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Evaluating a novel multi-species vector control tool for humanitarian crises: the efficacy of attractive targeted sugar baits among forcibly displaced populations in Northern Nigeria

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Abstract

Background Armed conflicts, natural disasters and forced population displacement have escalated dramatically since the 1990s. By mid-2024, the total number of people driven from their homes, often surviving in very harsh conditions for years, reached 122.6 million globally. In emergencies characterised by flimsy shelters, food insecurity, inadequate sanitation, poor access to health services and increased exposure to blood-feeding insects, diseases such as malaria, dengue and leishmaniasis cause high levels of morbidity and mortality. Conventional vector control interventions are inadequate in these settings due to operational and biological limitations. Novel vector control tools which are lightweight, easy to use and effective against multiple vector species are urgently needed to protect displaced populations.

Methods We conducted a 6-month, 2-arm community field trial in two internally displaced people camps in Maiduguri, Nigeria, to evaluate the entomological efficacy of attractive targeted sugar baits (ATSB). Monthly entomological monitoring measured changes in adult and immature vector density. Intervention acceptability was assessed using focus group discussions and a cross-sectional survey. To investigate environmental drivers of vector abundance, which might influence field outcomes, a hybrid approach of unsupervised and supervised machine learning regression models was developed using composite demographic, bioclimatic and ecological remote sensing data.

Results ATSB demonstrated a significant impact on indoor female *Anopheles gambiae* s.l. density (IRR: 0.140 [95% CI: 0.093–0.212]; $p < 0.0001$) and indoor blood-fed *An. gambiae* s.l. density (IRR: 0.0193 [95% credible interval: 0.0111–0.0356]). ATSB also significantly reduced indoor blood-fed *Aedes aegypti* (IRR: 0.0746 [95% credible intervals: 0.00884–0.502]). More than 97% of camp residents showed high levels of acceptance for ATSB, including willingness to pay. The strongest environmental predictors of *An. gambiae* s.l. occurrence were composite indices of vegetation water content, soil moisture, moist canopy, landcover diversity, urbanisation and normalised and enhanced vegetation index which together contributed to 73.5% of the final model.

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Conclusions Field trial findings strongly support the use of ATSB to control sympatric malaria and dengue vector populations in humanitarian crises. Remote sensing analysis identified key drivers of *An. gambiae* s.l. occurrence providing a high-resolution environmental profile where ATSB achieved an entomological impact against multiple vector species.

Keywords Conflict, Internally displaced persons, Malaria, Dengue, Temporary shelter, Vector control, Attractive targeted sugar baits

Background

Humanitarian crises are a singular event or series of events which critically threaten the health, safety, security and well-being of communities or populations [1]. Irrespective of whether the source is natural, climate-related or man-made, these crises may be accompanied by large-scale population movement, food insecurity and severe health systems disruptions [2]. In mid-2024, the United Nations High Commissioner for Refugees (UNHCR) estimated that 122.6 million people have been forcibly displaced worldwide, including 43.7 million refugees, 72.1 million internally displaced people (IDP) and 8 million asylum-seekers [3]. In 2025, the UN estimates that 305 million people will need humanitarian aid [4], and by 2030, two-thirds of the world's extreme poor will reside in areas of fragility, conflict and violence, with the latter driving 80% of all humanitarian needs [5]. Globally, approximately two-thirds of individuals affected by humanitarian crises inhabit malaria endemic areas [6], with almost complete geographical overlap between high burden malaria countries, regions of other intense vector-borne disease (VBD) transmission and ongoing emergencies, particularly in the World Health Organization (WHO) African region. Mass population displacement increases the risk of severe VBD epidemics, especially when individuals with little to no prior disease exposure move into areas of more intense transmission or when patients with subclinical infections transit into new settings [7]. Inadequate water, sanitation, and hygiene (WASH) facilities, drainage and waste management systems all contribute to high levels of vector breeding and increased disease transmission [8]; water storage in artificial containers in emergency settings is also highly conducive to the proliferation of arbovirus transmitting vector species, especially *Aedes* (*Ae.*) *aegypti* and *Ae. albopictus*, and the invasive malaria vector species, *Anopheles* (*An.*) *stephensi* [9, 10].

VBD transmission in many harsh humanitarian crises normally ramps up sharply and remains high where effective disease prevention is not established [11]. During the economic and civil conflict in Venezuela from 2000 to 2020, there was a 1200% increase in malaria cases [12]. More recently between 2021 and 2022, in Pakistan, catastrophic flooding affected more than 30 million people, leading to a 25% rise in malaria cases [13]. In 2023 in the Rohingya refugee camps in Cox's Bazar, Bangladesh, there were more than 17,500 reported dengue cases, including 3800 hospitalisations [14]. Finally, in the Central African Republic, where conflict has continued since 2008 and human rights abuse abounds, the highest measured nationwide mortality has been reported, with malaria being the leading cause [15].

Insecticide-treated nets (ITNs) and indoor residual spraying (IRS) are the cornerstones of malaria vector control in endemic, non-emergency settings, with a wealth of robust epidemiological data supporting their wide-scale deployment [13, 16–18]. While the WHO Global Malaria Program has endorsed the use of both ITNs and IRS during humanitarian emergencies [19, 20], their levels of protective efficacy are highly variable, as both interventions are contingent on living in a suitable housing or shelter structure which supports a hanging ITN or insecticidal treatment of an interior wall surface. In crisis settings, these core tools have not just insurmountable operational issues, including the need for a large, technically skilled workforce to implement repeated rounds of rotational IRS, very long lead times for ITN manufacture and high costs associated with importing IRS insecticides, heavy-duty spray tanks, personal protective equipment and heavy barrels of ITNs, but also biological limitations [21]. Consequently, malaria and other VBD deaths tend to rise sharply in the first weeks of an emergency and may stay very high for years in chronic situations [8]. While, in suitable permanent housing structures, ITNs and IRS can be very effective at controlling the few highly specialised *Anopheles* mosquito species that have strong preferences for human blood meals and bite at night, when people tend to be indoors and less mobile [16–18]; these interventions are unable to control the many other *Anopheles* populations that bite earlier in the evening or later in the morning, when most individuals are outdoors, nor do they provide protection from day-biting arbovirus vectors [22, 23]. ITNs and IRS also fail to control *Anopheles* vectors that enter houses at night, but exit early, are irritated, or repelled by the active ingredients (A.I.) used in ITNs and IRS. Collectively,

these 'unorthodox' mosquito species and behaviours occur in every endemic setting in Africa and sustain significant levels of persistent, residual malaria transmission [24, 25].

Recognising that the current armamentarium of vector control tools will be insufficient to eliminate malaria, there has been increasing momentum from commercial manufacturers, key donors and stakeholders to develop novel malaria and other multi-vector control tools, some of which may be appropriate for the emergency context. Attractive targeted sugar baits (ATSB) exploit the fundamental requirement of all mosquitoes to periodically feed on sugar (usually plant and floral nectars) to survive, by attracting mosquitoes to a sugar source to deliver an ingestion toxicant, thereby inducing vector mortality [26]. ATSB have several features which render them suitable for use during humanitarian crises, particularly in remote, insecure and inaccessible areas, or in mobile populations that move with little forewarning. They are light weight, highly portable, easily implementable and adaptable for diverse shelter types. ATSB also remain stable and viable when kept in storage for long time periods, allowing for stockpiling in strategic locations, ready for rapid deployment during crisis. Because populations affected by humanitarian emergencies suffer major trauma, it can be difficult to encourage behavioural change for beneficial good, which ATSB do not require. In these limited resource settings, communities may repurpose distributed humanitarian commodities, e.g. ITNs may be used for building structures, furniture or fishing; ATSB cannot be repurposed for any other function; therefore, distribution should lead to intended use.

To date, ATSB have been exclusively evaluated in stable settings to interrupt several mosquito and sand fly vector populations [27–31]. During initial semi-field trials, ATSB presented promising results in arid parts of West Africa, indicating that bait stations reduced numbers of female *Anopheles* with ≥ 3 gonotrophic cycles by 97.1% and sporozoite positive females by 97.8% [32, 33]. However, subsequent phase III cluster-randomised controlled trials (cRCT) in Mali, Kenya and Zambia reported minimal epidemiological impact on malaria transmission [34–37].

At the end of 2023, Africa hosted 35 million IDPs, 32.5 million were displaced by conflict and violence and 80% were concentrated in just five countries, the Democratic Republic of Congo, Ethiopia, Nigeria, Somalia and Sudan [38]. During the same time, four countries in sub-Saharan Africa accounted for ~ 50% of all global malaria cases and deaths, with Nigeria bearing the greatest burden (25.9% of all malaria cases and 30.9% of all malaria deaths) [39]. In the North of Nigeria, the protracted crisis and violent conflict have now entered the sixteenth year. Over 1.7 million people have been displaced from their homes in the second largest state, Borno State, of which approximately 853,000 live across 200 IDP camps [40]. As these camps have formed quickly, housing structures consist mostly of shelters constructed from plastic sheeting with a basic wooden frame. Nearly half of the IDP are living in these emergency shelters, while 33% reside in partially constructed or make-shift shelters with exposure to the elements [41]. We aimed to evaluate the entomological efficacy of ATSB against multiple vector species in IDP camps during a protracted humanitarian emergency in Northern Nigeria.

Methods

Study area and design

This study was a 6-month, 2-arm community-level field trial conducted in two urban IDP camps in Maiduguri Local Government Area (LGA), Borno State, Nigeria [Sabon Gari (11°50′50.4″N 13°06′32.0″E) and Doro (11°52′51.1″N 13°07′00.8″E)], which are separated by approximately 3.78 km. This area is characterised by hot, semi-arid climate, sparse savanna grasslands and shrubs, with land use dominated by subsistence agriculture, livestock grazing and increasing urbanisation. Population demographics for study camps are detailed in Additional file 1: Table S1. In Borno State, malaria accounts for more than 50% of mortality and morbidity, particularly in children < 5 years [42]. The peak malaria transmission seasons extend from July to October during the rainy season. The principal malaria vectors are *An. coluzzii* (98%) and, to a lesser extent, *An. arabiensis* (2%) (Allan R, Scherrer R, Estecha-Querol S, Weetman D, Paris L, Ba'abba Goni U, et al, The effectiveness of long-lasting spatial repellent against malaria in humanitarian crisis settings in Northern Nigeria. Lancet Infectious Diseases. 2025, under review). *Anopheles coluzzii* from Maiduguri LGA is resistant to pyrethroids with a 61.5% [95% CI: 58.8%–64.1%] frequency of voltage-gated sodium channel (*vgsc*)−995F and 15.6% [13.7%–17.6%] frequency of *vgsc*−1570Y mutations (Allan R, Scherrer R, Estecha-Querol S, Weetman D, Paris L, Ba'abba Goni U, et al, The effectiveness of long-lasting spatial repellent against malaria in humanitarian crisis settings in Northern Nigeria. Lancet Infectious Diseases. 2025, under review). Dengue also circulates in Borno State, coinciding with the main rainy season, but is underreported as most health facilities lack rapid diagnostic tests; a survey of febrile patients in Maiduguri detected anti-dengue IgM antibodies in 35% of participants [43].

ATSB intervention

The Sarabi v1.2 ATSB station (Westham Co., Hod-Hasharon, Israel) measures 24 cm × 31 cm. Each bait station

comprises a dark perforated membrane (4 pores/cm^2) that covers 16 wells containing 72 g date syrup-based bait, which acts as both the attractant and sugar meal, laced with dinotefuran (0.1% w/w), the A.I., and a bittering agent, Bitrex® (Johnson Matthey Group), to deter human consumption. Mosquitoes can probe and feed through small membrane pores of ~ 150 micron in diameter to access the sugar meal and A.I., while reducing the ability of non-target organisms to contact the bait. Dinotefuran (N-methyl-N′-nitro-N″-[(tetrahydro-3-furanyl)methyl]guanidine) is a neonicotinoid, which targets the nicotinic acetylcholine receptor (nAChR) in the insect central nervous system. Environmental assessment in Mali demonstrated that the ATSB toxicant poses limited risk to non-target organisms, including pollinators and humans [44].

Entomological monitoring

Following baseline entomological monitoring, 516 ATSB stations were installed to 172 shelters in Sabon Gari camp from 3 to 7 September 2024. Per shelter, 1 ATSB station was installed indoors, in the living room, and 2 ATSB stations were fixed outdoors on the side of each shelter; all ATSB stations were positioned 1.5–1.8 m above ground (Fig. 1A–D), according to the manufacturer's specifications. Despite the ATSB stations remaining efficacious for 6 months per manufacturer's recommendations, after entomological monitoring round 3, all original ATSB stations were replaced with new units; 507 ATSB stations were installed to 169 shelters from 9 to 13 December 2024. The control camp (Doro camp) received no vector control intervention as part of this field trial during the study period.

US Centers for Disease Control and Prevention light traps (CDC-LTs; John W Hock Company, USA) were used to sample adult, host-seeking vector populations for one trap night per month in a longitudinal cohort of 130 households per camp, over 6 months of follow-up. CDC-LTs were hung 1 m from the ground next to sleeping spaces indoors and were operational between 18:00 and 07:00 [45]. Indoor and outdoor resting adult vector populations were collected from the same houses using a 12-V battery-powered Prokopack aspirator the following morning [46]. Systematic sampling of indoor adult vectors resting on the walls, roofs, floors, furniture and household items was conducted in each room for up to 10 min, depending on the size of the house. Outdoor collections were performed from potential resting sites around the house, such as under roof eaves. One week after adult vector trapping, mosquito larvae/pupae were sampled from one water storage container per household using standard larval dippers. A minimum of 5 dips were used to sample each container, 4 from the corners/edges and 1 from the centre, with a short interval in between to allow for larvae/pupae that had descended to the bottom of the container to resurface. If containers could not be accessed using a dipper, then the water was poured out into a larger bucket, and immature stages were sampled in the same manner. Each month, ASTB coverage and condition were assessed in all 172 shelters in Sabon Gari camp during entomological monitoring activities. If any

Fig. 1 **A** Outdoor Sarabi v1.2 ATSB installation. **B** Indoor Prokopack aspiration and placement of Sarabi v1.2 ATSB. **C** ATSB information, education and communication (IEC) campaign. **D** Outdoor Prokopack aspiration and placement of Sarabi v1.2 ATSB. **E** Community engagement meeting during field trial baseline; **F** focus group discussion with men in Sabon Gari at 6 months post-intervention; and **G** focus group discussion with women in Sabon Gari at 6 months post-intervention

ATSB stations were missing, these were replaced immediately by the study team.

Laboratory analysis

Morphological identification

Adult *Anopheles* and *Aedes* were identified to species based on their morphology using dichotomous identification keys [47, 48]. The physiological status of female mosquitoes was further recorded. Adult *Culex* and phlebotomine sandflies were sexed and their physiological status recorded but not identified to species level. Thirty-two adult *Anopheles* were too damaged to be identified to species level (26 indoors and 6 outdoors) and were excluded from further analysis. A minority of adult *An. funestus* s.l. (18 indoors and 4 outdoors) and phlebotomine sandflies (3 indoors) were also collected but excluded from the primary analysis. Immature larvae/pupae were a visible mix of *Aedes* and *Culex* genera and were not identified further.

Molecular species identification

Molecular species ID was confirmed among a sub-set of *An. gambiae* s.l. and suspected *An. kingi* from both field trial arms. DNA was extracted from whole mosquitoes using the Qiagen DNeasy® blood and tissue kit (Qiagen, UK), according to manufacturer's instructions. Species identification of *An. gambiae* s.l. samples followed the qPCR melt curve protocol of [49] using Brilliant III Ultra-Fast SYBR Green Low ROX qPCR Master Mix (Agilent, UK) and was run on an Agilent AriaMx Real-Time PCR machine. Identifications were supplemented as necessary by the PCR method of [50] and/or the PCR–RFLP method of [51], where the qPCR result was unclear. Suspected *An. funestus* s.l. specimens were identified using the multiplex PCR protocol of [52]. In each PCR-based assay amplicons, differences in size which are species-diagnostic were visualised on 2% agarose gels. For all genotyping analyses of mosquitoes, positive and negative control samples were included in each set of reactions.

Specimens morphologically identified as suspected *An. kingi*, or any which failed to amplify using the genotyping methods above were PCR-amplified using LCO1490 and HCO2198 cytochrome oxidase I primers (COI) [53] with PCRs cleaned using the Qiagen PCR purification column kit (Qiagen, UK). Cleaned PCRs were sent for sequencing in forward and reverse directions by Eurofins genomics using the same primers. Resulting sequences were trimmed cleaned and aligned using CodonCode aligner software and checked for species identity using NCBI BLAST searches. Where necessary to attempt to improve species resolution, some of the samples sequenced at COI were additionally sequenced at the internal transcribed spacer 2 (ITS2) ribosomal DNA locus using primers ITS2A and ITS2B from [54]; bi-directional sequencing using the same PCR primers was performed by Eurofins genomics.

Intervention feasibility, acceptability and uptake monitoring

At field trial baseline (July 2024), a cross-sectional survey was conducted to enumerate households in both camps and to collect basic information about population demographics, household construction materials, vector control practices and intervention use, access to WASH and animal husbandry practices. Six months post ATSB distribution (February–March 2025), intervention acceptability was assessed among all participating households in Sabon Gari camp using a cross-sectional questionnaire. Focus group discussions (FGDs) were also conducted with men ($n=10$) and women ($n=10$) separately in Sabon Gari camp to assess intervention acceptability, perceived entomological impact and willingness to pay.

Satellite remote sensing analysis

Given that adult mosquitoes must engage in frequent sugar feeding to meet their energetic requirements, their local and positional abundance reflects the propensity to support multiple life-history behaviours, including host-seeking and blood-feeding, sugar foraging, mating, resting and oviposition. As such, vector abundance can be interpreted as an ecological proxy for the suitability of the environment to support these behavioural processes across the lifespan. To explore environmental drivers of vector abundance, a hybrid approach of unsupervised and supervised machine learning regression models was developed using composite demographic, bioclimatic and ecological remote sensing data. The full methodology is detailed in Additional file 1: Material S1, Tables S2–S4 and Figs. S1–S4 (Allan R, Scherrer R, Estecha-Querol S, Weetman D, Paris L, Ba'abba Goni U, et al, The effectiveness of long-lasting spatial repellent against malaria in humanitarian crisis settings in Northern Nigeria. Lancet Infectious Diseases. 2025, under review) [55].

Sample size

The primary entomological endpoints were adult *An. gambiae* s.l., *Ae. aegypti* and *Culex* spp. abundance indoors and outdoors, assessed by CDC-LTs and Prokopack aspirators. The sample size calculation of 130 households per field trial arm was powered on indoor female *An. gambiae* s.l. density. Baseline entomological monitoring estimated a coefficient of variation (CV) of 1.36 in *An. gambiae* s.l. counts between households; assuming an average of 20 female *An. gambiae* s.l. per household in the control arm, 80% power and a

5% significance level, this study was powered to detect a minimum intervention effect of 44.4%.

Statistical analysis

Based on observations of a high proportion of blood-fed vectors in CDC-LTs at baseline, both indoor collections (CDC-LTs and Prokopack aspirator collections) were pooled per species for analysis. Differences in vector population density were analysed using mixed effects negative binomial regression, with a fixed interaction between study arm and timepoint, a random intercept for household to account for repeated longitudinal measures and robust standard errors to adjust for clustering. Within-arm pre-post effects were estimated for the control arm, as $IRR_t = \exp(\gamma_t)$ (the time main effect at time t) and for the intervention arm as $IRR_t = \exp(\gamma_t + \delta_t)$ (time main effect plus the arm × time interaction at time t). Summary pre-post effect per arm was calculated as the geometric mean IRR and CIs were derived via the delta method using linear combinations of coefficients. Results are reported as incidence rate ratios (IRR) with corresponding 95% confidence intervals (CI). Depending on relative sample size per timepoint, sub-analyses by vector sex, physiological status, month and location were also performed. To analyse vector species with sparser count data through time, we fitted a Bayesian negative binomial regression with fixed effects for study arm, timepoint and their interaction and a random intercept for household to account for repeated measures. Posterior distributions were estimated using Markov chain Monte Carlo (MCMC) sampling with default priors. Each model was run with 12,500 MCMC iterations, a burn-in of 2500 and a final posterior sample size of 10,000. Posterior means of the log-linear coefficients for interaction terms between study arm and timepoint were exponentiated to derive IRRs, and the corresponding 95% equal-tailed credible intervals were derived by exponentiating the posterior lower and upper bounds of the log-scale coefficients. Congruence of estimated intervention impact was validated between mixed effects negative binomial regression models and Bayesian negative binomial regression models using the most abundant vector species as test data (*An. gambiae* s.l. and *Culex* spp. populations; Additional file 1: Table S5). Poisson regression was used to compare rates of vector blood-feeding pre- and post-intervention. The number of blood-fed females was modelled relative to the total number of females collected, which was included as a log offset to account for variation in sampling effort.

Entomological indices for immature vector surveys were calculated as follows: Container Index (CI) = containers positive for larvae or pupae*100 / containers inspected; Household Index (HI) = houses positive for larvae or pupae*100 / houses inspected; and Breteau Index (BI) = containers positive for larvae or pupae*100 / houses inspected and are presented in Additional file 1: Tables S6 and S7. We used a Bayesian multilevel logistic regression model to evaluate the impact of ATSB on the likelihood of detecting immature vectors breeding in household containers with a random intercept for household for repeated measurements. The outcome was binary (presence or absence of larvae/pupae), and models included fixed effects for study arm, timepoint and their interaction. Posterior distributions were estimated using MCMC sampling with default priors. Each model was run with 12,500 MCMC iterations, a burn-in of 2500 and a final posterior sample size of 10,000. Posterior means of the log-linear coefficients for interaction terms between study arm and timepoint were exponentiated to derive adjusted odds ratios (aOR) with corresponding 95% credible intervals.

Baseline household characteristics were compared between camps using Pearson's chi-squared tests or Fisher's exact tests (when responses were < 10). FGDs were recorded, manually transcribed and reported qualitatively. For all analyses, protective efficacy (PE) was calculated as (1 − IRR)*100. An alpha level of $p = 0.05$ was used for significance testing; p values were adjusted using Bonferroni correction when accounting for multiple testing. No missing data were reported. All statistical analyses were performed using StataNow/MP 19.5 (StataCorp LLC, College Station, TX). Data were visualised in RStudio v2024.12.1 + 563 [56].

Results

Baseline household characteristics per camp, including house construction, vector control practices, water storage practices, WASH access and animal husbandry, are summarised in Additional file 1: Table S8. Most households were constructed from plastic sheeting (79.43%; 251/316), with open windows (57.28%; 181/316) and few indoor toilets (37.03%; 117/316). Householders had access to waste collection (66.46%; 210/316) and drinking water located within minutes of their house (79.75%; 252/316). A minority of householders kept animals, principally poultry (10.44%; 33/316), dogs/cats (6.01%; 19/316) and goats (3.16%; 10/316). Householders reported using ITNs for vector control (70.57%; 223/316), with 80.38% (254/316) of household currently using ITNs. The majority of ITNs were received during mass distributions by the National Malaria Control Program/non-governmental organisations (31.96%; 101/316) or bought by householders in the shop/market (31.65%; 100/316). The proportion of current ITN users was comparable between intervention and control camps (75.37%

versus 84.07%, respectively; $\chi^2 = 1.51$; $p = 0.220$). Most householders reported receiving their ITNs years ago (62.34%; 197/316); the median age of ITNs was 2 years (SD ± 1.79 and 1.19, respectively) in both intervention and control camps.

One hundred and seventy-two shelters received the initial ATSB intervention, housing 804 individuals, with an average of 1.47 (SD ± 0.051) rooms per shelter. After the entomological round 3, 169 households received new ATSB; 3 families dropped out of the study. ATSB coverage and retention remained high throughout the field trial, with 98.82% of households reporting all present ATSB stations each month (1007/1019 responses). Twenty-two ATSB stations went missing (6 inside and 16 outside) and were replaced during the field trial: 16 at round 2, 2 at round 3, 2 at round 5 and 2 at round 6.

During 6 months of post-intervention entomological monitoring, a total of 18,156 adult *An. gambiae* s.l. were collected, 17,800 inside and 356 outside (Additional file 1: Tables S9 and S10); a total of 1189 *Aedes* spp. were collected, 834 inside and 355 outside (Additional file 1: Tables S11 and S12), of which 923 were identified as *Ae. aegypti* (Additional file 1: Tables S13 and S14); and a total of 225,393 *Culex* spp. were collected, 209,498 inside and 15,895 outside (Additional file 1: Tables S15 and S16). Molecular species identification classified the majority of *An. gambiae* s.l. as *An. coluzzii* (77%), with proportions comparable between field trial arms (Additional file 1: Material S2 and Table S17 [57]). Suspected *An. kingi* individuals were confirmed molecularly as *An. coustani* group or *An. squamosus.*

Post-intervention, ATSB demonstrated a significant impact on the density of indoor female *An. gambiae* s.l. over 6 months of follow-up (IRR: 0.140 [95% CI: 0.093–0.212]; $p < 0.0001$) (Table 1 and Fig. 2) and indoor blood-fed *An. gambiae* s.l. density for the first 2 months post-intervention, after which complete population suppression was observed in the intervention arm (IRR: 0.0193 [95% credible intervals: 0.0111–0.036]) (Table 1). A significant intervention effect was also evident for both sexes of indoor *An. gambiae* s.l. and all outdoor *An. gambiae* s.l. (Table 1). Following ATSB deployment, a significant reduction in the rate of indoor *An. gambiae* s.l. blood-feeding was observed in the intervention arm (IRR: 0.086 [95% CI: 0.0749–0.0978]; $p < 0.0001$); a parallel significant increase in the rate of indoor *An. gambiae* s.l. blood-feeding was apparent in the control arm (IRR: 1.962 [95% CI: 1.819–2.116]; $p < 0.0001$).

Similarly, post-intervention, ATSB demonstrated a significant impact on indoor blood-fed *Ae. aegypti* (IRR: 0.0746 [95% credible intervals: 0.00884–0.502]) for the first 2 months post-intervention, after which complete population suppression was observed in the intervention arm (Fig. 2). A significant intervention impact was also evident on outdoor *Aedes* spp. density across 6 months of follow-up (IRR: 0.0041 [0.0012–0.014]; $p < 0.001$; Additional file 1: Table S18). No significant intervention effect was observed for other *Aedes* spp. populations (Additional file 1: Table S18). Following ATSB deployment, a significant reduction in the rate of indoor *Ae. aegypti* blood-feeding was observed in the intervention arm (IRR: 0.136 [95% CI: 0.0325–0.568]; $p = 0.006$); a parallel significant increase in the rate of indoor *Ae. aegypti* blood-feeding was apparent in the control arm (IRR: 1.536 [95% CI: 1.130–2.086]; $p = 0.006$).

ATSB had a minimal reduction in *Culex* spp. population density (Additional file 1: Table S19) until month 5, when an upward trend in PE was observed, becoming statistically significant at the final follow-up (IRR: 0.448 [95% CI: 0.288–0.696]; $p < 0.0001$). Consistent with seasonal temporal trends, relative to baseline, most vector population densities declined over 6 months of follow-up; however, the reduction was markedly greater in the intervention arm (Additional file 1: Tables S20 and S21). ATSB did not have a sustained impact on presence of immature mosquito life stages among study households (aOR: 0.551 [95% credible intervals: 0.164–1.742]); however, a significant reduction was observed at 2, 5 and 6 months post-intervention (Additional file 1: Table S22).

After 6 months of use, ATSB were well received by male and female householders (Additional file 1: Table S23). In general, participants in both FGDs were initially hesitant about the intervention but then observed an entomological impact, which improved intervention acceptability. Householders reported minimal issues with ATSB installation and experienced no observable side effects. Householders also expressed a preference for ATSB over other vector control tools, a willingness to use ATSB in the future and to purchase the intervention. FGD observations were consistent with positive acceptability results from the cross-sectional survey (Additional file 1: Table S24).

The strongest environmental predictors (XGBoost Tuned; RMSE = 70.84; ROC AUC = 0.830) of *An. gambiae* s.l. occurrence were composite indices of vegetation water content, soil moisture, moist canopy, land cover diversity, urbanisation (human modification index, population density and built volume) and normalised and enhanced vegetation index which together contributed to 73.5% of the final model (Fig. 3 and Additional file 1: Fig. S3).

Discussion

Field trial findings demonstrated a significant impact of ATSB on pyrethroid-resistant *An. gambiae* s.l. populations across the household, as well as indoor blood-fed

Table 1 Protective efficacy of ATSB against wild pyrethroid-resistant *An. gambiae* s.l. populations in Northern Nigeria across 6 months of follow-up

Vector population	IRR [95% CI]	Protective efficacy [95% CI][g]	*p* value
Indoor[a] female *An. gambiae* s.l. (all physiological status)	0.140 [0.093–0.212][c]	85.95% [78.84%–90.67%]	< 0.0001
Indoor[a] *An. gambiae* s.l. (both sexes)	0.141 [0.094–0.210][c]	85.95% [78.98%–90.61%]	< 0.0001
Indoor[a] blood-fed *An. gambiae* s.l	0.0193 [0.0111–0.0356][d,e,f]	98.07% [96.44%–98.89%]	–
Outdoor *An. gambiae* s.l	0.0088 [0.0036–0.214][c]	99.12% [97.86%–99.64%]	< 0.0001
All[b] household *An. gambiae* s.l	0.140 [0.0949–0.206][c]	86.0% [79.40%–90.51%]	< 0.0001

[a] Indoor vector populations refer to *An. gambiae* s.l. collected in both CDC-LTs and by indoor Prokopack aspirators.

[b] II household vector populations refer to *An. gambiae* s.l. collected in CDC-LTs and by both indoor and outdoor Prokopack aspirators.

[c] IRR reported from mixed effects negative binomial regression, with a fixed interaction between study arm and timepoint, a random intercept for household to account for repeated measures and robust standard errors to accommodate clustering.

[d] IRR reported from Bayesian negative binomial regression models, with a fixed interaction between study arm and timepoint, a random intercept for household to account for repeated measures.

[e] Credible intervals which do not include 1 are considered statistically significant.

[f] Due to sparse/zero *An. gambiae* s.l. counts during later field trial months, this analysis was restricted to the baseline and the first 2 months post-intervention.

[g] PE = protective efficacy ((1 – IRR)*100)

Ae. aegypti and outdoor *Aedes* spp. populations. This is the first study to report an entomological effect of ATSB at the community-level against both sympatric malaria and dengue vector species, as well as to evaluate this tool in temporary shelters during a protracted humanitarian emergency. ATSB was received with high levels of participant acceptability, including willingness to pay, following perceived entomological effect, which is consistent with previous qualitative assessments of ATSB uptake [58, 59]. Given the inherent biological and operational restraints of 'gold standard' vector control interventions, study results strongly support future deployment of ATSB to control multi-vector populations in crisis settings, potentially expanding the toolbox of efficacious products for the emergency context.

Interestingly, our field trial observations do not align with previous multi-county cRCTs of ATSB, which failed to reduce malaria transmission or *Anopheles* parity [34, 35]. Discordance in entomological results may be explained by several factors, including insufficient

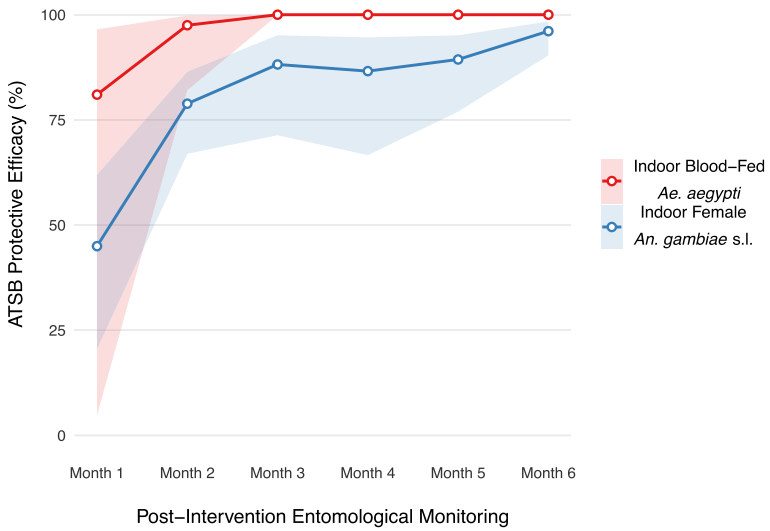

Fig. 2 Monthly protective efficacy of ATSB against indoor female *An. gambiae* s.l. and indoor blood-fed *Ae. aegypti* over 6 months of follow-up. Points indicate estimates of PE for each follow-up month (months 1–6), and shaded areas represent 95% confidence intervals. Baseline data (month 0) were included in the models for adjustment but not shown in the figure

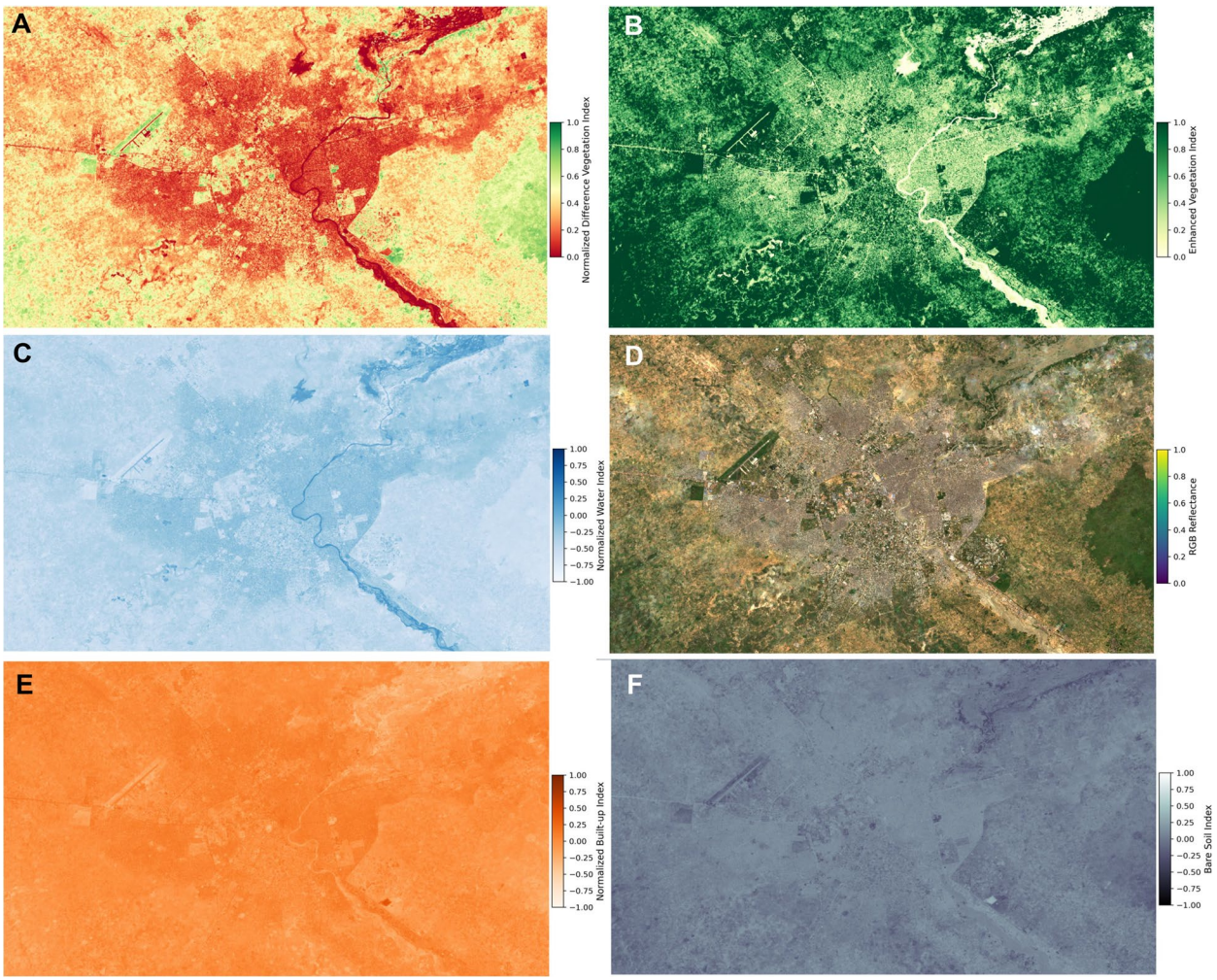

Fig. 3 Multispectral satellite imagery of Maiduguri during the 2024 rainy season, derived from the Sentinel-2 Surface Reflectance Harmonized dataset (COPERNICUS/S2_SR_HARMONIZED), displaying key environmental predictors of *An. gambiae* s.l. occurrence. Individual images were aggregated over a 4-month period, and the median spectral value of each pixel, within the area of interest (AOI), was computed to represent typical surface conditions, with cloud contamination and sensor noise excluded from analysis. **A** Normalised difference vegetation index (NDVI = (NIR – Red) / (NIR + Red); **B** enhanced vegetation index (EVI = 2.5*(NIR – Red) / (NIR + 6 Red – 7.5*Blue + 1); **C** normalised difference water index (NDWI = (Green – NIR) / (Green + NIH); **D** red, green, blue reflectance (RGB composite using reflectance bands: B2 = blue, B3 = green and B4 = red); **E** normalised difference built-up index (NDBI = (SWIR – NIR) / (SWIR + NIR)); and **F** bare soil index (BSI = ((SWIR + RED) – (NIR + BLUE)) / ((SWIR + RED) + (NIR + BLUE))

ATSB coverage per house/density per hectare, ATSB location, intervention durability and environmental differences between sites. In this study, we deployed ATSB as a combined indoor and outdoor intervention, installing two ATSB units outside and one inside per eligible structure. Earlier semi-field trials of ATSB in Mali evaluated this intervention inside houses, reporting a decrease in indoor female *An. gambiae* s.l. populations by 90% and a 3.8-fold reduction in proportion of female vectors that had completed four or more gonotrophic cycles [30]. Similarly, in Tanzania and Côte d'Ivoire, indoor experimental ATSB, using either chlorfenapyr or boric acid, were highly efficacious against wild *An. arabiensis* and *Cx. quinquefasciatus*, and *An. gambiae* s.l., respectively [31, 60]. With growing impetus from the vector control community to develop new tools to tackle outdoor malaria transmission and hot spots of residual infection [22, 61], later large-scale cRCTs installed two ATSB on outer house walls [44], thereby relying upon host-seeking, endophagic vectors to sugar feed outside either before or after house entry. However, the evidence for predominant outdoor foraging/resting behaviours in major vector species is variable. In some settings, the proportion of exophilic

An. gambiae s.l. may exceed 70% of the overall vector population [62], while in others, less than 15% of mosquitoes were collected outside [63], with plasticity in preferences driven by overlapping genetic adaptations [64], vector life-histories [65], microclimatic conditions [66] and community coverage of indoor insecticidal interventions [62]. Furthermore, such vector behaviours cannot be assumed to be uniform throughout a mosquito's lifespan. Newly emerged female mosquitoes have greater initial requirements for sugar versus blood [67] and may be more likely to feed from ATSB prior to finding a blood-meal. In this scenario, ATSB deployment will result in vector population suppression over time but may not initially reduce the more epidemiologically relevant cohort of older *Plasmodium*-infected mosquitoes to the same extent as their younger counterparts.

There were several other features of this field trial design and camp environment which may explain the significant intervention effect reported in this study compared to the ATSB cRCTs. IDP camp households were spatially clustered, often with multiple families residing under a single roof. While formal estimations of target ATSB number per area are forthcoming [34], dispersed settlement patterns, and resulting low intervention density was cited as one explanation for the lack of ATSB efficacy in Kenya and Zambia [34, 35]. In this field trial, we maintained high ATSB coverage by rigorous monitoring, with missing stations replaced immediately, and all units were pre-emptively exchanged anew after 3 months of community use. Previous large-scale durability monitoring of outdoor ATSB reported the median survival time was 7 months [68, 69], with vector mortality remaining higher than 80% as ATSB aged under field conditions [70]; while not yet quantified, indoor station longevity would be expected to be greater, without exposure to the elements. In this field trial, we adopted a more conservative approach by shortening the length of field use of each ATSB unit to establish proof-of-concept of this intervention in the emergency setting. Prospective ATSB deployment in this context could adopt a less frequent replacement scheme, supported by high levels of community acceptability and uptake.

Arguably, the most parsimonious reason for differences in ATSB impact between study sites is the relative abundance of alternate environmental sugar sources. Using a supervised machine learning approach, we identified green biomass and moisture indices, including vegetation water content, soil moisture, moist canopy, landcover diversity, urbanisation and normalised and enhanced vegetation index, as principal drivers of *An. gambiae* s.l. occurrence during the rainy season in Maiduguri. Further work is required to determine key ecological variables and associated thresholds which are predictive of ASTB performance. In general, once vector control tools have received a WHO recommendation from the Vector Control Advisory Group, the conventional paradigm has assumed that intervention efficacy is comparable across endemic areas, recognising that there are limited resources available for tool evaluation in every conceivable setting. Tailoring vector control intervention deployment at the sub-national level [71, 72] has been gaining prominence both in response to the biological threat of ubiquitous insecticide resistance among *Anopheles* populations [73, 74] and the availability of more expensive, yet more effective new dual-A.I. ITNs [16, 17, 75]. ATSB is an intervention that, by definition, will have variable efficacy based on the availability of environmental sugar sources. Contemporary frameworks for ATSB deployment are based upon predicted proportion of dyed/bait-feeding vectors [76]; our field trial results demonstrate the utility of remote sensing data to support intervention deployment and argue for inclusion of high-resolution ecological site characterisation as an additional dimension for decision-making regarding ATSB implementation. Comparing predominant environmental predictors across geographical locations may enable the identification of ecological settings where vector behaviours are more likely to support sustained ATSB impact, thereby informing targeted intervention deployment.

Our field trial results raise several unanswered questions regarding the mode of action of ATSB on vector populations. Following ATSB deployment, blood-feeding rate in the intervention arm dropped to 8.6% and 13.6% compared to baseline, for both *An. gambiae* s.l. and *Ae. aegypti*, respectively. After feeding on ATSB, it is anticipated that vector neurotoxicity is rapid, within 24–48 h. Results may indicate that dinotefuran ingestion can also inhibit blood-feeding, either by impairing host-seeking, disrupting probing, or overall reducing coordination and responsiveness to host semiochemical cues. To date, there is a paucity of data regarding the effect of sublethal oral dinotefuran exposure on vector blood-feeding, which warrants further investigation. The results of this field trial also suggest that indoor and outdoor ATSB may differentially contribute to the intervention effect per vector species; at the field trial baseline, 1.75% and 68.1% of household *An. gambiae* s.l. and *Ae. aegypti*, respectively, were found outside. To determine whether vector feeding was equitable across indoor and outdoor ATSB, future field trials should consider using different dye colours per station or newly developed scalable camera traps [77].

The field trial results should be interpreted in the context of several limitations. Assessment of the relative proportions of vectors feeding on natural sugar (by cold anthrone testing) [32] or ATSB (by treating bait stations with food dye or uranine) [78] was outside the scope of this study. It was also not feasible to rear immature vector stages to adulthood, meaning these entomological indices cannot be disaggregated by genera; however, the abundance of adult *Culex* in our field trial sites and lack of intervention effect on adults of these species likely explains the variable impact observed on immature vector life stages. Finally, additional measurements of vector parity to determine the effect of ATSB on reducing gonotrophic cycle number, blood-meal analysis to dissect changes in host feeding following dinotefuran exposure, and *Plasmodium* sporozoite rate to assess intervention impact on malaria transmission, were not possible due to financial constraints.

Conclusions

To date, the phase III epidemiological and entomological evidence for ATSB deployment has been mixed. This field trial is the first to report an entomological effect of ATSB at the community-level against both sympatric malaria and dengue vector populations, as well as to evaluate this innovative tool in temporary shelters during a protracted humanitarian crisis. Field trial findings indicated that combined indoor and outdoor ATSB can achieve rapid suppression of *An. gambiae* s.l. and *Ae. aegypti* populations within 2 months of implementation. We attribute the positive entomological outcomes reported herein to several key differences in field trial design, study site characteristics and intervention deployment, compared to prior cRCTs. In this field trial, ATSB were deployed as a combined indoor and outdoor intervention, with evidence indicating differential contributions in bait stations to intervention impact per major vector species. Households in this field trial were also spatially clustered, leading to high intervention density per area and complete ATSB coverage was maintained by rigorous monitoring, with missing stations replaced immediately, and all units pre-emptively exchanged anew after 3 months of community use. The results of this field also demonstrated the use of remote sensing data to support intervention deployment and argue for the inclusion of high-resolution environmental site characterisation as an additional dimension for decision-making regarding ATSB use. Given the inherent biological and operational restraints of the major vector control interventions, these field trial results strongly support future deployment of ATSB to control multi-vector populations in crisis settings, potentially expanding the toolbox of efficacious products for the emergency context.

Abbreviations

AI	Active ingredient
AOI	Area of interest
aOR	Adjusted odds ratio
ATSB	Attractive targeted sugar bait
AUC	Area under the curve
BI	Breteau index
BLAST	Basic local alignment search tool
BSI	Bare soil index
CDC	Centers for Disease Control and Prevention
CI	Confidence interval
CI	Container index
COI	Cytochrome oxidase I
cRCT	Cluster-randomised controlled trials
CV	Coefficient of variation
EVI	Enhanced vegetation index
FGD	Focus group discussion
IDP	Internally displaced person
IEC	Information, education communication
IgM	Immunoglobulin M
IRR	Incidence rate ratio
IRS	Indoor residual spraying
ITN	Insecticide-treated net
ITS2	Internal transcribed spacer 2
HI	Household index
LGA	Local Government Area
LT	Light trap
MCMC	Markov chain Monte Carlo
nAChR	Nicotinic acetylcholine receptor
NCBI	National Center for Biotechnology Information
NDBI	Normalised difference built-up index
NDVI	Normalised difference vegetation index
NDWI	Normalised difference water index
NIR	Near-infrared
PE	Protective efficacy
qPCR	Quantitative polymerase chain reaction
RFLP	Restriction fragment length polymorphism
RGB	Red, green, blue reflectance
RMSE	Root mean square deviation
ROC	Receiver operating characteristic
SWIR	Short-wave infrared
UNHCR	United Nations Higher Commissioner for Refugees
VBD	Vector-borne disease
Vgsc	Voltage-gated sodium channel
WASH	Water, sanitation, and hygiene
WHO	World Health Organization
XGBoost	Extreme Gradient Boosting

Supplementary Information

The online version contains supplementary material available at https://doi.org/10.1186/s12916-025-04449-5.

Additional file 1. Tables S1–S24, Materials S1 and S2 and Figs. S1–S4. Table S1 Population demographics of Sabon Gari (intervention) and Doro (control) camps. Table S2 Sixty demographic, bioclimatic and ecological remote sensing variables for initial multicollinearity analysis. Table S3 Final list of composite remote sensing variables for supervised machine learning analysis. Table S4 Comparative machine learning performance of different regression models. Table S5 Cross-validation of estimated protective efficacy of ATSB using Frequentist and Bayesian approaches across 6 months of follow-up in Northern Nigeria. Table S6 Household entomological indices for larval surveys in Northern Nigeria. Table S7 Household entomological indices for pupal surveys in Northern Nigeria. Table S8 Baseline household characteristics of Sabon Gari (intervention) and Doro (control) camps. Table S9 Baseline household characteristics of Sabon Gari (intervention) and Doro (control) camps. Table S10 Total *Anopheles gambiae* s.l. collected in the control arm, disaggregated by location, sampling method, sex and physiological status. Table S11 Total *Aedes* spp. collected in the intervention arm, disaggregated by location,

sampling method, sex and physiological status. Table S12 Total *Aedes* spp. collected in the control arm, disaggregated by location, sampling method, sex and physiological status. Table S13 Total *Aedes aegypti* collected in the intervention arm, disaggregated by location, sampling method, sex and physiological status. Table S14 Total *Aedes aegypti* collected in the intervention arm, disaggregated by location, sampling method, sex and physiological status. Table S15 Total *Aedes aegypti* collected in the intervention arm, disaggregated by location, sampling method, sex and physiological status. Table S16 Total *Culex* spp. collected in the control arm, disaggregated by location, sampling method, sex and physiological status. Table S17 Summary of vector molecular species identification disaggregated by field trial arm. Table S18 Protective efficacy of ATSB against wild *Aedes* spp. populations in Northern Nigeria across 6 months of follow-up. Table S19 Protective efficacy of ATSB against wild *Culex* spp. populations in Northern Nigeria across 6 months of follow-up. Table S20 Changes in vector density pre-post-intervention in Northern Nigeria in the intervention arm. Table S21 Changes in vector density pre-post intervention in Northern Nigeria in the control arm. Table S22 Protective efficacy of ATSB against wild immature vector populations in Northern Nigeria across 6 months of follow-up. Table S23 Focus group discussion (FGD) quotations regrading aspects of ATSB acceptability and feasibility at 6 months post-intervention. Table S24 ATSB acceptability at 6 months post-intervention. Material S1 Demographic, bioclimatic and ecological environmental variables, model selection and evaluation. Material S2 Vector molecular species identification. Fig. S1 Heatmap presenting pairwise Pearson's correlation coefficients between remote sensing variables and *Anopheles gambiae* s.l. (both sexes, indoor and outdoor collections) count data. Fig. S2 Heatmap presenting pairwise Pearson's correlation coefficients between final composite remote sensing variables and *Anopheles gambiae* s.l. (both sexes, indoor and outdoor collections) count data. Fig. S3 Proportional contribution of composite remote sensing variables to the final selected model (XGBoost Tuned). Fig. S4 Actual versus predicted *Anopheles gambiae* s.l. occurrence modelled using XGBoost (Tuned).

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Authors' contributions
RJA, DW, SEQ and LAM contributed to conception and design of the study. RA, MK, LAM, SA, MMI, UBG, GMA, MW, BNA, KLFC, DW, AG, and GN worked on acquisition and analysis of data. RJA and LAM drafted the manuscript and figures. All authors read and approved the final version of the manuscript. RJA, DW, and LAM have accessed and had verified the underlying data.

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Data availability
The datasets generated for or analysed are presented in the Article or the Appendix. All other relevant data are available from the corresponding author upon reasonable request. Nucleotide sequence data are available from NCBI GenBank under the accession numbers PV642914-PV642933 and PV629235-PV629253.

Declarations

Ethics approval and consent to participate
Ethical review and approval for this study was granted by the University of Maiduguri Teaching Hospital (reference: OHRP-IRB-FWA-00013572 UMTH/REC/24/31) and the Liverpool School of Tropical Medicine (reference: 24–021). Authorisation to conduct the study was obtained from the Ministry of Health and Human Services State Health Research Ethics Committee (reference: SHREC No.049/2024), the National Malaria Elimination Programme (reference: NMEP/IVM/MI/SI/64) and the Borno State Emergency Management Agency. Written informed consent was obtained from all heads of households which received the intervention and participated in the entomological monitoring.

Consent for publication
Not applicable.

Competing interests
The authors declare no competing interests.

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