PREVALENCE AND INTENSITY OF *EPISTYLIS* ON FISHES FROM TWO NORTH CAROLINA RESERVOIRS

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Abstract: Epistylis infestations occurred on 16 of 32 fish species collected from Mountain Island Lake and Lake Norman, North Carolina, during 1974 and 1975. Percichythids, centrarchids, and ictalurids were the fishes most commonly infested, while infestations were rare on clupeids, cyprinids, and percids. Epistylis was generally more prevalent on fishes during fall (September through November). Percichthyids, followed by centrarchids, had the highest intensity of infestation. Intensity of infestation increased with surface area and length of percichthyids, but not with that of centrarchids. Although the attachment site varied among fish species, Epistylis on fishes did not correlate with the abundance of Epistylis in the plankton. Dissolved orthophosphate, total phosphorus, alkalinity, and ammonia concentrations were positively correlated with the prevalence of Epistylis on white bass (Morone chrysops), redbreast sunfish (Lepomis auritus), bluegill (L. macrochirus), and largemouth bass (Micropterus salmoides). Total iron and dissolved oxygen concentrations were negatively correlated. Although temperature did not correlate with the prevalence of Epistylis was not found on fishes at water temperatures below 14 C.

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Epistylis, a stalked ciliated protozoan, is common in waters of the southeastern U.S. (Rogers 1971). *Epistylis* often attaches to the bony parts of fishes, and the attachment site is commonly accompanied by an inflamed lesion and erosion of spines or scales. As a result of these lesions, fishes are often rejected by fishermen. Very few fish mortalities have been attributed to *Epistylis*, but secondary bacterial infections by *Aeromonas hydrophila* were considered th ecause of several fish kills in North Carolina (Plumb 1973, Miller and Chapman 1977). Factors affecting the occurrence of *Epistylis* are unknown, but Rogers (1971) hypothesized that organic enrichment enhanced epizootics of *Epistylis*.

The objectives of this study were: 1) to determine the host specificity of *Epistylis*, 2) to characterize the seasonal abundance of *Epistylis* on fishes and relate its abundance to various water quality variables, 3) to determine characteristic areas of attachment on fishes, and 4) to define the relationship between intensity of *Epistylis* infestation to physical and biological characteristics of the host species.

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STUDY AREAS

Six study areas were located on Lake Norman and Mountain Island Lake of the Catawba River system in North Carolina. Mountain Island Lake and Lake Norman are warm monomictic reservoirs formed, respectively, in 1924 and 1963 for producing hydroelectric power and providing cooling water for steam electric stations. Lake Norman has a surface area of 13,517 ha and releases into Mountain Island Lake which has a surface area of 1,309 ha.

A lower lake cove (Station 4), a mid-lake cove (Station 10), an upper lake cove (Station 17), and the heated discharge of Marshall Steam Station (Station 14) were sampled on Lake Norman. The intake canal (Station 278) and the discharge canal (Station 276) of Riverbend Steam Station were sampled on Mountain Island Lake.

METHODS

Fish, plankton, and water chemistry samples were collected monthly at each station from September 1974 through December 1975. Fish were sampled using electrofishing gear and experimental gill nets. All fishes were examined externally for *Epistylis*. Most fishes with suspected infestations were measured, weighed, sexed, aged, and the location of infestation noted. Surface dimensions of suspected infestations were measured. Infestations were verified by microscopic examination of skin scraping. A ratio of the size of the infestation to the surface area of the fish provided a measure of intensity of infestation. The formulae for the surface area of perchichthyids and centrarchids were given by Olmsted, Sill, and Lewis (Unpublished manuscript, Environmental Sciences Unit, Duke Power Company, Huntersville, North Carolina).

Two, 1,000-1 plankton samples were collected at each station using a 0.2 m⁸/ gasoline powered pump. Each sample, taken from a depth of 0.3 m and a distance of 5 m from shore, was filtered through a 76 μ m mesh plankton net. The samples were concentrated to 50 ml and a 1 ml subsample microscopically examined for the abundance of *Epistylis*.

Data were collected for the following water quality variables: temperature, dissolved oxygen, pH, specific conductance (25 C), turbidity, alkalinity, hardness, nitrate-nitrite, ammonia, kjeldahl nitrogen, total iron, dissolved orthophosphate, total phosphorus, total organic carbon, and biochemical oxygen demand.

Many of the data were not amenable to analysis using parametric statistics. Prevalence data (percentage of the population infested) could not be normalized, but a transformation for the Neyman Type A distribution (Beall 1964) reduced the heterogeneity of variances. Homogeneity of variances was tested by a method described by Levene ((1960), cited by Keppel 1973). The prevalence of *Epistylis* on redbreast sunfish, bluegill, and largemouth bass was then compared among stations and seasons using a two-way analysis of variance. When analysis of variance results were significant, station and season means were compared using the least significant difference test (Keppel 1973).

The association between the prevalence of *Epistylis* on white bass, redbreast sunfish, bluegill, and largemouth bass and water quality variables was tested using productmoment correlation coefficients. Stepwise multiple regression techniques were used to construct models to attempt to explain the variation in *Epistylis* prevalence with water quality variables.

The attachment sites of *Epistylis* on fishes were compared between species using chi-square contingency tables.

Intensity of infestation data were normalized using a log transformation. Productmoment correlation coefficients were calculated for intensity of infestation with total surface area of the fish, total srength of the fish, age, and relative condition factor. Intensity of infestation data, collected during fall 1975, were compared among stations and months using one-way analysis of variance.

RESULTS

Host Specificity and Prevalence

Epistylis colonies were confirmed from 16 of the 32 fish species collected from Mountain Island Lake and Lake Norman (Table 1). Percichthyids, centrarchids, and ictalurids were most commonly infested; infestations were rare on clupeids, cyprinids, and percids.

Table 1.	The number of fish	es examined for	Epistylis	and infested	1 with Epistyli	s from
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(Etheostoma fusiforme) 2 50.0		193	1.6	64	0.0	
	darter	0	50.0			
		z	50.0			
(Estheostoma olmstedi) 4 0.0		4	0.0			
(Estheostoma olmstedi) 4 0.0 Yellow perch	- /	4	0.0			
(Perca flavescens) 547 0.2 23		K47	0.9	99	0.0	

Table 1 - Continued

White bass, striped bass, redbreast sunfish, bluegill, and largemouth bass were collected in sufficient numbers to inspect seasonal abundance of *Epistylis*. *Epistylis* was generally more prevalent on fishes during fall (September through November) (Table 2); no cases were observed during January, March, or April, and only a few during the remaining months. Results of chi-square contingency tables on the prevalence of *Epistylis* on fishes during fall 1975 indicated that *Epistylis* was more prevalent on percichthyids than centrarchids ($X^2 = 257$, df = 4, P < 0.0001), and that the prevalence of *Epistylis* was similar among species of the same family (centrarchids: $X^2 = 3.8$, df = 2, P > 0.10; perchichythyids: $X^2 = 1.3$, df = 1, P > 0.10).

Epistylis was common on white bass and striped bass collected during fall. The prevalence of *Epistylis* on white bass appeared higher at Station 14, but statistical comparisons were not made because very few white bass were collected at locations other than Station 14 (Table 2). The prevalence of *Epistylis* on striped bass was lower at the heated discharge station (Station 14) than other stations (Table 2).

		Lake Norman Stations					Mountain Island Lake Sections						
			4		10		14		17	2	276		278
Species	Season	%	, (N)	%	(N)	%	, (N)	%	(N)	%	(N)	%	(N)
White bass	Fall 1974	10	(10)		-	70	(10)	-	-			_	
	Winter 1975	0	(3)		-	0	(14)	0	(3)		-		-
	Spring 1975	0	(1)	0	(2)	0	(61)	0	(1)				-
	Summer 1975	0	(1)	0	(1)	50	(4)	-	-				
	Fall 1975	17	(6)		-	84	(45)	88	(8)		-		-
Striped bass	Fall 1974	39	(18)		-		-	-	-		_		
	Winter 1975		-		-		-	•	-		-		
	Spring 1975	0	(5)		-	0	(1)	•	-		-		-
	Summer 1975		-		-		-	-			-		-
	Fall 1975	80	(5)			46	(24)	100	(2)	100	(1)		-
Bluegill	Fall 1974	0	(50)	2	(180)	0	(29)	3	(231)	0	(56)	0	(12)
	Winter 1975	0	(1)	0	(51)	0	(100)	0	(38)	0	(41)		-
	Spring 1975	0	(36)	0	(56)	0	(55)	0	(80)	0	(17)	0) (3)
	Summer 1975	0	(23)	0	(68)	1	(168)	0	(159)	1	(76)	0) (10)
	Fall 1975	7	(14)	6	(180)	4	(228)	3	(263)	5	(38)	2	(82)
Redbreast sunfish	Fall 1974	0	(111)	0	(86)	0	(18)	10	(10)	4	(116)	12	(90)
	Winter 1975	0	(5)	0	(15)	0	(21)	0	(2)	0	(49)	0	(8)
	Spring 1975	0	(57)	0	(12)	0	(9)	0	(5)	0	(15)	0) (21)
	Summer 1975	0	(82)	0	(19)	7	(15)	0	(30)	1	(67)	6	i (99)
	Fall 1975	4	(48)	20	(35)	11	(28)	11	(37)	5	(82)	5	(136
Largemouth bass	Fall 1974	0	(6)	17	(23)	23	(13)	0	(17)	10	(20)	8	3 (13)
	Winter 1975	0	(1)	0	(26)	5	(59)	0	(19)	0	(111)	0) (7)
	Spring 1975	0	(22)	0	(48)	3	(60)	0	(41)	0	(222)	0) (11)
	Summer 1975	13	(8)	3	(31)	7	(41)	0	(32)	8	(38)	0) (29)
	Fall 1975	8	(13)	8	(48)	4	(171)	4	(94)	5	(120)	2	(43

Table 2. Prevalence (%) of *Epistylis* on fishes at stations on Lake Norman and Mountain Island Lake, North Carolina.

The temporal occurrence of *Epistylis* on redbreast sunfish and bluegill was similar, with the majority of the cases occurring during fall (Table 2). During summer, 2 cases were observed on bluegill and 7 cases were observed on redbreast sunfish. All of these cases occurred at either the heated discharge on Lake Norman (Station 14) or the 2 stations on Mountain Island Lake. Despite the extended peaks, no significant differences in the prevalence of *Epistylis* were observed among stations during any season for either redbreast sunfish ($\mathbf{F} = 1.1$; df = 5, 58; $\mathbf{P} > 0.10$) or bluegill ($\mathbf{F} = 1.4$; df = 5, 58; $\mathbf{F} > 0.10$). Significant differences (redbreast sunfish: $\mathbf{F} = 20.4$; df = 4, 58; $\mathbf{P} < 0.001$; bluegill: $\mathbf{F} = 19.5$; df = 4, 58; $\mathbf{P} < 0.001$) were observed among seasons with infestations being more prevalent during fall. No interaction was detected ($\mathbf{P} > 0.10$) between stations and seasons.

During 1975, peak prevalence of *Epistylis* on largemouth bass at Stations 4, 10, and 278 occurred from August through November, and during September and October at Station 17. *Epistylis* was found on largemouth bass at Station 14 in February and from May through December 1975, and at Station 276 in June, July, and September through November 1975. Analysis of variance of the prevalence of *Epistylis* on largemouth bass indicated significant differences among stations (F = 3.7; df = 58; (P < 0.01) and seasons (F = 5.1; df = 4, 58; P < 0.01); no interaction was apparent between stations and seasons (P > 0.10). The prevalence of *Epistylis* on largemouth bass was generally highest during the fall of 1974 and 1975 and during the summer of 1975 (Table 2). Infestations during winter (December, January, and February) and spring (March, April, and May) occurred only at Marshall discharge (Station 14). Prevalence during the winter at Station 14 was significantly greater than at other stations (P < 0.05), but prevalence during spring was not (P > 0.05).

Factors Affecting Prevalence

The prevalence of *Epistylis* on redbreast sunfish and bluegill appeared to increase with length, at least from 25 to 150 mm. This increase was not apparent with largemouth bass. The majority of the white bass and striped bass collected belonged to the same size class, so the relation of prevalence and length of these fishes could not be investigated.

The prevalence of *Epistylis* on fishes did not correlate with the abundance of unattached *Epistylis* zooids in the plankton. *Epistylis* in the plankton exhibited bimodal peaks of abundance—one occurring in November (maximum of 0.35 zooids/l) and a more pronounced peak in April (maximum of 12.1 zooids/l). The temporal occurrence of *Epistylis* was similar among stations despite large variation in water quality variables among stations. *Epistylis* was found in the plankton during November 1974 and 1975 and February, March, and April 1975.

The abundance of Epistylis in the plankton did not correlate with any of the measured water quality variables; however, the prevalence of Epistylis on white bass, redbreast sunfish, bluegill, and largemouth bass did correlate inversely with dissolved oxygen and directly with dissolved orthophosphate (Table 3). Alkalinity and ammonia

	Water Quality Variables							
	Dissolved Oxygen			Ammonia	Tota Iron			
White								
bass	0.26ª	0.355	0.27	0.29°	NS ^a			
Bluegill	-0.28°	0.33°	0.33°	0.28°	-0.21°			
Redbreast								
sunfish	0.35 ^b	0.37ь	0.39 ⁽	0.41 ^f	-0.29°			
Largemouth								
bass	-0.35 ^b	0.22 ^g	NS ^a	NS ^a	-0.27ª			
${}^{a}P < 0.01$ ${}^{b}P < 0.0005$ ${}^{o}P < 0.005$ ${}^{d}P < Not sign$	ificant	۲ Р	$ \stackrel{0.001}{<}_{0.0001}^{<}_{0.05}^{<} $	· · · · · · · · · · · · · · · · · · ·				

Table 3. Correlation coefficients for the prevalence of *Epistylis* on largemouth bass, bluegill, redbreast sunfish, and white bass with water quality variables from Lake Norman and Mountain Island Lake, North Carolina.

were directly correlated with the prevalence of *Epistylis* on white bass, redbreast sunfish, and bluegill (Table 3). The prevalence of *Epistylis* on redbreast sunfish, bluegill and largemouth bass was inversely correlated with total iron (Table 3. These correlations represent the variables which were at least common to 3 of the 4 species analyzed. Using stepwise multiple regression, a model for the prevalence of *Epistylis* was constructed for white bass, redbreast sunfish, bluegill, and largemouth bass. Dissolved oxygen (negative component) and either dissolved orthophosphates or total phosphorus (positive components) were present in all 4 models. Ammonia (positive component) and total iron (Negative component) were present in 2 of the 4 models. The variation explained by the models is as follows: redbreast sunfish, $R^2 = 0.35$ (4 variables); white bass, $R^2 = 0.27$ (3 variables); largemouth bass, $R^2 = 0.20$ (3 variables); and bluegill, $R^3 = 0.16$ (2 variables). These coefficients of determination are low, but the models are statistically significant (P < 0.05).

Site of Infestation

Primary sites of infestation varied significantly (P < 0.001) between redbreast sunfish, bluegill, and largemouth bass and these differed significantly (P < 0.001) from those of white bass and striped bass. *Epistylis* occurred exclusively on the sides of the body of adult striped bass and on the teeth (premaxillary and dentary) of small (< 30 cm) striped bass. Infestations on white bass occurred primarily on the teeth (50%) and sides of the body (43%) regardless of the size of the fish. The majority of the infestations on redbreast sunfish (75%) were found on the teeth. Infestations were also found on the teeth of bluegill (41%) and were abundant on the hard rays of the dorsal, anal, and pelvic fins (34%). Infestations on largemouth bass occurred on the teeth (33%), the ventral side of the dentary and articular bones (29%), and the sides of the body (29%). *Epistylis* was also found on *Lernaea* sp., a parasitic copepod, and on Floy dart tags. In these cases no *Epistylis* was found directly attached to the fish.

Intensity of Infestation

The intensity of infestation (area infested/surface area of fish) was significantly different (F = 20.7; df = 4, 264; P < 0.0001) among species. The mean intensities of white bass (4.7%, n = 51) and striped bass (5.6%, n = 23) infestations were similar and significantly greater (P < 0.05) than mean intensities of redbreast sunfish (1.1%, n = 61), bluegill (1.2%, n = 54), and largemouth bass (1.2% n = 76). Mean intensities were not different between redbreast sunfish, bluegill, and largemouth bass.

The intensity of infestation was positively correlated with surface area of white bass (r = 0.29) and striped bass (r = 0.77) and negatively correlated with the surface area of bluegill (r = -0.33). The intensity of infestation did not correlate with the surface area of redbreast sunfish and largemouth bass. Intensity of infestation correlations with length or age of fishes were similar to those observed with surface area.

Intensity of infestation did not vary among stations or months during fall when *Epistylis* was most prevalent. Intensity of infestation did not vary between sexes (P > 0.05). The intensity of infestation was negatively correlated with the relative condition of redbreast sunfish (r = -0.27), but not for the other species studied.

Microscopic examination of suspected *Epistylis* infestations was essential. Only 34 percent of the largemouth bass suspected of having *Epistylis* were actually infested. This resulted in part from numerous bass collected during March and April with lesions but no *Epistylis* colonies. Field identification was also low for bluegill (49%). Accuracy for field identification of *Epstylis* on striped bass, white bass, and redbreast sunfish was 93 percent, 76 percent, and 81 percent, respectively.

DISCUSSION

Ictalurids, percichyids, and centrarchids were the fishes most commonly found with Epistylis infestations. Within each of the families, the species collected were, in general, similarly effected. Chapman et al. (1976), Esch et al. (1976), Miller and Chapman (1977), and Plumb (1973) demonstrated that fishes other than ictalurids, percichthyids, and centrarchids were not appreciably affected by Epistylis.

The prevalence of *Epistylis* on striped bass and white bass was considerably higher than on other species. Dead striped bass with characteristic *Epistylis* lesions were found on Lake Norman during September and October, but no colonies were found on the dead fish. The prevalence and intensity of *Epistylis* infestations appeared high enough that mortalities probably resulted from *Epistylis* infestations on percichthyids. Although no fish kills on Lake Norman or Mountain Island Lake have been attributed to *Epistylis*, fish kills on Badin Lake, North Carolina, have been attributed to primary infestation by *Epistylis* and subsequent, secondary infections by *A. hydrophila* (Plumb 1973).

A. hydrophila is capable of primary invasion (Snieszko and Bullock 1968), although Miller and Chapman (1977), Plumb (1973), and Rogers (1971) suggested that Epistylis is the primary invader and A. hydrophila the secondary invader. Esch et al. (1976) and Miller and Chapman (1977) expressed concern as to which organism is the etiologic agent. We also share this concern, since we observed largemouth bass and bluegill, especially during spring and summer, with lesions and no visible *Epistylis*. This is especially apparent from the percentage of lesions examined with no *Epistylis* (e.g. largemouth bass 66%, bluegill 51%). More consequentially, Chapman et al. (1976) found A. hydrophila in 54 percent of fishes having lesions but no *Epistylis*; A. hydrophila was also present in 64 percent of the cultures made from fishes with *Epistylis*.

The prevalence of *Epistylis* was low on centrarchid fishes, and was similar among species. Because of the low prevalence and intensity of infestations, we feel that *Epistylis* had little impact on the centrarchids of Lake Norman or Mountain Island Lake. Furthermore, we received very few comments from fishermen concerning *Epistylis* on centrarchids; the majority of the complaints concerned *Epistylis* infestations on striped bass.

We are confident that a large proportion of the fishes with *Epistylis* recover from infestations. Evidence for this is the collection of healthy largemouth and striped bass wih extensive areas of regenerated and partially eroded scales in a pattern characteristic of *Epistylis* lesions on the sides of these species.

The probability of infestation should be directly related to length (Collard 1970) and surface area of the fish. Cloutman et al. (1978), Chapman et al. (1976) and Esch et al. (1976) documented an increase in the prevalence of *Epistylis* with the length of bluegill, redbreast sunfish, and largemouth bass. We observed a similar increase with the total length of bluegill and redbreast sunfish but not for largemouth bass. An increase in the prevalence of *Epistylis* with length and total surface area of largemouth bass was expected.

Epistylis was generally most prevalent on Lake Norman and Mountain Island Lake fishes during late summer and fall. Chapman et al. (1976) and Cloutman et al. (1978) also reported higher prevalence of *Epistylis* during summer and fall with peak prevalence occurring during September and October. Rogers (1971) noted that Epistylis infestations in the southeastern U.S. occurred in every month of the year, but were most prevalent during the winter and spring. Esch et al. (1976) found that the prevalence of Epistylis on centrarchid fishes from a South Carolina reservoir receiving heated effluents was lower in fall and winter with a peak in spring and a decline in summer. Esch et al. observed higher prevalence on largemouth bass taken from heated areas than those collected from unheated areas; other species of centrarchids were not similarly influenced by the heated effluents. We observed differences between the prevalence of Epistylis on largemouth bass collected at Marshall discharge (Station 14) and other stations, but these differences occurred mainly during winter and spring when infestations were absent from other stations. Despite unusually large sample sizes, no largemouth bass collected from Riverbend discharge (Station 276) during winter and spring were found with Epistylis infestations and prevalence of Epistylis on largemouth bass at Station 276 was similar to that of the unheated stations. The prevalence of Epistylis on white bass appeared higher at Station 14, but very few adult white bass were collected at other stations; when they were available at unheated stations, they generally had infestations. Prevalence of Epistylis on other species of fish appeared unaffected by the heated effluents.

The disparity in the prevalence of *Epistylis* on largemouth bass from the 2 discharge stations could not be explained by differences in water temperature or bass population densities; however it is, in part, explainable by the sources of cooling water of the 2 steam plants. During thermal stratification (May to October), Marshall Steam Station utilizes hypolimnetic water. Thus, high temperatures and low dissolved oxygen concentrations (< 6 mg/l) are apparent from late spring through fall in the surface waters at Station 14 and from late summer through fall at other stations. During spring, the Marshall Steam Station also affects the water quality of the surface waters by drawing water from deep turbidity currents that result during periods of rain. Often the increased turbidity is visually apparent only in the intake and discharge canals of Marshall Steam Station. This turbid water is not apparent outside the discharge flume.

Relatively high concentrations of dissolved orthophosphate and low concentrations of dissolved oxygen occurred during late spring, summer, and fall at Station 14 and during late summer and fall at other stations. We doubt that these variables directly affect *Epistylis*, but they may have been associated with the abundance of bacteria and water dispersed particles on which *Epistylis* feeds (Lom 1966). Bacteria are generally most abundant in bottom waters, especially at the end of thermal stratification (Kuznetsov 1970). Thus, the suspected abundance of bacteria occurs concurrently with the prevalence of *Epistylis* on fishes, the high concentrations of dissolved orthophosphate, and the low concentrations of dissolved oxygen. Water temperature probably played a role in the prevalence of *Epistylis*, since *Epistylis* was not found on fishes at water temperatures below 14 C. Also, stress that can result from high temperatures and low dissolved oxygen concentrations (Meyer 1970), probably contributed to the prevalence of *Epistylis*.

The prevalence of *Epistylis* on fishes did not correlate with the abundance of *Epistylis* in the plankton. Roche et al. (1978) compared the sizes of Epistylis on fishes to Epistylis in the plankton. They found that Epistylis in the plankton was considerably smaller than the Epistylis found on fishes. These 2 sizes of Epistylis may have represented 2 species, however. Lom (1966) observed considerable variation in the size and structure of Epistylis lwoffi on different hosts.

The intensity of infestation was higher on percichthyids than on centrarchids, and was similar among infested fishes of the same family. Intensity of infestation increased with the surface area and length of percichthids but not of centrarchids. Percichthyids were more susceptible to *Epistylis* invasion and development of extensive colonies of *Epistylis*. Adult striped bass and white bass usually had 6 or more independent lesions per fish, whereas centrarchids commonly had only 1 or 2 independent lesions. This could have made striped bass and white bass more susceptible to infection by A. hydrophila (Chapman et al. 1976).

The attachment of Epistylis on fishes appeared species-specific. The results of our study and the results of Chapman et al. (1976), Cloutman et al. (Unpublished), and Esch et al. (1976) showed considerable agreement in the location of Epistylis on an individual species.

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