Durability monitoring of Long-Lasting Insecticidal Nets in Uganda, distributed during the 2020 mass campaign

Study Progress report July 2022

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ii)   Abbreviations and acronyms

BCC	Behavior Change Communication
CDC	Center for Disease Control
COVID	Corona Virus Disease
GFTAM	Global Fund to Fight AIDS, Tuberculosis and Malaria
GPS	Global Positioning System
HPLC	High Performance Liquid Chromatography
IDRC	Infectious Diseases Research Collaboration
IRB	Institutional Review Board
KD	Knock down
LC	Local Council
LLIN	Long Lasting Insecticidal Net
MOH	Ministry of Health
NMCD	National Malaria Control Division
NMS	National Medical Store
ODK	Open Data Kit
PACE	Program for Accessible health Communication & Education
PBO	Piperonyl Butoxide
PCA	Principal Component Analysis
pHI	proportionate Hole Index
PMI	President's Malaria Initiative
PSI	Population Services International
QAQC	Quality Assurance Quality Control
RBM	Roll Back Malaria
SOP	Standard operating procedure
UBOS	Uganda Bureau of Statistics
UCC	Universal Coverage Campaigns
UMIS	Uganda Malaria Indicator Survey
UMRSP	Uganda Malaria Reduction Strategic Plan
USAID	United States Agency for International Development
VHT	village health team
WHO	World Health Organization
WHOPES	WHO Pesticide Evaluation Scheme

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vi) Operational Definitions

Attrition	Attrition: (opposite of survivorship) is the proportion of nets no longer in use as intended after a defined period after their distribution to households. Attrition can be categorized by the main reasons why a net is no longer used, namely decay (e.g. destroyed, so torn and worn out that it is considered useless for protection against mosquitoes), absence (e.g. stolen, given away, moved) or used for other purposes.
Durability of LLINs	The three elements to be considered in assessing the durability of LLINs are net survivorship/attrition, fabric integrity and insecticidal activity (bio-efficacy)
Household	Household will be defined as any single permanent or semi-permanent dwelling structure acting as the primary residence for a person or group of people that generally cook and eat together. Some households may include members who sleep in other dwelling structures within the same compound, if the members are still dependent on the head of household in the main household.
Household Head	The Household Head is an adult person or persons who primarily make decisions for the general household (e.g. decisions on healthcare, income, etc.), including emancipated minors.
Household resident	A Resident within each household will be defined as a person who intends to have a sleeping place primarily at that location for a period of the next 6 months. This may include people who sleep in a separate house within the same compound, if they are still dependent on the head of household for decisions on finances and health care.

Insecticidal activity (bio-efficacy)	The degree of knock-down, mortality or inhibition of blood-feeding induced in susceptible mosquitoes, as determined by standard WHO test procedures and criteria (i.e. cone bioassay) (WHO, 2005).
Long-lasting insecticidal net (LLIN)	Is factory-treated mosquito net made with netting material that has insecticide incorporated within or bound around the fibres. The net must retain its effective biological activity without re-treatment for at least 20 WHO standard washes under laboratory conditions and three years of recommended use under field conditions.
Physical or fabric integrity Survivorship of LLINS	Reflects the number, location, type (burn, tear, seam failure, nibbled or chewed by animals) and size of holes in each net. Is the proportion of distributed nets still available for use as intended in the households to which they were given after a defined period, e.g. 1, 2, 3 or more years.

vii)   Study summary

Title	Durability monitoring of Long-Lasting Insecticidal Nets in Uganda, distributed during the 2020 mass campaign
Background	Use of long-lasting insecticidal nets (LLINs) is the primary strategy for prevention of malaria in Uganda. Uganda distributes LLINs every 3 years to achieve universal coverage of the population (1net for every two persons). The Ministry of Health needs information on the durability of different LLIN products in local settings to provide country-level data to guide national program planning for long term coverage with LLINs.
Methods	The study will be a prospective study of a cohort of nets distributed through a mass campaign. Within the first 6 months following the mass campaign, a representative sample of campaign nets from the study location will be identified through a cluster household survey with all campaign nets from consenting households forming the study cohort. These nets will then be labelled with a unique identifier and their presence and physical condition assessed at this baseline and in three additional annual surveys together with household characteristics and use, care and repair behavior for the net. At each assessment (baseline, 12, 24 and 36 months), sub-samples of campaign nets will be selected for insecticide effectiveness testing (bio-assays and chemical analysis). A baseline chemical residue assessment is

| | expected to come from the routine post-procurement quality control (QAQC).

The study will be conducted in 3 districts in the Bunyoro region. The districts shall receive 5 brands of nets (Interceptor, PermaNet 2.0 & 3.0, Royal Guard and SafeNet) in the mass LLIN distribution campaign. Five sub counties, one with each of the five net brands, will be purposively selected for participation in the study. A 2 stage-sampling strategy will be used to select households for participation. Within each selected sub-county (cluster), 16 villages will be randomly selected from the existing UBOS census list for participation (UBOS, 2014). Villages will be mapped to form a sampling list of eligible households for enrolment. 10 households will be randomly selected from each sampled village and nets in the households followed up over three years. A standard questionnaire will be used to collect household and net data. Physical durability of LLINs will be assessed by counting the number of holes on the nets. The integrity of nets will be quantified based on the proportion of LLINs with any hole and the proportionate hole's index (pHI) for each net. A subsample of nets (30 LLINs per brand per site) will be withdrawn during baseline and each follow-up assessment (after replacement) to assess for insecticidal activity (bio efficacy and chemical analysis). |
|---|---|
| Primary Objectives | To assess the physical durability of 5 different brands of nets over a three-year period, estimate median LLIN survival and identify major determinants of field performance. |
| Secondary Objectives | To describe major behavioral aspects of net care and repair and their impact on physical durability.

To assess the insecticidal effectiveness (residue and bio-assay) over three years of field use |
| Outcomes | Net survival (proportion of LLINs survived at 12, 24 and 36 months)

Net durability (proportion of "physically durable" LLINs at 12, 24 and 36 months)

Bio-efficacy of different brands of LLINs in Uganda. |

1.0 Study Design Summary

The principal study design is that of a prospective study of a cohort of nets distributed through a mass campaign. The baseline round was conducted one to six months following the 2020/2021 mass campaign, during which a representative sample of campaign nets from the study locations were identified through a cluster household survey with all campaign nets from consenting households forming the study cohort. These nets were labeled with a unique identifier and their presence and physical condition was assessed. At each subsequent annual assessment (12-, 24- and 36-months following the mid-point of the distribution period) the presence and physical condition of each net in the study cohort will be reassessed and recorded, together with household characteristics and use, care and repair behavior for the net. These characteristics will be used to identify household- and respondent-level risk factors for net survivorship once the study is complete. In each of the four data collection rounds (including this baseline study) samples of campaign nets will be selected from outside the cohort for insecticide bio-effectiveness testing by bioassay.

The sample size follows the standard www.durabilitymonitoring.org guidance protocol. Based on this guidance, we included 160 households per study site (16 clusters with 10 households each), or 800 households in total. Given the mass distribution campaign strategy of one net for every 2 people in a household and assuming an average household size of 4.8 persons in the study sites, this would have resulted in registration of 414 LLINs in each subcounty, or 2070 LLINs in total.[1] This number of LLINs is estimated to be sufficient to detect a 7 percentage-point difference in median survival time across study sites, assuming the median survival is three years. These figures correspond to a median survival difference across sites of less than 0.5 years, the minimum difference which has historically been considered important to detect for the purposes of campaign planning.

A cluster design of ten households in 16 clusters per study site was set to achieve the required 160 households. Sub counties are subdivided into villages. At the first sampling level, 16 villages were selected in each subcounty with probability proportionate to population size from a list of all villages in the health district. At the second sampling level, within each selected village, the field team mapped the whole area (i.e. listed all inhabited houses where people live) and from the compiled list of eligible households the supervisor randomly selected 10 households with equal probability for each household using random number lists.

During the household interview, LLINs from the campaign were identified by matching the net brand label, color, shape and respondent recall of receiving the net from the campaign. Eligible cohort LLINs were tagged with a unique number. It was explained to the eligible households that the unique number tag will not inhibit the regular use of the net and they must be careful not to remove it. Households were geo-located to facilitate subsequent visits. In addition to ITNs from the 2020/2021 campaign, all other mosquito nets present in the selected households were recorded to capture full and comparable data of all nets in each household.

Thirty campaign LLINs were randomly sampled from households outside of the cohort but within each study site to undergo biological tests and evaluate insecticidal effectiveness. Participating households received a new replacement LLIN in exchange for the one withdrawn for the study. Bioassays for this study will be conducted by the Infectious Diseases Research Collaboration in Kampala, in accordance with standard WHO guidelines for cone and tunnel tests for pyrethroid ITNs and standard operating procedures produced by USAID, CDC and PMI for testing PBO-synergist and Royal guard ITN products.[2] Chemical content analysis will be conducted by Walloon Agricultural Research Centre (CRA-W) in Belgium.

1.1 LLIN Brands Monitored

The table below summarizes the five LLIN brands monitored for the study. Interceptor and SafeNet, are pyrethroid-only LLIN containing Alpha-cypermethrin as the active ingredient. Interceptor, together with permaNet2.0 (active ingredient is deltamethrin) were distributed in August 2020 while safeNet, the dual acting nets- permaNet3.0 (active ingredients are deltamethrin/Piperonylbutoxide) and Royal guard (active ingredient are alpha cypermethrin/ Pyriproxyfen) were distributed later in the year (November 2020). Therefore, the average amount of time elapsed between the distribution and baseline for permaNet2.0 in Kakindo cluster and interceptor in Buheesi cluster was 5.5 months and 6.0 months respectively. This duration was much shorter for the brands distributed later in the year. Permanet 3.0 in Kyabigambire was at 3.5months, Royal guard in Kigorobya town council was at 3.0 months and safeNet in Kiziranfumbi was at 3.75months respectively by the time the baseline survey was conducted. In total, 6,840 interceptor LLINs were distributed in Buheesi subcounty(Bunyangabu district), 29,240 PermaNet3.0LLINs in Kyabigambire(Hoima district), 23, 080 safeNet LLINs in Kiziranfumbi (Kikuube district), 9,360 permaNet2.0 LLINs in Kakiindo subcounty (Kakumiro district) and 4880 royal guard LLINs in Kigorobya town council (Hoima district).

TABLE 1: LLIN BRANDS DISTRIBUTED IN STUDY AREAS

	Interceptor	PermaNet2.0	SafeNet	PermaNet 3.0	Royal gurad
District	Bunyangabu	Kakumiro	Kikuube	Hoima	Hoima
Quantity	6,840	9360	23,080	29,240	4880
Study site	Buheesi subcounty	Kakiindo subcounty	Kiziranfumbi subcounty	Kyabigambire subcounty	Kigorobya town council
Distribution Date	August 15th–19th, 2020	August 15-19th 2020	November 7th-11th 2020	November 7th-11th 2020	November 7th-11th 2020
ITN type	Standard	Standard	standard	PBO-synergist	Dual-AI
Chemical content	Alpha-cypermethrin (100 mg/m^2)	Deltamethrine	Alpha-cypermethrine	Deltamethrin (2. g/kg sides, 4.0 g/kg roof; equivalent to approx. 118 mg/m^2 and 180 mg/m^2) PBO (25 g/kg roof only; equivalent to approx. 1100 mg/m^2)	Alpha-cypermethin /Pyriproxyfen
Fabric	Polyester	Polyester	Polyester	Sides: polyester Roof: polyethylene	polyethylene
Denier	100	100	100	Sides: 75 Roof: 100	120
Shape	Rectangular	Rectangular	Rectangular	Rectangular	Rectangular
Manufacturer	Shanghai Gongtai textile co ltd/BASF	Vestergaard SA	Mainpol GmbH	Vestergaard	Disease control technologies LLC China
Study site	Buheesi subcounty	Kakindo subcounty	Kiziranfumbi subcounty	Kyabigambire	Kigorobya town council

Average time between distribution and data collection (months)	6.0	5.8	4.0	3.6	3.2

1.2 Study Sites

The study was carried out in the Bunyoro and Toro regions, locations with moderate malaria burden (parasite prevalence: 9% - Uganda Malaria Indicator Survey, 2019). The study districts were purposively selected from the region in collaboration with NMCD to ensure that all net brands distributed in the 2020 UCC are represented.

Five sub-counties (clusters) from within the four districts of Hoima, Kikuube, Bunyangabu and Kakumiro were selected for net sampling. Each of the 5 sub-counties received different net brands distributed will be prospectively followed over the study period. Study villages and households were randomly selected from the sub counties.

FIGURE 1: STUDY SITE MAP SHOWING CLUSTERS FOR THE LLIN BRANDS
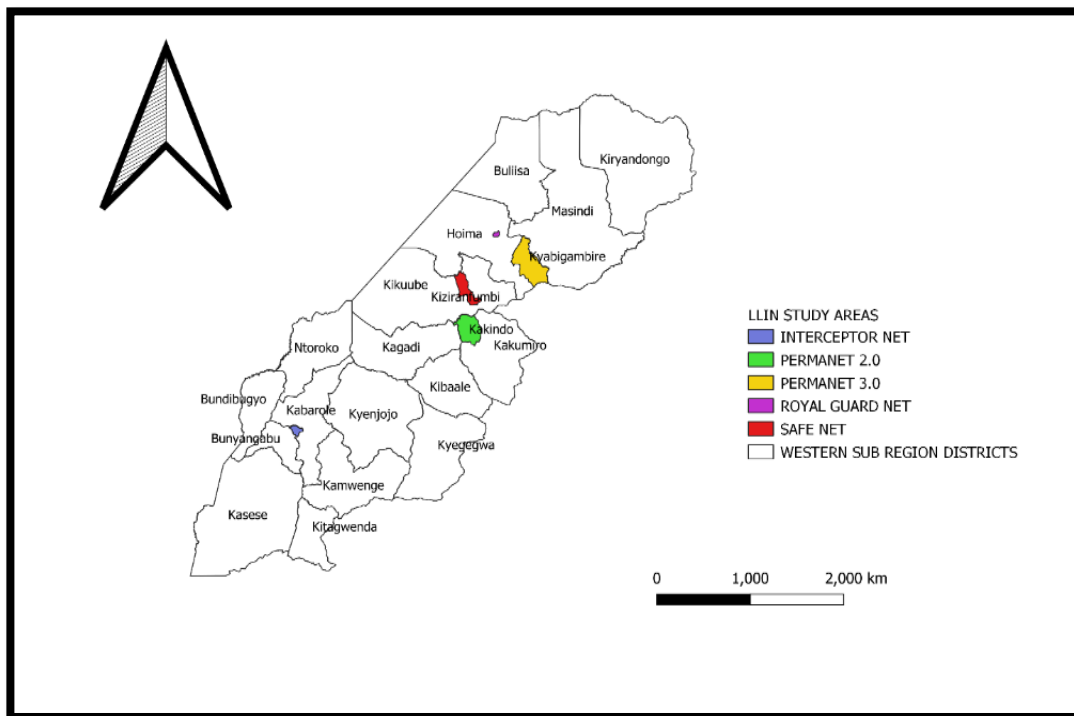
All districts have similar environmental, epidemiological, and population profiles. The districts are located in the western part of Uganda. Malaria is endemic throughout the year in Uganda, although peaks follow the rainy seasons in March-April and October-November. According to the 2018/2019 malaria indicator survey, the malaria prevalence was 9%. All districts experience a hot dry climate with hyperendemic malaria transmission. Agriculture is the main economic activity of the populations in all three districts.

Figure 2: Overview of study design
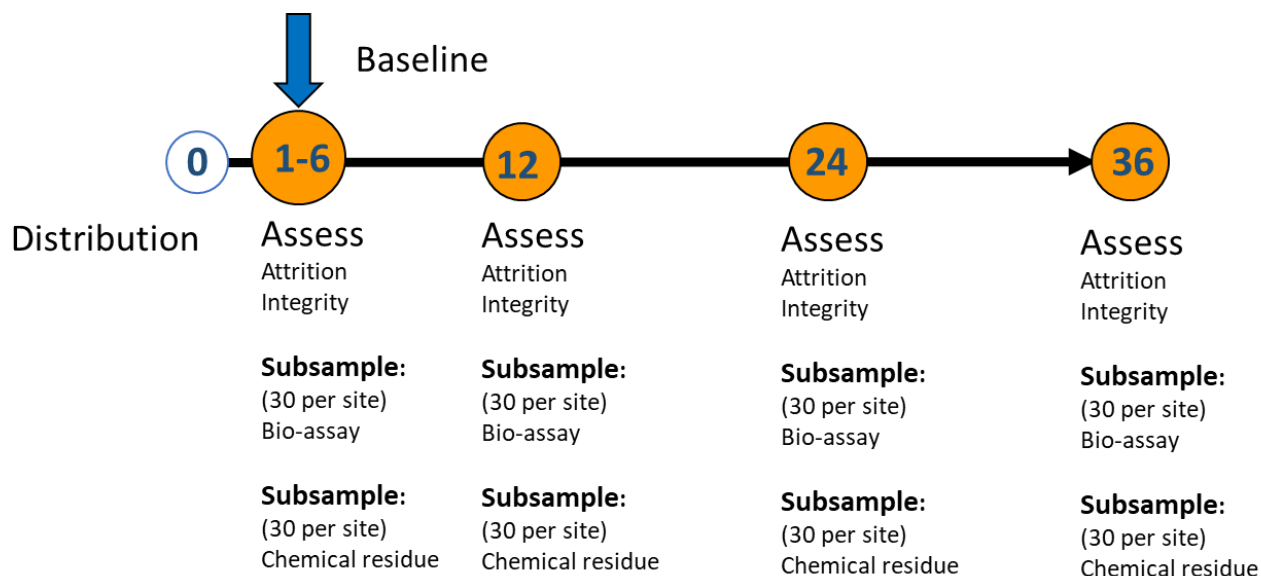
2.0 Study activities and Timelines

The study started in 2019 and will continue up to 36 months. Currently we are preparing for 24 Months survey data collection which will be conducted from August to December 2022. This study is implemented through a series of cascaded activities starting with protocol and data collection tool development to dissemination of results.   Below is the status of the study schedule

Below is the study activity schedule broken down into the preparatory and the implementation activity schedules. *Figure 3 and 4*.

Figure 3: LLIN Durability Monitoring study (Preparatory activities and Timelines)										
	Status	**2019**				**2020**				**2021**
Activity		**Jan - Mar**	**Apr - Jun**	**Jun - Sept**	**Oct - Dec**	**Jan - Mar**	**Apr - Jun**	**Jun - Sept**	**Oct - Dec**	**Jan - Mar**
Recruitment of the study core Research team	**Completed**			■						
Protocol Development and study planning meetings	**Completed**				■	■	■			
Ethical Approval	**Completed**									■
Development of Standard Operating Procedures (SOPs)	**Completed**								■	■
Participation in Micro planning and Net distribution campaign for the UCC 2020	**Completed**					■	■	■	■	
Training of Trainers training	**Completed**								■	

Activity	Status											
Development of training materials for the Data collector training	**Completed**										■	■
Research Ethics Training	**Completed**										■	
Baseline survey Data collector Training	**Completed**											■

Figure 4: LLIN Durability Monitoring study activities and Timelines (Implementation activities and Timelines)													
Activity	**Status**	**2021**				**2022**				**2023**			
		Jan - Mar	Apr - Jun	Jun - Sept	Oct - Dec	Jan - Mar	Apr - Jun	Jun - Sept	Oct - Dec	Jan - Mar	Apr - Jun	Jun - Sept	Oct - Dec
Baseline Survey		■											
Pilot test	**Completed**	■											
District Planning engagement, Community sensitization, mobilization, and entry	**Completed**	■											
Community surveys / Data collection	**Completed**	■											
Bioassay tests and Analysis	**Completed**		■	■	■	■							
Chemical residue (HPLC) tests and Analysis	**Completed**		■	■	■								
Report writing			■	■	■								
Results Dissemination	**Completed**					■	■						
12 Months survey						■	■						
Community surveys / Data collection (Kakindo and Buheesi Sub counties)	**Completed**			■	■								
Bioassay tests and Analysis	Ongoing						■	■	■				
Chemical residue (HPLC) tests and Analysis	Ongoing						■	■	■				
Results Dissemination	Pending								■	■			
24 Months survey									■	■			
Community surveys / Data collection (Kakindo and Buheesi Sub counties)	Pending							■	■				
Bioassay tests and Analysis	Pending									■	■	■	
Chemical residue (HPLC) tests and Analysis	Pending									■	■	■	
Results Dissemination	Pending												■
36 Months survey													■
Community surveys / Data collection (Kakindo and Buheesi Sub counties)	Pending										■	■	
Bioassay tests and Analysis	Pending												
Chemical residue (HPLC) tests and Analysis	Pending												
Results Dissemination	Pending												
Dissemination of comprehensive study results	Pending												

2.2 Preparatory Activities

2.2.1 Recruitment of the study core Research team

For successful study implementation, the study needed key staff to run the activities smoothly. These include the following:

1. The Principal Investigator (PI) whose main role is to lead the protocol development and coordinate the talent and skilled personnel on the study towards achieving the study objectives and utilization of the study results to inform policy.
2. A Research manager was also recruited whose role is to contribute to the protocol and data collection tool development, develop Standard Operating Procedures (SOPs), develop, and manage the study budgets and lead the study implementation.
3. A Research Coordinator was also recruited whose role is to lead the data analysis to ensure the output meets the study objectives, Review the study reports, write abstracts and manuscripts, and summarize the study findings in statistically sound formats for easy dissemination and data use.
4. A Research officer was also recruited whose role is to conduct the implementation of study activities in adherence to the protocol, participate in compiling study implementation reports, data cleaning and participate in performance review meetings.

Besides the study Research team, other personnel for support functions such as finance, Human Resource Management, procurement, and fleet management that support the study implementation.

2.2.1.2 Study synergist collaborations

The study is implemented in a collaboration with Population Services International (PSI), Infectious Diseases Research Collaboration and Walloon Agricultural Research Center (CRAW). Each collaborating institution contributes as described below.

1. PSI brings on board their experience in implementing net durability studies in more than 8 countries with similar study objectives. PSI contributed towards the protocol development and continues to contribute towards the implementation best practices learnt from other countries, data analysis, presentation formats and interpretation.
2. Walloon Agricultural Research Center (CRAW) conducts the High-Performance Liquid Chromatography Lab tests which are done at their laboratory in Belgium. CRAW has conducted these lab tests and analysis for all LLIN Net Durability studies conducted by PSI and therefore have adequate expertise authority in this field.
3. Infectious Disease Research Collaboration (IDRC) conducts the Bioassay Lab tests and analysis. IDRC does these tests at the insectary in Butabika inside the Central Public Health Laboratories (CPHL).

Figure 5: Study team structure and roles

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┌─────────────────────────┐   ┌─────────────────────────┐   ┌─────────────────────────┐
│    Study PI at PACE     │   │     Study PI at MOH     │   │     Study PI at PSI     │
│                         │   │                         │   │                         │
│  Provided oversight to  │◄─►│  Provided oversight to  │◄─►│  Provided oversight to  │
│  the study team during  │   │  the study team during  │   │   the study team        │
│  field work and         │   │  field work and         │   │   during field work     │
│  participated in the    │   │  participated in the    │   │                         │
│  supervision of data    │   │  supervision of data    │   │                         │
│  collection             │   │  collection             │   │                         │
└─────────────────────────┘   └─────────────────────────┘   └─────────────────────────┘
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┌───────────────────────────────┐          ┌───────────────────────────────┐
│ Study co investigators at PACE │          │ Study co investigators at MOH  │
│                                │          │                                │
│  Provided oversight to the     │◄────────►│  Provided oversight to the     │
│  study team during field work, │          │  study team during field work, │
│  participated in the RA        │          │  participated in the RA        │
│  Training, development of       │          │  Training and development of   │
│  fieldwork materials and       │          │  fieldwork materials           │
│  participated in supervision   │          │                                │
│  of data collection            │          │                                │
└───────────────────────────────┘          └───────────────────────────────┘
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```
┌─────────────────────────────────────────────┐
│            PACE Research manager            │
│                                             │
│  Provided oversight to the study team       │
│  during field work, facilitated in the RA   │
│  Training, development of fieldwork          │
│  materials and led the field supervision    │
│  team                                       │
└─────────────────────────────────────────────┘
```

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┌─────────────────────────────────┐    ┌─────────────────────────────────┐
│     PACE Research Officer       │    │    District Leadership (DHT)    │
│                                 │    │                                 │
│  participated in the RA         │    │  Provided political will,       │
│  Training, development of       │    │  supported the team in          │
│  fieldwork materials and was    │    │  community entry and            │
│  part of the field supervision  │    │  mobilization and participated  │
│  team                           │    │  in the field supervision       │
└─────────────────────────────────┘    └─────────────────────────────────┘
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```
┌─────────────────────────────────┐
│        Field supervisors        │
│                                 │
│  Supported the facilitation of  │
│  the RA Training and was part   │
│  of the field supervision team  │
└─────────────────────────────────┘
```

```
┌─────────────────────────────────┐
│   Volunteers / Data collectors  │
│                                 │
│  Participated in the RA         │
│  Training, community entry,     │
│  village mapping and            │
│  interviewing                   │
└─────────────────────────────────┘
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2.2.2 Protocol Development and study planning meetings

This was done in close collaboration with NMCD MOH. NMCD provided key information for purposes of sampling. This included the malaria zones of Uganda, the waving schedule of the 2020 Universal Coverage Campaign among others. During this activity, we had harmonization meetings with IDRC and PMI to avoid conducting the study in the same study areas unless with complementing study objectives. PSI is conducting the LLIN Durability Monitoring study in more than eight other countries and therefore, the study protocol, and data collection tools they are using were adopted and customized for this Ugandan context since the study objectives are the similar. This makes the study results comparable with those from other countries where the same net brands are studied.

The protocol was developed by the PACE and PSI research team and reviewed by the MOH study investigators. The team of investigators that contributed to the protocol development include Dr. Opigo Jimmy, Dr. Maiteki Catherine, Dr. Damian Rutazaana, Dr. Gonahasa Samuel, Mr. Kasule Mathias, Miss Rebecca Babirye, Mr. Stephen Poyer and Miss Phyellister Nakamya led the writing of the protocol. The review and approval were led by Dr. Opigo Jimmy. The institutions that contributed include PACE, PSI, MOH and IDRC.

The protocol was developed following the WHO LLIN Durability Monitoring studies guidelines to ensure that the international best standards were followed so that the study results are widely acceptable globally. This was done for all study aspects including physical integrity, Bioassay, and chemical residue (HPLC). Conventional thresholds for Proportionate Hole Index and Bio efficacy analysis were taken on.

The protocol went through several reviews and improved before submission for approval by the IRB. The last and approved version was version 1.0 which the study team follows in the implementation to date.

2.2.3 Participation in Micro planning for the UCC 2020

To facilitate sampling for the study, PACE participated in the micro planning for waves 1, 2 and 3. to access and obtain full acquaintance with the administrative units and their respective population specifics and other area profiling data necessary for the sampling of the study. PACE's participation in the micro planning also led to recommendations for the improvement of the net distribution campaign.

Consequently the districts sampled for the study area were within waves 2 and 3 (Kakumiro, Bunyangabu, Hoima and Kikuube). The micro planning data collected through our participation facilitated the sampling at the lower levels of sub county, village, and household level.

2.2.4 Ethical Approval

The protocol was submitted to Vector Control Division (VCD) Research Ethics Committee (REC) which approved the protocol with a condition of submitting for renewal at the end 2021 along with a summary progress of the study. In January 2021 the protocol was approved by the Uganda National Council of Science and Technology (UNCST) In November 2021, we applied for renewal of the ethical approval to the VCD REC and approval was granted valid up to the end of 2022. We will submit application for ethical approval renewal in November 2022 to get ethical approval for 2023.

Copies of the approved protocol and the ethical approval letters (VCD REC and UNCST) are attached as appendices 1 and 2 respectively.

2.2.5 Development of Standard Operating Procedures (SOPs)

This study involves a range of activities that must be conducted uniformly / homogeneously to collect high quality data. These include identifying the households eligible for interviews, consenting by the respondents, Conducting the Net Hole Assessment, Net Withdraw and storage and replacement. Consequently, SOPs were developed for these key activities. These include the following:

1. Identification of eligible households for participation in the study SOPs
2. Informed consent SOPs
3. Net withdraws, storage and replacement SOPs

2.2.6 Training of Trainers training

The training of trainers training was done by PSI (CO Investigator) team The training materials used for this study in other countries were used for this training with reference to the approved study protocol and data collection tools for the Ugandan study. This training took place on December 14th 2020 and was virtual on Microsoft teams.

The training covered several areas which include the following:

1. Introduction to the LLIN durability concept and LLIN durability monitoring

2. Study design and methods

3. Household listing and sampling

4. Questionnaire section

5. Labelling cohort nets

6. Hole Assessment

2.2.6.1 Training methodology

The training was highly participatory, and the main methodology used was power point presentations but with the trainers engaging the trainees more to stimulate discussions and questions for clarification where necessary.

Practical exercises were done for the sessions of hole assessment. This was facilitated by photos of nets with different hole sizes and numbers.

Demonstrations of how some of the tools shall be used were also made. These include the ruler and the hole assessment tally sheet.

Figure 6: A photo of demonstrating how the hole assessment shall be done using the ruler

Figure 7: A demonstration of how the tally sheet shall be filled

SHORT Side 1	5	10	15	20	Size 1	Size 2	Size 3	Size 4
Size 1	●●●●●	●●○○○	○○○○○	○○○○○	7			
Size 2	●●○○○	○○○○○	○○○○○	○○○○○		2		
Size 3	●○○○○	○○○○○	○○○○○	○○○○○			1	
Size 4	○○○○○	○○○○○	○○○○○	○○○○○				0

The header spanning "TOTAL" covers Size 1, Size 2, Size 3, Size 4 columns.

2.2.6.2 Training Evaluation

Evaluation of training participants to determine knowledge and skills and practice was done using pre and post tests.

2.2.7 Development of training materials for the Data collector training

PACE adopted the training materials and data collection tools from PSI hat had been used in other countries where the LLIN Durability Monitoring studies have been conducted. The PACE Research team revised and customized these to the Ugandan study protocol and profile specific information such as the net brands and study area and adopted for the study trainings. These training materials have been revised to include more explanation and illustrations where necessary to make the trainings more practical.

The content of study training materials includes the following:

1. Research Ethics
2. Study overview (Including the objectives, study area, operational definitions, Description of aspects of physical integrity, Bioassay efficacy and chemical residue / HPLC)
3. Sampling procedures and eligible household identification
4. Standard Operating Procedures
5. Hole Assessment and Proportionate Hole Index
6. Data Quality
7. Net sample withdraw procedures
8. Questionnaire review
9. Interviewing skills
10. PACE Organizational policies (including anti-fraud and safeguarding policies)

2.2.8 Research Ethics Training

This study involves interaction with human subjects including interviews and therefore it was very necessary that the study team complete a standard Research Ethics course and the data collection team in addition receive Research ethics training to ensure that the study participants are protected. Considering this, on 4th and 5th of December 2020, PACE conducted a Research Ethics training for 45 trainees in which all the prospective data collectors were trained in Research ethics and also given an overview of the study design and methodology. This training took place at Hotel Africana.

The training was facilitated by the PACE Research team and the study investigators from MOH. The PACE Research team included the study Principal Investigator (Phyellister Nakamya), the PACE Research manager (Dr. Alex Ndyabakira), the PACE research Coordinator (Dr. Betty Nambuusi Bukenya), and the PACE Research officer (Joel Kato). The investigators from MOH included the Global Fund National Facilitator at that time (Henry Katamba), Principal medical officer – Research and policy (Dr. Catherine Maiteki) and Dr. Damian Rutazaana.

In preparation for the training, the trainees completed an online Research Ethics certificate course on the CITI program platform. This course provided a robust foundation for the training since its content included the principles of Research Ethics and Human subjects' protection and background and context against which they were formulated. The PACE Research team as well completed this Research Ethics course first before even sharing the course link with the data collection team. These Research Ethics certificates are filed in the study cabin at the PACE Head Quarters office.

The content of this training included the following:

1. The Principles of Research ethics (Justice, Confidentiality, Respect for persons and Beneficence)
2. The consenting process
3. Study Standard Operating Procedures
4. Study Design and protocol overview
5. Questionnaire overview

The training was evaluated through a post and pretest before and after the training respectively. The questions of the evaluation were about the training content. The trainees that performed poorly (below 50%) were given more explanation by the PACE Research time to understand the Research Ethics principles. Those that even after further explanation showed lack of understanding of the Research Ethics principles did not join the baseline data collection team.

2.2.9 Development of the Data collection system

This is a longitudinal cohort study where data is collected more than one time and the same households and nets are followed over 3 years. This requires a long-term data collection system that can facilitate a series of data to be collected about the same household or net over the study life. PACE therefore contracted DERON Ltd to develop this system for the study. A system that operates both on the internet web browsers and android application was developed. The system could work offline as well for user friendly navigation and control of the challenge of poor network in the hard-to-reach rural places within the study area.

2.2.10 Data collector training

This is a cohort study that is designed to assess net durability following the WHO net durability study guidelines that must be followed strictly if the study is to generate scientifically robust results acceptable by the global community. For this to happen successfully, there is need to rigorously train the data collection team to fully understand the study SOPs and study assessment procedures.

Against this background, the PACE team conducted a data collectors' training of 41 Data collectors from February 1st to 7th February 2021. This training took place in Hoima at the District Head Quarter offices and was facilitated by the PACE Research team and the study investigators from MOH. The PACE Research team included the Research manager (Dr. Alex Ndyabakira), Research Coordinator (Dr. Betty Nambuusi Bukenya), Research officer (Joel Kato) and the Monitoring and Evaluation (M&E) Manager (Ivan Muhire)

The training was highly practical and included illustrations and sessions of hole assessment practice by each trainee and role play interviews. Trainee participation was encouraged as much as possible through out all sessions. The training content covered focused on equipping the trainees with the knowledge and skills to collect high quality data.

To enable the Research Assistants make reference where necessary, the study implementation materials which include the tally sheets, consent forms, net master lists, household master lists and the hard copy questionnaires as well were given to them. The Research Assistants also got the opportunity to practice using the data collection tools and master how to fill them before the actual exercise commenced.

This training was very beneficial to the Research Assistants because they helped them to fully understand the study procedures to be followed while Mapping, consenting, interviewing and conducting the hole assessment.

The content of this training included the following:

1. Research ethics
2. Study Protocol and Methodology
3. Data collection procedures and tools
4. Data Quality
5. PACE policies

The training was evaluated through a post and pretest before and after the training respectively. The questions of the evaluation were about the training content. The trainees that performed poorly (below 50%) were given more explanation by the PACE Research team to understand the particular concepts where they needed further explanations and illustrations.

2.3 Implementation Activities

2.3.0 Baseline Survey

2.3.1 Pilot test
After the field training, a pilot test was conducted with the establishing and testing the applicability of the knowledge and skills acquired through the field training and to identify any key issues that needed to be improved within the data collection strategies and tools to facilitate the collection of high-quality data in the sampled administrative units.

The pilot test took place in Bwanya cell village in Hoima East Division, Hoima city and Bulyango central village in Kitoba sub county, Hoima district. These are villages that were not sampled for the study. The Research Assistants were made into teams of 2 people each and each team was assigned a number / name. The Research Assistants were 39 in total, and they were divided into 19 teams and assigned team numbers from Team 1 up to Team 19 respectively. It is in these teams that they participated in the pilot test and the data collection activities. The activities conducted during the pilot test include consenting respondents, conducting the full interviews, conducting the hole assessment and filling the net and household master lists.

The pilot test was very beneficial to the Research Assistants as it gave them insights regarding how to do the hole assessment fast, how to fill the tally sheets and the net and household master lists faster and accurately. It gave Research Assistants the opportunity to get more acquainted with these tools and also the questionnaire itself. It built their confidence too in being freer with the respondents and creating rapport. The pilot test gave them exposure to the potentially common challenges that they would later encounter in the actual data collection which gave us the opportunity to think of solutions to these challenges early enough before data collection commenced.

One of the key issues identified was Households presenting nets of the same brand as that of interest in a particular sub county, yet these nets were not got through the campaign of 2020. This was addressed by advising the Research Assistants to always confirm with the respondent whether the particular net in question was received through the 2020 campaign. Additionally, to look at the year of distribution printed on the labels to verify if the nets being observed were from the 2020 campaign.

Several issues were identified in the flow of the electronic questionnaire and these were raised to the system developer (Deron Ltd) for rectification before the start of data collection.

2.3.2 District Planning engagement, Community sensitization, mobilization, and entry

Community entry was conducted at different levels. These included the district , sub county land village levels. At district level, the PACE Research team conducted district entry meeting with the district leadership of each of the 4 districts (Hoima, Kikuube, Kakumiro and Bunyangabu). During these meetings, we gave the district leader (DHO, CAO, LC V, Malaria focal person, VCO) an overview of the study and the support we expect the districts to give PACE throughout the entire study life. The district agreed to provide the support as well as participation in the supervision of the study.

. In Bunyangabu District, the District Health Team led by the DHO participated in the supervision of data collection, Hoima district leadership pledged to provided storage space at the district medical stores Additionally, The district leadership also provided us with information about the possible community events and structures such as market days and poor road networks that we needed to consider when planning the fieldwork / data collection.

At the sub county level. The study was introduced to the sub county leadership and discussed how they can support the study. The subcounty leadership provided the political support and security when and where necessary as need arises. They also assigned focal persons to support us in the mobilization of the VHTs and LC1 Chairpersons. This successful happened for all the sampled sub counties for the study.

At the village level and the entry and mobilization for this level was done by the Research Assistants. At village level, the community entry and mobilization activities included introduction of the study to the LC1 chairpersons and VHTs and giving them talking points to follow when explaining the study activities to community members during mapping and when asked individually.

During the village mapping, the Research Assistants also introduced the study to the community members and informed them that if their households were part of the sampled 10 households they would go back and interview them.

The mapping data was uploaded to the study server where it was accessed by the PACE Research team that did the sampling of the households within each village where mapping had been completed. This was followed by mobilization where the Research Assistants informed the LC1 chairpersons and the VHTs of the selected households and asked them to mobilize the household members to stay home on the days scheduled for interviews so they could find them and request their consent to be interviewed and also conduct hole assessment on their mosquito nets.

The talking points given by the Research Assistants to the VHTs and chairpersons were the following:

- ➢ The villages where the study was conducted were randomly selected
- ➢ The purpose of the study is to assess the durability of the mosquito nets distributed in the 2020 mass campaign
- ➢ The households where interviews were to be conducted were sampled randomly.
- ➢ This is not a health inspection it's a study
- ➢ Participation in the study is voluntary and out of free will and any refusal to participate shall not have attract any punishment

> ➤ The study team shall select some households for withdrawing nets from, but these nets shall be replaced with new nets of the same brand
> ➤ The study team shall also request to observe and assess the nets the sampled households received in the 2020 campaign.

2.3.2 Data collection

2.3.2.1 Study areas and administrative units
The LLIN DM baseline survey was done in the districts of Hoima, Kikuube, Bunyangabu and Kakumiro. Below the district level, the baseline was conducted was in 5 sub counties (One in Kakumiro, 2 in Hoima, one in Bunyangabu and one in Kikuube). Then in each sub county 16 villages were sampled apart from Kigorobya Town Council where only 8 were sampled but the household target was doubled. Then in each village 10 households were randomly sampled and data was collected from them. (for a detailed list of all the sampled admin units please refer to appendix 1).

2.3.2.2 Data collection methods and strategy
The LLIN baseline survey was quantitative in design according to the approved protocol and therefore data was collected using a structured questionnaire which was also used in other countries where the study was conducted. This questionnaire was programmed into a mobile application and installed on the study tablets.

The sub counties were covered one by one starting with Kakindo which had nets whose 6-month baseline period was about to expire. The entire team of 19 teams was placed in one sub county at a time and each of the 16 sampled villages was assigned one team of 2 people to do both the mapping and the interviews.

2.3.2.3 Data collection duration
The baseline survey data collection fieldwork (training, pilot test and data collection) took 41 days in total. The data collection took 36 days. The village mapping on average took 2 to 4 days depending on the number of households in the village and how distant the households were from each other within the village. The interviews on average took 2 to 3 days depending on the average number of nets the households in the village received. The field work started on 31/1/2021 and ended 12/3/2021.

2.3.2.4 Enrollment of eligible households into the study cohort

2.3.2.4.1 Household target and sampling procedure
We had a target of enrolling 800 households. 160 households from each sampled sub county. These are Kakindo sub county in Kakumiro district, Kigorobya T/C in Hoima, Kyabigamire sub county in Hoima district, Buheesi sub county in Bunyangabu and Kiziranfumbi sub county in Kikuube district. Within each sub county, we randomly sampled 10 villages and in each of these villages we randomly sampled 10 households. *(for a detailed list of all the sampled admin units please refer to appendix 1)*.

2.3.2.4.2 Village mapping
The first step to the enrollment of households was village mapping where GPS coordinates and the name of the household head of all the households in the villages were collected using an electronic system. This was to make sure that every household had a calculable and non-zero chance to be sampled. The Research Assistants together with the LC Chairperson and VHTs walked to each household in the sampled villages during mapping and collected this information.

After the mapping, the PACE Research team did the randomization / sampling of the households and then gave the lists of sampled households of which the first 10 were considered as the sampled households and were engaged first then the rest were for replacement in case the Research Assistants were not able to enroll any of the first 10 households.

2.3.2.4.3 Household Replacement

The randomized sample list given to the Research Assistants had more than 10 households. The first 10 households were considered as the sampled households and then the rest of the households on the list were for replacement in the event that any of the first 10 households could not be interviewed. However, this had to be done according to the replacement procedure. When replacing any household, the Research Assistants considered the households in consecutive or serial order, the first being the 11th household and then in case it was not available they moved on to the 12th and it is only when the 12th was not available that they moved to the 13th household. This is the criteria they followed in the replacement as they enrolled households into the study cohort.

2.3.2.4.4 Eligibility / Inclusion criteria

Regarding the eligibility of the households to be enrolled, the Research Assistants only enrolled the households that received the 2020 UCC campaign nets. These were considered even when they had lost all the campaign nets they received.

2.3.2.4.5 Exclusion criteria

The households that were excluded include the following:

- ➢ Those that did not receive nets from the 2020 campaign, those that refused to consent to be interviewed
- ➢ Those that did not have any adult of sound mind fit to be interviewed
- ➢ Those that were closed and no household member was available after 3 visits were replaced though eligible.

The sampled households were identified and located with the help of the LC1 chairperson and the VHT who were well acquainted with the households in their villages.

The Research Assistants used tools like household master list to keep a record of the enrolled households in every village. On this household master list, they recorded the village name and code, name of the household head, the household ID, the number of UCC nets enrolled in each household and the other nets found in each household.

The enrolled households can best be identified in the future rounds of data collection by following and asking for the head of household name after reaching the sampled villages.

2.3.2.5 Questionnaire administration

2.3.2.5.1 Target Respondents

The interviews were conducted at the sampled households or the replacements of the sampled households and the heads of households, their spouses or a representative of the household head that was well knowledgeable about the household were interviewed.

2.3.2.5.2 Consenting procedures

The households were consented in their local languages, and most were consented in Runyoro and the consent was documented by the consent forms in Runyoro. Both the respondent and the interviewer signed the consent form before starting any interview. In cases where the respondent could not read and write, the respondent identified someone who could read and write to be a witness on his or her behalf while the Research Assistants read the consent forms to the respondents. The witnesses in such cases also read through the consent forms on behalf of the respondent before signing. In such cases the respondent then put his or her thumb print in the designated space on the consent form.

2 consent forms were signed and one copy was left with the respondent and then the second was brought back to the study coordination / PACE Research team and we filed them together with the tally sheets for each household in suspension files which are kept in a locked metallic suit case at the PACE headquarters. These are only accessed by the study team when necessary which has all done the Research Ethics certificate and fully understand the concept of protecting human subject in research.

2.3.2.5.3 Interview administration

The research Assistants conducted the interviews in 950 households in the languages the respondents understood best which for most was Runyoro being the local language of the study region. The interviews were done using an electronic structured questionnaire and the responses were entered in an electronic system installed on the project tablets received by the Research Assistants.

The Research Assistants applied several interviewing skills where necessary and applicable such as probing, prompting, observation of nonverbal communication and not asking leading questions.

The interviews were conducted in observation of confidentiality and only the respondent was present during the interviews. The LC1 chairperson and the VHT were asked to kindly wait in a place where they could not listen to the interviews.

The interviews took an average of one and half hours to 2 hours depending on the number of nets found in the household and the number of holes the nets had. Where the nets were many and had many holes, the interviews took about 2 hours while where the nets were few and the holes were few also then interviews took about one hour.

At the end of every interview, the Research Assistants compensated the respondents with 10,000 shillings as communicated in the training. This was also documented on the payment forms where the respondents signed after receiving this money.

2.3.2.5.4 Enrollment of nets into the study cohort

This study has a cohort of nets which are the UCC 2020 nets received and still available within the enrolled households. The study particularly targets 5 brands which are; Permanent 2.0, Permanent 3.0, Royal guard, Interceptor and Safe net. The target for the net to be enrolled into the study cohort was 2400 in total and within these each of the 5 brands had a target of 480 nets. Each net brand was found in a different sub county. Below is a table showing the targets and location for each net brand.

Table 2: Targets and location for each net brand – LLIN Durability Monitoring Baseline survey – Physical durability cohort

District	Subcounty	Brand of interest	Target for cohort
Kakumiro	Kakindo	Permanent 2.0	480
Hoima	Kigorobya Town Council	Royal guard	480
Hoima	Kyabigambire	Permanent 3.0	480
Bunyangabu	Buheesi	Interceptor	480
Kikuube	Kiziranfumbi	Safe net	480

Within the enrolled households, we enrolled all the available 2020 UCC campaign nets into the study cohort. All these nets were tagged with labels that had unique ID numbers. The inclusion criteria for enrollment of nets into the study cohort include the following:

➤ The net was received from the 2020 UCC campaign
➤ The net is the brand of interest in that particular sub county where the household to which it belongs is located. (Permanent 2.0 for kakindo, Interceptor for Buheesi, Safe net for Kiziranfumbi, Royal guard for Kigorobya T/C and Permanent 3.0 for Kyabigambire)
➤ The net is available for observation and the respondent consents to tag it with an ID for follow up and also for net hole assessment.

The criteria for exclusion of nets include the following:

➤ Nets that were not received from the 2020 UCC campaign.
➤ Nets that were not the brand of interest in that particular subcounty where the net belongs.

All the enrolled nets were assessed for hole size and number after being tagged. This was done by use of marked rulers following the categories of the hole sizes communicated in the training where we had size 1, size 2, size 3 and size 4. The documentation of the net hole assessment was done by use of tally sheets. The criteria we followed for categorization based on which our rulers were marked is shown in the table below.

SIZE	CRITERIA
Size 1	>05cm to 2cm
Size 2	>2cm to 10cm
Size 3	>10cm to 25cm
Size 4	Above 25cm

The enrolled nets can best be identified in future rounds of assessment by looking for the net labels which were tagged on the enrolled nets. All the enrolled nets with their respective net IDs and households to which they belong were documented in the net master lists which we have filed and kept in a locked metallic suit case at the PACE head office.

2.3.2.6 Withdraw of nets for bioassay/chemical analysis

2.3.2.6.1 Targets for withdraw of nets for Bioassay

The study had 2 aspects considered and these are the physical durability which is assessed through conducting the net hole assessment and the insecticidal efficacy durability which is assessed through the bioassay laboratory assessments. This section focuses on the bioassay aspect of the study.

The study has a target of withdrawing 150 nets at each round of assessment including the baseline survey. In each sub county we have a target of 30 nets. Below is a table showing the breakdown of the targets for each brand in the baseline survey.

Table 3: Targets and location for each net brand – LLIN Durability Monitoring Baseline survey – Bio Assay			
District	Subcounty	Brand of interest	Target of samples to be withdrawn
Kakumiro	Kakindo	Permanent 2.0	30
Hoima	Kigorobya Town Council	Royal guard	30
Hoima	Kyabigambire	Permanent 3.0	30
Bunyangabu	Buheesi	Interceptor	30
Kikuube	Kiziranfumbi	Safe net	30

2.3.2.6.2 Bioassay net withdraw procedures

The Research Assistants withdrew nets for bioassay following the protocol . They randomly identified any 2 of the 10 households that were sampled and interviewed in 15 of the 16 sampled villages and then selected the next households on the left and engaged them for the net withdraw. Once they consented then the campaign nets in the household were checked to confirm that they were the nets of the particular brand of interest in that particular sub county and then withdrew nets one from each household. Each withdrawn net was replaced with a new one of the same brand that was withdrawn.

2.3.2.6.3 Bioassay eligibility criteria and questionnaire administration

In the process of withdrawing these nets, we also administered a short hard copy questionnaire to accompany the withdrawn net. These interviews as well as the consenting for the withdraw were also done in Runyoro.

The eligibility criteria for a net to qualify for withdraw included the following:

- ➤ The net should have been received from the 2020 UCC campaign
- ➤ The net should be the brand of interest in the particular sub county where the household belongs
- ➤ The household head should have consented to the withdraw of his net
- ➤ Preference was for nets that were already in use

All the withdrawn nets were packed in Ziplock bags together with their respective consent forms and questionnaires and then packed in metallic suitcases but separately by brand. It is in these suitcases in which they are to be taken to the IDRC laboratory that will do the bioassay assessments.

2.3.2.7 Field supervision/support

The field supervision was done by the PACE research team comprising of the Research manager, Research coordinator, Research Officer and the Field supervisor. We also got support supervision visits from the DHT members of districts where the study was done, the PIs from both PACE and MOH and the study Co Investigators from MOH.

Figure 11: *The PIs from both PACE (PACE ED - Nakamya Phellister) and MOH (Dr. Jimmy Opigo) holding the net in the middle right, the DHO Bunyangabu, UCC 2020 Coordinator, PACE Research manager on the left and the PACE Research coordinator and Team 6 RA(Grace Tusiime) on the extreme right doing a net hole assessment*

2.3.2.8 Summary of Data collected

| District	Subcounty	Net Brand	Targeted Households	Enrolled Households	Targeted nets	Enrolled Nets	% Achieved (Enrolled Nets)
Kakumiro	Kakindo	Permanet 2.0	160	160	480	415	86%
Hoima	Kigorobya T/C	Royal Guard	160	160	480	409	85%
Bunyangabu	Buheesi	Interceptor	160	160	480	404	84%
Hoima	Kyabigambire	Permanet 3.0	160	160	480	385	80%
Kikuube	Kiziranfumbi	Safenet	160	160	480	418	87%

Figure 12:Enrolled cohort study nets
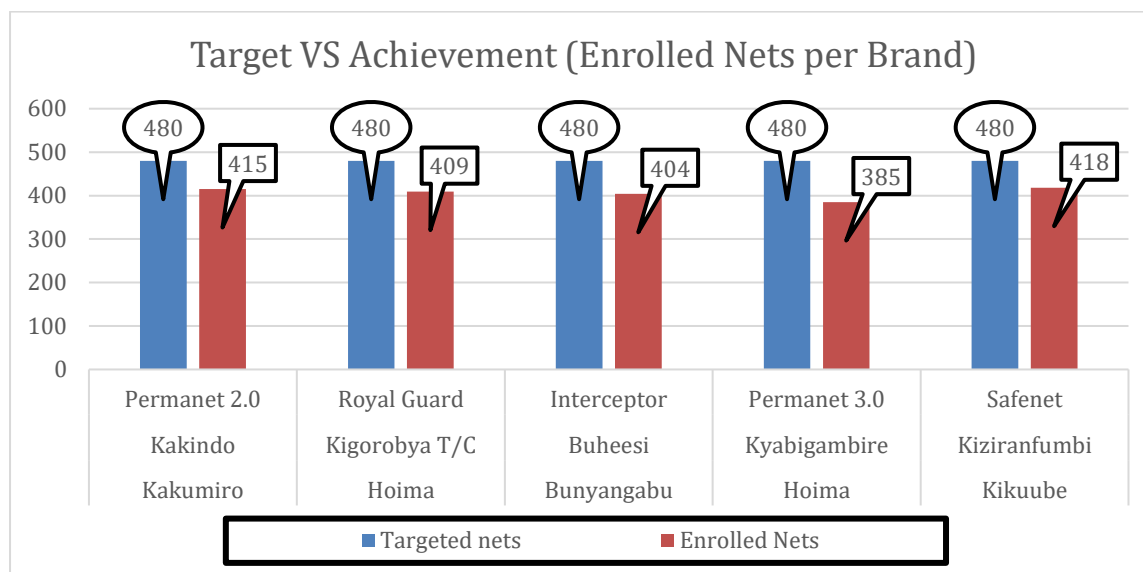
2.3.2.8 Baseline survey Logistics

In terms of logistics, the baseline survey took on several services and commodities. The services utilized include the following:

- ➢ Vehicle hire / transport services.
- ➢ Printing services
- ➢ Venue hire services
- ➢ Printing and photocopy services.
- ➢ Airtime The commodities we used include:
- ➢ Metallic suitcases which were used for storage of the nets withdrawn for bioassay assessment and the consent forms and tally sheets.
- ➢ Stationary such as flip charts, markers, notebooks, pens, stapling machines, punching machines for the field refresher training, ink pads and ink to facilitate thumb printing during consenting for interviews, rubber bands to bundle together the net labels during field work.
- ➢ Back bags for carrying field work materials to and from the field, zip lock bags for keeping the withdrawn bioassay nets.
- ➢ Sanitizers and masks for adherence to the COVID prevention guidelines.
- ➢ MOH and NMS provided the study with nets for replacement of the withdrawn bioassay nets.

2.3.2.8 COVID 19 risk mitigation

The LLIN Durability Monitoring baseline survey was implemented during the COVID times and therefore it was very important to have prevention measures in place against COVID 19. The prevention measures used include the following:

- ➢ Provision of sanitizer and masks to field teams, VHTs LC chairpersons Spacious vehicles (super customs) were used by the Research Assistants, and this allowed for social distancing even within the field vehicles to reduce the contact risk.
- ➢ During the interviews in which the Research Assistant interacted with in the community, social distancing was mandatory during the interviews.

> potential respondents were screened for COVID 19 risks. This screening was part of the data collection tool, and the screening questions were the first to be asked and the responses were assessed for the magnitude of the risk and where there was high risk and likelihood of the sampled household having COVID19, then the household was replaced with another household with no COVID 19 risk.

> The data was collected electronically to reduce the risk of transmitting COVID 19 through many people touching the same papers.

2.3.2.9 Challenges faced

> In some of the sampled villages,50% or more of the interviewed households received few nets (only 2). This led to fewer nets being enrolled than initially planned. 33 out of 72 villages (46%) had received 2 or less nets yet the average number of nets required for each village to meet the nets target was 30 implying a minimum of 3 nets from each of the 10 interviewed households. Out of these only 98 out of 800 (12%) had received less nets than what they were supposed to receive based on the number of household members.

> Some of the sampled villages (3 out of 72 villages) (Kasenyi, Busera and Kajoga) had some sampled households (two, four and one respectively) that had different native languages than the earlier anticipated and planned for. We amended the protocol and informed consent forms to cater for this.

> One of the recruited Research Assistants did not turn up for field work. This somehow distorted our previous arrangement of having 20 teams of 2 people each. We ended up having less teams (19 teams) and one of them had 3 people. This implied that we had fewer teams to provide support when needed.

> Some of the sampled households were not available on the days scheduled for interviews and were therefore replaced. This was mostly common when the days for interviews collided with community events such as market days.

> In some villages the VHTs and chairpersons did not know all the heads of the households for their respective villages. We therefore resorted to asking the neighbors of these households. This was most common in Kigorobya Town council where most of the residents were tenants.

2.3.2.10 Lessons Learnt

> Conducting weekly activity review and planning meetings with the Research Assistants is very important to draw lessons learnt and prevailing challenges so that immediate solutions and revisions in the implementation strategies are made as long as there is no deviation from the approved study protocol.

> Data and study document verification meetings are also very important to weed out any common errors and also identify any key missing data and documents and those that need correction while the team is still in the field and can go back to the households for clarification.

> Having communication platforms such as WhatsApp where challenges encountered in the field, immediate enquiries can be made and instruction given is very helpful and time saving in management and supervision of fieldwork.

> Involvement of LC1s and VHTs in the planning and implementation process is very key for the quick achievement of the intended targets.

> Profiling the sampled villages before starting work enables the study team to identify critical issues that may affect the quality of data collected such as language barrier and community norms, cultures and in regard to the study activities.

2.3.3 Insecticidal Effectiveness Assessment

Bioassays were performed against Interceptor, Permanent 2.0, Safenet, Permanent 3.0 and Royal Guard field samples to test for bio-efficacy. Standard WHO cone tests were used with an insectary-reared pyrethroid susceptible strain (*An. gambiae* Kisumu) and a pyrethroid resistant strain (*An. gambiae* Busia). The *An. gambiae* Busia strain was characterized prior to performing the bioassays using WHO susceptibility tests with deltamethrin 0.05% and Permethrin 0.75%. The susceptibility tests on the *An. gambiae* Kisumu resulted in 100% mortality with Deltamethrin and 98.04% mortality with Permethrin. This confirmed the susceptibility of the *An. gambiae* Kisumu strain.

Synergist tests were also conducted with deltamethrin (0.05%) + PBO (4%). These tests confirmed the presence of pyrethroid resistance (deltamethrin and permethrin resistance in the *An. gambiae* Busia strain. The PBO synergist tests with deltamethrin showed a greater than 30% increase in mortality compared to deltamethrin alone.

2.3.3.1 Net preparation

30cm x 30cm pieces were cut from standard positions 2, 3, 4 and 5 for each net sampled. Four total pieces were cut for each standard net and 6 net pieces were cut for the PBO net (Permanet 3.0 – 3 pieces in the roof – position 5 – instead of one). Netting samples were labeled with a net ID and the cutting position and were stored in a cool dry place at 4 degrees Celsius until when required for bio-efficacy testing. 3 standard nets namely, interceptor, permanet 2.0 and safe net were assessed in addition to two new generation LLINs namely permanet 3.0 and pyriproxyfen (royal guard).

2.3.3.2 Measuring Bio-efficacy

To measure the bio-efficacy of the LLINs, a susceptible colony of Kisumu strain An. gambiae s.s. mosquitoes susceptible to all four classes of insecticide namely; pyrethroids, organochlorines (DDT), carbamates and organophosphates was used in accordance to WHO cone bioassay procedures. Bio-efficacy tests were carried out using 2-5-day old unfed pyrethroid sensitive mosquitoes reared in the laboratory. The mosquito colony was maintained under laboratory conditions in our fully functional insectary at CPHL, Butabika.

Five mosquitoes at a time were introduced into WHO cones and four cones applied simultaneously onto the net sample with a three-minute exposure of the vectors. After exposure, females were grouped into batches of 10 or 20 in 200 mL plastic cups and maintained at 28°C ± 2°C and 80% ± 10% relative humidity with a sugar solution provided. For each sample this procedure was repeated twice, with a total of 40 test mosquitoes used. For each series a control was run with no exposure and results were included in the data base only if control mortality is less than 5%. Numbers of mosquitoes knocked down was recorded at 60 minutes and knock down rate calculated at 60 minutes (KD60). Percentage mortalities were recorded after 24 hours using immediate and delayed mortality as defined by WHO guidelines. If control mortality exceeded >10%, these results were discarded and the test repeated. Bioassays were carried out at ambient room temperatures and Relative Humidity with data recorded on bioassay forms.

In addition, to using pyrethroid based WHO Cone assays, we evaluated the bio-efficacy of new active ingredients in next generation LLINs such as PBO and Pyriproxyfen using recently adopted consensus protocols. This involved the use of established insecticide resistant colony lines to assess PBO and pyriproxyfen (insect growth regulator) activity.

2.3.3.3 Testing residual efficacy of pyrethroids

To test the residual efficacy of pyrethroids, insectary-raised,2-5-day old, unfed females of a pyrethroid susceptible strain were used. Five mosquitoes at a time will be introduced into WHO cones. For each of the four net pieces tested, 10 mosquitoes (2 cones) were completed on pieces in positions 2 to 5, resulting in 40 mosquitoes used for testing all four net pieces (plus 10 control susceptible mosquitoes on untreated netting surface for quality control. If control mortality exceeded 10%, the test was repeated. The knock-down effect was measured at 60 minutes after exposure (KD60) and mortality recorded after 24 hours of exposure.

2.3.3.4 Testing the overall impact of PBO nets on resistant mosquitoes

To evaluate the overall impact of PBO nets on resistant mosquitoes, a resistant strain was used in cone bioassays (using only net pieces with PBO on them). For PermaNet 3.0, 2 pieces cut from the roof of the net were tested with 10 pyrethroid resistant mosquitoes per piece. 10 control mosquitoes were used to validate the results. For each brand, LLINs from the field were evaluated against a new LLIN with the same active ingredient.

2.3.3.5 Testing the overall impact of alpha-cypermethrin and pyriproxyfen

Net pieces cut from the combination of alpha-cypermethrin and pyriproxyfen were exposed to both pyrethroid susceptible Kisumu colony mosquitoes and pyrethroid resistant mosquitoes. The assessment of pyriproxyfen was done using surviving resistant mosquitoes (from royal guard net exposure) which had been blood fed and their progeny evaluated by the presence or absence of egg batches. A blood fed control batch was used for comparison.

2.3.3.6 Methods of analysis

The table below provides a summary of the laboratory methods we shall use, citing examples of nets to be tested and applicable methods, according to standard operating procedures developed by WHO for standard LLINs and CDC for testing of PBO nets. The standard WHO thresholds for optimal effectiveness of KD60 ≥ 95% or 24-hour mortality ≥ 80% were followed. The proportion of nets that attained optimal effectiveness was the outcome of measure.

2.3.4 Chemical Residue – High Performance Liquid Chromatography (HPLC) Assessment and analysis

The chemical residue assessment and analysis was done by Walloon Agricultural Research Center (CRAW) in Belgium. These were done following the WHO guidelines for chemical residue (HPLC) assessment. The assessment procedures and tests conducted depend on the net insecticide and denier yarn. The details of how the assessment was done for each net brand are discussed below.

      Pre-treatment/storage before analysis : at room temperature in the original packaging, under shelter from direct sunlight.

      Storage : 3 months after analysis.

   1° PermaNet 2.0

      Nature :   deltamethrin 1.4 g/kg (for 100 and 150 denier) or 1.8 g/kg (for 75 denier) coated onto polyester, long-lasting insecticidal net (LN).

      Number :   30 samples.
                Each sample contains 5 pieces of minimum 25 cm x 25 cm, cut from the net according to the WHO specification 333/LN/1 (September 2020) and pooled together to form a sub-sample representative of the entire net.

2° PermaNet 3.0

Nature :   deltamethrin 2.1 g/kg (for 100 and 150 denier) or 2.8 g/kg (for 75 denier) coated onto
polyester for the sides and deltamethrin 4.0 g/kg + piperonyl butoxide 25.0 g/kg,
incorporated into polyethylene for the roof, long-lasting insecticidal net (LN).

Number : - PermaNet 3.0, sides : 30 samples.
Each sample contains 4 pieces of minimum 25 cm x 25 cm, cut from the sides of the net
according to the WHO specification 333+33/LN/1 (NET) (April 2019) and pooled
together to form a sub-sample representative of the entire net.

- PermaNet 3.0, roof : 30 samples.
Each sample contains 3 pieces of minimum 25 cm x 25 cm, cut from the roof of
the net according to the WHO specification 333+33/LN/1 (NETTING & NET) (April
2019) and pooled together to form a sub-sample representative of the entire
net.

Lab Id numbers : Mo 749/1 to Mo 749/30 for
the sides ; Mo 749/31 to
Mo 749/60 for the roof.

3° Interceptor

Nature :   alpha-cypermethrin 5.0 g/kg (for 100 denier) or 6.7 g/kg (for 75 denier), coated onto
polyester, long-lasting insecticidal net (LN).

Number : 30 samples.
Each sample contains 5 pieces of minimum 25 cm x 25 cm, cut from the net according to
the WHO specification 454/LN/1 (August 2015) and pooled together to form a sub-sample
representative of the entire net.

Lab Id numbers : Mo 750/1 to Mo 750/30.

4° SafeNet

Nature :   alpha-cypermethrin 5.0 g/kg (for 100 denier) or 6.7 g/kg (for 75 denier) long-lasting,
coated onto polyester insecticidal net (LN).

Number : 30 samples.
Each net sample contains 5 pieces of minimum 25 cm x 25 cm, cut from the net
according to the WHO specification 454/LN/1 (August 2015) and pooled together to
form a sub-sample representative of the entire net.

Lab Id numbers : Mo 751/1 to Mo 751/30.

5° Royal Guard

Nature :   alpha-cypermethrin 5.0 g/kg (for 150 denier) or 5.5 g/kg (for 120 denier) + pyriproxyfen
5.0 g/kg (for 150 denier) or 5.5 g/kg (for 120 denier), incorporated into polyethylene,
long-lasting insecticidal net (LN).

Number : 30 samples.

Each net sample contains 5 pieces of minimum 25 cm x 25 cm, cut from the net according to the WHO specification 454+715/LN (November 2018) and pooled together to form a sub-sample representative of the entire net.

Lab Id numbers : Mo 752/1 to Mo 752/30.

2.3.4.1 Performed analyses

Analyses	Methods or standards	Sub-contracting (name)	Accredited ISO17025 (A)	Dates
Laboratory sampling	*For PermaNet 2.0, Interceptor, SafeNet and Royal Guard* Pooling of the 5 pieces 1 to 5 of each net sample. Cutting of a circular piece of 100 cm² from the top of the net (piece 3) according to the CRA-W method PA-U10-NET001. This circular piece of 100 cm² is put aside in order to determine fabric weight. Cutting of a quarter from each piece 1 to 5, pooling of the 5 quarters together, cutting into small pieces of 5-10 mm square and homogenization in order to determine the active ingredient content. *For the sides of PermaNet 3.0* Pooling of the 4 pieces 1, 2, 4 and 5 of each net sample. Cutting of a circular piece of 100 cm² from one large side of the net (piece 2 or 5) according to the CRA-W method PA-U10-NET001. This circular piece of 100 cm² is put aside in order to determine fabric weight. Cutting of a quarter from each piece 1, 2, 4 and 5, pooling of the 4 quarters together, cutting into small pieces of 5-10 mm square and homogenization in order to determine the active ingredient content. *For the roof of PermaNet 3.0* Pooling of the 3 pieces 5.1 to 5.3 of each net sample. Cutting of a circular piece of 100 cm² from 1 piece of the net according to the CRA-W method PA-U10-NET001. This circular piece of 100 cm² is put aside in order to determine fabric weight. Cutting of a quarter from each piece 5.1 to 5.3, pooling of the 3 quarters together, cutting into small pieces of 5-10 mm square and homogenization in order to determine the active ingredient content.	-	-	from 2021/07/27 to 2021/08/09
Fabric weight (mass of net per m²)	**CRA-W PA-U10-NET001** (1 determination per net sample) Determination of the weight of the circular pieces of 100 cm² (see Laboratory sampling) and expression of the result as g of net per m².	-	-	2021/08/02, 2021/08/25, 2021/08/26, 2021/08/31 and 2021/09/02

Analyses	Methods or standards	Sub-contracting (name)	Accredited ISO17025 (A)	Dates
Deltamethrin content in PermaNet 2.0 and in PermaNet 3.0, sides	**CRA-W PA-U10-RESSM014** Based on CIPAC 333/LN/(M)/3 (HPLC-DAD), CIPAC Handbook M, page 66. (1 determinations per pooled net sample). Extraction by sonication and heating at 60°C for 30 minutes in heptane in presence of dicyclohexyl phthalate as internal standard and determination by normal phase High Performance Liquid Chromatography with UV Diode Array Detection (HPLC-DAD). (1 determination per pooled net sample)	-	**(A)**	from 2021/08/02 to 2021/08/06
Deltamethrin content in PermaNet 3.0, roof	**CRA-W PA-U10-RESSM021** Based on CIPAC 333/LN/M3/3 (HPLC-DAD), CIPAC Handbook O, page 43. (1 determination per pooled net sample). Extraction by refluxing at about 130°C for 30 minutes with xylene in presence of dicyclohexyl phthalate as internal standard and solvent exchange to hexane. Determination by normal phase High Performance Liquid Chromatography with UV Diode Array Detection (HPLC-DAD). (1 determination per pooled net sample)	-	**(A)**	from 2021/09/02 to 2021/09/08
Piperonyl butoxide content content in PermaNet 3.0, roof	**CRA-W PA-U10-RESSM026** Based on CIPAC 33/LN/(M)/3 (GC-FID), CIPAC Handbook N, page 112. (1 determination per pooled net sample). Extraction by refluxing at about 130°C for 30 minutes with xylene in presence of octadecane as internal standard. Determination by Gas Chromatography with Flame Ionisation Detection (GC-FID). (1 determination per pooled net sample)	-	-	from 2021/08/25 to 2021/09/08
Alpha-cypermethrin content in Interceptor and Safenet	**CIPAC method 454/LN/M2/3 and 570/LN/M/3** CIPAC Handbook P, page 63. Extraction by sonication for 30 minutes with heptane in presence of dicyclohexyl phthalate as internal standard and determination by Gas Chromatography with Flame Ionisation Detection (GC-FID). (1 determination per pooled net sample)	-	-	from 2021/08/26 to 2021/09/01

Analyses	Methods or standards	Sub-contracting (name)	Accredited ISO17025 (A)	Dates
Alpha-cypermethrin and pyriproxyfen content in Royal Guard	GC-FID method based on **CIPAC method 454/LN/M3/3 and 715/LN/M2/3** CIPAC Handbook P, page 73. Extraction by shaking and heating at about 85°C during 45 minutes with heptane in presence of dicyclohexyl phthalate as internal standard and determination by Gas Chromatography with Flame Ionisation Detection (GC-FID). (1 determination per pooled net sample)	-	-	from 2021/08/31 to 2021/09/02

A detailed chemical residue (HPLC) Report is attached for reference (See Appendix 6)

2.3.5 Data Management

The questionnaire was thoroughly tested prior to deployment. Skip patterns and filters, internal consistency checks, range checks and logical checks were programmed to support high quality data collection. Interviewer data was uploaded to a web-based database daily or stored on tablets until they could be transferred. A one-page paper questionnaire was completed for each ITN taken for bioassay and chemical content analysis. The questionnaire was stored with the ITN for transfer to the laboratory. At the end of the survey, the web-based database was downloaded and converted into a Stata data file for analysis. Data values were checked for internal consistency and logic, and coding was applied for non-response or missing values. All operations were documented in Stata "do" files.

2.3.6 Analysis

The household sample is considered approximately self-weighting and so no weights were applied during analysis. Estimates of sampling errors accounted for the clustered survey design.

A household wealth index was computed using standard principal component analysis (PCA) approaches and categorizing households into tertiles. The PCA was performed on variables representing household characteristics (e.g. wall, floor and roofing material) and household assets (e.g. mobile phone, radio, transport options and animals).

Attitudes towards nets and net care/repair were captured using Likert score questions, where respondents stated the extent to which they agreed or disagreed with a standard set of statements. Data from the Likert score questions were summarized into two summary scores (nets and net care/repair) by first recoding the four-level Likert scale to have a value of -2 for "strongly disagree", -1 for "disagree", +1 for "agree" and +2 for "strongly agree". These values for each response were then summed and divided by the number of statements to calculate an overall attitude score. An average score greater than 1 is interpreted as a household respondent with favorable attitudes to a given topic.

The physical integrity of campaign ITNs was assessed in accordance to WHO Guidelines, with the number of holes of size 0.5-2 cm diameter (size 1), 2-10cm diameter (size 2), 10-25 cm diameter (size 3) and >25 cm diameter (size 4) recorded for each net following examination by the team in a well-lit location.[3] Data from the ITN hole assessment was transformed into the proportionate Hole Index (pHI) for each ITN using standard weights defined by WHO:

pHI = Number of size 1holes + (No. of size 2 holes x 23) + (No. of size 3 holes x 196) + (No. of size 4 holes x 576)

Based on the pHI value, ITNs were categorized as "good", "serviceable" or "torn" as defined below. Note that "good" is a subset of all "serviceable" ITNs.

Good: pHI<64 (corresponding to a total hole surface area <0.01m²)
Serviceable: pHI<=642 (total hole surface area <=0.1 m²)
Torn: pHI>642 (total hole surface area >0.1m²)

The outcomes of insecticidal effectiveness will be based on the bioassay results performed by IDRC. For pyrethroid ITNs, standard WHO cone tests used a pyrethroid-sensitive Kisumu strain of *Anopheles gambiae* with five mosquitoes per cone, four sites tested on each net (standard positions 2, 3, 4 and roof) and two replicates per location (8 cone tests with 40 mosquitoes per net in total). The 60-minute knock-down (KD60) and the 24-hour mortality rate (mortality) will be measured.

Samples taken from ITNs selected for bioassays were packaged following standard procedures and shipped to CRA-W lab, Belgium for chemical content testing. Samples from the roof and sides will be tested separately for the PermaNet 3.0 brand. Outcome measures from these tests present the mean and median level of active ingredient across the net brand samples in mg/m² and compare these averages with manufacturer specifications for the insecticides used on the netting.

2.3.7 Baseline survey Results

2.3.7.1 Sample
Sixteen villages were targeted in four of the sub counties (Kyabigambire, Buheesi, Kakindo and Kiziranfumbi) and 8 villages in Kigorobya Town Council. 160 households were targeted in each study site (subcounty). The target for the number of nets in each sub county was 414 and this target was achieved and surpassed in each study site. The number of residents per household was on average 4.7 as expected. The highest number of residents per household was 4.9 (Buheesi subcounty, Bunyangabu district) and least was 4.5 (Kyabigambire subcounty, Hoima district). In total, 2,195 nets (106% of the total) were included in the 2020/2021 campaign cohort.

TABLE 4: TARGETED AND ACHIEVED SAMPLE SIZE

	Clusters		Households		Cohort ITNs	
Study site	Target	Achieved	Target	Achieved	Target	Achieved
Buheesi subcounty	16	16 (100.0%)	160	160 (100.0%)	414	422 (101.9%)
Kakiindo subcounty	16	16 (100.0%)	160	160 (100.0%)	414	459 (110.9%)
Kigorobya Town council	8	8 (100.0%)	160	160 (100.0%)	414	425 (102.7%)
Kiziranfumbi subcounty	16	16(100.0%)	160	160(100.0%)	414	433 (104.6%)
Kyabigambire subcounty	16	16(100.0%)	160	160(100.0%)	414	456 (110.1%)
Total	72	72 (100.0%)	800	800 (100.0%)	2070	2195 (106.0%)

Access to ITNs is an important determinant of ITN use: people need access before they can use an ITN (Table 17). Access can be measured at the household and population level: household access is defined as the proportion of households with one ITN for every two people in the household; population access is defined as the proportion of people that could sleep under an ITN assuming each ITN in a household was used by 2 people.

In all sites, the population access was above 85% which is the target for the UCC campaign 2020 / 2021, the lowest being in Kyabigambire (86%) and the highest being 91% for Kakindo, Kigorobya and Kiziranfumbi.

2.3.7.2 Household and Population ITN Access AND USE

TABLE 5: HOUSEHOLD AND POPULATION ITN ACCESS AND USE

Site	Households with 1 ITN for every 2 people	Population access	Population use
Buheesi sub county	N=160	N=782	N=782
Any ITN	84.4%	94.2%	77.7%
Campaign cohort ITNs (Interceptor)	68.1%	88.4%	67.9%
Other ITNs	10.6%	22.5%	9.8%
Kakindo	N=160	N=742	N=742
Any ITN	85.0%	94.1%	73.3%
Campaign cohort ITNs (Permanet 2.0)	77.5%	90.8%	64.2%
Other ITNs	6.3%	14.7%	9.3%
Kigorobya town council	N=159	N=677	N=677

Any ITN	86.8%	94.7%	65.0%
Campaign cohort ITNs (Royal Guard)	79.2%	90.7%	46.4%
Other ITNs	17.6%	28.5%	18.6%
Kiziranfumbi	N=160	N=718	N=718
Any ITN	87.5%	95.1%	78.3%
Campaign cohort ITNs (Safenet)	77.5%	91.2%	60.0%
Other ITNs	9.4%	25.8%	18.2%
Kyabigambire	N=160	N=681	N=681
Any ITN	88.1%	94.7%	71.2%
Campaign cohort ITNs (Permanet 3.0)	74.4%	86.3%	47.4%
Other ITNs	25.0%	37.0%	23.8%

2.3.7.3 Durability of Campaign ITNs

The durability of ITNs can be conceptualized as two components: *attrition*, or nets that are no longer present in the household; and the *physical integrity* of nets that are available for use in the household. At the time of the baseline survey (3.2 to 5.8 months post-distribution), total ITN attrition was similar across all sites ranging from 84% (Kyabigambire) to 96% (Buheesi sub county). In Kyabigambire which had the highest attrition, most of the nets no longer present in the household (11%) were being used by family elsewhere. This was different from the other four sites which range from 2% to 3% (*p*=0.000). In all sites there were nets reported as given away to others within a range between 2% to 7%. There were few discarded nets (also known as *attrition due to wear and tear*) and this was only in one site out of the five (Kakindo) and it was at 0.2%.

2.3.7.3.1 Campaign Cohort ITN Attrition

TABLE 6: CAMPAIGN COHORT ITN ATTRITION

Site	Months since mass distribution	Total campaign ITN attrition	ITNs given away to others	ITNs discarded*	ITNs Used by family elsewhere at baseline	ITNs lost for other/unknown reason
Buheesi sub county (N=422)	6.0	95.5%	2.1%	0.0%	2.4%	0.0%
Kakindo (N=459)	5.8	90.2%	6.5%	0.2%	3.1%	0.0%
Kigorobya town council (N=425)	3.2	95.3%	4.2%	0.0%	0.5%	0.0%
Kiziranfumbi (N=433)	4.0	93.3%	3.9%	0.0%	2.3%	0.5%
Kyabigambire (N=456)	3.6	84.4%	3.9%	0.0%	11.4%	0.2%

*Also known as attrition due to wear and tear.

2.3.7.3.2 Physical Integrity

The physical integrity component was assessed through the Proportionate Hole Index and the outcome was the proportion of nets surviving in serviceable condition. In all study sites, above

90% of the UCC nets were considered "serviceable". Kakindo had the highest proportion of torn nets (10%) and the lowest proportion of nets in "serviceable" condition (91%). Kigorobya town council had the highest proportion of nets found in "serviceable" condition (99%) and also had the lowest proportion of nets that had ever been used (47%)

TABLE 7: CAMPAIGN COHORT ITNS SURVIVING IN SERVICEABLE CONDITION

Site (Net Brand)	All cohort ITNs			Cohort ITNs ever used and present		
	n	Estimate	95% CI	n	Estimate	95% CI
Buheesi (Interceptor)	402	93.8%	[90.8%-95.8%]	321	92.8%	[89.2%-95.3%]
Kakindo (Permanent 2.0)	412	90.3%	[85.8%-93.5%]	277	87.0%	[81.8%-90.9%]
Kigorobya Town Council (Royal Guard)	403	98.8%	[94.1%-99.7%]	188	97.3%	[89.2%-99.4%]
Kiziranfumbi (SafeNet)	402	92.5%	[88.0%-95.5%]	254	89.0%	[82.6%-93.2%]
Kyabigambire (Permanent 3.0)	384	95.1%	[92.4%-96.8%]	205	91.2%	[86.5%-94.4%]

2.3.7.3.2.1 Causes of net damage

Figure 13 displays the reported causes of net damage, among the 216 households that reported having holes in their nets at the time of the survey (respondents could name more than one cause). The most reported cause of net damage across the five study areas was rats (71%). Damage by rats was relatively less common in Kigorobya town council (57%) compared with other sites. This was followed by tears (23.4%) and damage along corners (20%)

FIGURE 13: REPORTED CAUSES OF NET DAMAGE
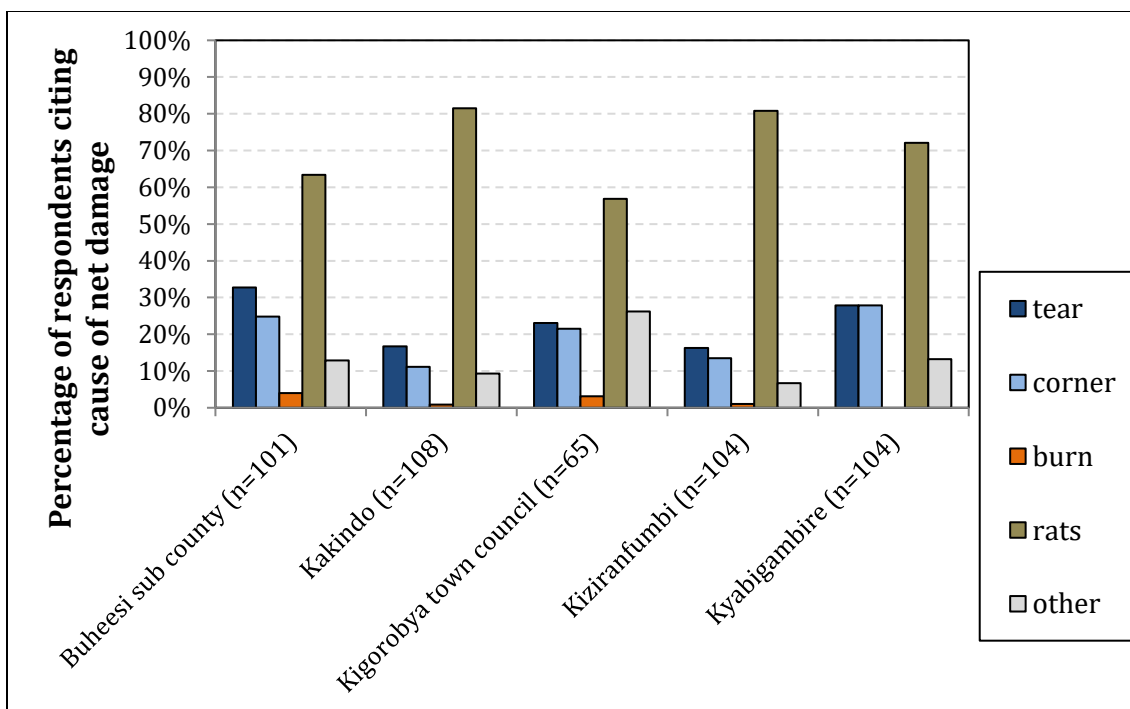
2.3.7.3.2.2 Net Repair practices

Respondents were asked if they had ever experienced holes in any of the mosquito nets they currently possess, including existing non-campaign nets (Table 12). Overall, 55.8% of all respondents reported holes, with no Kigorobya town council and Kyabigambire reporting the least proportions of households experiencing holes in the nets owned at 40.6% an 42.5% respectively. Other study sites had significantly higher proportions (p<0.001). Discussion of net care and repair was low across all sites as was hole repair amongst those households that had experienced them.

TABLE 8: NET CARE & REPAIR EXPERIENCE

Site	Household ever had holes in nets	Ever discussed net care and repair	Ever repaired a hole in a net*
Buheesi sub county (n=160)	63.1%	35.0%	21.8%
Kakindo (n=160)	67.5%	37.5%	22.0%
Kigorobya town council (n=160)	40.6%	36.3%	29.2%
Kiziranfumbi (n=160)	65.0%	23.8%	23.1%
Kyabigambire (n=160)	42.5%	30.0%	27.9%

* Among households reporting ever having holes in nets: Buheesi=101, Kakindo=108, Kigorobya TC =65, Kiziranfumbi=104, Kyabigambire=68; Total n=446

2.3.7.4 Bio efficacy Effectiveness

The outcome of measure was the proportion of nets that attained optimal effectiveness and for the Royal Guard component of growth regulation done by Pyriproxyfen, the outcome of measure

was the proportion of nets to which mosquitoes were exposed and they did not lay eggs after exposure. Below is a summary table of the results.

Table 9. Summary of Nets that attained Optimal effectiveness level per brand

Net Brand	Attained KD Optimal Effectiveness (>=95%)		Attained Mortality Optimal effectiveness (>=80%)		Attained Optimal effectiveness (Either KD or Mortality)		Growth Regulation effectiveness (Royal Guard)	
Interceptor	24	80.0%	0	0.0%	24	80.0%		
Permanent 2.0	20	66.7%	0	0.0%	20	66.7%		
SafeNet	30	100.0%	1	3.3%	30	100.0%		
Permanent 3.0 (Roof)	30	100.0%	27	90.0%	30	100.0%		
Permanent 3.0 (sides)	23	76.7%	18	60.0%	28	93.3%		
Royal Guard sides	30	100.0%	29	96.7%	30	100.0%		
Royal Guard Roof	30	100.0%	30	100.0%	30	100.0%		
Royal Guard Oviposition (Growth Regulation)							30	100%

A detailed Bio efficacy report is attached. See Appendix 5)

2.3.7.5 Chemical residue (HPLC)

The outcome measure for the chemical residue (HPLC) analysis was proportion of nets that fall within the range of the expected content / target dose of the content depending on the insecticide and the denier yarn for each net brand. Below is a summary of the chemical residue results. A detailed chemical residue report is attached. Each brand had a lower limit and upper limit within which the nets assessed were expected to fall at baseline (Between 0 – 6 Months).

Table 10: Proportion of nets within the chemical dose boundary per brand (n=30)

Brand	Insecticide	Percentage within target dose	Target dose for insecticide
Royal guard	Alpha-cypermethrin	93.3% (28/30)	5.5 g/kg ± 25 % (4.1 g/kg – 6.9 g/kg)
Royal guard	pyriproxyfen	50% (15/30)	5.5 g/kg ± 25 % for 120 (4.1 g/kg – 6.9 g/kg)
Permanet3.0 (Roof)	Deltamethrin	96.6% (29/30)	4.0 g/kg ± 25 % (3.0 g/kg – 5.0 g/kg)
Permanet3.0 (sides)	Deltamethrin	73.3% (22/30)	2.1 g/kg ± 25 % (1.6 g/kg – 2.6 g/kg)
Permanet3.0 (Roof)	Piperonyl Butoxide	43.3% (13/30)	25.0 g/kg ± 25 % (18.8 g/kg – 31.3 g/kg)
Interceptor	Alpha-cypermethrin	60% (18/30)	5.0 g/kg ± 25 % (3.8 g/kg – 6.3 g/kg)
Permanet 2.0	Deltamethrin	53.3% (16/30)	1.4 g/kg ± 25 % for 100 (1.1 g/kg – 1.8 g/kg)
SafeNet	Alpha-cypermethrin	26.6% (8/30)	5.0 g/kg ± 25 % (3.8 g/kg – 6.3 g/kg)

The possible factors that influence net durability and Bio efficacy of the nets include the net use, net handling practices, washing nets with detergent, hanging nets in the sun and mean washes which is a measure of how often the nets are washed. Below is a summary of these factors for the nets withdrawn for Bio efficacy and chemical residue assessments.

2.3.7.6 Handling of Bioassay Test ITNs

TABLE 11: HANDLING OF BIOASSAY TEST ITNS

	Kakindo	Buheesi	Kiziranfumbi	Kyabigambire	Kigorobya TC
	Permanet 2.0	(Interceptor)	Safenet	Permanet 3.0	Royal Guard
	N=30	N=30	N=30	N=30	N=30
Location found					
Hanging and folded up or tied	37%	30%	50%	27%	60%
Hanging loose over sleeping place	60%	70%	50%	63%	40%
Not hanging but not stored	3%	0%	0%	7%	0%
Stored away still in package	0%	0%	0%	3%	0%
Type of sleeping space (if used)					
Bed frame (finished)	20%	27%	40%	30%	57%
Bed frame (sticks)	53%	60%	23%	53%	30%
Foam mattress	17%	10%	23%	10%	10%
Ground	10%	0%	10%	0%	3%
Never used	0%	0%	0%	7%	0%
Reed mat	0%	3%	3%	0%	0%
Net users					
Adult only	57%	60%	50%	50%	53%
Older child, adolescent only	0%	10%	7%	3%	0%
Other	0%	0%	0%	7%	0%
Young child with adult	17%	10%	10%	7%	27%
Young children only	27%	20%	33%	33%	20%

TABLE 12: REPORTED USE OF BIOASSAY TEST ITNS

	Kakindo	Buheesi	Kiziranfumbi	Kyabigambire	Kigorobya TC
	Permanet 2.0	(Interceptor)	Safenet	Permanet 3.0	Royal Guard

	N=30	N=30	N=30	N=30	N=30
Used last night	100%	100%	100%	90%	93%
Used last week					
Every night (7 nights)	97%	100%	97%	87%	90%
Most nights (Net is not used at all-6)	3%	0%	0%	7%	3%
Net is not used at all	0%	0%	0%	3%	0%
Not used last week	0%	0%	0%	3%	7%
Some nights (1-Not used last week)	0%	0%	3%	0%	0%
Seasonal use					
Equally in rainy and dry season	97%	100%	100%	90%	93%
Mainly rains, sometimes also dry	0%	0%	0%	3%	3%
Not used at all	0%	0%	0%	7%	0%
Only rainy season	3%	0%	0%	0%	3%

TABLE 13: REPORTED WASHING OF BIOASSAY TEST ITNS

	Kakindo Permanet 2.0	Buheesi (Interceptor)	Kiziranfumbi Safenet	Kyabigambire Permanet 3.0	Kigorobya TC Royal Guard
	N=30	N=30	N=30	N=30	N=30
Ever washed	90%	70%	60%	47%	40%
Washes in the last 6 months among all nets					
Mean	2.0	1.0	0.9	0.6	0.6
Median	2	1	1	0	0
Washes in the last 6 months among washed nets					
Mean	2.2	1.5	1.4	1.4	1.4
Median	2	1	1	1	1
Soap used for last wash					

Detergent (OMO etc)	20%	13%	13%	7%	13%
Mix	0%	7%	10%	0%	3%
None	0%	7%	3%	0%	3%
Not Applicable	10%	23%	40%	53%	60%
Soap bar	70%	50%	33%	40%	20%
Where dried after last wash					
Dried in the shade	47%	40%	37%	33%	30%
Dried in the sun	43%	30%	23%	13%	10%
Not Applicable	10%	30%	40%	53%	60%

2.3.8 Baseline survey Report writing

The PACE study team Wrote the comprehensive baseline report including all the three study components (Physical integrity, Bioassay, and chemical residue). This report includes all the study components of physical durability, Bioassay efficacy and chemical residue. A copy was shared with MOH. The focus of the report is on addressing the study objectives of establishing the median survival and Bio efficacy of the five net brands being studied. It is however key to note that the Baseline survey results alone cannot be used to make conclusive deductions to answer the study objectives. *A copy of this report has been attached. See Appendix 4)*

2.3.9 Dissemination of Baseline results and data use for decision making

The PACE study team presents results to the programme, MOH management and stakeholders at scheduled meetings. In collaboration with NMCD, PACE team also presented results to the district leadership through the District Orientation and feedback meetings during the 12 months survey data collection to Hoima and Kikuube districts on 1st December 2021 and 10th of November 2021. Fora where results have been disseminated at national level include the following.

- IVM Technical Working Group on (Baseline study results-Durability)
- Malaria partners coordination meeting (Baseline study results)
- The Global Fund Sub Recipient (SR) performance review meetings at Serena Hotel on the 28th of February 2022 and another on the 10th of June 2022 at Serena hotel.
- IVM Technical Working Group on (Baseline study results- Chemical Analysis) on 19 May 2022
- World Malaria Scientific Conference on 22nd April 2022 (SBC results)

All these presentations are attached to this report. (See Appendices 7 – 10)

Key messages and recommendations that have emerged from the field observations and baseline results include:

    1) Rodents contribute significantly to net damages in the households.

    2) Net use was 73% below the UMRESP target of 85%;

3) At baseline one of the standard nets (Permanet 2.0) did not attain optimal effectiveness for either KD or mortality.

The programme engaged the district leadership during supervision visits and provided guidance on how to improve net use, care, and repair to maintain optimum net coverage and use levels. Implications on programme implementation are:

1. **Rodent control:** It was noted that rodents are attracted to nets that have been handled with food soiled hands, mostly by children given that household members often eat and sleep in the same space or spaces in close proximity. Rodent control should be an integral part of hygiene in the household and at community level. Messaging on rodent control is now integrated in the routine net and health messaging at all levels.
2. **Net use:** Net use at baseline was at 73% below the UMRESP target of 85%; to raise and sustain optimum net use levels, the programme should intensify net use, care and repair messaging as part of social behavior change. To this end the programme and partners have and continue disseminate the attendant guidelines through available channel (trainings, supervisions, community dialogues, radio talk shows etc
3. **Net effectiveness:** At baseline one of the standard nets (Permanet 2.0) did not attain optimal effectiveness for either KD or mortality. The programme has prioritized next generation nets in subsequent net campaigns.

A high-level dissemination meeting will be held once the 12 months survey data analyses is completed to share these results with the MOH and the key stakeholders. In addition, the NMCD will maintain close interaction with the study team to ensure that results and lessons learned from this study inform programme implementation in real time.

2.3.9.1 Recommendations for the Planning and strategy of the upcoming UCC 2023
1. There is need for IEC materials that clearly talk about the 24-hour period for hanging the nets in a shade before use. This comes from concerns and experiences shared by the communities of body itching after using the nets immediately after distribution. The 2023 campaign needs to emphasize this during the VHT or distribution teams trainings.
2. More strict measures of verifying that the registration and distribution teams are following the net allocation formular of one net per two people in each household as well as the capping limit are adhered to. Of the 800 households interviewed at baseline, 29% received more nets than they were supposed to receive based on the UCC2020 formular of one net per two people. Of these, 2% (18 households) received more than five nets which was the maximum capping set for the campaign.
3. As shown in the net attrition results above, use of nets by family members elsewhere outside the household accounts for 48% of the net attrition. According to feedback from the households not captured by study questionnaire, these are mainly boarding school

children. There is therefore need for the upcoming UCC 2023 to put into consideration the distribution of nets in boarding schools besides households.

2.3.9.2 Routine SBCC Recommendations drawn from baseline results

2.2.9.2.1 Effective communication channels

Interpersonal communication (VHTs) at 73% of 800 households, was the most cited channel for accessing malaria related information, followed by media (radio, newspapers, Television) cited by 53%. The most common net related messages heard and recalled were "use net every night" heard by 85%, followed by "hang net" heard by 47%, "Nets prevent malaria" was heard by 45%, "care for net" was heard by 44%. Only 10% heard "Repair net". Therefore, in the future LLIN UCCs, the IPC communication channel mostly the VHTs can be used for SBCC during and after the campaign.

2.2.9.2.2 Behaviors to target for SBCC (Net use, care and repair)

Positive attitudes towards net use and net care / repair, were at 65.9% and 47.6% respectively. Net access was at 95% and Net use at 73%. When we come to the net care practices, the mean and median washes in six months were 2 which implies that if this is upheld then in the 3 years the mean will be only 12 times which is below the threshold of 20 times. Washing nets a few times or only when necessary is a good behavior that preserves the net Bio efficacy and needs to be encouraged in the future net campaigns. On the other hand, the behaviors that need to be discouraged include the following:

1. Not folding or tying up hung nets during daytime which exposes them to a high risk of tears. This behavior was observed in 48% of the 800 households.
2. Using detergent or bleach to wash the nets which heavily depletes the insecticide. This was reported for 27% of the 2026 nets.
3. Drying nets on bush or fences. This was reported for 21% of the 2026 nets.
4. Storing food in the same room used for sleeping where the mosquito nets are hanged. This behavior was reported for 43% of the 2026 nets. The risk created by this behavior is the attraction of rodents to where the nets are hanged or kept, and the nets are consequently eaten by rodents. Consequently 71% of the holes observed in the nets were caused by rodents.
5. Use of nets from the previous campaign which are more than 3 years old. The baseline results showed that only 62% of the newly distributed nets of the 2020 campaign had ever been used yet 15% of the 2674 nets found in households were still being used yet they were from the previous campaign. This clearly shows that some households prefer to keep the new nets with better Bio efficacy, and they continue using the old nets from the previous campaign that have exceeded their three-year life effective life span. Households therefore need to be sensitized about the productive lifetime of the nets to appreciate why it's better for them to use the newly distributed nets.
6. When we come to the net repair practices, out of 55.7% of 800 households that had ever experienced holes in the nets, only 32.5% had ever discussed net care and repair while

only 24.8% had ever repaired a hole in the net. This clearly shows that there is need for more efforts to be dedicated towards SBCC about net repair.

2.2.9.2.3 Net Repurposing

It was observed that households repurpose old nets in different ways which include using them in gardens, using them to make ropes, make hanging lines for clothes and making poultry houses. During discussions with these households, they express the need for net repurposing guidelines to manage any environmental hazards that may result from repurposing old nets.

2.4.0 12 Months survey

This is a cohort study and therefore the 12 Months survey was conducted in the same study area and households that were recruited at baseline survey. The UCC 2020 campaign nets enrolled into the cohort at baseline were followed up at 12 Months for assessment. The 12 Months survey was conducted in two waves based on when the study sites received the nets in the campaign. Buheesi and Kakindo sub counties were covered in August 2021 while Kigorobya Town Council, Kiziranfumbi and Kyabigambire were covered in November 2021. In both these waves, Data collector trainings, district feedback and planning meetings were conducted before data collection. Below are the details of how these activities were conducted in the 12 months survey.

2.4.1 Data collector training

There were some changes in the household identification procedures and the data collection tools as well. In addition to this, the time lag between the baseline and the 12 Months survey necessitated having refresher trainings for the data collection team to fully understand the study SOPs and study assessment procedures.

The trainings took place in Hoima. The first one in August 2021 was at Hoima Resort Hotel from 16/8/2021 to 20/8/2021 and the second one in November 2021 took place at UNHCR Youth center from 3rd November 2021 to 9th November 2021. Both trainings were facilitated by Research manager (Dr. Alex Ndyabakira), Research officer (Joel Kato), Data management coordinator (Collin Baluku) and the Human Resources Manager (Olivia Nalunkuuma).

The training was highly practical and included illustrations and sessions of hole assessment practice by each trainee and role play interviews. The training content covered includes the following:

1. Research ethics
2. Study Protocol and Methodology
3. Data collection procedures and tools
4. Data Quality
5. PACE policies

The training was evaluated through a post and pretest before and after the training respectively. The questions of the evaluation were about the training content. The trainees that performed

poorly (below 50%) were given more explanation by the PACE Research team to understand the particular concepts where they needed further explanations and illustrations.

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1. Research ethics
2. Study Protocol and Methodology
3. Data collection procedures and tools
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Figure 14: Trainees practicing Net Hole Assessment during the training

2.4.2 Data collection

2.4.2.1 Data collection methodology and strategy

The LLIN 12 Months survey was quantitative in design according to the approved protocol and therefore data was collected using a structured questionnaire which was also used in other countries where the study was conducted. This questionnaire was programmed into a mobile application and installed on the study tablets.

The sub counties were covered one by one starting with Kakindo which had Permanent 2.0 nets and then Bunyangabu which had Interceptor was covered next. The entire team of 10 teams was placed in one sub county at a time. Within a sub county, the first 10 villages were covered by one team each and then the next 6 villages were covered by 2 teams each. The sixth team which had only one person was supported by the volunteer who was doing document review who had no specific team. Each village had between 9 to 10 eligible households.

All data collection was done electronically. The cohort survey tool was programmed in a customized app by Deron Ltd. The Bioassay data was collected using Kobo toolbox / ODK (Open Data Kit)

2.4.2.2 Survey duration
The 12 months survey took 24 days in total. The training took 5 days, the data collection in each village took on average took 2 to 4 days depending on the number of teams assigned that particular village, the number of eligible households in the village, the number of households interviewed, how distant the households were from each other within the village and the convenient availability of the respondents. Some respondents preferred to be interviewed at a later time by appointment hence pushing the data collection period further. The number of cohorts nets a household had was also very key in the determining the amount of time it took to complete data collection at a household and in the village subsequently.

2.4.2.3 Community sensitization, mobilization, and entry
We had the community entry at different levels. The first level was the district level, then the next was the sub county level and lastly the village level. At district level, the PACE Research team conducted a district entry meeting with the district leadership of each of the 2 districts (Kakumiro and Bunyangabu) and Hoima where we had the training. During these meetings, we gave the District Health leadership (DHO, Malaria focal person, VCO, Biostatistician and others) an overview of the 12 months survey and the support and role we expected from them.

The districts pledged support to the study and also pledged to participate in the supervision of data collection where possible. Regarding this, the Bunyangabu District Health Team led by the DHO participated in the supervision of data collection.

The district leadership also provided us with information about the possible community events and structures such market days and poor road networks that we needed to consider when planning the fieldwork / data collection.

After the district level entry and mobilization, we then went to the sub county level. At sub county level, we introduced the study to the sub county leadership and discussed with them how they can support the 12 months survey. The Buheesi sub county chief provided the study team meeting space at the sub county hall which we used for meeting throughout our stay in Buheesi sub county. The Buheesi Health Assistant also supported the study in compiling the contact list and mobilizing the VHTs of the study villages.

After the sub county level, the next was village level and the entry and mobilization for this level was done by the volunteers. At village level, the community entry and mobilization activities included introduction of the 12 months survey to the LC1 chairpersons and VHTs and also giving them talking points to follow when introducing the teams to the cohort households.

2.4.2.4 Engagement of eligible cohort households for 12 months survey

2.4.2.4.1 Eligibility / Inclusion criteria
Regarding the eligibility of the households to be engaged for interview at 12 months, the households that still had at least one net present out of those received from the UCC campaign

and had at least one net assessed at baseline were eligible for interview at 12 months. These were considered eligible even when they no longer had any net present at the time of 12 months survey.

2.4.2.4.2 Exclusion criteria

The households that were excluded include the following:

- ➢ Those that had lost all the UCC 2020 nets they received by the time of the baseline survey were not considered eligible for 12 months survey.
- ➢ Those that did not have any adult of sound mind fit to be interviewed

The full list of eligible cohort households for each village was given to the volunteers by the PACE Research Officer and they were identified and located with the help of the LC1 chairperson or the VHT who were well acquainted with the households in their villages.

The volunteers used the household master list to keep a record of the households still in the cohort in every village. On this household master list, they recorded the village name and code, name of the household head, the household ID, the number of UCC nets enrolled in each household and the other nets found in each household.

The cohort households can best be identified in the future rounds of data collection by following and asking for the head of household name after reaching the study villages.

2.4.2.5 Questionnaire administration

2.4.2.5.1 Target Respondents

The interviews were conducted at the cohort households with the heads of households, their spouses or a representative of the household head that was well knowledgeable about the household.

2.4.2.5.2 Consenting procedures

The households were consented y signing the hard copy consent forms for participation in the entire study of 3 years at baseline. However, at 12 months, the respondents gave their verbal consent to participate in the 12 months survey. This was done after the volunteers who collected data providing the relevant information about the participation in the 12 months survey adequate for making the decision to consent to participate in the interviews. The respondents were verbally consented in their local languages or other languages of choice they understood best.

For Bioassay, since new households are sampled for each time period survey, these households provided written consent to participate in the survey. They were consented in their local languages, and most were consented in Runyoro and the consent was documented by the consent forms in Runyoro. Both the respondent and the interviewer signed the consent form before administering the short questionnaire and withdrawing the Bioassay net from the household. In cases where the respondent could not read and write, the respondent identified someone who could read and write to be a witness on his or her behalf while the volunteers

read the consent forms to the respondents. The witnesses in such cases also read through the consent forms on behalf of the respondent before signing. In such cases the respondent then put his or her thumb print in the designated space on the consent form. 2 consent forms were signed, and one copy was left with the respondent and then the second was brought back to the study coordination / PACE Research team and we filed them together with the tally sheets for each household in suspension files which are kept in a locked metallic suit case at the PACE headquarters. These are only accessed by the study team when necessary, which has all done the Research Ethics certificate and fully understand the concept of protecting human subject in research.

2.4.2.5.3 Interview administration

The research volunteers conducted the interviews in the languages the respondents understood best which for most was Runyoro being the local language of the study region. The interviews were done using a structured questionnaire which was electronic, and the responses were entered in an electronic system installed on the project tablets received by the volunteers.

The volunteers applied several interviewing skills where necessary and applicable such as probing, prompting, observation of nonverbal communication and not asking leading questions.

The interviews were conducted in observation of confidentiality and only the respondent was present during the interviews. The LC1 chairperson and the VHT were asked to kindly wait in a place where they could not listen to the interviews.

The interviews took an average of one and half hours to 2 hours depending on the number of nets found in the household and the number of holes the nets had. Where the nets were many and had many holes, the interviews took about 2 hours while where the nets were few and the holes were few also then interviews took about one hour.

At the end of every interview, the volunteers compensated the respondents with 10,000 shillings as communicated in the training. This was also documented on the payment forms where the respondents signed after receiving this money.

These payment forms were filed and kept in locked cabins at the PACE head office with limited access. They are only accessed by the study team when necessary.

2.4.2.5.4 Assessment of cohort nets

This study has a cohort of nets which are the UCC 2020 nets received and still available within the eligible cohort households. The study particularly targets 5 brands which are; Permanent 2.0, Permanent 3.0, Royal guard, Interceptor and Safe net. All cohort nets eligible for assessment at 12 months were tagged with labels bearing unique ID numbers. The target for the nets to be assessed in each sub county is equivalent to the eligible cohort nets enrolled into the study at baseline. Below is a table showing the targets or number of eligible cohort nets and location for each net brand.

2.4.2.5.5 Summary of Data collected

District	Subcounty	Net Brand	Targeted Households (Eligible for 12 Months survey)	Households Interviewed	Targeted nets (Eligible for 12 Months survey)	Nets assessed at 12 Months)	% Achieved (Assessed Nets)
Kakumiro	Kakindo	Permanet 2.0	157	144	414	365	88%
Hoima	Kigorobya T/C	Royal Guard	156	144	405	329	81%
Bunyangabu	Buheesi	Interceptor	160	160	480	404	84%
Hoima	Kyabigambire	Permanet 3.0	155	152	385	326	85%
Kikuube	Kiziranfumbi	Safenet	157	149	405	323	80%

Within the cohort households, a list of the cohort nets in each cohort household was provided to the volunteers for reference while in the households. All these nets that were tagged with labels that had unique ID numbers were verified further by checking the brand labels. This was done because in some instances the household members had changed the study net labels to other non cohort nets after they had been plucked off by children or during washing. In addition to this, some of the cohort nets had lost the study net labels totally and therefore there was need for a verification / inclusion criterion for the nets to be assessed at 12 months. These included the following:

- ➢ The net was received from the 2020 UCC campaign
- ➢ The net is the brand of interest in that particular sub county where the household to which it belongs is located. (Permanent 2.0 for kakindo, Interceptor for Buheesi, Safe net for Kiziranfumbi, Royal guard for Kigorobya T/C and Permanent 3.0 for Kyabigambire)
- ➢ The net is either tagged with a study label with a unique ID or if it is missing, then the household members say it had a study label it just went off.

- The net can be traced and seen on the list of cohort nets for that particular household provided by the PACE Research officer.
- In cases where the study net label had been plucked off the net, then reference was made to the baseline database to check for who slept in the net of interest the night before the baseline survey was conducted at the household. Once the household members matched the study net label to who slept in it at baseline, then it was assessed.

The criteria for exclusion of nets include the following:

- Nets that had the study label, but the net brand label was showing that it was not the brand of interest for that particular sub county.
- Nets that had no study labels and could not in any way be linked to any study net label among those listed for that household.

All the confirmed cohort nets were assessed for hole size and number. This was done by use of marked rulers following the categories of the hole sizes communicated in the training where we had size 1, size 2, size 3 and size 4. The documentation of the net hole assessment was done by use of tally sheets. All confirmed cohort nets that were found with no study labels or where the study labels had become weak and would soon get plucked off, then a new study label was stitched to the net. The criteria we followed for categorization based on which our rulers were marked is shown in the table below.

SIZE	CRITERIA
Size 1	>05cm to 2cm
Size 2	>2cm to 10cm
Size 3	>10cm to 25cm
Size 4	>25cm

The cohort nets can best be identified in future rounds of assessment by looking for the net labels which were tagged on the enrolled nets. All the enrolled nets with their respective net IDs and households to which they belong were documented in the net master lists which we have filed and kept in a locked cabin at the PACE head office.

2.4.2.6 Withdraw of nets for bioassay/chemical analysis and interviews

2.4.2.6.1 Withdraw of nets for Bioassay

The study had 2 aspects considered and these are the physical durability which is assessed through conducting the net hole assessment and the insecticidal efficacy durability which is assessed through the bioassay laboratory assessments. This section focuses on the bioassay aspect of the study.

The study has a target of withdrawing 150 nets at each round of assessment including the 12 months survey. In each sub county we have a target of 30 nets. Below is a table showing the breakdown of the targets for each brand in the 12 months survey.

Table 15: Targets and location for each net brand – LLIN Durability Monitoring 12 months survey – Bioassay			
District	Subcounty	Brand of interest	Target for cohort
Kakumiro	Kakindo	Permanent 2.0	30
Hoima	Kigorobya Town Council	Royal guard	30
Hoima	Kyabigambire	Permanent 3.0	30
Bunyangabu	Buheesi	Interceptor	30
Kikuube	Kiziranfumbi	Safe net	30

2.4.2.6.1.1 Procedures for withdraw of nets for bioassays

The volunteers withdrew nets for Bioassay following the protocol as communicated in the training. They randomly identified any 2 of the cohort households that were interviewed in 15 of the 16 study villages and then selected the next households on the left and engaged them for the net withdraw. Once they consented then the campaign nets in the household were checked to confirm that they were the nets of the particular brand of interest in that particular sub county and then withdrew nets one from each household. Each withdrawn net was replaced with a new one of the same brand that was withdrawn.

2.4.2.6.1.2 Eligibility criteria for Bioassay nets and questionnaire administration

In the process of withdrawing these nets, we also administered a short electronic questionnaire on ODK to accompany the withdrawn net. These interviews as well as the consenting for the withdraw were also done in Runyoro.

The eligibility criteria for a net to qualify for withdraw included the following:

- ➢ The net should have been received from the 2020 UCC campaign
- ➢ The net should be the brand of interest in the particular sub county where the household belongs
- ➢ The household head should have consented to the withdraw of his net
- ➢ Preference was for nets that were already in use

All the withdrawn nets were packed in Ziplock bags together with their respective consent forms and questionnaires and then packed in metallic suitcases but separately by brand. It is in these suitcases in which they are to be taken to the IDRC laboratory that will do the bioassay analysis.

2.4.2.7 Field supervision/support

The field supervision was done by the PACE research team comprising of the Research manager, and Research Officer with the support of three field supervisors hired as consultants. We also got support supervision visits from the DHT members of districts where the study was done, the PIs from both PACE and MOH and the study Co Investigators from MOH.

Figure 16: *The PIs from both PACE (PACE Programs Lead – Rebecca Babirye in PACE T.Shirt, PACE Research manager on extreme left and MOH PI (Dr. Jimmy Opigo – Assistant commissioner NMCD in light blue shirt) observing the PACE Research officer and a volunteer conducting Net Hole Assessment*

Figures 17 and 18: Dr. Jimmy Opigo (PI from NMCD) in the blue shirt, Medard Rukaari (LLINs Coordinator) in brown NMCD jacket, the PACE Research manager and the PACE Research Officer in PACE T. Shirts supervising Net hole assessment in Hoima district

2.4.3 COVID 19 risk mmitigation

The LLIN Durability Monitoring study 12 months, August 2021 survey was implemented during the COVID pandemic and therefore it was very important to have prevention measures in place against COVID 19. The prevention measures used include the following:

➢ Every volunteer as well as the support staff (drivers) were given sanitizer and PACE branded masks. Sanitizer dispensers were also placed in each of the field cars for the volunteers to sanitize their hands every time they are entering the vehicles.

➢ Spacious vehicles (super customs) were used by the volunteers, and this allowed for social distancing even within the field vehicles to reduce the contact risk.

➢ The VHTs and LC Chairpersons that the volunteers interacted with for the several days were given masks which they were required to put on the entire field work exercise.

➢ During the interviews in which the volunteers interacted with in the community, social distancing was mandatory during the interviews.

➢ Before any interviews were conducted, the potential respondents were screened for COVID 19 risks. This screening was part of the data collection tool and the screening questions were the first to be asked and the responses were assessed for the magnitude of the risk and where there was high risk and likelihood of the cohort household having COVID19, then the interview for that particular household was pushed ahead to a time when the household would no longer be at risk as long as it would fall within the window period before expiry of the one month period of the nets. In cases where the

household risk of COVID was projected to be minimal at a time after one month period, then such households were simply left out.

➢ The data was collected electronically to reduce the risk of transmitting COVID 19 through many people touching the same papers.

2.4.4 Data Management

The questionnaire was thoroughly tested prior to deployment. Skip patterns and filters, internal consistency checks, range checks and logical checks were programmed to support high quality data collection. Interviewer data was uploaded to a web-based database daily or stored on tablets until they could be transferred. A one-page paper questionnaire was completed for each ITN taken for bioassay and chemical content analysis. The questionnaire was stored with the ITN for transfer to the laboratory. At the end of the survey, the web-based database was downloaded and converted into a Stata data file for analysis. Data values were checked for internal consistency and logic, and coding was applied for non-response or missing values. All operations were documented in Stata "do" files.

2.4.5 Analysis

Data analysis for the 12 months survey is ongoing and computation of the key indicators for all the study objectives will be done following the same parameters used at Baseline survey data analysis. The outcomes of measure will be the same as those for the Baseline survey.

3.0 Next Steps

The study team is currently preparing for the 24 months survey data collection while the Bioassay and chemical residue (HPLC) tests are ongoing as well at CPHL and CRAW Laboratories respectively. Upcoming activities include:

1. The 12 Months Bioassay tests and analysis is ongoing and is likely to be completed by the end of 2022.
2. The 12 Months chemical residue (HPLC) tests and analysis is ongoing and is likely to be completed by the end of 2022.
3. The 24 months survey data collection for wave 2 (Kakindo and Buheesi) is scheduled to take place in August to September 2022.
4. The 24 Months survey data collection for wave 3 (Kigorobya Town Council, Kyabigambire and Kiziranfumbi) is scheduled to take place in November to December 2022.

4.0 Conclusion

The PACE Durability study is being conducted to the expected standards, in keeping with Good Clinical Practice, and in close collaboration with the NMCD. The PACE study team and NMCD have continued to engage with both partners and stakeholders at the national and sub national levels.

The study team should prioritize the analysis and dissemination of the 12-month survey results to facilitate timely data use for decision making.