EFFECTS OF *MECISTOGASTER* SPP. (ODONATA: PSEUDOSTIGMATIDAE) AND *CULEX MOLLIS* (DIPTERA: CULICIDAE) ON LITTER DECOMPOSITION IN NEOTROPICAL TREEHOLE MICROCOSMS

Stephen P. Yanoviak
Department of Zoology, University of Oklahoma, Norman, Oklahoma 73019

Current address: Evergreen State College, Lab I, Olympia, Washington 98505

Abstract

I investigated the effects of a top predator, *Mecistogaster* spp. (Odonata: Pseudostigmatidae), on survivorship of the grazer *Culex mollis* (Diptera: Culicidae) and decomposition rates of leaf litter in treehole microcosms. In a factorial experiment using 200 ml cups, less litter mass remained when grazers (51%) and grazers plus predators (51%) were present, than without grazers (57%). Predators reduced mosquito survival, but had no indirect effect on litter decomposition rate. Mosquito larvae facilitated decomposition of litter and may have become food limited.

Key Words: damselflies, detritus, indirect effects, mosquitoes, predation

Resumen

Se investigaron los efectos del depredador *Mecistogaster spp*. (Odonata: Pseudostigmatidae) en la supervivencia del pacero *Culex mollis* (Diptera: Culicidae) y las tasas de descomposición de hojarasca en el microcosmos de huecos de árboles. El experimento se efectuó en vasos de 200 ml, utilizando un diseño factorial. La cantidad de hojarasca fue menor (51%) cuando los paceros y los grazer mas depredadores (51%) estuvieron presentes, que cuando no hubo paceros. La presencia de paceros redujo la sobrevivencia de mosquitos, pero no tuvo un efecto indirecto sobre la tasa de descomposición de la hojarasca. Las larvas de mosquito facilitaron la descomposición de la hojarasca y probablemente estuvieron limitados de alimento.

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Trophic cascades (i.e., indirect effects of predators on productivity at lower trophic levels) occur in large aquatic systems (e.g., Carpenter et al. 1985, Power 1990) and some terrestrial systems (e.g., Spiller & Schoener 1994, Moran & Hurd 1998). Fewer detailed investigations of trophic interactions have been conducted in small aquatic settings (but see e.g., Addicott 1974, Bradshaw & Holzapfel 1983, Cochran-Stafira & von Ende 1998, Srivastava & Lawton 1998).

Water-filled treeholes are formed when rain collects in branch crotches and other wood-lined cavities, and are common aquatic habitats in temperate hardwood and tropical forests (e.g., Kitching 1971). Unlike large aquatic systems such as lakes and streams, treeholes are experimentally tractable and can be reproduced as laboratory microcosms without compromising the scale of the natural system.

Detritus, especially leaf litter, is the base of food webs in most treeholes (Kitching 1971). Leaf litter in treeholes is primarily consumed by bacteria and fungi (Fish & Carpenter 1982), and mosquito larvae are typically the most common grazers on this

microfauna. In the lowland moist forest of Barro Colorado Island (BCI), Panama, larvae of *Aedes*, *Culex* and *Haemagogus* mosquitoes are the principal prey of top predators in treeholes, which include odonate larvae, *Dendrobates* tadpoles, and *Toxorhynchites* mosquito larvae (Fincke et al. 1997, Fincke 1999, Yanoviak 1999). Larvae of pseudostigmatid damselflies are the most common top predators in BCI treeholes (Fincke 1992a, 1999).

Carpenter (1982) and Fish & Carpenter (1982) showed that consumption of decomposer microbes by *Aedes triseriatus* (Say) larvae enhances litter degradation rates in treehole microcosms. Similar facilitation effects of grazers occur in other systems (e.g., Hargrave 1970, Barsdate et al. 1974), and several mechanisms have been suggested: 1) grazers fragment or directly consume leaf material in addition to the decomposer fauna (e.g., Fenchel 1970, Carpenter 1982); 2) grazing stimulates decomposer metabolism (e.g, Lopez et al. 1977); 3) grazing alters the composition of decomposer taxa (e.g, Harrison 1977, Hanlon & Anderson 1979); and 4) inorganic compounds excreted by grazers enhance growth of decomposers (e.g., Durbin et al. 1979).

Tropical treeholes support diverse predator assemblages (Kitching 1990, Fincke 1999) which can reduce mosquito survivorship (Fincke et al. 1997, Nannini & Juliano 1998), and influence life history characteristics such as development time and adult mass (Lounibos et al. 1993, Grill & Juliano 1996). These strong predator effects, in combination with mosquito effects on litter decomposition, suggest that treehole communities are prone to trophic cascades.

The purpose of this study was to determine the independent and interactive effects of a top predator and grazers on litter decomposition rates in treehole microcosms on BCI. I hypothesized that the consumption rate of leaf litter by decomposers would differ in simple food webs of different length (i.e., in the presence or absence of grazing mosquitoes, and with and without a predacious odonate). I tested the following predictions: 1) grazing by larvae of the treehole mosquito *Culex mollis* Dyar and Knab enhances leaf litter degradation; 2) predation by odonates reduces the survivorship (= successful pupation) and time to pupation of mosquitoes; and 3) the presence of an odonate indirectly reduces the degradation rate of leaf litter.

MATERIALS AND METHODS

Experiments were conducted in the laboratory building of the Smithsonian Tropical Research Institute on BCI during January and February 1998. Treehole microcosms consisted of plastic cups (8.5 cm height × 5 cm diameter) containing 125 ml filtered rain water. Treehole water was collected and pooled from 5 natural holes on BCI, and 3 ml was added to each cup as a microfauna inoculum. A strip of balsa wood (1 cm × 10 cm × 0.15 cm) served as a predator perch site and was included in all treatments. I added 200 ± 0.5 mg dried (≥ 10 d in air, then 48 h at 70°C) *Platypodium elegans* Vogel leaf litter as a nutrient source. I used leaflets of *P. elegans* because they are small and are often found in natural treeholes (pers. obs.). Most of the litter in each cup consisted of whole leaflets (each ≈ 15 cm²) collected from recent branch falls, but every cup also contained leaf fragments (each ≈ 1.0 cm²) to standardize the initial litter mass. I allowed the cup contents to stand for 48 hours prior to the start of the experiment to permit growth of bacteria and fungi. I maintained all cups at 23°C and periodically added rain water to compensate for evaporation.

The experimental design was a 2 × 2 factorial replicated six times, with presence/absence of predators and grazers as main effects. Treatment 1 contained odonates (predators) only, Treatment 2 contained mosquito larvae (grazers) only, and Treatment 3 contained both. Control cups lacked grazers and predators. Each cup in Treatments 1 and 3 received one medium size (15-18 mm) larva of the pseudostigmatid

damselfly *Mecistogaster* sp. The two species of *Mecistogaster* on BCI, *M. linearis* Fab. and *M. ornata* Rambur, are difficult to distinguish as larvae and were not differentiated in this experiment. Both species have similar foraging behavior and maximum growth rates (Fincke 1992b). I obtained odonate larvae from different natural treeholes and starved them for > 24 h before the start of the experiment. I biased the predators used in Treatment 3 toward smaller initial sizes (15-17 mm) because maximal growth and foraging rates of *Mecistogaster* tend to occur around that body length (Fincke 1992b, Fincke et al. 1997).

I used larvae of *C. mollis* as grazers in the experiment. This species is relatively common in natural treeholes on BCI (Fincke et al. 1997, Yanoviak 1999), and its egg rafts are easy to recognize and collect. Treatments 2 and 3 each received 25 neonate (< 12 h post hatch) *C. mollis* larvae on experiment days 0, 2, 4, and 6 (total = 100 larvae per cup). Mosquitoes added to a given treatment on a given day were reared from at least two different field-collected egg rafts (n > 16 rafts). Leaf litter and odonate densities were within the ranges observed for natural treeholes on BCI (pers. obs.). Although total mosquito density was biased toward the maximum found in small (< 500 ml) holes, the gradual addition of neonates over several days approximated mosquito recruitment patterns in natural and artificial holes augmented with fresh litter. Mosquitoes consumed by odonates in Treatment 3 were not replaced with new individuals.

I operationally defined mosquito survivorship as successful transformation to the pupa stage. I collected pupae from the cups daily and stored them in a drying oven (70°C) until the end of the experiment, when all pupae accumulated within a replication were weighed to the nearest 0.1 mg. Pupae were not separated by sex. Fresh weights (± 0.1 mg) and body lengths (± 0.25 mm, excluding caudal lamellae) of *Mecistogaster* larvae were recorded at the start and end of the experiment. I blotted the odonate larvae dry with a cotton cloth before all measurements. Leaf litter remaining in the cups at the end of the experiment was collected with forceps, rinsed in distilled water, dried (70°C, 48 h), and weighed to the nearest 0.1 mg. I terminated the experiment 31 days after the first mosquito input, when all larvae in Treatment 3 (predator plus grazers) pupated or were consumed.

Proportional data were arcsine-square root transformed (Sokal & Rohlf 1981) and homogeneity of variances was determined with F-tests prior to analyses. Odonate fresh weight and body length were highly correlated (N = 12, r = 0.99, P < 0.001), so only the change in weight was analyzed. One odonate larva in Treatment 3 molted during the experiment and was excluded from predator size comparisons. Means for the total number of *C. mollis* pupae, individual pupal mass (obtained by division), and days to appearance of the first pupa were compared across grazer and grazer plus predator treatments by ANOVA with Bonferroni adjustment for multiplicity.

<center>RESULTS</center>

The amount of leaf litter remaining at the end of the experiment was greater in the absence of mosquitoes, but there was no significant predator effect or grazer*predator interaction (Fig. 1). The average number of mosquitoes that successfully pupated was greater in the absence of predators (Table 1), but survival was low in all treatments; no more than 20 of the 100 mosquitoes in a replicate survived to pupation. Average individual pupal mass and the mean number of days to appearance of the first pupa did not differ between the two treatments containing mosquitoes (Table 1).

The foraging behavior of mosquito larvae surviving to the last week of the experiment differed from larvae that successfully pupated in the second week. Older larvae spent more time actively browsing leaf material and browsed a larger leaf area. These behavioral shifts were not anticipated and therefore not quantified.
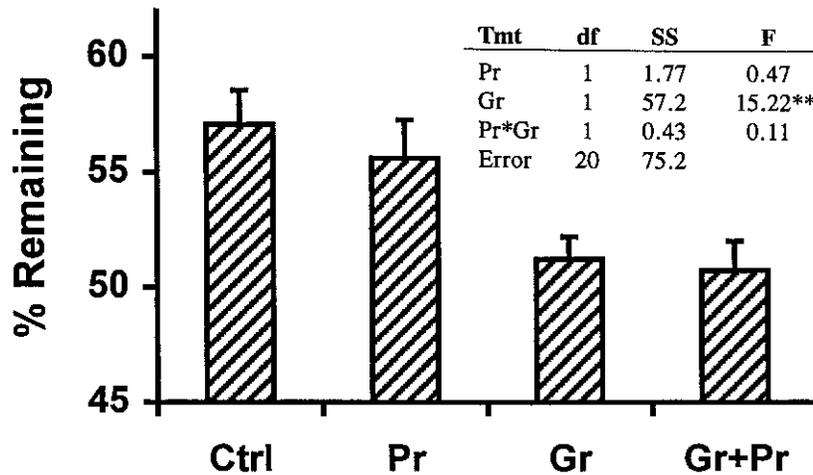
Fig. 1. Mean (+ SE) percent of litter mass remaining in each treatment (Tmt) at the end of the experiment. Ctrl = control, Pr = predator only, Gr = grazers only, Gr + Pr = grazers plus predators. ** = P < 0.05.

The mass of odonates in Treatment 1 (predators only), did not differ between the start and end of the experiment (paired t = 0.074, df = 5, P = 0.94). However, odonate mass in Treatment 3 (grazers plus predators) increased over the 31 days (paired t = 5.98, df = 4, P = 0.001; Fig. 2).

Treatments containing mosquitoes had comparatively clear water and no apparent microbial biofilm present on the leaf litter. In contrast, the water in the control cups and predator-only treatment was cloudy (presumed due to abundant bacteria) throughout the experiment and the leaves were covered with a conspicuous accumulation of decomposers (e.g., fungal strands), especially during the latter three weeks.

Discussion

The combination of species used in this study did not result in a true 3 trophic level system due to selective feeding by the odonates (= predator inefficiency; Fretwell 1977,

TABLE 1. COMPARISON OF MEAN (± SE) MOSQUITO VARIABLES AMONG TREATMENTS 2 (GRAZERS ONLY) AND 3 (GRAZERS PLUS PREDATOR). N = 6 FOR EACH MEAN; BONFERRONI ADJUSTED α = 0.0167.

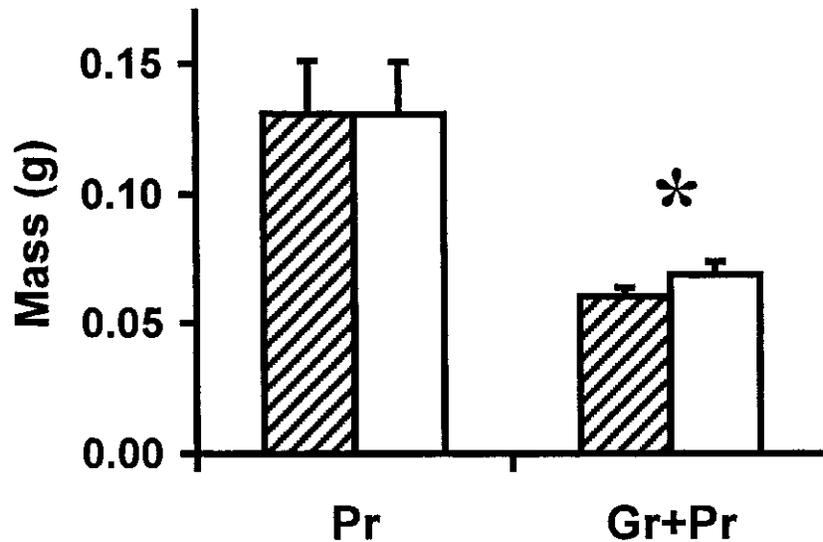| Variable	Grazers only	Grazers + Predator	$F_{1,10}$	P
Days to first pupa	9.17 ± 1.72	9.33 ± 0.82	0.05	0.83
Cumulative number of pupae	10.5 ± 2.95	3.33 ± 1.75	26.19	<0.001
Individual pupa mass (mg)	0.323 ± 0.068	0.251 ± 0.097	2.18	0.17

Fig. 2. Mean (+ SE) fresh mass of odonates at the start (hatched bars) and end (open bars) of the experiment in predator only (Pr) and grazer plus predator (Gr + Pr) treatments. N = 6 for Pr and N = 5 for Pr + Gr. * = P < 0.05.

1987). *Mecistogaster* depressed survivorship of *C. mollis*, but these odonates (and the other common pseudostigmatid on BCI, *Megaloprepus coerulatus* [Drury]) are size-selective predators, preferentially feeding on the largest prey available (Yanoviak 1999). This size-based refugium allows smaller (i.e., early instar) mosquito larvae to graze on and limit the growth of decomposers despite the presence of a predator. The lack of a significant predator*grazer interaction and the similarity in remaining litter biomass between the two treatments containing mosquitoes support this conclusion.

Although no trophic cascade effect occurred, my results are similar to those of Power (1990), who found that one of the top predators in her system did not control the dominant grazers due to the presence of refugia (Power et al. 1992). Other predators occurring in BCI treeholes, such as larvae of the aeshnid dragonflies *Gynacantha membranalis* Karsch and *Triacanthagyna dentata* Geijskes feed more voraciously and less selectively than pseudostigmatids (Yanoviak 1999). A duplicate experiment conducted with one of these species may yield a cascade effect.

Results of this study support the prediction that grazing by mosquito larvae facilitates the degradation of leaf litter in treehole microcosms. Although direct consumption of leaf material (e.g., Carpenter 1982) is the simplest explanation for this effect, there was no evidence (e.g., leaf fragmentation or abrasion) that *C. mollis* larvae consumed a measurable portion of the litter in this experiment. Alternative explanations for facilitation of litter decay cannot be addressed with data gathered in this study, but there is circumstantial evidence that grazing by mosquito larvae altered the composition of decomposer microbes. Specifically, the minimal fungal development in treatments containing *C. mollis* suggests that grazing controlled the growth of fungi that were abundant in non-grazer treatments.

Because odonates reduce mosquito abundance in artificial and natural tree holes (Fincke et al. 1997), the negative effect of *Mecistogaster* on *C. mollis* survivorship was

expected. Odonates occasionally eat mosquito pupae (Yanoviak 1999), so survivorship operationally defined as adult emergence rather than successful pupation may have produced an even stronger result.

I attribute low mosquito survivorship in the grazer-only treatment to nutrient limitation, which commonly affects the abundance and life history characteristics of treehole mosquitoes (e.g., Fish & Carpenter 1982, Carpenter 1983, Hard et al. 1989, Léonard & Juliano 1995). Evidence for food limitation in this study includes slower appearance of first pupae ($\approx$ 9 d compared to about 5 d for *C. mollis* reared on ad libitum yeast or fish food in the same laboratory), and the relative clarity of the water in treatments containing mosquitoes. In addition, differences in the foraging behavior of mosquito larvae surviving to the last week of the experiment compared to those that successfully pupated in the second week may be a reflection of starvation stress.

Predators can reduce mosquito abundance in treeholes (Fincke et al. 1997) and alter community parameters such as species richness (Yanoviak 1999), but "top-down" control of energy transfer among lower trophic levels may be relatively weak in this system. Although preliminary, my results suggest that this is true in the presence of size-selective predators.

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