Best wishes
Manny Pescador

5 Nymphal abnormalities in *Stenacron interpunctatum* (Ephemeroptera: Heptageniidae) from the Fenholloway River, Florida

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The nymphs of Stenacron interpunctatum *(Say) from the Fenholloway River, a stream severely impacted by cellulose mill wastes, were found to have abnormal gills while those from the Econfina River, a nearby and relatively unpolluted stream, were found to have normally developed gills. The abnormalities of* S. interpunctatum *nymphs from the Fenholloway include: 1. development of fibrilliform portions of the 7th abdominal gills; 2. development of extra long and multibranched fibrilliform portions of the abdominal gills; 3. unusually numerous tracheal branches of the abdominal gills; and 4. enlargement of the gill lamellae. Statistical analysis of the ratios of the abdominal gill length and width to body length, head length, mesonotal width and width of abdominal segment four showed that the nymphs of* S. interpunctatum *from the Fenholloway River have significantly larger gills, both in length and width than the nymphs from the Econfina.*

The gill abnormalities of the S. interpunctatum *nymphs appear to be a function of the extremely depressed dissolved oxygen and high BOD values below the cellulose plant. These abnormalities could be a form of adaptive response to this environmental stress. The enlargement of the gills and other associated gill structures not only increases the total respiratory surface area but perhaps minimizes gill movements or beatings during respiration, thus energy is conserved.*

Introduction

Mayflies are well known for their sensitivity to various forms of environmental stress that impact freshwater ecosystems. Consequently, they have been widely used as indicators of water quality (Brittain 1982). A few mayfly species, however, have been found to show a wide range of tolerance to various pollutants. One example is the heptageniid species, *Stenacron interpunctatum* (Hubbard and Peters 1978; Hilsenhoff 1987, 1988; Lenat 1991). The species is widely distributed,

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having basically the same geographic range as the genus. It is generally found throughout Florida in permanently flowing water (Berner and Pescador 1988).

Stenacron interpunctatum is generally considered a pollution tolerant species, but no morphological features that enable survival during long-term exposure to contaminated environments have been documented. The development of morphological abnormalities due to contaminants has been commonly observed in Chironomidae (Warwick 1985, 1988, 1989, 1990; Warwick *et al.* 1987; Warwick and Tisdale 1988; Pettigrove 1989; Dermott 1991). However, no attempt has been made to determine whether these abnormalities were an adaptive form of response to the contaminants.

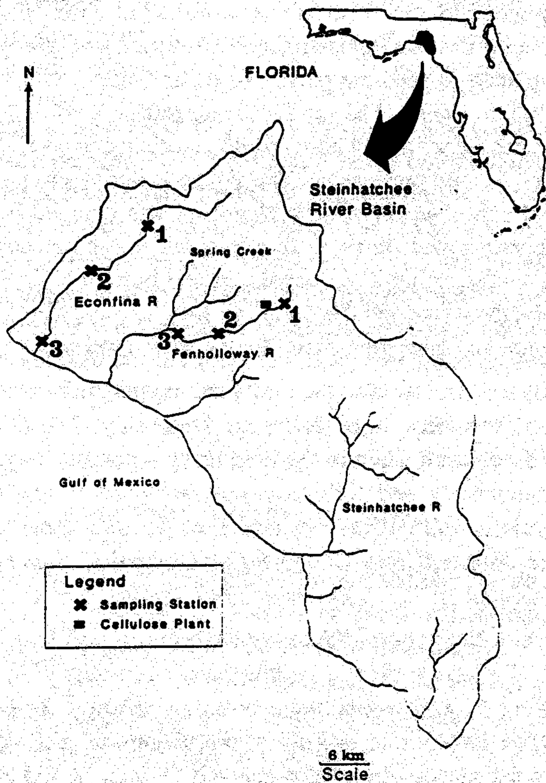
Although still preliminary, our study represents the first documentation of morphological abnormalities in mayflies exposed to a contaminated environment over a long period of time. One objective of the study is to describe the types of morphological abnormalities in *Stenacron interpunctatum* nymphs in the Fenholloway River. The second is to attempt to establish the probable cause and effect of these abnormalities.

Study Areas

The specimens of *Stenacron interpunctatum* used for this study were collected from the Fenholloway and Econfina rivers, with drainage areas of 1049 and 774 km^2, respectively. Both rivers are located in Taylor County, Florida within the coastal lowland Steinhatchee River Basin (Fig. 1). The Fenholloway and Econfina are small rivers originating in the San Pedro swamp and discharge into the Gulf of Mexico. Except for the industrial activities that are impacting the Fenholloway, both rivers have similar hydrological features (Fig. 2), types of vegetation and substrates consisting mainly of admixtures of sand, silt and clay, interspersed with a few exposed limestone rocks.

The Fenholloway River is a damaged system due to the long-term use of this stream as a receptacle for about 190 million L/day of cellulose mill wastes (Livingston and Fernald 1990). The cellulose plant, which has been in operation since 1954, is located along the river near Perry, Florida. During periods of low precipitation plant effluent makes up nearly the entire flow of the river. The Econfina, our reference system, is a relatively pristine river with no known point sources of pollution. The river has been selected as one of the potential candidate reference sites [benchmarks for disturbed streams in same region (Hughes et al. 1986)] in the proposed Ecoregions/Subregions of Florida by the Florida Department of Environmental Regulation (FDER). Both the Econfina and the Fenholloway rivers belong to the same ecoregion (Southern Coastal Plain) and subregion (Gulf Coast Flatwoods). The classic upstream and downstream approach to bioassessment of the impact of point source pollution is inapplicable in the study for two reasons: 1. our upstream sampling site (Station 1, Fig. 1) frequently experiences dryness due

Figure 1. Map of the study areas and sampling stations.



to voluminous water usage by the cellulose plant, and 2. *S. interpunctatum* does not occur or has never been collected in this site.

Materials and Methods

Benthic and water samples were collected from three stations each in the Fenholloway and Econfina rivers (Fig. 1). On the Fenholloway River, Station 1 was 4.4 km above the point of cellulose plant discharge, and stations 2 and 3 were 9.7 km and 26.4 km below the discharge, respectively. The three sampling stations on the Econfina River (Fig. 1) were located at various points along the river with Station 1 near the headwaters, Station 2 approximately midway along the river and Station 3 near the mouth of the river. The nymphs of *S. interpunctatum* along with other macroinvertebrates were collected bimonthly for one year (June 1991 to June 1992), using modified Hester-Dendy multiplate samplers (consisting of four columns of masonite multiplate samplers mounted on a concrete block). Each column of Hester-Dendy plates had a total surface area of 0.09 m^2. The Hester-Dendy plates were incubated in the river for approximately 60 days. Upon retrieval, the incubated samplers were placed in separate plastic bags, half-filled with stream water and stored in ice while in transit. The samples were refrigerated in the laboratory and subsequently processed within 24 hours by dismantling and carefully brushing with tap water over a 0.71 mm mesh sieve. The sieved material was preserved in 75 per cent ethanol and subsequently sorted, identified and counted using a dissecting microscope. Approximately three to five minutes of dip net collecting was also carried out bimonthly, using a rectangular frame bottom kick net to sample the various types of habitats around the sampling sites. Dip net samples were stored in 75 per cent ethanol and later processed like the sieved material above.

Water chemistry data such as the dissolved oxygen, temperature, pH, specific conductivity and turbidity were collected bimonthly. Dissolved oxygen and temperature readings were taken in the field using a portable oxygen meter, while the specific conductivity and turbidity were analyzed in the laboratory using standard conductivity and turbidity meters. Data for additional physico-chemical parameters were obtained from United States Geological Survey (U.S.G.S.) and FDER (Meadows et al. 1987, 1988, 1989, 1990, 1991).

Thirty-five and 25 nymphs of *S. interpunctatum* of various body sizes from the Econfina and Fenholloway rivers were measured and analyzed, respectively. Only 25 out of a total of 55 *S. interpunctatum* nymphs collected for one year from the Fenholloway River have either or both of the abdominal gills 4 intact. Thus, 25 nymphs became the sample size in this analysis. A total of 135 *S. interpunctatum* nymphs was collected from the Econfina River. We used a slightly larger sample size of 35 nymphs from this river.

The measurements of body length (BL) (excluding the caudal filaments), head length (HL), mesonotal width (MSW), abdominal gill 4 length (G4L) and width (G4W), and abdominal segment 4 width (AS4W) of the nymphs of *S. interpunctatum* were measured to the nearest 0.1 mm. The abdominal gills were temporarily mounted with 75 per cent ethanol on glass slides to facilitate the measurements. Nymphs were covered by a 22 x 22 x 1 mm glass cover slip during measurements of body parts. Figures 6-7 show how the nymphal structures were measured.

Ratios were computed for the following measurements of nymphal body parts from each river:

G4W to BL	G4L to BL
G4W to HL	G4L to HL
G4W to MSW	G4L to MSW
G4W to AS4W	G4L to AS4W

A Generalized Linear Models (GLM) Procedure (Schlotchaver and Littell 1987) was used to statistically analyze the data instead of the analysis of variance (ANOVA) in order to compensate for the unbalanced data set, 25 specimens from the Fenholloway River and 35 from the Econfina River. The Least Significant Difference T-test (LSD) (Schlotchaver and Littell 1987) was also used to analyze mean ratios of gill size to the body part size. A comparison of the growth relationships of body length, head length, mesonotal width and gill length and width from both river systems was also statistically analyzed using regression analysis.

Results

Physico-chemical Characteristics of the Study Areas

Tables 1-3 and Figs. 2-5 show some of the selected physico-chemical features of the Econfina and Fenholloway rivers. Both the Econfina and Fenholloway rivers have similar levels of mean water discharge (Fig. 2) except the latter below the cellulose plant shows less fluctuation in flow due to the steady release of plant effluent into the river. A summary of the U.S.G.S. discharge data on both rivers is presented in Table 2.

The mean water temperatures of the Fenholloway River are slightly higher than those of the Econfina River (Fig. 4). The specific conductivity values of the Fenholloway River are significantly higher than the Econfina River (Tables 1 and 3, Fig. 3). The dissolved oxygen concentrations are severely depressed in the Fenholloway River below the cellulose plant as compared to the Econfina River (Tables 1 and 3, Fig. 2). Our data show that the dissolved oxygen and specific conductivity below the cellulose plant have mean values of 2.3 mg/L (range 0.5-4.4 mg/L) and 1210 µmhos/cm (range 488-1830 µmhos/cm), respectively (Table 1). On the other hand, the mean values of the dissolved oxygen and specific conductivity of the three sampling stations from the Econfina River were 5.8 mg/L (range 3.4-8.3 mg/L) and 208 µmhos/cm (range 49.1-660 µmhos/cm) respectively (Table 1). Additionally, the BOD and Total N values below the cellulose plant are significantly higher than those from the Econfina River (Table 3).

Table 1. Mean values of selected water parameters from the Econfina and Fenholloway rivers (June 1991 to June 1992).

Station	#OBS.	Temp. (^{0}C)	D.O. (mg/L)	pH (s.u.)	Conductivity (µmhos/cm)	(N.T.U.)
Fenholloway R.						
Sta. 1	5	18.6	7.5	4.11	80.3	3.0
Sta. 2	7	23.6	2.0	6.52	1344	1.7
Sta. 3	7	22.3	2.6	6.64	1076	1.7
Sta. 2 and 3	x=	23.0	2.3	6.58	1210	1.7
Econfina R.						
Sta. 1	7	19.5	6.0	5.72	98.5	1.6
Sta. 2	7	20.5	5.1	6.30	253	2.3
Sta. 3	6	20.9	6.2	6.63	273	2.3
Sta. 1,2,3	x=	20.3	5.8	6.22	208	2.1

#OBS = number of observations

Table 2. Summary of United States Geological Survey discharge data from the Fenholloway and Econfina rivers (U.S.G.S - Data Report FL-91-4).

River System	U.S.G.S. Gage No.	Average (m^3/s)	Max. (m^3/s)	Min. (m^3/s)	Period of Record
Fenholloway	*02324400	1.56	90.9	0.01	1955-1991
Fenholloway	*02324500	3.77	136	0.02	1946-1991
Econfina	*02326000	4.11	71.9	0.07	1950-1991

*02324400 Approximately 8.4 km above discharge of cellulose plant

*02324500 Approximately 3.9 km below discharge of cellulose plant

*02326000 Approximately 22.5 km upstream from mouth

Table 3. Summary of Florida Department of Environmental Regulations water data for selected sites from the Fenholloway and Econfina rivers (1983-1988) (median values for each station) (unpublished data) TURB=turbidity (mg/L), SD=secchi depth (m), COLOUR=colour, TSS=total suspended solids (mg/L), DO=dissolved oxygen (mg/L), DOSAT=dissolved oxygen per cent saturation, BOD=biochemical oxygen demand (mg/L), COD=chemical oxygen demand (mg/L), pH=pH standard units, ALK=alkalinity (mg/L), NITRO=total nitrogen (mg/L), PHOS=total phosphorus (mg/L), CHLA=chlorophyll *a* (µg/L), COND=conductivity (µmhos).

River	#OBS	TURB	SD	COLOUR	TSS	DO	DOSAT	BOD	COD	pH	ALK	NITRO	PHOS	CHLA	COND
Fennholloway 4 km above Sta. 1	3	2.5	—	248	—	3.5	38	1.6	—	6.5	76	0.96	0.46	1	217
Fenholloway at Sta. 2	5	5.7	0.2	2100	8	0.6	4	6.8	203	7.1	106	7.29	1.19	5	1460
Fenholloway at Sta. 3	5	4.2	0.2	1030	6	0.7	8	4.0	178	6.8	122	5.61	0.94	1	1128
Fenholloway Sta. 2 and 3 $_x$		5.0	0.2	1565	7	0.7	6	5.4	191	7.0	114	6.5	1.07	3	1294
Econfina at Sta. 1	6	2.5	0.6	480	11	5.7	63	1.1	—	5.4	8	0.48	0.11	2	56
Econfina at Sta. 2	5	5.0	0.5	380	2	5.3	58	1.1	—	7.0	124	0.67	0.20	0	56
Econfina at Sta. 3	6	4.0	0.6	175	2	5.9	63	1.0	—	7.4	179	0.58	0.16	1	195
Econfina Sta. 1,2,3 $_x$		3.8	0.6	345	5	5.6	61	1.1	—	6.6	104	0.58	0.16	1	238

Figure 2. **Mean monthly water discharge and bimonthly dissolved oxygen of the Econfina and Fenholloway rivers. Top: discharge [United States Geological Survey Water Data Report, FL (1987-1991)-4)]; Bottom: Bimonthly mean dissolved oxygen of the three sampling stations from the Econfina River, and the two sampling stations below the cellulose plant from the Fenholloway River.**

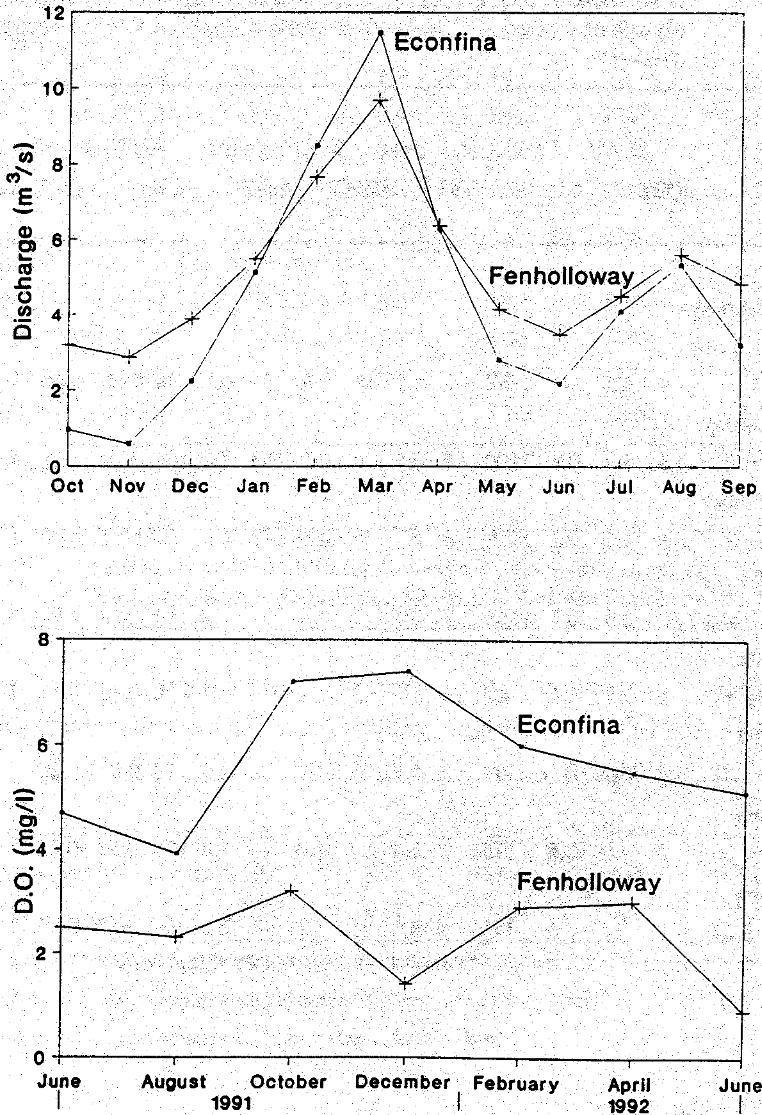
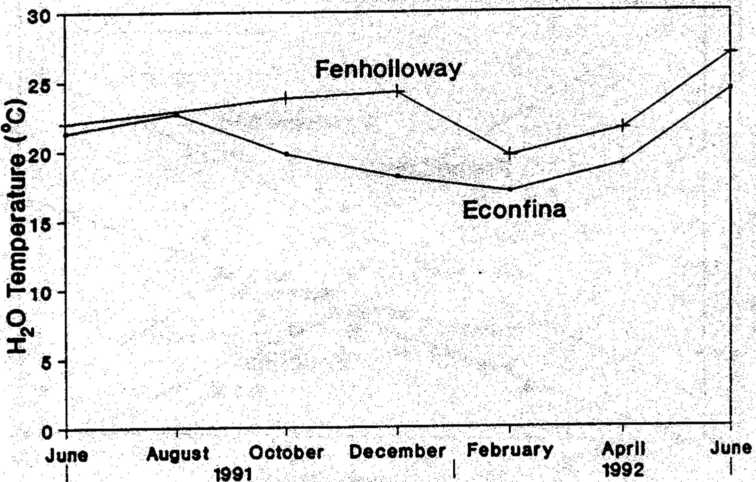


Figure 3. **Bimonthly mean temperature and conductivity of the three sampling stations from the Econfina River and the two sampling stations below the cellulose plant from the Fenholloway River; Top: temperature Bottom: conductivity.**



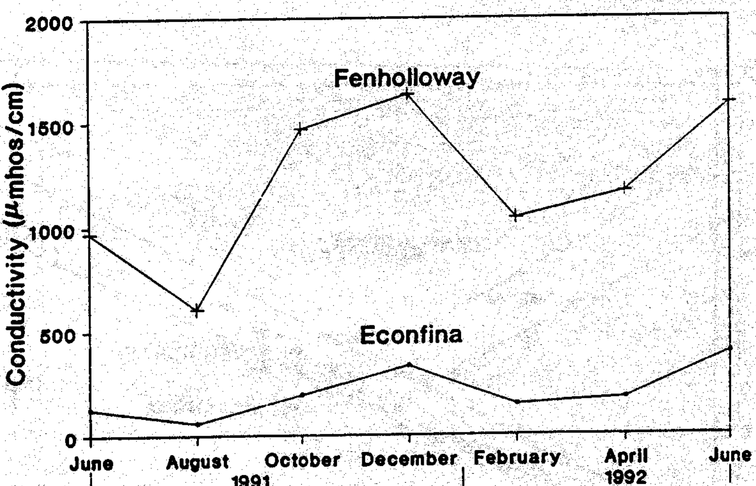
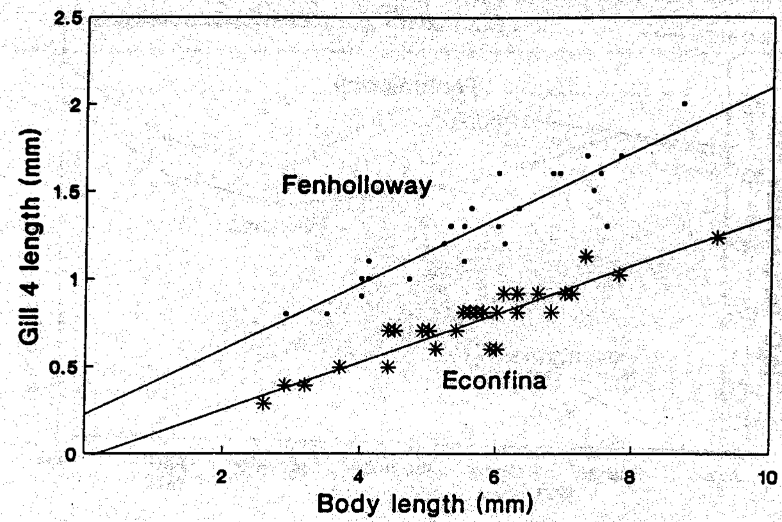


Figure 4. **Scatter diagrams and a regression line showing the linear relationship of gill 4 length to body length (Top) and gill 4 length to head length (Bottom) of *S. interpunctatum* nymphs from the Econfina and Fenholloway rivers.**



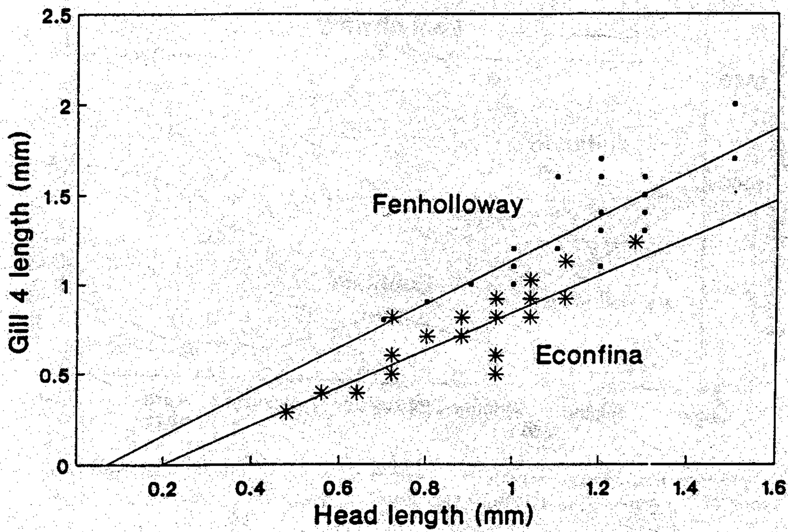
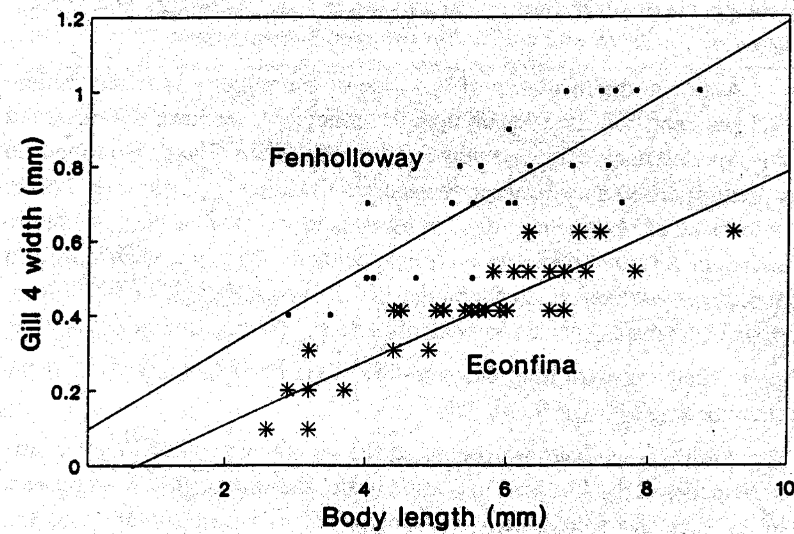
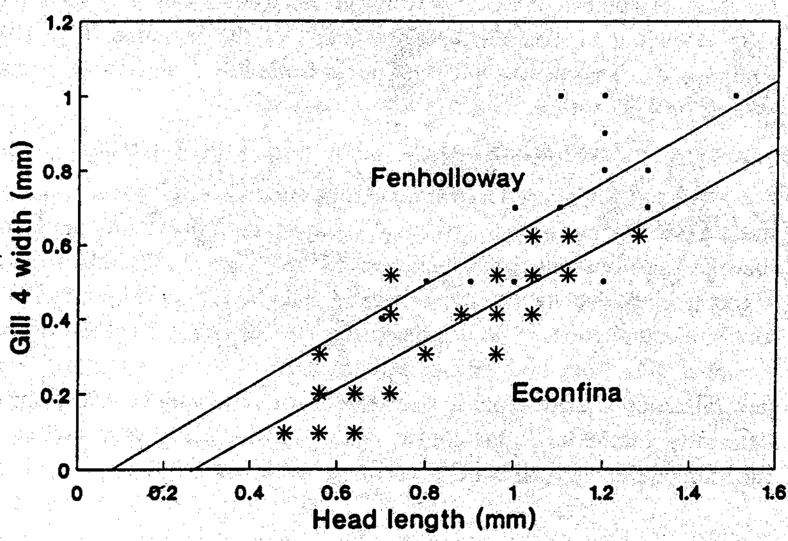


Figure 5. **Scatter diagram and a regression line showing the linear relationship of gill 4 width to body length (Top) and gill 4 width to head length (Bottom) of *S. interpunctatum* nymphs from the Econfina and Fenholloway rivers.**





Abnormalities of the Abdominal Gills of Stenacron *Nymphs*

Nymphs of *S. interpunctatum* from the Fenholloway River have the following abnormalities:

1. Development of fibrilliform portion of the left 7th abdominal gill (Figs. 8 and 11)

   Although the incidence of this type of abnormality is relatively low (7.3 per cent; four of 55 total nymphs collected), we have not observed this development in the nymphs from the Econfina River. We found no structural differences between these extra fibrilliform portions of the 7th with those of abdominal gills 1-6 other than the smaller size and fewer branches. It is unknown why all four specimens have an extra fibrilliform portion only on the left 7th abdominal gill. It is possible that the fibrilliform portions on the right side were detached during the processing and sorting.

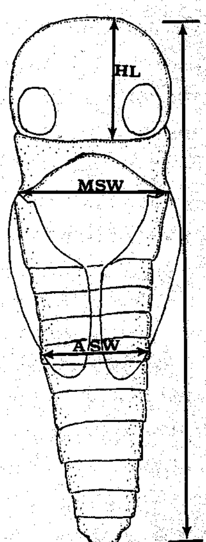
2. Development of extra long and multibranched fibrilliform portion of the abdominal gills (Figs. 9, 10, 12)

   Although we have not measured the length and counted the number of branches of the fibrilliform portion of the abdominal gills, it is apparent to us that the fibrilliform portions of *S. interpunctatum* from the Fenholloway River are much longer and more multibranched than the fibrilliform portions from Econfina River specimens. The gill tufts of the nymphs from the Fenholloway River extend well beyond the lateral margins of the gill lamellae (Figs. 9, 12) and most extend to the apex of the lamellae. A number of nymphs from the Econfina River may have the tufts extending beyond the lateral margins of the lamellae (Fig. 10). However, this is not as prominent as ones in Fenholloway River specimens, and the tufts do not extend to the lamellar apical margin.

3. Unusually numerous tracheal branches of the gills (Figs. 9, 10, 12, 13)

   We have not devised a technique to accurately count the number of trachea per unit surface area of the gill. However, it is visually apparent that the nymphs from the Fenholloway River (Figs. 9, 12, 13b) have a higher tracheal density than those from the Econfina River (Figs. 10, 13a). Until an actual count is done, comparing the number of tracheae of the nymphal gills from both rivers, caution must be exercised before any generalization is made regarding the extent and reliability of this specific deformity. Furthermore, the density of the tracheae could very well be a function of enlarged gill lamellae as discussed below.

Figures 6-10. Illustrations of nymph and gills showing linear measurements; 6) Numphal body parts: body length (BL), head length (HL), mesonotal width (MSW), abdominal segment 4 width (AS4W); 7) Abdominal gill: gill 4 length (G4L), gill 4 width (G4W); 8-10) Abdominal gills of *S. interpunctatum*, dorsal view: 8) Gill 7 (Fenholloway River), 9) Gill 4 (Fenholloway River), 10) Gill 4 (Econfina River).



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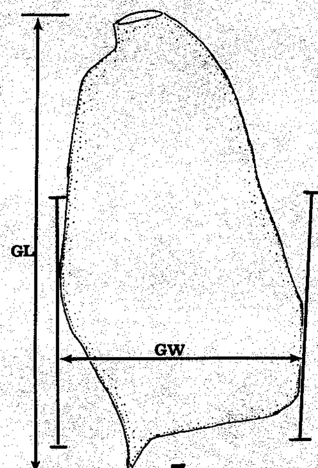

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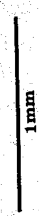

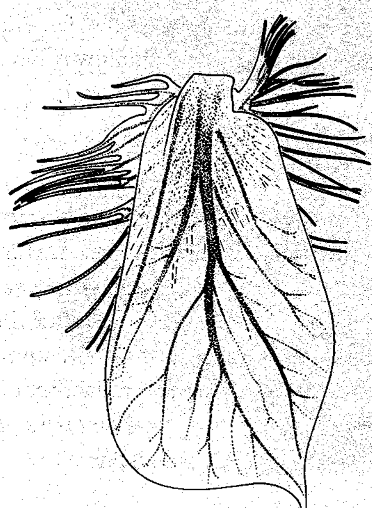

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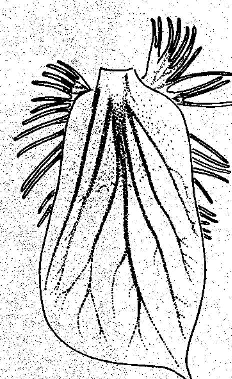

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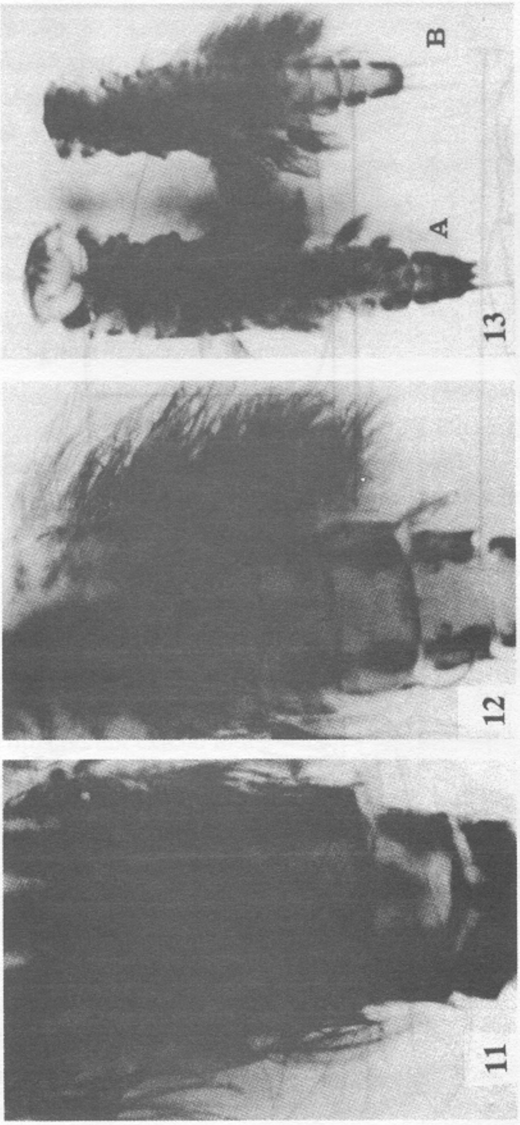
Figures 11-13. **Photomicrographs of the abdominal gills of *S. interpunctatum* from Fenholloway River: 11) Dorsal view of gill 7 (30X); 12) Ventral view showing fibrilliform portions and tracheation (22X); 13) Ventral view of nymphal gills. a) Econfina River, b) Fenholloway River (7X).**



4. Enlargement of the gill lamellae (Figs. 9, 10, 13).

   The gill lamellae of the nymphs from the Fenholloway River are significantly larger both in length and in width than the gill lamellae of the Econfina River specimens (Tables 4 and 5). Figures 9, 10 and 13 illustrate the significant differences in gill size of *S. interpunctatum* nymphs from the two rivers. Figures 9 and 10 were drawn from gills that were excised from nymphs of almost the same body length (7.2 and 7.3 mm) and equal head length (1.4 mm). The nymphal gill from the Fenholloway River (Fig. 9) is almost two times larger than the one from the Econfina River. Figures 4-5 compare relationships of the gill length and width to body length, head length, mesonotal width and width of the abdominal segment 4 of specimens of *S. interpunctatum* nymphs from the Fenholloway and Econfina rivers.

Results of the LSD tests (Table 6) show that the mean ratios of the gill length and width to body length, head length, mesonotal width and the width of abdominal segment 4 are significantly different ($p<0.05$) among nymphal populations of *S. interpunctatum*. For example, the mean ratios of gill length to body length of the nymphs from the Fenholloway River is much higher (0.228) than the Econfina River (0.172). Similarly, the mean ratios of gill width to body length of the nymphs from the Fenholloway River is 0.126 compared to 0.090 from the Econfina. From the results (Table 6), we can strongly conclude that the nymphal gills of *S. interpunctatum* from the Fenholloway River are significantly larger than those from the Econfina River.

A regression analysis comparing the growth relationships of body length, head length, mesonotal width and abdominal segment 4 width with gill 4 width and length indicate that the abdominal gills of *S. interpunctatum* nymphs from the Fenholloway River experience significantly faster growth, relative to body size, than those from the Econfina River (Table 7). For example, for every unit of growth of gill 4 width of the Econfina River nymphs, the body length increases approximately ten times (9.98) compared to approximately six times (6.39) for the nymphs from the Fenholloway River. Similarly, a unit increase of the gill length of the nymphs from the Econfina River produces a corresponding body length increase of approximately six times (6.64) compared to approximately four times (4.43) for the Fenholloway (Table 7). The data on growth relationships of gill size with the various nymphal body parts such as body length, head length, mesonotal width and abdominal segment 4 width strongly suggest that the abdominal gills of *S. interpunctatum* nymphs from the Fenholloway River experience more growth than those from the Econfina River.

Table 4. **Comparison of the abdominal gill 4 width and length (G4L) to body length (BL), head length (HL), mesonotal width (MSW) and abdominal segment 4 width (AS4W) of *Stenacron interpunctatum* nymphs from the Econfina River (all units are mm).**

Specimen#	G4W	G4L	BL	HL	MSW	AS4W
1	0.7	1.4	9.2	1.6	2.0	1.5
2	0.6	1.2	7.8	1.3	1.6	1.3
3	0.5	0.8	5.9	1.2	1.2	1.0
4	0.6	1.1	7.1	1.4	1.4	1.3
5	0.5	1.0	6.8	1.3	1.5	1.1
6	0.3	0.6	3.2	0.8	0.8	0.7
7	0.5	0.8	5.1	0.9	1.0	0.9
8	0.6	1.0	6.8	0.9	1.4	1.1
9	0.2	0.5	2.6	0.6	0.7	0.5
10	0.7	1.1	7.0	1.4	1.6	1.4
11	0.5	0.9	5.0	1.1	1.1	1.0
12	0.6	1.1	6.6	1.2	1.3	1.1
13	0.5	1.0	5.0	1.2	1.2	1.1
14	0.3	0.7	3.7	0.9	0.7	0.7
15	0.5	0.9	4.5	1.1	1.0	0.9
16	0.4	0.7	4.4	1.2	0.9	0.8
17	0.2	0.6	3.2	0.7	0.7	0.6
18	0.6	1.1	6.1	1.3	1.3	1.3
19	0.5	1.0	5.7	1.1	1.2	1.1
20	0.5	1.0	5.6	1.1	1.3	1.0
21	0.5	1.1	6.6	1.3	1.6	1.3
22	0.5	1.0	5.5	1.2	1.2	1.0
23	0.5	1.0	6.0	1.3	1.2	1.1

Table 4. (continued)

24	0.4	0.9	4.9	1.0	1.1	0.9
25	0.2	0.6	3.2	0.8	0.7	0.6
26	0.5	0.9	5.4	1.1	1.1	1.0
27	0.6	1.0	6.3	1.3	1.5	1.2
28	0.6	1.1	5.8	1.2	1.4	1.3
29	0.5	1.0	5.7	1.2	1.2	1.1
30	0.7	1.1	6.3	1.3	1.4	1.3
31	0.5	0.8	6.0	1.2	1.3	1.1
32	0.4	0.6	3.2	0.7	0.7	0.6
33	0.5	0.9	4.4	1.1	1.0	1.0
34	0.3	0.6	2.9	0.7	0.6	0.5
35	0.7	1.3	7.3	1.4	2.0	1.4

Discussion

Why are these gill abnormalities of *S. interpunctatum* occurring in the Fenholloway River and not in the Econfina River nor from the other streams in the area that we are presently and regularly collecting samples? Our results and observations are quite preliminary. More physico-chemical data will be collected and analyzed. Moreover, experimental data are needed before one can absolutely determine factors causing the development of gill abnormalities of this mayfly species in the Fenholloway River. It is fair to assume that the pollutants being discharged into the system are creating a stressful environment for benthic organisms. A number of studies have shown that several aquatic organisms develop morphological abnormalities when exposed to contaminated waters for an extended period of time. Bortone and Drysdale (1981) and Davis and Bortone (in press) have found a high incidence of masculinization among female poeciliid species, *Gambusia holbrooki, Heterandia formosa* and *Poecilia latippina* from the Fenholloway River. The authors surmised that pine oils coming from the cellulose plant are affecting the female endocrine system inducing the development of male characteristics in the females.

Table 5. **Comparison of the abdominal gill width (G4W) and length (G4L) to body length (BL), head length (HL), mesonotal width (MSW) and abdominal segment 4 width (AS4W) of *Stenacron interpunctatum* nymphs from the Fenholloway River (all units are mm).**

Specimen	G4W	G4L	BL	HL	MsW	AS4W
1	0.7	1.3	7.6	1.3	1.7	1.3
2	1.0	2.0	8.7	1.5	2.1	1.8
3	1.0	1.6	7.5	1.1	1.8	1.1
4	0.8	1.6	6.9	1.3	1.5	1.2
5	0.5	0.9	4.0	0.8	0.9	0.7
6	0.7	1.2	6.1	1.0	1.5	1.0
7	0.9	1.6	6.0	1.2	1.4	1.4
8	0.7	1.0	4.0	1.0	1.1	1.0
9	0.7	1.2	5.2	1.1	1.1	1.0
10	1.0	1.6	6.8	1.2	1.2	1.3
11	0.7	1.2	5.2	1.1	1.1	1.0
12	0.7	1.3	6.0	1.3	1.3	1.2
13	0.8	1.5	7.4	1.3	1.4	1.4
14	0.4	0.8	3.5	0.7	0.8	0.6
15	1.0	1.7	7.8	1.2	1.2	1.4
16	0.8	1.4	5.6	1.2	1.5	1.3
17	0.5	1.0	4.1	0.9	0.8	0.9
18	0.5	1.1	4.1	1.0	1.0	0.9
19	0.5	1.0	4.7	1.0	1.0	0.9
20	0.8	1.3	5.3	1.2	1.2	1.2
21	0.8	1.4	6.3	1.3	1.4	1.4
22	0.4	0.8	2.9	0.7	0.7	0.6
23	1.0	1.7	7.3	1.5	1.5	1.4
24	0.5	1.1	5.5	1.2	1.1	1.0
25	0.7	1.3	5.5	1.3	1.3	1.0

Table 6. Results of Least Significant Difference (LSD) Tests comparing the mean ratios of the abdominal gills to various parts of the body of *Stenacron interpunctatum* nymphs from the Fenholloway and Econfina rivers.

Body Parts Ratios	Fenholloway River Mean	Econfina River Mean
Gill 4 Width/Body Length	0.12613 A	0.09010 B
Gill 4 Width/Head Length	0.6347 A	0.4368 B
Gill 4 Width/Mesonotal Width	0.5782 A	0.4139 B
Gill 4 Width/Segment 4 Width	0.6563 A	0.4800 B
Gill 4 Length/Body Length	0.22802 A	0.17228 B
Gill 4 Length/Head Length	1.1455 A	0.8305 B
Gill 4 Length/Mesonotal Width	1.0468 A	0.7902 B
Gill 4 Length/Segment 4 Width	1.1890 A	0.9182 B

Comparison made at the 0.05 level of significance. Means with the same letter are not significantly different.

Several genera and species of chironomid midges have been reported to develop morphological deformities when exposed to various forms of toxic and organic contaminants. These deformities involved the deformation of the antennae and various structures of the larval mouthparts (Warwick 1985, 1989, 1990; Warwick et al. 1987; Pettigrove 1989; Dermott 1991). Interestingly, some of these genera (e.g., *Chironomus* and *Procladius*) have developed varying degrees of deformities when exposed to various sources and levels of pollution. As a result, an indexing scheme for classifying deformations in these genera has been proposed to illustrate the potential for using deformities as a biological technique for the detection and assessments of contaminants in aquatic ecosystems (Warwick 1985, 1989). Although the linkages between morphological deformities and contaminants are presently based on circumstantial evidence, and only limited experimental data, they are compatible with the theory that deformities represent responses to toxic substances introduced into the aquatic environment by humans, such as industrial wastes, agricultural pesticides and herbicides, trace metals and radionuclides (Warwick 1985).

Table 7. A comparison of growth relationships of body length (BL), head length (HL), mesonotal width (MSW), abdominal segment 4 width (AS4W) with abdominal gill 4 width (G4W) and length (G4L) of *Stenacron interpunctatum* nymphs from the Econfina and Fenholloway rivers using regression analysis.

Nymphal Body Structures	Econfina River	Fenholloway River
Body Length		
G4W	9.799	6.391
G4L	6.636	4.429
Head Length		
G4W	1.444	0.854
G4L	0.973	0.580
Mesonotal Width		
G4W	2.207	1.272
G4L	1.514	0.878
Abdominal Segment 4 Width		
G4W	1.802	1.176
G4L	1.187	0.806

Gills of mayfly nymphs are involved in absorption of both oxygen and dissolved ions (Dodds and Hisaw 1924; Filshie and Campbell 1984; Beaver 1990; Eriksen and Moeur 1990). The abnormalities occurring in the nymphs of *S. interpunctatum* could be a function of the depressed dissolved oxygen and high BOD levels in the Fenholloway River. Dodds and Hisaw (1924) found a direct correlation between the relative gill area and the oxygen content of the water in which some mayfly nymphs lived. Nymphs that live in areas with low dissolved oxygen usually have larger gills than those that are found in a highly oxygenated environment. Eriksen and Moeur (1990) found that the dissolved oxygen intake by tracheal gills of the *Siphlonurus occidentalis* nymphs is proportional to the gill's fraction of the total respiratory surface area. Decreasing dissolved oxygen also triggers faster beating rates of the nymphal gills of some mayflies (Eriksen 1963; Nagell 1973, 1974). The enlargement of the abdominal gills, increased tracheation and number of branches of the fibrilliform portions of the nymphal gills of *S. interpunctatum* from the Fenholloway River appear to be a form of adaptive response to the extremely depressed oxygen and high BOD levels in the river. The enlargement of the gills not only increases the total respiratory surface area but also minimizes gill movements or beatings, thus conserving energy during respiration. Further investigation and experimentation are needed to explain the development of such abnormalities of the gill.

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