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## CARD COUNT AND VIAL DEPLOYMENT/RETRIEVAL PROTOCOL

Forked from [CARD COUNT PROTOCOL](#)

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### ABSTRACT

Procedure to conduct "Crazy Yellow Ant Sampling on Tetiaroa"

This protocol provides instructions for performing card counts and deploying/retrieving vials in order to quantify the prevalence of yellow crazy ants, *Anoplolepis gracilipes*, in given transects around Onetahi, Tetiaroa using two kinds of ant baits—sugar water and peanut butter. In addition, the experiment tested yellow crazy ant preference for sugar water bait versus the peanut butter bait.

### GUIDELINES

- Do not trespass
- Analyze site safety before conducting a sample
- Avoid trampling flora
- If a point is inside a building, just go to the next point or as close to the point as possible
- If it starts raining, stop deploying vials and card counts

### MATERIALS

White flashcard (10cm x 10cm)  
Stopwatch  
EpiCollect5 mobile app  
Gaia mobile app  
Peanut butter vials  
Sugar water vials

### SAFETY WARNINGS

- ! Wear long pants and closed toed shoes—dense underbrush

### ETHICS STATEMENT

Unfortunately ants were harmed in the process.

### BEFORE START INSTRUCTIONS

- Recommend wearing long sleeves, pants, a sun hat, and closed toed shoes
- Recommend putting on sunscreen and bug spray

## Card Count Protocol

- 1 If needed, scrape the ground (at least a 10x10cm area) at the site so the card sits flat on the ground (i.e. if there are branches or leaf litter, scrape them away).
- 2 If you scraped the ground, wait 30 seconds before placing the card down. If you did not scrape the ground, place the card down without waiting.
- 3 Place the card down on the ground.
- 4 For 30 seconds, count the number of yellow crazy ants that cross the card.

- 5 After 30 seconds have passed, pick up the card and remove all ants from it.
- 6 Enter the number of ants counted in the card count into the appropriate field of the EpiCollect5 form.

## Vial Protocol

- 7 Preparation: Divide the atoll into a series of transects labeled A-Z. Then on each transect create points 1-X. The points should create a grid on the island.

- 7.1 Prepare 25 sugar plastic vials by soaking a napkin in sugar water and placing at the bottom of the vial

- 7.2 Prepare 12 peanut butter (PB) plastic vials by placing 1 TBS of peanut butter at the bottom of each vial, making sure not to get any PB on the sides of the vial.

- 8 Deployment: Navigate to the site using Gaia GPS

- 8.1 At each site, place one sugar vial. Place a peanut butter vial down directly next to the sugar water vial only if ants are spotted in the nearby surrounding area.

### Note

In the future, we would modify this procedure to deploy PB vials at every other site, not only when we saw ants surrounding, in order to standardize the procedure.

- 8.2 Fill out EpiCollect form detailing: deployed/retrieved, site number, and time collected (automatic)

- 8.3 Repeat steps 3-5 at each site. Go in reverse order in the following section for retrieval.

- 9 Retrieval: After 90 minutes, return to placed vials.

- 9.1 Pick up and quickly cap vials, not letting any ants escape in the process.

- 9.2 Fill out form on EpiCollect5. Count crazy ants on each vial. If there are too many ants, head back to lab. Freeze vials and count ants afterward.

- 10 Cleanup: Clean out test tubes and throw away samples.

