Northern Illinois University

Group Feeding and Predatory Behavior of the Flatworm

Dugesia tigrina on the Isopod Caecidota sp.

A Thesis Submitted to the

University Honors Program

In Partial Fulfillment of the

Requirements of the Baccalaureate Degree

With University Honors

Department of Biological Sciences

by

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DeKalb, Illinois

December 1998

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HONORS THESIS ABSTRACT THESIS SUBMISSION FORM

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THESIS TITLE: Group Feeding and Predatory Behavior of the Flatworm Dugesia tigrina on the Isopod Caecidota sp.

ADVISOR: Dr. Carl von Ende	ADVISOR'S DEPT: Biological Sciences
DISCIPLINE: Aquatic Ecology	YEAR: 1998
PAGE LENGTH: 14	BIBLIOGRAPHY: Yes

ILLUSTRATED: Graphs and tables

PUBLISHED: NO

ABSTRACT:

The purpose of this project was to evaluate the effect of predator group size on per capita ingestion rates of the flatworm *Dugesia tigrina*, and to determine whether an optimum group size exists. In previous experiements performes by Cash et al. (1993,1995), it was determined that when *Dugesia* was fed *Daphnia* and mosquito larvae, per capita ingestion rates were higher in predator groups of four and eight. My project was designed to determine if the same pattern would be found if the flatworms were given isopods (*Caecidotea* sp.) to prey upon, which are benthic organisms. To accomplish this, flatworms were maintained in groups of one, two , four, eight, ten, and sixteen, and were fed a constant ratio of three isopods per predator. Each experiment was

run for five days, counting and replacing eaten prey daily. A total of six experiments were performed, three of which included a mesh screening in the bottom of the dishes. Per capita ingestion rates were determined and analyzed. The results showed no significant difference in per capita ingestion rates between group sizes. However, there was a significant overall decrease in per capita rates among the flatworms in the dishes containing screen compared to those without the screen. It is hypothesized that the screen enabled the isopods to more easily escape predation by the flatworms. It is thought that, in addition to a food source, this may be one reason that isopods in natural environments are found on the undersides of dead leaves and vegetation.

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Introduction

It has long been known that the flatworm, Dugesia tigrina, is a group feeder (Cash et al, 1993, Jennings 1957, Pickavance 1971). Recent studies have been conducted to determine whether group size actually has any affect on the per capita ingestion rates of flatworms. Cash et al. (1993,1995) found that D. tigrina did indeed exhibit clear optimal group sizes in terms of maximum daily per capita ingestion rates when fed it on zooplankton (Daphnia) and mosquito larvae. Their studies clearly showed that a significant increase in per capita feeding rates occurred in predator group sizes of four to eight, when predator group size ranged from one to sixteen, and the predator:prey ratio was a constant 1:3. However, *Daphnia* and mosquito larvae spend most of their time in the water column. Flatworms also prev on benthic invertebrates in nature as well. I was interested in whether group size influenced per capita ingestion rates when D. tigrina fed on freshwater isopods (*Caecidotea*), a common benthic invertebrate. Therefore, in this study I examined whether group size influenced per capita feeding of D. tigrina on the isopods Caecidotea, following the experimental protocol of Cash et al. 1993, 1995. Isopods commonly are found on the undersides of leaves and dead vegetation. In preliminary experiments, it was observed that the isopods clumped together in dishes at higher densities, unless provided with an additional flat substrate such as window screening. Therefore, I compared predation rates at different flatworm group sizes, with and without an artificial substrate (window screening) for the isopods.

Materials and Methods

Experimental Design

A total of six experiments was performed consecutively between March 31, 1998 and June 13, 1998. I provided screening in three of the experiments and no screening in the three control experiments. In each experiment, I compared predation rates at six different predator:prey densities, 1:3, 2:6, 4:12, 8:24, 10:30, and 16:48, with a constant predator:prey ratio of 1:3. There were three replicates of each predator:prey density in each experiment. The individual flatworms, *Dugesia tigrina*, that were used during these experiments were collected locally in the Kishwakee River in the fall of 1997 and were maintained throughout the experiments at room temperature in the laboratory. They were maintained on both caddisfly larvae (*Hydropsychidae*) and isopods (*Caecidotea*) as prey while in the lab and when not being used in the actual experiments. The isopods were collected from Wilkinson Marsh, DeKalb County, IL several times during the period that the experiments were performed.

Experimental Protocol

All experiments followed a similar procedure. The experimental containers were circular finger bowls with a diameter of 10.6 cm and a height of 4.0 cm. The dishes were filled with aged tap water to a depth of 2.0 cm. The substrate used in the three screening experiments was a round piece of window screening with a diameter of 9.0 cm. Flatworms of similar size (total head to tail length while gliding 0.6-1.0 cm) were randomly assigned to dishes and maintained throughout each experiment in groups of one, two, four, eight, ten, and sixteen individuals per container, with a mean body length of 0.8 cm per dish. Flatworms were starved for four days prior to running each

experiment, and all experiments were run for five days. In all experiments, flatworms were provided with isopods of similar size. All isopods were of similar size throughout all experiments. All dead and eaten prey were removed and replaced daily, and the total number of prey eaten in each experimental container was recorded daily. All tailbuds were removed daily, so as to maintain predator:prey densities. Evaporated water was also replaced daily to maintain water depth at 2.0 cm. The experiments were run in an environmental chamber at 20°C with a 12:12 light:dark photoperiod. All replicates were positioned randomly within four rectangular plastic containers within the environmental chamber. After each experiment, all flatworms were returned to the laboratory culture and randomly assigned to containers for the next experiment.

Statistical Analysis

To determine whether screening affected predation rates at different predator/prey densities, per capita feeding rates were analyzed using a 3 factor nested factorial ANOVA design: experiments were nested within no-screening (control) and screening treatment levels and cross-classified with predator/prey densities. There were three replicates at each prey density in each experiment. Experiment 3 was excluded from the analysis because the data was so dissimilar to the others that it was considered that something unusual occurred in that experiment. In addition, each set of 2 control and 3 screening experiments was analyzed individually as a 2 factor ANOVA to determine whether there was an optimum group feeding size within the screening and control treatments. In these analyses, experiment was treated as a random factor and predator/prey density as a fixed factor. Trends were analyzed in all three analyses using

orthogonal polynomials. All data was analyzed with the NCSS97 (NCSS, 1997) statistical package.

Results

The daily per capita predation rates of *Dugesia tigrina* provided with isopods at a predator:prey ratio of 1:3 varied significantly between control and screening experiments (p=0.000004, Table 1). On average, predation rates were lower in the dishes with screening ($\bar{x} = 0.13$) than those without the screening ($\bar{x} = 0.33$). There was no significant difference in the number eaten at different densities (p = 0.10, Table 1), but there was a significant overall linear decrease in per capita ingestion as the predator group size increased (p = 0.0068, Figures 1, 2). There was a significant interaction between experiment and predator density (p = 0.0015), which meant that predation rates for different group sizes varied significantly between experiments.

Per capita ingestion rates were not found to be significantly different between predator densities for the control experiment (p = 0.546, Table 2); however, there was a significant interaction between experiment and predator density (p = 0.0055), indicating that the results for the control experiments were highly variable.

Per capita ingestion rates were not found to be significantly different between predation densities in the screening experiments (p = 0.074, Table 3); however, a linear as well as a quadratic trend that was marginally significant was found in the screening experiments. There was a general decrease in per capita ingestion rates as group size increased (linear, p = 0.0327; quadratic, p = 0.0524).

Discussion

In their experiments, Cash et al. (1993,1995) found a definite increase in per capita ingestion rates when D. tigrina fed on Daphnia in groups of four or eight. Therefore, these group sizes were labeled as the optimum group sizes. My studies, however, did not show any significant increase among specific feeding group sizes. Maximum peak levels were not achieved consistently with any one group size; therefore, no optimum group feeding size was discovered. This could be due in part to the difference in prey type. Whereas Daphnia and mosquito larvae, the prey used by Cash et al., spend their time in the water column, the isopods I used were benthic. Perhaps this made the prey easier to catch and therefore large predator groups offered no advantage in prey capture. The flatworms themselves tended to stay along the bottom and sides of the containers and on the surface of the water, but did not glide throughout the water column. Since isopods mainly remained on the bottom of the dishes, the likelihood of an encounter with a predator was increased. Perhaps this also reduced the advantage of group feeding. This is consistent with the observed pattern of an overall decrease in per capita ingestion rates as predator group size increased. Also, isopods are larger prey than Daphnia, so perhaps not as many were needed in order to satiate the flatworms. For example, one isopod may have fed several flatworms. So if one prey was eaten in a dish with one flatworm, and one prey was eaten in a dish containing four flatworms, then the per capita ingestion rates for the second container would be lower. Another possibility is based on the observation that the flatworms tended to attack newly added prey over old prey. After an isopod had been in a dish for awhile, it tended to be basically immobile,

whereas an isopod newly transferred to a dish tended to move around more. *Daphnia* are also very mobile organisms, so it may be that flatworms prefer more mobile prey.

There was a significantly lower overall per capita ingestion rate in the screening experiments as compared to control experiments. Perhaps this was due to an increased ability of the isopods to move about in the dishes with screening. It was observed that the isopods had some trouble moving about on the smooth glass bottom of the dishes without screening, whereas flatworms glided easily across the bottom and sides of the dishes. It may have been difficult to for the isopods to escape flatworm predation. At high densities the isopods clumped together into tightly knit balls of animals: perhaps this was a defense mechanism when they could not escape on the slippery bottom of the dishes. Clumping together may have reduced any single isopod's chance of being eaten. Providing the screen substrate, however, allowed the isopods a more sure footing; this may have allowed them an easier and faster escape when being pursued. The screening did not appear to affect the ability of the flatworms to move about. It is speculated that is one reason isopods in nature are found on substrate such as dead leaves and vegetation. In addition to providing them a source of food, the substrate may also provide them a greater advantage to escaping their natural predators.

Although no optimum group size was found when *D. tigrina* was fed isopods in these experiments, a very significant decrease in per capita ingestion rate was observed when isopods were provided with a screen substrate. To determine whether this was due to enhanced ability to escape predation or because of reduced clumping by the isopods is unknown at this time, and would require further investigation.

Table 1	ANOVA comparing predation rates of Dugesia tigrina on the isopod
	Caecidota sp. at different Dugesia group sizes with and without screening.

		Sum of	Mean		Prob
Treatment	DF	<u>Squares</u>	Square	<u>F-Ratio</u>	Level .
Screening (S)	1	0.99	0.99	6556.65	0.00
Experiment (E)	3	4.53E-04	1.51E-04	0.01	0.99
Group Size (G)	5	0.34	6.97E-02	2.26	0.10
S x G	5	0.15	3.01E-02	0.98	0.46
E x G (S)	15	0.46	3.08E-02	2.96	0.00
Error	60	0.62	1.04E-02		

Table 2ANOVA comparing predation rates of Dugesia tigrina on the isopodCaecidotea sp. at different Dugesia group sizes in the control condition
(no screening).

		Sum of	Mean		Prob	
Treatment	DF	Squares	<u>Square</u>	F-Ratio	Level	<u> •</u>
Experiment (E)	1	1.36E-04	1.36E-04	0.01	0.92	
Group Size (G)	5	0.27	5.53E-02	0.90	0.54	
ExG	5	0.30	6.17E-02	4.40	0.01	
Error	24	0.33	1.40E-02			

Table 3ANOVA comparing predation rates of *Dugesia tigrina* on the isopod*Caecidotea* sp. at different *Dugesia* group sizes in the screening treatment.

Treatment DF		Sum of Squares	Sum of Mean Squares Square		Prob <u>Level</u>	Prob <u>Level</u> .	
Experiment (E)	2	3.17E-04	1.58E-04	0.02	0.98		
Group Size (G)	5	0.21	4.17E-02	2.71	0.08		
ExG	10	0.15	0.02	1.92	0.07		
Error	24	0.28	8.02E-03				

Sheet1

Appendix 1									
Raw data collected in the control (no screening) treatments.									
	Experiment 1 Experiment 2 Experiment 3								
Replicate	Pred/prey ratio	Total Eaten	Per Capita	Total Eaten	Per Capita	Total Eaten	Per Capita		
A	1:3	2	0.4	3	0.6	0	0		
В	1:3	0	0	3	0.6	0	0		
С	1:3	2	0.4	2	0.4	2	0.4		
A	2:6	7	0.7	4	0.4	1	0.1		
В	2:6	5	0.5	4	0.4	2	0.2		
С	2:6	6	0.6	2	0.2	2	0.2		
A	4:12	6	0.3	6	0.3	4	0.2		
В	4:12	3	0.2	7	0.35	2	0.1		
С	4:12	11	0.55	6	0.3	3	0.15		
А	8:24	12	0.3	15	0.38	4	0.1		
В	8:24	21	0.53	14	0.35	6	0.15		
С	8:24	13	0.33	19	0.48	5	0.13		
A	10:30	21	0.42	20	0.14	8	0.16		
В	10:30	19	0.38	13	0.26	4	0.08		
С	10:30	25	0.5	18	0.36	6	0.12		
A	16:48	10	0.13	21	0.26	6	0.08		
В	16:48	9	0.11	15	0.19	15	0.19		
С	16:48	4	0.05	29	0.36	9	0.11		

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Sheet1

Appendix 2										
Raw data collected from the screening treatments										
	Experiment 4 Experiment 5 Experiement 6									
Replicate	Pred/Prey ratio	Total Eaten	Per Capita	Total Eaten	Per Capita	Total Eaten	Per Capita			
A	1:3	2	0.4	1	0.2	0	0			
В	1:3	2	0.4	0	0	3	0.6			
С	1:3	2	0.4	1	0.2	1	0.2			
A	2:6	2	0.2	2	0.2	1	0.1			
В	2:6	1	0.1	2	0.2	2	0.2			
С	2:6	1	0.1	2	0.2	0	0			
A	4:12	1	0.2	3	0.15	1	0.05			
В	4:12	0	0	2	0.1	5	0.25			
С	4:12	0	0	2	0.1	3	0.15			
A	8:24	2	0.05	5	0.125	6	0.15			
В	8:24	1	0.03	5	0.125	3	0.08			
С	8:24	2	0.05	3	0.075	5	0.13			
A	10:30	4	0.08	5	0.1	5	0.1			
В	10:30	2	0.04	9	0.18	4	0.08			
С	10:30	2	0.04	8	0.16	4	0.08			
Α	16:48	6	0.08	9	0.11	5	0.06			
В	16:48	13	0.16	9	0.11	6	0.08			
C	16:48	12	0.15	11	0.14	5	0.06			

Figure 1. Per capita predation rate $(x \pm 1 \text{ SE})$ of *Dugesia tigrina* on *Caecidotea* sp. in control and screen dishes for different group sizes of *D. tigrina*.







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