Evaluation of Bednets After 38 Months of Household Use in Northwest Ghana

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Abstract. A total of 255 bednets were collected 38 months after distribution in Lawra District of northwest Ghana to examine their physical condition and residual insecticide levels. Physical condition varied from nearly pristine to highly damaged. In 50 selected nets, 2023 holes $\geq 0.5$ cm and 31 holes $\geq 10$ cm were counted. The incidence of holes increases toward the bottom edge of the net. Seam failures were found in 50% of the nets. Repairs, mostly sewn, were evident in 64% of the nets. Using a combination of bromine x-ray fluorescence (XRF) spectrometry, high-pressure liquid chromatography, and cone bioassays, it was determined that 14.9% of the nets had retained full insecticidal strength. These results highlight the value of real-world data on bednet longevity to guide decisions regarding mosquito control strategies, bednet purchasing, frequency of bednet replacement, and product development.

INTRODUCTION

Several large-scale campaigns are currently underway to distribute long-lasting insecticide-treated bednets (LLINs) as part of an overall effort to reduce the malaria burden in sub-Saharan Africa. At this time most LLINs are being supplied by 2 manufacturers, but new LLIN products are under active development by several others. Standards for these products, both in terms of the fabric quality and the durability of the insecticide treatment, have been developed by the World Health Organization (WHO).

The WHO Pesticide Evaluation Scheme (WHOPES) regularly reviews performance data on new LLIN products and issues recommendations as to their effectiveness based on laboratory, experimental hut, and field test results. Although these types of studies are essential and provide valuable information, they are necessarily limited in scope so they cannot capture the effects of local climate, household wealth, use patterns, and many other factors that may profoundly impact the effective lifetime of LLINs.

Data concerning the real-world durability of LLINs are important for several reasons. Such data can be used to confirm that limited-scale testing actually reflects real-world performance and, if not, can be used to guide the design of new or modified methodologies to make them more predictive. Such data can also be used to aid manufacturers in the development of improved products by identifying shortcomings not discovered in small-scale tests. Furthermore, the data can assist those procuring bednets in deciding the most cost-effective products for use in a given distribution campaign. Agencies in charge of distributions can also use these data to determine when LLINs in the field should be replaced or treated, and which of these two courses of action is more appropriate. Finally, bednet longevity data would be valuable when deciding if LLIN distribution is a more cost-effective means of mosquito control compared with indoor residual spraying (IRS).

This report describes the findings from a study of a sample of bednets distributed in December 2002 in the Lawra District of the Upper West Region of Ghana. Lawra is a rural district and one of the poorest regions in the country, with an estimated 68.3% of the population living below the extreme poverty line of 700,000 cedis (US$86) per adult per year. It is located in the southern Sahel and experiences a cycle of wet, dry, and hot seasons. Malaria incidence is seasonal, and it may be presumed that bednet use is likewise. Household bednet coverage in northern Ghana prior to the 2002 distribution campaign was estimated at 4.4%. During the campaign, 14,600 LLINs were distributed and household bednet coverage 5 months post-campaign was determined to be 89.5%. Although the original plan was for Dawa nets (SiamDutch, Bangkok, Thailand) to be distributed, procurement difficulties at the time of distribution forced the use of a combination of Dawa nets, PermaNets (version 1.0; Vestergaard Frandsen, Lausanne, Switzerland), and deltamethrin-pretreated Siam-Dutch nets.

METHODS

Collection of bednets. Bednets were collected in February 2006, 38 months after distribution, in conjunction with a follow-up survey to evaluate use and coverage. During each household visit, the survey worker established whether the household still possessed a bednet distributed during the 2002 campaign. If one was present, the survey worker would offer a new PermaNet 2.0 LLIN in exchange for the campaign bednet and 5000 cedis (US$0.60). Recovered nets were immediately labeled and bagged individually in polyethylene bags. Survey workers returned the bednets and survey data to a central collection point. Money collected was used to help defray the cost of the survey.

A total of 250 PermaNet 2.0 bednets were donated by Vestergaard Frandsen to be used for exchange, and 7 or 8 bednets were given to each of the 32 survey workers. In this way used bednets could be recovered from every part of the district. After collection, the bednets were shipped to CDC laboratories in Atlanta for evaluation.

Physical evaluation. Recovered bednets were examined to identify the manufacturer and then repackaged into fresh polyethylene bags to facilitate storage. For most nets, the manufacturer labels provided identification, but in some cases the labels were either missing or illegible. In the case of PermaNet bednets, several features could be used for positive
identification if the label was compromised: the characteristic 4 × 8.5 cm dimensions of the label, the presence of a 10-cm band of reinforced fabric around the bottom edge of the net, and the existence of fabric tie-downs with no metal rings at the top edge of the net.

To examine the nets, a cube-shaped frame, approximately 180 cm on a side, was constructed using commercial PVC pipe and fittings. Black plastic sheeting was hung from the top rail of the frame to provide a contrasting background to aid examination and photography. Nets were then draped over the frame for examination.

Holes in the fabric panels were measured for size, and those 0.5 cm or larger were recorded. Because the netting tends to tear in a preferred direction, larger holes usually appeared as ovals. In these cases, hole size was measured along the long axis of the oval. The hole locations were measured as the vertical distance from the bottom edge of the net to the center of the hole. Holes on the top panels were recorded separately.

The condition of sewn seams was also noted. Most rectangular nets possessed a single vertical seam at one corner, with an additional seam attaching the top panel to the side panels. The presence of a 1-cm open length along the seam was judged to be a seam failure.

Evidence of repairs to the net fabric was also noted. In the case of sewn repairs, the length of the stitch was recorded as well.

**Insecticide retention.** Because it was expected that a large fraction of recovered nets would no longer have measurable deltamethrin levels, the nets were rapidly screened using bromine x-ray fluorescence (XRF) analysis. This method is non-destructive and can be carried out with virtually no sample preparation. It takes advantage of the fact that bromine atoms in the deltamethrin molecule emit x-rays at characteristic wavelengths upon exposure to ionizing radiation. Because bromine is not abundant in the inland environment, interference by bromine-containing contaminants is unlikely and it is assumed that all bromine detected in the sample is due to deltamethrin. This assumption was verified by examining a subset of samples using both XRF and high-pressure liquid chromatography (HPLC).

XRF analysis was conducted using an Innov-X Model XT-442 analyzer (Innov-X Systems, Woburn, MA). Screening was carried out by placing the storage bag containing the net on a benchtop and positioning the analyzer window against the bag so that there was ≈2 cm of sample between the window and the benchtop. In this way multiple layers of net could be examined simultaneously to average out any local variation of deltamethrin content. Analysis through the storage bag was possible because the bag is virtually transparent to both the incident and fluorescent x-rays used in the method. In addition to the convenience, sampling the nets while they were still in the bag prevented contamination of the analyzer window with deltamethrin. Before the nets were examined, the benchtop was tested with the analyzer to verify that it had no bromine content that would interfere with the results.

For quantitative XRF analysis, standard polyester net samples (SiamDutch) were prepared having known doses of deltamethrin (0, 4.25, 8.86, 24.10, 54.97, and 114.50 mg/m²) using pulverized K-O Tab WT (Bayer Environmental Science, Monheim-am-Rhein, Germany) deltamethrin concentrate and deionized water. These standards were used in conjunction with analyzer software to measure the deltamethrin 8-layer net specimens. In each case, a 45-sec data acquisition time was used.

HPLC determination of deltamethrin was carried out on 30 × 30 cm samples of net by first extracting the net specimen in 100 mL of a 77/23 v/v mixture of acetonitrile and deionized water for 1 hr at ambient temperature in an ultrasonic bath. HPLC analysis of the extract was carried out using a BAS 200B chromatograph (Bioanalytical Systems, West Lafayette, IN) fitted with a LiChrosphere 100 RP-18e column (125 × 4 mm, 5 μm) at ambient temperature, using acetonitrile/water (77/23 v/v) mobile phase at a 1.0 mL/min flow rate, and a UV detector set to 235 nm. Injector loop volume was 20 μL, and
the typical retention time for deltamethrin was 7.8 min. A 0.0444-mg/mL solution of deltamethrin (Chem Service, West Chester, PA) in acetonitrile was used as an external standard. The results for each sample were expressed as the average of 5 injections.

Cone bioassays were conducted according to WHOPES protocol using 19–21 pyrethroid-susceptible Anopheles gambiae (Kisumu) females per test sample.

RESULTS

Description of recovered bednets. All of the 250 replacement bednets were distributed by the survey workers, and a total of 255 bags containing used bednets were collected. There is no clear explanation for the appearance of 5 extra nets, but several of the recovered bags were found to contain only partial bednets, suggesting that they had been cut by their owners to customize their use. It is possible that some bags held different pieces of the same bednet.

Of the 255 bednets recovered, 149 (58.4%) were PermaNets, 64 (25.1%) were Dawa nets, 29 (11.4%) were SiamDutch nets not having the “Dawa” label, and 13 (5.1%) were either unidentifiable, partial nets or nets from other manufacturers. All PermaNets having legible labels indicated that they were the original version, now referred to as “version 1.0,” with a treatment date of December 2001. Dawa and SiamDutch nets were not dated.

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Physical examination. Not all of the bednets were suitable for systematic examination. Out of 71 nets selected randomly
from the total, 17 (24%; mostly Dawa nets) were too small to fit on the 180 × 180 cm examination frame and 4 (6%) were judged to be too badly tattered to provide useful data (Figure 1). The 50 remaining nets were examined for holes, seam integrity, and the evidence of repairs. This set consisted of 37 (74%) PermaNets, 12 (24%) Dawa nets, and 1 (2%) SiamDutch net. The mean basis weight of 12 PermaNet, 5 Dawa, and 3 SiamDutch nets were found to be 33.5, 33.4, and 34.3 g/m², respectively. Based on WHO specifications, this indicates that these nets were all constructed with 75-denier fiber.

A total of 2023 holes ≥ 0.5 cm were found in these nets (40.5 per net). Net condition varied widely (Figure 2), with 9 (16%) nets having < 10 holes and 7 (13%) nets having > 100 holes (including the 4 tattered nets). The distribution of hole sizes (Figure 3) shows that 1056 (52.1%) holes were 0.5–1.0 cm in diameter, with incidence of large holes decreasing predictably in proportion to hole size. A total of 31 holes > 10 cm in diameter were observed (0.62 per net). Seam failures were observed in 25 (50%) of the nets.

Holes were more prevalent at the lower portion of the side panels. Figure 4 shows the incidence of holes (per m² of net area) with respect to distance from the bottom edge of the net. Deviating from this trend was the low incidence of holes 0–9 cm from the bottom edge. This is because few holes were found in the reinforced bottom border of the PermaNets. The frequency of holes on the top panel (in terms of holes/m²) was essentially the same as the frequency at the upper portion of the side panels.

Evidence of repairs was found in 32 (64%) of the nets. All of the repaired nets had open holes, so no net was completely repaired. Three types of repairs were observed: stitches, knots, and patches (Figure 5). Stitch (darned) repairs were the most prevalent, with 107 observed (3.3 per repaired net). Some repairs were made by gathering the fabric and tying it into a knot to close a hole (13 knots observed; 0.4 per repaired net). Only one net was patched, having patches in 10 locations.

**Insecticide retention.** XRF screening results (Figure 6) show that the majority of nets have little or no remaining deltamethrin while a few nets retained a substantial dose. To verify that these results accurately reflect deltamethrin content, a 30 × 30 cm specimen was cut from each of 12 PermaNet samples, 6 having low XRF readings (to test for false negatives) and 6 having significant XRF readings (to test for false positives). Quantitative XRF analysis was done on the sample (folded into 8 layers), followed by a cone bioassay and HPLC analysis.

Close agreement between quantitative XRF and HPLC results is seen (Figure 7). The absence of spurious bromine contamination is evident by the lack of points appearing below the 1:1 trendline. Furthermore, cone bioassays show full insecticidal activity only in samples cut from bednets having a relative deltamethrin content of 500 or higher (Figure 8), corresponding to 14.9% of the bednets recovered.

**DISCUSSION**

LLINs offer protection from malaria by imposing both a physical and a chemical barrier to mosquitoes. This report documents the degree to which both of these barriers can become compromised during normal household use. This study was extremely limited, focusing only on polyester nets (Dawa and PermaNet 1.0) that are no longer on the market and examining the nets after only one time interval, in only one geographic setting. A similar, but more comprehensive, study of current nets in a variety of climatological and demographic settings would yield data useful for guiding future purchase decisions and the development of improved bednets.

The quantity and size of holes and the frequency of seam failures observed in this study suggest the need to raise the performance requirements for LLINs in terms of both fabric quality and seam strength. Because these nets were probably

![Figure 6. Distribution of relative deltamethrin content as measured by bromine x-ray fluorescence (XRF).](image-url)
used only seasonally and because an unknown number of unusable nets were probably discarded and therefore unavailable for this study, it is reasonable to assume that the development of holes and other deterioration occurred at a faster rate than the 38-month period suggests.

Other published studies addressing the physical deterioration of nets include that by Maxwell and others, who categorized polyester nets (70 denier) distributed in Tanzania as “intact” if they met the following criteria: < 20 holes, < 2 cm in diameter; < 5 holes, 2–5 cm in diameter; and < 2 holes, > 5 cm in diameter. Using this definition, 84.8% of the 4842 nets distributed in highland villages were intact 4–5 years after distribution, but only 56.6% of 5238 nets distributed in lowland villages were intact 3–4 years after distribution. This suggests that net longevity depends strongly on where they are being used.

A separate report by Shirayama and others investigated Olyset nets distributed in Lao PDR. After 2–3 years, 40% of these nets showed physical damage (holes and tears). Although this result suggests that polyethylene-based Olyset nets are more durable than polyester nets, a side-by-side study of the 2 types of nets would be necessary to remove bias introduced by geography, climate, and demographics.

Although the current study did not address the causes of hole formation, this issue deserves mentioning. Although some holes were caused by burns, as evidenced by melted fiber and localized heat shrinkage (Figure 9), most holes showed no such signs and were often associated with runs (Figure 10). The latter holes are believed to be caused by the fabric snagging on a sharp point (e.g., a bedframe corner, tree branch, etc) causing only 1 or 2 yarns to break. The resulting hole, though small, is a weak spot in the fabric. Further stresses around this weak point can cause the hole to enlarge. Enlarged holes, in turn, are more likely to catch on objects and become even larger. The distribution of hole sizes in this study is consistent with this scheme of random small hole formation followed by gradual enlargement. With regard to fabric integrity, current WHO testing guidelines address only bursting strength and do not consider its resistance to this mechanism of hole development.

Insecticide treatment is often regarded as backup protec-
tion in the event holes develop in a bednet. Because formation of holes is concurrent with insecticide loss, it is important to take both into account when determining the useful life of an LLIN. Lines and others\textsuperscript{10} demonstrated in experimental hut studies that permethrin-treated (200 mg/m\textsuperscript{2}) bednets having 8, 10 \times 20 cm holes could provide equivalent protection as an intact untreated bednet. Curtis and others,\textsuperscript{11} also using experimental huts, later showed that nets treated with cyfluthrin or lambda-cyhalothrin and having 6, 4 \times 4 cm holes significantly inhibited blood feeding and killed mosquitoes even after 15 months of domestic use. Other experimental hut studies\textsuperscript{12–14} of insecticide-treated nets have been conducted using nets having many small holes to make the study more realistic. Maxwell and others\textsuperscript{15} also demonstrated in field studies in Tanzania that physically damaged nets can still reduce parasitemia rates if the nets are retreated periodically.

Although the bednets in this study suffered extensive damage and insecticide loss during 38 months of household use, these findings and the evidence of repairs do confirm that, despite having little prior history with bednets, the population of Lawra District used the LLINs provided by the 2002 campaign. Ensuring that bednets that demonstrate long-lasting protection are made available continues to be the challenge.

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