degrees warmer at night than outdoors because of the high thermal mass of the thick mud blocks used for building the walls (appendix). Mud walls heated by sunlight during the day radiate heat at night, with the roof and walls trapping the hot air in the house. Outdoors, as night falls, the air temperature is cooled by the absence of solar radiation heating the ground, air movement, and by the often torrential rains that punctuated the study period. The daily pattern of indoor relative humidity was the opposite of that seen with temperature, with the relative humidity declining during the heat of the day and rising slowly after sunset through the night and until the first few hours after sunrise. Overall, the relative humidity outdoors was considerably lower than in houses during the day, whereas the opposite was true at night. The relative humidity of traditional thatched-roofed houses was lower than in other typologies of housing, but although this result was significant a difference of a few percentage points is unlikely to be of biological significance.

A traditional rural Gambian house leaks hot air from early evening throughout the night, carrying with it volatiles like carbon dioxide and human skin emanations, which mosquitoes use for host location.24,25 In essence, the house acts as a large concentrated attractant. The house is likely to make it easier for a mosquito to detect its next human-blood meal if a person is indoors rather than outdoors, since air expands when hot and will travel further than denser cooler air, making a person in the house detectable from further away than if they were outdoors. Mosquitoes might also be better able to detect a house at night when the wind strength is less than that during the day (appendix). The importance of wind direction in mosquito host location is uncertain since evidence suggests that host location can be downwind or upwind,13,26,27 although most mosquitoes are generally considered to fly upwind.28 In this study, the experimental houses were located along a north-south axis with major aquatic habitats being the extensive irrigated rice fields to the west. Polar plots show that the predominant wind direction at night is from the west (appendix), suggesting that in this case mosquitoes disperse with the wind, but they will also move up the turbulent airborne odour plumes emanating from the houses. We have also reported a similar finding in a neighbouring village.13 The rising concentrations of carbon dioxide that occur after people move indoors at night are likely to affect profoundly host location by A gambiae. Our study shows that it takes several hours for carbon dioxide to reach its highest concentration, so when people go to bed before midnight maximum concentrations of carbon dioxide in village houses might not be reached until after midnight. In The Gambia,29 and for many other parts of sub-Saharan Africa,30 more A gambiae bite indoors after midnight than before midnight. Therefore, carbon dioxide production indoors might partly explain the nocturnal biting pattern of this malaria vector.

The metal-roofed houses that are replacing thatchedroofed houses across much of sub-Saharan Africa are considerably hotter than thatched-roofed houses during the day. This difference in temperature is due to the heating effect of the metal roof and the much reduced permeability resulting from closing the eaves. Although hotter temperatures might increase the mortality of mosquitoes resting indoors, these houses are also uncomfortable for residents and are likely to lead to fewer people using their nets at night¹¹ and more choosing to sleep outside. We show here that ventilating metal-roofed houses by adding screened windows to the gable ends and screened doors will result in hot houses during the day but cooler temperatures at night, when people go to bed. After midnight the only house that was comfortable, according to the psychrometric analysis, was the metal-roofed house; in reality, since people go to bed wearing a T shirt or with a sheet, these conditions are comfortable for sleeping. This study, therefore, supports the development of screened windows and doors, which should be fitted to rural homes particularly those with metal roofs.

Our study has at least two limitations. First, the behaviour of volunteers sleeping in the experimental houses was untypical of villagers who move in and out of their houses, opening and closing the doors frequently before midnight.³¹ Thus, our estimates of protection afforded by screened doors are likely to be overestimated. Second, our experiments were limited to studies of one or two men in each house. Although many adult men sleep alone in single-roomed houses, these houses are mostly occupied by one or two women with several children. Multiple occupants are likely to increase the number of mosquitoes attracted to a house,³² although we would consider similar effect sizes of the interventions in higher occupancy houses.

This study not only provides fundamental insights into the house-entering behaviour of A gambiae, but also sheds light on practical measures that can be taken to reduce house entry of malaria mosquitoes and potentially reduce malaria. Our findings are likely to be relevant to similar typologies of housing in hot humid regions of sub-Saharan Africa, but further research is needed where house designs differ from those described in this study and where the climate is cooler. Our findings are likely to be of relevance to the transmission of malaria and also other vector-borne diseases in which transmission occurs indoors at night, such as lymphatic filariasis. A Roll Back Malaria, UN Development Programme and UN Habitat's Housing and Malaria consensus statement³³ asked, "What architectural features are protective against malaria?" Our study illustrates that sealing the eaves and adding screened doors will reduce house entry of mosquitoes substantially both in thatched-roofed and metal-roofed houses. Moreover, a metal-roofed house can be made as cool as a thatched-roofed house by encouraging airflow through the house by screening the doors and adding a screened window directly under the roof. The ventilated metalroofed house is hotter than a traditional thatched-roofed house during the day, which might increase the mortality of malaria mosquitoes; however, at night a ventilated metal-roofed house becomes as cool as a thatched-roofed house, making it more probable that people will sleep under an LLIN. Further changes in house structure are also required to prevent overcrowding, increase airflow, reduce indoor air pollution, and keep the occupants cooler.¹⁰ The transition of housing stock in sub-Saharan Africa from traditional thatched-roofed houses to metal-roofed houses is likely to have profound effects on the transmission of malaria and represents an enormous opportunity to reduce transmission further and keep malaria out in areas where malaria is eliminated but vectors remain.

Contributors

SWL, MP, JBK, and DJ conceived and designed the study. JBK designed the ventilated metal-roofed house. EJ and MJ carried out the fieldwork, supported by MP, BK, ALW, UD'A, and SWL. EJ, SWL, JB, DJ, and JBK analysed the data. All authors reviewed the final manuscript.

Declaration of interests

We declare no competing interests.

Acknowledgments

We thank the volunteers, land owners, and community of Wellingara for their support during the study. The houses were built by local craftsmen. The study was funded by the Sir Halley Stewart Trust. SWL and MP are supported by the Global Health Trials funded by the Medical Research Council–Department of International Development–Wellcome Trust. SWL and ALW are supported by the Global Challenges Research Fund.

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