Estimating functional survival of long-lasting insecticidal nets from field data

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Introduction
The durability of Long-Lasting Insecticidal Nets (LLINs) in the field has become a critical issue for the success of malaria control in areas where LLINs are being applied for malaria prevention for two main reasons: first, it has been shown by various modeling exercises [1] that increasing LLIN durability by one or two years on average would have a huge impact on the cost of malaria prevention, in the order of 500-700 million US dollars over 5 years; and second, there are increasing data suggesting that there is a wide variation of LLIN performance between different locations or populations. This implies a need to acquire country or region specific data on LLIN performance to feed into management decisions.

Such notion of “value for money” is particularly important in an environment of increasingly scarce resources [2] and has resulted in efforts to modify the practice of basing LLIN procurement decisions on price alone. For example, a meeting of African Ministers of Health during the World Health Assembly 2012 entitled ‘Optimizing funding for LLINs in a difficult financial environment’ concluded that “malaria endemic countries request that major donors such as Global Fund, PMI, and World Bank adopt policies that allow countries to select LLINs based on cost-effectiveness, incorporating both price and net performance” [3].

Procurement decisions for LLIN have to be based on verifiable criteria without exclusion of any brand that has obtained at least interim WHOPES recommendation irrespective of whether field data for such a product is available or not. Therefore, laboratory textile testing using an improved suite of tests currently under development will be the primary basis for procurement decisions. However, field data on LLIN survival will continue to be important for the validation of textile testing algorithms and to support country program management regarding number of LLINs needed to sustain universal coverage.

Significant progress has been made towards such performance-based procurement with the release in 2011 of the “Guidelines for monitoring the durability of LLINs under operational conditions” by WHO [4], which not only addresses some of the methodological issues but also encourages countries to incorporate assessment of LLIN performance as part of their distribution efforts. Furthermore, the “Guidelines for procuring public health pesticides” [5] issued by WHO in 2012 clearly states the importance of “value for money” and that “for LLINs, the criteria for comparison could be ‘cost per median year of net life under local conditions of use’” (page 20).

However, the current guidelines are not comprehensive enough to allow countries that have already started to collect data on LLIN performance to translate their findings.
into the required “median LLIN survival”, and an expansion of these guidelines is therefore urgently needed.

This document addresses primarily those program staff and researchers directly involved in the collection and analysis of LLIN durability data and should always be considered in conjunction with the existing guidance on the topic [4, 6]. It summarizes the current recommendations of durability measurement in the field and outlines where gaps still exist. It includes a description of the rationale to fill these gaps leading to simple and easy-to-use recommendations that can be used to obtain relevant data while further research is undertaken to fill existing gaps. In order to keep the document concise, supporting evidence for the approach chosen is presented in the annex, jointly with some practical tools for the field data collection and analysis. Finally, outstanding issues are summarized and recommendations for next steps provided.

Current recommendation on methodology and existing gaps
The “Guidelines for monitoring the durability of LLINs under operational conditions” [4] represent the most critical progress in recent years towards understanding and measuring durability of nets in general, and LLINs in particular. These guidelines highlight the need for local data on net performance in the field, since performance is expected not only to vary between products but also based on local conditions and user behaviors. The document clearly defines the three core elements of durability that need to be considered:

- **Survival/attrition**: net survival rate, sometimes also referred to as retention, is the proportion of distributed nets still present in the receiving households after a given time; the opposite of this rate – the loss – is the attrition rate. Within attrition, two major sub-groups are distinguished: loss due to destruction or discarding of the net (including use for alternative purposes), and “loss” due to the net being used at a location different from the original recipient household (i.e. nets that are stolen, sold or given away).

- **Physical or fabric integrity**: the physical condition with respect to holes and open seams of those nets found in a household after defined time intervals of use (e.g. annually) that have not been discarded or lost. These are referred to as the surviving nets.

- **Insecticidal activity**: the presence of effective insecticide on the LLIN is primarily measured by its effects on susceptible anopheline vectors through bio-assays such as cone or tunnel tests with the outcomes of knock down, mortality and feeding inhibition. The presence of insecticide is also captured by
Chemical residue analysis measuring the concentration of the insecticide in g/kg or mg/m$^2$ remaining at recommended time points.

The document outlines the two principle approaches to measure the three core elements of durability: first, cross-sectional household surveys (also called retrospective surveys) following up nets that were previously distributed, e.g. through a campaign; second, longitudinal or prospective surveys that actively distribute LLINs and follow them up over time assessing all three components at various time points. The main advantages and disadvantages of these two study designs can be summarized as follows:

- **Prospective studies** are the first choice, as they have the key advantage of allowing full control of distribution of LLINs and close monitoring of all outcomes, especially attrition and integrity. They have, however two disadvantages:
  - They take at least three years before reliable estimates of performance can be made.
  - In a “real life situation”, net users will discard a net when they feel its condition is so poor that it no longer serves its purpose. In a prospective study, however, where an ID-number is attached to the net and users are aware that the net will be followed up over time, they tend to keep a damaged net much longer than they normally would. This phenomenon is called the Hawthorne effect\(^1\), and leads to a lower attrition rate in prospective studies compared to retrospective surveys, as well as poorer physical conditions of the surviving nets. There is now increasing evidence that this is indeed the case (see Annex 1), meaning that the relative contribution of attrition and physical integrity to LLIN survival is different in a prospective study than in a real life situation.

- **Retrospective, cross-sectional studies** can provide information immediately, but have the following limitations:
  - They require sites where ideally only one net distribution has taken place at least one (and not more than three) years ago.
  - They rely on recall about nets received and what happened to those no longer present, or detailed lists of the distributions to specific households – either of which can be unreliable.

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\(^1\) The Hawthorne effect describes the change of behavior of study subjects by merely observing them or giving them attention. It is named after studies of worker behavior in the Hawthorne Works plant of Western Electric near Chicago, USA, in the 1930s.
The existing gap

The “Guidelines for monitoring the durability of LLINs under operational conditions” in conjunction with the recently updated “Guidelines for laboratory and field testing of long-lasting insecticidal mosquito nets” [6] provide clear recommendations on how each of the three core elements of durability should be measured:

- Reason for loss (attrition) through a questionnaire -- currently only designed for prospective studies.
- Physical integrity through the counting of holes in four hole size categories and summary of the counts in a proportionate Hole Index (pHI) weighted by the approximate surface area of the holes to provide a single measure of damage per net.
- Effectiveness of insecticide through standard bio-assay tests and chemical residue analysis based on appropriate sampling from each net.

But there is as yet no recommendation on how to combine measurement of the different components into one single assessment of durability or “median net survival” that could be used to compare data across studies and sites, and that could also be used to inform procurement decisions. This absence of recommendation was not by oversight but rather because at the time of writing the experts did not see sufficient evidence on some of the key questions to provide such a recommendation (see also section on outstanding issues). However, with the increasing focus on durability and the pressure to include these performance aspects as part of the considerations for procurement, there is an urgent need to review existing and emerging evidence and provide methods on how such a durability metric could be constructed – even if it is preliminary and needs to be revised at a later time once more comprehensive evidence is available (e.g. on the relationship between level of damage, insecticide residue and epidemiological protection).
Rationale for estimating functional net survival from field data

**Combining attrition and integrity**

There is growing evidence that any assessment of durability must always include at least attrition and integrity to be of any value (see annex 1 for details) because

- Integrity data of surviving nets may actually appear to improve over time as damaged nets are increasingly discarded.
- The contribution of each component, attrition and physical integrity, varies significantly between prospective and retrospective studies and between sites, and may also vary among different LLIN products.
- Attrition due to discarding can be “premature” if nets based on current evidence can still be considered in “serviceable” condition at the time they are discarded, and such loss is driven by people’s behavior rather than the qualities of the LLIN product.

Furthermore, only that part of attrition should be included in the calculation that directly links with the durability of the nets. The current guideline correctly distinguishes between loss by giving away the net to others to use either voluntarily (given to friends and relatives or sold) or involuntarily (stolen), and loss due to destruction, discarding or using it for alternative purposes because it is perceived as “useless”. However, while knowledge of the overall survival or retention rate certainly is of interest programmatically in order to know more about the movement or “social life” of the nets, it can be misleading when included in the durability estimation because:

- The rate of giving away nets very much depends on the supply situation (i.e. whether or not the household has enough, or more than enough, nets to cover all family members) as well as the general attitude towards sharing with others in the family, extended family or community. Neither of these considerations is directly linked to the qualities of the LLIN product, although the acceptability or preference for a type of net could influence the decision of which net to give away provided more than one brand was distributed to the same household (which is uncommon in campaigns and not recommended by the guideline for prospective studies) [4].
- The nets given away are still being used by someone, even if it is not the original recipient, and it is safe to assume that not all nets are discarded immediately. Rather, it is likely they are exposed to the same or similar rates of decay, although this cannot be confirmed in the survey. Including them as “lost” in the calculation therefore overestimates true losses and underestimates durability.
Therefore, only the attrition due to destruction and discarding or alternative use should be included in the estimation of functional LLIN survival (see interim recommendation below). Nets received but then given away for others to use should be excluded from the denominator as “outcome unknown”. A sample questionnaire module that can be used to collect such data in retrospective surveys is attached as annex 2.

**Categorization of proportionate Hole Index results**

The most crucial consideration in estimating LLIN durability is the question on how to utilize the proportionate Hole Index (pHI) data. The two main issues are the following:

- The pHI is an excellent and quite robust method to capture field data on the physical condition of a net into one single metric, but like many other parasitological data, it tends to be very skewed to the right (positive skew) with the vast majority of results near zero or very low, and very few with high values (see annex 1). With such non-normal distribution, the arithmetic mean is statistically a very poor measure of “central tendency” as it will depend on just a few high values which could be outliers. The median, on the other hand will – by definition – remain zero until at least 50% of all nets have any holes, which could be after 2-3 years and will also not well distinguish product performance. In addition, the shape of the distribution will change as the net cohort ages until a steady-state between rate of decay and rate of discarding is reached.

- The pHI value alone, even if statistically different for one product compared to another, does not in itself allow a determination of “end of useful life” unless a reference of what is still acceptable and what is not is developed. Furthermore, the absolute value of the pHI depends on the weights that are used, and these had changed over time before the current recommendation was reached, and may change again in the future. This makes differently calculated data impossible to compare.

The solution to the problem is to use pHI cut-offs that distinguish the following categories:

1. LLIN in “good” condition where there is no reduction of efficacy compared to an undamaged net
2. LLIN in “acceptable” condition in the sense that their effectiveness is somewhat reduced but still provide significantly more protection than no net at all.
3. LLIN “torn” where its protective efficacy for the user is in serious doubt and the net should be replaced as soon as possible.
These cut-offs should refer to an objective measure such as the “approximate total hole surface area”\(^2\) so that they can be:

- calculated for any weighting system of the pHl making results directly comparable irrespective of weights as long as they have some relationship to the size of the holes.
- easily adjusted once better data is available and additional knowledge of “how much damage on an LLIN is too much” becomes available.

Based on careful scrutiny of currently available evidence (see annex 1) the following categories and cut-offs are suggested (Table 1) using the weights/hole size category as recommended and described in detail by the guidelines for monitoring durability of LLINs [4,6]. These weights are based on the ratio of the average assumed surface area for each size category when compared to the smallest category\(^3\).

The 100 cm\(^2\) hole surface area for a "good" ITN is based on strong evidence from hut trials that there is no reduction of feeding inhibition compared to an intact ITN at that level. The evidence on the upper limit of 1000cm\(^2\) is less strong as not studied well, but there is reasonably good evidence that there still is protection/feeding inhibition between 900 and 2000cm\(^2\) hole surface area and therefore the approximate lower limit of this range has been chosen. These hole surface area values were then back-translated into pHl values based on the surface ratio from the table.

Table 1: Suggested categorization of proportionate Hole Index data

<table>
<thead>
<tr>
<th>Category</th>
<th>pHl value range</th>
<th>Approximate total hole surface area in cm(^2)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>If circle(^<em>) If rectangular(^</em>)</td>
</tr>
<tr>
<td>Good</td>
<td>0-64</td>
<td>&lt;79  &lt;100</td>
</tr>
<tr>
<td>Damaged</td>
<td>65-642</td>
<td>80-789 100-1,000</td>
</tr>
<tr>
<td>Too torn</td>
<td>643+</td>
<td>&gt;790  &gt;1,000</td>
</tr>
</tbody>
</table>

\(^*\)refers to the assumed functional shape of the hole

Such a categorization avoids the issues around the non-normal distribution of the pHl values and by creating dichotomous (yes/no) variables, allows a statistically straightforward analysis of various outcomes over time and between LLIN products. However, for such an analysis one additional category is needed, namely, the proportion of LLINs “serviceable” which comprises the categories of “good” and

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\(^2\) This assumes that the absolute hole surface area determines the ease of entry by the vector, not the relative size of holes to the total net surface, but evidence for this does not yet exist.

\(^3\) Weights are 1, 23, 196 and 576 for the four size categories of holes 0.5-2 cm, 2-10 cm, 10-25 cm, 25+ cm.
“acceptable” nets. These can be expected to still provide good or acceptable protection.

An additional advantage of this categorization is that it minimizes the misclassification of outcomes based on the variation or inaccuracy of the hole counts. Such misclassification inevitably occurs in large-scale surveys where integrity is measured in the field rather than under “laboratory” conditions with nets positioned over a frame to get an exact count and measure of the holes. The categorization, therefore, increases the robustness of the integrity metric.

**Incorporating insecticidal effectiveness**

The question of how to deal with insecticidal protection in the context of estimation of functional net survival remains challenging. The three main reasons are:

First, there is as yet no clear definition of the minimal effective dose still acceptable and its equivalent in bio-assay tests. The WHOPES cut-off of 80% of nets effective in WHO cone bioassays (with ≥95% Knock-Down or ≥80% mortality) or tunnel tests (≥80% mortality or ≥ 90% blood-feeding inhibition) is designated as the optimal level required after three years of use to decide on recommendation for public health use, not as an end-of life determination or a minimally acceptable performance. Existing data suggests that high Knock-Down rates and mortality can be achieved at very low levels of insecticide [7-11], much lower than what has been seen after three years in WHOPES Phase III evaluations (see WHOPES reports).

There is evidence that insecticide content and bio-assay results do not dramatically fall off after three years, but rather continue to decline at the same rate seen in the first three years [12]. In the early days of WHOPES Phase III testing WHO (Pierre Guillet, personal communication) had recommended a “minimal effectiveness” based on cone bio-assay tests of ≥75% KD or ≥50% mortality. This criterion had then been used in the field [13] establishing corresponding levels of chemical residue for a susceptible malaria vector strain. However, these minimal effectiveness criteria never appeared in any WHO guidelines. This issue should be revisited in upcoming discussions (see also 1st bullet under outstanding issues).

Second, bio-assays are difficult to do in the field. When done in an entomological laboratory they need – like chemical residue analysis – destructive sampling, meaning that the nets have to be removed and can no longer be followed. In addition, both tests are quite expensive and not available everywhere. As a result, usually only a small sub-sample of 30-50 nets is tested [6]. This excludes a determination of insecticidal outcome for each net in a cohort, which would be needed for a net survival estimate.

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4 Even if the exact count of holes from two surveyors on the same net differs somewhat, the net will in the vast majority of cases still fall in the same category of damage.
Third, field applicable tests for pyrethroid residue that allow a quantitative or at least a semi-quantitative assessment of a minimal dose would solve the problem if they can be shown to correlate well with the bio-available insecticide. A variety of such tests have been developed [14-19], but none of them is ready for general application in “routine” surveys.

In view of these challenges the best immediate solution is the following:

- Current WHOPES evaluations for LLIN include in phase II wash resistance and regeneration criteria in the laboratory. In addition, the phase III field testing assesses the performance of each LLIN brand after three years of field use requiring that at least 80% of surviving nets exhibit optimal bio-efficacy [6]. This usually includes the performance of the LLINs in several locations and settings as part of the evaluation. It can, therefore, be assumed that if a product has received WHOPES recommendation, there is sufficient evidence that insecticidal protection will last for at least three years. On this basis, it is recommended to exclude insecticidal effectiveness for the time being from the estimation of LLIN survival and base estimations on measures of attrition and integrity only

Once methods are available that allow the determination of the “insecticidal effectiveness” in the field (thus enabling all LLINs in the survey sample to be tested), incorporation of the insecticidal component can be re-considered.

Including precision of estimates

The final consideration for the estimation of functional LLIN survival refers to the need to include a measure of uncertainty or precision. This is part of the efforts to provide standardized, quality assured and sound evidence of LLIN performance that can be used in decision making. As has been outlined above, measurement of attrition will rely on respondent recall and hence will be subject to recall bias, especially in retrospective surveys. Similarly, measure of integrity and insecticidal effectiveness is prone to measurement errors. Therefore, for all estimates an adequate confidence interval should be calculated (at 95% or at least 90% level) in order to ensure that decisions are not based on chance variations. Such confidence intervals must take into account the sampling strategy, such as stratifications or over-sampling by applying sampling weights and adjustment of confidence intervals for a possible design effect.

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5 Monitoring residual efficacy by control programmes must always be encouraged and capacity to do so must be strengthened. However, until there is a method to test every net sampled in the field and enough evidence to define a cut-off for “failure”, when monitoring LLIN durability, the most important parameter should be physical integrity.
**Recommendation for calculating functional LLIN survival**

Based on the rationale outlined above and available evidence to date summarized in annex 1, the following recommendations are made for the estimation of functional LLIN survival. The analysis process is divided into two distinct steps:

- Estimate the “proportion of LLINs surviving to time x” from attrition, integrity and (once a minimal insecticidal effectiveness is defined) bio-assay and/or chemical residue data for each point in time for which data was collected.
- Plot these survival estimates of the LLIN against time of follow-up, compare results to reference curves of hypothesized survival assuming various median times of survival, and calculate “median LLIN survival” from two or more data points in time (as explained in detail below).

For all data handling and analyses the following general recommendations should be followed:

- Physical LLIN integrity should always be captured as count of holes by size category so that re-assessment can be done later should recommendations on weighting of the pHI change.
- All nets for which no definite outcome (attrition or integrity) can be determined during the field work for whatever reason (e.g. incomplete data or data entry error), as well as all nets which are still in the original package and have never been used at all should be censored and omitted from the analysis.
- For all survival estimates appropriate confidence intervals should be calculated taking account of the sampling design and using appropriate statistical procedures such as adjusting confidence intervals for the design effect and applying sampling weights in cases of unbalanced samples.
- Recommendations to calculate LLIN survival apply equally to prospective and retrospective data collections. However, based on the larger potential for recall bias in retrospective surveys, more than one data point in time (i.e. multiple surveys) should be used and/or results triangulated with prospective data from the same area.
- In order to minimize recall bias of “nets received from campaign” in retrospective studies, the identification of LLINs from a campaign should not only rely on the statement of the respondent but should include other independently collected information such as brand, source and age of net. In other words, a net claimed to be a campaign LLIN but which is not the brand that was distributed during that campaign or which was obtained before or far after the campaign date should not be counted as “received from campaign”.

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1. Estimate proportion of LLINs functionally surviving at time x

Given the current difficulties in determining a definite outcome regarding insecticidal effectiveness for each LLIN in the sample outlined in the previous section, the estimation is based on attrition and integrity data alone.

The indicator of “proportion of LLINs surviving to time x” after distribution is calculated as:

\[
\frac{\text{# of LN present and “serviceable” at time } x}{\text{# of LN originally received and not given away at time x}} \times 100
\]

As shown in Figure 1 below the numerator includes all LLINs that are still present from the original distribution and in a “serviceable” physical condition, i.e. not “too torn”, and the denominator includes all nets from the original distribution for which a definite outcome could be determined but excludes LLINs that have been reported as stolen or given away to others.

Figure 1: Composition of numerator and denominator for functional survival estimates

2. Estimating median survival time

Median LLIN survival is the time point at which the estimate of functional LLIN survival crosses the 50% mark. This requires follow-up of nets for a long time (prospective surveys) or carrying out a retrospective survey many years after the distribution. In order to facilitate interpretation of earlier data points, it is recommended to plot survival estimates with their corresponding confidence intervals against time in years (at least one decimal point) using a scatter plot where both x and y axis are continuous. These results can be compared against hypothetical survival curves showing the most likely S-shape of survival (see annex 1) with a range of median
survival times. An example is shown in Figure 2 and annex 3 provides the table of values to create such curves.

**Figure 2:** Example of (fictitious) LLIN survival data from prospective or retrospective surveys presented as survival curve and compared against hypothetical survival curves of defined median survival

If at least two data points in time are available, the median functional survival time can be estimated as:

\[
t_m = t_1 + \frac{(t_2 - t_1) \times (p_1 - 50)}{(p_1 - p_2)}
\]

Where \( t_m \) is the median survival time, \( t_1 \) and \( t_2 \) are the first and second time points in years, and \( p_1 \) and \( p_2 \) are the proportion surviving to the first and second time points respectively.

Using results from time points 2 and 3 of the (fictitious) data presented in Table 2 below, the calculation would be:

\[
t_m = 1.7 + \frac{(2.4 - 1.7) \times (83.4 - 50)}{(83.4 - 68.2)} = 3.2
\]

The confidence interval of the median net survival can be obtained by applying the formula to the lower and upper limits of \( p_1 \) and \( p_2 \) respectively.

However, data projected forward beyond the actual observation always have to be interpreted very carefully. Also, due to the initially flatter shape of the anticipated
survival curve (see Figure 2), projections toward the median should only be attempted if the first time point is at least 85% or lower. To demonstrate this point, the $t_m$ estimates for the (fictitious) data used in Figure 2 are presented below.

**Table 2:** Example of estimates of median LLIN survival using fictitious data

<table>
<thead>
<tr>
<th>Time point</th>
<th>Time in years</th>
<th>Functional survival</th>
<th>Median survival using last two data points (95% CI)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>1.0</td>
<td>93.3%</td>
<td>n.a.</td>
</tr>
<tr>
<td>2</td>
<td>1.7</td>
<td>83.4%</td>
<td>4.1 (3.7 to 4.5)</td>
</tr>
<tr>
<td>3</td>
<td>2.4</td>
<td>68.2%</td>
<td>3.2 (3.0 to 3.5)</td>
</tr>
<tr>
<td>4</td>
<td>3.1</td>
<td>53.2%</td>
<td>3.3 (3.0 to 3.5)</td>
</tr>
<tr>
<td>5</td>
<td>4.3</td>
<td>31.6%</td>
<td>3.3 (3.0 to 3.6)</td>
</tr>
</tbody>
</table>

It is also recommended to include, wherever possible, more than two data points in time to reliably estimate functional LLIN survival.

In order to facilitate analysis of such data, a tool to prepare survival graphs and calculate median survival times will be made available to countries and partners.

**Outstanding issues**

The most important outstanding issues are:

- To ensure a high level of standardization, there is a need to develop standard operation procedures (SOP) for all steps of LLIN survival estimation surveys, make available standardized tools and templates, and establish strict norms of quality control.
- To include durability performance into the WHOPES evaluation criteria for LLIN and thereby create a stimulus for manufacturers to improve performance of their products, WHOPES should consider a minimum requirement of “x% of LLIN that survive to three years of field use in ‘serviceable’ condition”.
- To understand better the determinants of mosquito entry into a damaged net and to improve – if needed – the weighting system for hole counts in the proportionate Hole Index, there is a need to study the relationship between hole size and position on an effective LLIN and the influence of total net size compared to the size of the hole.
- To define a) the cut-offs to be used to determine “end of useful life” and b) how the cut-offs need to be adjusted with increasing vector resistance, there is a need to explore the relationship between net damage, remaining insecticide and feeding inhibition in susceptible and resistant vectors in hut trials.
- To establish the epidemiological impact of damaged LLIN, there is a need to study the impact of damage, age and insecticide levels of LLIN on malariometric parameters in children and adults (malaria incidence, prevalence and anemia).
In order to include insecticidal effectiveness in the future, it is important to accelerate the development of field tests that reliably predict protective effectiveness of the LLINs.

**Conclusion**
This technical guidance document provides to country programs and partners a method for calculating the functional survival of LLINs from field data obtained from prospective or retrospective surveys, as well as a method to estimate the median survival time of LLINs. It is based on the best available evidence to date. Some gaps in knowledge still exist; these methods will need to be reviewed and revised in the future as more data become available.

**Next steps**

**Countries**
- Analyze available data according to this document as long as all necessary input is available
- Include in country work plans the collection and analysis of data on LLIN survival according to existing recommendations and guidance
- Share the results from LLIN survival analyses with other partners so that a better understanding of the dynamic of LLIN survival can be obtained
- Where sufficiently reliable information exist, include this in their planning for malaria prevention using LLINs

**Partners**
- Support countries in the collection and analysis of LLIN survival data directly and by building capacity
- Undertake research to address the outstanding issues identified in this technical document
- Actively contribute to improvement of this document and these methods in the future

To support countries and partners, WHO will disseminate and promote this guidance note on estimating LLIN longevity and accompanying tools; facilitate the collection, analysis and sharing of results of comparable LLIN survival data by providing training and technical support; and review these methods as new information emerges.
References


Green MD et al. Evaluation of a rapid colorimetric field test to assess the
effective life of long-lasting insecticide-treated mosquito nets in the Lao PDR. 

Annex 1

Presentation of evidence behind rationale to calculate functional LLINs survival

Evidence for the need to have both attrition and integrity data

Data on proportion of lost nets given away to others to use

In the published literature to date, only one article addresses the question of redistribution of nets within the community after a campaign. Khatib et al. [1] analyzed mosquito net coverage and use in Rufiji District in Tanzania and found that for the six months following free ITN distribution to children under five, the proportion of nets obtained for free from “other sources” grew to 16% of all nets in use. The authors conclude that this most likely represents a redistribution within families or communities.

More detailed information comes from unpublished post-campaign or durability studies. In Nigeria, the NetWorks project and the SuNMaP project carried out 10 state-representative household surveys between 1.5 and 16.7 months after a LLIN mass campaign. The questionnaire sought to determine the fate of LLINs received during the campaign that were then since lost. In addition, NetWorks is undertaking a post-campaign durability study in three states in Nigeria where the same questions were asked and for which the “one-year” and “two-year” results are available. Pooling these “lost” nets provides data on 780 campaign LLINs no longer in the household.

Figure 1: Main reason for attrition by time since distribution from 780 “lost” campaigns nets from Nigeria (NetWorks & SuNMaP projects, unpublished)
As shown in Figure 1, the proportion of nets given away to others or stolen, i.e. still being used somewhere else, was over 90% of all “lost” nets in the first six months following distribution. This proportion then declined to 62.7% (95% CI 52.9 to 71.5) 12 to 17 months after distribution, and dropped again to 58.9% (50.7 to 66.9) after 24-30 months. A detailed breakdown of the reasons for attrition (Figure 2) shows that relatives were the dominant recipients of nets given away. Nets that were thrown away constituted the second most common source of attrition, followed by nets destroyed accidentally (usually by fire), with this fraction increasing after 24-30 months.

Figure 2: Detailed reason of attrition from Nigeria 12-30 months after distribution

This picture is very similar to results from an ongoing prospective study by CDC/PMI in Kenya (Figure 3) where more than 70% of “lost” nets were moved, stolen, sold or given away after 30 months and only about 25-30% were discarded or destroyed [2].

Figure 3: Reasons for attrition 30 months after distribution from CDC durability study in Kenya (John Gimnig, personal communication)
This data demonstrates that attrition due to giving away nets for others to use comprises a significant proportion at least in the first 2-3 years, and therefore justifies that these nets are not considered in the estimation of functional LLINs survival.

Data on torn nets being discarded and therefore distorting results if integrity alone is considered

The first indication that torn nets are being discarded leading to stagnation or even improvement of the measure of physical integrity over time comes from the 2008 multi-country WHOPES Phase III study on Olyset® LLINs [3]. Physical condition of surviving nets was assessed after three and five years of field use in five countries in sub-Saharan Africa. The outcome measure was “mean number of holes per net”. Although mean hole number varied between countries with a range of 11.6 to 40.7 after three years, there was no overall difference between the three and five years measure of physical integrity. While in two countries the results were the same, the mean number of holes determined in another country was lower after five years, and only in one country did it increase.

The second set of data comes from work done by the Mentor Initiative in Eastern Chad, where two different types of polyester-based and one polyethylene-based LLINs were distributed among local and refugee populations. The physical condition of these nets was followed after one, two and three years. The initial data showed a high proportion of nets “too torn” using a proportionate hole index equivalent to what is being suggested in this document [4]. The follow-up data after two years (Figure 4) showed a further deterioration, but after three years the measure of integrity seemed to dramatically improve for polyester nets and, to some extent, for polyethylene nets as well (Richard Allan, personal communication). At the same time, attrition rate increased from 7.7% in the first survey to 21.1% in the second, and 59.7% in the third, and this rate was more pronounced for polyester nets. This trend suggests that rate of discarding was higher for polyester nets, explaining the larger “improvement” in the integrity measure.
Data on “Hawthorn effect” in prospective, longitudinal studies, i.e. damaged nets are kept much longer than in situations where nets are not known to be followed-up

A direct comparison of the trend in the proportion of “serviceable” or “good” LLINs between cross sectional, retrospective and prospective longitudinal data is available from Western Uganda. Pooled data from several WHOPES III studies [5,6] between 2000 and 2011 in Kyenjojo District provides 6,998 observations on the physical condition of 1,410 polyester LLINs with follow-up times between three and 54 months. In addition, measurement of physical integrity was included in a subsample of two representative, cross-sectional household surveys in 2011 and 2012 in nine Districts in the area, including Kyenjojo (Malaria Consortium, unpublished data). Hole assessment was done for 1,905 nets (91% LLINs) using a proportionate Hole Index and cut-off values for categorization, as were used for all data in this document. Time since net was obtained from the respondent interview was used to determine age of nets in the retrospective surveys. As shown in Figure 5, the proportion of “serviceable” nets (i.e. not “too torn”) continuously declined in the prospective studies but remained almost at the same level in the retrospective data, suggesting that damaged nets were discarded. The proportion of “good” nets declined continuously over time in the retrospective data but at a much lower rate compared to the prospective data.
Previously presented data (Figures 2 and 3) on the retrospective surveys in Nigeria provide further evidence that after 12-17 months the proportion of destroyed or discarded LLINs was higher than the respective rate in the prospective CDC/PMI study in Kenya, and even more so after 24-30 months.

**Data on discarding of nets in reasonable condition**

Evidence from Ethiopia suggests that discarding of LLINs can be “premature”, i.e. discarded nets are still in “serviceable” condition, thereby masking the product attributable performance. In a cross-sectional survey, Batisso and co-workers [7] captured the loss of previously owned nets and measured the physical condition of the nets found in the households using a proportionate hole index and a categorization similar to that proposed in this document. Of nets three years or older, only 17% were “too torn” and 47% were still in “good” condition, similar to the data from Uganda in Figure 5. One third of the households had lost nets in the past three years and 25% of households had discarded nets because they were considered “too old”, but 34% of the discarded nets were only one year old or less according to the respondents. On the other hand, of the nets that were still in the household but were no longer used because they were considered “too torn”, only 44% were actually “too torn” by the PHI categorization and 14% were still in “good” condition (Figure 6). This suggests that the point at which users considered the nets as no longer usable differed from what can be considered as still “serviceable” based on existing evidence (see next section).
**Figure 6:** Physical condition of nets not used because considered “too torn” by users in Ethiopia [7]

**Evidence to decide on pHl cut-off for a “good”, “acceptable” and “torn” net**

Here data is presented to support the suggestion of a) categorizing the pHl values as the best way of analysis and b) selecting the preliminary cut-offs as presented in Table 1 in the main text.

**Data on non-normal distribution of pHl**

Evidence showing the highly skewed distribution of pHl data in retrospective as well as prospective surveys and their change over time is shown in Figure 7 using the previously described data from Western Uganda (Malaria Consortium, unpublished). In both types of surveys about 80% of assessed nets had no holes at all in the first two years, and even for older nets this proportion was still quite high although there were more damaged nets in the prospective study. The results also show the huge discrepancy between median and mean and the fact that at all time points the Inter-Quartile Range (IQR) included 0. This demonstrates that neither median nor mean is a very good tool to distinguish performance of LLINs in the field, supporting the argument for a categorization as a better approach.
Figure 7: Distribution of pH1 data for retrospective and prospective data from Western Uganda

Entomological data on protection of damaged insecticide treated nets

A considerable body of evidence exists regarding the entomological impact of damage to untreated and treated nets, mainly from experimental hut trials with vector feeding inhibition being the main outcome indicator.

As early as 1982, Port and Boreham [8] studied the effect of natural damage of untreated nets in The Gambia by inviting volunteers to sleep with their nets in an experimental hut. Many of the nets had some holes and the amount of damage was assessed by the “sum of circumference of holes and length of split in cm”. The study demonstrates two things: a) that in an untreated net, level of feeding inhibition decreases with the amount of damage; and b) that even with damage between 50 and 130 cm “hole circumference”, there was still some protection compared to no net at all, with feeding inhibition between 25% and 73%.

A similar result was shown by Irish et al. [9] in experimental hut trials in Benin using different levels of artificially produced holes and a pyrethroid-resistant strain of Culex quinquefasciatus as vector. They found that untreated nets with 96 cm² total hole surface (6 holes 4 cm x 4 cm) had 60% feeding success compared to 36% in intact untreated nets and this increased to 69% with 320 cm² hole surface (40 holes 2 cm x 2 cm). The authors demonstrate that treatment...
with insecticide (40mg/m² alpha-cypermethrin) does not provide much protection if the vector is resistance. Compared to the untreated nets, feeding inhibition was only 16-27%. In other words, there was no increasing loss of protection with increasing damage.

Darriet et al. [10] assessed blood feeding success of *Anopheles gambiae* s.s. in hut trials in Côte d’Ivoire comparing intact untreated nets to damaged untreated nets into which 225 holes of 2 cm x 2 cm were cut (0.8% of net surface and 900 cm² of total hole surface). Results show that feeding success almost tripled, increasing from 24% for intact nets to 68% for damaged nets. Damaged nets were still slightly better than no net at all, however, which resulted in 83% feeding success.

The fact that any protection added by the pyrethroid in damaged nets is lost if the vectors (*Anopheles gambiae*) are resistant was also impressively demonstrated by Asidi et al. [11]. Using naturally damaged nets from villagers in South Benin and treating them with 18 mg/m² lambda-cyhalothrin before the experimental hut trial, they showed that no feeding inhibition at all could be achieved with resistant vectors. This was true regardless of whether the mean total hole surface was less or more than 15 cm². However, the study also demonstrates that with sensitive strains the pyrethroid treatment provided excellent protection even with damage up to 49 cm² hole surface.

Assuming then that a vector is sensitive to pyrethroids, what is the limit at which a sufficiently treated ITN or LLIN still provides “good” protection and can be considered equal to an intact ITN? Vatandoost et al. [12] showed in a tunnel test with *Anopheles stephensi* and treatment with lambda-cyhalothrin that a small amount of damage (9 holes 1 cm x 1 cm or 9 cm²) provided almost complete feeding inhibition (97.5%-100%) even if the dose was reduced to 2.5mg/m³, 12.5% of the WHO recommended dose.

There are three studies that demonstrate that artificial holes of 96 cm² (6 holes of 4 cm x 4 cm) in ITN provide excellent feeding inhibition in hut trials. Curtis et al. [13] used cyfluthrin (50mg/m²) and lambda-cyhalothrin (10mg/m²) and found that even after 15 months of field use, i.e. considerably reduced levels of insecticide, these holed ITN still had only 20% to 25% feeding success. Similar results were obtained by Maxwell and co-workers [14] with alpha-cypermethrin and lambda-cyhalothrin showing that a dose of 10mg/m² alphacypermethrin worked as well on these holed ITN as 40 mg/m². Finally, Malima et al. [15] treated nets with 20mg/m² alphacypermethrin and tested them with 96 cm² of hole surface immediately and after 20 months of field use. Initial feeding inhibition was 69% and after 1.5 years was still 30%.

The question then is what can be defined conservatively as an upper limit of acceptable protection, and here only two studies from hut trials are available to date. Pless et al. [16] studied nylon nets treated with various formulations of permethrin in The Gambia, and instead of 6 holes of 4 cm x 4 cm they cut 6 holes of 10 cm x 10 cm into the nets to produce a total hole surface of 600 cm². For all treatments they found a 80-90% feeding inhibition compared to untreated nets using susceptible *A. gambiae* even after three washes. Carnevale et al. [17] undertook three hut trials in Burkina Faso and Congo Republic with nets treated with either
permethrin or deltamethrin and deliberately damaged to various levels. They either cut 50 or 70 holes sized 10 cm x 10 cm (i.e. 500 cm$^2$ and 700 cm$^2$ total hole surface) or 80 holes of 5.5 cm x 5.5 cm, equivalent to 2,420 cm$^2$ or between 0.5% and 5% of the total net surface. In the first trial in Burkina (Soumousso) they found a 62% reduction in *A. gambiae* and a 67% reduction of *A. funestus* caught in hut traps comparing permethrin treated nets with 500 cm$^2$ or 700 cm$^2$ hole surface to equally holed nets without insecticide. In the second trial in Burkina Faso (Vallée du Kou), nets with 2,420 cm$^2$ hole surface were treated with either 25 mg/m$^2$ deltamethrin or 50 mg/m$^2$ and compared to equally holed nets without treatment. Man-biting inhibition of the holed treated nets was 32% for the lower dose and 44-56% of the higher dose, with vector mortality (*A. gambiae*) varying between 73% and 93%. In the final trial in Congo Republic, nets received holes of either 560 cm$^2$, 1,125 cm$^2$ or 2,250 cm$^2$ hole surface and were treated with either 25 mg/m$^2$, 12.5 mg/m$^2$ or 6.25 mg/m$^2$ deltamethrin. Results showed a 64-67% reduction in man-biting of *An. gambiae* at the highest dose and 57-62% reduction at the middle dose irrespective of the size of holes. Only at the lowest deltamethrin dose was biting inhibition seen at moderate levels (37% reduction) in nets with 560 cm$^2$ hole surface, and there was no reduction at higher levels of damage.

**Epidemiological data on impact of net damage on parasitaemia**

Data on the epidemiological impact of damaged nets or LLINs is more scarce, with only four published studies (and one submitted for publication), all of which looked at the parasitaemia of children as a function of the condition of the net they used. For example, a study conducted in Vietnam looked at the association of hole size with risk of malaria infection. Owners of nets with larger holes (>20 cm$^2$) have a high risk of malaria infection (OR 1.36; 95% CI 0.55-3.39) than owners of nets with smaller holes (0.25-19.9 cm$^2$) (OR 0.87; 95% CI 0.32-2.35) or no holes (OR 0.88; 95% CI 0.39-2.02)[18]. In addition, one modeling study was published which suggests that the functional survival of the LLINs (i.e. reducing transmission) not only depends on the physical and chemical decay but also on the transmission intensity, and could be significantly longer in low transmission settings. [19]

Clarke and co-workers [20] studied the effect of net condition on child parasitaemia in 1996 in 48 villages in The Gambia. Most of the nets used by 914 children 6-59 months of age were untreated, with only 58 being effective ITN. Nets were categorized as in “good condition” if they had less than 5 holes of not more than 2 cm in diameter, which is 6-8 cm$^2$ total hole surface. Any net with more holes was considered in “poor condition” without further differentiation. They found that untreated nets even with a few small holes, i.e. in “good condition”, still had a 51% protective effect against *P. falciparum* parasitaemia and that the best protection was availed by ITN irrespective their physical condition, with 69% protective effect.

Maxwell and colleagues [21] studied parasite rates in children 6 months to 12 years of age in eight villages in Tanzania in two surveys (1999 and 2000), where in three villages nets were provided and regularly treated with alpha-cypermethrin. Nets were assessed for their physical
condition and categorized as “intact” if they had fewer than 20 holes of <2 cm diameter, fewer than 5 holes <2-5 cm or fewer than 2 holes >5 cm, which can be estimated to be around 30-35 cm² total surface area. Malaria parasite rates in children 6-24 months, 24-59 months and 6-12 years showed absolutely no difference for children using “intact” ITN or ITN with >35 cm² hole surface in the 1999 survey, but a 25-38%point reduction compared to villages without nets. In the 2000 survey, children under 5 showed a trend to acquire even lower parasite rates if they used “torn” ITN, with only the 6-12 year age group showing a 13%-point increase in parasitaemia if the ITN was torn.

Rehman et al [22] examined the relationship between net damage and parasitaemia in two rounds of household surveys in 2009 and 2010 in Equatorial Guinea and Malawi involving a total of 14,807 children less than 15 years of age. Nets included untreated nets, conventionally treated ITNs and LLINs. Physical condition was not assessed as recommended by WHO [4], but rather categorized on the basis of whether or not any holes were present, and whether any present holes were larger or smaller than about 3.3 cm (D-size battery). Overall parasite rates did not vary dramatically between intact LLINs or ITNs (34%), slightly damaged LLINs or ITNs (30%), and LLINs or ITNs with larger holes (33%). However, in a logistic regression analysis adjusting for confounders, intact and slightly damaged LLINs or ITNs had a significant protective effect compared to no net or a torn untreated net (OR 0.65, 95% CI 0.55 to 0.79), while more damaged LLINs only had a marginal protective effect (OR 0.95, 95% CI 0.71 to 1.28). No direct comparison between torn and intact LLINs is provided, but it can be estimated that the OR for more damaged LLINs was 1.46, though it is unknown how damaged these nets really were. These data show that LLINs with small amounts of damage still function as well as intact LLINs, but also show that very damaged LLINs have an insignificant level of protection.

In the context of malaria control efforts in Western Uganda, physical condition of nets was measured by Malaria Consortium in a random sample of nets from two representative, cross-sectional surveys in July 2011 and October 2012, 18-30 months after mass distribution of LLINs in the area. From a total sample of 1,598 and 3,938 households, 592 and 1,313 nets were assessed for physical integrity, respectively, using a proportionate hole index as recommended by WHO. Of these nets, 702 (43%) had been used the previous night by children 6-59 months of age for whom data on malaria parasitaemia were also obtained. Of the LLINs, 64% were in “good” condition based on the categorization proposed in this document, 16% had some damage, 7% serious damage and 13% were “too torn”. Interestingly, this pattern did not differ between the two surveys 12 months apart, providing further evidence that old nets were discarded at a certain level of damage. The results for malaria parasitemia by physical condition of the LLINs are shown in Figure 8. Although there was a slight increase of parasitaemia with increasing damage initially, the parasitaemia rate for children under “too torn” LLINs was not higher than for those with moderate damage. In a logistic regression model of the likelihood of being parasitaemic adjusting for age of the child, district, age of net, wealth quintiles and whether the child had acute fever, the odds ratio for parasitaemia for “too torn” LLINs compared to “serviceable” LLINs was 1.01 (95% CI 0.45 to 2.40) and no trend with increasing damage could be detected. Conversely, LLINs in “good” condition did not
provide better protection than more damaged LLINs (adj. OR 0.89, 95% CI 0.51 to 1.55). These data (submitted) suggest that, in the setting of Western Uganda, even seriously torn LLINs still provide some protection for children and nets are discarded before they lose their protective effect.

**Figure 8:** Malaria parasite rates in children 6-59 months (N=702) as a function of physical condition of the LLINs they used from Western Uganda

![Figure 8](image)

**Evidence for the shape of the survival curves**

Data on slow early losses comes from a series of 29 post-distribution surveys that assessed the overall retention of nets (i.e. the proportion of nets still present in the household that received them), some of which have been published [23-34] and some of which are unpublished survey results from Malaria Consortium and the NetWorks project. Results are shown in Figure 9 against the hypothesized LLINs survival curves and include one data point after five years [32] and one outlier with high retention after seven years from a project in Tanzania where the community has been intensely followed [34]. It must be kept in mind that the non-retention captured in these surveys includes the giving away of nets not relevant to LLINs survival but common in the early post-distribution phase (Figure 1). This suggests that the early losses based on discarded nets due to damage follows indeed a rather flat curve in the first year.
Data on the long tale of the distribution of net survival is presented in Figure 10 and comes from 12 household interview surveys carried out in Uganda (Malaria Consortium, unpublished) where the standard question on “when was this net obtained” was included in the net roster, but in these cases without the limitation of a maximum net age of three years as recommended in the standard DHS or MIS questionnaire. This shows that there is always a small number of nets that are very well-kept, reaching an age of several years and up to 10-12 years. Although a certain recall bias cannot be excluded in these surveys, there is little doubt from other anecdotal evidence and more recent surveys that long survival of some nets is not uncommon.

Figure 10: Age distribution of nets from 12 household surveys in Uganda (N=2569)
Finally, some data is presented supporting the S-shape of the net survival curve for nets (Figure 11). This comes from two post-campaign surveys in Nigeria (Kano and Anambra) undertaken by the SuNMaP project. Here the questionnaire included a module asking about nets that the households had owned at the time of the campaign or in the preceding year and which no longer were in their possession. Reasons for the loss as well as the age of the net at the time of discarding were recorded (see also annex 2). The sample included 111 lost nets, and plotting an inverse cumulative distribution of the age of the nets shows that this is very much an S-shaped curve.

**Figure 11**: Distribution of age of nets when lost from two post-campaign surveys in Kano and Anambra, Nigeria (N=111)
References


34. Maxwell CA et al. Comparison of coverage with insecticide-treated nets in a Tanzanian town and villages where nets and insecticide are either marketed or provided free of charge. *Malaria Journal*, 2006, 5:44.
## Annex 2

### Questionnaire module to capture and differentiate attrition in retrospective surveys

We would now like to ask some questions about the mosquito nets the household received from the campaign.

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| Q21| Are all these nets still in the possession of the household?                                         | Yes        | 1    |
|    |                                                                                                    | No         | 0    |

| Q22| If not, how many of the nets are still in the possession of the household?                          |             |      |

| Calculate the number of missing nets (Q20 minus Q22), record the number and proceed to Q23-Q25 for each net lost |

<table>
<thead>
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| Q23| Can you tell me what happened to the net?                                                          | Net was stolen | 1 |
|    |                                                                                                    | Net was destroyed accidentally | 2 |
|    |                                                                                                    | Net was sold | 3 |
|    |                                                                                                    | Net was given away to relatives | 4 |
|    |                                                                                                    | Net was given away to others | 5 |
|    |                                                                                                    | Net was thrown away | 6 |
|    |                                                                                                    | Material used for other purpose | 7 |
|    |                                                                                                    | Other | 8 |
|    |                                                                                                    | Don’t know | 9 |

| Q24| Why did you not keep this net?                                                                    | Net was too torn, too many holes | 1 |
|    |                                                                                                    | Net was too dirty | 2 |
|    |                                                                                                    | Net was not needed at the time | 3 |
|    |                                                                                                    | We did not like this net | 4 |
|    |                                                                                                    | Needed the money | 5 |
|    |                                                                                                    | Other | 6 |
|    |                                                                                                    | Don’t know | 9 |

| Q25| How old was the net when you discarded, gave away or otherwise lost it?                          | Age in months if less than 1 year |
|    |                                                                                                    | Age in years if above 1 year |
|    |                                                                                                    | Don’t remember | 98 |

Please enter the following information for each net “lost”
## Annex 3

### Reference LLINs survival curves

Preliminary reference survival table indicating the proportion of nets surviving

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**Figure 12:** preliminary reference survival curves