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Larval Feeding

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Protocol status: Working
We use this protocol and it's working. This protocol is current as of 19 February 2025 and will be updated as needed.

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Abstract

Larval feeding protocol developed by the Applied Science and Technologies department of Lee County Mosquito Control District for the purpose of mass rearing *Aedes aegypti* larvae for the sterile insect technique program.

Guidelines

Larval feeding and food preparation protocol for Aedes aegypti reared in Wolbaki larval rearing systems. (Updated 19 Feb 2025)

Materials

Feed Inspection

- 600 micron sieve
- 5 gallon buckets with lids, or other suitable container
- Zeigler Tropical Pond Meal, 50lb bag

Larval Feeding

- Fish feed powder (Ziegler Tropical Pond Meal 33%), pre-sifted
- 16 oz deli cups, or other suitable container
- Measuring scoop set
- Spray bottle
- Filtered water, aged tap water, or RO water

Safety warnings

Do not use feed if spoilage or contamination is suspected. First consult AST supervisor. If larvae are not behaving as expected or there are concerns for water quality, consult AST supervisor for instructions on how to proceed.

Feed Inspection

1 Inspect the bag and label to ensure that the correct product has been delivered. Inspect the bag for damage in the form of tears and leaks. If the bag is intact, the bag can be stored in the feed room. It does not require refrigeration, provided it is used within six months and kept in a cool and dry location.

2 Upon opening the bag, note any off odors or changes in color and texture relative to previous batches. The material is a fine powder with a light brown color and an odor similar to wheat. Report any suspected changes immediately and before using it for larval feeding.

2.1 When the last remaining bag has been opened, report it to the supervisor so that additional bags can be reordered immediately.

3 Obtain a 600 micron sieve to remove large particles from the feed. Sieving to 600 microns removes these larger particles and has helped to maintain consistency in larval rearing. Sieve until no fine particles are seen falling (~5 minutes).

4 Aliquot fine, sieved particles into smaller batches for ease of use. Store in the feed room when not being used. Discard any aliquots if you suspect the quality of the feed has changed, or if you notice any change in larval behavior and mortality.

Larval Feeding

5 Household measuring spoons (Imperial volumes) are used to deliver the larval feed. Refer to Table 1 for the weights associated with the different spoon volumes.

	Volume (Imperial)	Volume (ml)	Weight (g)
	1/8 tsp	0.63	0.33
	1/4 tsp	1.25	0.75
	1/2 tsp	2.5	1.5
	3/4 tsp	3.75	2.1
	1 tsp	5	3
	1 tbsp	15	8

Table 1: *Measuring spoon volume and approximate weights of leveled feed in each spoon.*

6 Observe water quality and general larval health of a few trays. If there is an excess of feed remaining, abnormal larval behavior (reduced negative phototaxis, margin crowding, abnormal movements, etc.), instar asynchrony (3 or more instars), or excessive mortality, then delay feeding and/or reduce the amount of feed delivered. Also note the presence of surface scum, foul odors, or the presence of calliphorid and phorid flies.

6.1 After observing the trays and noting any changes in water quality, push the trays back into place. Make note of any trays that will receive a reduced amount of feed, if any.

7 Determine the amount of food needed depending on rearing density (10,000/tray for release batch, 5,000/tray for colony) and day of rearing. Use table 2 for colony (5,000/tray) and table 3 for release (10,000/tray).

	Food weight (g)	Volume (spoon)	Food (mg)/larva
	0.75	1/4 tsp	0.15
	0.75	1/4 tsp	0.15
	1.5	1/2 tsp	0.3
	3	1 tsp	0.6
	6	2 tsp	1.2
	6	2 tsp	1.2
	0	0	0

Table 2: *Feeding weights (g), volumes, and food (mg) per larva for colony batches at a density of 5,000 larvae per tray. Note: Day 1 represents the day of hatching with vacuum. No feeding occurs on Day 7 as the trays will be drained. No feeding occurs after draining.*

	Food weight (g)	Volume (spoon)	Food (mg)/larva
	1.5	1/2 tsp	0.15
	1.5	1/2 tsp	0.15
	3	1 tsp	0.3
	6	2 tsp	0.6
	8	1 tbsp	0.8
	8	1 tbsp	0.8

	Food weight (g)	Volume (spoon)	Food (mg)/larva
	0	0	0

Table 3: *Feeding weights (g), volumes, and food (mg) per larva for colony batches at a density of 10,000 larvae per tray. Note: Day 1 represents the day of hatching with vacuum. No feeding occurs on Day 7 as the trays will be drained. No feeding occurs after draining.*

8 Once the amount of feed per tray has been determined, obtain an aliquot of pre-sifted feed and select the proper measuring spoon (or spoons) needed to deliver the material.

9 Starting at the bottom of the rack, pull the lowermost tray out approximately 6-8″ and evenly dispense the feed onto the water surface using the necessary measuring spoon. Once completed, continue to move upwards along the rack, pulling out trays and dispensing feed into each.

10 Obtain a spray bottle filled with water. Gently spray the tray with water to sink and disperse the feed as well as to break up any large chunks of feed. Once sprayed, gently push the tray back into position and begin spraying the tray below. Work your way down the rack system until all trays have been sprayed and the feed adequately dispersed.

10.1 If excessive water loss due to evaporation has been observed, top off water levels in trays as needed.

11 Once manual feeding is complete, pour out any remaining water in the spray bottle and let it dry. Clean the measuring spoons and store any unused aliquots of food in the feed room.