



The Use of Parasitoid Wasps (*Leptopilina heterotoma*) in Biology Laboratory Courses

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Abstract

The parasitoid wasp, *Leptopilina heterotoma*, infects the second instar stage larvae of the fruit fly, *Drosophila melanogaster*. This process can be used to enhance interest of students in parasitology, genetics, and developmental biology courses. Students can conduct a variety of studies, including determining the efficacy of infectivity of various mutants of fruit flies. First, students subculture various mutants of fruit flies and wait until second instar larvae have developed. They then remove the adult flies and coat the inside of the plug with honey, which will serve as the food source for the wasps. They then anaesthetize the wasps and add them to the vials with the fruit fly larvae. They can then count surviving adult fruit flies after the flies should have completed their life cycle in both infected and uninfected flies. The wasps that infect the larvae take twenty-eight days to hatch and produce a new source of wasps. Other species of these non-stinging parasitoid wasps have also been sold commercially for pest control. Pros and cons of using parasitoid wasps in this manner can be added to round out the laboratory experience for the student.

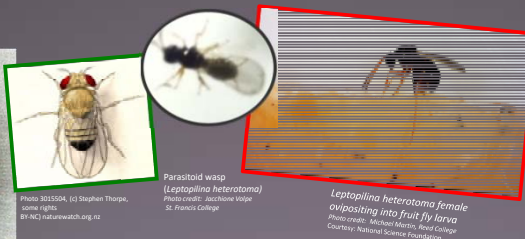


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Parasitoid wasp (*Leptopilina heterotoma*) Photo credit: Jonathan Vogel, St. Francis College

Leptopilina heterotoma female ovipositing into fruit fly larva Photo credit: Michael Senter, Reed College Courtesy: National Science Foundation



Orcon TR-C35Q Live Trichogramma, 3 Squares/12,000 Eggs

Materials and Methods

We were given two vials of flies from the Govind lab at the City College of New York. One contained fruit flies alone (*Drosophila melanogaster*) and the other contained fruit flies that had had parasitoid wasps (*Leptopilina heterotoma*) introduced. Many had infected the fruit fly larvae. The evidence was that the pupae of the infected fly larvae looked much darker than uninfected ones. After 28 days, many wasps had hatched out of the fruit fly pupae. The students in the Parasitology course observed the flies and wasps and learned about their life cycles and relationships. New vials of fruit flies were cultured, and were reared till larvae appeared (about a week). Adults were cleared by two different methods—drowning and pouring off or by fly nap and clearing. Students then introduced fly napped wasps to the vials. We did not sex them (the females have longer antennae and a more pointed abdomen). After a month, the vials were observed again. The vials that had more than seven wasps introduced produced new wasps, and the fly napped and cleared adults yielded more results than the vials in which the adults were drowned.

A group from the Developmental Biology course took it a step further. Four groups of students participated in this project. *Drosophila* were separated according to first and third instar larval stages, and placed in different vials with food cultures. Two adults wasps were inserted into each vial with the exception of Group 1's Vial 1 which was infected with the pupa that were suspected to be infected by wasps. In order to provide a food source for the wasps, the foam stopper was dipped into honey. The larvae were observed weekly and observations were recorded. The number of infected larva were counted in each vial and data was recorded.

Results

Vials that contained *Drosophila* that had first instar larvae were successfully infected, whereas those with third instar larvae were not. The literature has indicated that *Leptopilina heterotoma* successfully infect fruit fly larvae at the second instar stage. The students results were consistent with this. The third instar stage is probably too developed for the wasp to infect.

	Group 1 First instar	Group 2 Third instar	Group 3 Third instar	Group 4 First instar
Week 1	<ul style="list-style-type: none"> Vial 1 Infected <i>Drosophila</i> pupa contained 10 wasps Vial 2 did not have any wasps, only <i>Drosophila</i> were observed. 	<ul style="list-style-type: none"> Vial 3 contained third instar larva <i>Drosophila</i>; after infection with wasps there were not visible wasps Vial 4 did not have any observable wasps, only <i>Drosophila</i> were present. 	Vial 5 Same Observations as Group 2	Vial 7 There were wasps in the vial and most <i>Drosophila</i> appeared as dark colored pupa (a mix of larvae were present)
Week 2	<ul style="list-style-type: none"> Vial 1—Increase in the number of wasps, approximately 50 were observed Vial 2—there were no wasps, only <i>Drosophila</i> were seen. 	<ul style="list-style-type: none"> Vial 3 did not present any visible wasps Vial 4 did not appear to have any wasps, only <i>Drosophila</i> were observed. 	Vial 6 Same Observations as group 2	Vial 7 There were 15 visible wasps and 30 <i>Drosophila</i> observed

Discussion

The students in the Parasitology and Developmental Biology courses at St. Francis College were introduced to the system of parasitoid wasp—fruit fly infection successfully. The animals are easy to grow and manipulate and tangible results were obtained after two distinct experiments. We suggest that further experiments such as infecting different fly mutants or species of flies with *Leptopilina* spp., or other species be conducted under various conditions. Todd Schlenke at Reed College maintains a variety of parasitoid wasps, and, according to his web site, “We maintain a large number of live strains of parasitoid wasps that infect *Drosophila*. Our lab policy is to make these strains available to everyone once we have published on them.” (schlenke@reed.edu)

Selected references

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Introduction

There are many species of parasitoid wasps that infect various life stages of insects, especially eggs, larvae and pupae. The endoparasitoid females use ovipositors to inject their eggs in these life stages. Because most of these species are non-stinging to humans, they have exploited in biological control. For example, they have been recently released in Minnesota to try to combat the emerald ash borer. These species (*Muscidifurax raptorellus*, *Muscidifurax zaraptor* and *Spalangia cameroni*) are used against horse and house flies, and the tiny *Trichogramma* spp. are commercially available to assay over 200 species of garden and crop pests. We were recently made aware of species that parasitize the easily laboratory reared *Drosophila melanogaster*. These wasps include: *Leptopilina*, *Ganaspis*, *Asobara*, *Aphaereta* *Trichopria* and *Pachycrepoideus* spp. Infecting fruit flies with a parasitoid wasp can be a model to teach students about a variety of topics including ecology, life cycles, behavior, life cycles, life history strategies, mechanisms of parasitism, and developmental stages. The flies and wasps are very easy to raise and lend themselves to investigative student-driven projects. We expand upon the use of the parasitoid wasps for the first time at St. Francis College in our new Parasitology and Developmental Biology courses. We welcome any suggestions and ideas for further projects.