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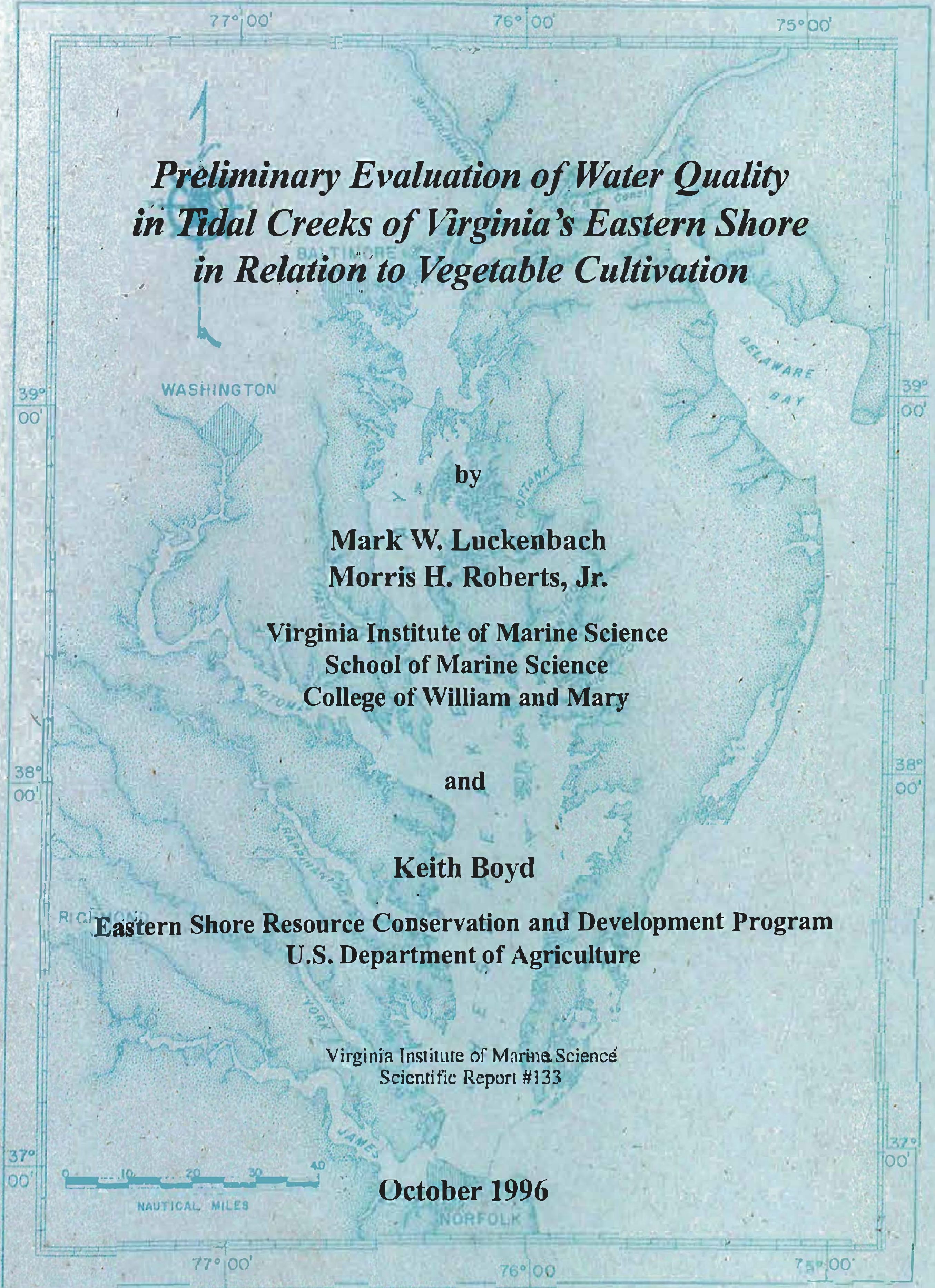

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Acknowledgments

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Summary

In response to concerns raised about the impacts of vegetable cultivation using plastic ground covers on water quality, we have initiated a broad-scale, systematic study of water quality in seaside tidal creeks of Virginia's Eastern Shore. Our objective was to determine if acute toxicity associated with heavy metals or pesticides was more prevalent in tidal creeks with drainage areas which include this agricultural practice than in those which do not. Though such correlations do not confirm cause and effect, they may serve as the basis for future, more targeted investigations and for some immediate changes in land management practices which, regardless of the specific cause, are likely to produce some remediation.

Eleven study sites, located in six different watersheds, were selected to evaluate acute toxicity from heavy metals and organic pesticides. Land use patterns and acreage within each watershed was determined from aerial photographs. The amount of vegetable plasti-culture in the watersheds of the study sites ranged from 0-13% of total acreage. An assay for heavy metals, based upon enzyme inhibition in a bacterial strain, was used to determine if up to seven metals (including copper) were present at acutely toxic levels. Both water samples and aqueous extracts of sediment samples were tested. A continuous series of 96 hr *in situ* bioassays using the grass shrimp, *Palaemonetes pugio*, were conducted from Aug. 1, 1996 - Sept. 22, 1996 at each station to assay for toxicity from organic pesticides. Grass shrimp are known to be quite sensitive to insecticides and the *in situ* bioassay approach provides a continuous means of monitoring for toxic events.

Results from the MetPAD™ assays demonstrate metals toxicity on one occasion in water samples from three creeks, two with vegetable plastic-culture and one essentially without on at least one occasion and a trace of metal toxicity at two other locations. The sensitivity of the test in saline waters such as these is comparable to that for freshwater, but is less than the sensitivity of bivalve embryos and larvae. The critical point is that though no particular metal is implicated by these data, there is evidence of metals toxicity.

The major finding of this study was a relationship between toxic events and rainfall with clearly different degrees of toxic impacts across watersheds. This observation is strongly suggestive of the importance run-off management. Some sites downstream of vegetable farms experienced moderate toxic events only after major storm events, while one site experienced complete mortality of test organisms after most rainfalls. This study does not indicate that vegetable farming with the use of plastic ground covers *per se* results in damage to aquatic ecosystems, but it does suggest that failure to provide some containment of run-off poses environmental threats. Based upon these preliminary findings we urge the implementation of good run-off management through (1) the elimination of direct ditching into tidal creeks, (2) the expanded use of vegetated buffer strips, and (3) where practical, the use of retention ponds for containing storm water run-off.

Introduction

The economic and cultural importance of both agriculture and the seafood industry on Virginia's Eastern Shore is well-known. Both have been mainstays of the local economy for generations and both are undergoing changes in some production methods. Cultivation of vegetables using plastic ground covers (often referred to as plasti-culture) as a means of controlling soil moisture, reducing pesticide requirements and increasing yields has been expanding in both counties on the Eastern Shore. Similarly, intensive culture of shellfish is expanding rapidly as an alternative to harvesting declining wild stocks of clams and oysters. These new practices in both industries provide opportunities for continuing economic development within the region.

One of the problems facing the seafood industry in recent years has been declining water quality. In the shellfish aquaculture industry water quality is a particular concern because the industry depends upon rearing very sensitive early life stages in hatcheries and nurseries adjacent to uplands and using water pumped from tidal creeks. Over the past several years many shellfish hatcheries have experienced water quality problems. At times these problems have been related to large-scale run-off in the greater Chesapeake Bay basin which reduced salinity and at other times to blooms of toxic dinoflagellates (Luckenbach *et al.* 1993). Recently concerns have been expressed by some in the shellfish aquaculture industry about the impacts of run-off into their immediate watersheds, particularly those from vegetable fields with plastic ground covers.

Among the effects observed in shellfish hatcheries have been chronic feeding inhibition and shell deformation in larvae and acute lethality to larvae and juveniles. The effects have been reported both by the industry and directly observed by one of us (Luckenbach). Though the source of toxic substances responsible for these problems have not been determined, the evidence is consistent with both heavy metal and organic (biogenic or anthropogenic) toxicant contamination.

The water quality problems experienced by some shellfish hatcheries raise broader environmental concerns related to impacts on living resources in tidal waters along Virginia's Eastern Shore. Because the existing evidence for impacts on water quality was limited in geographic distribution, poorly quantified and sometimes anecdotal, we have initiated a broad-scale, systemic study of water quality in tidal creeks on the Eastern Shore. In the initial phase of this study we sought to determine if a relationship exists between acutely toxic events in tidal creeks on the seaside of the Eastern Shore and the presence of vegetable plasti-culture in the watershed.

Relating water quality to specific land use can be a difficult task, particularly when many activities in a watershed may contribute the same or similar materials to non-point source inputs. In the case of this study, we have attempted to compare a few aspects of water quality in several creeks within the context of broad land uses in the watershed, with the premise that correlations between water quality and land use do not confirm cause and effect, but may serve as the basis for future, more targeted investigations.

Objectives

Our primary objective was to evaluate water quality in tidal creeks along the seaside of Virginia's Eastern Shore in relation to the presence or absence of vegetable cultivation using plastic ground cover within the watershed. Specifically, we sought to determine if acute toxicity associated with heavy metals or pesticides was more prevalent in tidal creeks with drainage areas which included this agricultural practice than in those which do not. Further, we sought to distinguish between impacts due to toxicity and those due to salinity or dissolved oxygen changes associated with run-off. Our goal was to provide a quantitative assessment of water quality at sufficient numbers of sites to permit correlative evaluation of the potential impact of plastic cultivation. We specifically did not set out to identify particular chemical sources of toxicity or to establish unequivocally the origin of suspected toxicant substances. In fact, we argue that such an approach would at this stage be too narrowly focused and fail to address the primary issue of establishing whether there are widespread water quality problems associated with this practice.

Methods

Site selection

We selected study sites in the headwaters of six tidal creeks on the seaside of the Eastern Shore (see Figs. 1-6) based on the following criteria: (1) a clearly definable watershed, (2) a minimum water depth of 1 ft at low tide, (3) salinities above 5‰ at low tide, and (4) accessibility by boat at high tide. Four of these creeks had vegetable (predominately tomato) cultivation using plastic ground covers within their watersheds; two other creeks were thought to lack any cultivation using plastic ground covers, but otherwise appeared to have similar land use. Two sites within Parting Creek, one with and one without tomato cultivation in the watershed, were initially selected but later abandoned when it was clear that the minimum depth criterion was not met. In addition to the headwater sites, five other sites were selected farther downstream of tomato cultivation. The Gargathy Creek 2 (**G2**) site was located about 1 km downstream from **G1** near the site of a shellfish hatchery which has experienced water quality problems (see Fig. 2). Folly Creek sites **FO2** and **FO3** were located along a transect from the creek headwaters to near the ocean inlet (Fig. 3). Finney Creek (**F**), which drains an area containing vegetable cultivation, and Nickawampus Creek (**N**), which has no plastic cultivation, merge into Wachapreague Channel about 1 km upstream of the **W1** site (Fig. 4). **W1** was located in Wachapreague Channel near the VIMS Eastern Shore Laboratory where we have a long record of good water quality and successful bivalve culture. **W2**, located in a marsh creek near the inlet to Wachapreague Channel (Fig. 4), was the collection site for grass shrimp used in the bioassays described below. The Phillips Creek site (**P**, Fig. 5) was located in the headwaters of a creek without any tomato cultivation in the watershed and the Indiantown Creek site (**I**, Fig. 6) was located downstream of a large tomato field.

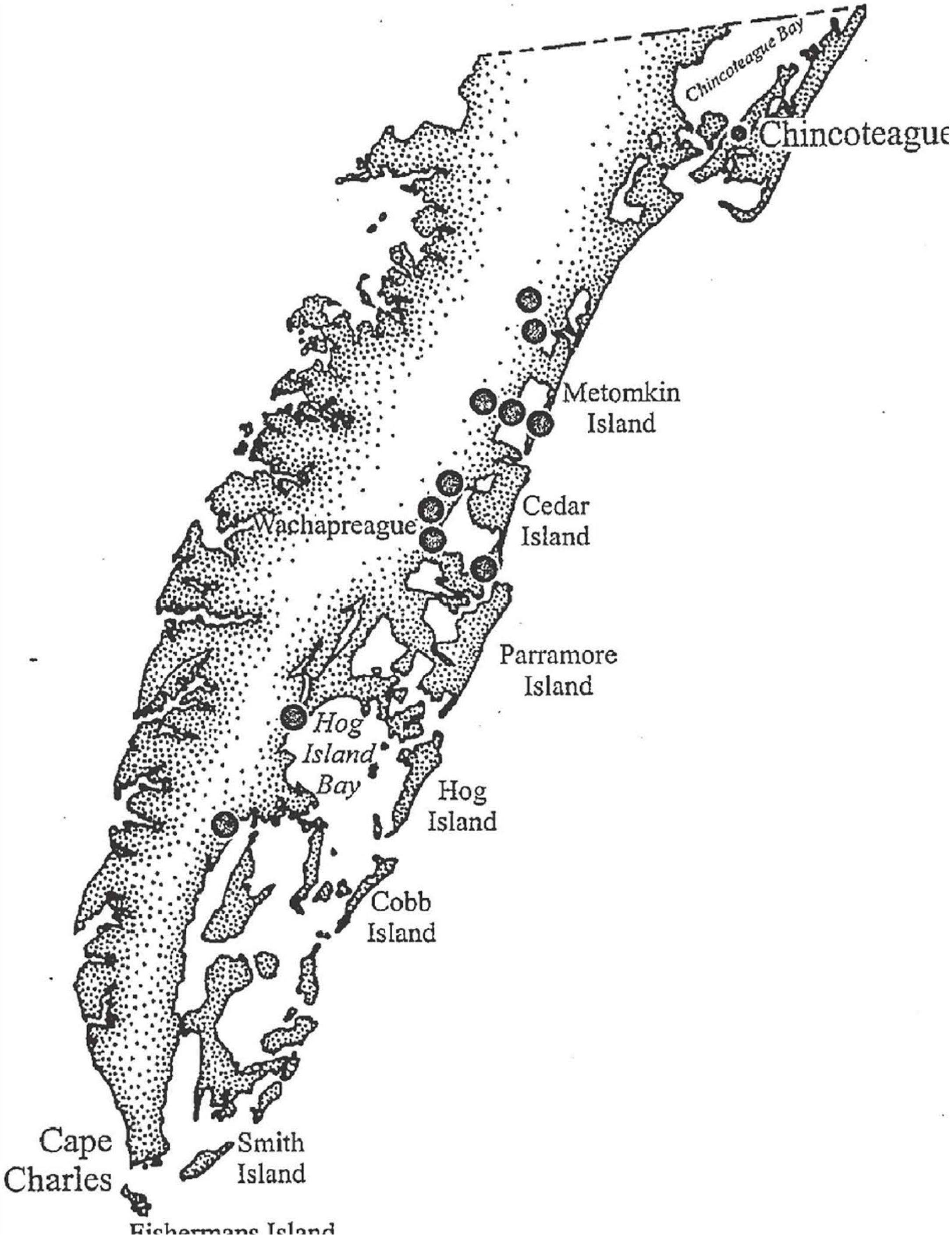


Figure 1. Study sites for *in situ* bioassays and collection of samples for metals toxicity tests.

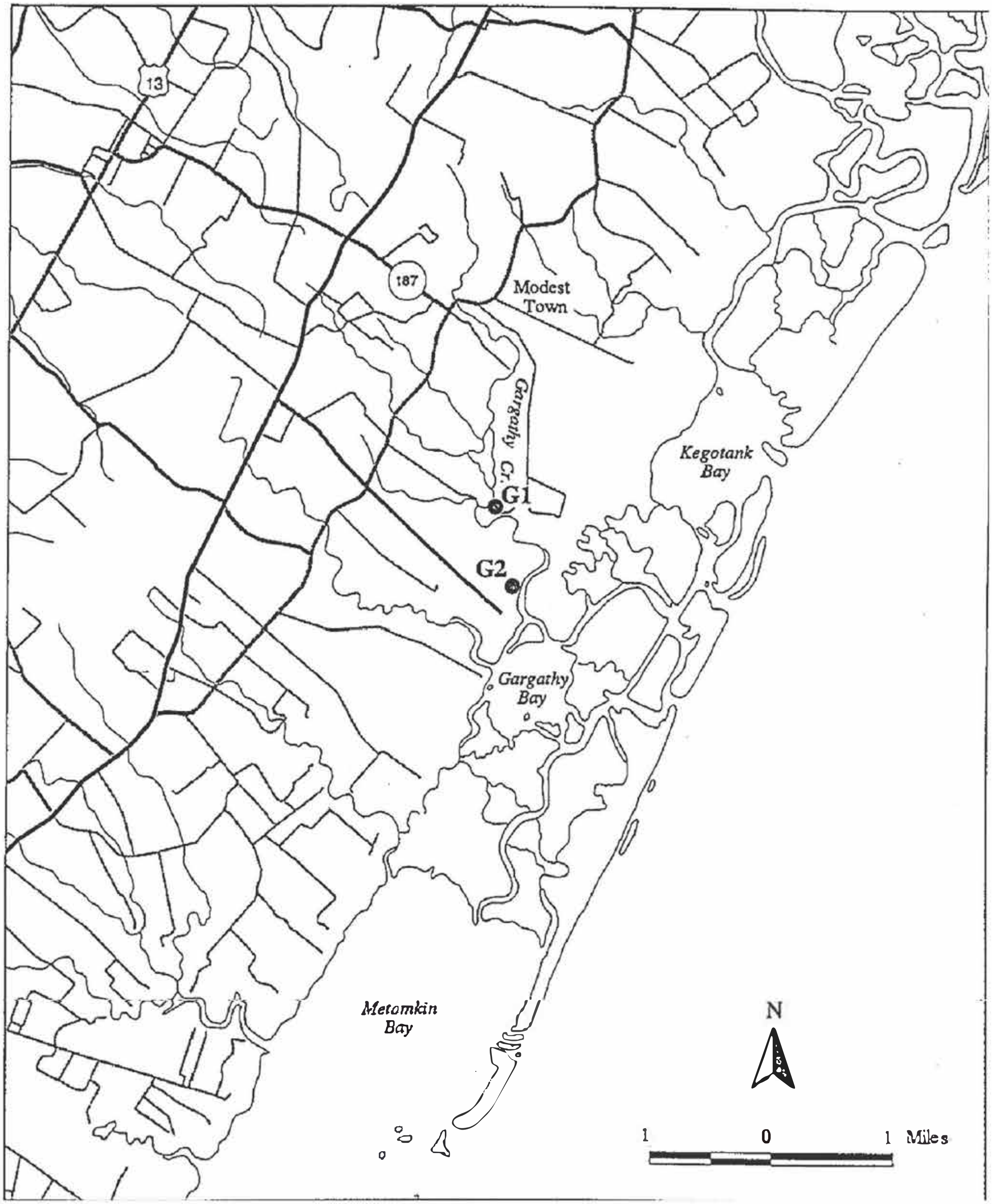


Figure 2. Gargathy Creek study sites. G1 indicates the headwater site near a tomato field and G2 indicates the downstream site near a shellfish hatchery

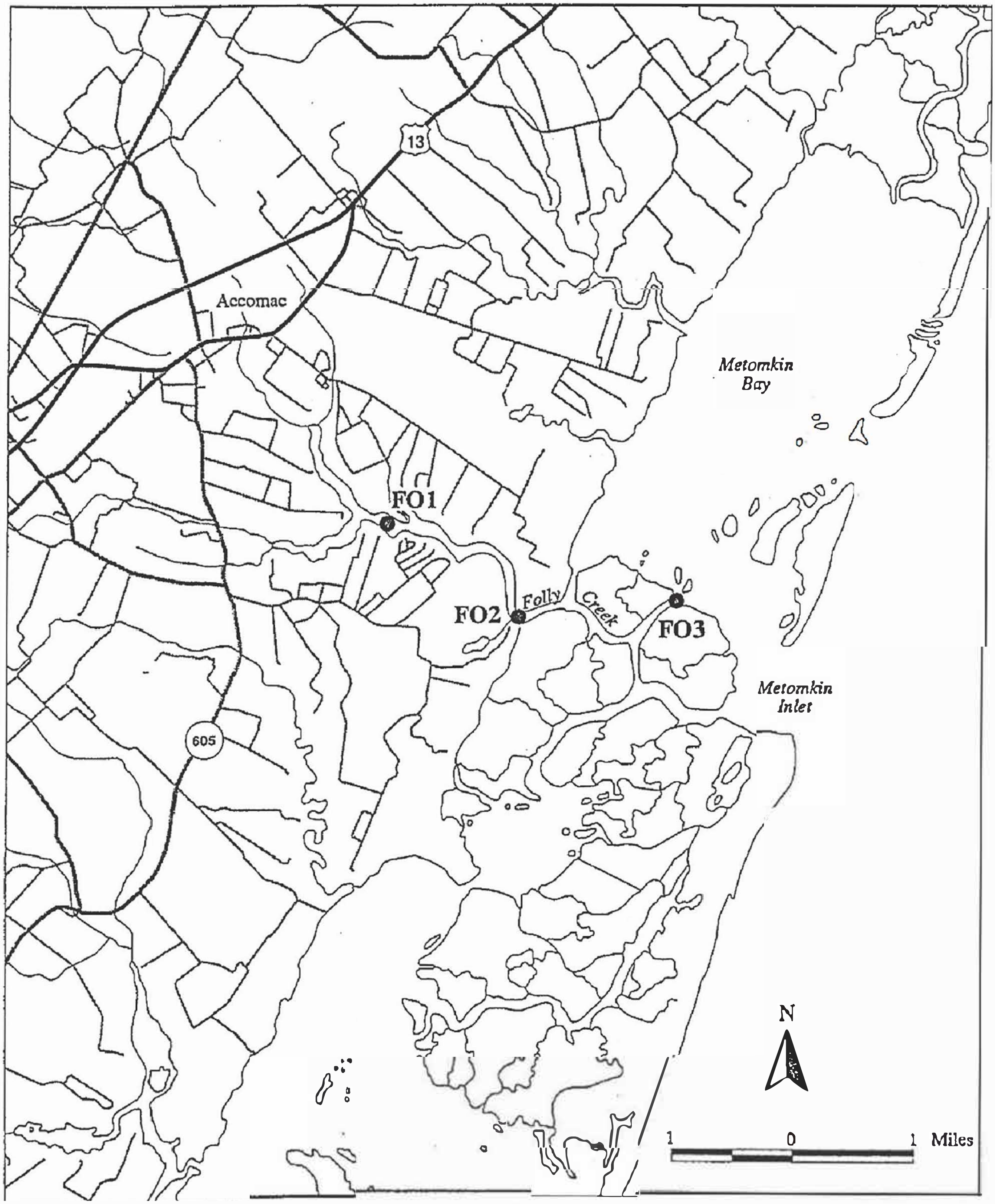


Figure 3. Folly Creek study sites. FO1, FO2 and FO3 lie along a transect from the headwaters to the inlet

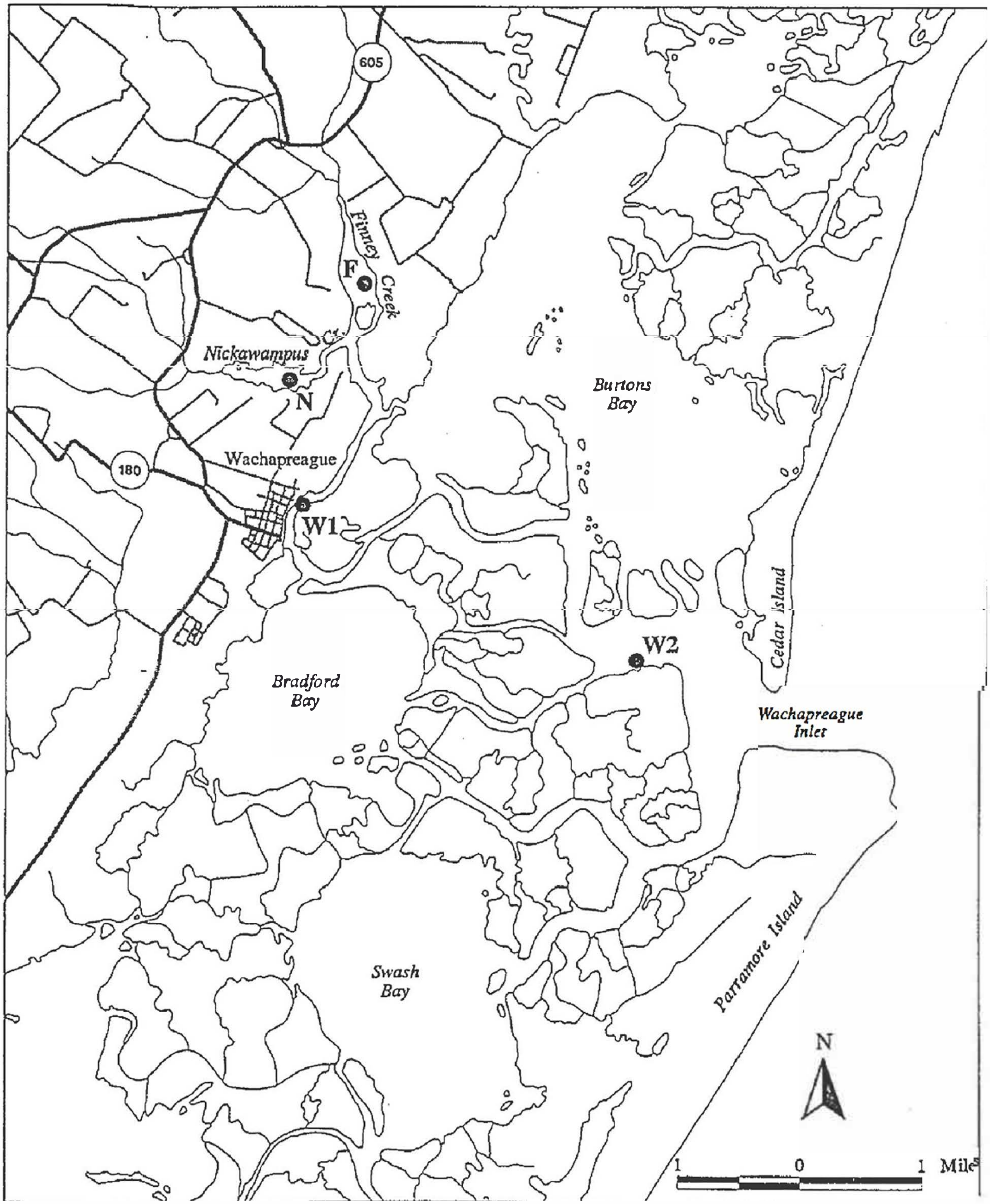


Figure 4. Study sites in Finney Creek (F), Nickawampus Creek (N), Wachapreague Channel (W1), and Wachapreague Inlet (W2)

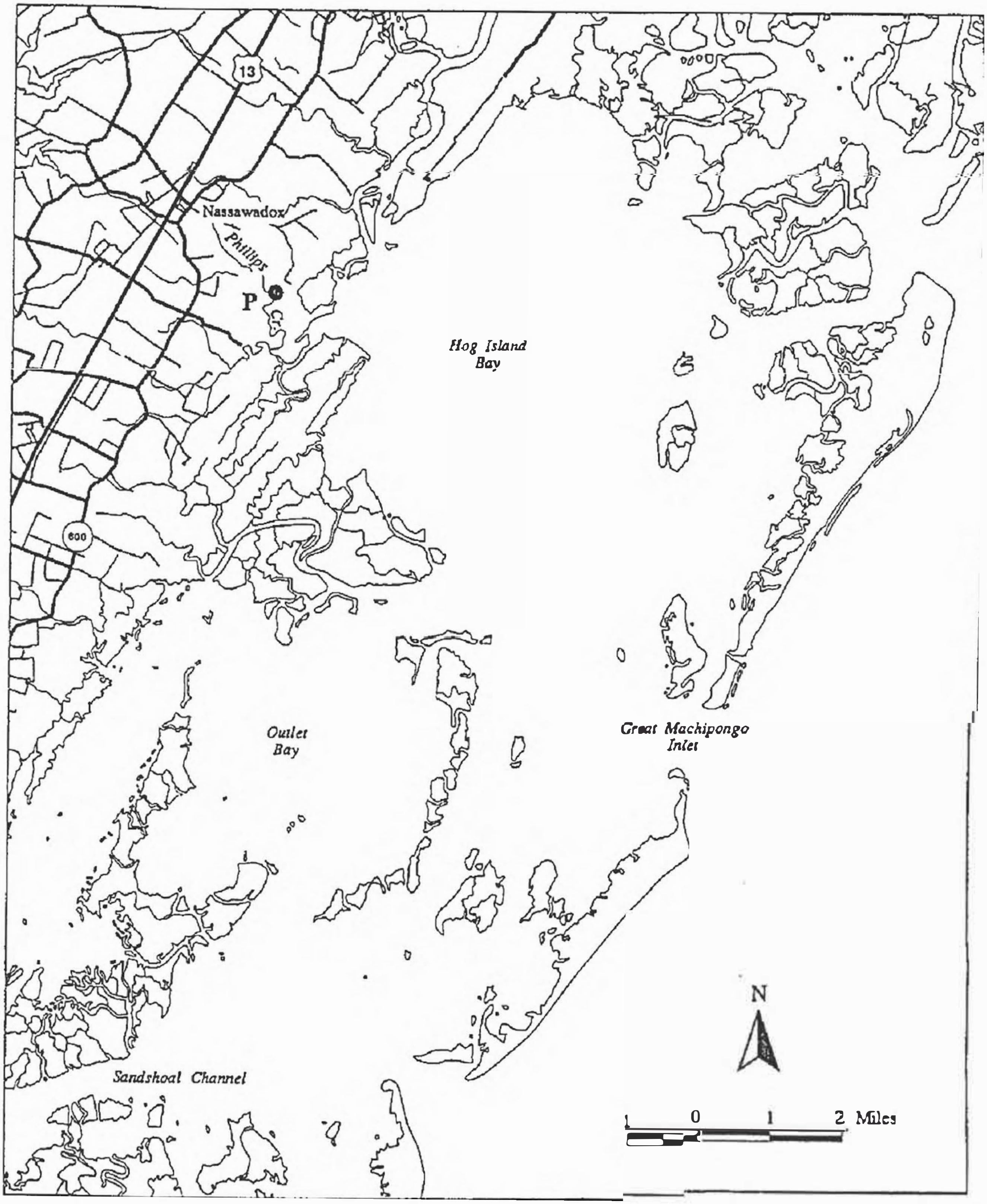


Figure 5. Phillips Creek Study site (P)

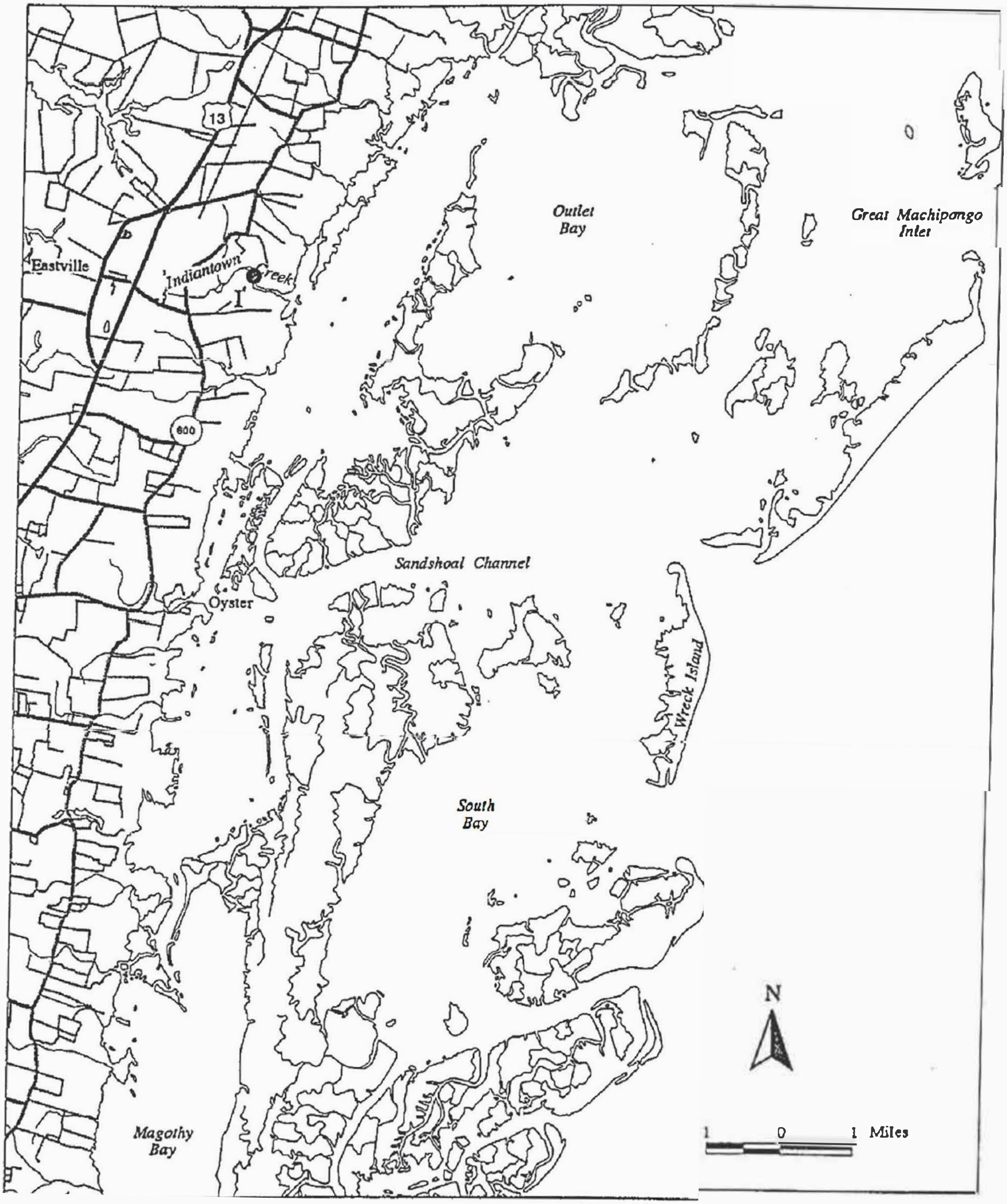


Figure 6. Indiantown Creek Study site (I)

Characterization of watersheds

Watershed boundaries were interpolated from U.S. Geological Survey 7.5 minute topographic quadrangle maps. Elevations for these quadrangles were developed in 1968 with photo revisions in 1979 and are expressed with 5 ft contour intervals. Watersheds were outlined on these maps and the total acreage estimated with a planimeter.

Land use within the watersheds was characterized using aerial photographs taken by the USDA, Farm Services Agency in June 1995. First, we used color slides to distinguish plastic culture from other agriculture within the area and categorized land use into one of five categories: urban/residential, woodland, marsh and open water, total cropland and plastic culture. Actual land use acreage was then determined from rectified, black and white, National Photography Program (NAPP) photos using a planimeter.

Physical parameters

Rain gauges placed within or near each of the watersheds were used to collect rainfall data over 48 hr periods throughout the study. Three battery-powered, submersible water quality sensors were deployed at selected sites at various times throughout the study (see Table 1). These sensors (a DataSond™ manufactured by Hydrolab and two analogous meters manufactured by YSI) measured temperature, salinity, dissolved oxygen (for the YSI™ meters only), turbidity and pH at 15 min intervals and recorded them to an internal data logger. In addition to these semi-continuous records, we measured salinity and water temperature with hand held instruments on

Table 1. Station locations, deployment dates and parameters measured using Hydrolab Datasond™ - and YSI meters.

Station	Deployment Dates	Parameters Measured
Gargathy Creek, upstream (G1)	8/29 - 9/2	temperature, salinity, D.O.
Gargathy Creek, upstream (G1)	9/18 - 9/22	temperature, salinity, D.O., turbidity, pH
Finney Creek (F)	9/6 - 9/10	temperature, salinity, turbidity, pH
Finney Creek (F)	9/18 - 9/22	temperature, salinity, D.O., turbidity, pH
Folly Creek, upstream (FO1)	9/18 - 9/22	temperature, salinity, turbidity, pH
Indiantown Creek (I)	9/6 - 9/10	temperature, salinity, D.O., turbidity, pH

every visit to all sites throughout the study.

Metal toxicity assays

As noted above, some of the problems experienced by bivalve hatcheries are consistent with heavy metal toxicity. Tomato cultivation in this region relies upon the application of copper sulfate as a bactericide, but it is unknown whether there are other heavy metal inputs into the watersheds. Thus, rather than specifically targeting copper (and possibly missing other sources of metal toxicity), we employed a commercially available assay which is specific for heavy metals toxicity, MetPAD™ (Bitton, et al., 1992). This procedure is based upon inhibition of β-galactosidase in a non-pathogenic strain of *Escherichia coli* exposed to aqueous solutions of metals at environmentally relevant concentrations (Table 2). This assay is not sensitive to pesticides or polycyclic aromatic hydrocarbons (Bitton, et al., 1994).

Table 2. Minimum inhibition concentrations for various metals in the MetPAD™ assay (from Bitton, et al., 1992).

Metal	Minimum Inhibition Concentration (mg/L)
Cadmium	0.3
Chromium	25.0
Copper	0.5
Mercury	0.5
Lead	5.0
Nickel	8.0
Zinc	0.5

Water column and sediment samples for these assays were taken from all sites on August 13, September 10, and September 22. Water samples were taken approximately ½ m below the surface and sediments were collected from the upper 3 cm. Water samples were analyzed both with and without filtration through 0.45 µm fiber glass filters to evaluate any effect of the sometimes high suspended solids loads. Five gram portions of oven-dried sediment samples were extracted with 20 ml distilled water shaken for two hours at room temperature and centrifuged for 30 minutes. The centrifugate was then used for analysis. This extraction procedure was strong enough to strip bioavailable metals, whereas acid extraction would have come closer to stripping off all metals from the sediment.

Briefly, the MetPAD analysis involves incubating the test bacterium for 1½ hours in control solutions with and without metals or unknown samples, spotting droplets of the incubated samples on assay pads and incubating the pads an additional 1 to 1½ hr. If there is no toxic response, the spots turn a distinct red/purple color indicating enzyme activity. In samples containing toxic levels of metals, the sample area remains yellow indicating a lack of enzyme activity.

A limited number of samples were assayed to determine whether the test method is equally sensitive in fresh water (the medium in which it was developed) and salt water. Tests for sensitivity were limited to copper, cadmium, and lead. Only for cadmium was sensitivity reduced (at least 10-fold), a result anticipated because cadmium forms chloride complexes that are not bioavailable, thus reducing the apparent toxicity of cadmium to saline organisms (Sunda et al., 1978, DeLisle and Roberts, 1988).

In situ Bioassays

To evaluate the potential water quality problems associated with insecticide and other inputs into tidal creeks, we conducted *in situ* bioassays using the grass shrimp *Palaemonetes pugio*. We adopted this method (as opposed to direct water analyses for insecticides) to fulfill the need for a sensitive, continuous measure of toxicity. Since inputs of pesticides and other materials from upland sources into tidal waters will vary with application schedules, rainfall events and tidal stage, and further, concentrations may be ephemeral, it was necessary to employ an evaluation procedure which integrates water quality over time. *P. pugio* is a common inhabitant of tidal mashes and creeks. This species has a wide salinity and dissolved oxygen tolerance and is relatively robust with respect handling tolerances. This species has been shown, however, to be quite sensitive to insecticides (Baughman, 1986; Scott, *et al.*, 1990 , see Table 3). Decapods such as *P. pugio* are also known to be sensitive to various heavy metals as well as other materials in one or more life stages (Bradley and Roberts, 1987).

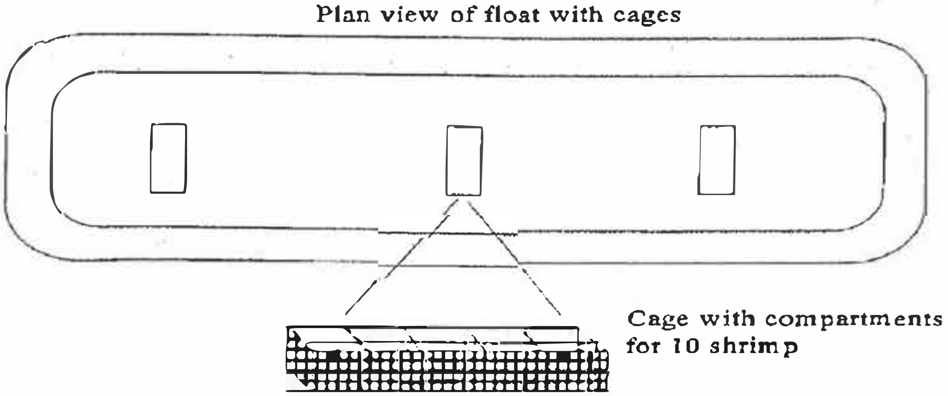
Table 3. **Acute toxicity levels for adult *Palaemonetes pugio* to selected pesticides. Values are concentrations which cause mortality to 50% of the animals in 96 hr laboratory tests conducted in 20‰ seawater. Values in parenthesis are 95% confidence intervals. (Data from Scott et al. 1990).**

Insecticide	96 hr LC_{50} (95% CI) in mg/L
Azinphosmethyl	1.05 (0.91-1.21)
Endosulfan	1.01 (0.72-1.43)
Fenvalerate	0.052 (0.043-0.063)

Field bioassays were conducted using *P. pugio* in the following manner. Adult shrimp were collected in the creeks and marshes adjacent to site W2, transported to the lab and held in aquaria in filtered seawater adjusted to 25‰ salinity. Collections were made throughout the study and shrimp were held for no more than two days prior to use. Shrimp were not fed during this holding period, but some cannibalism apparently occurred.

On field deployment days one adult shrimp (gravid females excluded) was placed into each of ten small exposure cells within a wire mesh cage (Fig. 7). Cages were constructed of 1/16 inch mesh galvanized wire with styrofoam lids for floatation. After filling the cages with shrimp, they were placed in large coolers filled with seawater for transport to field sites. At each of the field sites described above three grass shrimp cages were tethered within a larger float (2 ft x 8 ft PVC ring with an attached 1 ft deep wire basket made of 1 inch mesh galvanized wire) (Fig. 7). These larger floats provided a secure, floating mooring for the small cages. They are also widely recognized in the region as oyster culture floats and generally not tampered with. Thus, at each deployment, each site received 30 *P. pugio* (10 shrimp/cage x 3 cages/float). Two days after deployment each cage was inspected, all shrimp categorized as live, dead or missing and the cage returned to the water. On day 4 the cages were retrieved, the status of the shrimp again noted and new cages with new shrimp deployed. A continuous series of 96 hr *in situ* bioassays were conducted in this manner from August 1 to September 22, 1996.

Figure 7. Schematic of deployment technique for *in situ* bioassays.



Results

Land use patterns - Each of the six watersheds contained some acreage in four of the categories (urban/residential, woodland, marsh and open water, and cropland). Four watersheds had plastic culture acreage varying from 5 to 13% of the watershed and among the two sites which were selected to have no plastic culture, one of the sites was found to contain <1% of the total acreage in plastic culture (Table 4).

Table 4. **Land use patterns within the watersheds for each upstream station.**

Site	Watershed Acreage (% total) marsh & open water	woodland	urban/ residential	cropland	plastic culture	Total Acreage
Phillips	143 (11%)	429 (32%)	50 (4%)	704 (53%)	0	1326
Nickawampus	140 (5%)	1167 (38%)	145 (5%)	1616 (51%)	25 (‹ 1%)	3093
Indiantown	15 (1%)	448 (35%)	55 (4%)	608 (47%)	175 (13%)	1301
Gargathy	247 (7%)	1161 (33%)	128 (4%)	1782 (51%)	179 (5%)	3497
Finney	125 (2%)	2549 (40%)	200 (3%)	2983 (46%)	597 (9%)	6454
Folly	263 (6%)	1413 (32%)	306 (7%)	2123 (48%)	301 (7%)	4406

Physical parameters - One of the YSI meters malfunctioned and failed to give accurate measures for dissolved oxygen. Those stations affected by this loss are noted in Table 3. Although values for both salinity and dissolved oxygen varied widely at different sites throughout the study (see Figs. 8 - 11), levels lethal to *P. pugio* were observed only once. At the G2 station in Gargathy Creek dissolved oxygen fell below 1 mg/L for 1 hr between 5:38 and 6:38 AM on the morning of 15 Sept. (Fig. 8b). Although these levels, if sustained, may be toxic to *P. pugio*, mortality of shrimp was not observed in at that station at that time (see Fig. 20). Less severe hypoxia (1 mg/L < D.O. < 2 mg/L) was recorded at G2 between 5:32 and 8:38 AM on 16 Sept. and at G1 between 4:15 and 7:15 AM on 30 Aug. (Fig. 8a &b), but these levels are not acutely lethal to *P. pugio*.

Rainfall data for the individual sites are reported together with shrimp survival in Figs. 13 - 23. Both total rainfall and the time of rainfall events varied between watersheds. Rainfall in excess of 50 mm within 48 hrs was recorded at Gargathy and Folly creeks on Sept. 12 and at Indiantown Creek on Aug. 25 and Sept. 12 (see Figs. 18-19 & 21-22). Although there were some heavy rainfall events during the study, average rainfall for the month of August was close to the 50 year average; September rainfall was approximately 50% above the 50 year average (Table 5). It is noteworthy that during the month of July, prior to the initiation of this study, that rainfall was over 3 times the 50 year average for the area (Table 5).

Table 5. Rainfall recorded at Painter, Virginia, 1996 and 50-year average values for July - September. (Data supplied by the Eastern Shore Agricultural Research and Extension Center, Virginia Tech University).

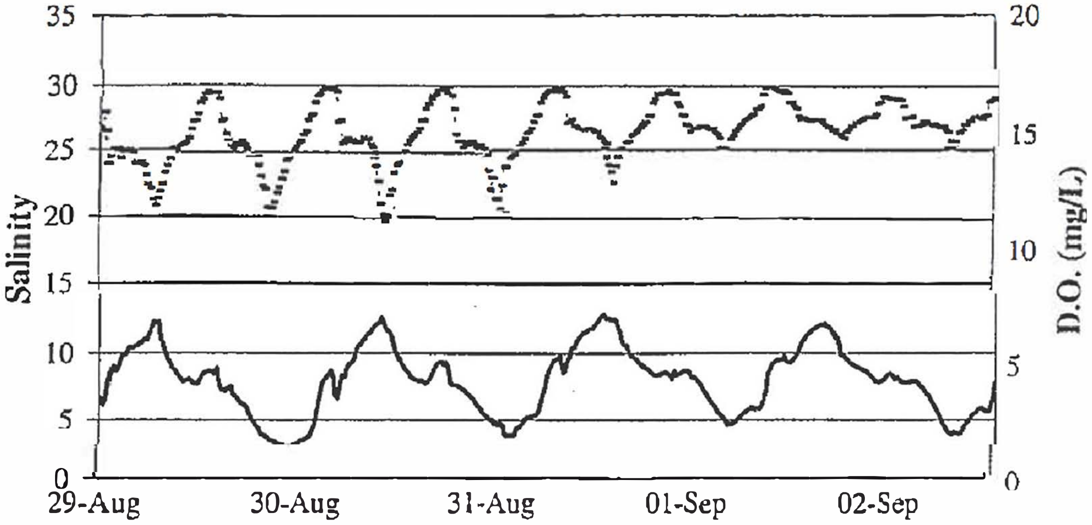
Month	1996 (amount in mm)	50-Year Average (amount in mm)
July	365.0	108.7
August	105.7	109.7
September	129.3	86.9

Metal toxicity - Analysis of the complete array of samples collected has been delayed because the manufacturer of the kit has on two occasions delivered lots of bacterial reagent that were already killed as a result of a freezer failure in their facility. We have a complete set of results for unfiltered water and sediment samples for August 13 and a selection of filtered samples for August 13 and September 10. These data are portrayed in Fig. 12.

Evidence of metals toxicity was observed in the filtered water sample collected from Gargathy Creek at the upstream site on September 10, with lesser toxicity observed at the midstream site. Metals toxicity was also observed in filtered water samples collected from Finney and Nickawampus Creeks on August 13. A trace of toxicity was observed in an unfiltered water samples from the Wachapreague lab station and Indiantown Creek, also on August 13 ("trace of toxicity" in this case means a slight reddening of the spot). Negative controls for all of these samples produced no color change, indicating that there was no contamination of the reagents with any metals.

Fiqure 8. Salinity and dissolved oxygen at the Gargathy Creek upstream station (G1) during the periods (a) 8/29-9/2 and (b) 9/18-9/22. (dash line = salinity; solid line = dissolved oxygen)

(a)



(b)

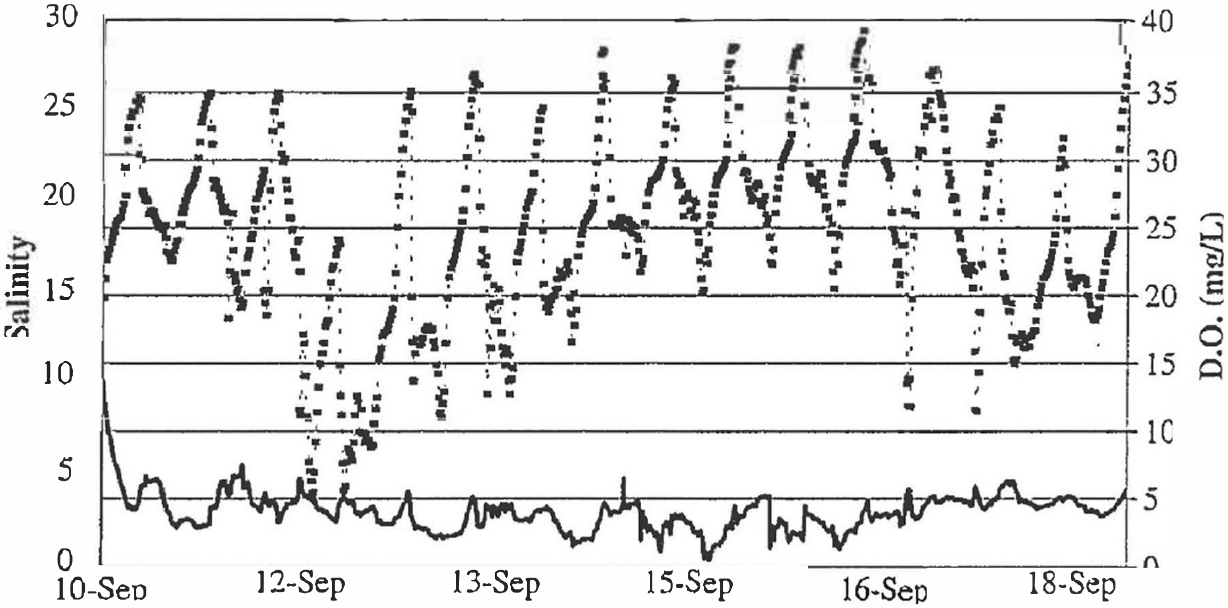
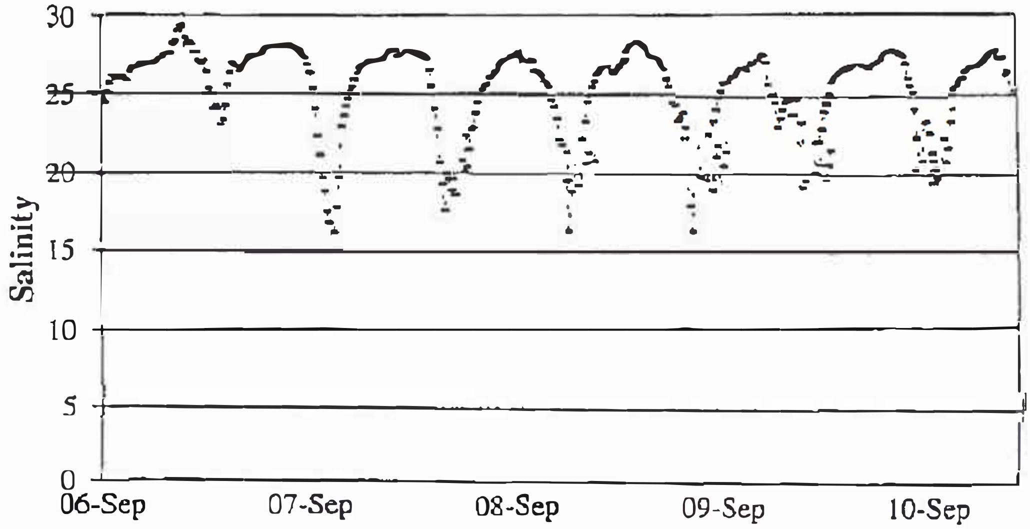


Figure 9. Salinity and dissolved oxygen at the Finney Creek station (F) during the periods (a) 9/6-9/10 and (b) 9/18-9/22. (dash line = salinity; solid line = dissolved oxygen)

(a)



(b)

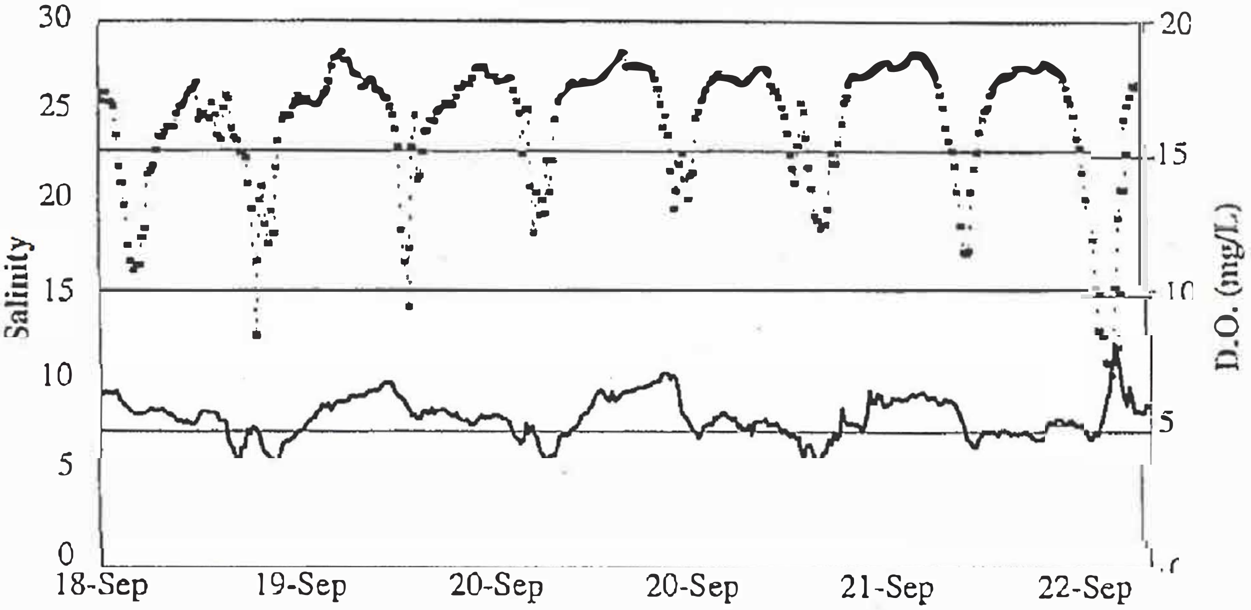


Figure 10. Salinity at the Folly Creek upstream station (FO1) from 9/18-9/10.
(dash line = salinity; solid line = dissolved oxygen)

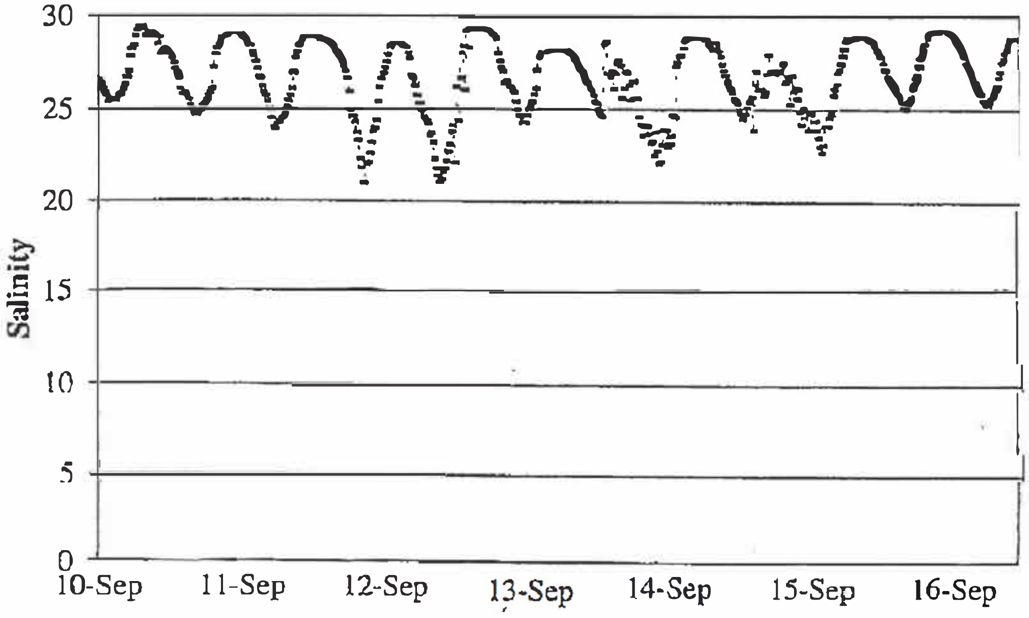


Figure 11. Salinity and dissolved oxygen at the Indiantown Creek station (I) from 9/6-9/10.
(dash line = salinity; solid line = dissolved oxygen)

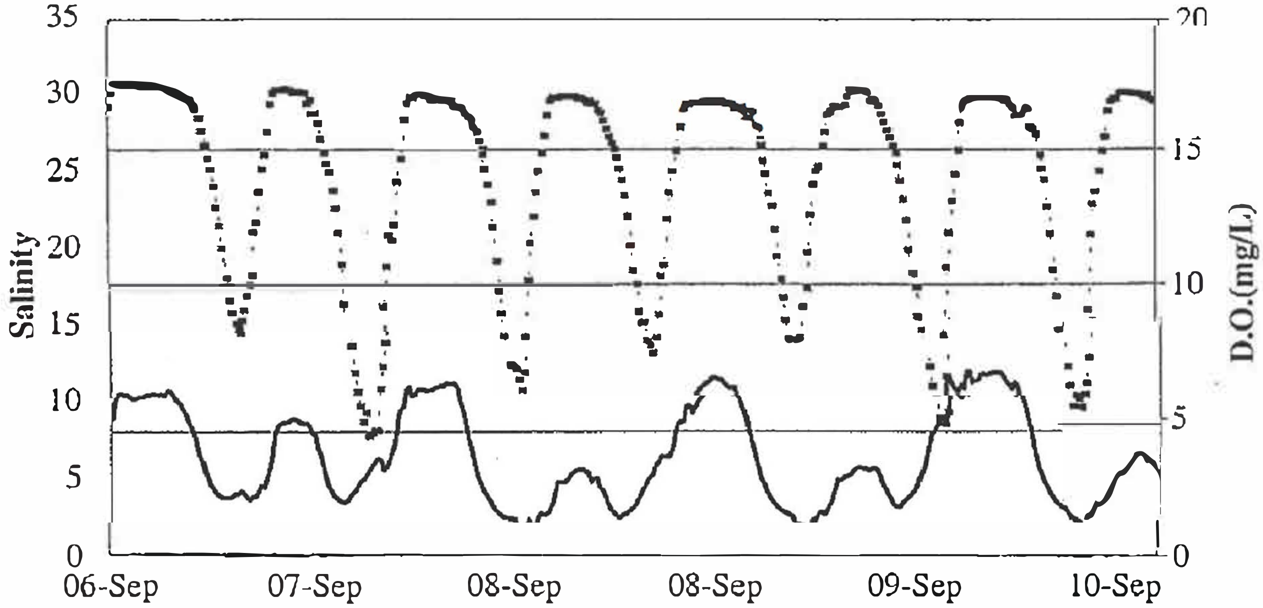


Figure 12. Occurrence of metals toxicity in water and sediment samples as indicated by the MetPAD™ Test. Blank locations indicate that sample analyses are incomplete.

		8/13		9/10		9/22	
		U	F	U	F	U	F
Gargathy, Upstream	Water	○	○		●		
	Sediment	○		○			
Gargathy, Midstream	Water	○	○		◉		
	Sediment	○		○			
Folly, Upstream	Water	○	○		○		
	Sediment	○					
Folly, Midstream	Water	○			○		
	Sediment	○					
Folly, Downstream	Water	○					
	Sediment	○					
Finney	Water	○	●		○		
	Sediment	○					
Nickawampus	Water	○	●		○		
	Sediment	○					
Wachapreague Lab	Water	○					
Wachapreague, Field	Water	○					
	Sediment	○					
Wachapreague Inlet	Water	○					
	Sediment	○					
Phillips	Water	○			○		
	Sediment	○					
Indiantown	Water	○			○		
	Sediment	○					

Metal Toxicity	
None	○
Trace	○
Slight	◉
Distinct	●

In situ bioassays - Some loss of shrimp occurred during the early part of the study due to escape from the cages and positioning of some cages in too shallow portions of creeks. Also, the Folly Creek station located near Metompkin Inlet (**FO3**) was subject to strong wind- and tidal-driven currents which frequently disturbed the cages and subjected the shrimp to physical damage. These data have been omitted this report. Only data for deployments resulting in exposure for a full 96 hr are included herein.

Survival of shrimp in the laboratory control and at the **W2** field control site was generally between 90 and 100% throughout the study, with only two instances below 80% survival (Figs. 13 & 14, respectively), indicating that our handling procedures were appropriate.

Phillips Creek, which lacked any plastic culture within its watershed, had good survival throughout the study period, always exceeding 85% (Fig. 15). No effect was evident after a 45 mm rainfall recorded on Sept. 12.

Survival of *P. pugio* at the Nickawampus Creek site was more variable, ranging from 100 to <50% (Fig. 16). Survival of the shrimp was reduced at this site for several days following a large rain event on Aug. 27, but no effect was evident after a large rain on Sept. 12. This site, originally selected to have no plastic culture, did in fact have a minimal amount of the watershed covered in plastic culture (<1%). However, the primary difference between this watershed and that of Phillips Creek would appear to be the total acreage, which is some roughly 3x greater for Nickawampus, and the smaller proportion of marshlands at Nickawampus (Table 4). We interpret these results as revealing a greater impact of the watershed on water quality at this site, but think that it is unlikely to be related to the presence of plastic vegetable culture.

Finney Creek (**F**) lies adjacent to and has a drainage area about twice the size of Nickawampus (Fig. 4, Table 4). Five percent of its watershed is occupied with plastic cultivation of vegetables and, perhaps more importantly a large field lies adjacent to the creek just upstream of the sample site. Survival of *P. pugio* at this site was generally above 80%, with significant mortalities occurring after large rainfalls (Fig. 17). Following a 38 mm rainfall recorded on Aug. 27, shrimp survival was below 35% for 4 days, indicating a sustained toxic event. Less impact was observed after rainfalls between 20 and 25 mm (Fig. 17).

The site in Wachapreague Channel (**W1**) is located about 1 km downstream of the confluence of Nickawampus and Finney Creeks. As noted earlier, it is a site at which we have over 30 years of experience in culturing bivalve larvae and have generally found that water quality is excellent. Indeed, survival of grass shrimp was good throughout the study, dropping to 80% on two occasions which coincided with large rainfall events (Fig. 18). It is evident that most of the watershed effects noted in Nickawampus and Finney Creeks have been ameliorated at this downstream site.

The Folly Creek sites (**FO1** & **FO2**) also generally had good survival (Figs. 19 & 20, respectively). At the upstream site (**FO1**) mortalities greater than 20% were observed on only two occasions—Aug. 5 and Sept. 10. These losses may have been associated with rainfall events, but the pattern is equivocal (Fig. 19). Survival at **FO2** approached 50% on Sept. 6 and was low for a week (Fig. 20), but we suspect that high winds associated with Tropical Storm Fran were responsible for damaging shrimp at this site because of its greater exposure to wind and waves.

The two sites in Gargathy Creek (**G1** & **G2**) both experienced significant mortalities (Figs. 21 & 22, respectively). At **G1** rainfall events of any magnitude were followed by complete mortality of *P. pugio* (Fig. 21). Though a rain gauge was not installed in this watershed until Aug. 3, we know from gauges elsewhere (e.g., see Fig. 18) the a major rain event occurred on Aug. 1. This was followed by mortalities at **G1** recorded on Aug. 3 and 5. Each subsequent rainfall event which we recorded here was associated with shrimp mortality. Data from our *in situ* meters (Figs. 8a & 8b) and manual measurements do not reveal lethal salinities or dissolved oxygen levels at the site, so we interpret these mortalities to be associated with inputs of toxicant substances. The downstream site **G2** had similar patterns of survival associated with rainfall events, but mortalities were never complete at this site (Fig. 22).

The Indiantown Creek watershed contains the greatest proportion of plastic cultivation among the sites in the study (Table 4). Survival of grass shrimp at the site varied considerably from 0 to 100%; however, mortalities greater than 50% were observed only after rainfall >50 mm/48 hr (Fig. 23). Smaller rainfall amounts of 10 - 20 mm/48 hrs did not result in noticeable toxic events.

Figure 13. Survival of *P. pugio* in 96 hr bioassays in laboratory control tank.

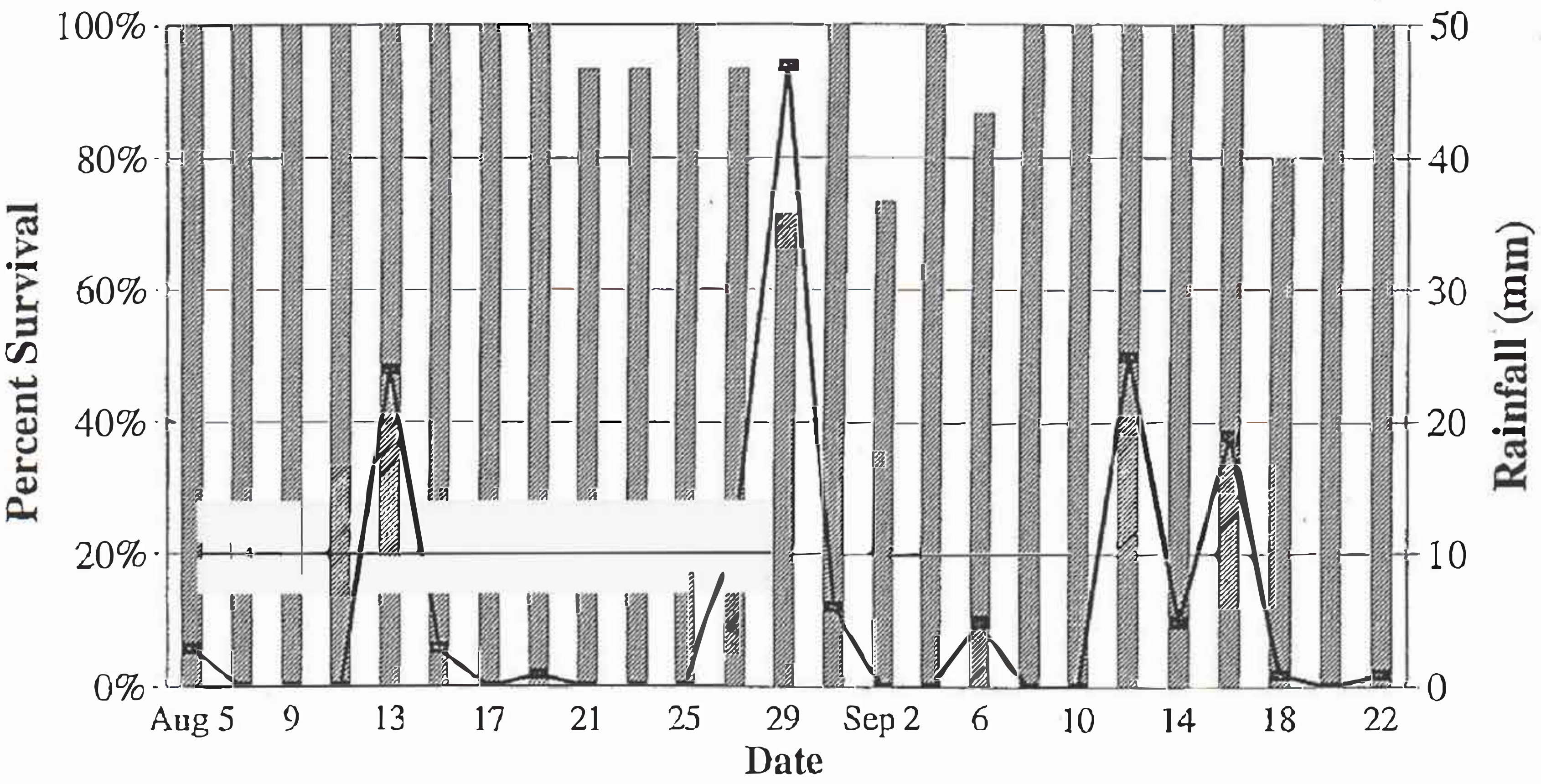


Figure 14. Survival of *P. pugio* in 96 hr bioassays at the Wachapreague 2 site (W2) and rainfall for approximately 48 hr period preceding the sampling date.

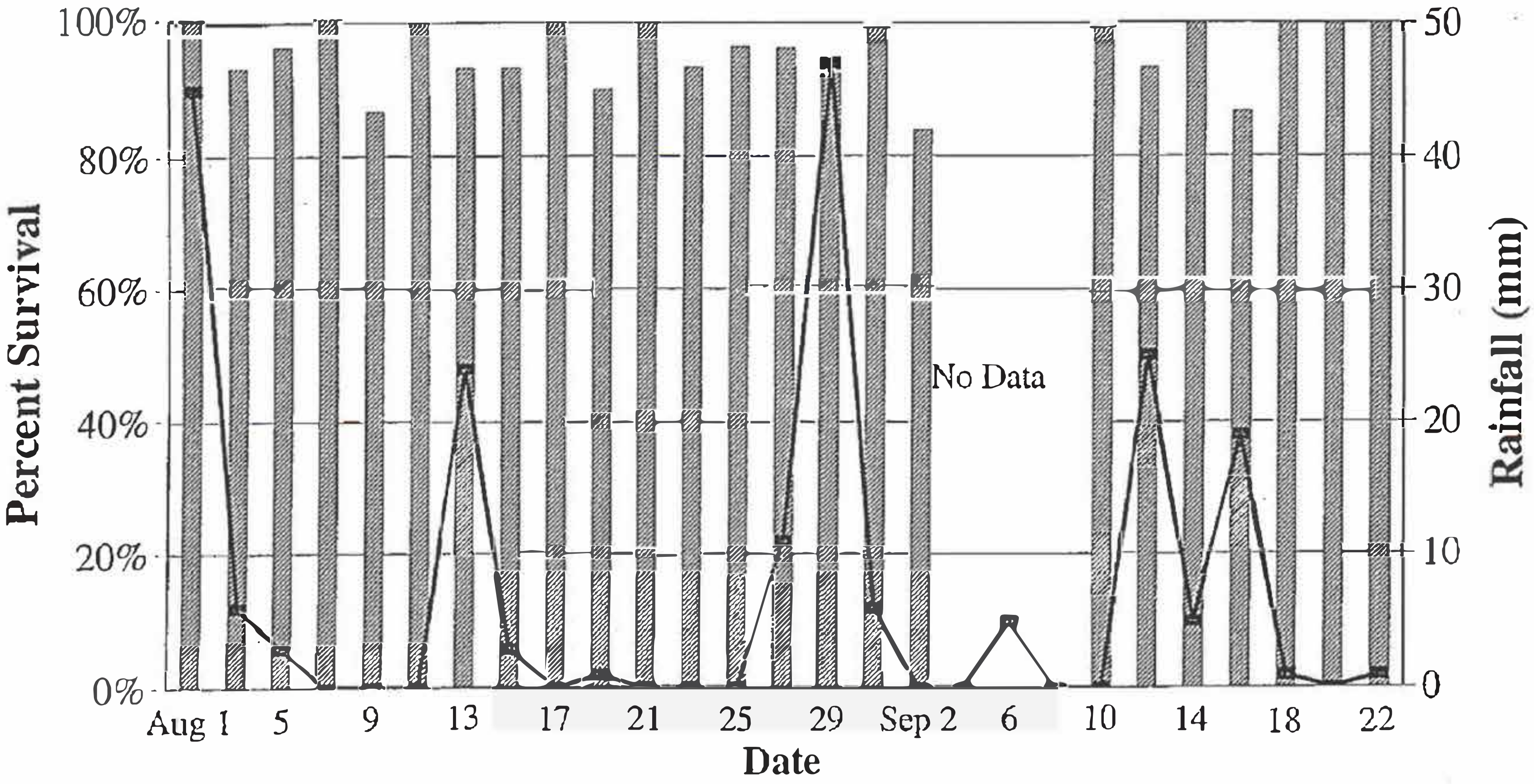


Figure 15. Survival of *P. pugio* in 96 hr bioassays at the Phillips Creek site (P) and rainfall for the approximately 48 hr period preceding the sampling date.

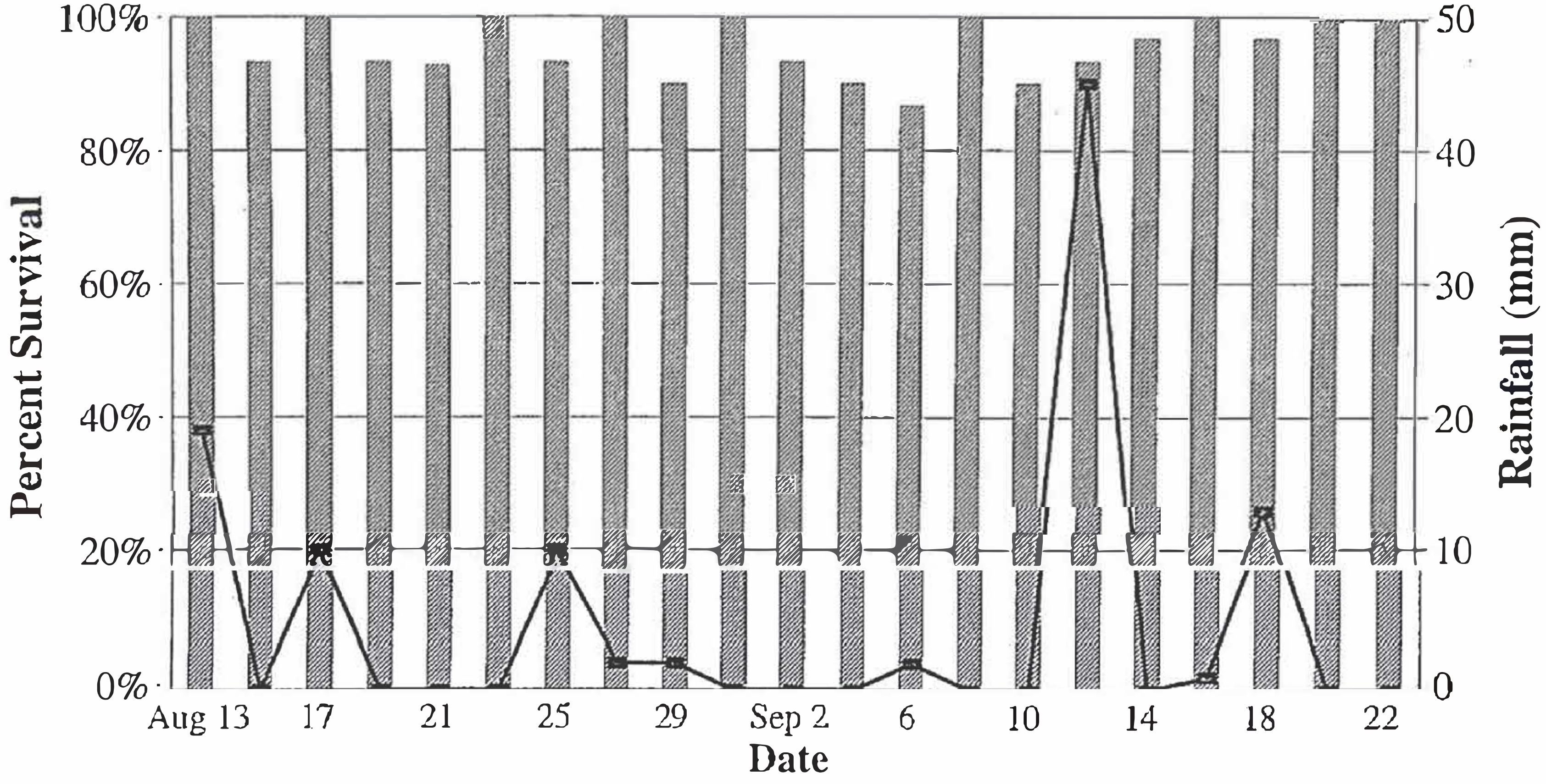


Figure 16. Survival of *P. pugio* in 96 hr bioassays at the Nickawampus Creek site (N) and rainfall for the approximately 48 hr period preceding the sampling date.

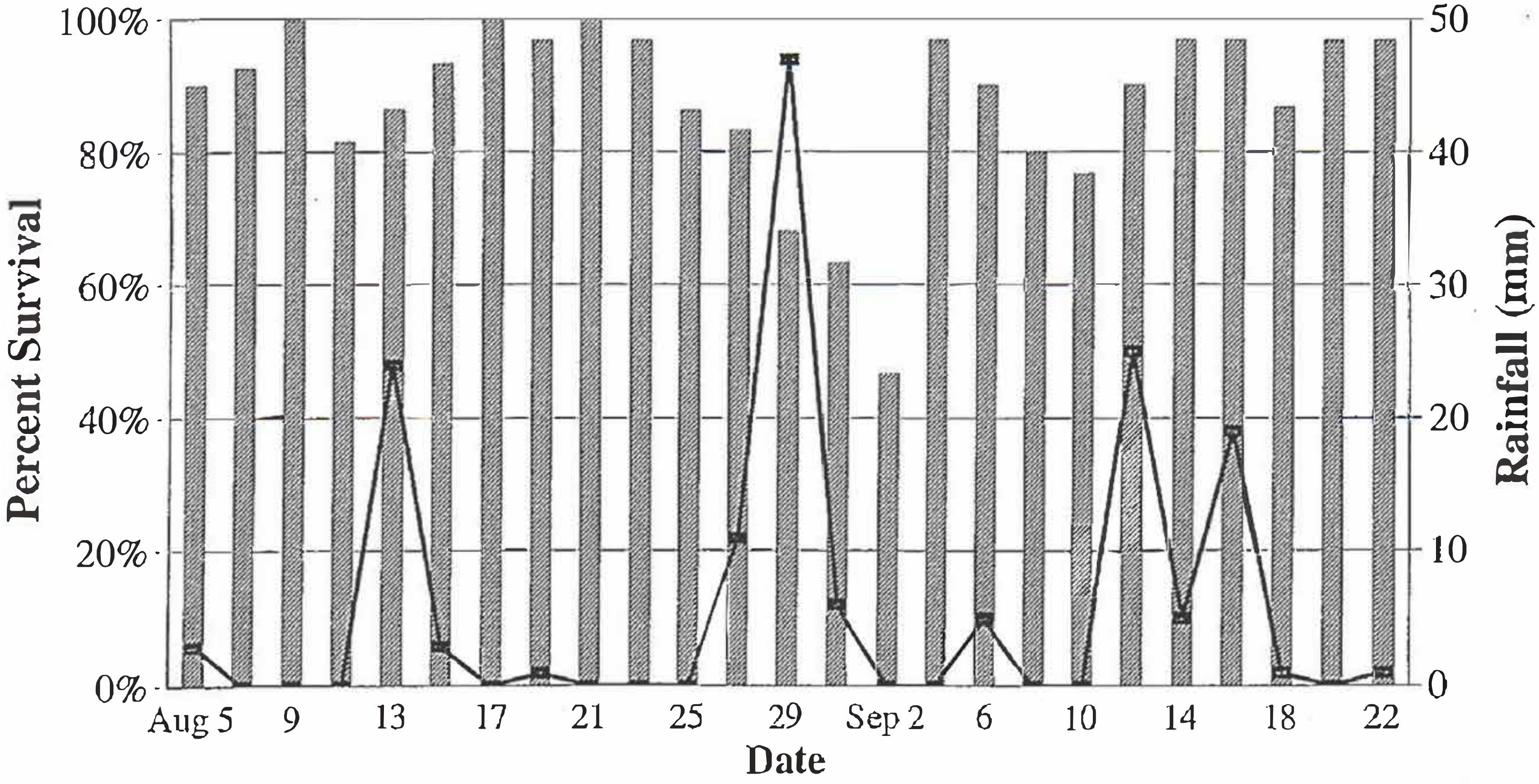


Figure 17. Survival of *P. pugio* in 96 hr bioassays at the Finney Creek site (F) and rainfall for the approximately 48 hr period preceding the sampling date.

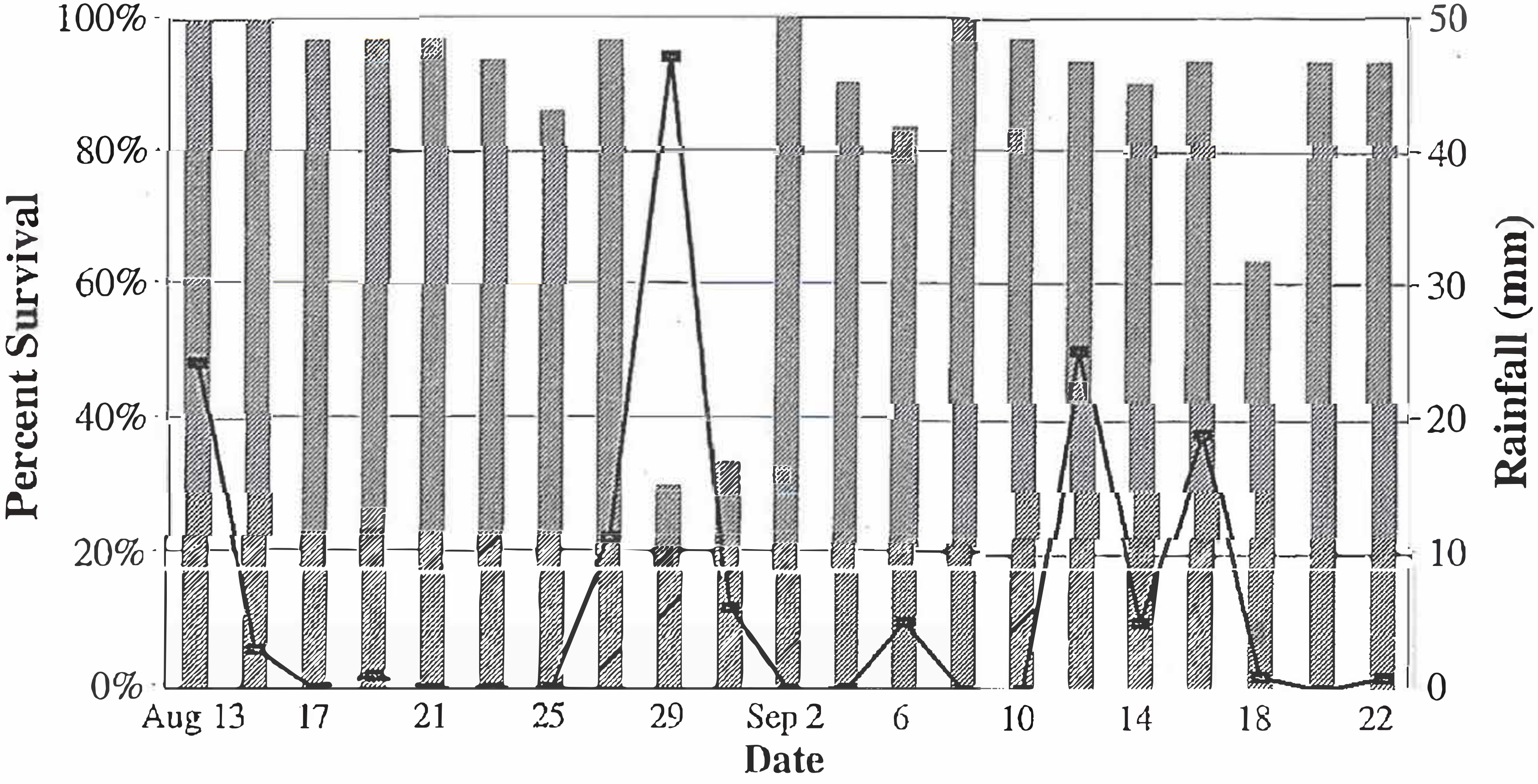


Figure 18. Survival of *P. pugio* in 96 hr bioassays at the Wachapreague Channel 1 site (W1) and rainfall for the approximately 48 hr period preceding the sampling date.

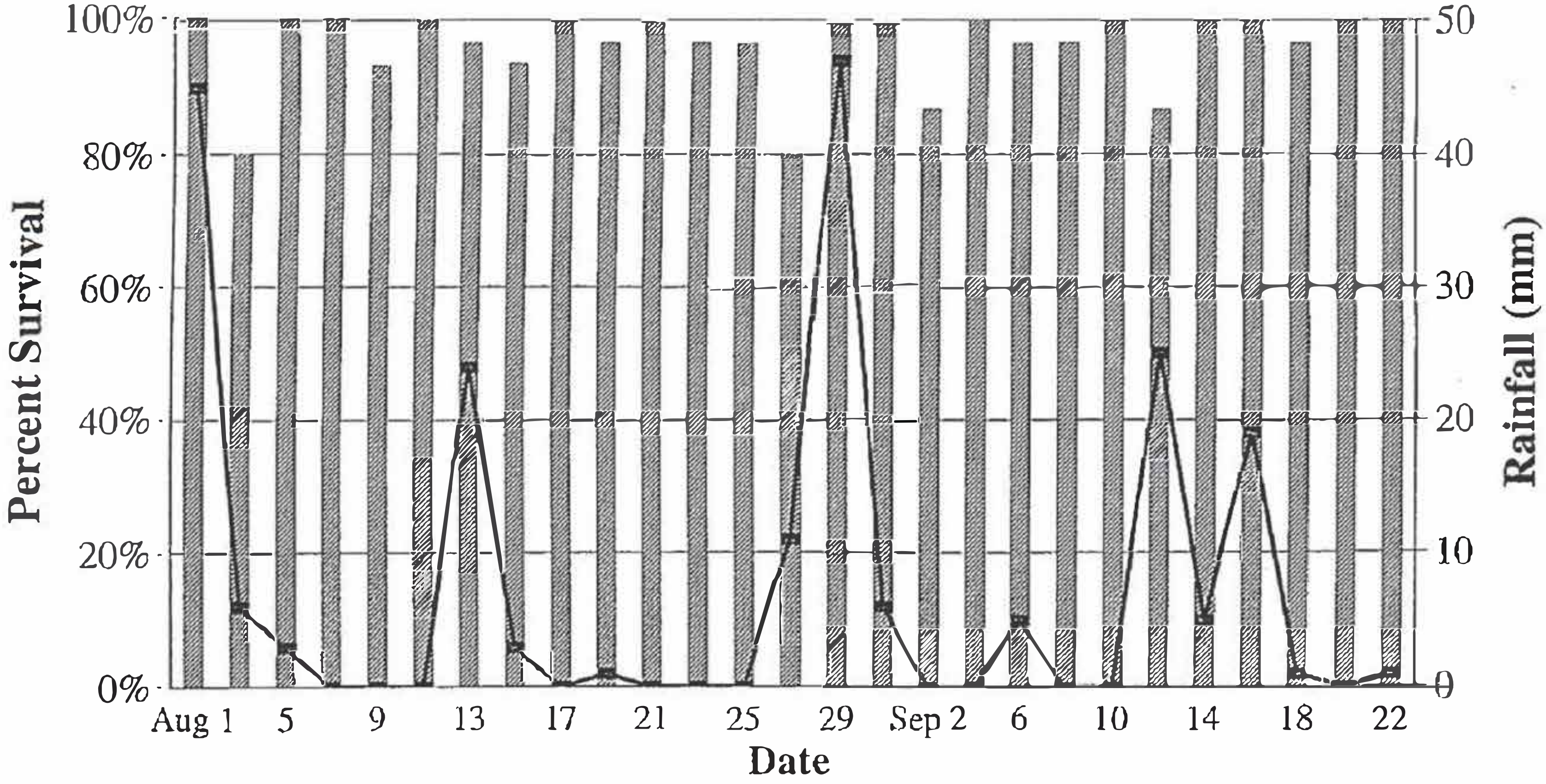


Figure 19. *Survival of P. pugio in 96 hr bioassays at the Gargathy Creek upstream site (G1) and rainfall for the approximately 48 hr period preceding the sampling date.*

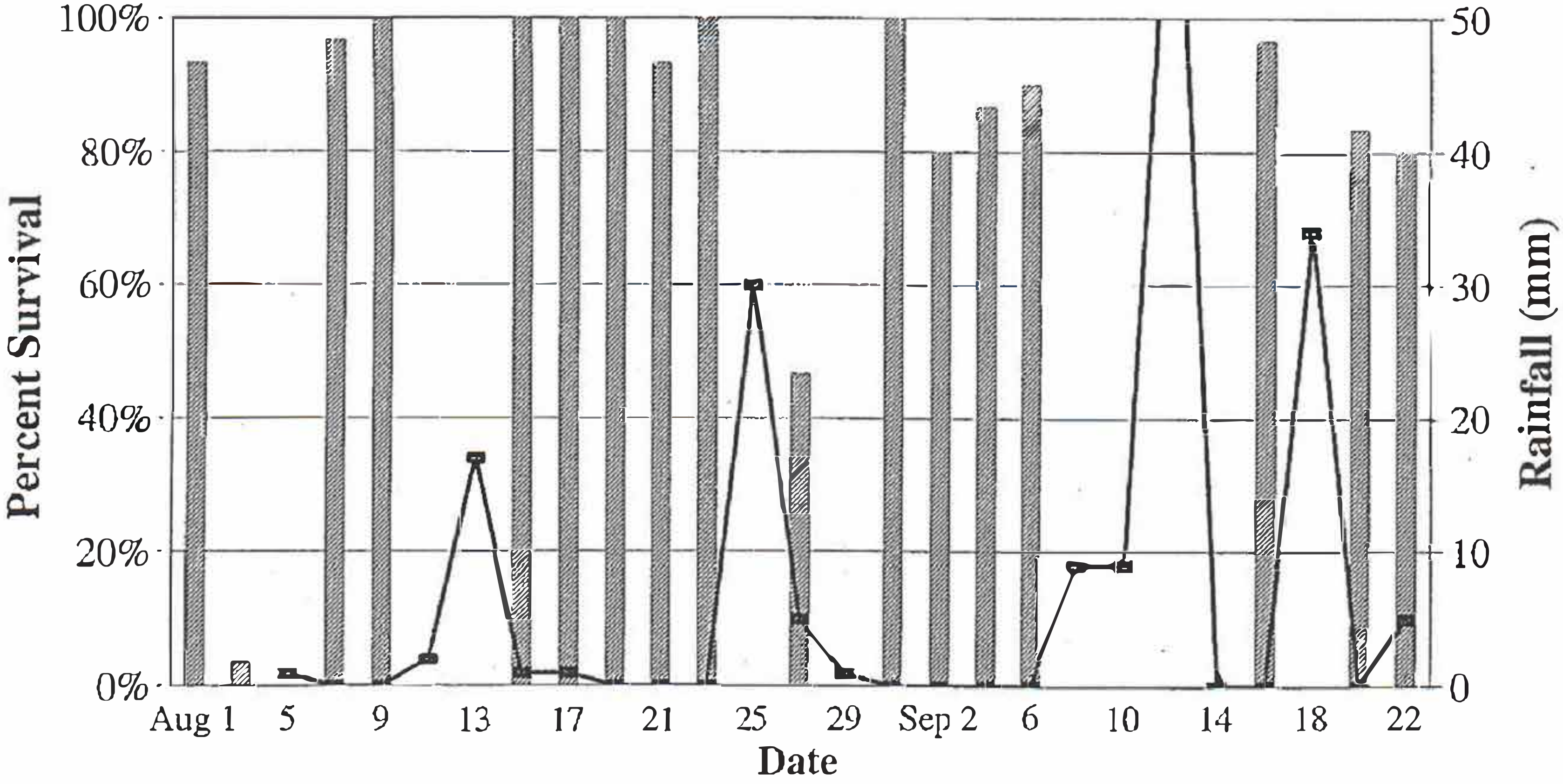


Figure 20. Survival of *P. pugio* in 96 hr bioassays at the Gargathy Creek downstream site (G2) and rainfall for the approximately 48 hr period preceding the sampling date.

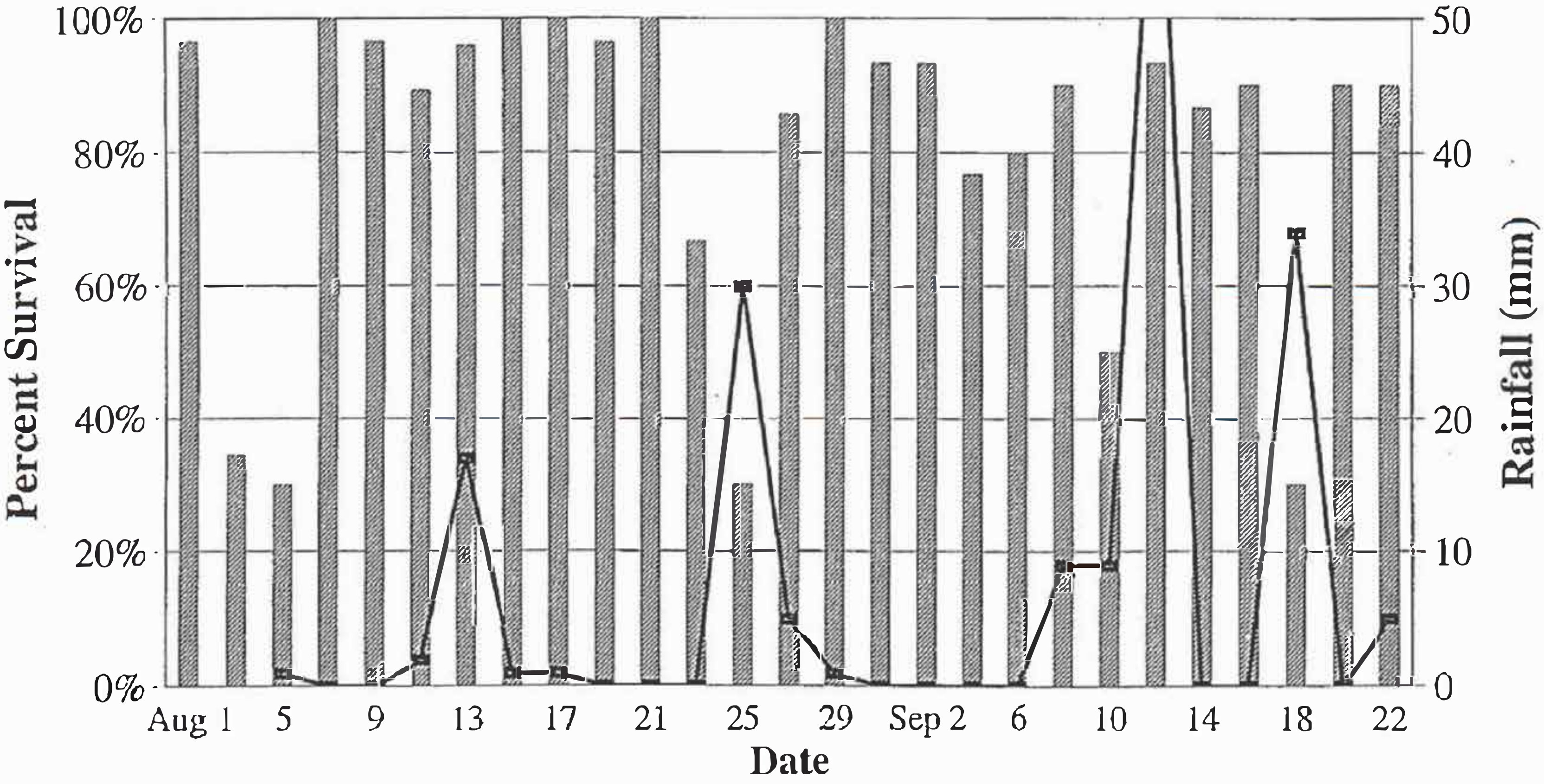


Figure 21. Survival of *P. pugio* in 96 hr bioassays at the Folly Creek upstream site (FO1) and rainfall for the approximately 48 hr period preceding the sampling date.

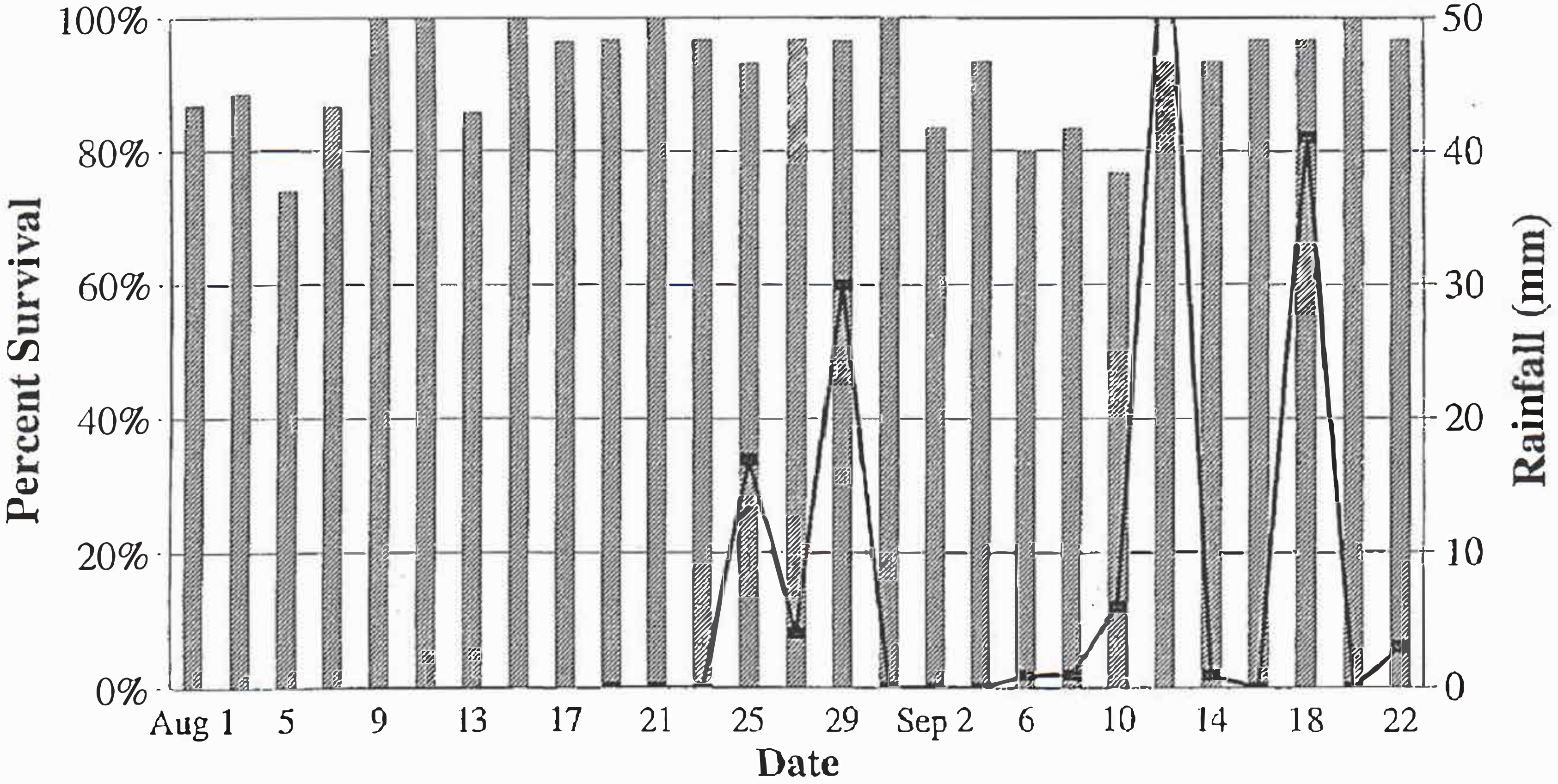


Figure 22. Survival of *P. pugio* in 96 hr bioassays at the Folly Creek midstream site (FO2) and rainfall for the approximately 48 hr period preceding the sampling date.

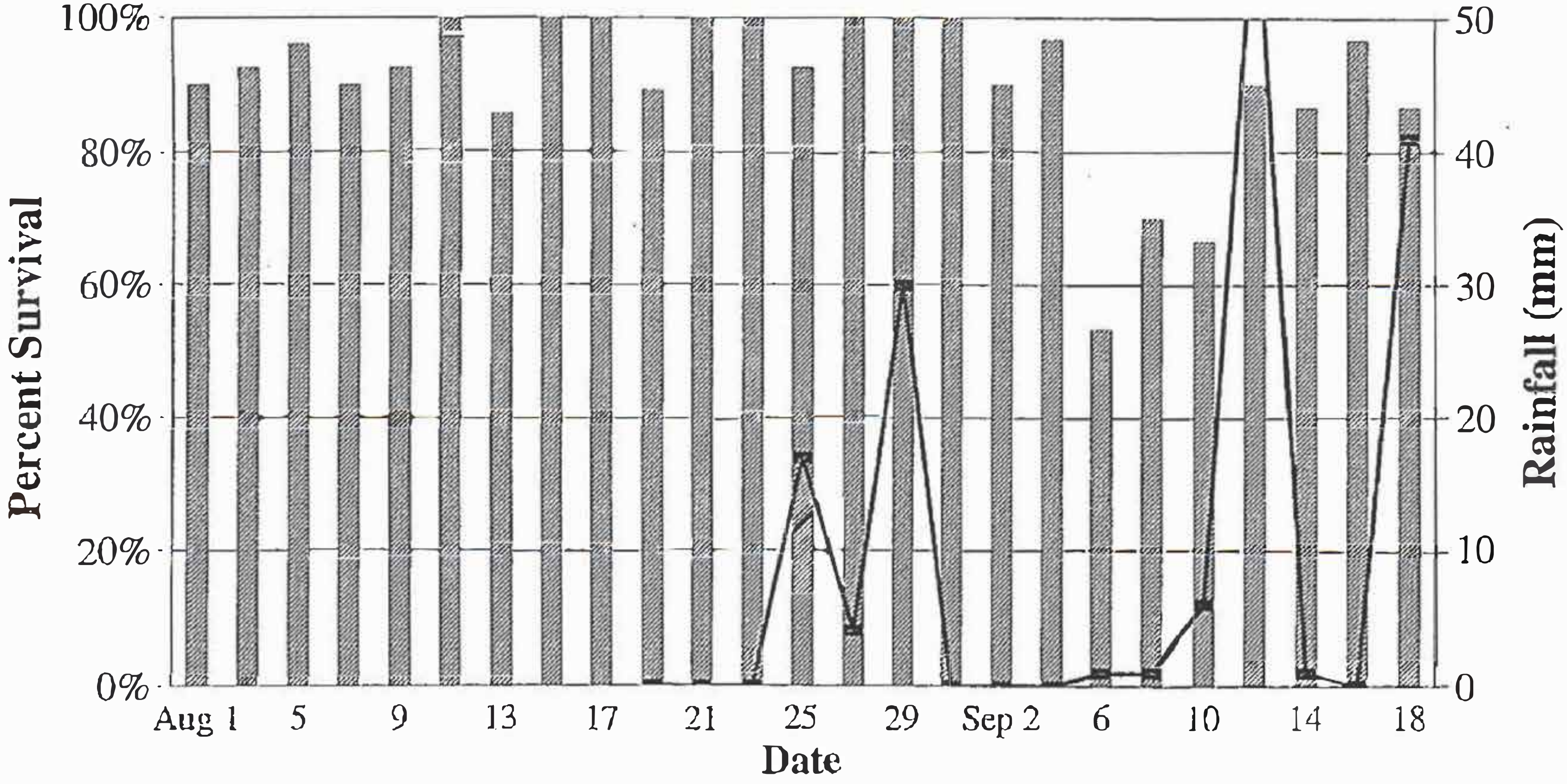
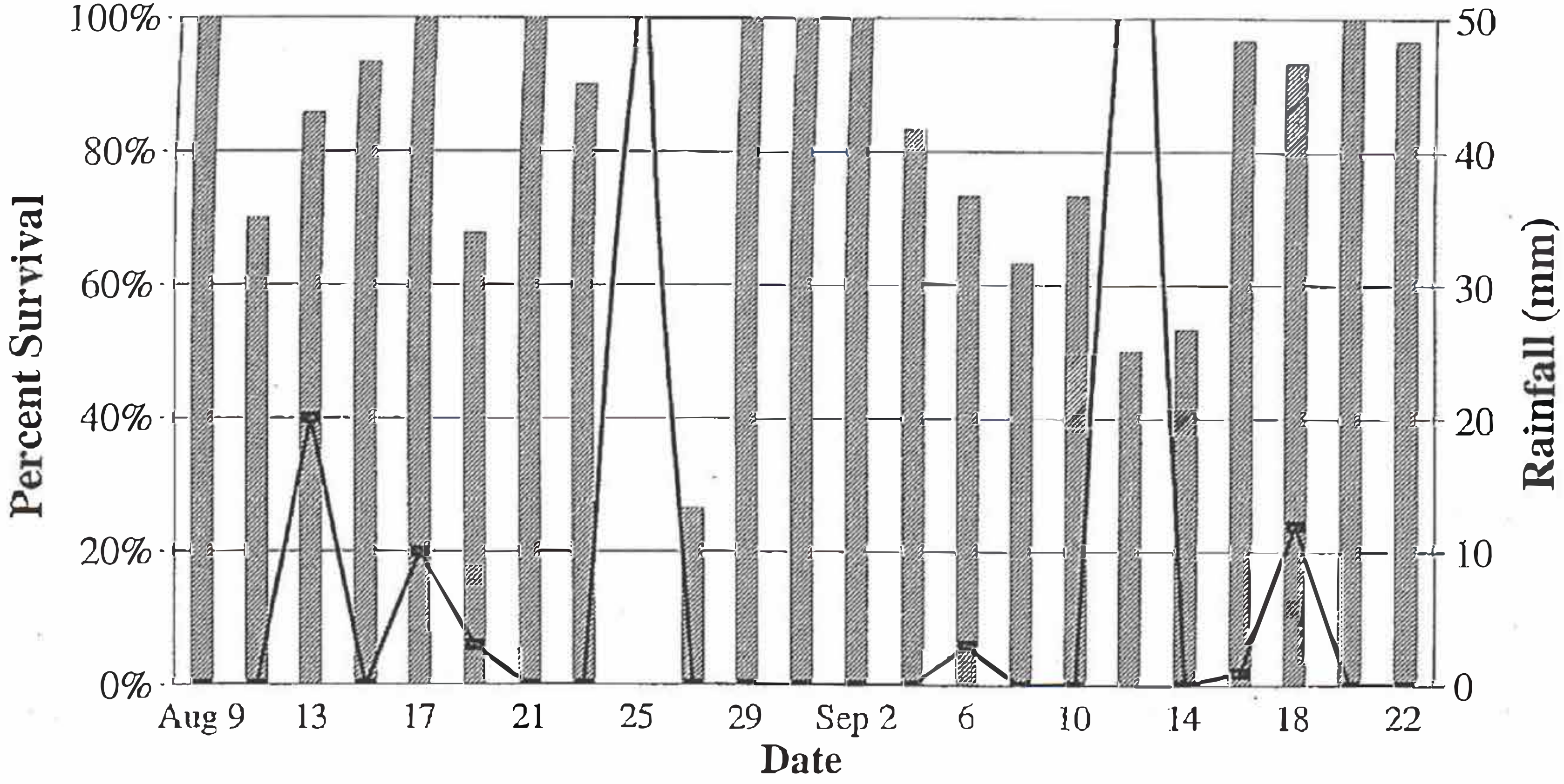


Figure 23. Survival of *P. pugio* in 96 hr bioassays at the Indiantown Creek site (I) and rainfall for the approximately 48 hr period preceding the sampling date.



Discussion

This study was not designed to identify particular fields or toxicant substances responsible for problems reported in shellfish hatcheries. Rather, it sought to determine whether there were widespread water quality problems on the seaside Eastern Shore which might be associated with vegetable cultivation. Our approach was rigorous in the selection of multiple sites and the use of sequential 96 hr bioassays, but it was general its evaluation of metal and pesticide toxicities. Our primary objective in this study was to lay the groundwork for future water quality studies by evaluating the spatial extent and timing of toxic events.

There are several important limitations to this study. First, the selected watersheds contain only a limited range of coverage by plastic vegetable cultivation (0-13%). We do not know how this compares to the full range of values for all creeks across the entire Eastern Shore. Sites which we found to contain a greater coverage by plastic culture failed to meet other selection criteria (e.g., water depth or salinity). Further, the timing of this study, late summer, corresponds to only a portion of the agricultural season and does not correspond to the time during which most bivalve embryo/larva culture is concentrated. Differing acreage, pesticide applications and rainfall patterns earlier in the season may have yielded different results.

Our choice of assay techniques, MetPAD™ for metals and *P. pugio* survival for organic pesticides and other materials, necessarily influence our findings. The enzymatic bioassay for metals lacked the sensitivity to detect concentrations of copper which might cause lethal effects in molluscan larvae, but, with its sensitivity to numerous metals, provided a means of determining broadly whether acute metal toxicity was a problem at any of the study sites. Further, the rapid test completion meant that a single technician could process a larger number of samples that could have been processed using a less specific test involving bivalve embryos as the test species. The *in situ* bioassay approach using *P. pugio* provided good sensitivity to certain organic pesticides commonly used on tomatoes (Table 2), but its sensitivity to all pesticides is not known. Moreover, we were only able to evaluate acutely lethal, not chronic or sublethal, effects with this study. As noted above, this study did not identify specific toxins responsible for the observed mortalities. Finally, though our results suggest water quality impacts associated with land use, we have only characterized land use patterns within the watersheds in a cursory fashion. More detailed information on land use, pesticide applications and management of individual fields would be desirable.

The major finding of this study was the relationship between toxic events and rainfall within several watersheds. At the upstream location in Gargathy Creek complete mortality of shrimp was observed after virtually every rainfall event and the available data indicate that this mortality was not associated with low salinity or dissolved oxygen levels. Coupled with observations of direct run-off from an adjacent tomato field, the implication of this finding is that agricultural practices in the immediate watershed are impacting living aquatic resources at this site. A similar, but less severe, pattern of mortality in relation to rainfall was observed at the downstream site in Gargathy Creek. Mortality of shrimp was also observed at Indiantown Creek, but generally only after rainfall events in excess of 50 mm/48 hr. The Finney Creek site experienced approximately 70% mortality after a large rainfall, but over 30% mortality was observed in Nickawampus Creek (which essentially lacks vegetable cultivation) following the same rainfall. Upstream stations at Folly Creek and Phillips Creek, along with downstream stations at Folly Creek and Wachapreague, experienced only minimal mortalities.

The MetPAD data provide limited evidence that metals may be contributing to observed toxicity at selected creeks. The unavoidable nonavailability of data for many samples and the limited sampling scheme limited in frequency precludes an unequivocal evaluation of a relationship to runoff for metals. Nevertheless, whenever metals toxicity was implicated there has a rainfall event of some magnitude. The converse is not true; to wit that there was observed metals toxicity whenever there was a rainfall event.

Taken in total our findings suggest that toxic events from organic pesticides and metals in the headwaters of selected creeks on the Eastern Shore are related to inputs from the immediate watershed. Correlative evidence suggests that vegetable farming (particularly tomato cultivation) may be a significant contributor to these toxic inputs. However, our findings also suggest that management practices on the tomato fields may dramatically affect the likelihood of impacts on living resources resulting from runoff into adjacent tidal creeks. The tomato field adjacent to the upstream site on Gargathy Creek drains directly into the tidal creek via a series of dug ditches and erosional channels. In contrast, the tomato field adjacent to the Indiantown Creek site has wide vegetated buffer strips on all of its boundaries and much of the run-off feeds into a retention pond. It is important to note, however, that this retention pond is the result of an impoundment created over 30 years ago and may not represent a viable management option for many farms today.

It is significant to note that some mortality of grass shrimp was observed in most creeks following storm water run-off events. For instance, Nickawampus Creek, which has only a very minimal amount of vegetable cultivation within its watershed, experienced over 50% mortality in the test organisms after a rain event approaching 40 mm within a 48 hr period. Only Phillips Creek, which has extensive tidal marshes through which all terrigenous inputs pass, did not exhibit toxic impacts during our study.

In a very extensive study in South Carolina, Scott et al. (1990) investigated very similar issues to those which have been raised here. They found that the use of plastic ground covers on tomato fields increased productivity by 25% over test fields grown without ground covers. These ground covers were observed to increase total run-off from the field, but lower concentrations of pesticides were found in streams which drained these fields compared to ones draining fields which did not use plastic covers. Scott and co-workers noted, however, that toxic events were more frequent and followed smaller rain events in watersheds with plastic cultivation. After experimentation with different field management options, they concluded that (1) effective use of integrated pest management to determine pesticide application, (2) the use of less toxic pesticides and (3) the use of retention ponds to reduce run-off served to greatly reduce the impacts of this agricultural practice on water quality and living resources in tidal marsh creeks.

Determining the effects of specific land management practices on aquatic ecosystem health is seldom a trivial matter. Because numerous land uses and management approaches are typically found within a watershed, each with a unique potential for having an impact on the receiving water, care must be taken in extrapolating from correlative studies. To fully evaluate the impacts of vegetable cultivation techniques on water quality, we will require further study. Future work addressing this issue on Virginia's Eastern Shore should expand the watersheds under consideration, especially to include bayside creeks. Further studies need to be conducted during the spring and early summer when agricultural practices may vary and many aquatic organisms are in reproductive phases. The inclusion of chronic, as well as acute, indicators of ecosystem health are needed, and future studies should evaluate impacts on more species, including natural assemblages. When toxic events are observed in particular water bodies, the specific pesticides and metals present need to be quantitatively identified, the chemical form of each specified, and the terrestrial source investigated. Finally, collaborative efforts, involving research scientists and farmers are needed to evaluate management practices and their potential to improve water quality and thereby the living resources in coastal waters.

Despite the above caveats, our findings, as well as those from previous studies, make it clear that implementing best management land use practices which minimize direct run-off from vegetable fields into tidal creeks provide a straightforward, practical approach towards reducing environmental impacts. The apparent relationship between toxic impacts and run-off from some vegetable fields observed during this study are, we believe, compelling justification for judicious management of run-off and we urge that careful attention be paid to this matter by vegetable farmers.

Conclusions and Recommendations

This study does not indicate that vegetable farming with the use of plastic ground covers *per se* results in damage to aquatic ecosystems. Over the range of sites which we studied, the amount of plastic vegetable cultivation varied from 0 - 13% of the watershed and management techniques for individual fields also differed. Following heavy rainfall some degree of toxic impacts was observed at most upstream sites in our study, regardless of the amount of vegetable cultivation in the watershed.

We did not systematically evaluate management practices on individual fields and we have no data on pesticide applications to any of the fields within the study area. Nevertheless, our study clearly points to differing toxic impacts across watersheds and is strongly suggestive of the importance of run-off management. Contrasting the results from Indiantown Creek and Gargathy Creek makes it clear that efforts to contain run-off may have a dramatic effect on downstream water quality. Though we cannot entirely discount the possible roles of differing pesticide applications and other land uses within these watersheds, it is quite clear that containment of run-off from vegetable fields (and other areas which receive biocides) is a requisite component of wise resource management.

Clearly, further monitoring of water quality and studies directed towards evaluating land management approaches are needed. Improving our understanding of how the broad spectrum of land uses and management practices on the Eastern Shore affect water quality would seem to be vital improving the health of aquatic resources in tidal waters. This will be an ongoing research need as the patterns of land use continue to change in coming years. There is an obvious trend toward increased use of land for residential/commercial purposes which could accelerate.

Even in advance of further study, there are sound management practices which can be put into practice immediately. We urge the implementation of good run-off management through (1) the elimination of direct ditching into tidal creeks, (2) the use of vegetated buffer strips, and (3) where practical, the use of retention ponds for containing storm water run-off.

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