



SOP: Cutting and preserving net samples for chemical analysis

May 2022

Title	Cutting and preserving net samples for chemical analysis
Document number	I2I-SOP-005
Version number	2
Date first published	30/10/2020
Date last revised	24/05/2022

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Timeline

Version	Date	Reviewed by	Institution
1	30/10/2020	Angus Spiers	I2I
2	24/05/2022	Angus Spiers Rosemary Lees	I2I LSTM

Version Control¹

Version	Date	Updated by	Description of update(s)
2	April – May 2022	Alex Wright, Natalie Lissenden	Purpose, materials & equipment, data collection

¹ Historical versions of SOPs can be found on the I2I website (<https://innovationtoimpact.org/>)

			sheet information, glossary of terms and references added.
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Related documents

- I2I Best Practice SOP Library, 30 October 2020 (<https://innovationtoimpact.org/>)
- WHO Guidelines for laboratory and field testing of long-lasting insecticidal nets (WHO, 2013)

1. Purpose

Chemical analysis is conducted on LLINs (long-lasting insecticidal nets) in order to determine the between- and within-net variability of insecticide concentration. This SOP outlines the method to cut and preserve net samples for chemical analysis.

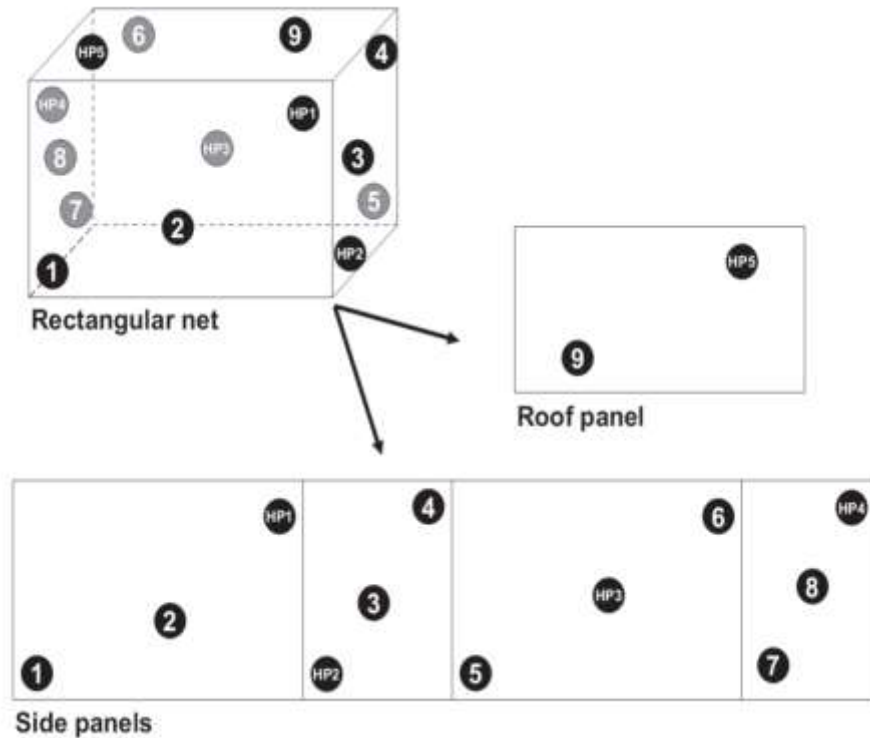
2. Background

After cutting or testing, all netting samples should be properly labelled, wrapped individually in aluminium foil and stored at 4-10°C until they can be analysed for their insecticide content to determine their wash-resistance index. The insecticide content of each net sample should be analysed to estimate between- and within-net variation, and the density of netting (i.e. mass of net per unit area) should be measured. The samples should be analysed by the methods published by the Collaborative International Pesticides Analytical Council (CIPAC) or, if those are unavailable, with tests devised by the manufacturer and validated. The results should be expressed in grams of active ingredient per kilogram as well as in milligrams of active ingredient per square metre of netting material. The decrease in insecticide content after successive washes can be used to estimate the wash-resistance index of the LLIN.

Preparation of nets for testing four candidate LLINs are required for laboratory studies, from at least two different production batches. From each net, 14 pieces (25 cm x 25 cm) are sampled, as shown in Figure 1. The tests conducted on the 56 pieces are as follows: - Eight pieces (four unwashed and four washed) are used to estimate regeneration time. - 28 net pieces are used to

evaluate wash-resistance. Four pieces are tested after 1, 3, 5, 10, 15, 20 and 25 washes (4 x 7 = 28 bioassays), although only 20 washes are considered standard procedure for determining wash-resistance. If the manufacturer's claim cites more than 20 washes, additional net pieces may be cut and used for further washing and bioassays. After bioassays, the net pieces are tested in chemical assays to determine the wash-resistance index. - 20 pieces (five pieces from four nets) are wrapped in aluminium foil and held at 4-10°C for chemical analysis in order to determine the between- and within net variability. Net pieces should be handled with care to avoid contamination or excessive abrasion. Nets should be stored wrapped in aluminium foil at 30°C between washes.

Figure 1. Sampling scheme for 14 pieces of netting from each net, including positions HP1–HP5 for chemical assay. A different sampling scheme is required for combination nets.



3. Materials & Equipment

- a. Cutting pieces from whole nets

- i. Lab coat
 - ii. Gloves
 - iii. Stencil
 - iv. 70% ethanol
 - v. Paper towel
 - vi. Marker pen
 - vii. Scissors
- b. Preserving net samples
- i. Labels with protocol number, position, date, etc
 - ii. Stapler
 - iii. Aluminum foil
 - iv. +4°C Refrigerator
 - v. Bag sealer (if available)

4. Procedure

Cutting net pieces from whole nets

- a. **Wear personal protective equipment (lab coat, gloves, respirator)**
- b. Preparation of the cardboard stencil and supplies
- i. Clean benchtop with 70% ethanol, then wipe with damp paper towel and dry with clean dry paper towel
 - ii. Draw squares (30cm x30cm) on two pieces of cardboard with a marker pen
 - iii. Cut out the squares to obtain two separate cardboard stencils of 30cm x 30cm
 - iv. Clean a pair of scissors with 70% ethanol and leave to dry on a clean benchtop
- c. Preparation of net sample
- i. Remove net from package and put on clean table
 - 1. Start with control net and move to insecticide treated net pieces
 - 2. Change gloves for nets with different insecticides

- ii. Identify areas to cut from the sides and top of the net for the chemical analysis according to the diagram below.
- iii. Measure carefully with a ruler, mark around the cutting location with a permanent marker pen Cut out the net sample carefully following the drawn line
- iv. Cut out five pieces of netting material (30cm x 30cm) from each net as shown in Figure 1 above.

Preserving net samples

- d. Prepare labels for net samples. Include the protocol number, net ID, position, date of cutting, technician initial
- e. Staple label on netting sample
- f. Wrap each sample in aluminium foil and label the foil
- g. Place all samples for each net together in one clean, labelled foil or plastic container
- h. Store at 4-10°C
- i. Return whole net to the original package, and seal using a bag sealer. Return to storage room.
- j. Send to laboratory for chemical analysis

Collection and reporting of data

Ensure the following data is recorded in the data collection sheets:

- Net piece ID information
 - Study Code
 - Date net piece put in fridge
 - Lot or batch number
 - Active ingredients
 - Fridge box ID
 - Piece ID number
 - Disposal/removal date

- Staff initials of disposal
- Net cutting record
 - Net ID number
 - Study protocol number
 - Date net received
 - Whole net tag number (if needed)
 - Position of net piece
 - Date cut
 - Initials of staff who cut net
 - Piece ID number- coded for position on whole net

5. Glossary of terms

LLIN	Long-lasting insecticidal nets
SOP	Standard Operating Procedure
WHO	World Health Organization

6. References

WHO. (2013). Guidelines for laboratory and field-testing of long-lasting insecticidal nets. In *WHO/HTM/NTD/WHOPES/20131*. World Health Organization.

