SOP: Mini chamber test

October 2026

Title	Mini chamber test
Document number	I2I-SOP-XXX
Version number	1
Date first published	27/10/2026
Date last revised	27/10/2026

Prepared by

Name	Role	Institution
Katherine Gleave	Author	LSTM
Amy Lewis	Author	LSTM

Timeline

Version	Date	Reviewed by	Institution
1	27/10/2026	Rosemary Lees	LSTM

Version Control

Version	Date	Updated by	Description of update(s)

1. Purpose

The purpose of this document is to provide step-by-step instructions for conducting experiments to measure mosquito mortality of insecticide treated nets (ITNs) using the mini chamber test. This test allows mosquitoes to come into contact with, and pass through, ITNs to feed on a meal source, in a space large enough to enable more flight than a standard World Health Organisation (WHO) cone test.

2. Materials

- Calibrated data logger
- Assay record sheets
- Gloves
- Parafilm
- Scissors (designated for LLIN use only)
- Holding cups
- Rubber bands
- Cotton wool
- Labelling tape
- Permanent pens
- 10% sugar solution
- Net samples (negative control, positive control, treatment net)
- HEPA-filtered mouth aspirators (one for each net treatment)
- 5% decon solution (for cleaning)
- Unfed female mosquiotes (3-5 days old)
- Foil to wrap nets
- Plastic pots with approximate dimensions (12 x 9.2 x 7.6 cm or 450ml/16oz internal volume)

Details about dimensions, materials and how to construct the mini chambers can be found in Appendix 1.

3. Methods

Preparation of the chambers

- When preparing the pots for the first time, cut two holes in half of the pots and one hole in the other half of the pots. These holes can be any size as they will be covered with cotton wool and are used for aspirating the mosquitoes in and out of the chambers. The second hole in half of the pots is for the sugar and should be covered with mesh.
- Cut the net pieces into 15x15cm squares with two 1cm holes in the middle (Figure 1) – if these are new nets, air for seven days before use. Make a record of the net code.
- Place the net piece in the middle of two pots and use laboratory tape to secure the pots together. Wrap the centre of the pots with Parafilm, ensuring there are no gaps for mosquitoes to escape.
- Label the chambers using tape
- Secure the chambers horizontally, either using stands or cotton wool so they won't move.

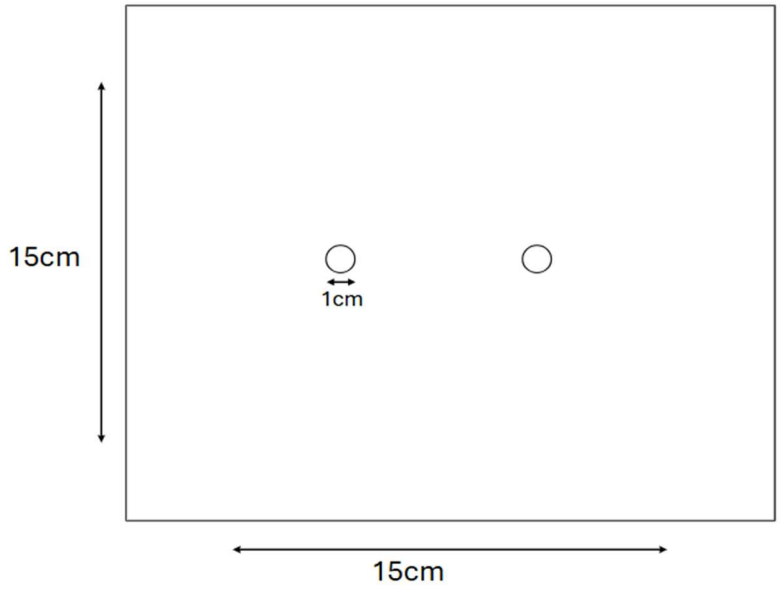


Figure 1. Size of the net pieces and holes in the net pieces used in the mini chamber test.

Performing the assay

- Prepare cups of 10 unfed females – one cup of 10 mosquitoes per chamber. Acclimitise the mosquitoes for 1 hour in the testing room.
- Aspirate the mosquitoes into compartment 1 of the chambers through one of the pre-cut holes and plug the hole with cotton wool.
- Secure cotton wool soaked in 10% sugar solution over the mesh on compartment 2 using tape or parafilm.
- Allow the assay to run for six hours.
- At the end of the exposure time, aspirate the mosquitoes out of the chambers and into holding cups. Label the cups with date of exposure, mosquito strain and net type.
- Add cotton wool soaked in 10% sugar solution to each of the cups. Replace the sugar every 24 hours for 72 hours.
- Record knock down 1 hour after exposure and mortality at 24, 48 and 72 hours post exposure.

Equipment cleaning

- Remove the parafilm, tape, cotton wool, mesh and net pieces.
- Wrap the net pieces in foil and store appropriately.
- Soak the chambers in 5% Decon for two hours.
- Rinse the chambers twice using tap water and a final third time using distilled water. Leave the chambers to air dry.
- Repeat this process at the end of every test week or when testing with new net pieces.

Data sheet

Mini Chambers Test Record Sheet

Test Date:				Test Net Type:			
Colony (Species and Generation):		Mosquito Age (days):					
Mosquitoes aspirated by (initials):							
Test Replicate:		Number of Mosquitoes Per Test		Length of Test:		**Test Start Time**	**Test End Time**

Temp and Humidity Instrument Number **Tolerance range= Set temperature at 26°C +/- 2°C and Humidity at 70% +/- 10%.**	**Exposure start**		**Exposure end**		**24 hours**		**48 hours**		**72 hours**	
	Temp (°C)	Humidity (%RH)	Temp (°C)	Humidity (%RH)	Temp (°C)	Humidity (%RH)	Temp (°C)	Humidity (%RH)	Temp (°C)	Humidity (%RH)

Test Observations:

Cup I. D	Net Type	KD/Dead After Exposure	KD/Dead 24 hours	KD/Dead 48 hours	KD/Dead 72 hours	Total Mosquitoes at Test End

Cup I. D	**Net Type**	**KD/Dead After Exposure**	**KD/Dead 24 hours**	**KD/Dead 48 hours**	**KD/Dead 72 hours**	**Total Mosquitoes at Test End**

4. References

Lewis, A., Jones-Brown, R., Harvey, G., Praulins, G., Reid, E., Lugenge, A. G., Mechan, F., Gleave, K., & Lees, R. S. (2026). The mini chamber test: a novel bioassay for bioefficacy testing of chlorfenapyr treated nets used in malaria control. *Gates Open Res, 10*, 14. https://doi.org/10.12688/gatesopenres.16381.1