COMPARISON OF ECTOSYMBIOTIC GILL INHABITANTS OF FISH FROM TWO AREAS

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COMPARISON OF ECTOSYMBIOTIC GILL INHABITANTS OF FISH FROM TWO AREAS

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PREFACE

This thesis concerns the effects of water quality on aquatic life in Stillwater Creek, a tributary of the Cimmaron River in northcentral Oklahoma. The objective was to determine if differences in water quality were reflected in the differences in the communities of gill inhabitants of fish from Stillwater Creek.

I thank Dr. O. Eugene Maughan of the Oklahoma Cooperative Fishery Research Unit and Dr. S. L. Burks of the Water Quality Research Laboratory for making available equipment and space for this project. The advice and assistance of Elaine Stebler of the Water Quality Research Laboratory is acknowledged. Dr. William D. Warde of the Oklahoma State University Statistics Department set up the experimental design and assisted in the data analysis. Dr. Mike Douglas was also consulted during data analysis.

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To my wife Donna I owe completion of this work. Her endless patience and service as my sole technician made this project possible.

Finally, to my parents I extend my gratitude for teaching me to appreciate the outdoors and thus beginning my education.

TABLE OF CONTENTS

LIST OF TABLES

FIGURE

CHAPTER I

INTRODUCTION

The reactions of aquatic organisms to pollution can be detected at at least two levels of biological organization. First, within individuals, responses such as changes in blood chemistry, metabolism, morphology, histology, behavior, and basic condition of the individual can be measured. At the community level alterations commonly associated with pollution occur in the total number of individuals, number of species, diversity, and the relative importance of some species. Changes in at least one of these two major levels usually are studied in evaluating the effects of pollution.

Short term pollution may escape notice if only abiotic variables are monitored, especially if the monitoring is done only infrequently. However, the aquatic community, or at least certain species, generally exhibit changes at one of the two biological levels if the pollutant is toxic. Therefore biologists use the biota to detect recent environmental perturbations. In short, aquatic communities may be excellent indicators of the amount of stress that is currently in the system or has been in the system in the recent past.

Several major groups of organisms including algae, macroinvertebrates, and fish have been used extensively in biomonitoring programs. However, the relationships among fish, the ectosymbionts residing on their gills, and water quality have not been well studied.

My objective was to determine the effects of altered water quality, the result of rural and urban runoff and treated sewage effluent, on gill ectosymbionts of Gambusia affinis and Lepomis cyanellus. The interaction between these symbionts and their fish hosts in this setting depends largely on two groups of factors: 1) the effects of pollutants upon susceptibility of fish to the commensals and/or parasites and 2) the effects of pollutants upon the commensals and/or parasites themselves.

CHAPTER II

LITERATURE REVIEW

Pollution and Fish Susceptibility to Disease

Many fish and shellfish diseases and abnormalities have been reported to be associated with marine pollution (Sinderman 1979). These diseases and abnormalities include fin erosion, ulcers, shell disease, lymphocystis, bacterial diseases, neoplasms, skeletal defects, genetic anomalies, and suppression of the immune system. For example, Stich et al. (1976) found that skin papillomas in flatfish species appeared to be related to pollution discharges, and Mearns and Sherwood (1977) in a study of 151 fish species off the coast of southern California, found only fin erosion to be correleted with waste discharge.

Numerous studies of fish health in relation to pollution of fresh water have also been conducted. For example Brown et al. (1977, 1979) investigated the effects of different levels of pollution on the health of fish from three rivers. In a majority of cases the number of diseased fish was positively correlated with the level of pollution. Specifically, the gills of several fish species inhabiting polluted water were found to harbor more monogenetic trematodes than the same fish species found in similar but comparatively unpolluted habitats (Hanek and Fernando 1978a, 1978b, Skinner 1982). Other authors also have reported anomalies in fish in close proximity to polluted areas. Among these afflictions are listlessness, weight loss, lesions, exophthalmia

(Young 1964), neoplasms (Dawe et al. 1964; Young 1964; Matsudo and Chamberlain 1976; Kraybill et al. 1977), spinal curvature (Wunder 1976), fin erosion (Perkins et al. 1972; Wunder 1976), papillomas (Young 1964; Grizzle 1981), lymphocystis (Perkins et al. 1972, Overstreet and Howse 1977), epidermal ulcers (Perkins et al. 1972; Mahoney et al. 1973), blindness, skin hemorrhages (Mahoney et al. 1973), and death (Young 1964; Pippy and Hare 1969; Cardeilhac et al. 1981).

Effects of pollution on the gill are of particular interest because this tissue is closely and continuously irrigated by the water and because water comes in close proximity to the tissue fluids. Gill damage such as hyperplasia, fused and swollen lamellae, clubbing of filaments, hemorrhage, excess mucus production, and sloughing of epithelial cells have been reported to result from exposure to pesticides, heavy metals, organic toxicants, nitrogenous waste products, detergents, and chlorine (Abel 1974; Eller 1975; Bass and Heath 1975; 1977; Bass et al. 1977; Skinner 1982). These reported changes in gill tissue may have important implications for those organisms residing or feeding on the gills, such as the peritrich protozoans Epistylis and Glossatella (= Apiosoma) and monogenean flatworms.

Another factor that strongly influences susceptibility of fish to disease is the suppression of defense mechanisms following exposure to toxicants. Generally, susceptibility to a pathogen is increased by experimental exposure to a sublethal stressor (Flagg and Hinck 1978; Roales and Perlmutter 1977; Rodsaether et al. 1977; Hetrick et al. 1979; Knittel 1980, Ewing et al. 1982; Snarski 1982). Conversely, it has been demonstrated that the toxicity of certain compounds is increased by prior infection with a pathogen (Boyce and Yamada 1977; Guth et al.

1977; Pasco and Cram 1977; Iwama and Greer 1980; Moles 1980).

Actual suppression of the immune system in fish is difficult to demonstrate but some pollutants have been shown to inhibit the production of various immunological components (Street and Sharma 1975, Sarot and Perlmutter 1976, Roales and Perlmutter 1977, 1980, O'Neill 1981). Enzymological changes have also been documented in response to stress (Sastry and Agrawall 1979a, 1979b, Verma et al. 1979).

Pollution and Protozoan Survival

Until recently the use of protozoans as bioindicators has been largely neglected because of difficulties in their preservation and identification. Identification often depends on observing live specimens (Hynes 1960); thus samples must be examined within hours of collection. Furthermore, alterations in the community structure and composition may result from environmental changes within the holding vessel prior to examination (Cairns 1978).

However, recently, protozoa have been used in evaluations of water quality and are recognized to have certain advantages for this type of work. They have a larger surface to volume ratio than fish, insects, and other organisms commonly used in biomonitoring, and the body covering is typically thin and therefore may be more permeable to toxicants than that of many other aquatic organisms. As Noland (1925 p.449) said of protozoans: "There are few organisms that come in closer contact with their surroundings."

Several authors have compared the ecology of protozoans in clean and polluted environments. Certain species of ciliates have been shown to be associated with definite ranges of saprobic activity in activated

sludge processes (Curds 1965, 1973; Curds and Cockburn 1970). In addition, exposure to heavy metals and sewage-induced eutrophic conditions were found to result in reduction of diversity, number of species, and colonization rates as well as alteration of periodicity of population fluctuations (Carter and Cameron 1973; Gibson and Grice 1977; Cairns et al. 1980; Moraitou-Apostolopoulou and Ignatiades 1980; Dive et al. 1980; Honig et al. 1980). In contrast, Suehiro and Tezuka (1981) concluded that interspecific interactions were more important in regulating seasonal changes in the ciliate populations of the bottom sediments of a sewage polluted river than were food availability or occurrence of toxins.

Few investigators have studied aquatic animals as substrates for ectocommensal/ectoparasitic protozoans in relation to water quality as was done in this study. Henebry and Ridgeway (1979), however, examined planktonic copepods and cladocerans from a eutrophic lake for the presence of ectocommensal ciliates and found infestation levels higher than those reported from other habitats. Similarly, in a study of mosquito larvae from streams and ponds in Singapore, Laird (1959) found ectocommensal protozoa considered to be pollution indicators although no documentation of pollution was obtained. Similarly, Antipa (1977) found that one species of commensal protozoa of freshwater bivalve molluscs was more sensitive to organic pollution than was the host, suggesting that this protozoan might also be considered a pollution indicator.

Pollution Sources

The current study on gill ectosymbionts of fish was undertaken on Stillwater Creek, which receives three sources of pollution: municipal

sewage, urban runoff, and rural runoff. Among the common stressors of aquatic communities are sewage contamination and the associated products of chlorine treatment. Nevertheless, in a detailed survey of the protozoan fauna of a stream receiving treated sewage effluent, Small (1973) reported the greatest diversity each season at a location less than one mile below a combined wastewater discharge. At this so-called "septic" station Epistylis and Trichophrya, genera which contain species that are known ectocommensals of fish, were collected.

Elevated ammonia and lowered dissolved oxygen concentrations are the primary deleterious characteristics of untreated sewage, but chlorine or chlorine residuals comprise the most toxic component of treated sewage effluents (Cross 1950; Zillich 1972; Brungs 1973; Gehrs and Eyman 1974; Brungs 1976; Katz and Cohen 1976; Alexander et al. 1977; Bellanca and Bailey 1977; Dickson et al. 1977; Ward and DeGrave 1978). Tsai (1973) characterized the condition below 149 secondary sewage plants in Virgina, Maryland, and Pennsylvania and found that a noxious environment existed below outfalls, primarily as a result of chlorine and turbidity.

Cairns and Plafkin (1975) found the more frequent the exposure to hypochlorite the greater the reduction in protozoan numbers in fresh water. Experimental exposure to chlorine resulted in a substantial reduction in the total number of individuals. Diversity was not significantly altered by three exposures in a two-hour period at concentrations less than 1.45 ppm but at shorter exposure intervals and higher concentrations the number of species was reduced and the most tolerant species became relatively more important.

Runoff from streets (urban runoff) is typically highly contaminated

and similar to secondary sewage in its effects (Sartor and Boyd 1972; Yu et al. 1975; Wullschleger et al. 1976; Porcella and Sorensen 1980; Heaney et al. 1981). Some receiving water parameters that may be altered significantly are B.O.D., C.O.D., suspended solids, and concentrations of oil and grease, heavy metals, nutrients, nitrites, nitrates, phosphates, sulfates, and other salts. The more important instream biological effects are on biomass, diversity, and P/R ratios. In urban situations where sanitary and storm sewers are combined, toxicants in runoff may affect the biological purification process in sewage treatment plants. Also, impacts on receiving water may be more severe where the dilution capacity is not adequate.

The effects of rural runoff are subtle but eutrophication due to nutrient input and contamination with chemicals are of principal concern (Cope 1965; Evans and Duseja 1973; Kilmer et al. 1974; McElroy et al. 1975; Skaggs et al. 1981; Sullivan 1983). In general if fertilizers and pesticides are judiciously applied, concentrations in runoff are negligible (Kilmer et al. 1974, Skaggs et al. 1981). However heavy rains after application may increase concentrations to toxic levels (Evans and Duseja 1973).

Literature Summary

Biomonitoring is a suitable method for detecting change in the aquatic community and protozoans may be reliable pollution indicators. In combination with other methods their use may aid biologists in judging the impact of pollutants on aquatic communities.

Fish, the hosts of the protozoans used in this study, are themselves subject to physiological, immunological, and histological

alterations induced by environmental changes. Of particular concern are immunological and changes as they relate to the host's defense against ectosymbionts histological changes as they relate to the condition of the host's gills. Finally, the suspected sources of pollution in this project, sewage effluent, and urban and rural runoff, clearly have been shown to cause severe alterations in the biological and chemical environment of aquatic ecosystems. Faster and more reliable methods to assess the impact of pollution are always needed and ectocommensal protozoans may in part meet this need.

CHAPTER III

DESCRIPTION OF STUDY AREA

Stillwater Creek is a tributary of the Cimarron River in Payne County, Oklahoma (Fig. 1), and courses from northwest to southeast just south of the city of Stillwater, population approximately 35,000. The confluence with the Cimarron occurs approximately 14.9 km southeast (downstream) of the Stillwater city limit. Five major tributaries join Stillwater Creek within the study area. These are: 1) Harrington Creek; 2) North Stillwater Creek; 3) Cow Creek; 4) Boomer Creek; and 5) Brush Creek. On four of these streams there are major impoundments. Lake Carl Blackwell, the largest at 7820 hectares, impounds the upper reaches of Stillwater Creek proper. Lake McMurtry is located on North Stillwater Creek near Carl Blackwell. Ham's Lake, located on Harrington Creek, is a private impoundment of 41 hectares. Lastly, Boomer Lake is situated on Boomer Creek within the Stillwater city limit.

Nonurban land within the drainage basin is used primarily for grazing and cultivation. Stillwater is the sole municipality of significant size within this area. The Stillwater sewage treatment plant (STP) is located on Brush Creek immediately upstream from the confluence of Brush and Stillwater Creeks. This confluence is downstream from the city limit and approximately 11.7 km upstream from the Cimarron River.

This study was conducted at two locations on Stillwater Creek (Fig. 1). Location A, including three sites (1,2, and 3), is located

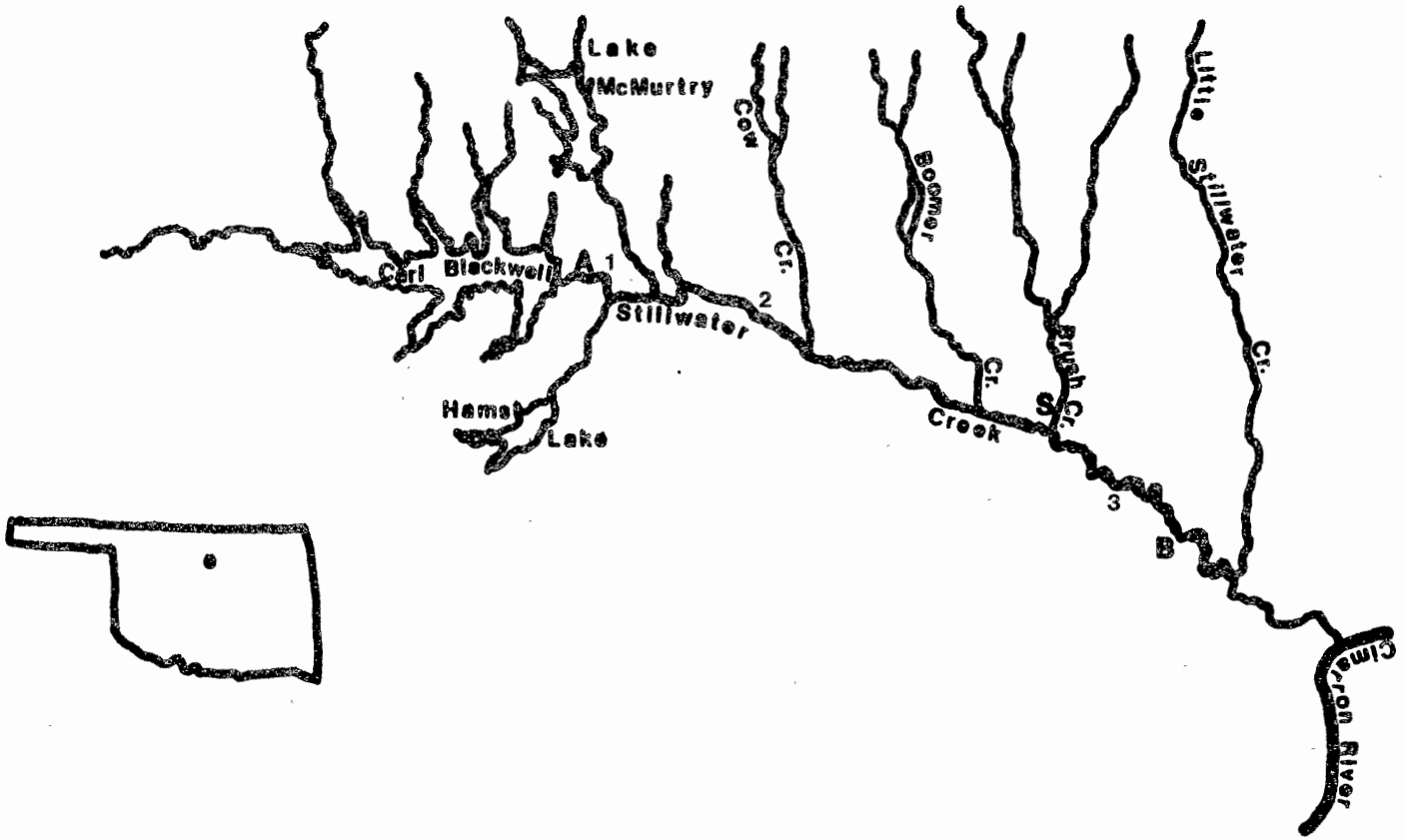

Figure 1. Map of study area showing location A, location B, the Stillwater sewage treatment plant(S), and three stations used by the Oklahoma Department of Water Resources.

immediately below Lake Carl Blackwell (SW 1/2, Sec. 10, T19N, R1E). Location B, also including three sites (4,5, and 6), is located downstream from the Stillwater STP (NW 1/4, Sec. 11, T18N, R3E). Location A has relatively clean water and location B, downstream from the STP, has water of poorer quality. Stream flow was measured at three sites in 1982 by the Oklahoma Department of Water Resources (Fig. 1).

CHAPTER IV

METHODS AND MATERIALS

Three collections were made during the summer of 1981 and four during 1982. Two species of fish were collected for examination: green sunfish, Lepomis cyanellus Rafinesque and mosquitofish, Gambusia affinis Baird and Girard. Analysis of the variation encountered in preliminary samples indicated that a monthly sample of six individuals of each species at each site would be necessary to detect significant differences between locations A and B. Dissolved oxygen concentrations and temperature also were measured at each site.

All fish were collected with a Type VII Smith and Root backpack-mounted electroshocker and placed in buckets with creek water, each species in a separate bucket, for transportation to the laboratory. In 1981 50% of the creek water in each bucket was replaced in the laboratory with deionized-water which had been passed through an activated carbon filter.

Fish were examined as soon as was practicable. Four gill arches, from the same side, left or right, were excised from each fish for examination with a light microscope. Symbiotic organisms were identified to the lowest taxonomic level practicable and were counted. Additional data collected or calculated included length, weight, age (scale annuli 3 method), sex, and condition factor [K = (grams x 10)/inches] of each fish, diversity of organisms found on each fish, elapsed time from

capture to workup, and the location (left or right) of the gill arches were removed. Diversity (Wilhm and Dorris 1968) of the ectosymbiont community found on each fish was calculated using the equation of Wilhn and Dorris (1968).

$$d = 1 \sum_{i=1}^{s} {}^{n_i}/_{n} \log_2 \left({}^{n_i}/_{n} \right).$$

This diversity index is independent of sample size making comparisons valid between fish harboring different numbers of organisms or organism groups. Relative density, defined by Margolis et al. (1982) as the mean number of individuals of a particular taxon per host, for all hosts examined, was also determined.

Means of these data were calculated for each fish, host, species, site, and location for each month. Correlation coefficients between appropriate variables and F-tests were determined using the Statistical Analysis System computer package (Helwig and Council 1979). F-tests were performed to detect any significant differences between locations with respect to relative density of each taxon and of the total ectosymbiont community. Protozoan spores, probably of the order Myxosporidia were not included in comparisons of total community density because they occurred sporadically and their populations varied enormously.

CHAPTER V

RESULTS AND DISCUSSION

Abiotic Features

Stillwater Creek flow during spring 1982 was 0.3 cfs and 0.2 cfs respectively at two sites above the city and 8.0 cfs just below the outfall from the sewage treatment plant. Stream recharge between the upstream site nearest the plant and the outfall accounted for 96% of the stream volume as measured below the outfall. Sewage plant effluent was a large portion of this recharge.

The dissolved oxygen concentrations observed were inconsistent (Table 1). Measurements generally were taken between 0700 and 1000 hours and occasionally were taken later in the day. The lowest and highest measurements were encountered at the downstream location. Large variation in oxygen levels is characteristic of organically enriched aquatic environments although no pattern consistent with enrichment was observed.

In this study water quality in collecting buckets was influenced by holding time. During the time fish were held, changes in the water quality may have affected ectosymbiont populations. These changes vary with holding times and include shifts in temperature and dissolved oxygen, metabolite, and bacteria concentrations. Clearly the holding times in 1981 (Table 2) would have had a greater effect on the ectosymbionts than in 1982. In addition, holding times may have been

Table 1. Temperature (°C) and dissolved oxygen (mg/l) levels.

	1981						1982							
	May		June-July		Sept.		June		July		Aug.		Sept.	
Locations	A	B	A	B	A	B	A	B	A	B	A	B	A	B
Temp (°C)	19.9	22.7	28.9	26.9	21.6	25.2	24.0	22.0	28.7	29.7	28.0	24.4	21.8	18.9
D.O. (ppm)	3.0	3.4	6.3	2.7	4.8	4.4	4.8	5.6	4.5	11.0	5.0	5.0	6.8	5.2

Table 2. Holding times (hours) for Gambusia and Lepomis by year, month, and location.

	1981						1982							
	May		June-July		Sept.		June		July		Aug.		Sept.	
	A	B	A	B	A	B	A	B	A	B	A	B	A	B
Gambusia	40.9	84.8	57.9*	123.3	279.0*	82.6	26.1	9.3	27.5	17.3	23.8	20.2	23.6	20.6
S.D.	21.9	46.5	27.7	59.2	54.9	34.1	14.9	11.2	12.0	3.4	9.1	10.8	10.1	2.4
Lepomis	40.0*	84.7	56.7	87.2	188.2	73.3	24.4	20.0	23.2	25.4	23.2	17.7	25.7	14.2
S.D.	22.6	55.2	27.2	56.9	146.5	46.3	14.2	17.6	10.4	12.8	13.6	12.3	13.4	5.3

* Denotes a significant difference between locations.

excessive for a species-association analysis involving protozoans to accurately reflect the actual relationships present in the stream. It has been reported that, for protozoans, samples should be examined within 8 hours to avoid temporal and water quality related changes in the community (Cairns 1978).

Ectosymbionts of Mosquitofish

Of the 211 Gambusia affinis examined, 184 (87.2%) harbored at least one ectosymbiont. No consistently significant ($\alpha = .05$) differences between locations in relative densities of any ectosymbionts were found (Table 3).

Monogenean flatworms, unidentified ciliates, and flagellates were always at least as abundant at location A as at B. Monogeneans have been found to flourish in eutrophic conditions (Hanek and Fernando 1978a, 1978b). The fact that monogeneans were found in larger numbers at location A, a relatively clean habitat, than at location B, which received sewage effluent, is therefore surprising. Perhaps the trematode offspring found a suitable host more readily at the three upstream sites in the absence of flow than at location B where there were currents. A similar influence of currents was suggested by Davis and Huffman (1977). Another explanation is the possible presence of chlorine at location B and not at location A, although not documented for monogeneans, chlorine in low concentrations is toxic to many aquatic organisms (Cairns and Plafkin 1975, Dickson et al. 1977).

The percent of the total ectosymbiont load attributable to each group was used to determine the dominant taxa in the mosquitofish community (Table 4). Only monogeneans, Trichodina sp., Scyphidia sp.,

Table 3. Relative densities and standard deviations of Gambusia ectosymbionts.

Location	1981						1982							
	May		June-July		Sept.		June		July		Aug.		Sept.	
	A	B	A	B	A	B	A	B	A	B	A	B	A	B
Monogeneans	3.2	0.0	2.3	1.2	3.1	1.3	3.6	0.5	6.7*	0.0	4.1	0.2	2.3	1.1
S.D.	4.9	0.0	2.9	1.9	3.3	1.8	3.5	1.0	5.6	0.0	8.2	0.4	1.6	2.1
Flagellates	0.7	0.0	0.7	0.0	1.3	1.7	1.7	0.0	0.4	0.0	18.6	0.0	0.0	0.0
S.D.	3.1	0.0	2.1	0.0	5.7	7.3	7.3	0.0	1.7	0.0	79.0	0.0	0.0	0.0
Trichodina	0.1	0.2	0.0	0.0	0.0	0.3	0.1	0.0	0.1	0.0	0.0	0.1	0.0	0.0
S.D.	0.2	0.4	0.0	0.0	0.0	1.2	0.2	0.0	0.2	0.0	0.0	0.3	0.0	0.0
Scyphidia	0.4	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	1.1	0.0	0.2	0.1
S.D.	1.5	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	3.8	0.0	0.5	0.2
Glossatella	7.9	106.5	4.1	5.0	1.7	6.9	6.3	102.5	2.8	1.3	19.2	81.4	8.4	6.6
S.D.	8.6	274.3	6.6	11.3	4.9	15.0	5.8	96.1	6.2	1.0	28.8	157.9	16.4	10.6
Spores	0.0	0.0	5.1	8.9	123.9	0.0	0.0	0.0	140.3	0.0	0.0	0.8	277.8	0.0
S.D.	0.0	0.0	21.7	40.6	524.4	0.0	0.0	0.0	435.0	0.0	0.0	2.3	826.4	0.0
Ciliates	0.0	5.3	0.0	0.2	0.0	0.1	0.1	0.5	0.2	0.3	0.0	0.1	0.1	3.0
S.D.	0.0	8.1	0.0	0.5	0.0	0.2	0.2	1.0	0.7	0.5	0.0	0.3	0.5	11.3
Totals	12.3	112.0	12.2	15.3	130.0	10.3	11.8	103.5	150.5	1.6	43.0	81.8	288.8	10.8

* Denotes a significant difference between locations.

1/ Probably of the protozoan order Myxosporidia.

Table 4. Ectosymbiont dominance as percent of the total organism load per individual *Gambusia*.

Locations	1981						1982							
	May		June-July		Sept.		June		July		Aug.		Sept.	
	A	B	A	B	A	B	A	B	A	B	A	B	A	B
Monogeneans	40.0	0.0	44.9	50.4	85.4	52.8	41.7	10.0	81.3	0.0	26.8	6.7	44.4	29.8
Trichodina	0.6	6.6	0.0	0.0	0.0	8.0	0.6	0.0	0.2	0.0	0.0	6.7	0.0	0.0
Scyphidia	2.9	0.0	0.0	0.0	0.0	0.1	0.0	0.0	0.0	0.0	6.5	0.0	0.0	7.1
Glossatella	56.5	70.0	55.1	35.1	14.6	36.8	57.3	89.4	18.0	83.3	66.7	80.0	53.5	53.6
Ciliates	0.0	23.3	0.0	14.4	0.0	2.2	0.4	0.6	0.5	16.7	0.0	6.7	1.3	9.4

Glossatella sp., and unidentified ciliates were used in the analysis because these comprised almost the entire community found on mosquitofish gill. In all cases, trematodes and Glossatella were the dominant groups and in combination they made up at least 70% of the load in each monthly location mean and greater than 90% in 8 of the 14 location/months. No similar data were found in the literature for Glossatella, but a single species of monogenean has been reported to account for as much as 84.7% of the total parasite load on the gills of Lepomis gibbosus (Hanek and Fernando 1978a). Of the other taxa, only unidentified ciliates constituted greater than 8% of the organism load in any monthly location mean and never exceeded 24%. Hence, the mosquitofish community was overwhelmingly dominated by two taxa, monogeneans and Glossatella sp., while unidentified ciliates were occasionally important. Nevertheless, no consistently significant (α = .05) pair-wise linear relationships were detected among the ectosymbionts.

Theoretically, an ecosystem under stress has a less complex community structure and exhibits a lower diversity than one which is not stressed. Although diversity of ectosymbionts on Gambusia was not consistently and significantly different between locations, the diversity of ectosymbiont groups was higher at location A than at B in all 4 monthly samples in 1982 and 2 of the 3 monthly samples in 1981 (Table 5). In the one month in 1981 when the diversity of location A was lower than that at B (September 1981) mean holding time at location A (279 hours) was the longest in the study. This may have reduced the numbers of protozoa and resulted in the low diversity (d = .07) observed at location A on that date.

The diversities reported here are lower than those reported by

Table 5. Host condition parameters and ectosymbiont diversity parameters for Gambusia, means and standard deviations.

	1981						1982							
	May		June-July		Sept.		June		July		Aug.		Sept.	
	A	B	A	B	A	B	A	B	A	B	A	B	A	B
Length (mm)	38.1	36.7	25.9	34.8	28.2	29.6	35.8	34.8	36.2	39.3	27.2	29.2	29.8	27.7
S.D.	7.8	8.3	6.8	10.0	5.6	6.4	7.6	6.9	7.6	4.2	4.9	7.0	6.1	4.5
Weight (gm)	0.6	0.6	0.2	0.6	0.2	0.3	0.4	0.4	0.5	0.7	0.2	0.2	0.2	0.2
S.D.	0.3	0.5	0.3	0.5	0.2	0.2	0.5	0.3	0.3	0.3	0.1	0.2	0.2	0.1
Condition factor	1.4	1.6	1.6	1.6	1.2	1.5	1.4	1.4	1.4	1.9	1.3	1.2	1.3*	1.4
S.D.	0.3	0.6	0.3	0.4	0.2	0.6	0.3	0.2	0.2	0.3	0.1	0.2	0.1	0.2
Age (yrs.)	0.9	0.8	0.3	0.6	0.2	0.3	1.0	1.0	0.8*	1.3	0.3	0.3	0.1	0.2
S.D.	0.5	0.4	0.4	0.7	0.4	0.5	0.6	0.0	0.4	0.5	0.5	0.5	0.3	0.4
Taxa/fish	1.7	1.4	1.4	0.9	1.0	1.3	1.8	1.5	1.6	1.0	1.9*	1.0	1.9	1.1
S.D.	0.9	1.1	0.8	0.7	0.5	0.8	0.7	0.6	0.9	0.8	0.8	1.1	0.8	0.7
Species Diversity S.D.	0.5	0.4	0.4	0.2	0.1	0.3	0.6	0.3	0.4	0.3	0.5	0.3	0.7	0.2
	0.6	0.6	0.5	0.4	0.2	0.4	0.5	0.5	0.5	0.5	0.4	0.6	0.5	0.3
Number fish exam.	18	4	18	4[1/]	18[2/]	9	18[2/]	18	18	11	18[3/]	21	18	18[4/]

* Denotes a significant difference between locations.

1/ Weight obtained for only 3 of these 4 fish.

2/ Weight obtained for only 17 of these 18 fish.

3/ Age obtained for only 16 of these 18 fish.

4/ Weight obtained for only 17 and weight for only 16 of these 18 fish.

Cloutman (1975) for largemouth bass, bluegill, and warmouth in his examination of the entire fish. Diversity is a function of both the number of taxa and number of individuals in each taxon. Thus, it is not suprising that the number of taxa per fish exactly paralleled diversity (Table 5).

Prevalence, expressed as the percentage of fish infected by a particular ectosymbiont group, reflects the extent of occurrence for each ectosymbiont group on Gambusia (Table 6). Only Glossatella attained a prevalence of 100% and frequently was 50%. Zero to 13.6% prevalence has been reported for this organism from several host species (Molnar et al. 1974). Monogeneans were always in excess of 65% prevalence at location A but never more than 45% at location B, the location receiving sewage effluent. Again this result is not consistant with what has been reported for monogeneans occurring in eutrophic conditions (Hanek and Fernando 1978a, 1978b). However, the prevalence levels reported here are within the range typically reported. Flagellates were always at least as prevalent at location A as at location B, but the levels were very low in comparison with monogeneans and Glossatella. Prevalence of taxa other than monogeneans and Glossatella only occasionally reached 20%.

It is notable that Tripartiella, Trichophrya, and Epistylis, organisms which occurred on green sunfish, were never found on mosquitofish. The prevalence of Epistylis has been reported to be between 0.0% and 28.0% on species other than mosquitofish (Lom and Chapman 1976; Chapman et al. 1977; Lewis et al. 1977; Cloutman et al. 1978; Roche et al. 1978). In a study in which three mosquitofish were examined, however, no Epistylis were observed, a finding consistent with

Table 6. Prevalence of ectosymbionts on Gambusia as percentage of fish infected.

Location	1981						1982							
	May		June-July		Sept.		June		July		Aug.		Sept.	
	A	B	A	B	A	B	A	B	A	B	A	B	A	B
Monogeneans	72.2	0.0	66.7	42.8	77.7	61.1	77.8	25.0	94.4	0.0	83.3	22.2	88.9	33.3
Flagellates	5.6	0.0	11.1	0.0	5.6	5.6	5.6	0.0	5.6	0.0	5.6	0.0	0.0	0.0
Trichodina	5.6	18.2	0.0	0.0	0.0	11.1	5.6	0.0	5.6	0.0	0.0	11.1	0.0	0.0
Scyphidia	11.1	0.0	0.0	0.0	0.0	5.6	0.0	0.0	0.0	0.0	61.1	0.0	11.1	5.6
Glossatella	72.2	63.6	66.7	33.3	16.7	33.3	88.9	100.0	50.0	75.0	61.1	55.6	83.3	50.0
Spores*	0.0	0.0	5.6	4.8	11.1	5.6	0.0	0.0	11.1	0.0	0.0	11.1	11.1	0.0
Unid. Ciliates	0.0	45.5	0.0	14.3	0.0	5.6	5.6	50.0	16.7	25.0	0.0	11.1	5.6	16.7

* Probably of the protozoan order Myxosporidia.

the present study (Lewis et al. 1977).

Ectosymbionts of Green Sunfish

Of the 225 green sunfish examined, 224 (99.5%) harbored at least one ectosymbiont. No consistently significant ($\alpha = .05$) differences in relative densities of taxa between locations were found (Table 7). However, in five of the seven monthly samples the relative densities of monogeneans were greater at location A than at B and two of these five differences were statistically significant ($\alpha = .05$). In 1981 the relative density of Trichodina was higher at location B, but in 1982 it was higher at location A. Neither of these differences was statistically significant ($\alpha = .05$). The holding times were longer in 1981 (Table 8), which may account for the differences in relative density between the two locations.

Epistylis was more abundant on green sunfish at location A than at location B throughout the study, but again none of these differences were significant ($\alpha = .05$). This abundance pattern is the reverse of what might be expected. Because Epistylis is a bacteriovore, location B which receives sewage effluent might be expected to have higher concentrations of potential food, bacteria. However, low flow and the possible presence of chlorine at location B may be responsible for the differences observed between the two locations.

As percent of the total ectosymbiont load per fish, monogeneans and Trichodina combined varied from 57% to 99% dominance in the 14 monthly location means for Lepomis (Table 9). This observation is consistent with the report that a single species of monogenean may comprise more than 80% of the total parasite load on Lepomis gibbosus (Hanek and Fernando 1978a). Tripartiella and Epistylis, individually, comprised

Table 7. *Lepomis* ectosymbiont relative densities and standard deviations.

Location	1981						1982							
	May		June-July		Sept.		June		July		Aug.		Sept.	
	A	B	A	B	A	B	A	B	A	B	A	B	A	B
Monogeneans	11.4*	4.3	23.1	13.1	24.9*	8.6	25.4*	9.8	20.9	9.8	16.1	22.9	9.6	22.6
S.D.	5.3	3.5	10.2	12.5	18.0	7.1	10.3	4.8	11.4	5.6	8.7	9.4	5.9	22.6
Flagellates	0.0	0.2	0.0	7.3	0.0	0.0	0.4	8.0	0.0	4.7	0.0	0.0	0.0	6.2
S.D.	0.0	0.4	0.0	24.1	0.5	0.0	0.0	0.9	31.5	0.0	14.9	0.0	0.0	13.9
Trichodina	24.2	305.6	54.8	72.7	39.0	50.9	25.8	22.6	26.4	0.6	12.5	1.8	6.5	5.0
S.D.	28.6	573.1	67.8	115.7	96.3	67.2	41.4	55.0	36.2	0.8	22.0	2.6	15.0	7.3
Tripartiella	1/	1/	1/	1/	4.7	0.0	0.1	0.8	0.1	0.0	10.2	9.7	3.5	125.0
S.D.					10.6	0.0	0.2	3.1	0.5	0.0	23.8	20.2	5.8	172.5
Epistylis	15.9	0.1	5.6	0.2	5.9	1.6	22.4	14.4	23.2	0.0	27.9	4.8	1.7	0.0
S.D.	37.9	0.3	23.8	0.9	15.1	6.6	33.6	25.3	29.5	0.0	51.8	20.0	6.8	0.0
Scyphidia	0.0	0.2	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.1	0.0	0.0
S.D.	0.0	0.6	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.2	0.0	0.0
Ichthyophthirius	0.2	0.6	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
S.D.	0.4	0.6	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Glossatella	16.7	0.3	0.1	4.3	0.7	0.4	4.3	16.6	8.8	0.1	1.9	6.7	0.2	0.6
S.D.	41.1	0.7	0.3	18.1	1.8	1.2	9.5	50.4	33.0	0.5	4.8	20.1	0.5	1.2
Trichophrya	0.3	0.6	0.0	0.0	0.0	0.0	0.1	70.1	0.1	0.1	0.0	0.0	0.0	0.0
S.D.	1.4	0.6	0.0	0.0	0.0	0.0	0.3	249.1	0.5	0.3	0.0	0.0	0.0	0.0

Table 7. Continued.

Location	1981						1982							
	May		June-July		Sept.		June		July		Aug.		Sept.	
	A	B	A	B	A	B	A	B	A	B	A	B	A	B
Spores[2/]	0.0*	51.8	21.7	93.1	19.1	16.3	13.0	0.0	0.0	34.4	5.3	0.0	7.8	0.0
S.D.	0.0	148.1	44.3	120.3	81.1	51.8	34.6	0.0	0.0	137.5	17.2	0.0	24.9	0.0
Ciliates	0.0	0.7	2.6	0.4	0.3	0.4	0.2	0.4	0.3	0.0	0.0	0.2	0.4	0.0
S.D.	0.0	1.6	7.9	1.4	1.0	1.2	0.6	1.1	0.6	0.0	0.0	0.0	1.0	0.0
Totals	68.5	363.2	107.9	191.1	94.7	78.9	91.3	135.1	87.8	45.0	78.6	46.2	29.7	164.4

* Denotes a significant difference between locations.

1/ Tripartiella included with Trichodina in these samples.

2/ Probably of the protozoan order Myxosporidia.

Table 8. Host condition parameters and ectosymbiont diversity parameters for Lepomis, means and standard deviations.

	1981						1982							
	May		June-July		Sept.		June		July		Aug.		Sept.	
	A	B	A	B	A	B	A	B	A	B	A	B	A	B
Length (mm)	63.9*	83.6	94.4	99.5	81.8	95.8	83.3	71.2	71.2	70.8	65.3	59.4	68.6*	47.2
S.D.	16.1	7.6	26.5	17.5	18.7	23.8	14.9	11.7	14.8	19.6	9.4	10.4	6.1	5.0
Weight (gm)	4.2	9.1	14.2	14.8	8.9	14.0	9.8	5.9	5.8	6.0	4.2	3.0	4.6	1.5
S.D.	3.2	2.6	10.2	6.4	8.6	8.1	4.6	3.1	3.7	5.3	2.0	1.7	1.3	0.4
Condition factor	2.2	2.5	2.4	2.4	2.2	2.1	2.44	2.79	2.4*	2.2	2.3	2.2	2.3	2.3
S.D.	0.2	0.2	0.4	0.3	0.2	0.5	0.2	2.08	0.1	0.2	0.2	0.1	0.1	0.2
Age	1.2	1.1	1.9	1.7	1.7	1.2	1.8*	1.1	1.2	1.4	1.2	1.0	1.0	1.0
S.D.	0.4	0.3	0.7	0.6	0.7	0.7	0.5	0.3	0.4	0.6	0.4	0.3	0.0	0.0
Taxa/fish	3.1	2.7	2.6	2.4	2.6	2.2	3.4	3.7	3.4*	1.6	3.5*	2.5	2.4	2.6
S.D.	0.4	1.1	0.5	0.8	1.6	0.8	1.2	1.3	1.1	0.5	1.5	1.4	1.4	1.5
Diversity	1.1*	0.4	0.9	0.7	0.7	0.6	1.1	1.1	1.2*	0.3	1.1*	0.7	0.8	0.5
S.D.	0.4	0.4	0.3	0.4	0.7	0.4	0.5	0.6	0.5	0.4	0.6	0.6	0.7	0.5
Number fish exam.	18	10	18[1/]	19	18[2/]	18[2/]	18	13	18	16	18	18	18	5

* Denotes a significant ($\alpha = 0.05$) difference between locations.

1/ Weight obtained for only 17 of these 18 fish. 2/ Age obtained for only 17 of these 18 fish.

Table 9. Ectosymbiont dominance as percent of the total organism load per individual Lepomis.

Location	1981						1982							
	May		June-July		Sept.		June		July		Aug.		Sept.	
	A	B	A	B	A	B	A	B	A	B	A	B	A	B
Monogeneans	32.5%	22.2	45.3	41.6	63.9	27.4	49.9	35.8	41.0	90.7	43.6	68.4	67.8	61.6
Trichodina	34.3	76.0	49.0	55.3	23.6	68.5	23.6	22.9	28.1	8.4	18.0	3.5	15.7	3.1
Tripartiella	1/	1/	1/	1/	6.7	010	0.1	0.6	0.2	0.0	9.6	14.9	9.8	35.2
Epistylis	16.3	0.0	3.1	0.1	3.4	2.7	21.7	15.3	23.5	0.0	26.0	3.6	4.6	0.0
Scyphidia	0.0	0.7	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.1	0.0	0.0
Glossatella	15.8	0.5	0.2	1.7	1.5	0.6	4.1	14.3	6.2	0.7	2.9	910	0.6	0.1
Trichophrya	1.0	0.0	0.0	0.0	0.0	0.0	0.1	10.7	0.5	0.3	0.0	0.0	0.0	0.0
Ciliates	0.0	0.6	2.4	1.3	0.8	0.9	0.4	0.4	0.5	0.0	0.0	0.5	1.6	0.0

1/ Tripartiella included with Trichodina in these samples.

more than 20% of the total load on occasions but neither exceeded 35.2%. The community inhabiting green sunfish was typically made up of monogeneans and Trichodina, although other taxa frequently were important. However, no statistically significant ($\alpha = .05$) pairwise relationships were detected between taxa as measured by correlation coefficients.

Diversity values were lower than those previously reported by Cloutman (1975) for several fish species, and there were no consistently significant ($\alpha = .05$) differences between locations A and B (Table 8). However, the diversity at location A was higher than at B for 6 of the 7 monthly samples and significantly higher at 3 of these 6. Similarly, the number of taxa per fish was higher at location A in 5 of the 7 monthly samples, and significantly different in 2 of these 5. Green sunfish from location A seemed to support a higher diversity of ectosymbionts than those from B. Perhaps the organisms encountered were not tolerant of the water quality at location B despite the probable food supply for bacterivores. Chlorine, a common sewage disinfectant, has been shown to be toxic to protozoans (Cairns and Plafkin 1975). This toxicity combined with a much greater water flow at location B, may have been responsible for the lower diversity found there.

Prevalence of each taxon was also calculated for the ectosymbionts on green sunfish. Monogeneans attained 100% prevalence in 3 of the 6 monthly location means in 1981 and in all 8 such means from 1982 (Table 10). Trichodina was 100% prevalent in 1 location mean in 1981 and 2 in 1982 (Table 10). Both these taxa contain species which have been reported at greater than 90% prevalence (Chappel 1969; Boxrucker 1979). In contrast with mosquitofish, individual prevalence of three other

Table 10. Prevalence of ectosymbionts on Lepomis expressed as percentage of fish infected.

Location	1981						1982							
	May		June-July		Sept.		June		July		Aug.		Sept.	
	A	B	A	B	A	B	A	B	A	B	A	B	A	B
Monogeneans	88.9	90.0	100.0	100.0	88.9	100.0	100.0	100.0	100.0	100.0	100.0	100.0	100.0	100.0
Flagellates	0.0	20.0	0.0	10.5	5.6	0.0	0.0	23.1	11.1	0.0	16.7	0.0	0.0	20.0
Trichodina	94.4	90.0	100.0	89.5	61.1	83.3	94.4	100.0	100.0	43.8	83.3	50.0	22.2	60.0
Tripartiella	1/	1/	1/	1/	27.8	0.0	5.6	7.7	5.6	0.0	50.0	33.3	38.9	60.0
Epistylis	33.3	10.0	5.6	5.3	22.2	11.1	66.7	53.8	72.2	0.0	61.1	11.1	11.1	0.0
Scyphidia	0.0	10.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Ichthyophthinius	16.7	10.0	0.0	0.0	0.0	5.6	0.0	0.0	0.0	0.0	0.0	5.6	0.0	0.0
Glossatella	72.2	10.0	11.1	15.8	22.2	16.7	55.6	46.2	27.8	6.3	38.9	44.4	11.1	20.0
Trichophrya	0.0	0.0	0.0	0.0	0.0	0.0	11.1	30.8	5.6	6.3	0.0	0.0	0.0	0.0
Spores2/	0.0	30.0	33.3	42.1	5.6	5.6	16.7	0.0	0.0	6.3	11.1	0.0	11.1	0.0
Unid. Ciliates	0.0	30.0	44.4	15.8	16.7	16.7	11.1	15.4	16.7	0.0	0.0	5.6	22.2	0.0

1/ Tripartiella included with Trichodina in these samples.

2/ Probably of the protozoan order Myxosporidia.

taxa, Tripartiella, Epistylis, and Glossatella, was at least 50% in at least one monthly location mean during the study. Prevalence of four of the remaining 6 groups found on green sunfish reached 20% in at least one such sample.

Ectosymbionts: Host and Nonhost Influences

The effects of various biotic and abiotic factors on hosts and their parasite populations have previously been investigated (Esch et al. 1975, Snieszko 1974). Although a decrease in parasite populations can result when the host is subjected to stress, an increase in parasite numbers is typically expected. This expectation is based on the assumption that the hosts' defenses are diminished by exposure to a stressor (Ewing et al. 1982; Snarski 1982; Roales and Perlmutter 1977, 1980). However, for this study no such reasoning can be introduced because none of the organisms encountered except Ichthyophthirius and spores,probably of the protozoan order Myxosporidia, were subjected to the defense mechanisms associated with the interior of the host.

In this study the ectosymbiont's environment is considered from the standpoint of two major components: the water and the host. Although many of the taxa ecountered were bacteriovores, none of the groups exhibited consistently greater density or prevalence at location B, an area receiving sewage effluent and suspected of having a higher concentration of food organisms, bacteria. Low water flow at location A as compared to B and the possible contamination of location B with chlorine, a sewage disinfectant, are suggested explanations for these findings.

The effects of the second environmental component, the host, are

also important. In this study several aspects of the host are considered: age, length, weight, condition, defense mechanisms employed by the host, and the condition of the host's integument. Many studies have correlated parasite densities and prevalence with host age and length. In this study no consistently significant correlations were found between ectosymbiont population parameters and these variables. Neither were weight nor condition factor found to be correlated positively with ectosymbiont parameters.

The likely most important defense operative in this study was the hosts' mucus secretions. Internal aspects of the fishes' immune system are probably not important here because the organisms encountered rarely come into contact with the internal body fluids of their fish hosts. Fish from both locations appeared to have normal amounts of mucus, providing no insight as to the ectosymbiont occurrence and infestation levels observed.

The hosts' epithelium, specifically gill tissue, is the attachment site for many of the organisms such as the monogeneans and a majority of the protozoan taxa encountered. Several authors have reported that poor water quality can cause hyperplasia, fused and swollen lamellae, clubbing of filaments, excess mucus production, and sloughing of epithelial cells (Abel 1974; Eller 1975; Bass and Heath 1975, 1977; Bass et al. 1977; Skinner 1982). However, the gills of the hosts examined, with few exceptions, appeared to be free of gross abnormalities which might interfere with ectosymbiont attachment. Consequently, the condition of the hosts' integument was judged to be not important in limiting organism attachment.

Another factor which may have been important in regulating

ectosymbiont numbers was host availability. If more suitable hosts are available, then there is a greater probability of ectosymbiont offspring finding a suitable host. The sampling goal of six fish (hosts) per site was attained for both hosts in every sample at location A but it was frequently impossible to attain this goal at location B for either host species (Tables 5 and 7). Lower probability of encounter between susceptible host and ectosymbiont was lower at location B would decrease the probability of a ectosymbiont population sustaining itself.

Additionally, other species of fish encountered at location A in moderately large numbers included bluegill and longear sunfish, both close relatives of green sunfish. If the ectosymbionts found on green sunfish are not species-specific, then the number of suitable hosts available may be very large at location A. In contrast only red shiners were encountered in large numbers at location B, and then only in early summer of 1982 during their spawning migration. Red shiners are not closely related to mosquitofish or green sunfish and are not permanent residents of the sample sites. Therefore they probably did not serve as a reservoir for ectosymbionts at location B.

CHAPTER VI

SUMMARY AND CONCLUSIONS

During the summers of 1981 and 1982 211 Gambusia affinis and 225 Lepomis cyanellus were collected from two locations on Stillwater Creek in northcentral Oklahoma. There were three known major differences between the two locations. Location B received urban runoff and treated sewage effluent and always had measurable flow, and location A received no urban runoff or treated sewage effluent and usually exhibited no measurable flow.

Four gill arches were removed from each fish and examined for the numbers and taxa of ectosymbionts. Seven taxa were associated with Gambusia, and eleven taxa were associated with Lepomis.

Monogeneans and Glossatella were the dominant taxa on Gambusia and constituted at least 70% of the community in all samples. The Lepomis community was more complex. Monogeneans and Trichodina were generally dominant but Epistylis, Tripartiella, and Trichophrya were occasionally numerous. In addition three organisms occurred on Lepomis which were never recovered from Gambusia: Tripartiella, Trichophrya, and Epistylis. During this study the Gambusia community was overwhelmingly dominated by two taxa, but the Lepomis community was more often composed of three or four equally numerous taxa.

No consistently significant relationships were detected among the ectosymbiont taxa as measured by correlation coefficients. Similarly,

no consistent relationships were noted between ectosymbionts and temperature or dissolved oxygen concentrations.

Neither host exhibited consistently significant location differences in relative density of the total ectosymbiont population or in relative density of each taxa individually. On Gambusia, however, monogeneans, unidentified ciliates, and flagellates were generally more abundant at location A; and on Lepomis, monogeneans and Epistylis were generally more abundant at location A. Because suitable food is likely more available at location B, Epistylis and monogeneans were expected to be more abundant there. However, differences in stream flow or water chemistry or both probably resulted in greater abundance at location A.

Only Glossatella and monogeneans were observed to have high prevalence on Gambusia. Glossatella prevalence was generally 50% or higher and was the only organism to attain 100% prevalence in at least one monthly location mean for Gambusia. Monogeneans were always in excess of 65% at location A but never exceeded 45% at location B. Again, water currents and chemistry are suggested as possible explanations for the apparent contradiction of location preferance for monogeneans.

Trichodina and monogeneans both attained a prevalence of 100% on green sunfish during this study. Trichodina was 100% prevalent in 3 of 14 monthly location samples and monogeneans were 100% prevalent in 11 of 14 monthly location samples. Tripartiella, Epistylis, and Glossatella were recovered on at least 50% of the hosts examined in at least one location/month for green sunfish.

Stream flow and pollution may be responsible for the ectosymbiont community differences noted between locations. For the most part these

differences were inconsistent and seldom statistically significant. Nevertheless, the observed differences in ectosymbiont diversity seem to indicate that the conditions which exist at location B do represent a stress on the biotic community, which may be biologically significant, validating the continued research into the use of these organisms in biological monitoring programs. Much work remains to be done, however, in documenting the ecological relationships and standardizing the identification of the many ectosymbionts which inhabit the gills of fish before these creatures can be employed reliably as biological indicators.

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APPENDIX A

TEMPERATURE (°C) DATA BY YEAR, MONTH, AND SITE

Table 11. Temperature (°C) levels for 1981 and 1982 by year, month, and site.

	1981			1982			
	May	June-July	Sept.	June	July	Aug.	Sept.
Site 1	20.4	27.1	24.0	23.0	29.0	29.5	21.2
Site 2	19.7	29.8	20.5	23.5	29.0	27.1	21.2
Site 3	19.5	29.8	20.4	25.5	28.2	27.5	23.0
Site 4	22.8	27.0	26.8	20.9	30.0	24.0	18.5
Site 5	22.8	26.7	24.6	21.2	29.8	24.8	19.0
Site 6	22.5	27.0	24.2	22.0	30.5	25.0	20.0

APPENDIX B

DISSOLVED OXYGEN (mg/l) DATA BY YEAR, MONTH, AND SITE

Table 12. Dissolved oxygen (mg/l) data by year, month, and site.

	1981			1982			
	May	June-July	Sept.	June	July	Aug.	Sept.
Site 1	4.5	9.5	6.2	4.4	2.8	4.1	5.9
Site 2	2.0	3.5	3.9	5.8	4.6	5.0	7.4
Site 3	2.6	5.5	4.2	4.2	6.1	5.9	7.0
Site 4	2.8	2.8	5.2	5.8	10.4	5.1	5.5
Site 5	3.5	2.6	3.9	5.9	12.2	4.9	5.1
Site 6	3.9	2.6	4.1	5.9	11.6	4.8	5.6

APPENDIX C

DATA MEANS FOR *LEPOMIS* BY YEAR, MONTH, AND SITE

Table 13. *Lepomis* data means for May 1981 by site.

	Site					
	1	2	3	4	5	6
Length (mm)	62.5	58.2	71.2	81.6	-	85.6
Weight (gm)	3.9	3.3	5.6	8.5	-	9.7
Condition factor	2.4	2.1	2.2	2.5	-	2.5
Age (yrs.)	1.0	1.2	1.3	1.2	-	1.0
Holding time (hrs.)	40.0	39.7	40.3	73.4	-	96.0
Taxa/fish	3.3	3.2	2.7	2.8	-	2.6
Diversity	1.3	1.1	0.9	0.2	-	0.5
No. fish examined	6	6	6	4	0	5
Trematodes	11.2	11.3	11.8	2.2	-	6.4
Flagellates	0.0	0.0	0.0	0.2	-	0.2
Trichodina	44.8	19.3	8.3	514.2	-	97.0
Tripartiella[1]	-	-	-	-	-	-
Epistylis	9.2	7.5	31.2	0.2	-	0.0
Scyphidia	0.0	0.0	0.0	0.0	-	0.4
Ichthyophthirius	0.2	0.2	0.2	0.0	-	0.0
Glossatella	35.5	12.2	2.5	0.4	-	0.2
Trichophrya	0.0	0.0	1.0	0.0	-	0.0
Spores	0.0	0.0	0.0	102.0	-	1.6
Ciliates	0.0	0.0	0.0	0.4	-	1.0
Total organisms	100.9	50.5	55.5	619.6	-	106.8

[1] *Tripartiella* included with *Trichodina*.

Table 14. *Lepomis* data means for June-July 1981 by site.

	Site					
	1	2	3	4	5	6
Length (mm)	91.5	90.4	99.7	94.2	109.3	95.7
Weight (gm)	12.5	11.7	17.9	12.9	19.0	12.9
Condition factor	2.5	2.3	2.6	2.4	2.4	2.4
Age (yrs.)	2.0	1.8	2.0	1.7	1.8	1.7
Holding time (hrs.)	44.0	61.6	64.0	84.8	56.2	115.9
Taxa/fish	2.5	2.6	2.7	2.3	2.0	2.7
Diversity	0.8	0.8	1.0	0.9	0.5	0.8
No. fish examined	6	5	7	6	6	7
Monogeneans	19.8	21.4	27.0	5.8	8.5	23.3
Flagellates	0.0	0.0	0.0	6.3	16.7	0.0
Trichodina	74.0	54.8	38.3	15.2	118.8	82.6
Tripartiella[1]	-	-	-	-	-	-
Epistylis	16.8	0.0	0.0	0.0	0.0	0.6
Scyphidia	0.0	010	0.0	0.0	0.0	0.0
Ichthyophthirius	0.0	0.0	0.0	0.0	0.0	0.0
Glossatella	0.2	0.2	0.0	0.3	0.0	11.4
Trichophrya	0.0	0.0	0.0	0.0	0.0	0.0
Spores	1.5	23.8	37.6	128.3	61.5	90.0
Ciliates	0.3	0.8	5.7	0.2	0.0	1.0
Total organisms	112.6	101.0	108.6	156.1	205.5	208.9

[1] *Tripartiella* included with *Trichodina*.

Table 15. *Lepomis* data means for September 1981 by site.

	Site					
	1	2	3	4	5	6
Length (mm)	79.5	78.8	87.0	85.5	102.2	99.8
Weight (gm)	7.6	7.9	11.3	12.4	15.8	13.8
Condition factor	2.4	2.2	2.1	2.0	2.3	2.2
Age (yrs.)	1.4	1.7	2.0	1.3	1.0	1.3
Holding time (hrs.)	70.8	219.7	274.2	87.2	62.2	70.7
Taxa/fish	3.5	2.8	1.3	2.0	2.5	2.2
Diversity	1.1	0.8	0.2	0.6	0.8	0.5
No. fish examined	6	6	6	6	6	6
Monogeneans	19.7	19.0	36.0	9.0	8.3	8.3
Flagellates	0.3	0.0	0.0	0.0	0.0	0.0
Trichodina	109.0	4.3	3.7	52.0	42.8	58.0
Tripartiella	8.5	5.5	0.0	0.0	0.0	0.0
Epistylis	16.0	1.7	0.0	0.2	4.7	0.0
Scyphidia	0.0	0.0	0.0	0.0	0.0	0.0
Ichthyophthirius	0.0	0.0	0.0	0.2	0.0	0.0
Glossatella	0.3	1.8	0.0	0.3	0.0	1.0
Trichophrya	0.0	0.0	0.0	0.0	0.0	0.0
Spores	0.0	0.0	57.3	0.2	41.3	7.5
Ciliates	0.2	0.7	0.2	0.8	0.3	0.0
Total organisms	154.0	33.0	97.2	62.7	97.4	74.8

Table 16. *Lepomis* data means for June 1982 by site.

	Site					
	1	2	3	4	5	6
Length (mm)	81.5	91.3	77.0	61.0	73.2	71.0
Weight (gm)	8.8	11.7	8.8	3.0	6.8	5.5
Condition factor	2.4	2.5	2.4	2.2	3.4	2.3
Age (yrs.)	1.8	1.8	1.7	1.0	1.2	1.0
Holding time (hrs.)	21.3	28.5	23.5	6.0	9.5	32.8
Taxa/fish	3.3	3.0	4.0	3.0	3.7	3.8
Diversity	1.1	0.9	1.3	1.3	1.2	1.0
No. fish examined	6	6	6	1	6	6
Monogeneans	21.5	26.2	28.5	10.0	9.0	10.5
Flagellates	0.0	0.0	0.0	0.0	0.7	0.2
Trichodina	31.2	21.7	24.7	3.0	10.5	38.0
Tripartiella	0.0	0.0	0.2	0.0	1.8	0.0
Epistylis	9.8	9.7	47.8	0.0	22.3	8.8
Scyphidia	0.0	0.0	0.0	0.0	0.0	0.0
Ichthyophthirius	0.0	0.0	0.0	0.0	0.0	0.0
Glossatella	0.5	9.3	3.0	3.0	35.3	0.2
Trichophrya	0.0	0.3	0.0	0.0	0.0	152.0
Spores	21.7	4.8	12.5	0.0	0.0	0.0
Ciliates	0.3	0.0	0.3	0.0	0.0	0.8
Total organisms	85.0	72.0	117.0	16.0	79.6	210.5

Table 17. *Lepomis* data means for July 1982 by site.

	Site					
	1	2	3	4	5	6
Length (mm)	80.8	62.8	69.8	67.4	68.0	77.4
Weight (gm)	7.9	3.8	5.7	5.0	6.4	6.5
Condition factor	2.3	2.5	2.3	2.2	2.2	2.1
Age (yrs.)	1.4	1.0	1.2	1.2	1.7	1.2
Holding time (hrs.)	31.8	14.3	23.3	32.8	23.0	21.0
Taxa/fish	3.7	3.8	2.8	1.6	1.7	1.4
Diversity	1.4	1.4	0.9	0.2	0.5	0.2
No. fish examined	6	6	6	6	6	6
Monogeneans	28.2	16.3	18.3	12.8	6.8	10.4
Flagellates	1.7	22.3	0.0	0.0	0.0	0.0
Trichodina	52.3	22.8	4.0	0.4	1.0	0.2
Tripartiella	0.0	0.3	0.0	0.0	0.0	0.0
Epistylis	42.3	9.8	17.3	0.0	0.0	0.0
Scyphidia	0.0	0.0	0.0	0.0	0.0	0.0
Ichthyophthirius	0.0	0.0	0.0	0.0	0.0	0.0
Glossatella	0.0	24.8	1.5	0.4	0.0	0.0
Trichophrya	0.0	0.0	0.3	0.0	0.0	0.2
Spores	0.0	0.0	0.0	0.0	0.0	110.0
Ciliates	0.5	0.5	0.0	0.0	0.0	0.0
Total organisms	125.0	196.8	41.4	13.6	7.8	120.8

Table 18. *Lepomis* data means for August 1982 by site.

	Site					
	1	2	3	4	5	6
Length (mm)	63.5	62.5	70.0	66.2	57.8	54.3
Weight (gm)	3.9	3.8	5.0	4.0	2.7	2.3
Condition factor	2.5	2.3	2.3	2.2	2.1	2.2
Age (yrs.)	1.0	1.3	1.3	1.2	1.0	0.8
Holding time (hrs.)	20.3	17.3	31.8	14.7	24.3	14.0
Taxa/fish	3.3	3.3	3.8	2.7	2.0	2.8
Diversity	1.0	1.1	1.3	0.7	0.5	0.9
No. fish examined	6	6	6	6	6	6
Monogeneans	13.7	11.2	23.3	27.7	20.0	21.0
Flagellates	4.0	0.0	10.2	0.0	0.0	0.0
Trichodina	25.8	4.2	7.5	1.7	1.5	2.3
Tripartiella	4.0	20.5	6.2	7.2	10.0	12.0
Epistylis	45.0	19.0	19.8	14.5	0.0	0.0
Scyphidia	0.0	0.0	0.0	0.2	0.0	0.0
Ichthyophthirius	0.0	0.0	0.0	0.0	0.0	0.0
Glossatella	0.3	5.0	0.3	1.5	1.8	16.7
Trichophrya	0.0	0.0	0.0	0.0	0.0	0.0
Spores	0.0	0.0	15.8	0.0	0.0	0.0
Ciliates	0.0	0.0	0.0	0.7	0.0	0.0
Total organisms	92.8	59.9	83.1	53.5	33.3	52.2

Table 19. *Lepomis* data means for September 1982 by site.

	Site					
	1	2	3	4	5	6
Length (mm)	64.2	67.8	73.8	40.0	49.0	-
Weight (gm)	3.7	4.3	5.7	1.0	1.6	-
Condition factor	2.3	2.2	2.3	2.6	2.3	-
Age (yrs.)	1.0	1.0	1.0	1.0	1.0	-
Holding time (hrs.)	28.0	15.0	34.2	5.0	16.5	-
Taxa/fish	1.8	1.3	4.0	2.0	2.8	-
Diversity	0.5	0.3	1.5	0.4	0.5	-
No. fish examined	6	6	6	1	4	0
Monogeneans	10.7	6.8	11.3	10.0	32.0	-
Flagellates	0.0	0.0	0.0	0.0	7.8	-
Trichodina	1.8	0.3	17.3	1.0	6.0	-
Tripartiella	0.2	0.0	10.3	0.0	153.3	-
Epistylis	0.0	0.3	4.8	0.0	0.0	-
Scyphidia	0.0	0.0	0.0	0.0	0.0	-
Ichthyophthirius	0.0	0.0	0.0	0.0	0.0	-
Glossatella	0.0	0.0	0.5	0.0	0.8	-
Trichophrya	0.0	0.0	0.0	0.0	0.0	-
Spores	16.7	0.0	6.7	0.0	0.0	-
Ciliates	0.7	0.0	0.7	0.0	0.0	-
Total organisms	30.1	7.4	51.6	11.0	202.9	-

APPENDIX D

DATA MEANS FOR *GAMBUSIA* BY YEAR, MONTH, AND SITE

Table 20. *Gambusia* data means for May 1981 by site.

	Site					
	1	2	3	4	5	6
Length (mm)	39.3	36.2	38.7	45.2	-	29.7
Weight (gm)	0.6	0.5	0.6	0.9	-	0.3
Condition factor	1.4	1.4	1.4	1.6	-	1.7
Age (yrs.)	1.2	0.7	0.8	1.0	-	0.7
Holding time (hrs.)	42.7	39.5	40.5	86.4	-	83.5
Taxa/fish	1.5	1.5	2.0	1.0	-	1.7
Diversity	0.5	0.3	0.6	0.4	-	0.4
No. fish examined	6	6	6	6	0	6
Monogeneans	5.5	1.0	3.2	0.0	-	0.0
Flagellates	0.0	0.0	2.2	0.0	-	0.0
Trichodina	0.0	0.0	0.2	0.2	-	0.2
Scyphidia	0.0	0.3	1.0	0.0	-	191.8
Glossatella	6.2	9.5	8.2	4.0	-	0.0
Spores	0.0	0.0	0.0	0.0	-	0.0
Ciliates	0.0	0.0	0.0	0.2	-	9.5
Total organisms	11.7	10.8	14.8	4.4	-	201.5

Table 21. *Gambusia* data means for June-July 1981 by site.

	Site					
	1	2	3	4	5	6
Length (mm)	30.1	23.0	24.1	40.7	32.9	30.9
Weight (gm)	0.4	0.1	0.1	0.9	0.5	0.3
Condition factor	1.7	1.7	1.5	1.7	1.8	1.4
Age (yrs.)	0.3	0.3	0.2	0.9	0.6	0.4
Holding time (hrs.)	45.7	62.8	65.0	130.6	124.1	115.1
Taxa/fish	1.2	1.6	1.6	0.7	1.6	0.4
Diversity	0.1	0.5	0.5	0.0	0.5	0.0
No. fish examined	6	5	7	7	7	7
Monogeneans	2.2	4.2	1.1	1.0	1.6	0.3
Flagellates	0.0	0.0	1.7	0.0	2.3	0.0
Trichodina	0.0	0.0	0.0	0.0	0.0	0.0
Scyphidia	0.0	0.0	0.0	0.0	0.0	0.0
Glossatella	2.2	3.2	6.3	3.3	1.1	10.4
Spores	15.3	0.0	0.0	0.0	26.6	0.0
Ciliates	0.0	0.0	0.0	0.0	0.6	0.0
Total organisms	19.7	7.4	9.1	4.3	32.2	10.7

Table 22. *Gambusia* data means for September 1981 by site.

	Site					
	1	2	3	4	5	6
Length (mm)	31.3	29.8	23.5	30.0	28.0	30.5
Weight (gm)	0.3	0.2	0.1	0.3	0.2	0.3
Condition factor	1.3	1.2	1.0	1.5	1.4	1.7
Age (yrs.)	0.5	0.0	0.0	0.4	0.2	0.3
Holding time (hrs.)	233.5	315.8	287.7	80.3	75.8	91.5
Taxa/fish	0.7	1.0	1.3	1.5	1.2	1.2
Diversity	0.0	0.0	0.2	0.4	0.3	0.2
No. fish examined	6	6	6	6	6	6
Monogeneans	1.3	4.5	3.5	1.7	1.7	0.7
Flagellates	4.0	0.0	0.0	5.2	0.2	0.0
Trichodina	0.0	0.0	0.0	0.8	0.0	0.0
Scyphidia	0.0	0.0	0.0	0.0	0.0	0.2
Glossatella	0.0	0.0	5.2	0.7	11.7	8.5
Spores	0.0	370.8	0.8	0.0	0.0	0.0
Ciliates	0.0	0.0	0.0	0.2	0.0	0.0
Total organisms	5.3	375.3	9.5	8.6	13.6	9.4

Table 23. *Gambusia* data means for June 1982 by site.

	Site					
	1	2	3	4	5	6
Length (mm)	34.3	35.7	37.3	-	32.0	35.7
Weight (gm)	0.5	0.4	0.5	-	0.3	0.5
Condition factor	1.5	1.3	1.4	-	1.3	1.4
Age (yrs.)	1.0	1.0	1.0	-	1.0	1.0
Holding time (hrs.)	18.5	29.5	30.2	-	4.0	11.0
Taxa/fish	2.0	2.0	1.5	-	2.0	1.3
Diversity	0.8	0.7	0.5	-	0.9	0.1
No. fish examined	6	6	6	0	1	3
Monogeneans	6.0	2.5	2.3	-	2.0	0.0
Flagellates	0.0	0.2	0.0	-	0.0	0.0
Trichodina	0.0	0.2	0.0	-	0.0	0.0
Scyphidia	0.0	0.0	0.0	-	0.0	0.0
Glossatella	7.2	6.8	4.8	-	3.0	35.7
Spores	0.0	0.0	0.0	-	0.0	0.0
Ciliates	0.2	0.0	0.0	-	0.0	0.7
Total organisms	13.4	14.7	7.1	-	5.0	36.4

Table 24. *Gambusia* data means for July 1982 by site.

	Site					
	1	2	3	4	5	6
Length (mm)	34.5	35.2	39.0	41.0	38.7	-
Weight (gm)	0.4	0.4	0.6	1/	0.7	-
Condition factor	1.3	1.3	1.5	1/	1.8	-
Age (yrs.)	0.7	0.8	0.8	1.0	1.3	-
Holding time (hrs.)	40.8	18.7	23.0	13.0	18.7	-
Taxa/fish	1.8	1.3	1.7	0.0	1.3	-
Diversity	0.6	0.2	0.5	0.0	0.3	-
No. fish examined	6	6	6	1	3	0
Monogeneans	6.3	8.7	5.0	0.0	0.0	-
Flagellates	0.0	0.0	1.2	0.0	0.0	-
Trichodina	0.0	0.2	0.0	0.0	0.0	-
Scyphidia	0.0	0.0	0.0	0.0	0.0	-
Glossatella	6.3	0.5	1.7	0.0	1.7	-
Spores	0.0	420.8	0.0	0.0	0.0	-
Ciliates	0.5	0.0	0.0	0.0	0.3	-
Total organisms	13.1	430.2	7.9	0.0	2.0	-

1/Only one fish was collected at site 4 and no weight was obtained for it.

Table 25. *Gambusia* data means for August 1982 by site.

	Site					
	1	2	3	4	5	6
Length (mm)	31.0	23.3	27.2	29.2	33.5	21.0
Weight (gm)	0.3	0.1	0.2	0.2	0.3	0.1
Condition factor	1.3	1.4	1.3	1.3	1.1	1.2
Age (yrs.)	0.7	0.0	0.0	0.3	0.5	0.0
Holding time (hrs.)	20.3	18.3	32.8	20.3	18.0	24.0
Taxa/fish	2.0	2.0	1.8	0.8	1.0	2.0
Diversity	0.7	0.6	0.3	0.3	0.0	0.9
No. fish examined	6	6	6	6	2	1
Monogeneans	3.0	8.0	1.2	0.2	0.0	1.0
Flagellates	0.0	55.8	0.0	0.0	0.0	0.0
Trichodina	0.0	0.0	0.0	0.2	0.0	0.0
Scyphidia	0.7	0.0	2.7	0.0	0.0	0.0
Glossatella	6.0	17.5	34.0	72.5	148.0	2.0
Spores	0.0	0.0	0.0	1.2	0.0	0.0
Ciliates	0.0	0.0	0.0	0.2	0.0	0.0
Total organisms	9.7	81.3	37.9	74.3	148.0	3.0

Table 26. *Gambusia* data means for September 1982 by site.

	Site					
	1	2	3	4	5	6
Length (mm)	35.2	26.7	27.5	27.7	28.7	26.7
Weight (gm)	0.4	0.1	0.2	0.2	0.2	0.2
Condition factor	1.4	1.2	1.3	1.4	1.4	1.5
Age (yrs.)	0.3	0.0	0.0	0.5	0.0	0.2
Holding time (hrs.)	20.3	18.5	31.8	23.7	19.0	19.2
Taxa/fish	1.8	2.2	1.7	1.7	0.8	0.7
Diversity	0.6	0.8	0.6	0.5	0.1	0.0
No. fish examined	6	6	6	6	6	6
Monogeneans	1.7	3.0	2.2	1.3	2.0	0.0
Flagellates	0.0	0.0	0.0	0.0	0.0	0.0
Trichodina	0.0	0.0	0.0	0.0	0.0	0.0
Scyphidia	0.2	0.3	0.0	0.0	0.0	0.2
Glossatella	2.8	11.0	11.3	11.3	3.3	5.0
Spores	500.0	0.0	333.3	0.0	0.0	0.0
Ciliates	0.3	0.0	0.0	9.0	0.0	0.0
Total organisms	505.0	14.3	346.8	21.6	5.3	5.2

VITA 2

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