



The Gnatwork

Intended use of resource / data

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Title of resource

GN_20: Protocol for use of sugar bait FTA card stations

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DOI

Description

Protocol for use of sugar bait FTA card stations. Describes sugar bait FTA card station design created at LSHTM, and how to use these stations with a potential to monitor leishmaniasis transmission in an area.

Protocol from the Gnatwork project “The use of FTA cards to monitor *Leishmania* infection and infectiousness in sand flies and midges” created for field work in Ethiopia and Ghana (summer 2019).

Intended use

Scientific research use and training purposes.

Restrictions on use

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Resource history

N/A

GN_20: Protocol for use of sugar bait FTA card stations

When using this protocol, the following should be referenced:

Mojca Kristan, Tom Walker, Matt Rogers (London School of Hygiene and Tropical Medicine)

A. Introduction

Male and female sand flies and midges require sugar for their energetic requirements. They can use a variety of different sugar sources including nectar and fermenting fruit. These FTA card bait stations are based on research showing that various mosquito and sand fly species are attracted to and will feed from sugar-baited traps [1-4], and recent findings demonstrating that infectious mosquitoes excrete detectable malaria sporozoites onto FTA cards during sugar feeding [5].

The sugar bait FTA card stations were designed and made at London School of Hygiene and Tropical Medicine (LSHTM).

B. Materials

Trap components

Each trap is comprised of:

- A large black Perspex disc with string attachments for hanging (protective shield against rain)
- A smaller transparent Perspex disk with string attachments for hanging (this will hold the four tubes)
- Four tubes with hole in orange lid (we used 50 ml conical-bottom centrifuge tubes)



Other equipment

- FTA classic cards (Whatman® FTA® card technology, FTA classic card with 4 sample areas per card, Sigma-Aldrich WHAWB120205)
- Absorbent cotton wool
- Sealable plastic bags

Reagents

- Sucrose powder (Sucrose for microbiology, ACS reagent, >99.0%, Sigma-Aldrich 84100)

C. Method

C.1 Cut out each individual FTA card (there are four together). The correct “active” side is the one with black circle drawn, so make sure to leave this circle marking when you cut it out. This is the side on which the insects will feed. Use gloves and clean scissors.

C.2 Place cotton wool into each tube and make sure it fills the tube completely. Then thoroughly wet (fill) with ~50% sucrose solution. Cotton wool should reach the top of the tube. This will ensure that cotton wool and FTA card remain wet.

C.3 With clean forceps or gloves place one FTA card circle on top of the cotton wool and ensure it is wet with the sugar solution and secure at the top of the tube. Screw on the orange “lid” to the tube.



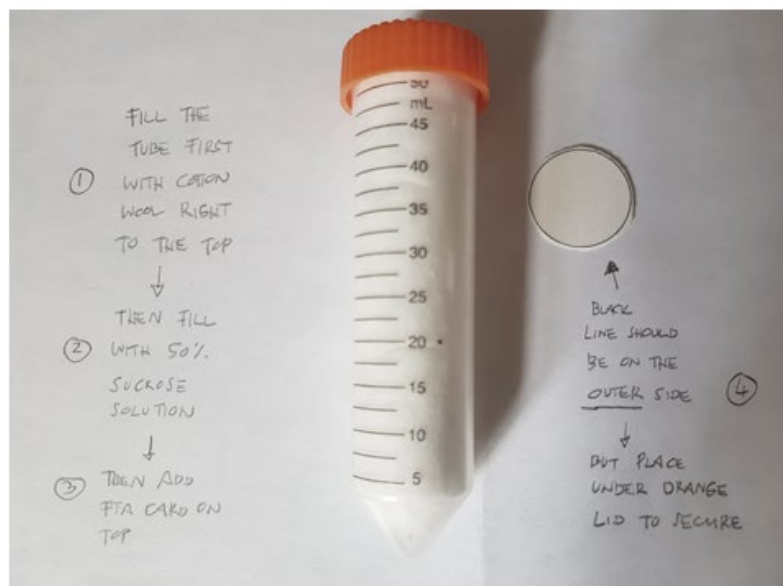
Cut out each FTA card circle but make sure the black edge remains visible – this is the

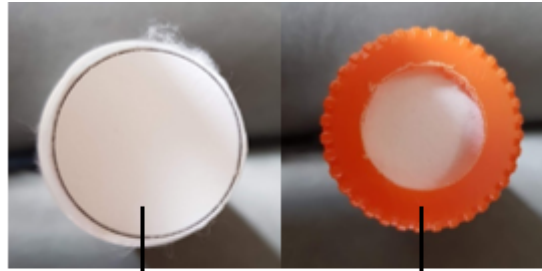
FTA card pack

FTA card pack (open) showing 4 FTAs



Materials to assemble the tubes





Fill the tube with cotton wool and place FTA card on top – active side needs to be facing outwards!

Secure FTA card with a lid

- C.4 Hang the sugar bait trap in a secure location outdoors – or where you know that sand flies or midges can be found, ideally in a sheltered area. If possible, choose an area where *Leishmania*-positive sand flies (or midges) were caught previously or where there are many people with leishmaniasis.
- C.5 Each day check the FTA cards are still wet with sugar solution and top up as necessary.
- C.6 Collect and replace the FTA cards every 2-3 days.
- C.7 Remove FTA cards from plastic pouches with clean forceps. Clean forceps by wiping with 70% (v/v) ethanol.
- C.8 Allow the FTA cards to dry completely at room temperature on a clean surface and store each one individually in a small sealable plastic bag. Label it with the date of collection and location details.

Place used dried FTA card into a small sealable bag, close it and label properly.



- C.9 Redeploy the bait station with fresh cotton wool, sugar, and FTA cards.

Next steps

- C.10 Extract gDNA or RNA from collected FTA cards.
- C.11 If extracting RNA, generate cDNA.
- C.12 Use qRT-PCR for detection of *Leishmania* parasites.

D. References

1. Qualls, W.A. *et al.* (2015). Control of sand flies with attractive toxic sugar baits (ATSB) and potential impact on non-target organisms in Morocco. *Parasit Vectors*, 8: 87.
2. Beier, J.C. *et al.* (2012). Attractive toxic sugar bait (ATSB) methods decimate populations of *Anopheles malaria* vectors in arid environments regardless of the local availability of favoured sugar-source blossoms. *Malar J*, 11: p. 31.
3. Muller, G.C. and Y. Schlein (2011). Different methods of using attractive sugar baits (ATSB) for the control of *Phlebotomus papatasi*. *J Vector Ecol*, 36 (Suppl 1): S64-70.
4. Muller, G.C. and Y. Schlein (2008). Efficacy of toxic sugar baits against adult cistern-dwelling *Anopheles claviger*. *Trans R Soc Trop Med Hyg*, 102(5): 480-4.
5. Brugman, V.A. *et al.* (2018). Detection of malaria sporozoites expelled during mosquito sugar feeding. *Sci Rep*, 8(1): 7545.