

INDICATORS OF STREAM HEALTH: THE USE OF BENTHIC MACROINVERTEBRATES
AND AMPHIBIANS IN AN AGRICULTURALLY IMPACTED AREA, SOUTHWEST
GEORGIA

by

TARA KRISTIN MUENZ

(Under the direction of Stephen W. Golladay and George Vellidis)

ABSTRACT

Benthic macroinvertebrates and amphibians (frogs and salamanders) were used to assess differences amongst buffered (fenced from cattle over 20 years ago) and unbuffered streams within an agricultural landscape in southwest Georgia from 2002-2003. Water quality, physical and vegetative parameters as well as macroinvertebrate metrics (% Crustacea, % EPT, % Elmidae, and % Diptera) showed differences between treatments, suggesting that fenced sites are recovering from any cattle activity incurred and that conservation buffers are effective at mitigating these effects. However, only one amphibian survey method captured these differences, *Eurycea cirrigera* larvae captured within macroinvertebrate collections, of which highest captures were at the fenced sites. Feeding preference of *E.cirrigera* was also examined, and electivity indices suggest slight positive selection for a subfamily of the Chironomidae, the Tanypodinae. Certain amphibians are good candidates as ecological indicators, however more information is needed on responses and tolerances to disturbance from the microhabitat to landscape levels.

INDEX WORDS: Agriculture, Stream health, Bioassessment, Biomonitoring, Indicators, Macroinvertebrates, Amphibians, Salamanders, Treefrogs, Fencing, Cattle grazing, Riparian buffers, *Eurycea cirrigera*, Electivity, Prey preference, Coastal Plain

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B.A., Miami University, 1998

A Thesis Submitted to the Graduate Faculty of the University of Georgia in Partial
Fulfillment of the Requirements for the Degree

MASTER OF SCIENCE

ATHENS, GEORGIA

2004

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DEDICATION

To the water and the land....

Conservation is a state of harmony between people and the land. By land is meant all of the things on, over, or in the earth. Harmony with land is like harmony with a friend; you cannot cherish his right hand and chop off his left. This is to say, you cannot love game and hate predators; you cannot conserve the waters and waste the ranges; you cannot build the forest and mine the farm. The land is one organism.

-Aldo Leopold 1966

ACKNOWLEDGEMENTS

A great many people and experiences have contributed to my thesis, and to my growth as a person. I would not be who I am and where I am today if not for them...

Mille grazie to my parents, Kathy and Don, for all their love and support, and openness to ‘Tara’s latest revelations.’ I couldn’t have arrived this far without you and your kindness, you gave and still give, beyond yourselves; thanks for believing in me, vi amo molto!

Thanks to all my family and friends, for their support and interest in this project and all the stories that went along with it. The Athens crew up in Ecology, I will always remember our dinners together conversing topics in conservation biology... and our laughter. BDY, thank you for listening to my ideas and frustrations, and thank you for your advice and ideas for this project, you’re a gem ☺

I am grateful to the wonderful folk at Ichauway, who provided so much physical, emotional, and technical support. It was an incredible opportunity to be around you and this ‘place.’ I could always count on relief from a good game of ultimate, and the unending sandy roads of Ichauway to run like the wind. Thanks to the aquatics and herpetology labs, and plant geeksters (Lee Anne Bledsoe, Frankie Hudson, Raynie Bambarger, Jan Battle, Anna Liner, Slaton Varner Wheeler, Shannon Hoss, Eamonn Leonard, and Brian Yahn) who toughed it out in the rugged terrain of southwest Georgia to help me with my field work and picked a bug or thousand; I learned so much from each of you ☺ Thanks to Liz Cox, who upon receipt of my hundreds of journal requests, always had a smile on her face (and she was quick too!). Thanks also to Jean Brock for her unending patience with my GIS questions. Statistical help was

provided by many, but extra thank-you's go to Mike Connor, Scott Chapal, and Micah Perkins. Thanks also to Kay Kirkman, for her advice and help with the vegetation aspect of this study. And I know this may sound strange, but I have to give props to good 'ol truck # 36, which numerous times got me safely to my study sites and through the cow herds, over the bumpy land, and most often, out of the mud.

If it were not for the Brownlee family and their kindness and openness to this project, I would not have had such a fantastic adventure, nor would this project have ever taken off. I also would probably still be sitting stuck in a ditch or in the mud in one of their peanut fields today if they hadn't rescued me. I would also like to thank the family for sharing their time, and thoughts and knowledge of the land. Some of the most beautiful sunrises I've ever seen have come over those hills, and that's where I learned "there's a difference in livin' and livin' well."

Thanks also to the many kind folks in Blakey, Georgia, especially the district conservationists with the NRCS, namely Joe Wilson. And also to Fred and Sheila, who had awaiting, the best food after a long day out in the field, at Smitty's Deli. My belly was always, always, happy.

The folks at NESPAL in Tifton played a major role in the water quality aspect of this study, from Rodney meeting me every two weeks at the Newton Petro for my grab samples, to the analyses of water samples. Thanks so much for all your time and hard work.

I won't get into the whole story of the long road to finding, obtaining, and creating the coverboards, but it involves the kindness of many folks. First to Steve Cross for finding suitable wood, and to Sean Kelly and Grant Crumbaugh who kindly took the lead on tearing apart the final source: an old barn. And thanks go to Peter Jones for putting those old barn scraps together to make a coverboard even a salamander could love.

Thanks also to my wonderful committee, Steve Golladay, George Vellidis, Mary Freeman, and Lora Smith, for their patience, willingness to teach, and time and thought that went into the development and final culmination of this study. Each of you offered a unique piece to my development and understanding of the many ‘ologies,’ and the science of being both a scientist and educator.

I am also grateful to the following foundations and organizations that funded this project: the Robert W. Woodruff Foundation, the Joseph W. Jones Ecological Research Center, the University of Georgia, and funds from an Environmental Protection Agency 319(h) grant.

Thank you God for teaching me how to accept the things I cannot change, the courage to change the things that I can, and the wisdom to know the difference...

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CHAPTER 1

INTRODUCTION, LITERATURE REVIEW, AND THESIS FORMAT

A healthy stream ecosystem, as defined by Meyer (1997), involves key concepts such as ‘sustainability’ and ‘resilience,’ incorporating both ecological integrity (maintaining structure and function) and societal values. Freshwater ecosystems worldwide have been subject to a variety of anthropogenic disturbances, severely altering the health of these systems. In fact, over 60% of reported waterway problems within the U.S. have been associated with land altering practices such as agriculture, which continue to be a major contributor of non-point source (NPS) pollution to Georgia’s streams and rivers (EPA 1994). Hydraulic alterations of waterways, alteration of flow rates, and the disruption of wildlife habitats through changes in chemical concentrations and increases in sedimentation, are additional consequences of intensive, high production agriculture (Schultz et al., 1995). Degradation of waterways will continue unless appropriate management techniques are employed. Conservation buffers are one management strategy that has become widely accepted to help reduce agricultural impacts on surface and ground water systems.

Conservation Buffers

Conservation buffers are small areas or strips of land permanently maintained in vegetation designed to intercept and effectively mitigate nonpoint source (NPS) pollutants such as sediment, nutrients, and pesticides, within and from farm fields. Buffers also provide critically important habitat, supporting a diversity of aquatic and terrestrial wildlife, and in some cases they may also function as habitat corridors in a highly fragmented agricultural landscape (Noss, 1983; Lowrance et al., 1985). Vegetative filter strips, contour buffers, cattle exclusion fencing, and maintenance of riparian forests are buffer types that are suggested as Best Management Practices (BMPs), designed for specific problems associated with different agricultural production systems. In many cases, such conservation practices have demonstrated a

reduction of agricultural impacts on adjacent streams (e.g. Osborne et al., 1993; Lowrance et al., 1995; Edwards et al., 1997; Raffaele et al., 1997; Vellidis et al., 2003).

Efforts to improve the health of riparian areas typically focus on establishing vegetative or forested buffers along streams. Several studies have shown the effectiveness of buffers at removing sediment and nutrients such as $\text{NO}_3\text{-N}$ (Lowrance, 1992; Verchot et al., 1997), as well as controlling pesticide transport from surface and subsurface flow (Lowrance et al., 1997). Vellidis et al. (2002) found that herbicide (atrazine and alachlor) concentrations and loads in surface runoff and shallow groundwater were reduced significantly during transit through a restored riparian buffer system. Furthermore, at least three separate studies specific to the Gulf Coastal Plain region have shown that concentrations and loads of nitrogen and phosphorus in subsurface flow and surface runoff are significantly reduced after passage through a riparian forest (Lowrance et al., 1983 and 1984; Peterjohn and Correll, 1984; Jacobs and Gilliam, 1985). Vellidis et al. (2003) reported that a reestablished riparian forest downslope from agricultural production sites retained 65 % of N and 68 % of P entering the buffer. Finally, past research has shown that grass filter strips can effectively reduce pollutants from animal wastes (Bingham et al., 1980). The exclusion of cattle from streams has shown to produce the same effect as restoring a vegetated buffer, with substantial reductions in fecal coliform, suspended solids and nutrient loads, and increases in aquatic insect diversity (Owens, 1996; Line et al., 2000; Thomas, 2002).

Although an extensive amount of data is available, essential questions concerning buffer effectiveness still remain. Limited data exist in the Southeastern U.S. and even fewer studies are available with regard to buffer effectiveness in the Coastal Plain of Georgia. Data available from these studies predominately consist of plot-size treatments, not larger whole farm studies.

Additionally, only certain kinds of buffer practices in certain types of agricultural settings have been examined. Finally, conservation has been centered on the protection and restoration of public lands even though more than 70% of the entire landmass of the conterminous U.S. and Hawaii, and over 90% of Georgia resides in private lands (NRCS 1996). Some of the most modified ecosystems reside on private lands, and if we are to successfully conserve and understand these systems, we must consider private as well as public land. To encourage conservation, private landowners must be partners in sustaining and restoring natural communities (Knight, 1999; Norton, 2000).

Biological Indicators and Water Quality Monitoring

Water quality monitoring is one tool used to assess the health of aquatic systems, and in turn, the effectiveness of conservation buffers. Water quality monitoring is a general term that encompasses a multitude of indicators (physical, chemical, and biological). It is used to evaluate changes in the environment, most often due to anthropogenic disturbances, with the intention of using the information for restoration and management programs (Rosenberg and Resh, 1993). Biological monitoring in particular, has been recognized as one of the most useful tools in assessing water quality because biota are sensitive to environmental disturbances, show a wide range of responses, and are continuous monitors of their 'living' environment (Chandler, 1970; Karr and Chu, 1999; Linke et al., 1999). Ideally a useful or 'good biological indicator' would have the combined attributes of being holistic, early warning, diagnostic, abundant and tractable, readily sampled, occurring in high enough numbers for comparison (Rapport, 1992). More simply, the biota's condition would reveal and communicate the consequences of human activities for an aquatic system, and as Karr and Chu (1999) state, "a biota's condition offers the most comprehensive indication of ecological risks in a particular place."

Benthic macroinvertebrate assemblages have been often used in biological monitoring, especially to assess stream disturbances (Rosenberg and Resh, 1993). Considered excellent indicators of localized conditions, macroinvertebrates offer a spectrum of responses, inhabiting a wide range of environments while integrating the effects of short-term environmental variation (e.g. Lenat et al., 1980; Abel, 1989; Barbour et al., 1999). In fact, invertebrates have been shown to be sensitive indicators of changes in stream quality due to agricultural impacts (Lenat, 1984; Berkman et al., 1986; Gregory, 1996; Davis et al., 2003). More specifically, studies by Gregory (1996) and Davis et al. (2003) have also shown macroinvertebrates communities as valuable tools to evaluate the effectiveness of BMPs on streams affected by land-use disturbances in the Gulf Coastal Plain.

The monitoring of fish assemblages is also an integral part of monitoring programs, with extensive life history information available for many species. Although both groups of organisms fit many of the criteria for 'good indicators,' there are several disadvantages for each; quantitative samples are difficult to obtain, species diversity may vary due to factors other than water quality, and in the case of invertebrates, the effort required to sort and identify specimens can be extensive. Yet studies have found macroinvertebrate and fish assemblages extremely useful in monitoring programs, with the advantages far outweighing the disadvantages (Berkman et al., 1986; Nerbonne and Vondracek, 2001).

Amphibians as Bioindicators

Amphibians are thought to be highly sensitive to disturbances due to their complex life histories, which use both terrestrial and aquatic environments, require specific microhabitats, and specialized physiological adaptations (Wake, 1990; Blaustein et al., 1994; Stebbins and Cohen, 1995). These adaptations can cause them to be susceptible to minor environmental perturbations

which alter their ability to seek cover from predators or forage on primary prey such as zooplankton, insects, and other invertebrates (Welsh and Ollivier, 1998). Amphibians, especially salamanders, are excellent monitors of local conditions because they remain in fairly confined home ranges for their entire lives (Blaustein and Wake, 1995), their life stages involve many parts of the environment, and they are relatively long-lived species, having extended developmental times (Petranka, 1998). Because of their semi-aquatic lifestyles, amphibians are also likely to become exposed to chemical contaminants that are applied to agricultural fields, acting as a barometer of environmental conditions (Semlitsch, 2003). According to studies by Storm et al. (1994), Ash (1997), and Hunter (1995), salamanders were found to be sensitive to environmental factors such as habitat destruction or alteration, contaminants in soil and water, drought, and acidification. The state of Georgia supports an extremely diverse assemblage of stream amphibians, with over 80 species occupying a variety of freshwater niches from seeps and headwaters to higher-order streams (Corn et al, 2003). Furthermore, many species have lotic-dwelling larval stages which are highly specialized in using stream microhabitats for both cover and foraging (Welsch and Ollivier, 1998). In small headwater streams, salamanders can occur in high numbers and provide another tool to assess stream habitat quality where other species assemblages, such as fish, may be less diverse or absent (Rocco, 1999; Corn et al., 2003).

Numerous studies within the western U.S. have incorporated amphibians into assessments of impacts of forest management practices within riparian zones (e.g. Gomez and Anthony, 1996; Wilkins and Peterson, 2000; Welsh and Lind, 2002), yet few have examined disturbance resulting from agricultural practices such as grazing. The state of Ohio Environmental Protection Agency (EPA) has even established an amphibian IBI (index of biotic integrity) for isolated wetlands and is developing similar indices for headwater streams

(Micacchion, 2002). Although a multitude of reasons favor the use of amphibians as ecological indicators, few studies in the eastern U.S. have been specifically designed to examine amphibian responses to disturbances within aquatic ecosystems, or have used them within a water quality monitoring program (but see Chazal and Niewiarowski, 1998; Bowers et al., 2000; Homyack and Giuliano, 2002; Ryan et al., 2002; Willson and Dorcas, 2002).

Pervasive amphibian population declines in North America are attributed to environmental alterations from wetland drainage, timber harvesting, agriculture, urbanization, stream pollution and siltation, and the introduction of exotic species (e.g., Blaustein and Wake, 1995; Petranka, 1998; Welsh and Lind, 2002). These disturbances alter the hydrodynamics of stream and river ecosystems that support amphibians, fill and drain wetland breeding sites, and remove natural vegetation in upland areas that are critical for feeding and refugia (Semlitsch, 2003). Within the state of Georgia, several amphibian species abundances and ranges have been reduced primarily due to habitat loss (GADNR 1999), but as yet, an effective monitoring and conservation program has not been established. Amphibian conservation efforts are worthy of establishment, as these taxa play a vital role in aquatic ecosystem dynamics, and their sensitivity to land disturbance and pollution may serve as an indication of ecosystem health (Vitt et al., 1990; Semlitsch, 2003). One possible tool for amphibian conservation is their inclusion in biological monitoring programs. This would offer crucial information to better understand the ecology of amphibians, such as environmental limitations, and perhaps also serve to educate and include humans in conservation solutions. Past, present, and future threats also need to be identified, and incorporated into programs to promote riparian protection, including perennial headwater streams that are critical habitat to significant amphibian populations (Corn et al., 2003).

Objectives

The objectives of this study were to evaluate differences in the macroinvertebrate and amphibian community composition and abundance, riparian composition, physical habitat composition, and overall water quality between buffered (fenced from cattle access) and unbuffered (unfenced; cattle have access to streams) sites in southwest Georgia. It was important to also determine potentially useful metrics for stream health in this region, and to address the role amphibian species may play in bioassessment methods, with the hope of extending conservation efforts for this faunal group. Finally, I wanted to examine feeding preferences of salamander larvae from streams with a history of disturbance due to agricultural practices, and understand if their abundance levels are associated with prey availability.

Thesis format

This thesis is written in manuscript format with chapters 2 and 3 as separate manuscripts. Chapter 1 is a general introduction to the thesis, providing background information on agricultural influences on stream health and the management tools that have been used to restore and monitor impacted streams, such as conservation buffers and biological monitoring. Chapter 2 describes the effect of stream bank fencing as a form of conservation buffer in southwest Georgia, and also discusses the potential for using amphibians in biological monitoring programs. Chapter 3 presents information on the diet and feeding behavior of the southern two-lined salamander, *Eurycea cirrigera* in streams with adjacent agricultural land-use. Chapter 4 offers conclusions and recommendations from this study.

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CHAPTER 2

CONSERVATION BUFFER EFFECTIVENESS: BENTHIC MACROINVERTEBRATES AND AMPHIBIANS AS INDICATORS OF STREAM HEALTH IN AN AGRICULTURALLY IMPACTED AREA, SOUTHWEST GEORGIA.

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ABSTRACT

Agricultural land use continues to be a major contributor of non-point source (NPS) pollution to Georgia's streams and rivers, associated with over 60% of reported waterway problems. Conservation buffers such as streambank fencing are one strategy adopted by various federal and state agencies to aid in the reduction of impacts on surface and ground water systems. To evaluate the impact of grazing livestock, we studied three unfenced (UF-1, UF-2, and UF-3) and two fenced (~20 yrs; F-1 and F-2) stream reaches within a diversified row crop and beef cattle operation located on a tributary of the Lower Chattahoochee River in Early County, southwest Georgia. We measured a suite of indicators including amphibian diversity and abundance, aquatic macroinvertebrate populations, vegetative structure, and water quality and physical parameters. Variability among sites and treatments existed, with those sites in the same treatment as most similar (e.g. UF-2 and UF-3), and disturbances from a nearby eroding gully strongly effecting unfenced site UF-1. Higher percentages of more sensitive macroinvertebrate metrics such as Ephemeroptera-Plecoptera-Trichoptera (EPT), Elmidae (Coleoptera), Crustacea (Decapoda and Amphipoda), and clingers were found at the fenced sites, whereas percentages of Dipterans and dominant family were highest at the unfenced sites. Fenced sites also showed lower and more stable levels of nitrate-N, suspended solids, and fecal coliforms. Percent canopy cover in the riparian area and over the stream was similar amongst sites (except UF-1), however vertical vegetative coverage and % leaf litter were greatest at fenced sites ($p < 0.0001$). Results revealed no differences between sites in salamander abundance from surveys within the riparian area. However, larval salamanders (*Eurycea cirrigera*) captured within bimonthly invertebrate samples were significantly more abundant at fenced sites ($p < 0.0001$), most likely due to lower sedimentation levels and greater heterogeneity of the stream substratum. Treefrogs captured

within PVC pipe refugia were dominated by *Hyla squirella* (87% of captures) and displayed an interesting yet puzzling pattern as both the overall highest and lowest abundance levels were found at fenced sites ($p < 0.0001$). Amphibians demonstrated a strong potential for use in biomonitoring programs, however, further life history information is needed for each species and lifestage, from variables at the microhabitat to landscape levels. Overall, differences in measurements (chemical, physical, biological) suggested that sites fenced from cattle many years ago demonstrate better water quality and habitat structure and that conservation buffers are benefiting these streams.

Key Words: Agriculture, stream health, bioassessment, biomonitoring, indicators, macroinvertebrates, amphibians, salamanders, Plethodontidae, treefrogs, Hylidae, fencing, cattle grazing, riparian buffers, Coastal Plain

INTRODUCTION

Aquatic ecosystems worldwide have been subject to a wide variety of anthropogenic disturbances, severely altering the health of these systems. In fact, over 60% of reported waterway problems within the U.S. have been associated with land altering practices such as agriculture, which continue to be a major contributor of non-point source (NPS) pollution to Georgia's streams and rivers (EPA 1994). Hydraulic alterations of waterways, alteration of flow rates, and the disruption of wildlife habitats through changes in chemical concentrations and increases in sedimentation, are additional consequences of intensive, high production agriculture (Schultz et al., 1995). Degradation of waterways will continue unless appropriate management techniques are employed. Conservation buffers are one management strategy that has become widely accepted to help reduce agricultural impacts on surface and ground water systems.

Conservation buffers

Conservation buffers are small areas or strips of land permanently maintained in vegetation designed to intercept and effectively mitigate nonpoint source (NPS) pollutants such as sediment, nutrients, and pesticides, within and from farm fields. Buffers also provide critically important habitat, supporting a diversity of aquatic and terrestrial wildlife, and in some cases they may also function as habitat corridors in a highly fragmented agricultural landscape (Noss, 1983; Lowrance et al., 1985). Vegetative filter strips, contour buffers, cattle exclusion fencing, and riparian forests are buffer types that are suggested as Best Management Practices (BMPs), designed for specific problems associated with different agricultural production systems. In many cases, such conservation practices have demonstrated a reduction of agricultural impacts on adjacent streams (e.g. Osborne et al., 1993; Lowrance et al., 1995; Edwards et al., 1997; Raffaele et al., 1997; Vellidis et al., 2003).

The benefits of buffers are well known and several studies have shown effects such as the removal of sediment and nutrients such as $\text{NO}_3\text{-N}$ (Lowrance, 1992; Verchot et al., 1997; Vellidis et al., 2003), as well as controlling pesticide transport from surface and subsurface flow (Lowrance et al., 1997; Vellidis et al., 2002). Although an extensive amount of data is available, there still remain essential questions concerning buffer effectiveness. Limited data exist in the Southeastern U.S. and even fewer studies are available with regard to buffer effectiveness in the Coastal Plain. Data available from these studies predominately consist of plot-size treatments, not larger whole farm studies. Additionally, only certain kinds of buffer practices in certain types of agricultural settings have been examined. Finally, conservation has been centered on the protection and restoration of public lands even though more than 70% of the entire landmass of

the conterminous U.S. and Hawaii, and over 90% of Georgia resides in private lands (NRCS 1996).

Biological Indicators and Water Quality Monitoring

Water quality monitoring is one tool used to assess the health of aquatic systems, and in turn, the effectiveness of conservation buffers. Biological monitoring in particular, has been recognized as one of the most useful tools in assessing water quality because biota are sensitive to environmental disturbances, show a wide range of responses, and are continuous monitors of their 'living' environment (Chandler, 1970; Karr and Chu, 1999; Linke et al., 1999). Ideally a useful or 'good biological indicator' would have the combined attributes of being holistic, early warning, diagnostic, abundant and tractable, readily sampled, occurring in high enough numbers for comparison (Rapport, 1992). Benthic macroinvertebrate assemblages have been often used in biological monitoring, especially to assess stream disturbances (Rosenberg and Resh, 1993). Considered excellent indicators of localized conditions, macroinvertebrates offer a spectrum of responses, inhabiting a wide range of environments while integrating the effects of short-term environmental variation (e.g. Lenat et al., 1980; Abel, 1989; Barbour et al., 1999). In fact, invertebrates have been shown to be sensitive indicators of changes in stream quality due to agricultural impacts (Lenat, 1984; Berkman et al., 1986; Gregory, 1996; Davis et al., 2003). More specifically, studies by Gregory (1996) and Davis et al. (2003) have also shown macroinvertebrates communities as valuable tools to evaluate the effectiveness of BMPs on streams affected by land-use disturbances in the Gulf Coastal Plain.

The monitoring of fish assemblages is also an integral part of monitoring programs, with extensive life history information available for many species. Although both groups of organisms fit many of the criteria for 'good indicators,' there are several disadvantages for each;

quantitative samples are difficult to obtain, species diversity may vary due to factors other than water quality, and in the case of invertebrates, the effort required to sort and identify specimens can be extensive. Yet studies have found both macroinvertebrates and fish extremely useful in monitoring programs, with the advantages far outweighing the disadvantages (Berkman et al., 1986; Nerbonne and Vondracek, 2001).

Amphibians as Bioindicators

Amphibians are thought to be highly sensitive to disturbances due to their complex life histories, which use both terrestrial and aquatic environments, require specific microhabitats, and specialized physiological adaptations (Wake, 1990; Blaustein et al., 1994; Stebbins and Cohen, 1995). These adaptations can cause them to be susceptible to minor environmental perturbations which alter their ability to seek cover from predators or forage on primary prey such as zooplankton, insects, and other invertebrates (Welsh and Ollivier, 1998). Amphibians, especially salamanders, are excellent monitors of local conditions because they have fairly limited home ranges (Blaustein and Wake, 1995), their life stages involve many parts of the environment, and they are relatively long-lived species, having extended developmental times (Petranka, 1998). The state of Georgia supports an extremely diverse assemblage of stream amphibians, with over 80 species occupying a variety of freshwater niches from seeps and headwaters to higher-order streams (Corn et al, 2003). Furthermore, many species have lotic-dwelling larval stages which are highly specialized in using stream microhabitats for both cover and foraging (Welsch and Ollivier, 1998). In small headwater streams, salamanders can occur in high numbers and provide another tool to assess stream habitat quality where other species assemblages, such as fish, may be less diverse or absent (Rocco, 1999; Corn et al., 2003).

Numerous studies within the western U.S. have incorporated amphibians into assessments of impacts of forest management practices within riparian zones (e.g. Gomez and Anthony, 1996; Wilkins and Peterson, 2000; Welsh and Lind, 2002). The state of Ohio Environmental Protection Agency (EPA) has even established an amphibian IBI (index of biotic integrity) for isolated wetlands and is developing similar indices for headwater streams (Micacchion, 2002). Although a multitude of reasons favor the use of amphibians as ecological indicators, few studies in the eastern U.S. have been specifically designed to examine amphibian responses to disturbances within aquatic ecosystems, or have used them within a water quality monitoring program (but see Chazal and Niewiarowski, 1998; Bowers et al., 2000; Homyack and Giuliano, 2002; Ryan et al., 2002; Willson and Dorcas, 2002).

Pervasive amphibian population declines in North American are attributed to environmental alterations from wetland drainage, timber harvesting, agriculture, urbanization, stream pollution and siltation, and the introduction of exotic species (e.g., Blaustein and Wake, 1995; Petranka, 1998; Welsh and Lind, 2002). Within the state of Georgia, several amphibian species abundances and ranges have been reduced primarily due to habitat loss (GADNR 1999), but presently an effective monitoring and conservation program has not been established. Amphibian conservation efforts are worthy of establishment, as these taxa play a vital role in aquatic ecosystem dynamics, and their sensitivity to land disturbance and pollution may serve as an indication of ecosystem health (Vitt et al., 1990; Semlitsch, 2003). One possible tool for amphibian conservation is their inclusion in biological monitoring programs. This would offer crucial information to better understand the ecology of amphibians, such as environmental limitations, and perhaps also serve to educate and include humans in conservation solutions.

Potential threats also need to be identified, along with the development of strategies promoting riparian protection.

The objectives of this study were to evaluate differences in the macroinvertebrate and amphibian community composition and abundance, riparian composition, physical habitat composition, and overall water quality between buffered (fenced from cattle access) and unbuffered (unfenced streams; cattle have access to streams) sites in southwest Georgia. It was important to also determine potentially useful metrics for stream water quality in this region, and to address the role amphibian species may play in bioassessment methods, with a hope to extend conservation efforts for this faunal group.

Study Area Description

The study was located in Early County, Georgia which is located within the Eastern Gulf Coastal Plain of southwest Georgia, in the physiographic district known as the Fall Line Hills (FLH) (Figure 2.1). The Fall Line is the boundary between the older crystalline rocks of the Piedmont Physiographic Province and the younger unconfined Cretaceous and Tertiary sediments of the Coastal Plain Province. The southern boundary of the FLH separates this District from the younger (Eocene-Paleocene) low-lying Dougherty Plain District (Clark and Zisa 1976, USGS 1996). The FLH is an area characterized by flat-topped ridges and little level land, and is dissected by frequently meandering streams, creating valleys that lie 15-75m below the adjacent ridge tops. Underlain by easily eroded sands, clays and gravels, this region contains numerous sinkholes and springs, with narrow stream terraces (Brantly 1916, Clark and Zisa 1976, USGS 1996). The erodability of the strata, coupled with the high altitude of the plain above the rivers, abundant rainfall, cultivation of the land, and timber removal, have caused the area to experience extensive erosion, forming steep gullies or washes (SWG RDC 1998).

Geomorphically, streams in this region are sinuous, typically lacking riffles and shoals common to the Piedmont. However, as is implied by the name, streams flowing near the area of contact (the Fall Line) undergo abrupt changes in gradient, exhibited by the presence of shoals and rapids. Deeply incised into underlying aquifers (Claiborne, Clayton, and Providence aquifers) these streams also receive considerable amounts of ground-water discharge (USGS 1996).

Ultisols and entisols are the two major soil orders present in the region, and characterize the pattern of soil leaching and runoff potential for the area due to their sandy and loamy horizons, which are subject to active erosion. The upland areas have level to strongly sloping, well drained soils, most of which comprise a sandy or loamy surface layer and loamy or clayey subsoil (Soil survey of Calhoun and Early Counties, 1985). Floodplains are dominated by nearly level, poorly drained loamy soils whereas the low stream terraces have a moderately well-drained, loamy surface layer and clayey subsoil.

The climate of this subtropical temperate region is warm and humid, with long hot summers, due to the close proximity to and the moist tropical air from the Gulf of Mexico. The winters are short and snowfall is rare because minimum temperatures dip below freezing for only brief periods of time; on average there are 235-255 frost free days per year (Southeast Regional Climate Center, SERCC). Average monthly temperatures range from 2.7-15.3 °C in January to 20.8-33.3°C in July (SERCC). Average annual precipitation is 141.8cm, with the average minimum rainfall occurring in October (6.6cm) and the maximum in January (15.9cm) (SERCC).

Until the early 1800's, the longleaf pine (*Pinus palustris*) and its associated communities covered and dominated the upland landscape of Early County, and more extensively, the Coastal

Plain of the southeastern United States (Early Co. Historical Society, 2002; Georgia Wildlife Federation, 2001). For centuries, American Indians altered the virgin forest (Stewart 1956), followed by extensive clearing and farming by European settlers (Turner and Ruscher, 1988). Between the years of 1866-1890, Coastal Plain forests were also extensively cut, and by 1895 virgin pine timber was exhausted (Plummer, 1975). From 1960-1980 the county experienced a significant change in land-use. Much of the forestland was converted to agricultural use, in which forestland now makes up 48% of Early County (USDA/SCS 1981). The dominant forest types of the region today are longleaf-slash pine (*Pinus palustris* - *Pinus elliottii*) and loblolly-shortleaf pine (*Pinus taeda* - *Pinus echinata*), with oak-gum-cypress (*Quercus-Nyssa-Taxodium*) occurring along river floodplains. Presently, the riparian area of the study sites is dominated by a second-growth mixed forest comprised of tree species such as *Magnolia grandiflora*, *M. virginica*, *Nyssa biflora*, *Quercus nigra*, and *Q. rubrum* (pers obs).

As previously mentioned, agriculture has been a dominant land use within southwest Georgia, and as one of the Nation's most productive agricultural regions, it leads the state and Nation in many sectors of agricultural production (SWG RDC 1998). From its beginning in 1818, Early County has been an agricultural area where even almost two-hundred years later, the local economy is still determined by the local crop year (Early Co. Historical Society, 2002). The county is presently a mix of cropland and pasture, with over 50% of the county in farmland. Subsistence crops such as corn, oats, rye, and wheat were grown by early settlers. After the Civil War, farmers urgently needed a cash crop to rebuild the farming operation. Cotton was selected and it continues to be one of the main crops grown along with corn, soybeans, sorghum grain, small grain, peanuts, and pecans. Due to the high amount of frost free days, it is not uncommon for the land to be double cropped. One of many farmland management concerns in the county

has been soil erosion and low soil fertility. Despite these problems, crops have responded well to nutrient additions and irrigation (SWG RDC 1998).

The study was conducted on the Brownlee Farm, located within the 68 km² sub-watershed of Factory Creek in Early County (Figure 2.2). The farm is a diversified row crop and beef cattle operation that has been in production for over 30 years. The stream site is the location of the first factory in the county, built in 1855 for spinning cotton, hence its name. Factory Creek is a 2nd order tributary of the Lower Chattahoochee River, which has been designated as impaired under the Clean Water Act Section 303(d) due to concerns of altered lead, dissolved oxygen, and fecal coliform bacteria concentrations (U.S. EPA, 2000). The Chattahoochee River, named for a Creek Indian word meaning ‘painted rock,’ drains an area of 22,714 km² and flows 692 km to its confluence with the Flint River. One of the most heavily used water resources in Georgia, the flow of the river is controlled predominantly by hydropower plants releasing water for the production of electricity (USGS 1996).

The Brownlee Farm was used as a demonstration site for the Georgia Stream Buffer Initiative (GSBI), a statewide interagency partnership with the purpose of increasing buffer awareness and understanding among Georgia landowners, while accelerating the adoption of conservation buffers across the state. The GSBI, headed by the Upper Ocmulgee River Resource Conservation and Development Council, was created in response to Georgia’s low participation in buffer enrollment for the National Buffer Initiative and overall low enrollment in conservation assistance programs such as the Conservation Reserve Program (CRP). The Initiative is based on education and Best Management Practice (BMP) implementation and demonstration, with over 20 demonstration sites throughout Georgia, in three different production systems: cropland, grazing, and silviculture. Demonstration sites were located primarily on private or working

lands, with the intention of using these sites for landowner education on the anticipated improvements from management techniques incorporating conservation buffers. Thus water and wildlife monitoring of these demonstration sites was a critical component and objective of the project.

MATERIALS AND METHODS

Overview of Study

The landowners were contacted prior to the commencement of this study, and consulted for access to stream reaches and informed of the project's activities and progress throughout its duration. National Resource Conservation Service (NRCS) district conservationists for the county were also consulted and used to facilitate communication with the participating landowners.

Five sampling sites in the Factory Creek basin were selected for biological, chemical, and physical assessment (Figures 2.3 and 2.4). All sites were located within the Brownlee farm, three sites were unfenced (UF), which will be referred to herein as UF-1, UF-2 and UF-3, and two were fenced sites (F) or reference sites, F-1 and F-2. Study site descriptions are as follows: **UF-1** was located near a heavily eroding gully and was surrounded by cotton fields and pastures planted to rye grass (*Secale cereale*) and bahiagrass (*Paspalum notatum*). Prior to this study, the upland area surrounding the gully was fenced from cattle and seeded with grass as a conservation effort to restore the area and prevent sediment from washing into the nearby stream. Due to drought conditions, little vegetation growth was established, thus allowing sediment loads to continue to wash into the stream following heavy rainfall (Figure 2.5). During the entire study, cattle had direct access to the stream and traveled down steep slopes (8%) for water, further contributing to slope erosion.

UF-2 and **UF-3** were surrounded predominately by pastures planted to bahiagrass. Cattle had direct access to the streams prior to and during the study. Compacted soil and severe bank erosion was evident. Understory vegetation was sparse, if present, tree roots were exposed, and many trees had fallen both within the stream (especially at site UF-3) and in the floodplain (Figures 2.6 and 2.7).

F-1 and **F-2** were located within the same sub-basin. Both streams were fenced from cattle for 25 years or more prior to the commencement of this study and were located down slope from pastures, cotton fields, and pine plantations (Brownlee, personal communication.). This area of the property was logged over 70 years ago and allowed to revegetate naturally (Figures 2.8 and 2.9). The pine plantations were planted to loblolly pine at intervals of 7 and 15 years ago (Figure 2.4).

Reference sites (F-1 & F-2) were chosen based on preliminary biotic index scores using the Adopt-A-Stream index for stream macroinvertebrates, which assigns an index value (i.e. excellent, good, fair, or poor water quality) based on the tolerance and sensitivity of aquatic taxa (GADNR, 2002). From a preliminary sample, site UF-1 rated as ‘poor’ with only four orders present (diptera, hemiptera, zygoptera (sub-order), and odonata), whereas the reference site F-1 rated ‘good’ and contained seven orders (ephemeroptera, plecoptera, trichoptera, hemiptera, coleoptera, diptera, and decapoda). The stream reaches selected for this study offered the best possible reference conditions, which are often difficult to find in a region dominated by a long history of environmental alterations.

At each stream site, a representative 100 meter transect was selected and marked for sampling purposes. Streams were sampled biweekly for water quality from February 2002 until March 2003. Bimonthly collections of macroinvertebrates occurred from February 2002 until

February 2003 for a total of seven sampling dates. Herpetofaunal surveys were also conducted on a bimonthly basis (except tree pipe refugia which occurred monthly), beginning March 2002 through March 2003. Tree pipes were surveyed from June 2002 to March 2003. Vegetation surveys occurred once at each site and were conducted during the months of September and October 2002.

Physical Measurements

Stream habitat evaluations were conducted once at each site, with physical characterizations consisting of general land use, description of stream origin and type, and measurements of channel morphological characteristics, including stream bankfull width and depth. Physical habitat assessments were also conducted once using the ‘EPA Rapid Bioassessment Protocols for Use in Wadeable Streams and Rivers;’ rating stream habitat parameters on a numerical scale of 0 to 20 (highest) for each sampling reach (Barbour et al., 1999). To provide for a habitat ranking, ratings were totaled and compared to a reference condition, which in this study were the fenced sites.

Stream flow velocity (Marsh McBirney® Flowmate) and depth were measured at 20 cm intervals across each cross-stream transect on invertebrate sampling dates. In addition, on biweekly grab sample dates and invertebrate sampling dates, temperature and dissolved oxygen concentrations were measured in the field with a dissolved oxygen meter (YSI Model 50B, Yellow Springs, OH). In order to obtain a representation of conditions in the benthic habitat, readings were taken at the sediment water interface. Water depth was measured with stationary staff gauges, although at some sites gauges periodically were dislodged during spates. Temperature loggers (HOBO® Temperature logger, Pocasset, MA) were placed on a selected tree

at each site in the riparian area, set to record air temperature (° C) every hour. Loggers were launched, recovered, and data downloaded every 75 days from February 2002 to March 2003.

Chemical and Biological Water Quality Measurements

Grab samples of 5000 ml were collected biweekly from each stream. All samples were kept on ice until processing. Nitrate (NO₃-N), ammonium (NH₄-N), ortho-phosphate (PO₄-P), and chloride (Cl⁻) concentrations were determined using a Brann and Luebbe TRAACS 800 autoanalyzer following EPA approved colorimetric techniques (Greenberg et al., 1992). Total kjeldahl nitrogen (TKN) was quantified using an EPA approved digestion and titration technique (Tecator Autosampler 1035/1038) (Greenberg et al., 1992). Total suspended solids concentrations were determined using conventional techniques, and were dried at 103-105 °C (Method 2540 D) (Greenberg et al., 1992). Turbidity measurements were made within 24 hrs of sample collection, and unfiltered, room temperature samples were analyzed for apparent color with a platinum-cobalt standard scale (HACH, DR/2000 Spectrophotometer, Method #120). At each grab sample date, one 100mL water sample from each site was collected in a single-use sterile sampling bag, stored on ice, and analyzed for *Fecal coliform* and *Fecal streptococci* colonies within six hours of collection following conventional membrane filtration techniques (Greenberg et al., 1992). Due to laboratory analytical availability, on each grab sample date from February 2002-June2002 only three stream sites were sampled for bacterial concentrations.

Aquatic Macroinvertebrates

Bimonthly, benthic macroinvertebrates were sampled using a 500 µm mesh Hess sampler (Hess, 1941). Within the selected 100 m reach at each site, three random stream transects were chosen. At each transect, composite samples comprised two Hess collections, sampling representative habitat types within the stream channel. Streambed composition (sand, gravel,

roots, etc.) was visually estimated by the line intersect method. Benthic stream material such as leaf and woody debris, sand, and pebbles was also washed into each sample to be separated and analyzed later for organic content. Samples were rinsed into plastic bags, preserved in the field with 70% ethanol and stained with rose Bengal dye. In the laboratory, processing of entire samples involved washing invertebrates from organic matter through a 1mm and 500 µm sieve to be later sorted. Identifications were then made to the lowest practical taxonomic level, most often genus, under a low power dissecting microscope and light compound microscope (Berner and Pescador 1988; Stewart and Stark 1988; Pescador et al. 1995; Epler 1996 and 2001; Wiggins 1996; Needham et al. 2000; Thorp and Covich 2001). Larval Chironomidae (Diptera) samples from two dates, February and August 2002, were mounted on microscope slides in CMC and identified to genus (Epler, 2001). Samples of more than 500 individuals were subsampled and adjusted to a final volume of 100 ml with 70% ethanol, placed on a magnetic stirrer to produce a homogenous solution, and then three 5 ml subsamples were removed with a wide-bore pipette (Hax and Golladay, 1993).

Coarse organic material was removed from invertebrate samples, and oven-dried (70° C, 24h), ground in a Wiley-Mill, subsampled, weighed, ashed, and reweighed to obtain ash-free dry mass (AFDM) as an estimate of benthic organic matter.

Amphibians

Five methods were used to measure amphibian diversity and abundance: (1) searches of natural cover objects along transects (Jaeger, 1994); (2) artificial cover boards (Fellers and Drost, 1994); (3) tree pipe surveys (Boughton, 1997), (4) dip netting (Heyer et al., 1994), and (5) larvae collected within Hess samples. Reptiles and amphibians observed opportunistically were also noted. Artificial and natural cover searches were conducted bimonthly (March 2002 – March

2003), tree pipes were surveyed on a monthly basis (June 2002- March 2003), and dipnetting occurred once in May of 2002. The sampling design at each site comprised natural/artificial cover transects and tree pipes, all located within a 100 m x 4 m strip along the stream (Figure 2.10). Surveys were conducted on only one side of the channel due to narrow stream terraces. All species of amphibians encountered were identified and released at the point of capture. The presence of any malformations, disease, or ectoparasites were recorded for each individual. Microhabitat measures at the time of sampling included air and soil temperature, cloud cover and weather conditions.

Artificial cover arrays consisted of five cover board stations, each spaced 20m apart at sampling transects. Each station included three pairs of boards (30 cm x 30 cm x 2.54 cm) , with one set located 4 m, 2 m, and 0 m (from the stream edge), for a total of 6 boards at each station, and 30 boards at each site. Boards were placed flush to the mineral soil, secured by a sod staple, which ran through a hole drilled in the board, and then covered with leaf litter. Many sections of the stream reaches were so deeply incised that in some cases boards were not able to be placed at water level. For example, a 0 m cover board pair was 0 m horizontally from the stream's edge, yet still was more than 0.5 m vertically from the water. Cover board material came from an old collapsed barn, which exposed the unpainted pine siding to weathering conditions for more than a year. Surveys began six weeks after boards were placed at the study site. Board location was also recorded for each amphibian captured.

The natural cover transects were surveyed by two observers, turning (and replacing) all natural cover objects (logs, sticks, rocks) within the transect area. In addition to the previously mentioned parameters collected, number of cover objects turned was also recorded.

White polyvinyl chloride (PVC) pipes provided artificial refugia for arboreal frogs and were placed at each cover board station to capture hylid treefrogs (Moulton et al., 1996; Boughton, 1997). Two pipes were hung vertically on trees at each station, for a total of ten pipes at each site. Tree pipes consisted of 5 cm diameter x 61 cm long, schedule 40 PVC capped at one end. Drain holes were placed ca. 10 cm above the cap and 0.61 cm lengths of 0.64 cm diameter rope were placed in tree pipes to allow arboreal rodents to escape. Water was added to the pipes to provide a moist environment. Pipes were painted brown on the outside for concealment and each pipe was marked with station (A, B, C, D or E) and direction location (north or south). Tree pipes were mounted on nails placed on the north and south face of trees at approximately 2 m above ground. Trees were chosen based on proximity to cover board station, with all trees located within the 4 m-width area. Each trap was removed from the tree for inspection, and cleaned of anoles, insects, and spider webs. Frogs were removed for identification and returned to the same pipe from which they were found. Pipes were put in place four weeks prior to the first survey period and were checked every month between 09:00 and 16:00 from June 2002 – March 2003. Tree pipes were hung on 12 different species of trees, predominantly consisting of a mixture of hardwood species, however one tree was a snag and one was a pine (*Pinus taeda*). Most sites had at least 3 different tree species upon which tree pipes were hung, and overlapped in species with other sites. Most tree pipes were hung on two species, *Liriodendron tulipifera* (Tulip tree) and *Nyssa biflora* (Swamp tupelo).

Sampling for amphibian larvae occurred during May 2002 using a 4 mm mesh dipnet. Within each stream site, 5 cross stream belt transects were established, placing one belt every 20 m (Figure 2.10). Belt transects were 1m wide and extended from bank to bank. Three composite sweep samples were taken within each belt, sampling all habitat types (i.e. leaf beds, sand,

gravel). Species and number of larvae collected at each site were recorded for species richness accounts.

Salamander larvae recovered from preserved invertebrate samples were identified (Petranka, 1998) and enumerated, and used for feeding electivity studies (Chapter 3).

Vegetation surveys

To characterize forest structure riparian habitat composition, a belt transect design was utilized along each stream reach (Figure 2.11). Each transect consisted of four consecutive plots (5 m x 25 m). Due to narrow floodplain terraces on one side of the stream channel, transects were positioned on the wider terrace (the same side on which herpetofaunal transects were conducted). The belt transects ran along and followed the natural configuration of the channel, with the stream's edge marked as 0 m and used as the reference point for plot placement.

All overstory and understory trees > 2.5 cm diameter breast height (DBH) were identified to species and DBH was recorded. For each transect, mean density, basal area, and importance values were determined for each species. Within each plot, all shrubs (DBH < 2.5 cm, but >30 cm in height) were identified to species and % coverage of each species was estimated using seven cover classes (Appendix A.I). Coarse woody debris (CWD) greater than 5 cm in diameter was quantified by size class based on diameter and length classes (Appendix A.II). Standing dead trees (snags) were noted and DBH of each was recorded. A robel pole (cover estimation technique) was used to estimate vertical obstruction of the understory (Robel et al., 1969). A pole (180 cm length) was divided into six 30 cm segments and positioned vertically in the vegetation 8 m from the center of the plot. The amount of pole obscured from view by vegetation was recorded for each pole segment. Two pole readings per plot were taken, one in the upstream direction and then one downstream, observing the pole from a fixed height of 1 m.

Ground layer coverage type was estimated across each plot using a line-transect method. Across each plot lengthwise, a vertical pole was dropped along a transect at 1m increments, noting the cover in each of the following categories: sand, live vegetation, wood, leaf litter, and other organic matter.

Nested within each plot, five 1 m x 1 m frame quadrats (sub-plots) were established, one at each corner, and one at the center of the plot. Within each sub-plot, ground cover (<30 cm in height) species presence and % coverage of graminoids, forbs, woody debris (all sizes; coarse and fine), and bare ground were recorded using the same cover classes as for the shrubs (Appendix A.I). Vegetation for all plots and sub-plots was identified to species level when possible (Wunderlin, 1998). Percent canopy cover was determined with a spherical densitometer (Model-C, R.E.Lemmon Forest Densiometers, Bartlesville, OK) with a measurement in the center of each subplot, in the downstream direction. Two densitometer readings were also taken at each invertebrate transect collection, one upstream and one downstream from the middle of the stream channel. Soil compression was measured within each quadrat of the subplot with a pocket penetrometer, which measures the compressive strength of the soil (kg/cm^2). On both sides of the stream channel, within the selected transect area, floodplain width was measured and slope was estimated with a percent scale clinometer.

Data Analysis

Multiple parameters and metrics shown in previous studies to be valuable indicators of water quality were tested in this analysis, and new indicators were developed that might prove effective in assessment of streams disturbed by agriculture. Invertebrate metrics were chosen based on those found valuable for bioassessment (and for this region) and those able to be included due to taxonomic resolution (see Rosenberg and Resh, 1993; Barbour et al.1996; Davis,

2000) (Table 2.1). A multimetric approach using an index of biological integrity (IBI) for benthic macroinvertebrates was not utilized in this study because either many of the organisms were in early instars or the expertise to identify the individuals to species was not available. Oligochaeta, Hydracarina, and Copepoda were not adequately sampled during Hess invertebrate collections, and were excluded from the analyses.

Principal components analysis (PCA), an ordination technique, was used to visualize and compare physical, vegetative, water quality parameters, and macroinvertebrate metrics among sites. The object of the analysis is to represent a data set containing many variables with a smaller number of composite variables known as principal components or axes. Some parameters were measured with more than one technique (i.e. soil coverage methods), and if a strong correlation was evident between data ($r^2 > 0.70$), then the more rapid technique was used in the PCA. PCA options were Euclidean distance and cutoff r^2 was set at 0.25 (McCune and Mefford, 1995). In each PCA, three axes were selected for interpretation based on broken-stick eigenvalues. An outlier analysis was conducted to identify sites that were located more than two standard deviations from all other sites. Each stream was designated with separate symbols and the results of the ordination were interpreted by looking at each site for all dates combined to see if differences were apparent. Ellipses were subjectively drawn around the central locations of points from each site to guide and aid the observation of patterns.

Relationships between the axes and variables are important to pay attention to when interpreting the PCA ordinations. First, variables or vectors furthest from the origin explain the most variation. Variables close together are positively correlated, with those on opposite sides of the axis negatively correlated, and those at right angles are not correlated. Finally, sites close together are similar and those far apart are dissimilar.

Based on the results of the PCA, variation in each metric and parameter was compared between stream sites for all dates combined. Comparisons were performed using a Kruskal-Wallis Test ($p < 0.05$) (SAS Institute, Inc. 2002). Box plots were then constructed to illustrate the differences and similarities at the study sites. Due to different collection dates for bacterial samples, statistical analyses were not used for this data. Coverage percentages used in analyses for shrub and ground cover classes were generated by using the median value of the cover class midpoint (i.e., class 2 (1-5 % cover) would use 2.5 as the coverage percentage).

Abundance data were reported for all hydrid species combined and all salamander species combined, and for any individual species that was found statistically significant. Comparisons were performed using a Kruskal-Wallis Test ($p < 0.05$) (SAS Institute, Inc.2002).

Overstory plant species importance values (IV) were calculated for each site by averaging the relative dominance (as expressed by basal area), relative density, and relative frequency for each site. Jaccard's index of similarity (JI) was used to compare vegetation by site (Ludwig and Reynolds 1988).

The Georgia Adopt-A-Stream (AAS) index score and ranking (e.g. excellent, good, fair, or poor water quality) for stream macroinvertebrates was calculated for each stream during all sampling dates to provide information for the AAS database and provide a comparison to other methods of bioassessment evaluated within this study (GA DNR, 2000). Georgia Adopt-A-Stream is a volunteer stream monitoring program with the goals of increasing public awareness of the State's water quality issues and providing citizens with tools to protect and evaluate their local waterways, while also providing critical baseline data.

RESULTS

Physical Parameters

Throughout the study all streams maintained flow, with maximum velocities occurring during the winter months (Dec 2002 and Feb 2003) (Figure 2.12). Overall, site UF-3 had highest flow velocity whereas fenced sites F-1 and F-2 had the lowest velocities ($p=0.0011$). Stream temperatures and dissolved oxygen levels, were statistically similar at all sites. However, site UF-2, a wider and deeper stream, had noticeably lower dissolved oxygen concentrations for a majority of the study, with higher stream temperatures in the spring to early fall season (Apr-Oct 2002) and lower temperatures in the winter/early spring seasons (Nov 2002-Mar 2003) (Figure 2.13)(Table 2.2).

The PCA for physical parameters determined that the first two axes explained 49.4% of the variation, and cumulatively 65.4% with the inclusion of the third axis (Figure 2.14; Axis 3 not shown). Percent open canopy (over the stream) was negatively correlated with Axis 1 ($r^2 = 0.48$), whereas % leaves, stream depth, and AFDM (ash-free dry mass; kg/m^2) were positively correlated with Axis 1 ($r^2 = 0.52, 0.83, \text{ and } 0.53$, respectively). Dissolved oxygen levels were positively correlated with Axis 2 ($r^2 = 0.52$), and one variable, % pebble/rock was positively correlated with Axis 3 ($r^2 = 0.57$). Fenced sites F-1 and F-2 and unfenced site UF-2 tended to have deeper water depths, lower percentages of open canopy, higher amounts of benthic organic matter (AFDM), and higher percentages of leaf debris and wood/roots. Unfenced site UF-1, the gully site, was characterized by higher percentages of open canopy and exposed streambed, and was similar to site UF-3 with high percentages of sand/silt in the streambed.

The unfenced sites differed in physical parameters measured, whereas fenced sites were very similar. Site UF-1 had the highest % sand/silt, % exposed streambed, % open canopy (over

the stream), and width to depth ratios, and lowest % wood/roots, % leaves, and AFDM ($p < 0.05$) (Table 2.2). Unfenced site UF-2, overall the widest and deepest stream, had the highest % leaves and AFDM, and lowest width: depth ratios ($p < 0.01$) (Table 2.2). UF-1 also had the lowest % pebble/rock. The third unfenced site, UF-3, was overall the narrowest in width, and maintained the fastest velocities with also higher percentages of pebbles within the streambed ($p < 0.01$) (Table 2.2). Fenced sites F-1 and F-2 overall had slower stream velocities, the lowest percentages of sand/silt, and highest % wood/roots (Table 2.2).

From the EPA physical habitat assessment ranking, fenced and unfenced streams displayed many critical differences in stream habitat quality. Overall habitat scores were highest for fenced sites F-1(174) and F-2 (143), indicating better habitat quality (Barbour et al., 1999) (Appendix B). Unfenced site UF-2 (128) was relatively similar to fenced sites, yet scored lower in parameters such as channel sinuosity, bank stability, and vegetative protection. The other two unfenced sites UF-1 (65) and UF-3 (80) scored lower in habitat quality, with extremely poor rankings in bank stability, vegetative protection, epifaunal substrate, pool substrate and variability, channel flow status, and sediment deposition. Habitat diversity was also highest at the fenced sites with variable riffle-pool-run habitat available, and only 100% run habitat available at any of the unfenced sites. Diverse inorganic and organic substrate composition was also lacking at the unfenced sites, and predominately consisted of sand (85-99% of substrate components), with little detritus (sticks, wood, coarse plant materials; 2-15 %). At the fenced sites sand was still the dominant component (55-65%) yet more substrata types were present at these sites: cobble (5%), gravel (15-20%), and detritus (20-25%). F-1 had especially higher percentages of boulder substrate (20%).

Water Quality

The first two axes from the water quality PCA explained 56.3% of the variation, with no parameters explained by the third axis (Figure 2.15). NH₄-N, suspended solids, and turbidity were positively correlated with Axis 1 ($r^2 = 0.48, 0.53, \text{ and } 0.61$, respectively). Total-Nitrogen and NO₃-N were strongly positively correlated with Axis 2 ($r^2 = 0.94 \text{ and } 0.77$, respectively). Site UF-3 was characterized by higher levels of Total-N and NO₃-N, whereas site UF-2 had lower levels. The unfenced sites were similar in that they had relatively higher levels of Cl⁻, TKN, suspended solids, turbidity, NH₄-N, and PO₄-P, with site UF-1 having the highest levels of all sites. Both unfenced sites F-1 and F-2 were characterized by low levels of nutrients and sediments.

Fenced sites were relatively similar in their nutrient and sediment concentrations over time, whereas the unfenced sites, although similar in some parameters, had greater variation in bacterial and nutrient concentrations (Table 2.3; Figure 2.16). Turbidity and suspended solid levels were significantly lowest at the fenced sites ($p < 0.0001$) and trends show these sites to be more stable over time, with unfenced sites showing pulses of suspended sediments after storms (Figure 2.17). The lowest mean concentrations for TKN, PO₄-P and NH₄-N were also measured at fenced sites ($p < 0.0001$), and were consistently lower than unfenced sites (Figures 2.18 and 2.19). Patterns at the unfenced sites varied, with the highest PO₄-P and NH₄-N levels at two of the three unfenced sites, UF-1 and UF-2 ($p < 0.0001$) (Figures 2.18 and 2.19). Both the highest and lowest nitrate levels however, were found at unfenced sites UF-3 (highest) and site UF-2 (lowest) ($p < 0.0001$) (Figure 2.16). Similar patterns were also found for concentrations of Total-N (Figure 2.20), where again, highest mean levels were found at unfenced site UF-3, and lowest levels at site UF-2 ($p < 0.0001$).

Bacterial concentrations between study streams were not analyzed statistically because all sites were not sampled on each date, yet patterns were able to be inferred from geometric means and trend line data. Mean fecal coliform (FC) and fecal streptococci (FS) levels varied over time, with overall highest FC levels peaking in the fall season (Aug-Sept-Oct 2002) and highest FS levels occurring in the spring season (Feb-Mar-Apr 2002) and again in the winter season (Nov-Dec 2002) (Figure 2.21). Highest concentrations of both fecal forms were consistently found at unfenced sites UF-3 and UF-2, and lowest levels were found at fenced sites F-1 and F-2, and unfenced site UF-1. Overall, mean bacterial concentrations appeared to fluctuate, especially at the unfenced sites, with small fluctuations at the fenced sites (Figure 2.21). In the case of FS levels, UF-1 also appeared with low, constant levels. In comparison to Georgia's water quality standards established by the Georgia Department of Natural Resources-Environmental Protection Division, mean FC concentrations were higher than the designated state standard of 200 CFU/100 mL during the recreational season of May-Oct (Figure 2.21) at all the unfenced sites (Table 2.4). No site concentrations were above the standard of 1000 CFU/100 mL for waters designated for drinking and fishing (November-April). According to ratios established by Geldrich et al. (1967), mean FC to FS ratios mostly stayed between the range of 0.1-0.7, indicating bacterial contaminations originated from livestock (FC/FS>4: human contamination; 0.1<FC/FS<0.7: livestock; FC/FS<0.1: wildlife) (Table 2.5).

Macroinvertebrates

A total of 23,840 individual organisms were collected at the five study sites during the seven sampling dates (Appendix C). Forty-two taxa were identified, with greatest numbers of taxa in the orders Trichoptera (10 taxa) and Diptera (9 taxa), followed by Coleoptera (5 taxa) and Odonata (4 taxa). Throughout the study, abundance at all sites was dominated by two main

groups of macroinvertebrates: Dipterans (87%), of which 88% were of the family Chironomidae, and Coleoptera (8%) of which 73% were of the family Elmidae (Figure 2.22). Diptera were found consistently in all samples, and were the most abundant at the unfenced sites ($p < 0.0001$), whereas Coleoptera were significantly higher at the fenced sites ($p < 0.0001$), yet not present on all sampling dates (Figure 2.23).

Throughout the study overall mean taxa richness peaked twice, during the early summer and winter sampling seasons (June and December 2002), with decreases in the late summer and early fall (August and October 2002) (Figure 2.24). Fenced sites F-1 and F-2 had overall significantly greater invertebrate richness than the unfenced sites, with site UF-1 having the lowest numbers of taxa ($p < 0.0001$). Overall mean taxa richness per site ranged from 0-16 taxa, with mean numbers of taxa collected ranging from 0-6 at the unfenced sites, and 5-10 at the fenced sites. EPT taxa richness was very similar to overall taxa richness patterns, with peaks at the June and December 2002 dates, although some sites (F-1 and F-2) peaked in the fall (October 2002) (Figure 2.24). Overall, EPT taxa richness was very low, especially at site UF-1, which had a mean range of 0-2 taxa, and was highest at the fenced sites, which ranged from 0-8 taxa ($p < 0.0001$).

Mean invertebrate abundance (no. /m²) varied by site and season and no overall pattern was evident (Figure 2.25). The fenced sites peaked in different seasons, and abundance at site UF-1 and UF-2 appeared somewhat consistent through time, whereas at UF-3 levels fluctuated with patterns similar to taxa richness. All of the sites had similar numbers of stream invertebrates, except UF-1, which had significantly lower abundances ($p < 0.0001$). Dipteran abundance (no. /m²) patterns varied through time, with lowest numbers at site UF-1 ($p < 0.0001$) (Figure 2.23). Sites UF-2 and F-1 showed season fluctuations while the remaining sites displayed

somewhat consistent abundance levels. Coleoptera abundance however, showed highest levels at the fenced sites, with peaks in the spring (April 2002) and early winter seasons (December 2002) and lowest numbers at UF-1 ($p < 0.0001$) (Figure 2.23). The number of individuals grouped as clingers (those being mostly Coleoptera) was also highest at the fenced sites exhibiting similar trends to Coleoptera abundance ($p < 0.0001$) (Figure 2.26). Crustacean abundance, which was predominantly Amphipoda, displayed highest numbers at the fenced sites ($p < 0.0001$). Abundance at fenced sites differed slightly, with one high peak in the early spring at site F-1, which then fell to low abundances for the duration of the study, and two peaks at F-2 during the early summer and fall/winter seasons (June, October, December 2002) (Figure 2.26).

Certain invertebrate taxa were found only at particular stream sites. Although many were low in abundance, some occurring once throughout the study, they are worthy of mention. The fenced sites contained more exclusive taxa, and many of those taxa have been noted in the North Carolina Biological Index as being sensitive to environmental disturbances (Lenat, 1993). The unique taxa at fenced site F-1 were: *Dixa* and *Dixella* (Dixidae), *Perlesta* (Perlidae), *Lepidostoma* (Lepidostomatidae), and *Molanna* (Molannidae), all of which are sensitive taxa (Lenat, 1993). Site F-2 also had sensitive taxa such as *Lype* (Psychomyiidae) and *Hydroptila* (Hydroptilidae), as well as less sensitive invertebrates such as Curculionidae, *Trepobaks* (Gerridae), and *Habrophlebia* (Leptophlebiidae). The unfenced stream sites had fewer numbers of unique taxa, mostly of the orders Coleoptera and Diptera, with none listed as sensitive. Site UF-1 had three taxa: *Hydrobiomorpha* (Hydrophilidae), *Prionocyphon* (Scirtidae), and one Hemipteran family, the Hebridae. Only found at site UF-2 were *Ancyronyx* (Elmidae) and *Nemotelus* (Stratiomyidae). Site UF-2 also harbored noticeably greater numbers of Mollusca than at any other sites (Appendix C). Site UF-3 had no unique taxa.

From the invertebrate metric PCA, the first two axes explained 47.5% of the variation, and 56.8% with the inclusion of the third axis (Figure 2.27; Axis 3 not shown). Axis 1 represents a shift from a group consisting of EPT and Elmidae to dipterans. Fenced sites F-1 and F-2 were positively associated with axis 1 and the metrics % EPT/Chironomidae, % Elmidae, % clingers, and % EPT ($r^2=0.69$, 0.70 , 0.68 , and 0.67 , respectively). A cluster of points from predominately the unfenced sites were characterized by high % Diptera and % Dominant Family, which were both negatively correlated with the first axis ($r^2=0.86$ and 0.61 , respectively). The dominant family metric consisted mostly of chironomids, however some samples were dominated by elmids, simuliids, ceratopogonids (at UF-1), and tipulids. In addition, % Chironomidae ($r^2=0.49$) was negatively correlated with the second axis. Percentages of Ceratopogonidae ($r^2=0.72$) and burrowers ($r^2=0.91$) were both positively strongly correlated with axis 2 and associated with a group of points consisting mostly from site UF-1. The crustaceans, % Amphipoda ($r^2=0.22$) and % Decapoda ($r^2=0.53$) along with a chironomid subfamily, % Tanypodinae ($r^2=0.56$) were all negatively correlated with axis 3, associated with a mixture of sites dating mostly from February 2002.

Crustacean metrics such as % Crustacea, % Amphipoda and % Decapoda were significantly higher at the fenced sites ($p=0.001$) (Figure 2.28). No crustaceans were found at unfenced sites UF-1 and UF-3. Of all invertebrate samples, only one amphipod genus was identified, *Crangonyx*, and two decapod genera, *Cambarus* and *Procambarus*, all which are more tolerant taxa. (Lenat,1993) (Appendix C). The metrics % clingers, % EPT, and % Elmidae were also found highest at the fenced sites ($p<0.0001$) (Figure 2.28). One metric was found lowest at cattle exclusion sites, % Diptera ($p<0.0001$) (Figure 2.29). Site UF-1 the gully site, was characterized by the lowest percentages of many metrics including EPT, Chironomidae,

Tanypodinae, Elmidae, and EPT/Chironomidae ($p < 0.001$) (Figures 2.28 and 2.29). Collectively, the unfenced sites showed higher percentages of Dipterans, with lower percentages of Elmidae and Crustacea (Amphipoda and Decapoda) ($p < 0.001$).

Eleven total genera of chironomids were identified from the August 2002 sample, within three subfamilies: the Chironominae, Orthocladiinae, and Tanypodinae. Lowest diversity occurred at unfenced site UF-1 (2 genera) and greatest diversity at fenced site F-2 (9 genera). Only one genera was found at all sites, *Polypedilum*, and site UF-2 had noticeably higher abundance levels of *Saetheria*, whereas highest numbers of *Tanytarsini* were found at F-2. The following genera were also identified: *Ablabesymia*, *Cryptochironomous*, *Krenosmittia*, *Pseudochironomous*, *Thienemannimyia*, *Tribelos*, *Xestochironomous*, and *Zavrelimyia*.

The Georgia Adopt-A-Stream index for stream macroinvertebrates showed sites ranking anywhere from ‘poor’ to ‘good’ water quality (Figure 2.30). Both fenced sites overall scored a ‘good’ rating, with highly variable ranges for sites F-1 (6-25) and F-2 (9-33). Site UF-1 (1-13) ranked the lowest overall, ‘poor,’ and the other two unfenced sites UF-2 (4-18) and UF-3 (4-18) had similar ranges and both scored an overall ‘fair’ water quality rating. None of the sites scored ‘excellent’ except in December 2002 at fenced site F-2.

Amphibians

A total of 21 herpetofaunal species were observed during the study, although not all of these were target species during transect surveys (Appendix D). Of amphibians, three species of frogs within the Hylidae utilized tree pipe refugia: Gray treefrog (*Hyla chrysoscelis*), Green treefrog (*H. cinerea*), and the Squirrel treefrog (*H. squirella*). One Barking treefrog (*H. gratiosa*) was found within a treepipe at unfenced site B-3, after the conclusion of this study. Four salamander species were observed during natural and artificial cover searches, all within the

Plethodontidae: Apalachicola Dusky Salamander (*Desmognathus apalachicolae*), Southern Two-lined Salamander (*Eurycea cirrigera*), Southeastern Slimy Salamander (*Plethodon grobmani*), and the Red Salamander (*Pseudotriton ruber*). Species richness was estimated using these two groups, and no differences between sites were found, however from the ANOVA test it is suggested that there is some pattern, with richness estimates tending to be higher at the fenced sites ($p=0.04$). Capture rates were not high enough to analyze data on coverboard or natural cover occurrences for any salamanders. However, captures of these species were included in analyses for all species.

Cover board and natural cover searches

Searches for salamanders using natural and artificial cover (cover boards) occurred over seven dates, from March 2002-March 2003, however problems occurred with cover boards during the survey months of May and July 2002, when major rain events occurred. The data for cover board searches was not used for these two months, due to the large loss (over 50%) of boards.

Of the four salamander species found during the study, *E. cirrigera* composed 67% of total captures, while *P. ruber* composed 17%, *D. apalachicolae* 12%, and *P. grobmani* 4% (Figure 2.31). A total of 92 individuals were found during the entire study under natural and artificial cover (Table 2.6). Salamander abundance levels were very similar between survey methods, finding 39 individuals within natural cover surveys and 44 individuals during artificial cover surveys (using five survey dates when cover boards were present) (Figure 2.32). Abundance levels (using natural cover data) appeared to fluctuate over time with lowest levels in summer (July 2002) and late fall (November 2002), and more observations of salamanders during the fall and winter seasons (September 2002 and January 2003) (Figure 2.33). Mean

abundance of salamanders did not differ between sites for natural cover searches ($p=0.19$) or for both search types combined ($p=0.41$), however cover board searches showed slightly more salamanders at the fenced sites F-1 and F-2 than the other sites ($p=0.005$) (Figure 2.34). No differences amongst individual species abundances were found when combining survey methods, or from individual methods, yet *P. ruber* found by natural cover surveys, was in highest abundances at unfenced site UF-1, and this difference approached statistical significance ($p=0.06$) (Figure 2.35).

Only one species, the southeastern slimy salamander, *P. grobmani*, was not detected by both search methods; it was found only during natural cover searches (under leaf litter) and not under coverboards. At unfenced site UF-1, two species, *P. grobmani* and *D. apalachicola* were not detected at all during the entire study, and at another unfenced site, UF-3, no *D. apalachicola* were found. Of salamanders found under cover boards, most were found (56% of total captures) at boards located closest to the stream (0m), followed by boards located at 4m (24%) and 2m (20%) ($p=0.03$).

Salamander larvae captured within invertebrate collections

Forty-one southern two-lined salamander larvae, *E. cirrigera*, were captured during invertebrate collections (Table 2.6). Larval abundances were highest during the late winter and fall (February 2002/2003 and October 2002), and lowest in the summer and early winter (June, August, and December 2002) (Figure 2.36). Levels of abundance at each site differed significantly ($p<0.0001$), with highest numbers at fenced sites F-1 ($n=19$) and F-2 ($n=18$) (Figure 2.37).

Correlations between larval abundance and stream measurements made at the time of the invertebrate sampling, showed no strong relationships. Nor was a relationship present between

the abundance of invertebrates within those collections. However, the amount of organic matter present in the environment (AFDM), could play a role in the presence of larval salamanders ($r=0.40$, $p=0.05$).

Dipnetting

During May 2002 dipnetting, all salamander larvae captured were identified as *E. cirrigera*. Six individuals were captured, with none captured at sites UF-1 and F-1, three captured at UF-2, one at UF-3, and two at site F-2. Dipnetting surveys did not continue after the May 2002 survey primarily due to the disruption of streambed habitat that occurred, which could possibly have affected invertebrate collections needed for this study. It was also concluded that dipnetting yielded the same species of salamander larvae as the Hess collections, and therefore unnecessary.

Tree pipe surveys

A total of 408 individual treefrogs were captured from ten surveys conducted from June 2002-March 2003 (Table 2.6). *Hyla squirella* was the most abundant species, comprising 87% ($n=355$), followed by *H. cinerea* (11%, $n=45$) and *H. chrysoscelis* (2%, $n=8$) (Figure 2.38). All species were present at each site, except for unfenced site UF-3, at which no *H. chrysoscelis* were found.

Treefrog abundance fluctuated over time and was lowest in the summer and early winter (June-Sept 2002 and December 2002), gradually increasing in numbers to peak in the late fall (November 2002) and early spring (March 2003) (Figure 2.39). Numbers of frogs captured per date for all sites combined ranged from 2 to 107 individuals. Although no patterns amongst treatments were evident, the fenced sites showed an interesting pattern. Site F-2 had the highest

mean number of individual treefrogs, whereas the lowest abundances occurred at site F-1 ($p < 0.0001$) (Figure 2.40).

Vegetation

Eighty-five plant species were identified in the floodplain forest [(40 trees/shrubs, 18 forbs, 16 vines, 10 graminoids, and 1 unidentified moss species) (Appendix E)]. Of all species, 4 are listed as introduced or non-native, and 18% as disturbance species (Wunderlin 1998). Mean species richness was significantly higher at the fenced sites F-1 (42 species total) and F-2 (43 species), and unfenced site UF-1 (34 species) than at the other sites ($p < 0.0001$) (Figure 2.41).

Low vegetative similarity occurred between sites (Table 2.7). The greatest affinity existed between sites that were similar in treatment, with fenced sites F-1 and F-2 having the greatest similarity (46.6%) followed by fenced sites UF-2 and UF-3 (26.3%). There were some species that were unique to individual sites (UF-1 = 18 species, UF-2 = 2 species, UF-3 = 3 species, F-1 = 12 species, F-2 = 9 species). Few species were common to all sites, however the following were present within at least three of the five sites: *Acer rubrum*, *Arundinaria gigantea*, *Ligustrum sinense*, *Magnolia virginiana*, *Nyssa biflora*, *Prunus serotina*, *Quercus alba*, *Quercus nigra*, *Smilax walteri*, *Toxicodendron radicans*, and *Woodwardia areolata*. The greatest richness among plant families was seen within the Smilacaceae (7 species), and Fagaceae (5 species) families.

Magnolia virginiana was the dominant canopy species at both of the fenced sites and at unfenced site UF-3, while *Alnus serrulata* was the dominant species at site UF-1 and *Liriodendron tulipifera* at site UF-2 (Table 2.8). Mean density of canopy trees was greatest at the fenced sites and unfenced site UF-1, however site UF-1 was dominated by *A. serrulata*, a species

noted for its shrub-like, and multiple branching structures, which probably also contributed to the low basal area of 19.65 m²/ha measured at this site (Table 2.9). The highest basal area was 86.28 m²/ha, measured at unfenced site UF-2.

PCA for riparian parameter data determined that the first two axes accounted for 50% of the variation among sites, and 67% with inclusion of the third axis (Figure 2.42; third axis not shown). Four riparian parameters were positively correlated with the first axis: % open canopy (within the riparian area), % sand (ground coverage type), coarse woody debris frequency (CWD), and the volume of coarse woody debris ($r^2=0.37, 0.76, 0.37, \text{ and } 0.53$, respectively). One variable was negatively correlated with axis 1, % leaf litter cover ($r^2=0.76$). Percent coverage of the robel pole (vertical obstruction) and % total woody debris (all size classes of debris) were positively correlated with axis 2 ($r^2=0.53 \text{ and } 0.31$, respectively), whereas one variable, soil compaction, was negatively correlated with axis 2 ($r^2=0.76$). Snag frequency showed a negative correlation with axis 3, while % vegetative cover showed a positive correlation with this axis ($r^2=0.61 \text{ and } 0.33$, respectively). Fenced sites were characterized by higher percentages of leaf litter cover and lower percentages of sand coverage, open canopy, and coarse woody debris (CWD) occurrence and volume. Unfenced sites UF-2 and UF-3 were characterized by higher soil compaction and lower percent cover of woody debris, ground cover, and vertical structure. Such patterns were not consistent for one site, UF-1.

Floodplain width ranged from 10 m (site UF-1) to over 30 m (site UF-2) (Table 2.9). Most sites had steep, narrow stream terraces on at least one side of the stream, contributing to large variation in % slope. Percent canopy cover within the riparian zone was similar to the canopy cover measured over the stream channel at all sites, except site UF-1 which consistently had the lowest canopy cover ($p<0.0001$) (Table 2.9). Soil compaction (compressive strength of

the soil) within the riparian area was highest at two of the three unfenced sites UF-2 and UF-3, and lowest at both fenced sites F-1 and F-2 along with site UF-1 ($p < 0.0001$). Two variables that described the ground layer, % woody and % vegetative cover, did not significantly differ at any of the sites, however these two variables tended to be higher at a fenced site (F-2) and an unfenced site (UF-1) ($p > 0.05$). Percent cover of the ground by leaf matter was highest at the fenced sites, whereas cover by only sand was highest at two of the three unfenced sites, UF-1 and UF-2 ($p < 0.001$). Vertical structure of the understory also was highest at both fenced sites, and at site UF-1 ($p = 0.0001$).

DISCUSSION

Buffer effects and site differences

Numerous studies within North America have documented the positive effects of buffers such as streambank fencing, on water quality and aquatic biota (e.g. Lowrance et al., 1983 and 1984; Peterjohn and Correll, 1984; Jacobs and Gilliam, 1985; Owens, 1996; Line et al., 2000; Thomas, 2002; Vellidis et al., 2003). This study, located in the Gulf Coastal Plain of Georgia, also showed significant effects of buffers on physical, chemical, and vegetative measures, as well as benthic macroinvertebrate communities, suggesting that stream sites fenced over twenty years ago are recovering from any previous impacts incurred by cattle activity. However, amphibian abundance and diversity did not show similar positive responses between treatments, yet overall patterns suggest better habitat at the fenced sites. Variability amongst sites and treatments also existed, with those sites in the same treatment being most similar, and disturbances from a nearby gully strongly effecting one of the unfenced sites.

Water Quality and Physical Parameters

Contrary to studies that found increases in stream temperatures and channel width, and decreases in dissolved oxygen at sites impacted by cattle activity (Kauffman and Krueger, 1984; Maloney et al., 1998; Belsky et al., 1999; Strand and Merritt, 1999), most sites (except for UF-1) within this study were similar in those measurements. Although site UF-2, a deeper stream, showed more variability in temperature measurements. Channel morphology was extremely dynamic at site UF-1, and was filled with sediments from the gully, resulting in an aggraded and braided channel that changed with each storm event (pers obs.). Canopy cover, a major influencing factor on water temperature (Fletcher et al., 2000), did not differ amongst sites (except site UF-1) and may be reason for similar DO concentrations. The least canopy coverage occurred at UF-1 and was most likely due to the shorter height of the dominant canopy species, *Alnus serrulata*, which after 10-20 years reaches heights of only 8-12 ft (Godfrey, 1988). Although this site had lower canopy cover, it appeared to have been an adequate amount to maintain temperatures similar to other sites.

Water quality at fenced streams appeared more stable with lower peaks in response to rainfall or storms and showed lower and less variable bacterial, nutrient and sediment concentrations. Similar results were noted by Thomas (2002), who found that restricting cattle access to streams in the Georgia Piedmont resulted in decreases in nutrients (17 to 72%) and fecal coliform (95%). Elevated nutrients, turbidity, sedimentation, and greater percent sand/silt within the channel bed were also found in Coastal Plain stream sites influenced by agriculture (Davis et al., 2003). In this study it is also important to note that no FC concentrations were above the Georgia state standards for drinking and fishing (November through April), nor were nitrate concentrations above the EPA established 10mg/L standard for drinking water. Nutrient

and bacteria levels were generally lower compared to other studies examining intensively grazed areas (Barker and Sewell, 1973; Doran et al., 1981; Thomas, 2002). Stream habitat in fenced streams was also more diverse, with more wood/roots and organic matter and less sand/silt within the stream channel, providing for higher EPA habitat assessment scores.

Vegetation

A protected riparian zone in an agriculturally influenced area is widely viewed as a critical component in the protection of surface water quality, instream habitat, and the biotic integrity of aquatic ecosystems (Roth et al., 1996; Stevens and Cummins, 1999; Paine and Ribic, 2002). Detritus, a critical habitat and principal component of the food base for many streams, is produced by the terrestrial and emergent plant communities of the surrounding riparian areas (Felley, 1992). Most studies that have examined the impacts of cattle on riparian habitat have noted decreases in leaf litter accumulation, with access sites having higher levels of soil compaction and greater percentage of bare ground (Belskey et al., 1999). Similar results were found in this study, however even amongst similar treatments, vegetation structure and soil stability were shaped and affected by factors other than cattle access/exclusion. Unfenced site UF-1 showed similar high percentages of vertical cover to fenced sites F-1 and F-2. From personal observation it was apparent that cattle activity was lower at site UF-1, perhaps due to the steep slopes surrounding this area.

Soil compaction at unfenced site UF-1 was statistically similar to fenced sites, whereas high percentages of soil covered only by sand was similar to unfenced sites. High levels of soil compaction, as seen at unfenced sites UF-2 and UF-3, is a common result of cattle trampling and reduces plant productivity and plant cover (Kauffman and Krueger, 1984; Belskey et al., 1999). The soil compaction however, was not the dominating factor shaping plant structure at site UF-1.

The soil at UF-1 was predominantly moisture-laden, loose sand, resulting from soil erosion in the nearby gully. It was apparent that effects from the gully on riparian soil structure were localized, and that stable substrate at this site was available either upstream or outside the study area from the gully, where sand was not constantly washed in during flooding events. Perhaps this pattern of increased stability downstream from the gully had an effect on the riparian plant community, and thus similar percentages of vegetative cover were observed between UF-1 and fenced sites.

The effects of cattle activity on plant species richness have been inconsistent, with reports of increases, decreases, or no change (Sarr, 2002). In this study, plant species richness was similar between unfenced site UF-1 and the fenced sites, however overall similarity indices were low, with greatest similarity observed between sites that were in the same treatment. It has been suggested that in the presence of cattle disturbance, riparian diversity is often increased by invasion of upland or nonnative species (Belsky et al., 1999), with overall biodiversity including non-native and weedy species, and species with high edge affinities (Noss, 1983). Perhaps this effect may be cause for such differences in this study. At the gully site UF-1, a gradient of change was apparent as the number of herbaceous species increased further from the area of impact. Also, many of the nonnative and weedy species identified within the study were present at this site, which may explain the similarity in species richness to the fenced sites.

Macroinvertebrates

A number of variables determine the composition of aquatic invertebrate communities, one of which, stream habitat, is strongly influenced by streambed substrate and riparian vegetation (Strand and Merritt, 1999). Invertebrate communities are also influenced by regional land characteristics (i.e. geology) and by the presence of disturbances such as those resulting from grazing activities within the riparian area (Strand and Merritt, 1999). Within the Gulf

Coastal Plain, streams are typically sandy-bottomed, low to medium gradient, where one would expect to find Chironomidae as the dominant taxa (i.e. Felley, 1992; Gregory, 1996; Davis et al., 2003). The streams within this study are somewhat unusual in that cobble and coarse particles were present, typically a characteristic of more northern streams. Even so, over 70 % of taxa identified within this study were Chironomidae. Chironomids have been found in association with streams having higher levels of pollution (i.e. sedimentation), and tend to increase in abundance with disturbances such as agriculture (Kerans, 1992; Clements, 1994; Strand and Merritt, 1999; Davis et al., 2003). In this study, abundances for this taxa group were not found to be significantly different amongst treatments, nor did Chironomidae characterize unfenced sites as has been reported (Davis et al., 2003; Reed, 2003). However, diversity within the Chironomidae differed amongst sites, with highest diversity and number of sensitive chironomid taxa at the fenced sites.

The impacts of agriculture (i.e. increased sediment, nutrients, and oxygen demand) can also result in an increase of macroinvertebrate diversity by eliminating less tolerant organisms, providing for a shift to more environmentally tolerant generalists (Richards et al., 1993; Delong and Brusven, 1998). In this study, differences in richness estimates were found between sites, however higher richness of EPT and all taxa combined, occurred at the sites with the least disturbance (fenced sites) and more habitat heterogeneity. Numbers of 'unique' (and sensitive) taxa were also highest at fenced sites, which has been predicted (i.e. Lenat, 1984). Richness estimates were comparable to recent studies within the Gulf Coastal Plain (Gregory, 1996; Davis et al., 2003). However, estimates were influenced by difficulties incurred from the lack of taxonomic resolution of some taxa groups, predominantly the Ephemeroptera and Plecoptera.

Consistent with other studies evaluating the impacts of buffers using macroinvertebrates, higher percentages of more sensitive groups such as EPT, Crustacea (Decapoda and Amphipoda), and clingers were found at the fenced sites. Percentages of more tolerant taxa, the Dipterans and dominant family, were highest at the unfenced sites (except for UF-1) (Lenat, 1984; Davis et al., 2003). Higher percentages of ceratopogonidae and burrowers appeared to characterize the gully site (UF-1), which in the case of burrowers, are expected to increase with increasing disturbance (Barbour et al., 1999). One metric, % Elmidae, showed strong differences amongst treatments, with higher percentages at the fenced sites. The Elmidae (Riffle beetles), are inhabitants of the swifter portions of streams such as riffles, which have coarse sediments, high in oxygen concentration (Merritt and Cummins, 1996). Coleopterans as a group display a wide range of tolerances to disturbances (Gilbert 1989), and in a study by Lenat (1984), Elmidae were not found to be affected by agricultural runoff. However, many sensitive taxa occur within the Elmidae, such as *Microcylloepus* and *Stenelmis* which were most common in this study.

Of invertebrate composition and habit metrics examined in this study, percentages of burrowers, clingers, crustaceans (amphipods and decapods), EPT, EPT/Chironomidae, Dipterans, and Elmidae were found most useful for perennial streams within the Fall Line Hills District. Although taxonomic resolution of taxa groups within the EPT was low, richness estimates were different amongst sites, and abundances of these taxa (35 ind/m²) were much higher than studies within the Coastal Plain which found extremely low numbers (0-5 ind/m²) (Gregory, 1999; Davis et. al, 2003). Percent Elmidae, a metric developed in this study, appeared to be appropriate and captured differences in the study streams, as they were reliably collected and occurred in large enough quantities for comparison amongst sites (Rosenberg and Resh, 1993; Barbour et al., 1999).

Amphibians

Only one amphibian survey method used in this study, salamander larvae collected with a Hess sampler, revealed distinct statistical differences between buffer treatments. However, overall patterns of abundance, coupled with the presence of larvae, suggest better habitat may exist at the fenced sites. Larvae captured within stream invertebrate samples were significantly greater in abundance at the fenced sites, whereas salamander surveys within the riparian area did not show significant differences in abundance. An interesting, yet puzzling result was found from treefrog surveys, in which both highest and lowest abundances of frogs were found at the fenced sites.

Many studies have observed that both amphibian abundance and richness are lower in disturbed habitat than in undisturbed habitat (Chazal and Niewiarowski, 1998). However, some studies (e.g. Homyack and Giuliano, 2002) found no statistical differences in numbers and species of herpetofauna between fenced (1-2 years) and unfenced streams, even though fenced streams showed greater amounts of cover (i.e. vertical obstruction, litter cover), an important habitat component for amphibians (Petranka, 1998). They attributed the lack of differences in herpetofauna to similarities in water quality and the need for a longer recovery time for vegetative diversity and structure (>2-4 years). They also noted that within a year of streambank fencing a substantial herbaceous layer normally returned (Kauffman and Kruger, 1984), yet suggested that may be insufficient time for measurable improvements in water quality and thus noticeable responses by herpetofauna.

In this study, streams were fenced for over twenty years, and showed substantial differences in vegetative, chemical, and physical stream parameters as well as differences in macroinvertebrate communities. Yet, differences in salamander abundance within the riparian

zone were not apparent. Important components of salamander habitat are suitable woody debris and leaf litter (Pough et al., 1987; Petranka, 1993). Leaf litter was highest at the fenced sites in this study, but coarse woody debris showed no difference. Other microhabitat measures such as air/soil temperature, canopy cover, and leaf litter depth (with the exception of soil moisture), were not found by Hyde and Simons (2001) to explain variations in salamander abundance and distribution in their study within the Great Smoky Mountains National Park. However, large-scale habitat characteristics including disturbance history were found useful. Of those measured in this study, canopy cover and soil/air temperature were not different amongst treatments. Perhaps twenty years is long enough for the recovery of vegetative and chemical parameters, yet not long enough for the recovery of salamander populations. Petranka et al. (1993) and Pough et al. (1987) noted that recovery required about 60 years in areas disturbed by clearcutting, and Hyde and Simons (2001) also suggested that the effects of habitat disturbance (logging or agriculture) on salamander diversity and abundance may persist for more than 60 years.

Willson and Dorcas (2003) examined the effectiveness of buffer-zone systems to support stream-dwelling salamander populations in N.C., and suggested that a simple buffer zone of forested habitat was insufficient to maintain stream conditions that support high salamander abundances. Moreover, they found that salamander abundance was most closely linked to the amount of disturbed habitat within the entire watershed, and that conservation efforts must consider land use from this perspective. Perhaps a number of these factors influenced salamander abundance within this study, coupled with the need for increased survey area or trapping objects to increase capture rates for statistical analyses.

In this study, southern two-lined salamander larvae, *Eurycea cirrigera*, were the only amphibians that showed statistical differences in abundance among treatments, with higher

abundances at fenced sites. Multiple physical habitat characteristics can affect the presence and distribution of larval stream salamanders, such as stream velocity, streambed substrate, and the effects from anthropogenic stream disturbances (Welsh and Lind, 1996; Baumgartner et al., 1999; Smith and Grossman, 2003). The presence of lower sediment levels and greater heterogeneity within the streambed at fenced sites, are two factors that may help to explain these differences. Smith and Grossman (2003) examined stream microhabitat use by larval *E. cirrigera*, in Georgia Piedmont streams, and found a close association of increased larval abundance with increased substrata heterogeneity. Larvae were found to occupy stream areas with larger substratum, avoiding those with high levels of silt and embeddedness. The presence of pools at the fenced sites, which were lacking at the unfenced sites, may also have provided suitable habitat for larvae (Petranka, 1984). Welsh and Ollivier (1998) also found significantly lower abundances of amphibian individuals in streams impacted by sediment, although these effects were species specific. Sensitivity to fine sediments, as some models suggest, is a shared vulnerability of many stream amphibians, and is probably a reliance on interstitial spaces within the streambed which provide crucial habitat for cover and foraging.

The capture of treefrogs in this study also did not show statistical differences amongst treatments, in fact, fenced sites displayed both the highest and lowest abundances. A number of microhabitat variables such as tree type (hardwood or pine), DBH, distance to breeding sites, and midstory vegetation appear to influence the capture rate of treefrogs within PVC pipes (Boughton et al., 2000). These relationships also vary with treefrog species. For example, total species capture rate was found to increase with increased DBH, and significantly more captures occurred in pipes hung in hardwoods than pipes in pines (Boughton et al., 2000). Only one of twenty-five trees used in this study for treepipe surveys was a pine. Although different amongst

sites, DBH and tree type did not appear to be a major cause in differences in this study. Unfenced site UF-1 was overall significantly lower in DBH than F-1, yet had higher abundances of treefrogs. Midstory vegetation, a factor found to be strongly correlated with *H. cinerea* captures (Boughton et al., 2000), was similar between fenced sites in this study, yet overall treefrog abundance differed. Proximity to and availability of breeding areas is another factor that may explain such extremities in treefrog abundance amongst fenced sites. Little is known about the life history of wetland-breeding amphibians away from breeding sites, let alone the distance that most species in the southeast can disperse from these areas. However it is generally accepted that individuals may disperse some distance from breeding sites (2 km for *H. squirella* and *H. cinerea*; Franz, 1991) with variation among species and life stages, and in response to the availability of habitat (Dodd, 1996). Information regarding breeding sites is lacking in this study, but perhaps distance or simply the presence/absence of sites coupled with a fragmented landscape were factors resulting in abundance differences. Lastly, each hylid species has different environmental requirements, including those more or less specific to their breeding and non-breeding habitat. For example, *Hyla squirella*, common to natural and suburban areas (Zacharow et al., 2003), dominated captures in this study and was found by Zacharow et al. (2003) to be in higher abundances near forest edges. *H. squirella* is also known to breed in more temporary ponds, grassy pools and ditches, dispersing further from water than *H. cinerea*, which breeds in more permanent ponds and marshes (Goin, 1958). Although this relationship cannot be determined from the data, it is apparent that at the landscape-level, this study area is highly fragmented, and associated factors such as edge effect and isolation, may act as barriers to migration (Dodd and Smith, 2003).

Site differences

Weigel et al. (2000) noted that when comparing multiple streams it is essential to account for inherent stream variability. In Sarr's (2002) review of livestock exclosures in the western U.S., he states that upon exclusion, riparian areas may show different recovery paths depending on the level of past disturbances. An interesting aspect of my study arises from the fact that sites did indeed show variability, where in some cases, stream sites reacted or rather displayed similar and dissimilar effects to buffering treatments. Variability could be attributed to the geographic location of each site and the surrounding past and present land uses. For example, fenced sites F-1 and F-2 have a history of disturbance from logging and cattle activity. Even though these sites are fenced and a buffer does exist, they are down slope from grazing and crop fields (Figure 2.4), which may be cause for some similarities to unfenced sites. Unfenced site UF-1 also has a history of disturbance, with cattle intrusion as one impacting factor, but major alterations seemed to apparently originate from an upland eroding gully. Yet in some cases, this site appeared similar to the fenced sites (i.e. vegetative measures). Lastly, unfenced sites UF-2 and UF-3 were very similar in most measurements, reflecting similar patterns of adjacent and upstream land use.

Biological monitoring and the use of amphibians

As aquatic ecosystems continue to suffer from land use disturbances such as agriculture, the need to track the condition of such systems increases (Fore et al., 1996). Amphibian monitoring is important because threats to their populations are two-fold within an agricultural landscape, including alterations of habitat crucial to hibernation and migration to and from breeding sites, while also facing the threat of pollution from agrochemicals such as pesticides and fertilizers (Beebee, 1996). Understanding the changes that occur to aquatic biota (such as amphibians) in relation to disturbance, forms the basis of many biomonitoring methodologies

used in aquatic ecosystems (Cao et al., 1997). Resident aquatic biota such as macroinvertebrates and fish have been widely adopted to evaluate water quality because they are considered good indicators of localized disturbances, revealing and expressing the consequences of human activities on an aquatic system (Lenat et al., 1980; Abel, 1989; Barbour et al., 1999).

Amphibians exhibit traits of a good indicator, and their inclusion in monitoring and conservation programs has been suggested in many studies (i.e. Welsh and Ollivier, 1998; Rocco, 1999; Boughton, 2000; Micacchion, 2002; Semlitsch, 2003). However, efforts to establish such programs have been hindered due to the lack of long-term data, uncertainties associated with their varied and complex life histories, their detectability, stability, and fluctuation of populations, species-specific habitat requirements, occurrence in the landscape (home ranges), and a poor understanding of sampling technique accuracy (Hyde, 2001; Dodd, 2003). A number of biases have been associated with amphibian monitoring methods such as species distribution, sampling methods (pipe diameter or board length/width), location, residency, seasonal activity, and response to environmental conditions such as disturbance (Zacharow et al., 2003). In this study, it was difficult to determine a specific reason why most salamander and treefrog survey techniques did not show differences amongst sites. Overall observations of salamanders were low, but perhaps if a larger number of survey objects were used, or a larger area searched, differences would have been detected. However, one of the most crucial elements to understanding and deciphering the impact of disturbance on amphibian populations is the knowledge of individual species life history traits including habitat associations. Although the use of similar survey methods has been successful in other studies, each amphibian species may yet have different requirements for using treepipes or coverboards. Further life history information is needed for each amphibian species and lifestage, from variables at the

microhabitat to macro-landscape levels, including response and tolerance levels to disturbance (i.e. pollution), distance to nearest breeding areas, and home ranges.

Amphibians are ‘sensitive species,’ and demonstrate a strong potential for use in biomonitoring programs. Ultimately, a metric or tool used in biological monitoring programs is chosen because it reflects specific and predictable responses of organisms to changes in the environment. Perhaps *Eurycea cirrigera* (larvae) and *Hyla squirella* show promise as ‘indicator species,’ for they were found consistently at both fenced and unfenced sites and in high enough numbers for comparison. *H. squirella* has also been found in high abundances within other studies (Goin, 1958; Boughton et al., 2000; Zacharow et al., 2003). The Hess sampler did detect differences in larval salamander abundance among sites and shows promise as a metric and tool for amphibian biological monitoring. Captures were rapid and quantifiable with no sampler biases, and enough individuals were captured to detect significant differences.

It is suggested that the most effective approach to monitoring aquatic health is one that is synthetic, adopting a group of relevant metrics, while recognizing the need for approaches that consider the ‘many attributes of biological condition simultaneously’ (Karr and Chu, 1999). The inclusion of both macroinvertebrates and amphibians in biological assessment would concurrently provide for a more complete and strengthened assessment of a site’s condition. It would also offer a broader spectrum of responses, as both groups are highly sensitive to disturbances, and are linked to aquatic and terrestrial environments for their life history stages. If we seriously begin to acquire the missing information needed to incorporate amphibians into monitoring programs, another biological ‘tool’ would be available, especially in the case where other species, such as fish, may be poor or absent.

CONCLUSIONS

Differences in measurements (chemical, physical, biotic) between buffered (streams fenced from cattle) and unbuffered sites were apparent in this study, suggesting that sites fenced from cattle many years ago have better water quality and habitat, and that conservation buffers are benefiting these streams. Variability amongst sites and treatments also existed, with those sites in the same treatment being most similar, and disturbances from a nearby gully strongly affecting one of the unfenced sites. Rapid assessment tools for soil coverage and vertical obstruction were found useful in detecting differences amongst sites, and should be considered for monitoring programs. Ground layer coverage type, estimated across each plot using a line-transect method for soil coverage appeared to capture similar differences amongst treatments than conventional plot surveys methods. The vertical cover estimation technique incorporated in our study using a robel pole, proved to be a useful and rapid tool for demonstrating vegetative differences amongst sites.

Of invertebrate metrics examined in this study, percentages of burrowers, clingers, crustaceans (amphipods and decapods), EPT, EPT/Chironomidae, Dipterans, and Elmidae were found most useful for perennial streams within the Fall Line Hills District of Georgia. Amphibian abundance and diversity did not show similar positive responses between treatments for surveys within the riparian area, however salamander larvae collected within invertebrate collections did display significant differences. A number of factors could play a role the abundance of amphibians, ranging from those controlled by the study design, such as small sample size or survey area, to variables beyond our control such as distance to nearest breeding site (for hylids) and the degree of disturbance these sites incurred previous to this study. Salamander larvae captured with the Hess sampler were most likely influenced by lower

sediment levels and increased streambed heterogeneity at the fenced sites. Larval captures with a Hess sampler proved useful as a metric and tool for assessment of site differences, and should be used in correlation with other biotic indicators such as benthic macroinvertebrates. Amphibians appear as useful tools in biological monitoring, however a consortium of life history traits and responses to disturbance variables ranging from the microhabitat to landscape levels needs to be further examined.

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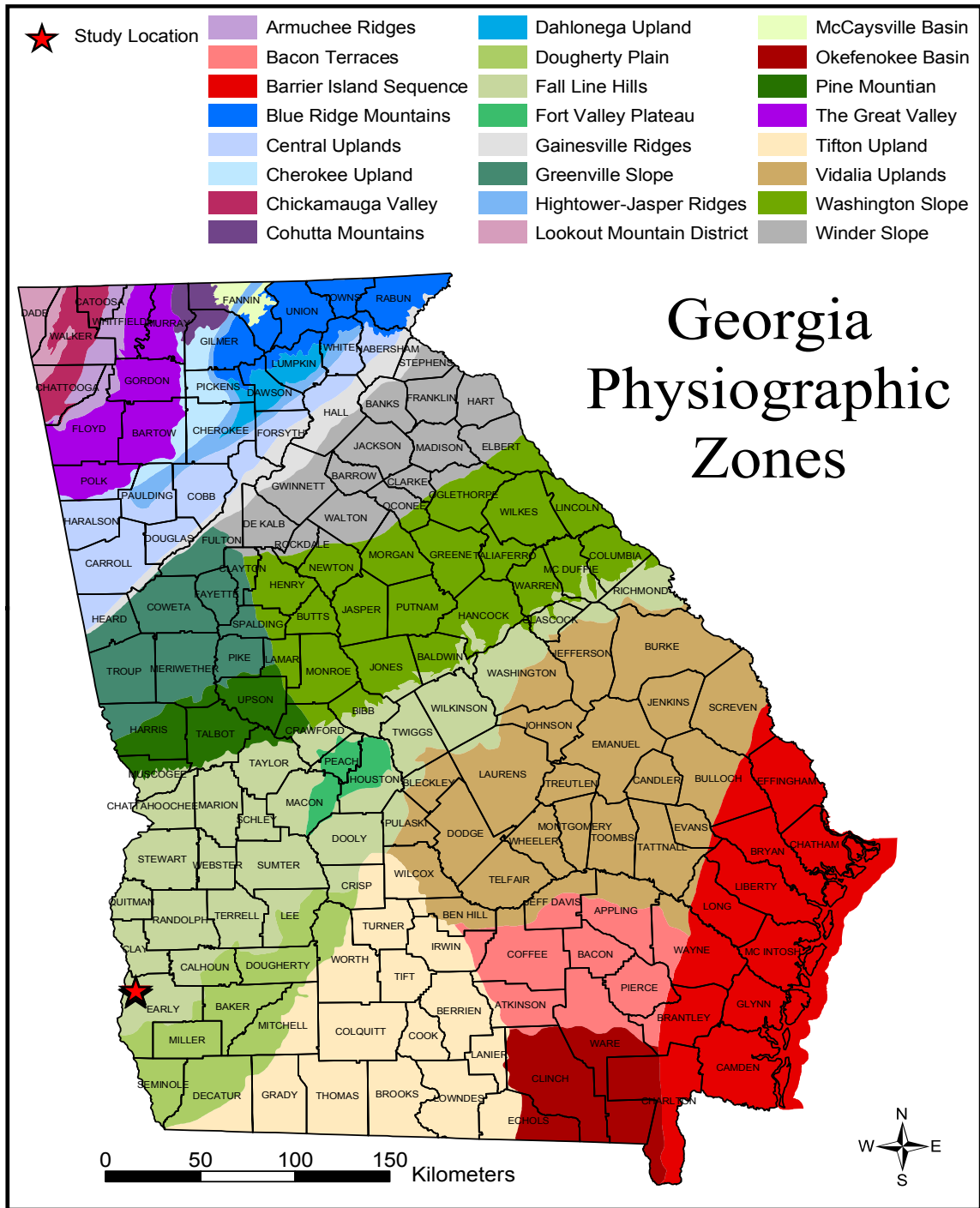


Figure 2.1. The Physiographic Zones of Georgia. Note the Fall Line Hills District (FLH) and the study site location in Early County, Georgia.

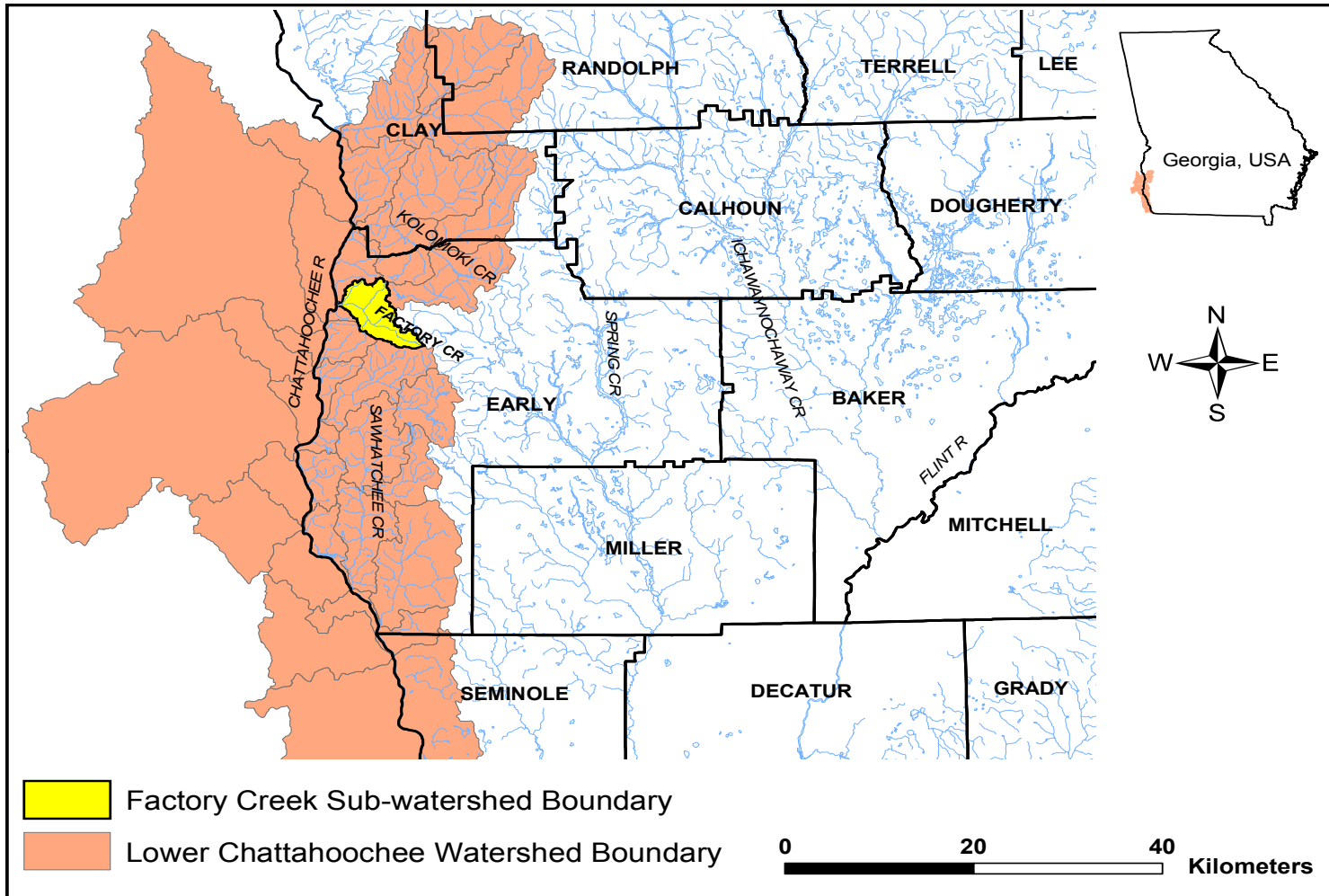


Figure 2.2. Study area location and watershed boundaries.

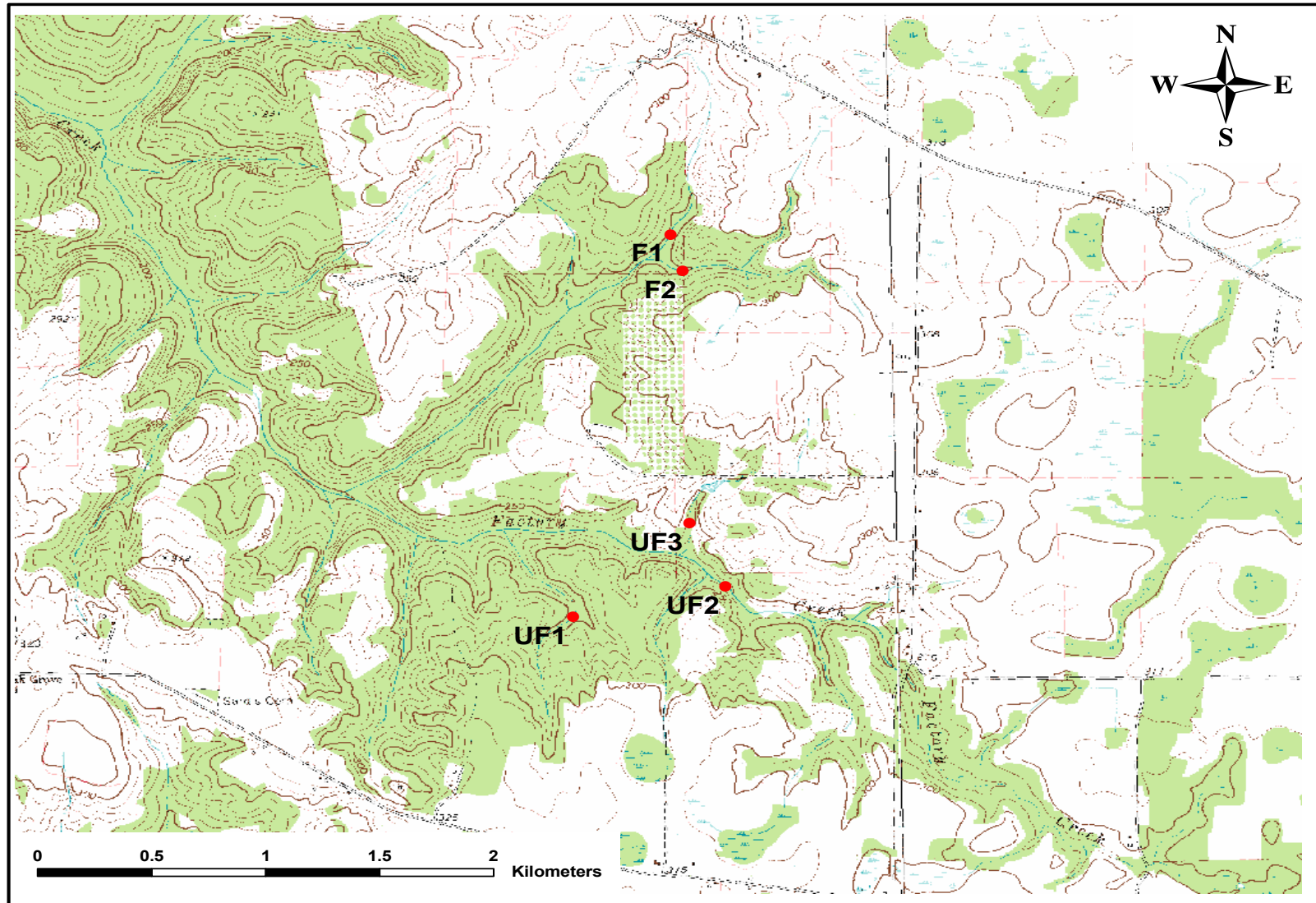


Figure 2.3. Sampling site location. Sites UF-1, UF-2, and UF-3 allow cattle access to stream. Sites F-1 and F-2 are fenced from cattle access (USGS DRG 1:24000 topographic Quadrangle; 1970 and 1973).

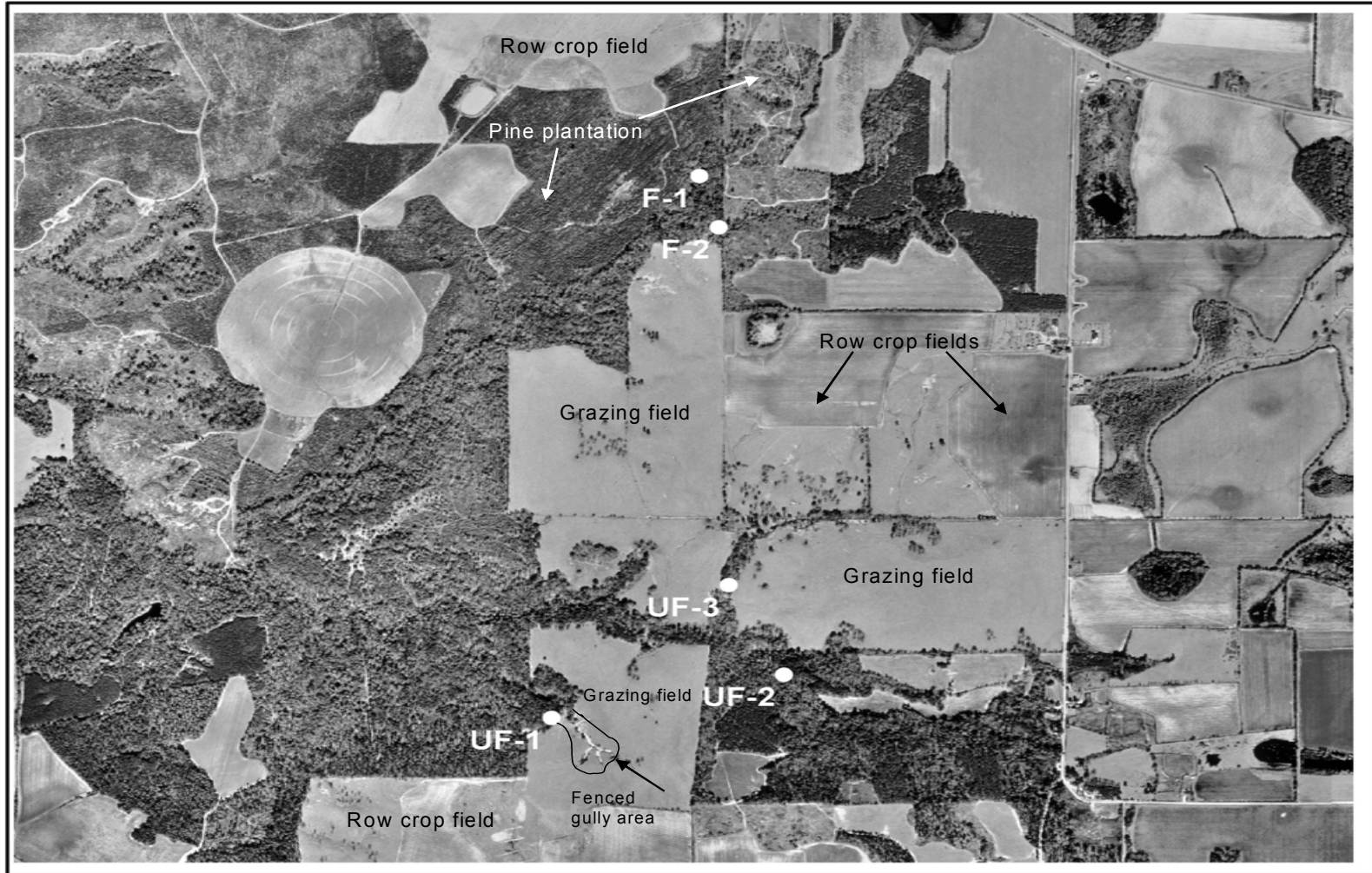


Figure 2.4. Land-use on Brownlee farm surrounding study sites, Early County, Georgia. (NAPP geo-rectified photo, GA DNR- EPD; 1999).



Figure 2.5. View of unfenced site UF-1. Note that this stream is located near a heavily eroding gully. Photo taken after rain event, March 2003.



Figure 2.6. View of unfenced site UF-2. Photo taken in winter 2003



Figure 2.7. View of unfenced site UF-3. Photo taken in summer 2002



Figure 2.8. View of fenced site F-1. Photo taken in winter 2002



Figure 2.9. View of fenced site F-2. Photo taken in summer 2002

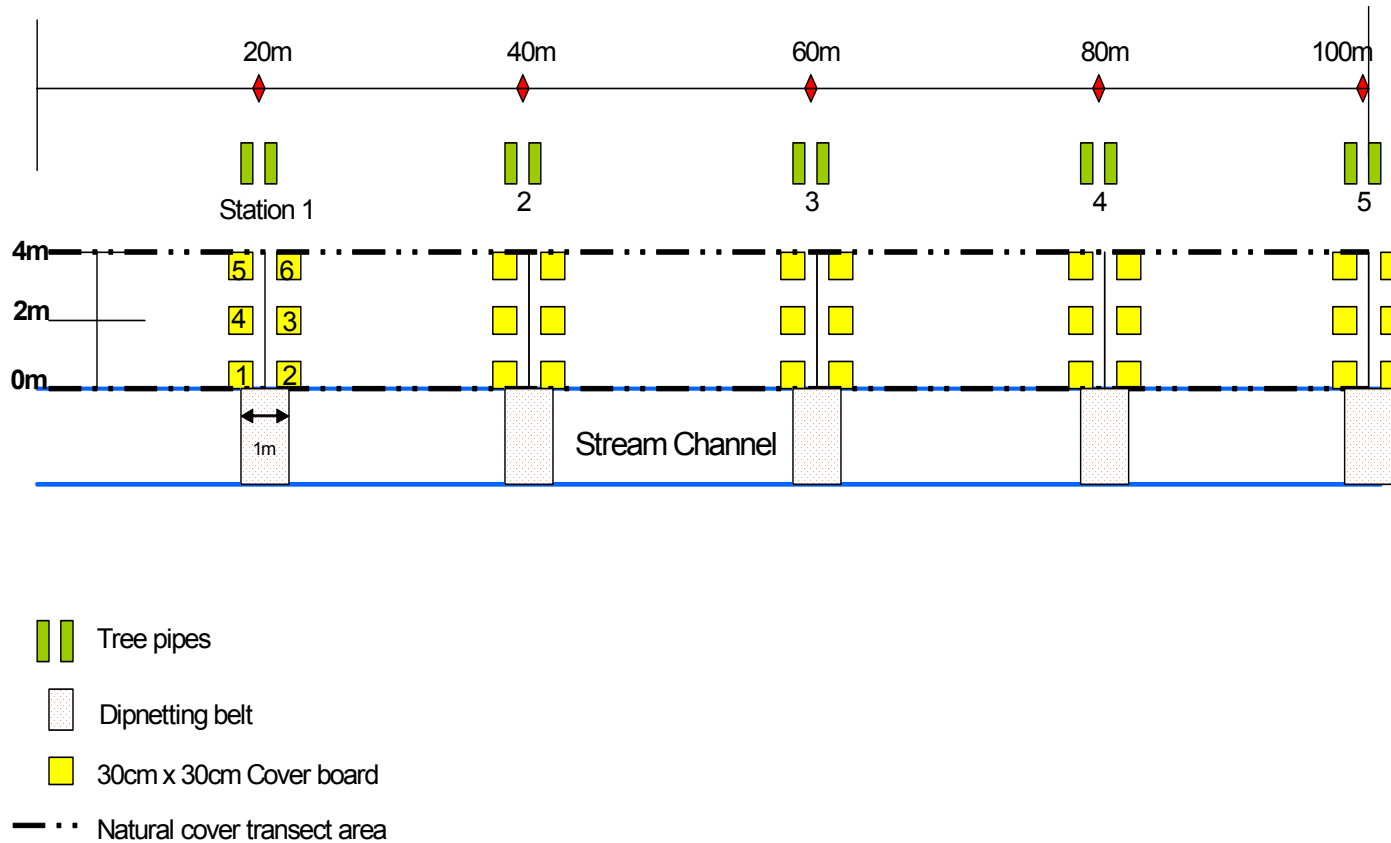


Figure 2.10. Amphibian sampling framework at each site. A natural cover transect, an artificial cover board transect, dipnetting belts, and tree pipes. The cover board transects consisted of 5 stations each, spaced 20 m apart, with 6 cover boards at each station. Dipnetting belts (5), 1 m in width, were spaced 20 m apart. One pair of tree pipes was placed on the same tree at each station, one facing north and one facing south for a total of 10 pipes at each site.

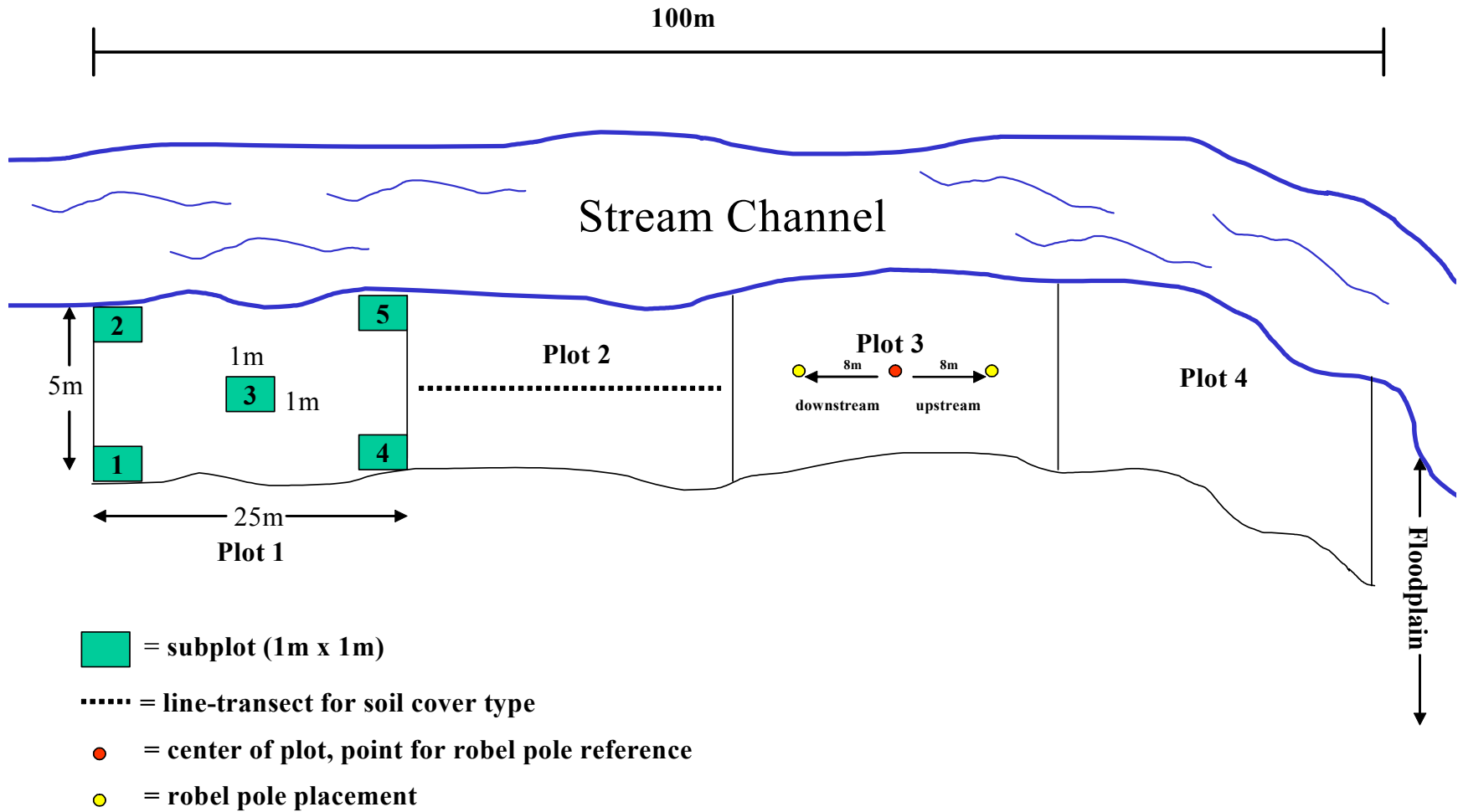


Figure 2.11. Plot layout for vegetation survey. Methods consisted of five (5) 1 m x 1 m sub-plots within each of the four (4) 5 m x 25 m plots, located on one side of the stream site (the same side as amphibian surveys). Total survey reach was 100-m in length.

Table 2.1. Expected responses of invertebrate metrics to increased stress. % Elmidae was designed for this study and the remaining metrics have been found useful in other studies.

Metric	Increasing Stress
EPT abundance	Decrease
No. Families	Decrease
% Amphipoda	Decrease
% burrowers	Increase
% Chironomidae	Increase
% clingers	Decrease
% Crustacea	Variable
% Diptera	Increase
% dominant family	Increase
% Ephemeroptera	Decrease
% EPT/ Chironomidae	Decrease
% non Diptera	Decrease
% Plecoptera	Decrease
% Trichoptera	Decrease
% Elmidae	Decrease

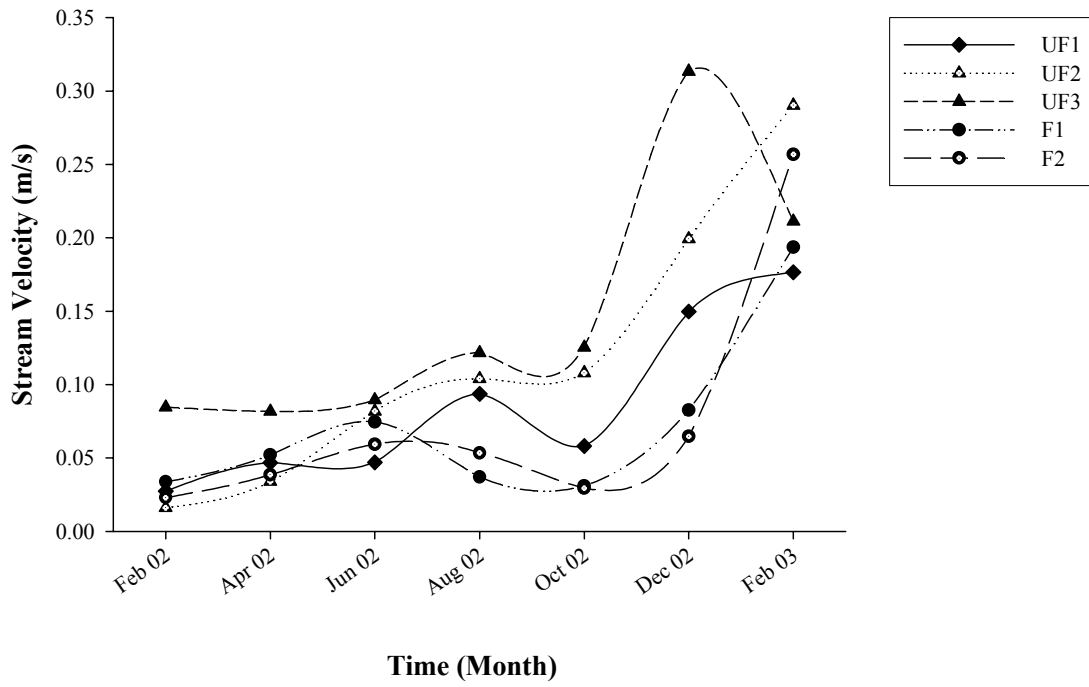


Figure 2.12. Mean bimonthly measurements of flow velocity (m/s) at each site from February 2002 to February 2003.

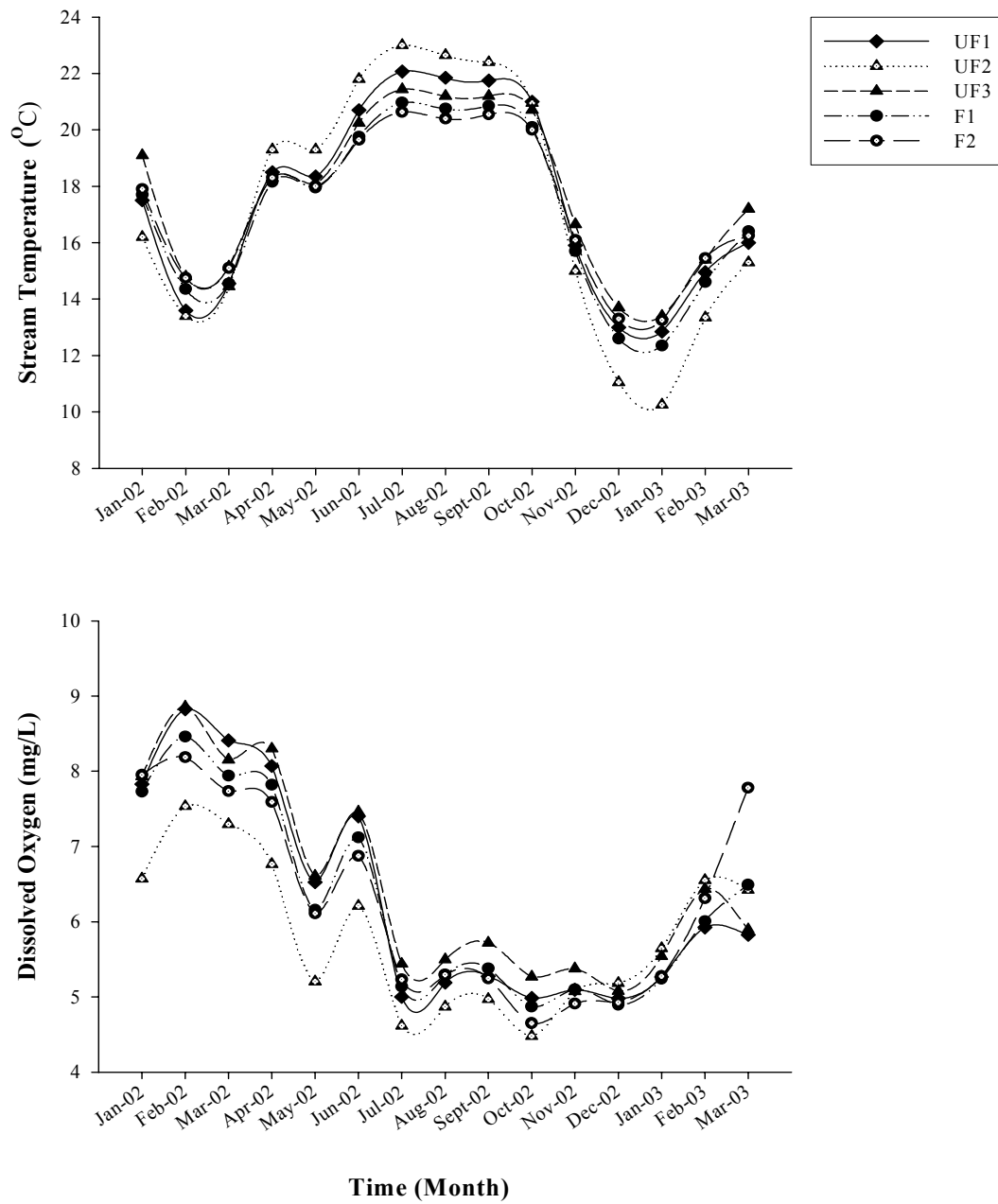


Figure 2.13. Mean monthly measurements of stream temperature and dissolved oxygen from January 2002 to March 2003 at all sites.

Table 2.2 Mean, minimum, and maximum physical stream measurements for all sites, January 2002 – March 2003. For each site and parameter, values with different letters are significantly different (Kruskal-Wallis test with respective p-value).

Parameter	Mean (Min. – Max.)					p-value
	UF1	UF2	UF3	F1	F2	
Width (m)	2.1cb (0.7 - 5.7)	2.9a (1.7 - 4.5)	1.7c (1.2 - 2.4)	2.2b (1.3 - 3.4)	1.9cb (1.5 - 2.5)	<0.0001
Depth (cm)	1.76d (0.29 - 3.95)	11.79a (7.09 - 24.41)	4.44c (2.63 - 8.18)	7.71b (4.59 - 6.58)	6.37b (1.81 - 11.9)	<0.0001
Width: Depth (m)	61a	7 c	10cb	12b	11cb	<0.0001
Stream Temperature (°C)	19.23a (13.5 - 23.3)	17.83a (12.1 - 23.0)	18.93a (14.4 - 22.9)	17.87a (13.0 -21.5)	18.73a (15.2 -21.6)	0.8984
Dissolved oxygen (mg/L)	7.04a (4.35 - 8.87)	6.55a (4.50 - 8.09)	7.08a (5.28 - 9.22)	6.59a (5.11 - 8.48)	6.99a (5.26 - 8.70)	0.5407
Velocity (m/s)	0.09ab (0.01 - 0.27)	0.12ab (0 - 0.34)	0.15a (0.05 - 0.35)	0.07b (0.002 - 0.28)	0.07b (0.01 - 0.35)	0.0011
Sand/silt (%)	68.16a (0 - 100)	56.88ab (0 - 93.3)	57.16ab (0 - 100)	42.50b (0 - 81.8)	53.37ab (29.3 - 84.2)	0.0130
Leaves (%)	9.76b (0 - 36.4)	27.51a (0 - 100)	14.60ab (0 - 55.9)	15.16ab (0 - 47.8)	17.24ab (2.6 - 37.96)	0.0436
Wood/roots (%)	0.41d (0 - 5.7)	11.0bc (0 - 40)	4.92cd (0 - 23.5)	25.36a (0 - 70)	18.29ab (0 -51.4)	<0.0001
Algae (%)	0.49a (0 - 10.3)	0a 0	0a 0	0.13a (0 - 2.8)	0.39a (0 - 8.1)	0.7356
Pebbles (%)	5.95c (0 - 50)	6.30c (0 - 30.3)	21.71a (0 - 82.86)	15.73ab (0 - 54.2)	10.88abc (0 - 28.1)	0.0004
Exposed streambed (%)	15.06a (0 - 62.5)	0.20b (0 - 4.1)	2.94b (0 - 18.18)	1.12b (0 - 12)	0.28b (0 - 5.9)	<0.0001
Canopy opening (over stream) (%)	26a (5-52)	6b (2-23)	8b (3-20)	6b (1-17)	8b (0-27)	<0.0001
AFDM (kg/m ²)	0.071c (0.002-0.204)	0.473a (0.051-1.327)	0.152bc (0.01-0.736)	0.199bc (0.031-0.896)	0.244ab (0.012-1.048)	<0.0001

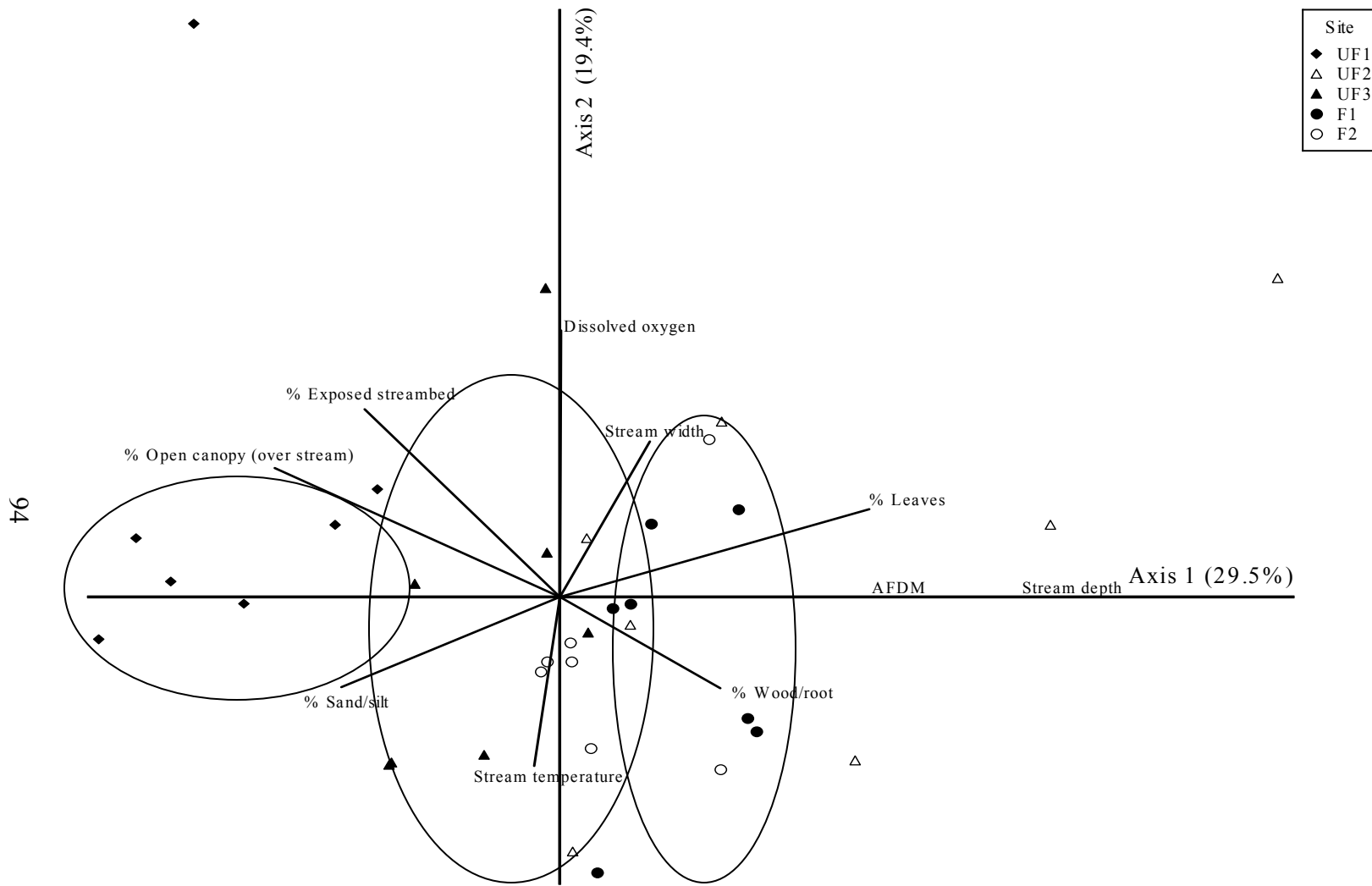


Figure 2.14. First and second axes of the principal components analysis (PCA) for physical stream parameters at all sites. Points represent individual site/dates and vectors indicate physical stream variables. Percent variation is explained in parentheses on each axis.

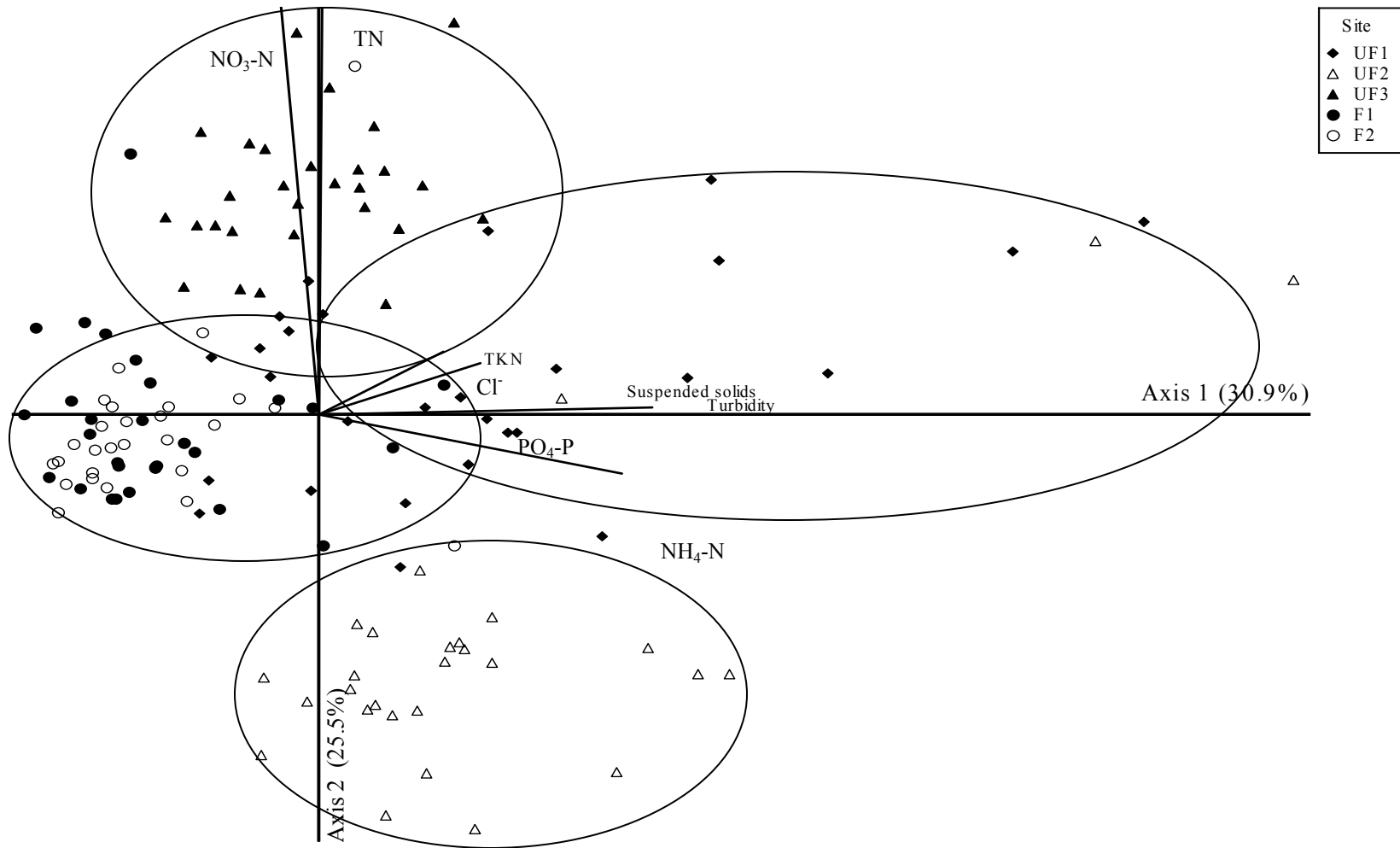


Figure 2.15. First and second axes of the principal components analysis (PCA) for stream water quality parameters at all sites. Points represent individual site/dates and vectors indicate water quality variables. Percent variation is explained in parentheses on each axis.

Table 2.3. Water Quality Parameters. Mean, minimum, and maximum chemical measurements for study sites, February 2002 to March 2003.(Kruskal-Wallis test with respective p-value).

Parameter	Mean (Min. - Max.)					p-value
	UF1	UF2	UF3	F1	F2	
Turbidity (nm)	78.50ab (21.0 – 392.5)	88.97a (16.5 – 369)	42.93b (3.0 – 100.5)	29.19c (0 – 234.5)	17.91c (0 – 102)	<0.0001
Suspended Solids (mg/L)	5.90a (1.42 – 27.08)	3.45a (0.43 – 11.51)	2.98a (0.20 – 7.14)	0.81b (0 – 5.21)	0.77b (0 – 3.67)	<0.0001
NO ₃ -N (mg/L)	0.57b (0.46 – 0.70)	0.24c (0.17 – 0.42)	0.80a (0.71 – 0.97)	0.57b (0.50 – 0.80)	0.56b (0.29 – 0.67)	<0.0001
PO ₄ -P (mg/L)	0.03a (0 – 0.08)	0.02ab (0 – 0.06)	0.01bc (0 – 0.07)	0.01c (0 – 0.05)	0.01c (0 – 0.05)	<0.0001
NH ₄ -N (mg/L)	0.05a (0.02 – 0.11)	0.06a (0.03 – 0.18)	0.03b (0.01 – 0.09)	0.02c (0.01 – 0.007)	0.02c (0 – 0.06)	<0.0001
TKN (mg/L)	0.23a (0.004 – 0.70)	0.21ab (0 – 0.56)	0.15abc (0 – 0.36)	0.14bc (0 – 0.30)	0.13c (0 – 0.75)	0.0005
Total-N (mg/L)	0.81b (0.52 – 1.21)	0.45d (0.19 – 0.92)	0.95a (0.79 – 1.13)	0.71c (0.52 – 1.07)	0.69c (0.52 – 1.32)	<0.0001
Cl ⁻ (mg/L)	3.51c (3.09 – 4.51)	3.95b (3.48 – 5.21)	4.27a (3.83 – 5.33)	3.49c (3.10 – 4.30)	3.59c (3.24 – 4.40)	<0.0001
f.coliform col/100 mL	281 (10 – 1100)	418 (60 – 1500)	532 (120 – 1800)	156 (28 – 760)	237 (23 – 3200)	N/A
f.streptococci col/100mL	671 (16 – 2300)	1452 (100 – 12,000)	1593 (17 – 10,000)	1222 (40 – 13,000)	631 (13 – 5,500)	N/A
†Ph	5.06	5.32	5.12	5.03	5.05	N/A
†Alkalinity	8.60	9.11	5.45	3.81	4.75	N/A

†denotes measurements taken once during the 2003 sampling season

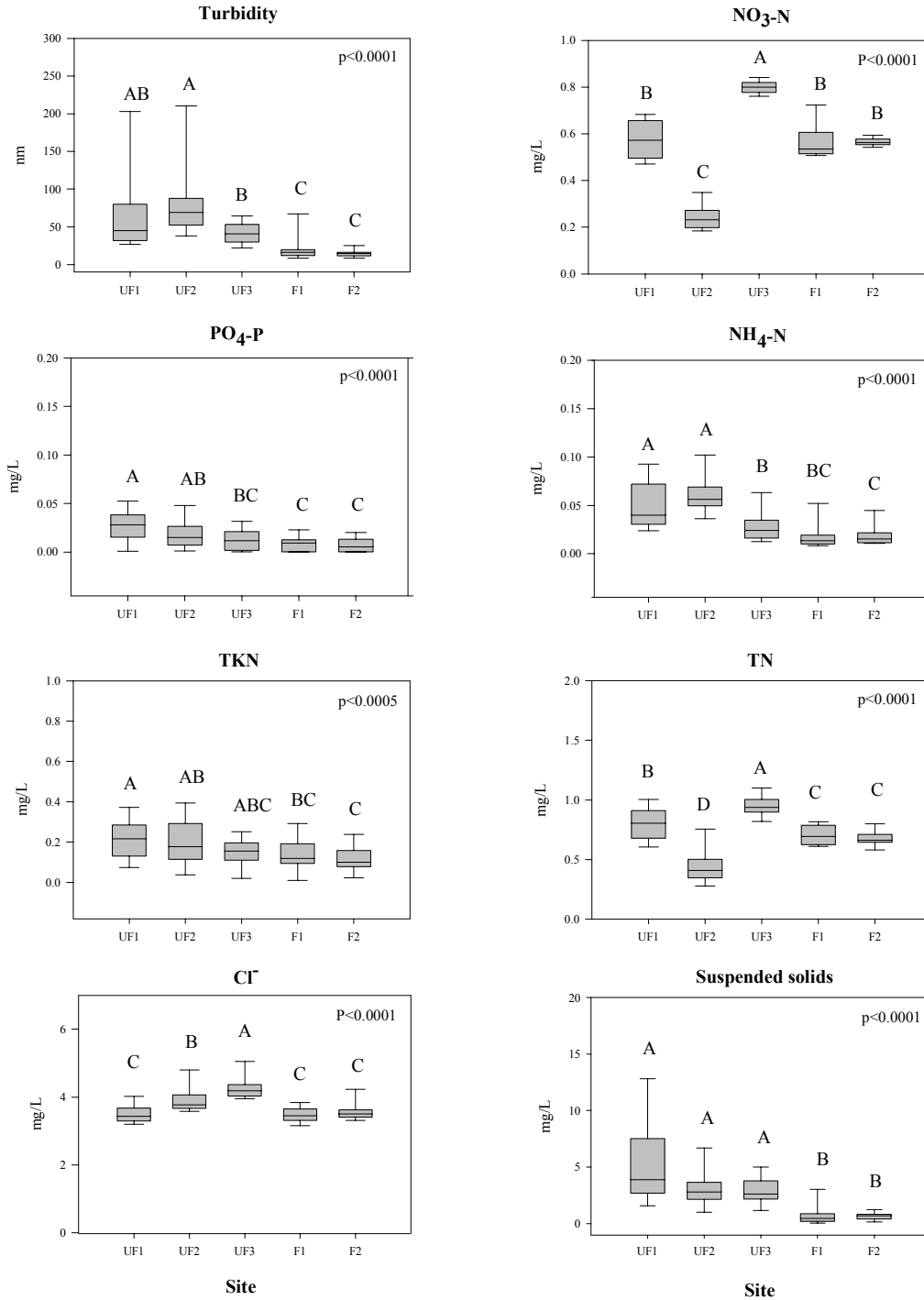


Figure 2.16. Box plots of water quality parameters for all sites (median, 25th and 75th percentiles, maximum value, minimum value and outliers). For each stream and parameter, values with different letters are significantly different (Kruskal-Wallis One Way ANOVA on ranks test with respective p-value).

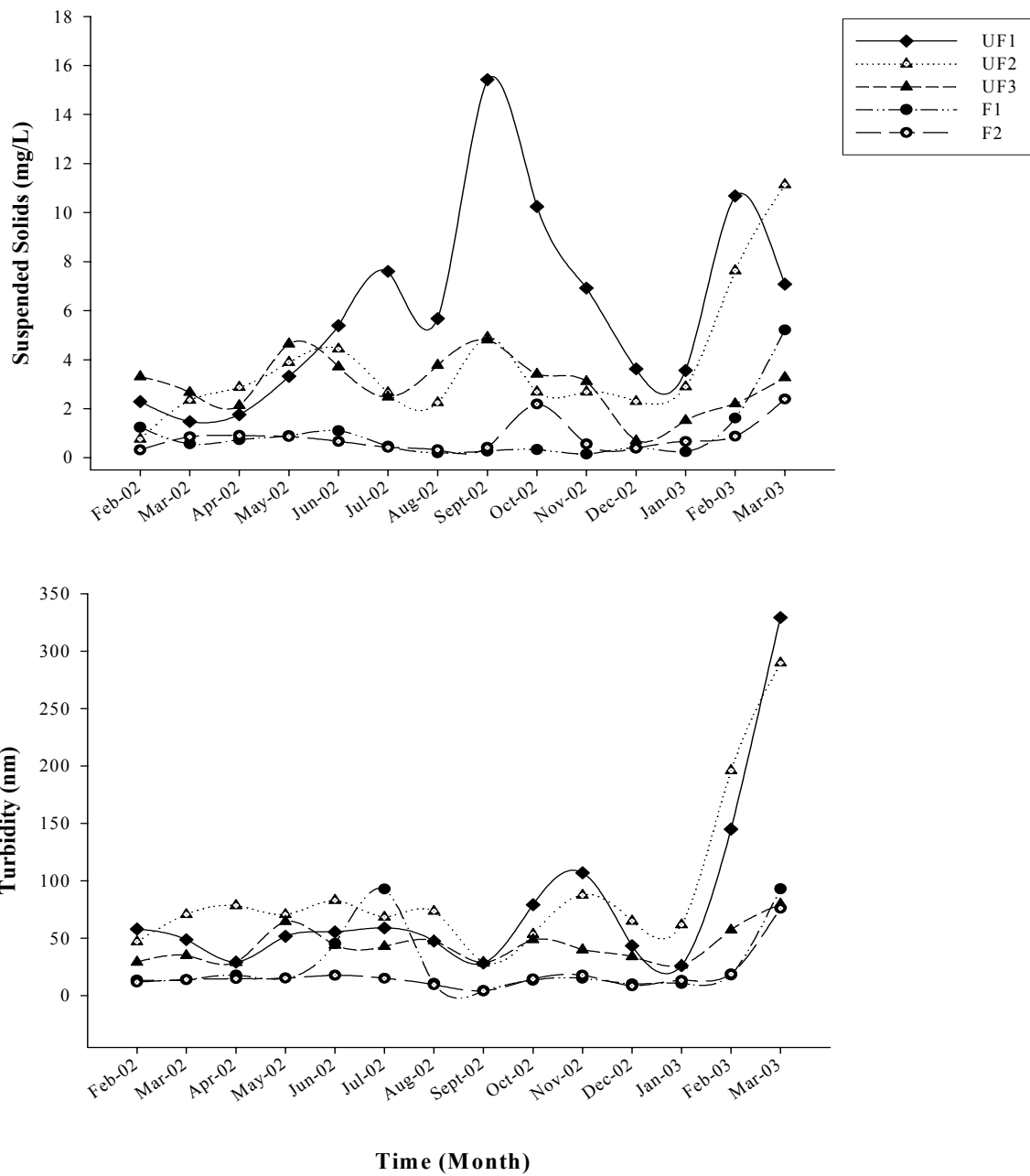


Figure 2.17. Monthly trends of suspended solids and turbidity at each site from February 2002 to March 2003.

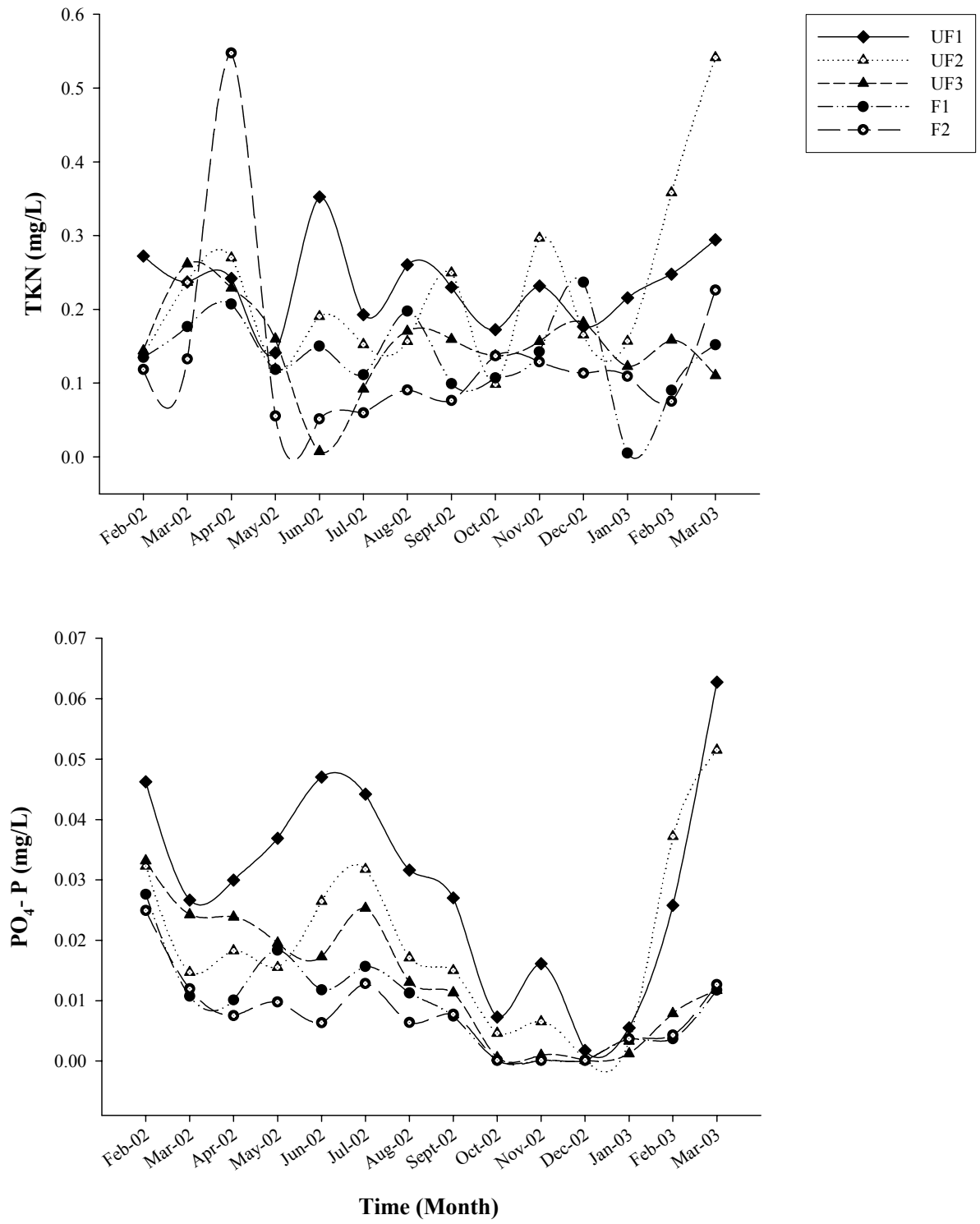


Figure 2.18. Monthly trends of TKN and phosphate at each site from February 2002 to March 2003.

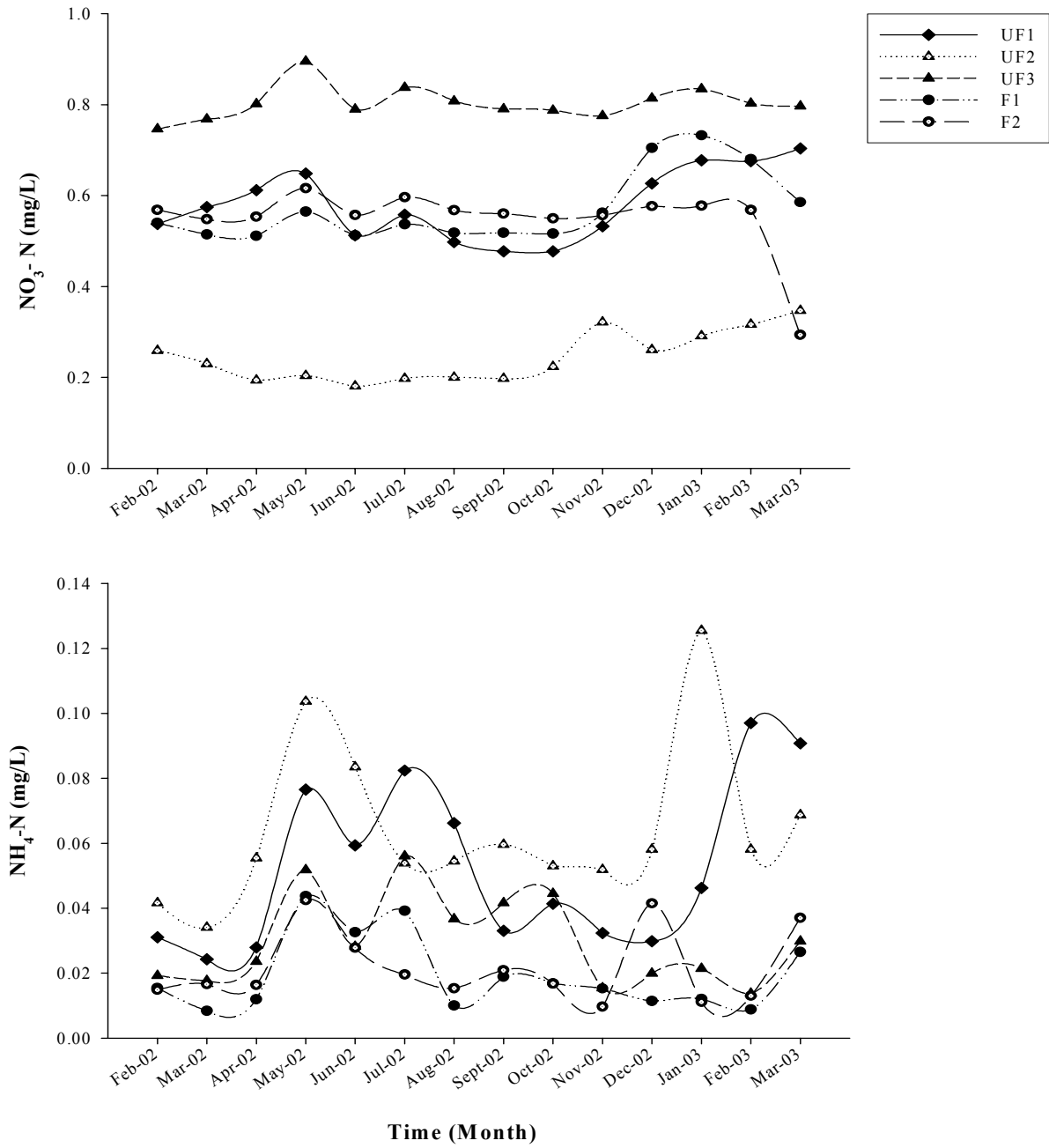


Figure 2.19. Monthly trends of nitrate and ammonium at each site from February 2002 to March 2003.

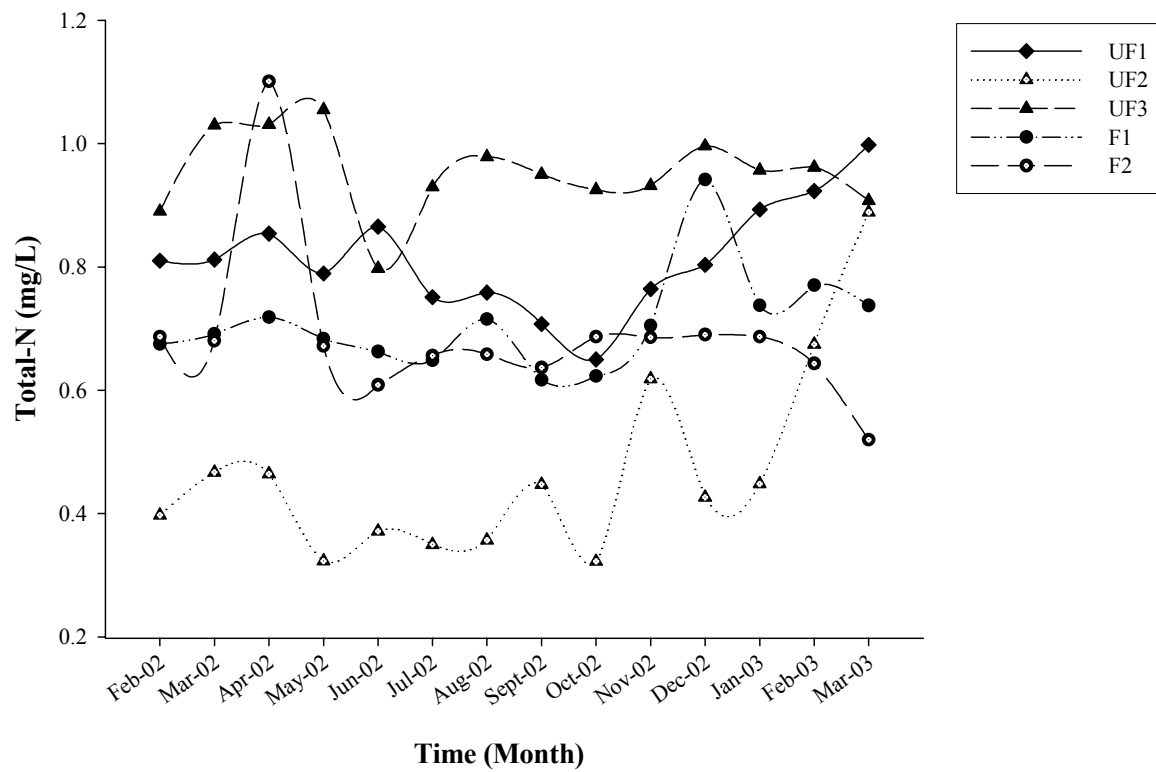


Figure 2.20. Monthly trends of Total-N at each site from February 2002 to March 2003.

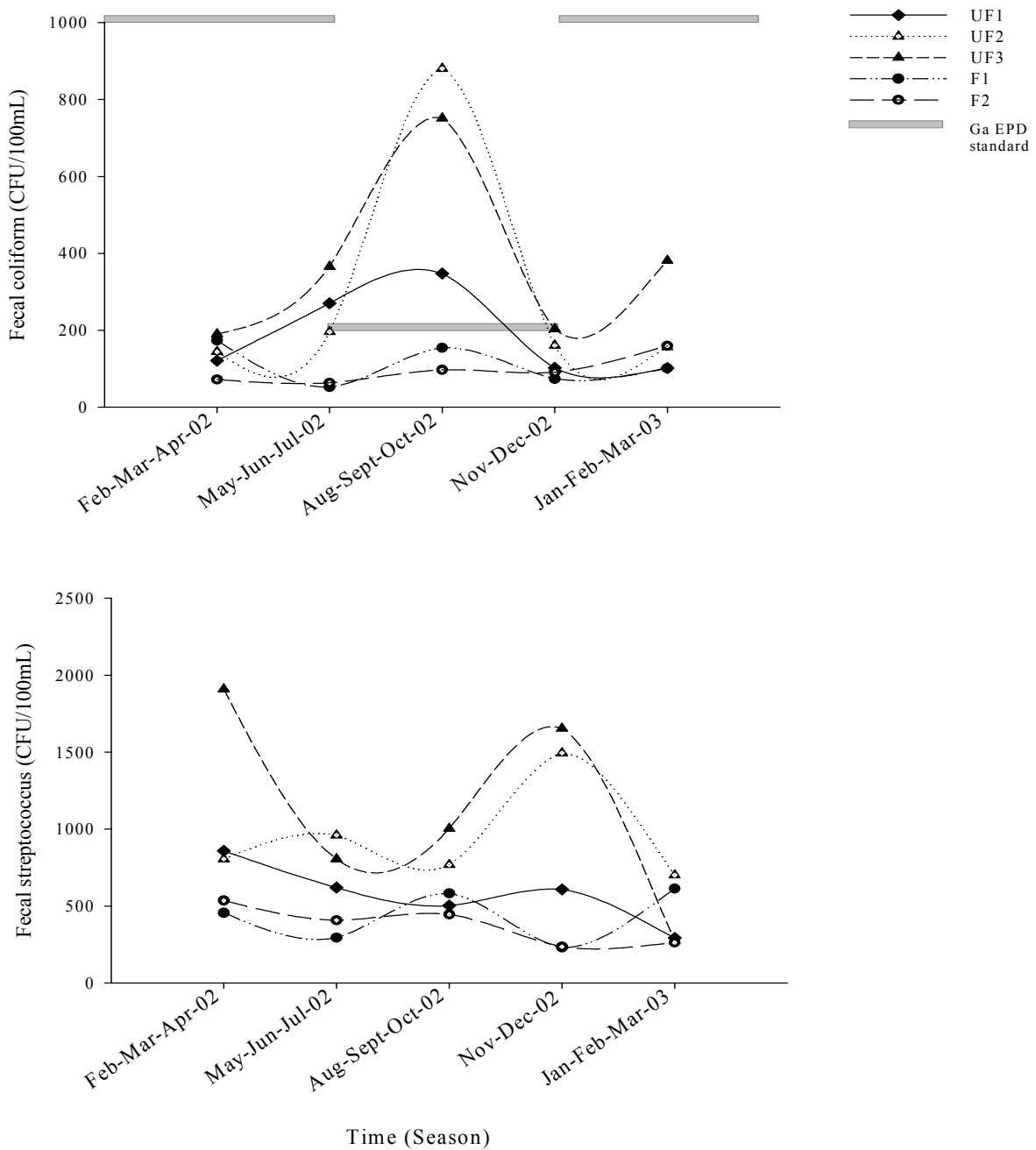


Figure 2.21. Mean seasonal fecal coliform and fecal streptococci levels at each site from February 2002 to March 2003 (CFU/100mL). Barred line indicates the Georgia state water quality standards for all waters during the recreational season (200 CFU/100mL; May-October) and for waters designated for drinking and fishing (1000 CFU/100mL; November-April) (GaDNR, 2000) for fecal coliform concentrations.

Table 2.4. Geometric means of fecal coliform (A) and fecal streptococcus (B) levels (CFU/100mL) by season, February 2002 to March 2003.

A.	UF1	UF2	UF3	F1	F2
Feb/Mar/Apr 2002	121	144	190	173	71
May/Jun/Jul 2002	270	196	366	52	63
Aug/Sept/Oct 2002	347	880	751	154	97
Nov/Dec 2002	102	161	204	74	91
Jan/Feb/Mar 2003	102	157	381	101	159
Average:	189	608	378	111	96

B.	UF1	UF2	UF3	F1	F2
Feb/Mar/Apr 2002	619	960	806	294	407
May/Jun/Jul 2002	502	767	1002	581	445
Aug/Sept/Oct 2002	607	1494	1654	230	235
Nov/Dec 2002	293	700	273	612	262
Jan/Feb/Mar 2003	94	226	411	303	308
Average:	423	830	830	404	331

Table 2.5. Mean fecal coliform (FC) to fecal streptococcus (FS) concentration ratios by season at all sites, from February 2002 to March 2003. FC/FS>4: human contamination; 0.1<FC/FS<0.7: livestock; FC/FS<0.1: wildlife; (Geldrich et.al., 1968).

	UF1	UF2	UF3	F1	F2
Feb/Mar/Apr 2002	0.20	0.15	0.24	0.59	0.18
May/Jun/Jul 2002	0.54	0.26	0.36	0.09	0.14
Aug/Sept/Oct 2002	0.57	0.59	0.45	0.67	0.41
Nov/Dec 2002	0.35	0.23	0.75	0.12	0.35
Jan/Feb/Mar 2003	1.09	0.70	0.93	0.33	0.52
Average:	0.55	0.38	0.55	0.36	0.32

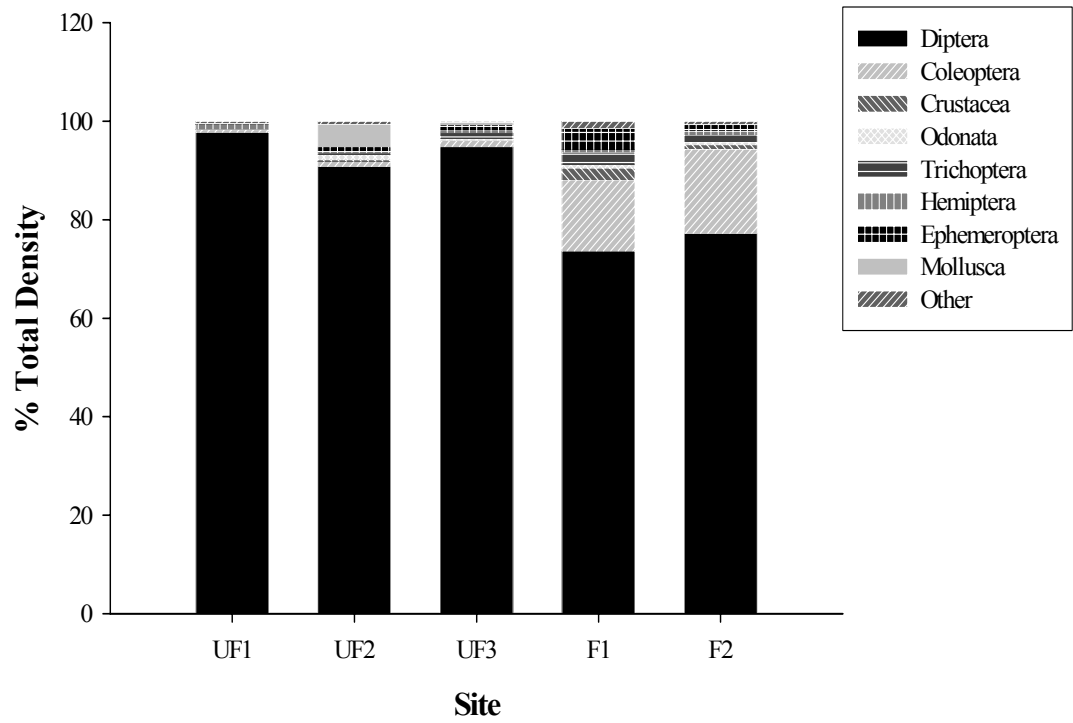


Figure 2.22. Percent of total density for each invertebrate group for all dates combined at all study sites.

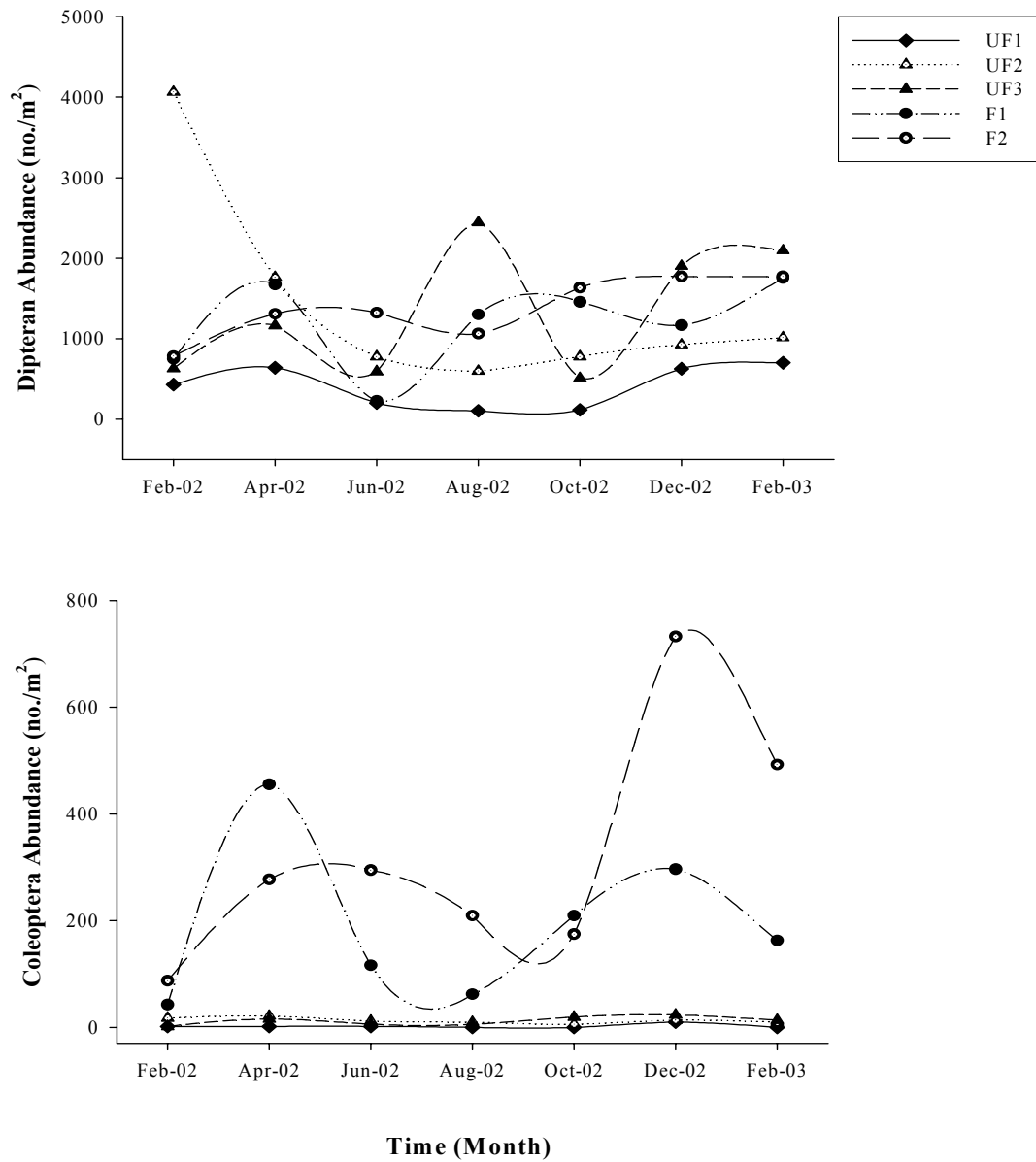


Figure 2.23. Diptera and Coleoptera abundance (no./m²) at each site from February 2002 to February 2003.

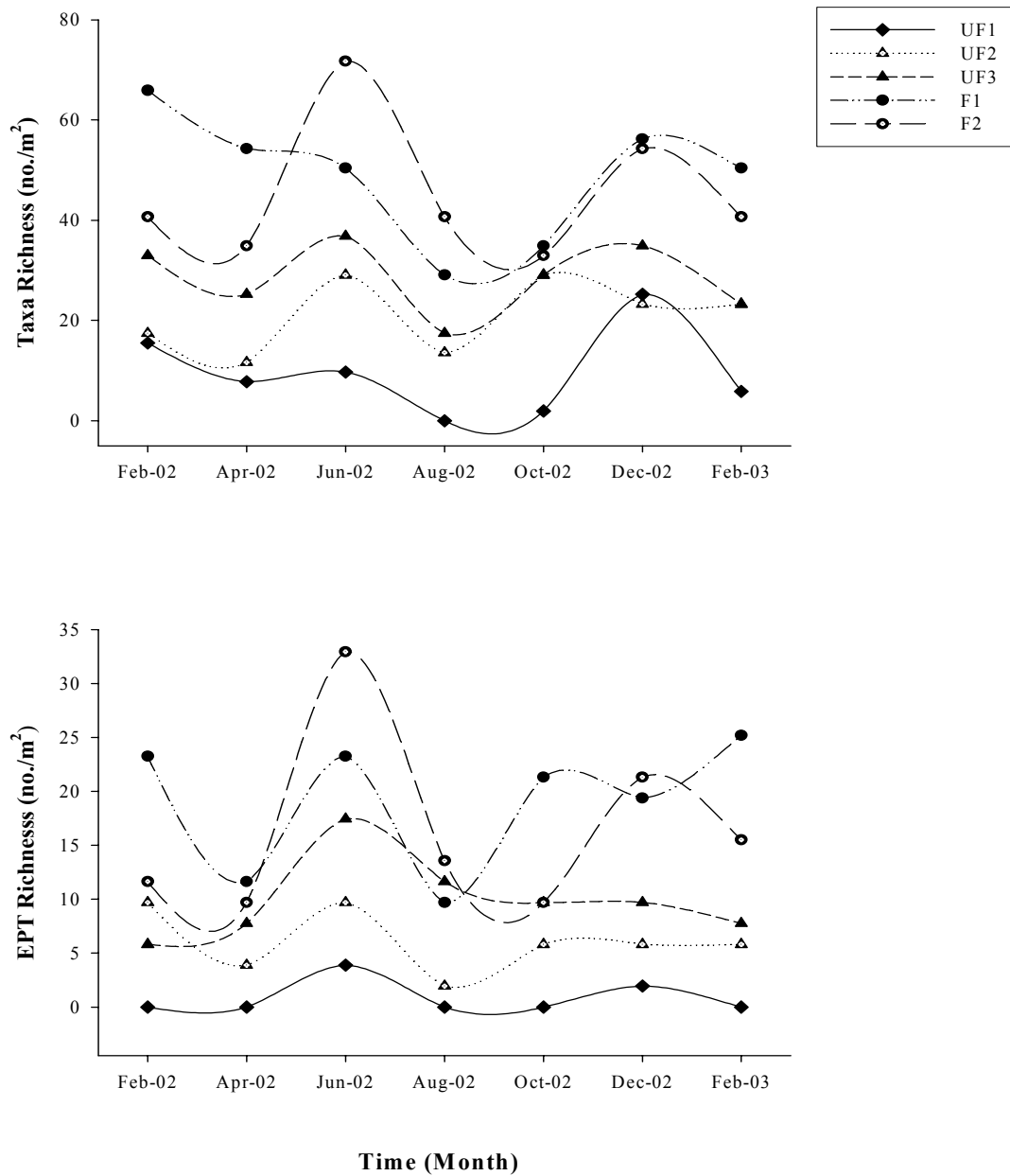


Figure 2.24. Mean benthic macroinvertebrate (all species) and EPT (Ephemeroptera-Plecoptera-Trichoptera) taxa richness (no./m²) at each site from February 2002 to February 2003. Both total species and EPT richness was highest at both fenced sites F1 and F2 and lowest at unfenced site UF1 ($p < 0.0001$).

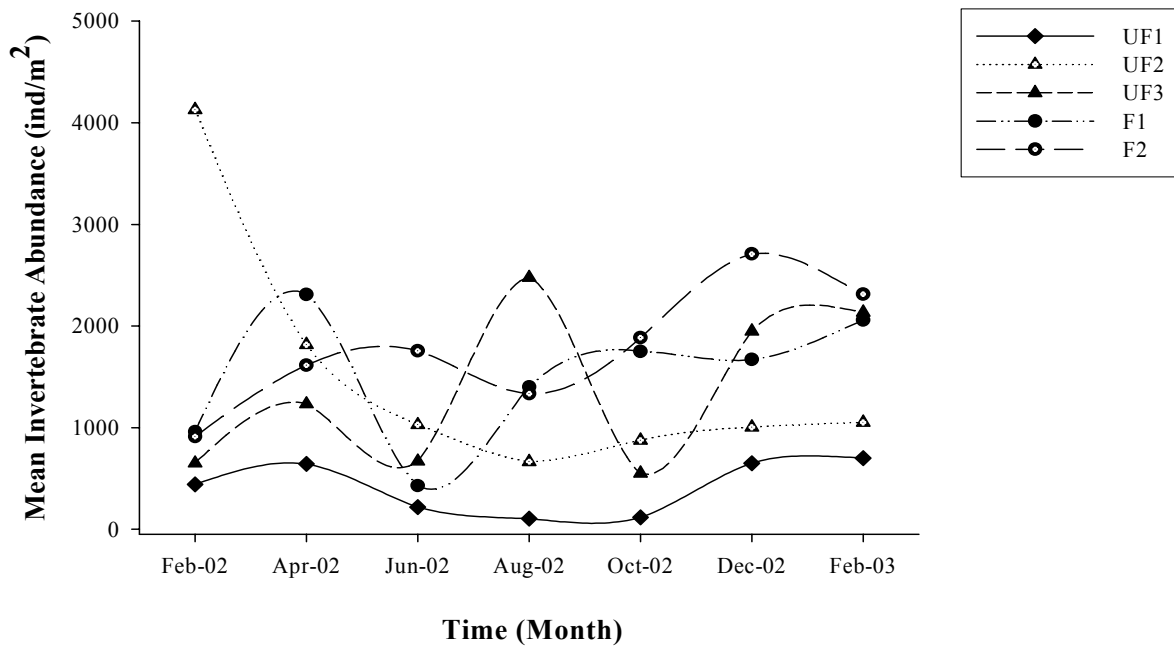


Figure 2.25. Mean benthic macroinvertebrate abundance (ind/m²) at each site from February 2002 to February 2003. Abundance was significantly lowest at site UF1, whereas all other sites were similar in abundance levels ($p < 0.0001$).

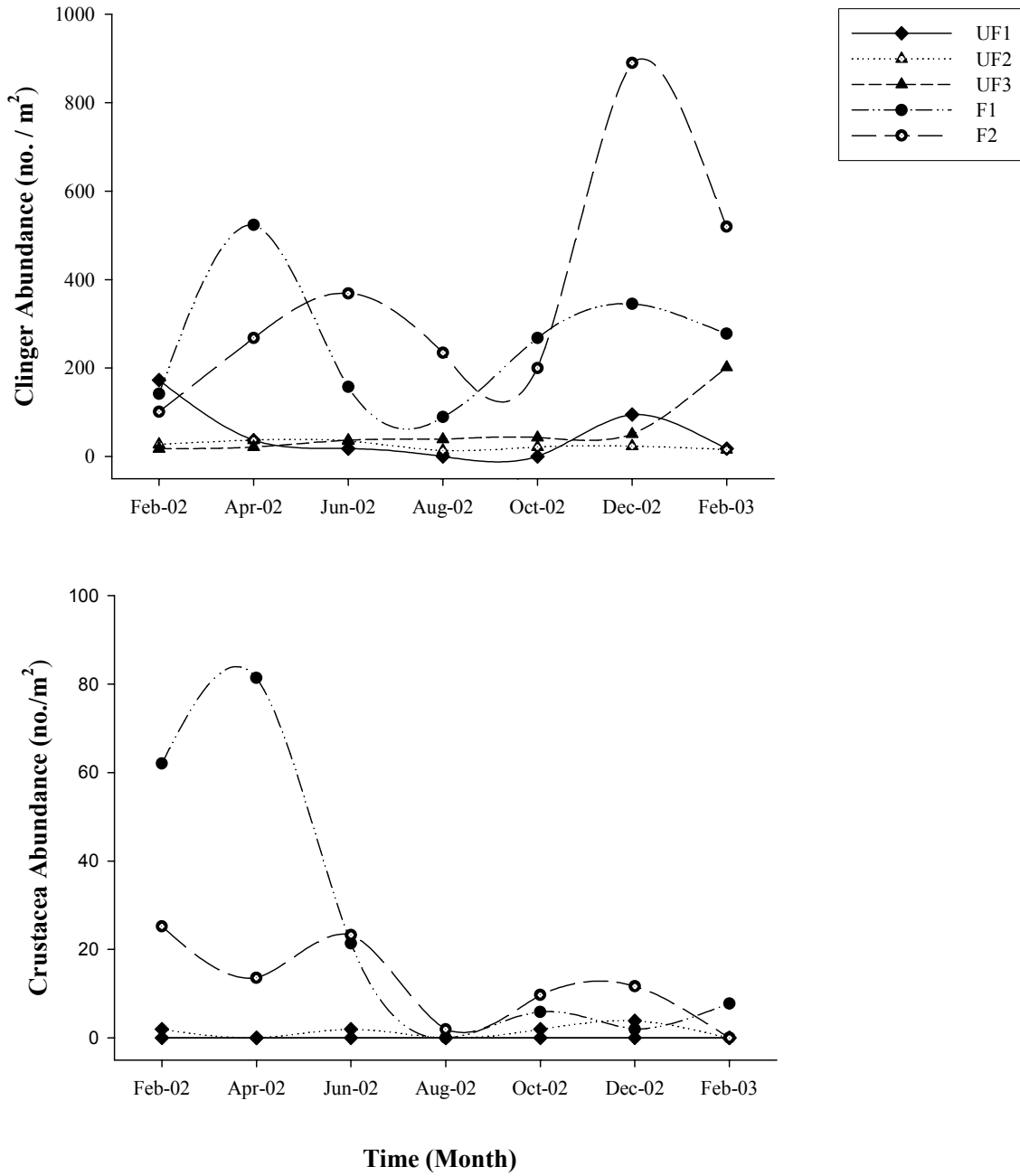


Figure 2.26. Clinger and Crustacean abundance (no./m²) at each site from February 2002 to February 2003.

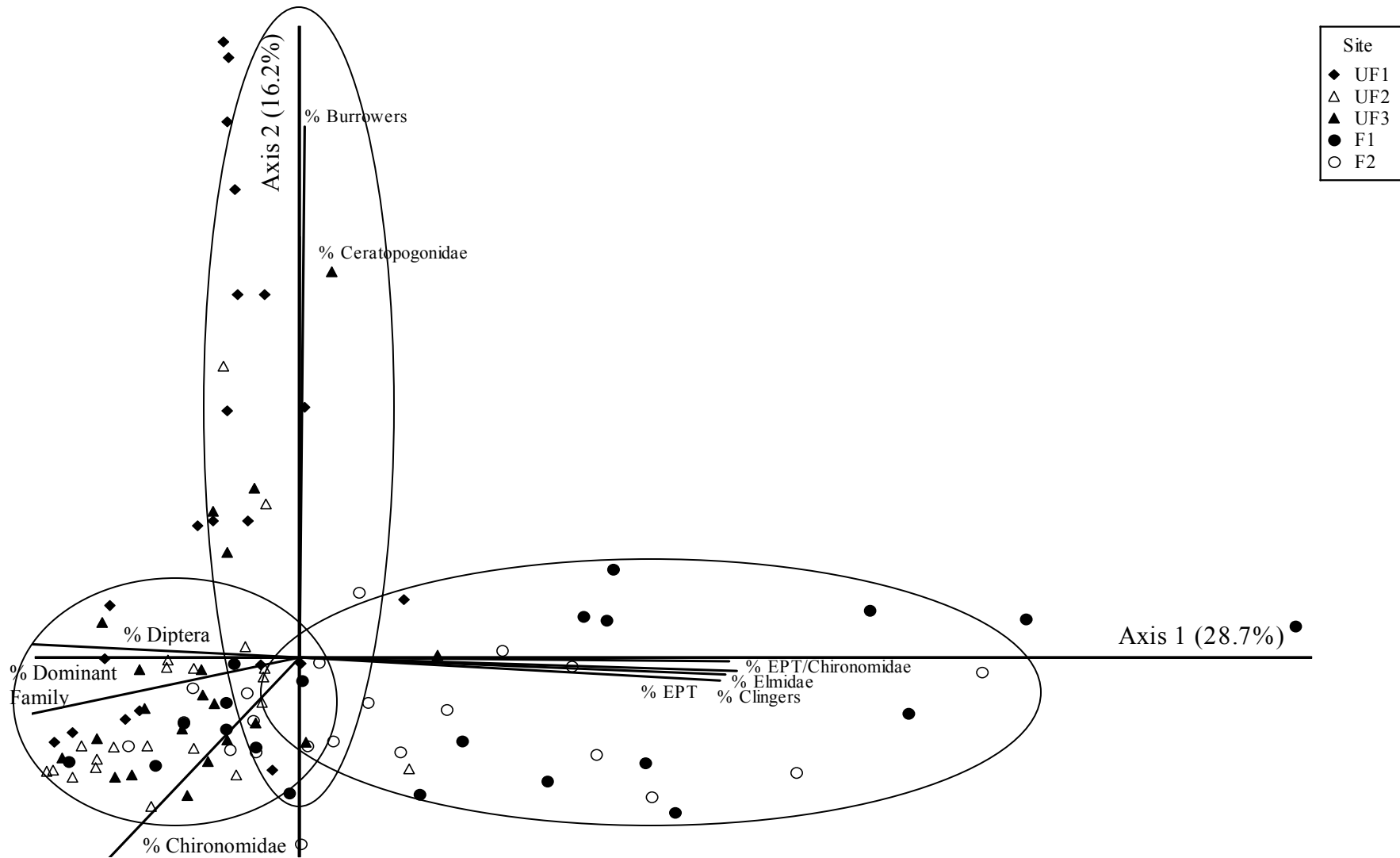
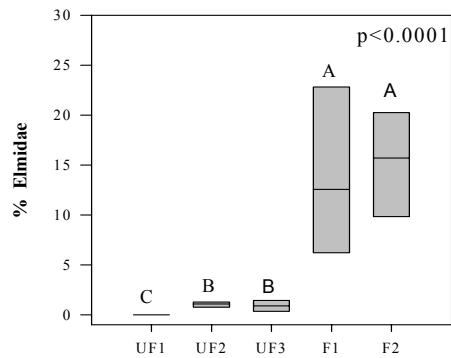
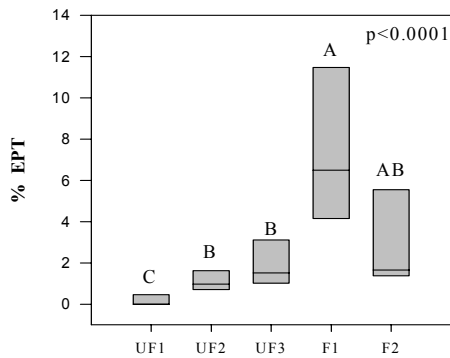
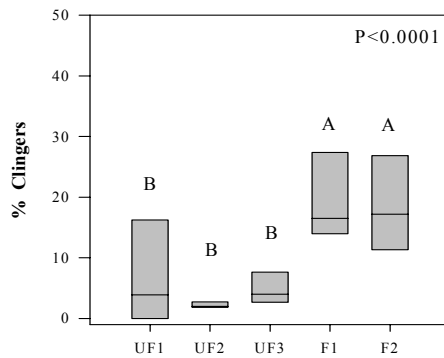
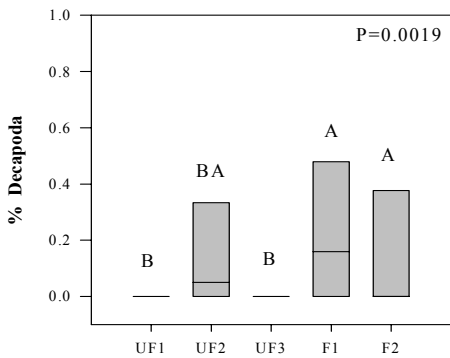
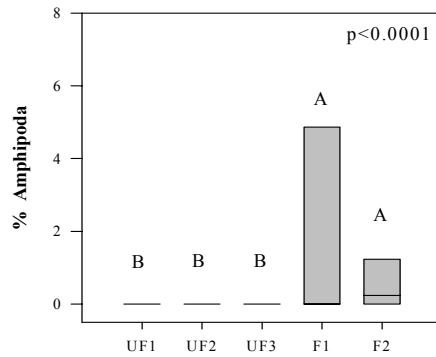
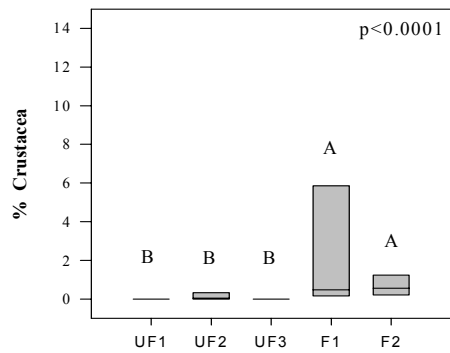


Figure 2.27. First and second axes of the principal components analysis (PCA) for benthic macroinvertebrate metrics at all sites. Points represent individual site/dates and vectors indicate metrics. Percent variation is explained in parentheses on each axis.



Site

Site

Figure 2.28. Box plots of invertebrate metrics for all sites (median, 25th and 75th percentiles, maximum value, minimum value and outliers). For each stream site and metric, values with different letters are significantly different (Kruskal-Wallis test with respective p-value).

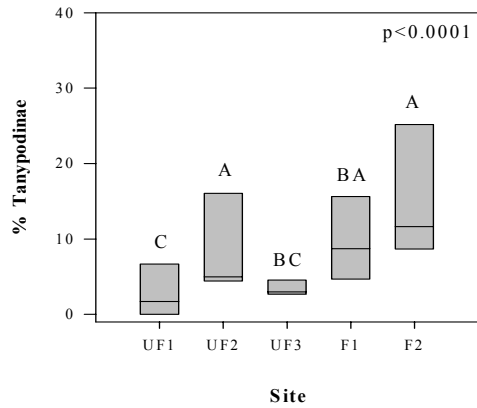
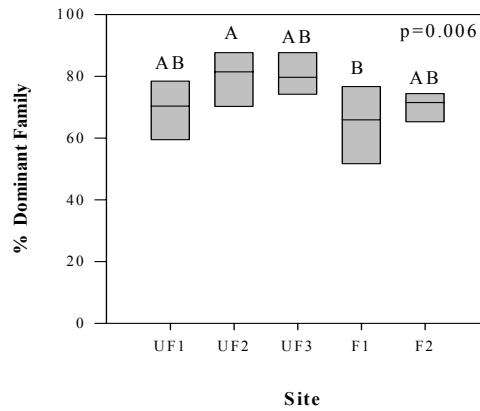
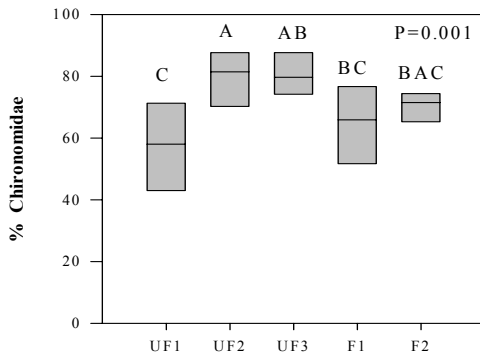
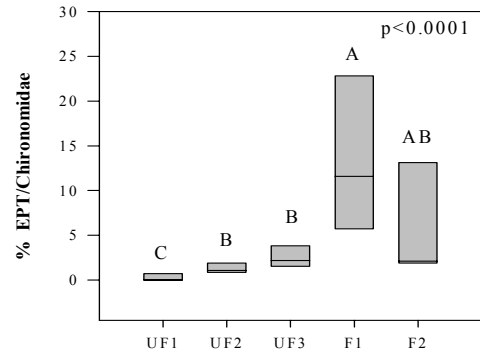
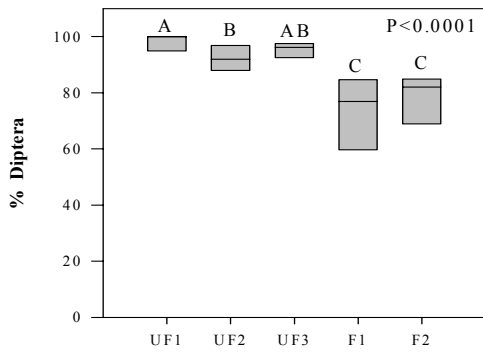


Figure 2.29. (Cont'd) Box plots of invertebrate metrics for all sites (median, 25th and 75th percentiles, maximum value, minimum value and outliers). For each stream site and metric, values with different letters are significantly different (Kruskal-Wallis test with respective p-value).

Georgia Adopt-A-Stream Index for Macroinvertebrates

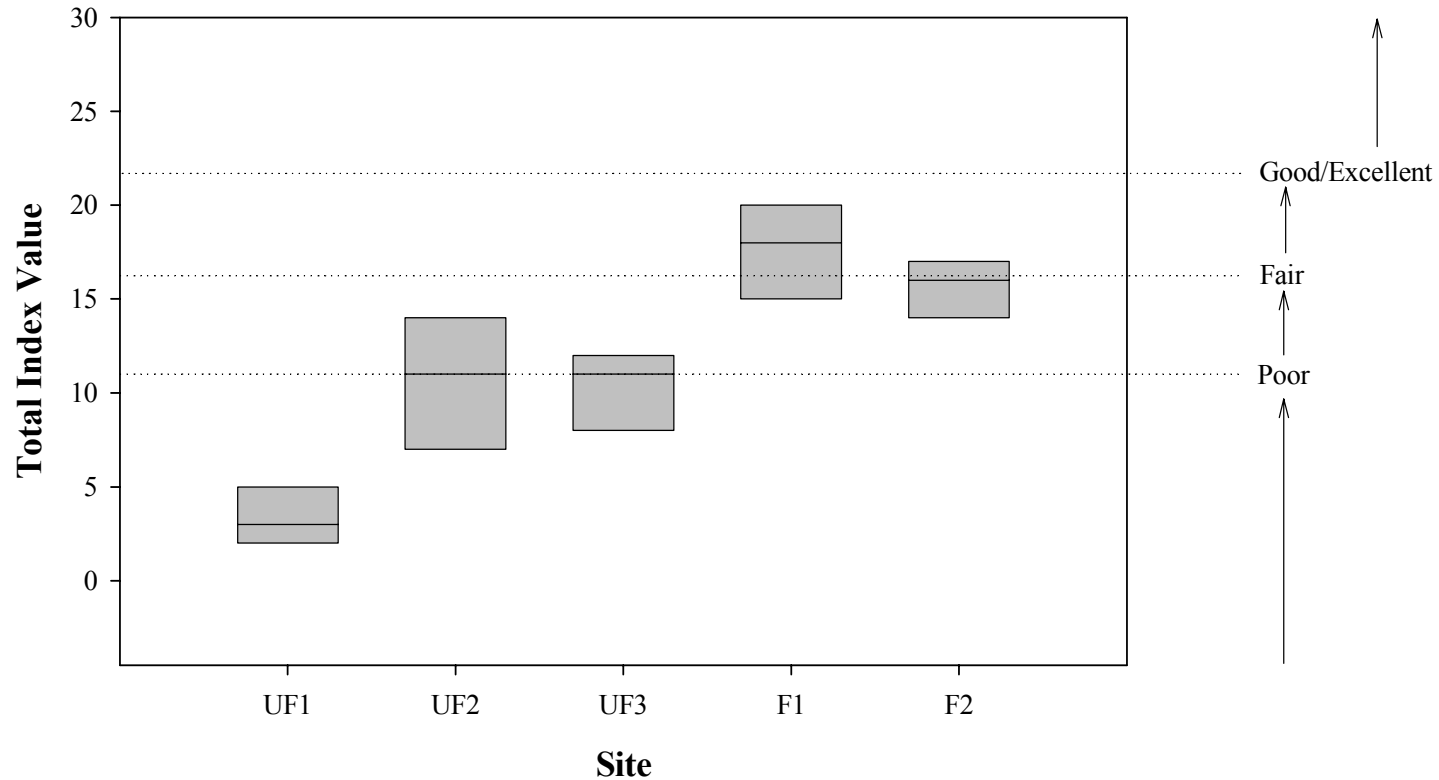


Figure 2.30. Box plots for Georgia Adopt-A-Stream macroinvertebrate index values and ratings for water quality at each site from February 2002 to February 2003 (median, 25th and 75th percentiles, maximum value, minimum value and outliers). Water quality rating is based on index values such as the following: Excellent (>22), Good (17-22), Fair (11-16), and Poor (<11) (GA DNR 2000).

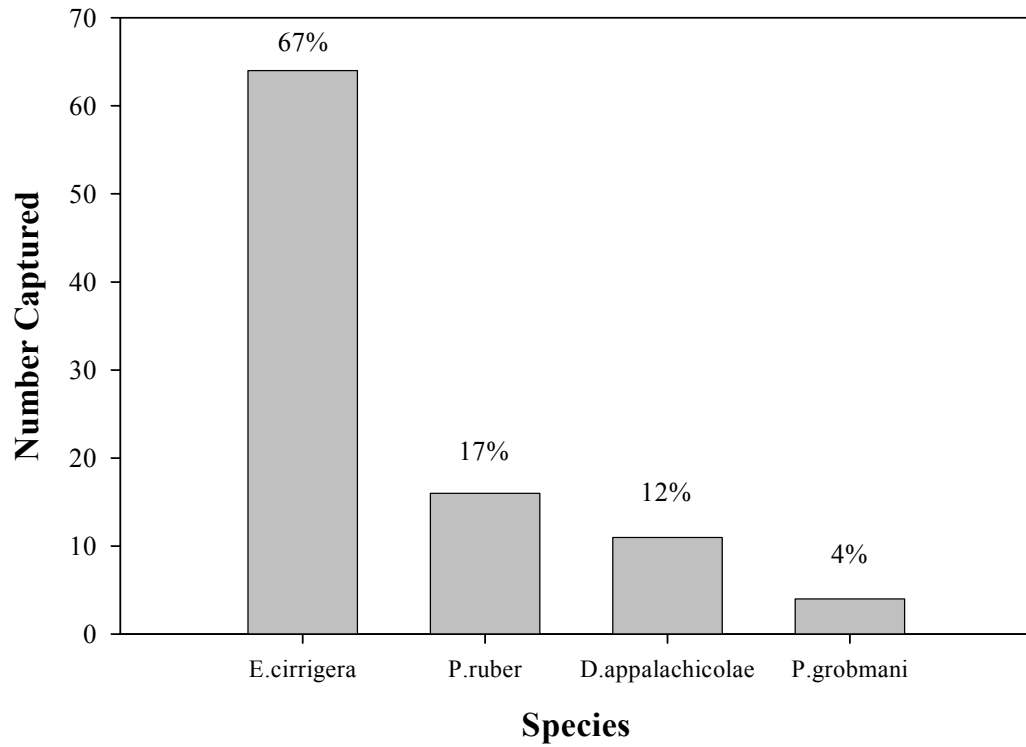


Figure 2.31. Total number of Plethodontid salamanders observed during natural cover and artificial cover searches, Early County, Georgia. Percentages represent that of total individuals.

Table 2.6. Abundances of amphibians species (no. individuals) found during: A) natural cover searches, artificial cover searches (cover boards) and invertebrate collections (Hess sampler), and in B) tree pipe refugia, at each site for all dates combined.

A. Species/Site	Natural Cover					Artificial Cover					Hess Sampler				
	UF1	UF2	UF3	F1	F2	UF1	UF2	UF3	F1	F2	UF1	UF2	UF3	F1	F2
Plethodontidae															
<i>Desmognathus apalachicola</i> (A)	--	2	--	3	--	--	1	--	3	2					
<i>Eurcycea cirrigera</i> (A)	11	3	7	3	7	7	1	7	11	7					
<i>Eurcycea cirrigera</i> (L)											1	--	1	19	18
<i>Plethodon grobmani</i> (A)	--	1	1	1	1	--	--	--	--	--					
<i>Pseudotriton ruber</i> (A)	7	1	2	--	1	--	--	--	1	3					

(A)=adult form, (L) =larval form

B. Species/Site	Tree Pipe Refugia				
	UF1	UF2	UF3	F1	F2
Hylidae					
<i>Hyla chrysoscelis</i>	2	1	--	1	4
<i>Hyla cinerea</i>	9	11	1	4	20
<i>Hyla squirella</i>	73	45	84	35	118

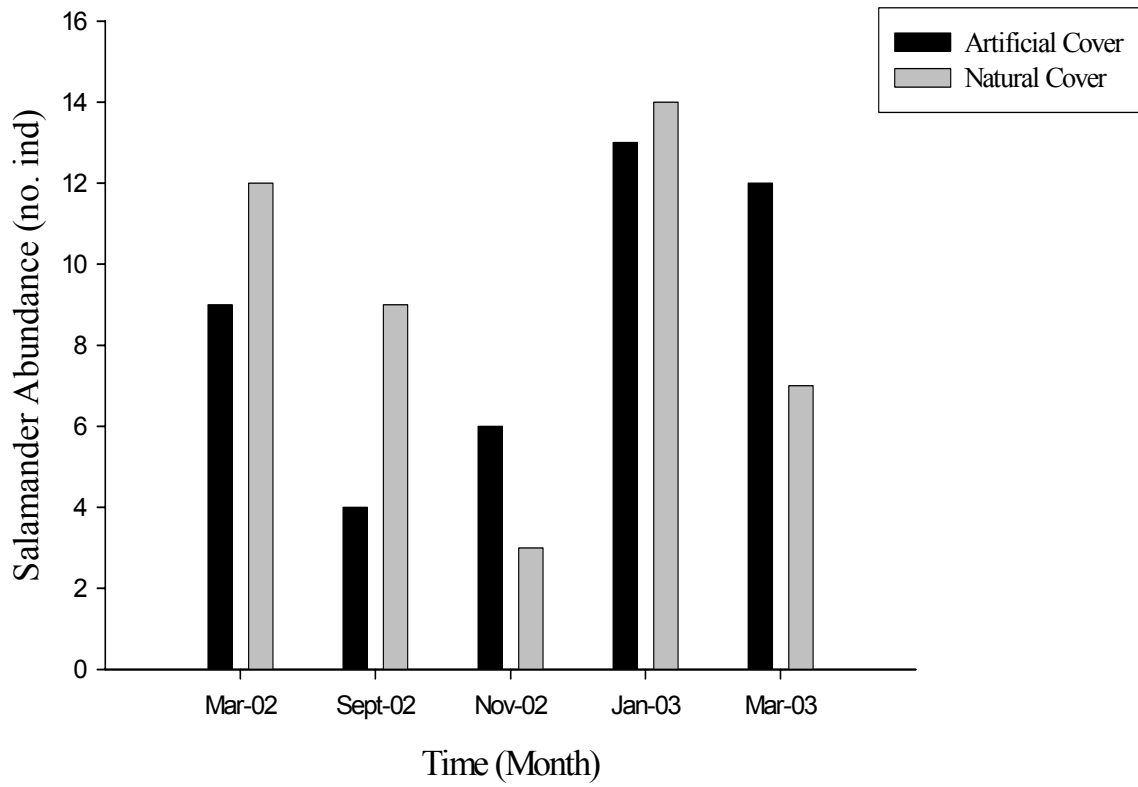


Figure 2.32. Abundance of salamanders (no. ind) encountered during natural cover and artificial cover searches for five dates (March/September/November 2002, and January/March 2003).

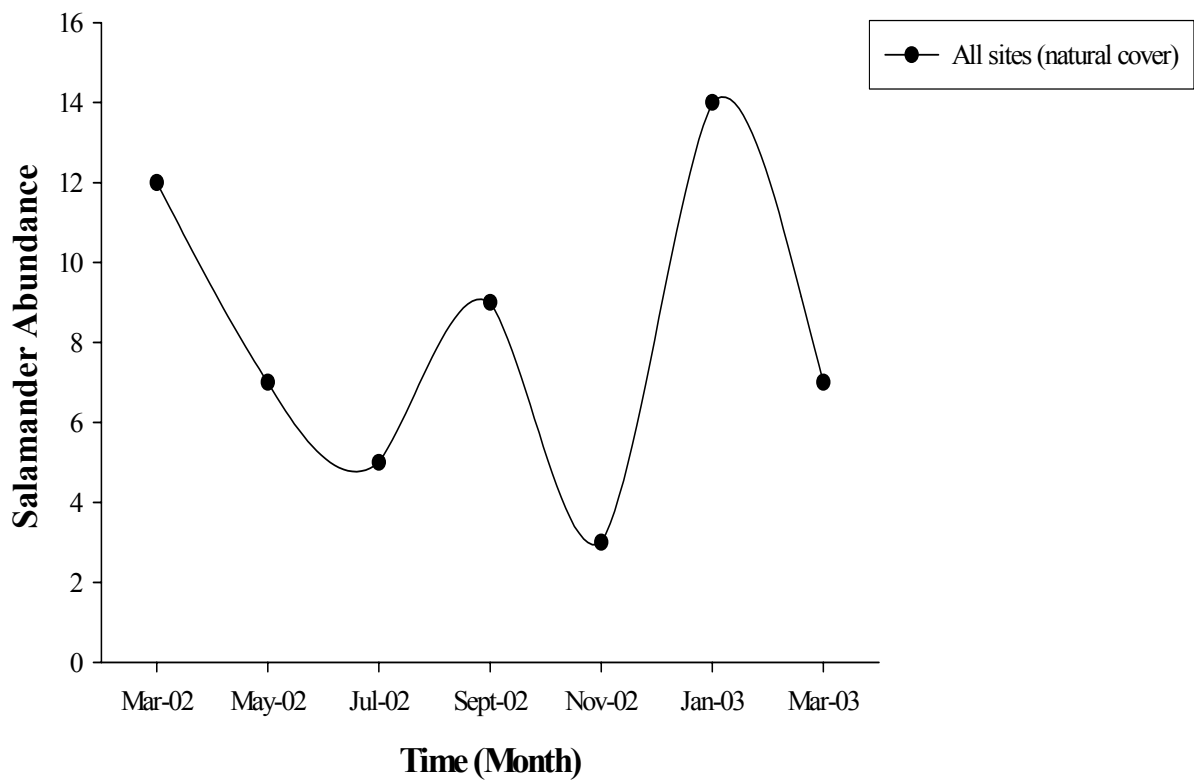


Figure 2.33. Salamander abundance (using natural cover survey data) for all sites combined, from March 2002 to March 2003.

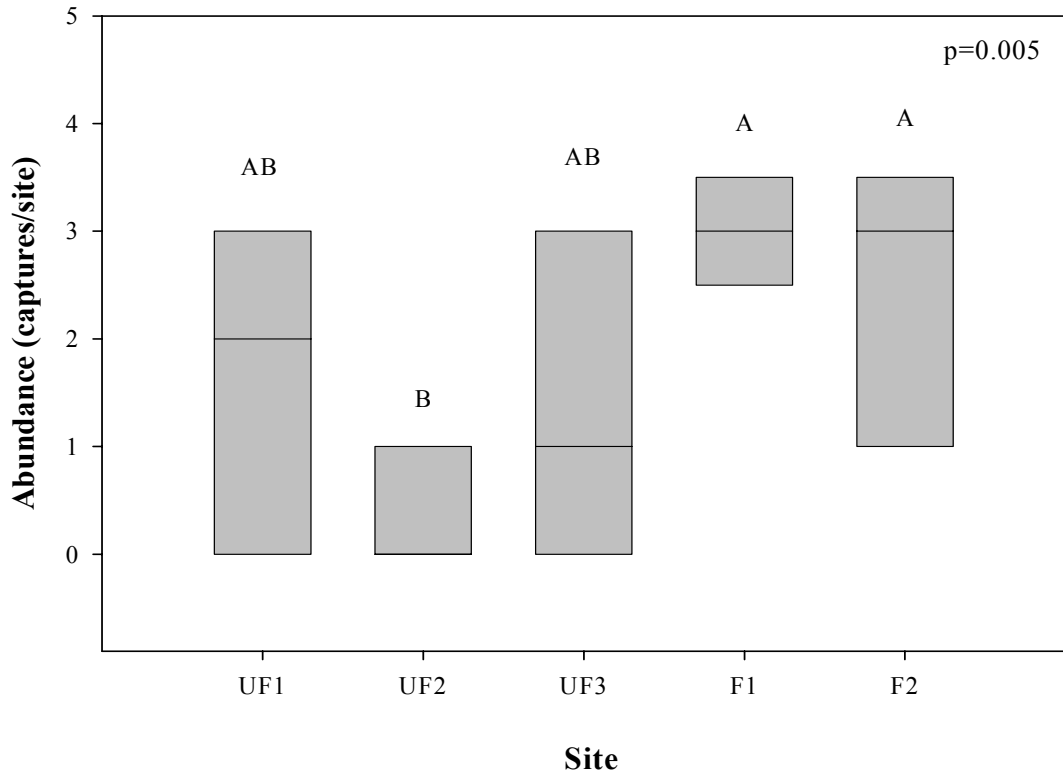


Figure 2.34. Box plots of salamander abundance at each site using cover board searches (median, 25th and 75th percentiles, maximum value, minimum value and outliers). Values with different letters are significantly different (Kruskal-Wallis test with respective p-value).

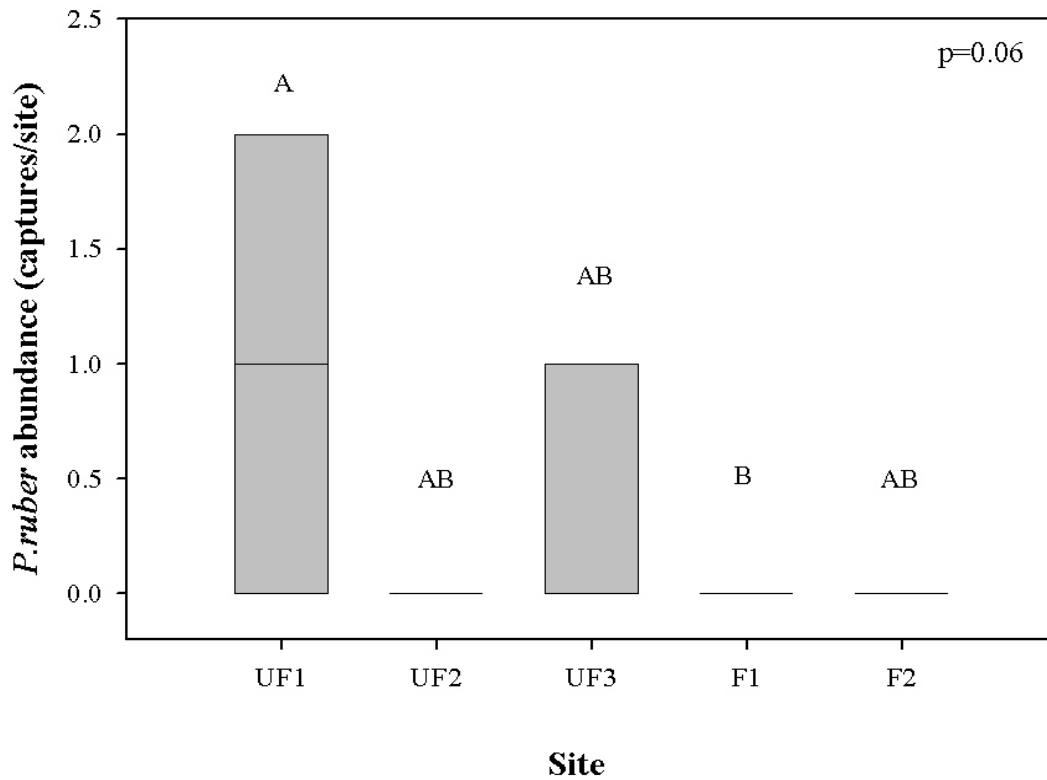


Figure 2.35. Box plots of the abundance of the red salamander, *Pseudotriton ruber*, found through natural cover searches, at each site (median, 25th and 75th percentiles, maximum value, minimum value and outliers). Values with different letters are significantly different (Kruskal-Wallis test with respective p-value).

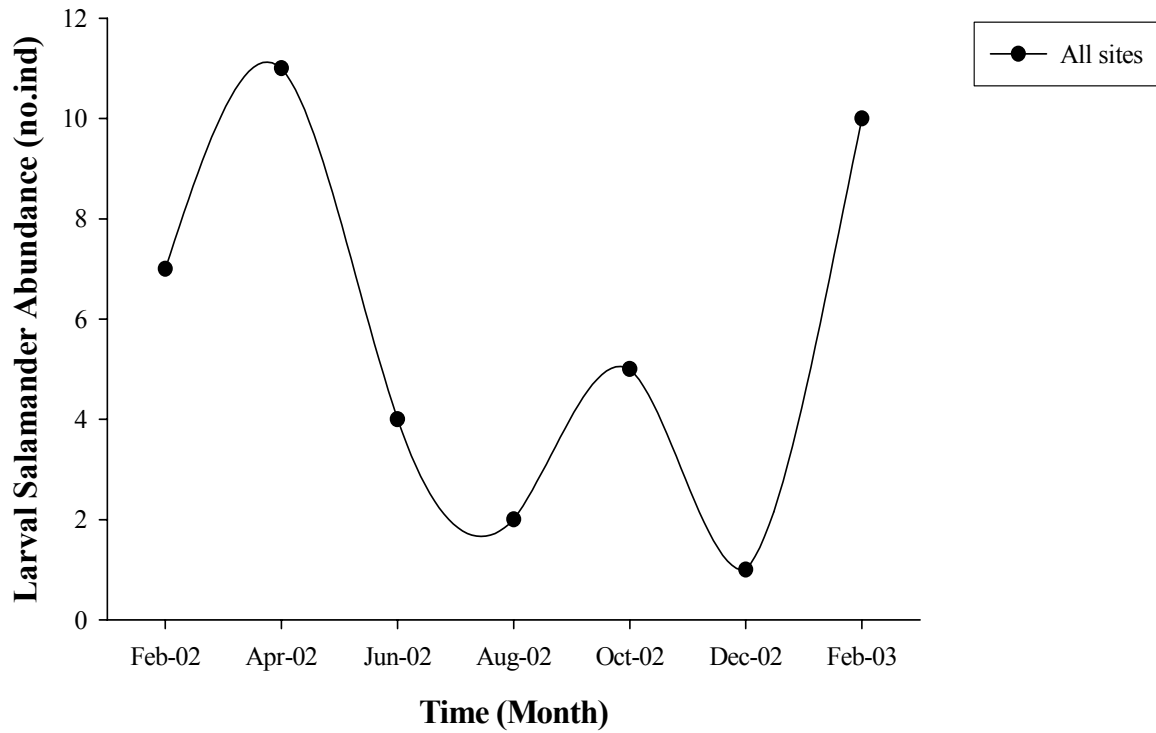


Figure 2.36. Abundance of salamander larvae (no.ind), *Eurycea cirrigera*, captured within bimonthly invertebrate collections, from February 2002 to February 2003.

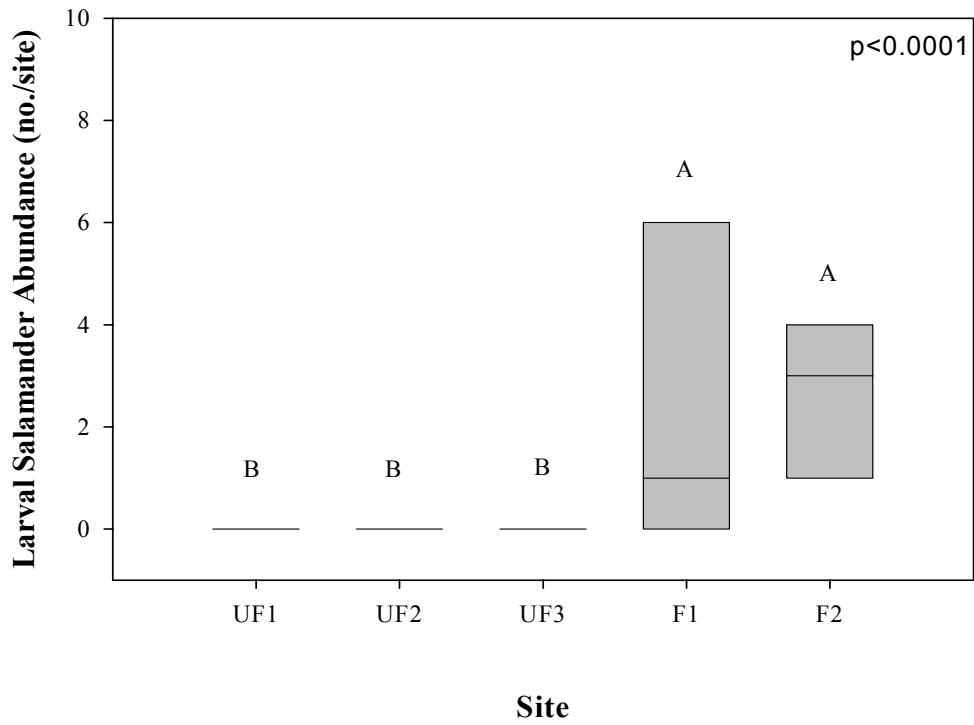


Figure 2.37. Box plots of the abundance of the southern two-lined salamander, *E. cirrigera*, captured within bimonthly invertebrate collections, February 2002-February 2003 (median, 25th and 75th percentiles, maximum value, minimum value and outliers). Values with different letters are significantly different (Kruskal-Wallis test with respective p-value).

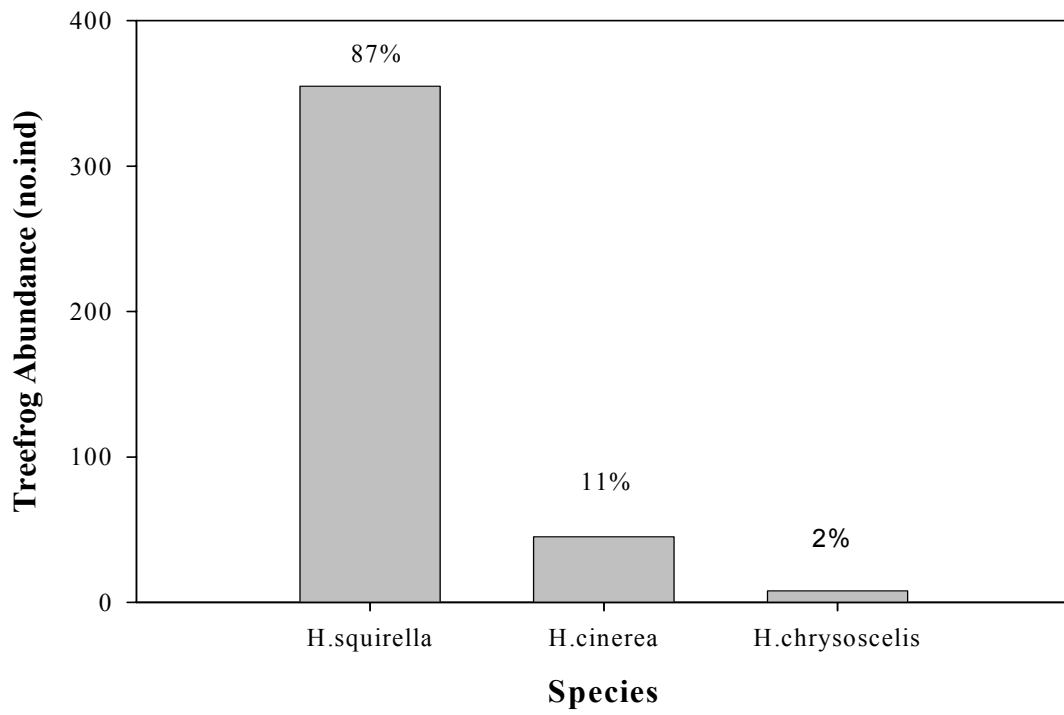


Figure 2.38. Total number of hylid treefrogs captured in PVC pipe traps, from June 2002 to March 2003, Early County, Georgia.

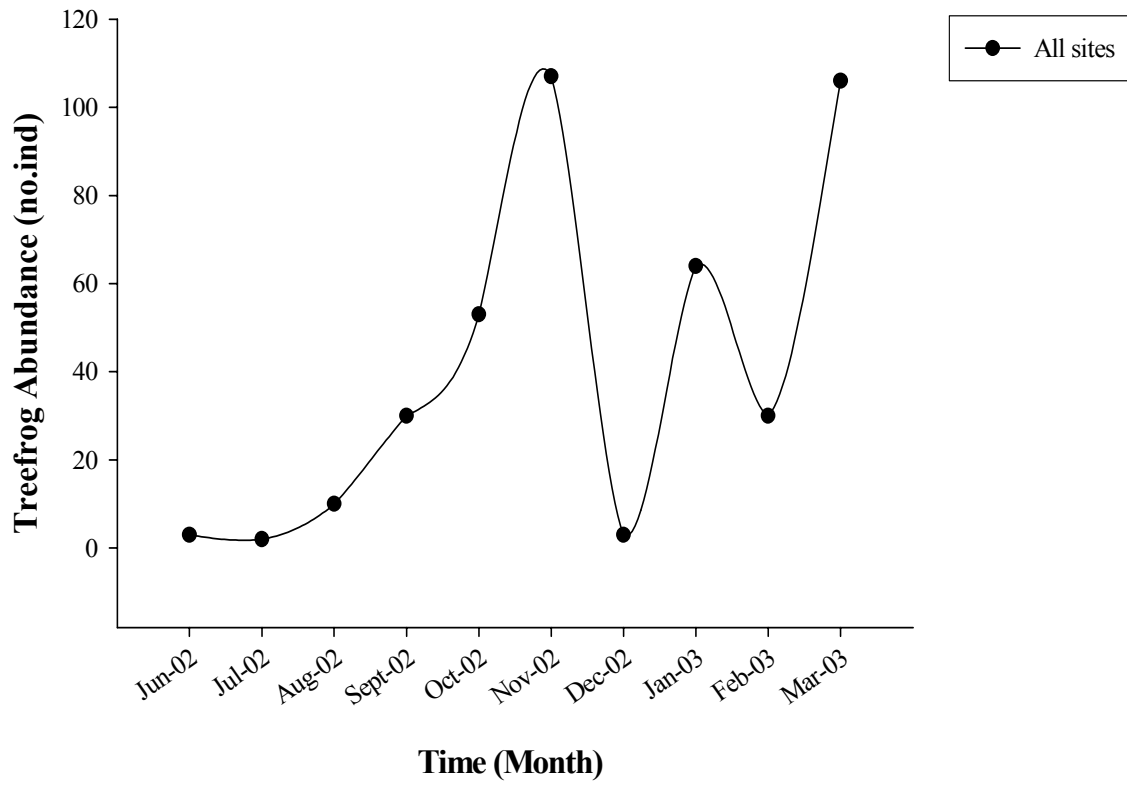


Figure 2.39. Seasonal abundances of treefrogs at all sites, from June 2002 to March 2003. Percentages represent that of total individuals.

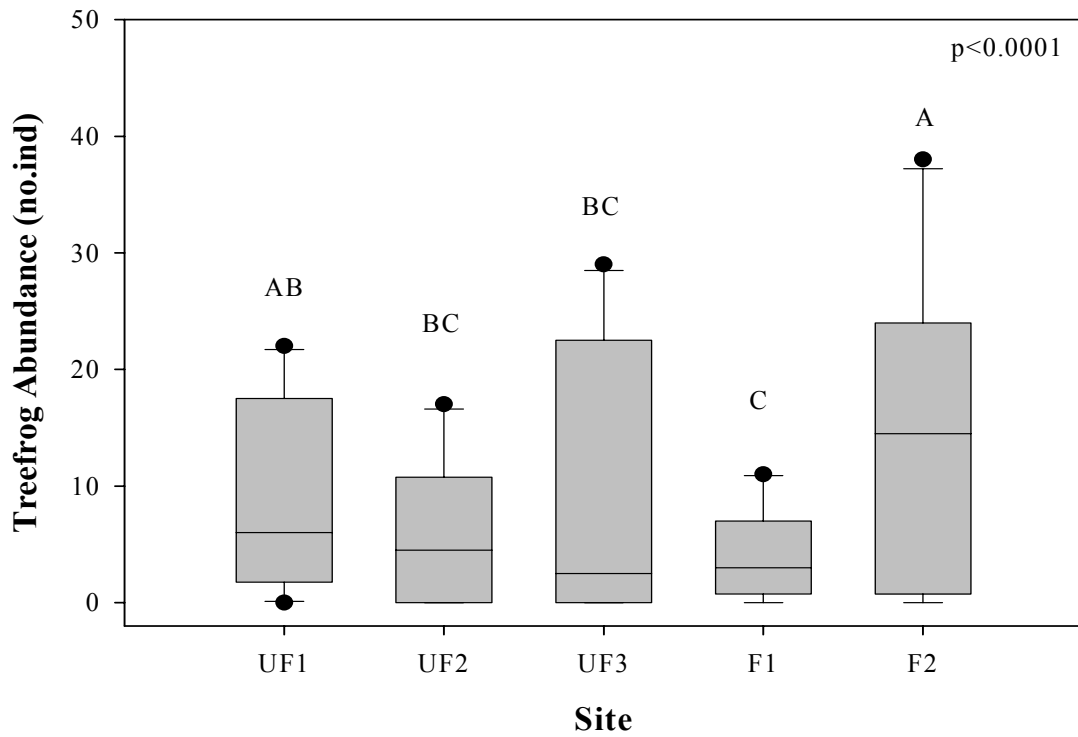


Figure 2.40. Box plots of treefrog abundance at each site, from June 2002 to March 2003 (median, 25th and 75th percentiles, maximum value, minimum value and outliers). Values with different letters are significantly different (Kruskal-Wallis test with respective p-value).

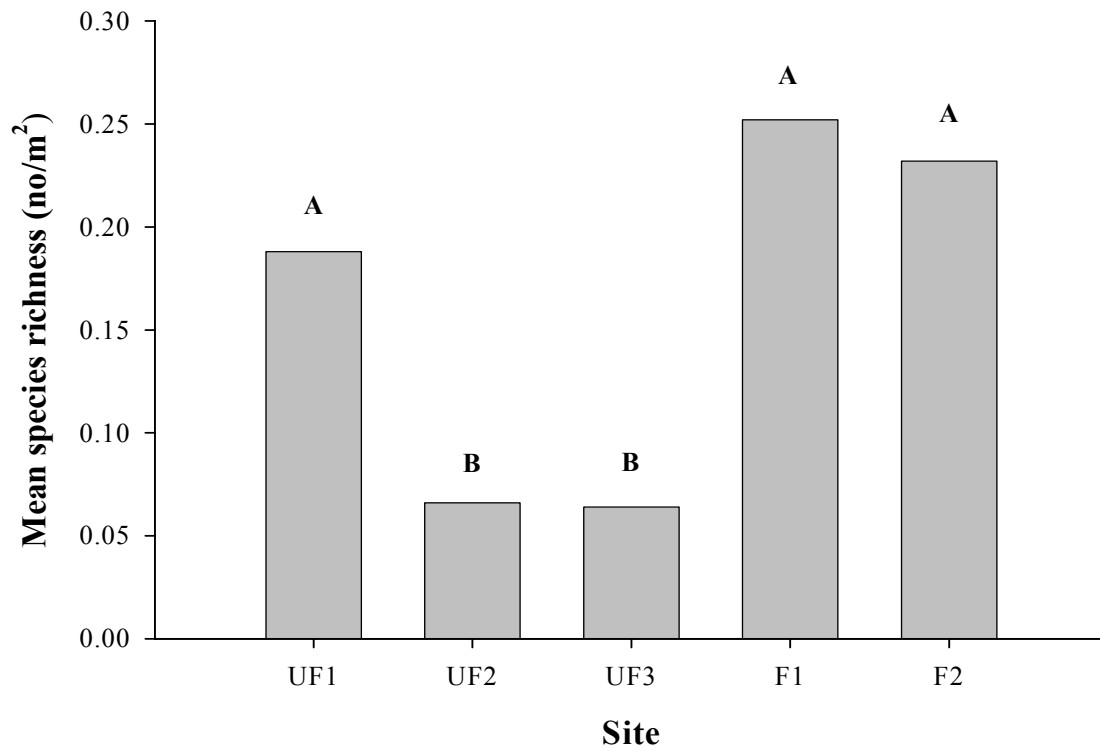


Figure 2.41. Mean vegetative species richness at each site. Columns marked with different letters are significantly different (Kruskal-Wallis test, $p < 0.0001$).

Table 2.7. Jaccards' Index (JI) of percent vegetative species similarity between sites.

SITE	UF-1	UF-2	UF-3	F-1	F-2
B-1	--	15.4%	13.6%	20.0%	20.6%
B-2	15.4%	--	26.3%	10.0%	15.2%
B-3	13.6%	26.3%	--	15.7%	17.6%
R-1	20.0%	10.0%	15.7%	--	46.6%
R-2	20.6%	15.2%	17.6%	46.6%	--

Note: $JI = a / (a + b + c)$ where a = the number of species in common between two sites (A, B), b = the number of species at site A not at site B, and c = the number of species at site B and not at site A.

Table 2.8. Importance values (IV) for canopy riparian species at each site, calculated as the sum of relative density, relative dominance, and relative frequency divided by 3 (basis of 100).

	UF1	UF2	UF3	F1	F2
<i>Acer rubrum</i>	0.9	1.8	7.7	7.2	3.7
<i>Alnus serrulata</i>	44.3	--	--	--	--
<i>Celtis laevigata</i>	--	--	2.8	--	--
<i>Cornus florida</i>	--	--	--	1.0	1.2
<i>Diospyrus virginiana</i>	--	--	--	--	1.2
<i>Halesia diptera</i>	--	--	--	3.8	2.3
<i>Ligustrum sinense</i>	--	10.2	--	--	--
<i>Liquidambar styraciflua</i>	1.8	7.7	--	4.4	11.2
<i>Liriodendron tulipifera</i>	--	38.8	23.2	13.7	24.3
<i>Magnolia grandiflora</i>	--	14.4	--	--	9.2
<i>Magnolia virginiana</i>	29.8	--	34.8	42.5	41.8
<i>Nyssa biflora</i>	19.6	4.0	10.8	12.0	5.6
<i>Oxydendron arboreum</i>	--	--	2.0	--	--
<i>Pinus taeda</i>	--	--	--	3.7	--
<i>Prunus serotina</i>	--	2.5	5.1	1.9	--
<i>Quercus alba</i>	--	--	9.1	1.6	--
<i>Quercus nigra</i>	--	18.8	4.5	--	--
<i>Salix nigra</i>	3.6	--	--	--	--
<i>Sassafras albidum</i>	--	--	--	1.4	--
<i>Symplocos tinctoria</i>	--	--	--	5.0	--
<i>Unknown 1</i>	--	--	--	0.9	--
<i>Unknown 2</i>	--	--	--	0.9	--
<i>Unknown 3</i>	--	1.8	--	--	--

Table 2.9. Summary of riparian structural characteristics: Mean, minimum, and maximum measurements at each site. For each site and parameter, values with different letters are significantly different (Kruskal-Wallis test with respective p-value).

Parameter	Mean (Min. – Max.)					p-value
	UF1	UF2	UF3	F1	F2	
Basal Area (m ² /ha)	19.65	86.28	41.73	54.14	66.55	N/A
Floodplain Width (m)	15 (10-23)	30 (23-36)	20 (15-23)	25 (18-30)	25 (24-27)	N/A
Slope (%)	22a (7-29)	18a (5-48)	12a (9-18)	15a (6-27)	18a (5-38)	0.9252
Canopy Opening (riparian) (%)	21a (4-54)	7b (0-29)	8b (1-24)	5b (1-16)	7b (1-23)	<0.0001
Sand Cover (%)	79a (61-100)	74a (50-92)	49ab (15-77)	15b (0-31)	12b (0-31)	0.0004
Leaf Litter Cover (%)	13b (0-23)	25b (8-46)	42ab (23-77)	83a (69-100)	75a (46-100)	0.0003
Vegetative Cover (%)	4a (0-8)	0a	2a (0-8)	2a (0-8)	10a (0-15)	0.1429
Robel Pole Coverage (%)	50a (0-100)	4b (0-18)	2b (0-14)	14a (3-31)	23a (2-68)	0.0001
Woody (%)	4a (0-8)	0a	2a (0-8)	0a	4a (0-8)	0.2930
CWD Frequency (no./100m ²)	20a (7.2-32)	6.8a (3.2-8)	18.6a (4-34.4)	13.2a (11.2-17.6)	10.4a (5.6-13.6)	0.1257
CWD Volume (m ³)	1.39a	0.53a	1.66a	0.58a	0.54a	0.0711
Tree Frequency (no./100m ²)	17.4a (8-40)	8b (6.4-9.6)	7.2b (3.2-12.8)	15a (12-17.6)	12.4ab (9.6-15.2)	0.1373
Snag Frequency (no./100m ²)	1.4a (0-2.4)	0.4a (0-1.6)	0.6a (0-1.6)	2.2a (0.8-3.4)	0.4a (0-1.6)	0.0599
Soil Temperature (°C)	17.25a (11.3-25)	16.62a (9.4-26)	17.41a (11.3-25.9)	17.71a (12.1-24.9)	18.45a (12.4-25.3)	0.7029
Soil Compression (kg/cm ²)	0.50c (0-4.5)	2.12a (0-4.5)	0.97b (0-4.5)	0.45c (0-1.75)	0.44c (0-3.25)	<0.0001

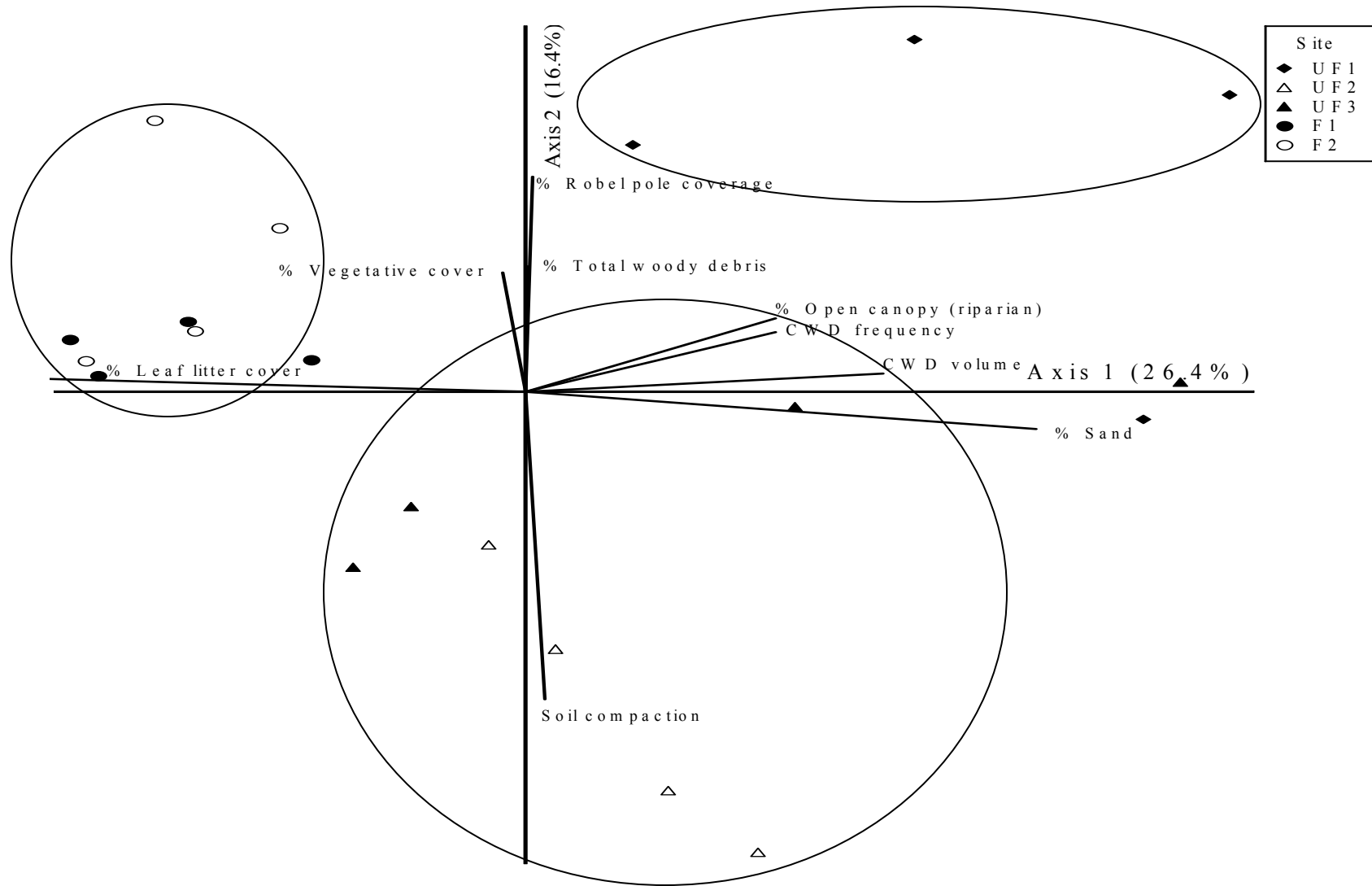


Figure 2.42. First and second axes of the principal components analysis (PCA) for riparian vegetative and soil parameters at all sites. Points represent individual site/dates and vectors indicate riparian variables. Percent variation is explained in parentheses on each axis.

CHAPTER 3

GET IN MY BELLY! FEEDING PREFERENCES BY THE STREAM DWELLING LARVAL SALAMANDER, *EURYCEA CIRRIGERA*, IN AN IMPACTED AGRICULTURAL AREA, SOUTHWEST GEORGIA.

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ABSTRACT

Feeding preference of the southern two-lined salamander, *Eurycea cirrigera*, was studied in streams affected by agricultural practices in southwest Georgia. Larvae were collected within bimonthly benthic macroinvertebrate samples using a 500 μ m mesh Hess sampler from February 2002 to February 2003. Five stream reaches were sampled, two of which were fenced from cattle (F-1 and F-2) and three allowed cattle access (UF-1, UF-2, and UF-3). The entire digestive tract was removed from larvae and stomach contents were examined to better understand prey selection in streams with differing intensities of adjacent agricultural land-use. Forty salamander larvae were recovered from the invertebrate collections, with significantly higher captures at fenced sites than unfenced sites ($p < 0.0001$). Invertebrates were enumerated in 34 salamander stomachs, with Chironomidae comprising the largest percentage of individuals found, both in stomach contents and habitat collections. Electivity values showed a wide range of variability among individual salamanders, however, overall indices suggest slight positive selection for a subfamily of the Chironomidae, the Tanypodinae. It appears that *E. cirrigera* larvae select for Tanypodinae, however, this invertebrate group was found at all stream sites, suggesting that other or additional factors than prey abundance, such as stream habitat quality, may also influence larval salamander abundance.

Key words: Electivity, *Eurycea cirrigera*, southern two-lined salamander, prey, food-preference, agriculture, streams, benthic macroinvertebrates, chironomid, Tanypodinae

INTRODUCTION

Many salamander larvae such as the southern two-lined salamander, *Eurcyea cirrigera* depend upon habitats and refugia within stream ecosystems which are crucial to their development and overall survival (Petranka, 1998). However, land-use practices such as agriculture pose a threat to streams, degrading them by altering natural flow regimes, disrupting habitat, and altering water quality (Schultz et al., 1995). In addition to chemical and physical effects, agricultural land-use has also been found to alter benthic macroinvertebrate assemblages, a major prey source for larval salamanders (Strand and Merritt, 1999; Davis, 2000).

Southern two-lined salamanders survive in a wide range of habitats and are distributed from southern West Virginia to eastern Illinois and south into northern Florida and eastern Louisiana. Living at stream margins and in forest floor habitats in both the juvenile and adult stages, they are totally aquatic during their first two to three years (Duellman and Wood, 1954). Larvae are primarily benthic, found mostly in slow-moving pools except when drifting downstream in fast currents (Petranka, 1998). Throughout the larval period they feed within the substrate, roaming stream bottoms in search of small prey using chemical, tactile, and visual cues (Petranka, 1998). Noted as opportunistic generalists, Petranka (1984) examined the feeding behavior of *Eurycea bislineata* larvae, and found them to feed on the same proportion and size of prey as either small or large larvae (Petranka, 1998). However, Zaret (1980) found them to be gape limited in the maximum size of prey that could be swallowed due to the physical constraint of the jaw width. Although current knowledge of the feeding ecology of larval salamanders is relatively well documented (e.g. Caldwell and Houtcooper, 1973; Petranka, 1984), effects of land use alterations on prey selection have yet to be noted.

We examined feeding preferences of salamander larvae that were collected within benthic macroinvertebrate samples from Coastal Plain streams in Southwest Georgia, as part of a study looking at the impacts of cattle grazing on stream health (see Chapter 2). The larvae collected were used to determine (1) diet composition of larval two-lined salamanders, *Eurycea cirrigera*, in streams with a history of disturbance due to agricultural practices, and (2) prey selection as compared with prey distribution and abundance. This study also sought to provide greater insight into the abundance of *E. cirrigera*, relating to land-use, and to address the following question (3) is salamander larval abundance related to abundance of prey items and/or prey type?

METHODS

Study Site Description

Stream sites for this study were located on a diversified row crop and beef cattle farm within southwest Georgia in the Fall Line Hills physiographic district. The area is characterized by frequently meandering streams underlain by easily eroded sands, clays and gravel. Located 15-75 m below the adjacent ridge tops, these streams experience extensive amounts of erosion, forming steep gullies or washes (SWG RDC 1998), and receive considerable amounts of ground-water discharge (USGS 1996). The climate is warm and humid subtropical, with short winters. Average monthly temperatures range from 3-15 ° C in January to 21-33°C in July (Southeast Regional Climate Center, SERCC). Average annual precipitation is 142 cm, with the average minimum monthly rainfall occurring in October (7 cm) and the maximum in January (16 cm) (SERCC).

Five 100 m stream reaches were selected for physical, chemical, and biological assessment. All were located in Factory Creek sub-watershed, a 2nd order tributary of the Lower

Chattahoochee River. Three stream sites were unfenced from cattle access, and will be referred to herein as UF-1, UF-2 and UF-3, and two were fenced, F-1 and F-2, which have been limited from cattle access for over 20 years (M. Brownlee, property owner, pers comm.). The canopy cover in the riparian area was dominated by species such as Southern Magnolia, *Magnolia grandiflora*; Sweetbay Magnolia, *M. virginiana*; Swamp Tupelo, *Nyssa biflora*; Sweetgum, *Liquidambar styraciflua*; and Yellow Poplar, *Liriodendron tulipifera*. All streams were perennial, with an average width of 2.0 m, an average depth of 0.09 m, and an average velocity of 0.01 m/s. Stream temperatures ranged from 12.1°C in December to 23.3°C in August, and dissolved oxygen from 4.4 mg/L in October to 9.2 mg/L in February.

Macroinvertebrate and Salamander Larvae Collection

Salamander larvae were recovered from preserved invertebrate collections made bimonthly from February 2002-February 2003 with a 500 µm mesh Hess sampler (Hess, 1946). Collections were made anywhere from 09:00hrs to 16:00hrs (EST). Within the selected 100 m reach at each site, three random stream transects were chosen. At each transect, composite samples were taken comprising two Hess collections, sampling representative habitat types within the stream channel. Samples were rinsed into plastic bags, preserved in the field with 70% ethanol and stained with rose Bengal dye. In the laboratory, sample processing involved rinsing material (e.g. invertebrates, salamander larvae, sand, organic matter) through a 1 mm and 500 µm sieve. Salamander specimens were identified (Petranka, 1998), and blotted dry to remove excess moisture and weighed to the nearest 0.01g. Snout-vent length (SVL) of the larvae was measured in mm. The entire digestive tract was then removed for gut content analysis. Invertebrates from gastrointestinal (GI) tracts and invertebrate captures (with Hess sampler) were enumerated and identified with a low power dissecting microscope and light compound

microscope to lowest taxonomic level possible, usually order or family, but in some cases to genus (Berner and Pescador, 1988; Stewart and Stark, 1988; Thorp and Covich, 1991; Pescador et al., 1995; Epler, 1996 and 2001; Wiggins, 1996; Needham et al., 2000). Larval Chironomidae (Diptera) within invertebrate captures from two dates (February and August 2002) were mounted on microscope slides in CMC and identified to genus (Epler, 2001). Samples of more than 500 individuals were subsampled and adjusted to a final volume of 100ml with 70% ethanol, placed on a magnetic stirrer to produce a homogenous solution, and then three 5-ml subsamples were removed with a wide-bore pipette (Hax and Golladay, 1993). Larval Chironomidae within GI tracts were processed in a similar manner, and identified to genus when possible.

Statistical analysis

Pearson's correlation coefficient was used to compare the relative abundance of selected prey taxa in the benthos and the salamander's GI tracts. Differences amongst abundance of salamander larvae at each site were compared using a Kruskal-Wallis One Way ANOVA on ranks (SAS Institute, Inc. 2002).

Feeding electivity indices are used to elucidate relationships between prey availability and diet composition and have been used to compare the feeding habits of organisms such as fish, frogs, and salamanders (e.g. Parker, 1994; Baker, 2002; Hirai and Matsui, 2002). Strauss's (1979) linear index of feeding electivity was used in this study to evaluate prey selection based on the abundance of prey in the habitat. Strauss' index was selected because it addresses potential biases based on disparate samples sizes of gut contents and habitat, and is noted as a more statistically reliable index with a less complex variance structure (Strauss, 1976). The linear index is calculated as follows:

$$L = r_i - p_i$$

Where r_i is the relative abundance of each prey item (i) in the gut and p_i is the relative abundance of each prey item in the habitat. This index gives a value ranging from -1 to +1, with values near zero indicating neutral selection or opportunistic feeding, positive values indicating selectivity for a prey item (relative to its availability in the habitat), and negative values indicating avoidance. For this study, relative patterns were reported based on whether scores were positive or negative. For statistical purposes of analysis, only those taxa represented in both the stomach contents of salamander larvae and within the Hess collections (environment) were used. Due to the mesh size of the Hess sampler and sieving methods of the invertebrates from the original study, Crustacean taxa (e.g. Cladocera, Copepoda, Ostracoda) were not found within the habitat samples and thus were not available for electivity calculations. This study therefore focused on the largest taxa group present, both numerically and as biomass, the larval Chironomidae. Sufficient identifications and sample sizes were also available for the subfamily Tanypodinae, thereby permitting evaluation of this taxa group as well as the Non-Tanypodinae using feeding electivity measures.

RESULTS

Salamander Abundance

Forty individual salamander larvae were recovered from the Hess collections, all of which were identified as the southern two-lined salamander, *Eurycea cirrigera*. Larvae were found at each sampling date with over 90% of the individuals collected from fenced sites F-1 (n=18) and F-2 (n=20), and total number of captures for all seasons combined was significantly higher at these sites than at the unfenced sites ($p < 0.0001$) (Figure 3.1). Larvae were found at all of the study sites except unfenced site UF-2. Numbers of captures for all sites combined were

highest during April 2002 and February 2003 (Figure 3.1). Snout-Vent-Length (SVL) ranged from 7 mm-35 mm.

Salamander Diet Composition

Of the 40 GI tracts examined, 34 contained macroinvertebrates, with 293 individual invertebrates identified (Table 3.1). The relative number of dietary items varied amongst sample dates. Average number of prey items for all taxa at all sites was highest in the late summer/fall (August and October 2002), and lowest number of prey items within GI tracts were found in the summer (June 2002) and winter (December 2002 and February 2002/2003) (Figure 3.2).

Individuals of the Order Diptera were the most frequently observed composing overall, 60% of total prey consumed (Figure 3.3). Of Dipterans, 98% were composed of the subfamily Chironomidae (midge larvae). Because they were the dominant prey type in every season (except in April 2002), their abundance pattern throughout the study paralleled the pattern of abundance for all taxa, with highest densities occurring in the late summer/fall (August and October 2002), and lowest densities occurring during the summer, spring, and winter months (February, April, June, December 2002 and February 2003) (Figure 3.2). Three major subfamilies accounted for a majority of the Chironomidae that were identified, Chironominae, Orthoclaadiinae, and Tanypodinae, of which greatest numbers of individuals were found within the Non-Tanypodinae (Chironominae 58.3% of chironomids; Orthoclaadiinae 2.4 %.) (Table 3.1). The Tanypodinae composed 39.3% of chironomids. Genera of the Chironomidae were difficult to identify, possibly due to damage incurred during digestion, however the following genera were noted: *Ablabesmyia*, *Microspectra*, *Polypedilum*, *Thienemannimyia*, and *Zavrelimyia*.

Crustaceans were also frequently observed (38% of total) and comprised cladocerans (28% of total), copepods (6% of total), and ostracods (4 % of total) (Figure 3.3). Crustacean

abundance was highest in April 2002, and the Cladoceran, Chydoridae (water flea), was noted in the diet only during this time. Overall, rare taxa found in the GI tracts included Collembola, Coleoptera (Elmidae), and Hydracarina, and comprised less than 2% of total individuals (Table 3.1).

Benthic Macroinvertebrate Community

A total 7,560 individual organisms were identified, representing 30 genera. As found in the GI tracts of salamander larvae, invertebrate collections from the environment were also dominated by Dipterans (87%), of which 89% were of the family Chironomidae. Average densities of individuals for all taxa combined and for the Family Chironomidae were both highest in the months of August and December 2002 and lowest in February 2002 (Table 3.2). Of Chironomid subfamilies, Tanypodinae comprised 27% of total chironomids, and Non-Tanypodinae (Chironominae 70% and Orthoclaadiinae 3%) comprised 73%. Tanypods were present at all stream sites, however no discernible differences were found between mean site abundances ($p < 0.0001$). A subsample of the chironomids was identified to subfamily and genus, finding higher densities of Chironominae than Tanypodinae or Orthoclaadiinae. The most common chironomid genera identified were: *Ablabesmyia*, *Polypedilum*, *Saetheria*, *Thienemannimyia*, *Zavreliomyia*, and the tribe Tanytarsini.

Prey Preferences

Strauss's linear index showed a wide range of individual salamander variability and seasonal variability in electivity for Chironomidae, Tanypodinae, and Non-Tanypodinae (Figure 3.4). Selection for Chironomidae was mostly positive through time, except in February 2002 (no selection) and April 2002 (slight avoidance). However, overall indices suggest no selection for Chironomidae ($L_0 = -0.043$) (Figure 3.4). Tanypodinae selection was consistently positive

through time, suggesting slight selection ($L_0 = 0.228$), and Non-Tanypodinae were consistently negative, suggesting slight non-selection or avoidance ($L_0 = -0.349$) (Figure 3.4).

DISCUSSION AND CONCLUSIONS

Salamanders of the genus *Eurycea* have been regarded as opportunistic consumers, feeding on what is available within their environment, but are limited by gape size in the prey they consume (Zaret, 1980; Petranka, 1984). However, this study suggests that larval *Eurycea cirrigera* select for a particular taxonomic group, the Tanypodinae. Studies such as Petranka (1984) based their conclusions of opportunistic feeding by examining the effects of prey size and prey type on selection, without calculating prey preference indices or linking the availability of prey within the environment. Burton (1976) even suggested that feeding selectivity should be examined through quantitative studies of stream macroinvertebrate abundance. This study compared diet composition with prey availability, and found great variability in selection for prey taxa through time. However, based on the linear electivity index, it appears that *E.cirrigera* larvae show slight selection for Tanypodinae. This finding, coupled with the observation that Chironomidae larvae alone comprised over half of *E.cirrigera* diets, and were found consistently in GI tracts, suggests some preference or selection for this group.

Numerous macroinvertebrate taxa collected in benthic samples were not found in salamander GI tracts, however the dominant group, the Diptera and more specifically, the Chironomidae, were similar in percentages in both GI tracts and benthic collections. Similar to other studies in the northern U.S. (Kentucky, Indiana and New York) which examined the feeding behavior and diet of *Eurycea bislineata*, this study found small stream benthic invertebrates such as fly pupae, chironomid larvae, copepods, and ostracods as the most frequently consumed prey (Caldwell and Houtcooper, 1973; Burton, 1976; Petranka, 1984).

However, Plecoptera nymphs, which were also common to these studies, were not ingested. Burton (1976) noted a seasonal shift in diet, with chironomids as an important prey source for *E. bislineata* during warm weather and copepods during cooler weather. Unlike this study, chironomids were found to be a major prey source throughout the entire year, composing at least 50% of individuals per sampling date, except in the spring month of April, when Crustaceans (Chydoridae) were the main prey type. Burton also found Chydoridae to be an important component of *E. bislineata* diet, composing 61 % of the total number of prey during the month of October. In this study, there appears to be low selection for Tanypodinae in April and strong negative selection for Non-Tanypodinae overall (Figure 3.4), perhaps reflecting the shift of selection to the Chydoridae. Although the Chydoridae were too small to be captured within invertebrate samples, their high abundance in GI tracts suggests that *E. cirrigera* may sometimes be selectively feeding on the Chydoridae.

The Chironomidae, which in this study were the dominant taxonomic group both as prey consumed and available prey, are the most widely distributed and frequently most abundant insects in freshwater ecosystems (Armitage et al., 1995). An ecologically important group of organisms, they have been found to display a multitude of morphological, physiological, and behavioral adaptations and sensitivities to environmental stresses and disturbances (Armitage et al., 1995; Coffman and Ferrington, 1996; Epler, 2002). As commonly found in electivity studies, the most abundant taxa group (both as available prey in the environment and within stomachs), commonly brings positive electivity scores, indicating selection. However, this was not the case for the Family Chironomidae, or the Non-Tanypodinae, which gave either negative electivity scores or scores close to zero. Although they were the second most abundant subfamily found in GI tracts, Tanypodinae within the environment were present in relatively low abundances.

Aside from abundance, a multitude of factors play a role in prey selection, including those relating to general life history. Tanypodinae larvae are epibenthic predators, crawling or swimming freely within the water column feeding on oligochaetes and other soft bodied invertebrates (Mason, 1998). In contrast, Chironominae are more cryptic, living on or in the benthos in silk-lined tubes (Mason, 1998). Petranka (1998) noted *E. cirrigera* use primarily visual cues to detect prey. This study suggests that tanypods, which are more conspicuous or mobile, are more likely to enter a larval salamander's perceptive field compared to other chironomid subfamilies. Tanypodinae also have rather long bullet-shaped head capsules typically mounted on an elongated thorax, and depending on species, fourth instar larva of Tanypodinae can also attain lengths greater than other subfamilies (Wiederholm, 1983), perhaps making them more conspicuous than other Chironomidae.

Larval *Eurycea cirrigera* were significantly more abundant at sites fenced from cattle than those with cattle access (Chapter 2). This paper explores other reasons or factors for these differences in salamander abundance, one of which is prey type and the abundance of prey within a salamander's habitat. Although *E. cirrigera* abundance was correlated with Tanypodinae abundance, this invertebrate group was found at all stream sites, suggesting that factors other than prey abundance, including stream habitat, may account for differences in larval salamander abundance. Land-use such as agriculture has played a major role in altering stream habitat, structure and function. For example, documented results of cattle grazing include higher levels of sedimentation and nutrient inputs to streams, destruction of riparian and in-stream habitat, and changes in aquatic biota community composition, including decreases in aquatic insect diversity and increases in more stress tolerant taxa (Strand and Merritt, 1999; Thomas, 2002). The Tanypodinae as a group exhibit a wide range of tolerance levels, however some

species (e.g. *Paramerina*) are highly sensitive (Lenat, 1993). Species level identification of chironomids is needed to gain a better understanding of the tolerance levels of invertebrates available as prey, and if land-use affects such as agriculture play a role in shaping the presence of these taxa. For, if indeed *E. cirrigera* abundance is affected at all by a certain prey type that may be sensitive to stresses associated with such land-use practices, then conservation of stream habitats is imperative. Although current populations of *E. cirrigera* in the U.S. appear to be stable, more knowledge of their life history and ecological needs would be invaluable to their conservation. Further exploration into prey type relationships and salamander abundance would greatly contribute to this need.

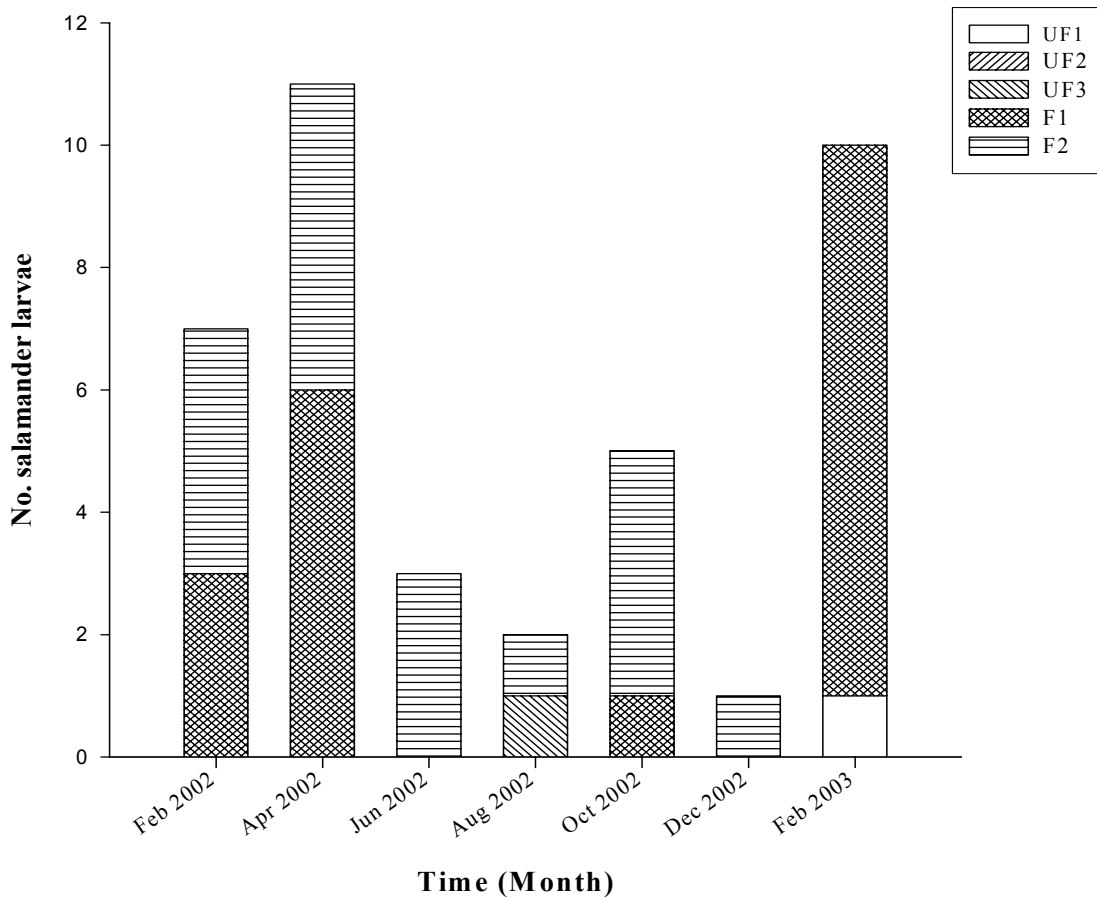


Figure 3.1. Abundance of Southern Two-lined salamander larvae, *Eurycea cirrigera*, collected with a Hess sampler from February 2002 to February 2003 (N=40). Abundances were significantly higher at fenced sites F-1 and F-2 ($p < 0.0001$).

Table 3.1. Seasonal variation in the composition of invertebrate taxa within the diets of larval southern two-lined salamanders, *Eurycea cirrigera*, collected in Early County, Georgia. The values are expressed as an average of the total number (and percent) of dietary items for each stomach per date. (N) = the number of stomachs enumerated for the respective date.

Taxon	Date						
	Feb 2002 N = 5	April 2002 N = 11	June 2002 N = 2	Aug 2002 N = 2	Oct 2002 N = 5	Dec 2002 N = 1	Feb 2003 N = 8
Coleoptera							
Elmidae larvae	0.2 (5.9)	--	--	--	--	--	--
<i>Microcylloepus sp. (adult)</i>	--	--	0.5 (12.5)	--	--	--	--
Unknown larvae	--	--	0.5 (12.5)	--	--	--	--
Collembola	0.2 (5.9)	--	--	--	--	--	--
Crustacea							
Chydoridae	--	7.4 (70.6)	--	--	--	--	--
Copepoda	--	0.9 (8.7)	--	--	--	--	0.1 (3.4)
Calanoida	--	0.3 (2.6)	--	0.5 (3.6)	--	--	--
Cyclopoida	--	0.1 (0.9)	--	--	--	--	--
Ostracoda	--	0.4 (3.5)	--	--	1 (5.3)	--	0.5 (13.8)
Diptera							
Ceratopogonidae	--	0.1 (0.9)	--	--	--	--	2.4 (65.4)
Chironomidae (unknown)	--	0.5 (4.3)	--	4.0 (28.6)	11.8 (63.8)	--	0.6 (17.2)
Chironominae	0.4 (11.8)	0.5 (4.3)	0.5 (12.5)	9.0 (64.3)	3.2 (17.0)	--	--
Orthocladiinae	--	--	--	--	0.4 (2.1)	--	--
Tanypodinae	1.6 (47.1)	0.4 (3.5)	2.5 (62.5)	0.5 (3.6)	2.8 (14.9)	1 (100.0)	--
Empididae							
<i>Hamerodromia sp.</i>	0.2 (5.9)	--	--	--	--	--	--
Tipulidae							
<i>Hexatoma sp.</i>	0.2 (5.9)	--	--	--	--	--	--
Unknown pupae	0.2 (5.9)	--	--	--	0.2 (1.1)	--	--
Hydracarina	--	--	--	--	0.2 (1.1)	--	--
Unidentified	0.4 (11.8)	0.1 (0.9)	--	--	--	--	--
Average no. of individuals	3.4	10.4	4.0	14.0	18.8	1.0	3.6

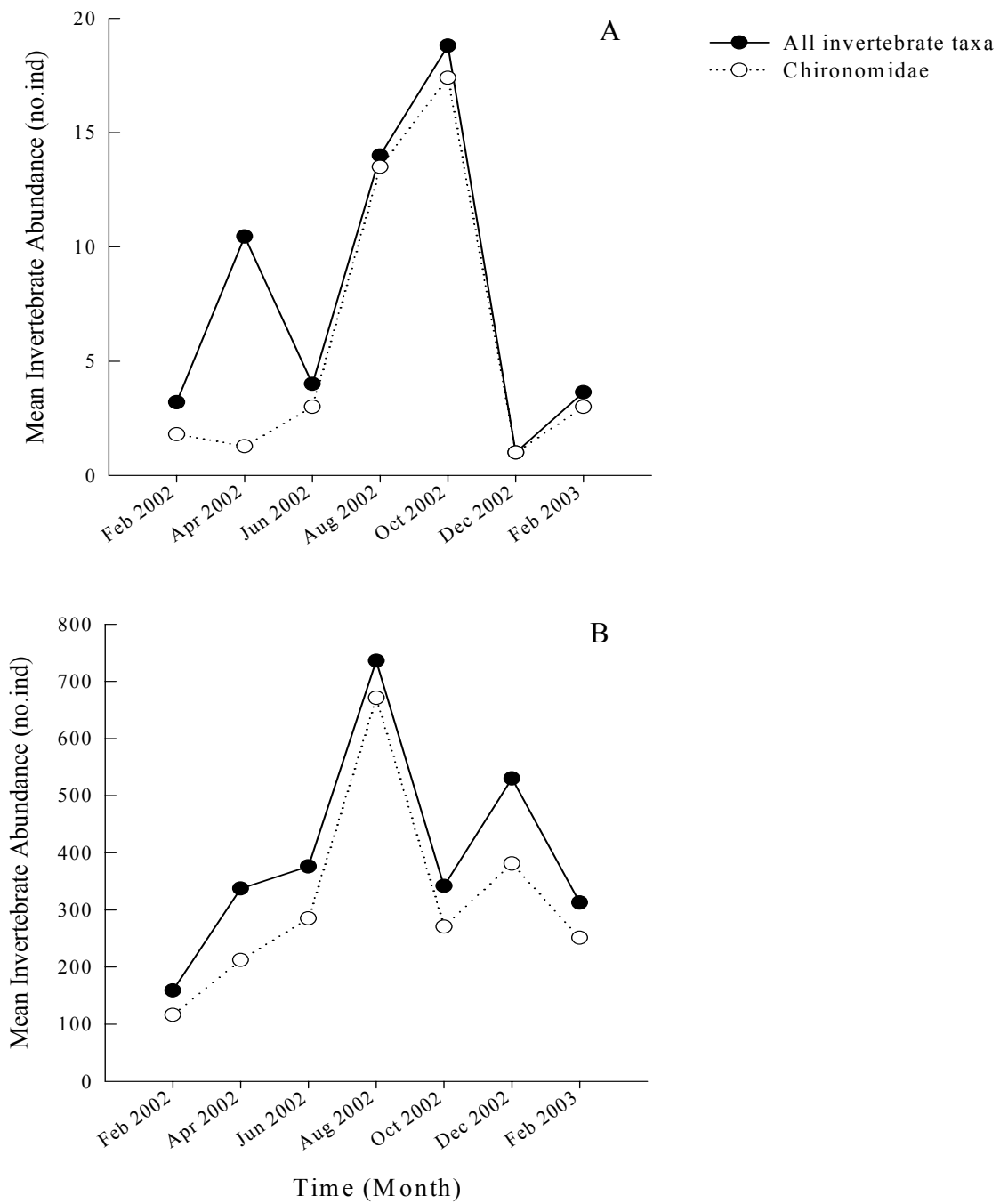


Figure 3.2. Aquatic invertebrate abundance (no.ind) for all taxa combined and of the Family Chironomidae, within salamander gastrointestinal tracts (A) and benthic macroinvertebrate Hess collections (B) from February 2002 to February 2003.

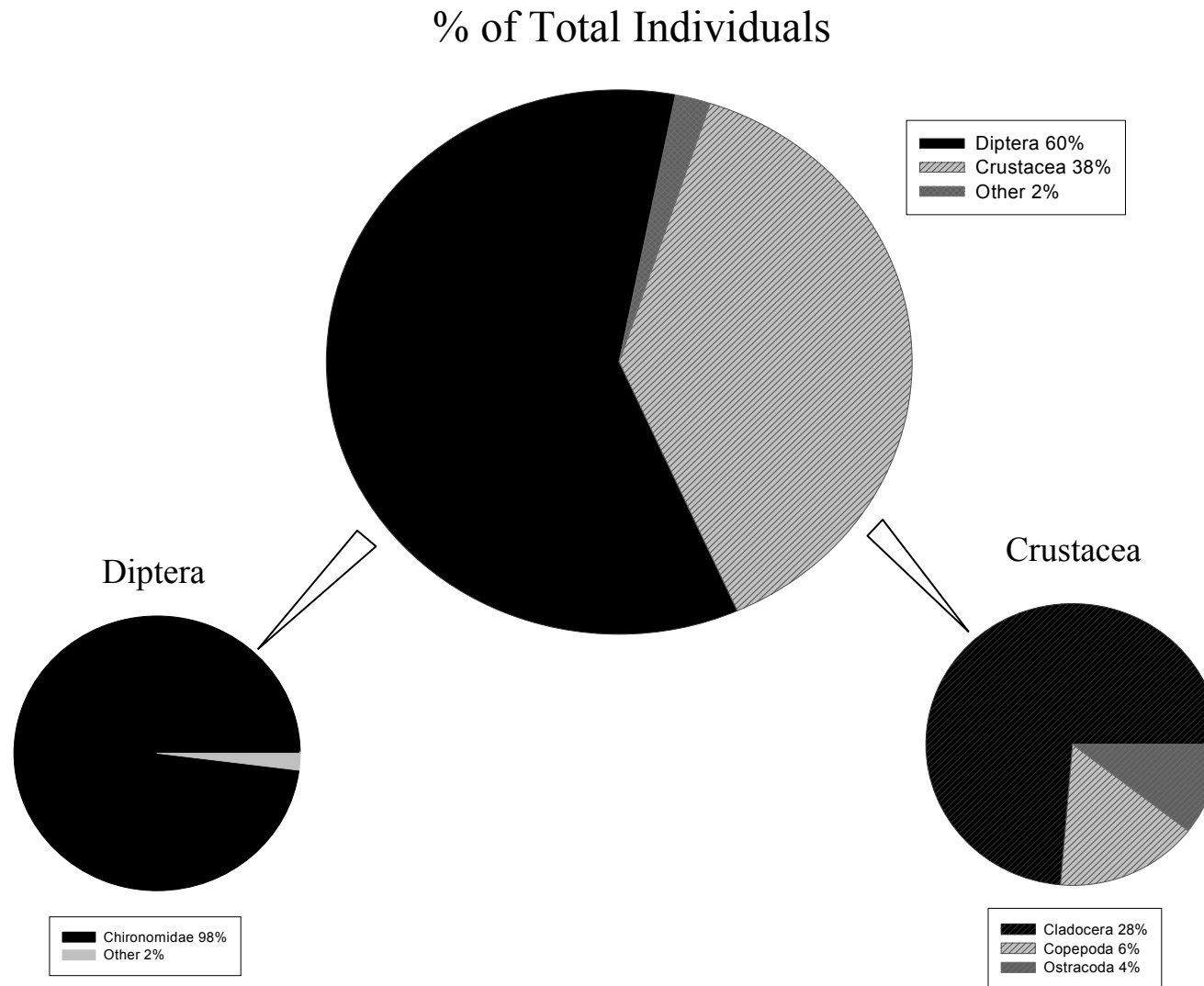


Figure 3.3. Percentage of each taxa present (of total individuals) in gastrointestinal tracts of salamander larvae.

Table 3.2. Community composition of common benthic macroinvertebrates collected by a Hess sampler in Early County, Georgia. Expressed as an average density (rounded to nearest whole number) with percentage of total organisms in parentheses.

Taxon	Date						
	Feb 2002	April 2002	June 2002	Aug 2002	Oct 2002	Dec 2002	Feb 2003
Amphipoda							
Crangonyctidae							
<i>Crangonyx sp.</i>	3 (2.18)	8 (2.41)	6 (1.59)	--	1 (0.30)	2 (0.37)	--
Coleoptera							
Elmidae							
<i>Microcylloepus (adult)</i>	1 (0.65)	10 (2.96)	5 (1.33)	1 (0.14)	2 (0.72)	1 (0.18)	1 (0.32)
<i>Stenelmis (adult)</i>	--	1 (0.20)	--	--	1 (0.30)	--	1 (0.42)
<i>Stenelmis (larvae)</i>	7 (4.58)	51 (14.98)	49 (12.98)	29 (3.87)	27 (8.38)	47 (8.62)	18 (5.70)
Decapoda	3 (2.18)	--	--	--	1 (0.30)	5 (0.92)	1 (0.42)
Diptera							
Ceratopogonidae	6 (4.14)	32 (9.36)	6 (1.46)	11 (1.42)	18 (2.59)	18 (3.30)	19 (2.85)
Chironomidae (other)	63 (41.18)	173 (51.14)	262 (69.40)	656 (88.95)	244 (75.70)	332 (60.92)	208 (65.72)
Tanypodinae	52 (33.99)	40 (11.58)	23 (6.09)	15 (2.03)	21 (6.52)	49 (9.00)	43 (13.71)
Simuliidae							
<i>Simulium sp.</i>	2 (1.31)	--	1 (0.27)	4 (0.48)	--	1 (0.18)	1 (0.32)
Tipulidae							
<i>Hexatoma sp.</i>	1 (0.65)	1 (0.30)	5 (1.19)	--	--	--	--
<i>Pseudolimmophila sp.</i>	5 (3.27)	1 (0.30)	1 (0.27)	--	--	9 (1.65)	3 (0.95)
<i>Tipula sp.</i>	3 (1.74)	1 (0.30)	--	--	--	--	1 (0.32)
Ephemeroptera							
Baetidae	--	--	1 (0.27)	1 (0.14)	1 (0.30)	16 (2.94)	--
Heptageniidae							
<i>Stenonema sp.</i>	--	1 (0.30)	2 (0.53)	1 (0.14)	2 (0.72)	18 (3.30)	8 (2.53)
Hemiptera							
Vellidae							
<i>Rhagovelia sp.</i>	--	2 (0.49)	1 (0.27)	4 (0.54)	3 (1.03)	--	--
Hydracarina	--	--	1 (0.27)	1 (0.14)	--	16 (2.94)	1 (0.32)
Odonata							
Caloptergidae							
<i>Calopteryx sp.</i>	1 (0.65)	--	1 (0.27)	--	--	--	1 (0.32)
Gomphidae							
<i>Progomphus sp.</i>	--	--	1 (0.27)	--	1 (0.30)	6 (1.10)	--
Plecoptera	2 (1.53)	6 (1.68)	--	--	--	--	1 (0.32)

Table 3.2. Cont'd.

Taxon	Date						
	Feb 2002	April 2002	June 2002	Aug 2002	Oct 2002	Dec 2002	Feb 2003
Trichoptera							
Hydropsychidae							
<i>Diplectrona sp.</i>	1 (0.65)	--	1 (0.27)	--	--	--	5 (1.48)
<i>Hydropsyche sp.</i>	--	--	1 (0.27)	--	--	--	1 (0.32)
Lepidostomatidae							
<i>Lepidostoma sp.</i>	1 (0.65)	1 (0.30)	--	--	--	--	2 (0.53)
Leptoceridae							
<i>Ceraclea sp.</i>	--	--	1 (0.27)	--	--	--	--
Odontoceridae							
<i>Psilotreta sp.</i>	--	--	4 (1.06)	1 (0.14)	--	--	--
Average no. individuals	153	338	378	738	322	545	316

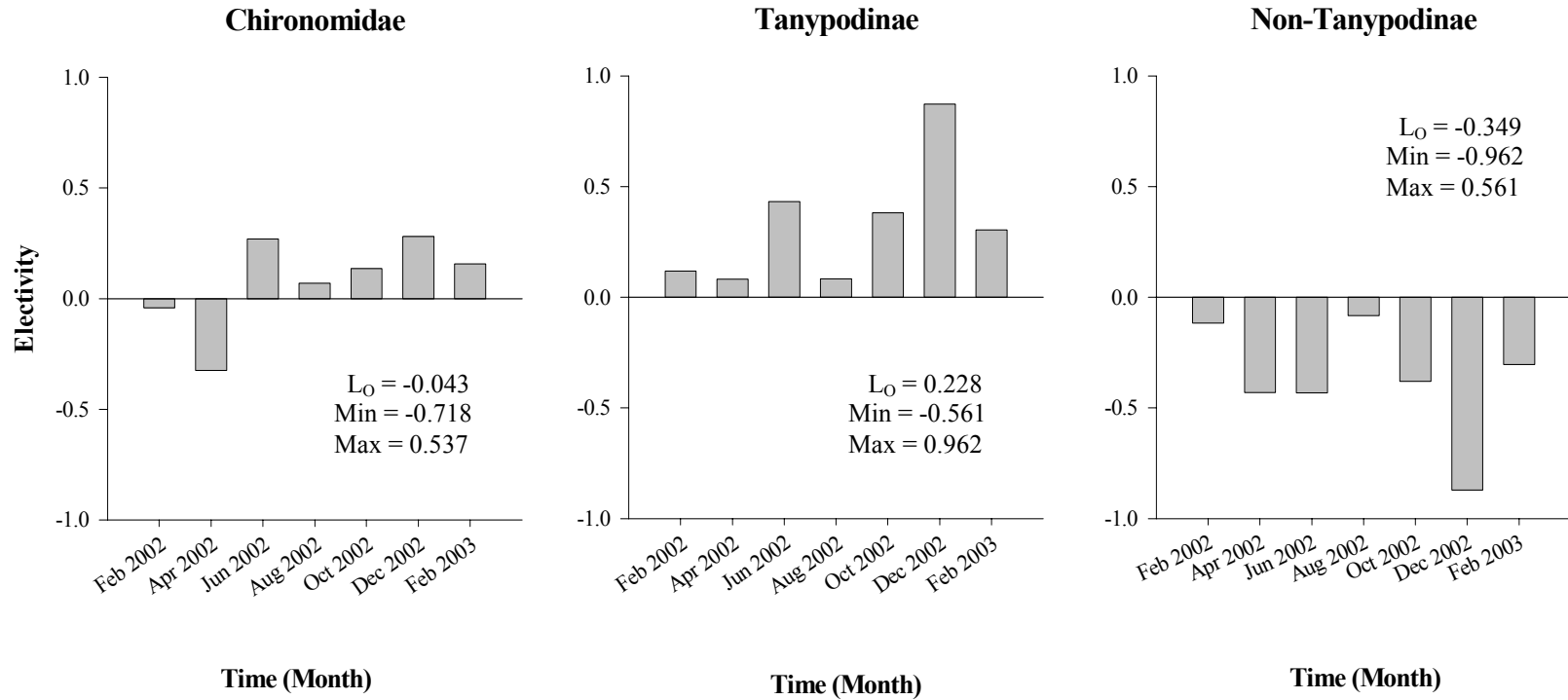


Figure 3.4. Linear electivity for Chironomidae, Tanypodinae, and Non-Tanypodinae prey consumed by larval *Eurycea cirrigera* for each sampling date. Electivity index values (L_0) are for all dates combined, and ranges of index values (reported as minimum and maximum values) are from all dates and individual salamander larvae in the study.

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CHAPTER 4
CONCLUSIONS

Anthropogenic disturbances such as those resulting from agricultural practices destabilize stream ecosystem function (Stevens and Cummins, 1999). In particular, grazing in riparian areas can have impacts on water quality through the removal of streamside vegetation, alteration of channels and banks through destabilization, compaction of soils, and deposition of wastes directly into streams. The most apparent effects are elevated nutrient loads, increased sediment transport, degradation of riparian and aquatic habitat, and increased erosion and runoff (Armour et al. 1991).

Differences in measurements (chemical, physical, biotic) between buffered (streams fenced from cattle) and unbuffered sites were apparent in this study, suggesting that sites fenced from cattle many years ago have better water quality and habitat, and that conservation buffers are benefiting these streams. Variability amongst sites and treatments also existed, with those sites in the same treatment being most similar, and disturbances from a nearby gully strongly effecting one of the unfenced sites. Rapid assessment tools for soil coverage and vertical obstruction were found useful in detecting differences amongst sites, and should be considered for monitoring programs. Ground layer coverage type, estimated across each plot using a line-transect method for soil coverage appeared to capture similar differences amongst treatments than conventional plot surveys methods. A Robel pole also proved useful in estimating the cover of vertical obstruction of the understory, and was quick in technique. Of invertebrate metrics examined in this study, percentages of burrowers, clingers, crustaceans (amphipods and decapods), EPT, EPT/Chironomidae, Dipterans, and Elmidae were found most useful for perennial streams within the Fall Line Hills District of Georgia. Amphibian abundance and diversity did not show similar positive responses between treatments for surveys within the riparian area, however salamander larvae collected within invertebrate collections did display

significant differences. A number of factors could play a role the abundance of amphibians, ranging from those controlled by the study design, such as small sample size or survey area, to variables beyond our control such as distance to nearest breeding site (for hylids) and the degree of disturbance these sites incurred previous to this study. Salamander larvae captured with the Hess sampler were most likely influenced by lower sediment levels and increased streambed heterogeneity at the fenced sites. Larval captures with a Hess sampler proved useful as a metric and tool for assessment of site differences, and should be used in correlation with other biotic indicators such as benthic macroinvertebrates. Amphibians appear as useful tools in biological monitoring, however a consortium of life history traits and responses to disturbance variables ranging from the microhabitat to landscape levels needs to be further examined.

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APPENDICES

Appendix A. Vegetation plot and subplot cover classes

I. Shrub and ground cover classes

<u>% cover</u>	<u>Class</u>
<1	1
1-5	2
6-15	3
16-25	4
26-50	5
51-75	6
76-100	7

II. Coarse woody debris (CWD) classes

<u>Diameter (cm)</u>	<u>Length (m)</u>	<u>Class</u>
5-10	<1	1
11-15	1-2	2
16-20	2-3	3
> 20	>3	4

Appendix B. EPA physical habitat scores for each stream reach. (EPA Rapid Bioassessment Protocols for use in Streams and Wadeable Rivers: Periphyton, Benthic Macroinvertebrates, and Fish, Second Edition-Form 3)

Habitat Parameter	Condition Score				
	UF-1	UF-2	UF-3	F-1	F-2
Epifaunal Substrate/ Available Cover	4	10	3	17	11
Pool Substrate Characterization	6	11	6	19	13
Pool Variability	0	0	1	14	9
Sediment Deposition	1	17	3	16	9
Channel Flow Status	6	20	10	19	15
Channel Alteration	18	19	18	19	19
Channel Sinuosity	11	9	15	18	15
Bank Stability					
Left Bank	1	8	1	8	8
Right Bank	1	4	1	8	8
Vegetative Protection					
Left Bank	2	6	2	8	8
Right Bank	1	4	2	8	8
Riparian Vegetative Zone Width					
Left Bank	7	10	9	10	10
Right Bank	7	10	9	10	10
Total Score	65	128	80	174	143

Appendix C. Mean abundance (individuals/m²) of invertebrate taxa collected with a Hess sampler, for each site.

TAXA	UF-1	UF-2	UF-3	F-1	F-2
AMPHIPODA					
Crangonyctidae					
<i>Crangonyx</i>	--	0.28	--	19.10	8.03
COLEOPTERA					
Unknown	0.28	0.28	--	--	0.55
Curculionidae	--	--	--	--	0.28
Elmidae					
<i>Ancyronyx</i>	--	0.28	--	--	--
<i>Microcyllopeus</i>	0.55	0.28	0.28	26.02	67.84
<i>Stenelmis</i>	0.83	12.18	11.35	161.13	251.94
Hydrophilidae					
<i>Hydrobiomorpha</i>	0.28	--	--	--	--
Ptilodactylidae					
<i>Ancyrtarsus</i>	--	--	0.55	1.94	0.83
Scirtidae					
<i>Prionocyphon</i>	0.28	--	--	--	--
DECAPODA (unknown)	--	0.28	--	--	0.28
<i>Cambarus</i>	--	0.55	--	1.94	1.94
<i>Procambarus</i>	--	0.28	--	1.66	1.94
DIPTERA					
Unknown	7.75	16.06	41.25	31	21.69
Ceratopogonidae	47.91	52.88	42.36	67.76	85.27
Chironomidae larvae	294.29	1331.13	1031.84	1046.79	1236.71
Tanypodinae larvae	22.70	140.37	36	132.34	174.42
Dixidae					
<i>Dixa</i>	--	--	--	0.28	--
<i>Dixella</i>	--	--	--	0.28	--
Empididae					
<i>Clinocerca</i>	--	0.28	0.55	--	--
<i>Hamerodromia</i>	0.83	--	3.32	0.83	1.11
Simuliidae					
<i>Simulium</i>	42.91	1.94	32.12	3.05	8.31

Appendix C. Cont'd

TAXA	UF-1	UF-2	UF-3	F-1	F-2
DIPTERA cont'd					
Stratiomyidae					
<i>Nemotelus</i>	--	0.28	--	--	--
Tabanidae	0.28	2.77	--	0.28	0.28
Tipulidae	1.94	0.28	--	1.11	0.55
<i>Hexatoma</i>	0.83	7.75	9.95	2.77	5.27
<i>Pseudolimnophila</i>	1.38	1.38	23.26	13.83	9.15
<i>Tipula</i>	0.28	--	0.28	4.43	4.43
EPHEMEROPTERA					
Unknown	0.28	1.11	1.11	1.38	2.21
Baetidae	--	0.83	1.66	0.28	5.83
Heptageniidae	--	--	0.83	--	--
<i>Stenonema</i>	--	3.32	7.47	42.08	16.06
Ephemeridae					
<i>Hexagenia</i>	--	8.87	--	0.28	0.28
Leptophlebiidae					
<i>Habroplebia</i>	--	--	--	--	0.83
HEMIPTERA					
Unknown	--	--	--	0.83	--
Gerridae					
<i>Trepobaks</i>	--	--	--	--	0.28
Hebridae	0.28	--	--	--	--
Vellidae	--	--	--	--	0.55
<i>Microvelia</i>	0.83	--	0.55	0.83	0.28
<i>Rhagovelia</i>	2.77	4.43	8.31	4.71	11.63
ODONATA					
Unknown	--	0.83	--	--	--
Aeshnidae					
<i>Coryphaeschna</i>	--	0.55	0.28	--	0.55
Calopterygidae					
<i>Calopteryx</i>	0.28	0.83	0.28	3.60	1.94
Cordulegastidae					
<i>Cordulsgaster</i>	--	0.28	0.28	0.28	1.38

Appendix C. Cont'd

TAXA	UF-1	UF-2	UF-3	F-1	F-2
Odonata Cont'd					
Gomphidae	--	0.55	--	0.28	--
<i>Progomphus</i>	--	7.19	0.55	3.32	3.32
MEGALOPTERA					
Unknown	--	--	--	0.28	--
Corydalidae					
<i>Chauloides</i>	--	0.28	--	--	0.28
<i>Nigronia</i>	--	0.28	--	0.28	--
MOLLUSCA	0.55	43.47	3.32	0.28	0.55
PLECOPTERA					
Unknown	--	0.28	--	3.60	0.83
Chloroperlidae	--	--	0.28	1.38	1.11
Perlidae	--	--	--	1.11	--
<i>Beloneuria</i>	--	--	--	9.71	0.83
<i>Perlesta</i>	--	--	--	0.55	--
Perlodidae	--	0.28	0.28	--	--
TRICHOPTERA					
Unknown	--	--	4.96	1.66	3.05
Hydropsychidae	--	--	1.38	1.11	0.83
<i>Cheumatopsyche</i>	--	--	0.55	0.28	--
<i>Diplectronea</i>	--	1.11	1.38	9.95	4.43
<i>Hydropsyche</i>	0.28	1.94	2.49	3.32	2.77
Hydroptilidae					
<i>Hydroptila</i>	--	--	--	--	0.83
Lepidostomatidae					
<i>Lepidostoma</i>	--	--	--	4.43	--
Leptoceridae					
<i>Ceraclea</i>	--	0.28	--	0.83	0.55
<i>Oecetis</i>	--	0.28	0.28	4.16	4.43
Molannidae					
<i>Molanna</i>	--	--	--	0.83	0.55
Odontoceridae					
<i>Psilotreta</i>	--	--	--	0.83	6.08
Psychomyiidae					
<i>Lype</i>	--	--	--	--	0.28

Appendix D. Herpetofaunal species found at study sites in Early County, Georgia, March 2002-March 2003. An ‘X’ denotes species was observed at or near the respective site.

Scientific name	Common name	UF-1	UF-2	UF-3	F-1	F-2
Frogs and Toads						
Hylidae						
<i>Acris sp.</i>	Cricket Frog				X	
<i>Hyla chrysoscelis</i>	Cope’s Gray Treefrog	X	X		X	X
<i>Hyla cinerea</i>	Green Treefrog	X	X	X	X	X
<i>Hyla gratiosa</i>	Barking Treefrog			X*		
<i>Hyla squirella</i>	Squirrel Treefrog	X	X	X	X	X
Ranidae						
<i>Rana catesbeiana</i>	American Bullfrog			X		
<i>Rana clamitans</i>	Bronze Frog	X		X		
<i>Rana sphenoccephala</i>	Southern Leopard Frog	X	X		X*	X
Salamanders						
Plethodontidae						
<i>Desmognathus apalachicola(A)</i>	Apalachicola Dusky Salamander		X		X	X
<i>Eurycea cirrigera(A)</i>	Southern Two-lined Salamander	X	X	X	X	X
<i>E.cirrigera(L)</i>	Southern Two-lined Salamander	X		X	X	X
<i>Plethodon grobmani (A)</i>	Southeastern Slimy Salamander		X	X	X	X
<i>Pseudotriton ruber(A)</i>	Red Salamander	X	X	X	X	X
<i>P.ruber (L)</i>	Red Salamander	X	X*			
Lizards						
Polychridae						
<i>Anolis carolinensis</i>	Green Anole	X	X	X	X	X
Scincidae						
<i>Eumeces fasciatus</i>	Common Five-lined Skink	X*				
<i>Scincella lateralis</i>	Little Brown Skink	X				

* Denotes species not found during designated study survey dates
(Taxonomy for reptiles and amphibians follows Crother, 2000)

A=Adult form; L= Larval form;

Appendix D. Cont'd

Scientific name	Common name	UF-1	UF-2	UF-3	F-1	F-2
Snakes						
Colubridae						
<i>Masticophis flagellum</i>	Coachwhip	X*				
<i>Nerodia fasciata</i>	Banded Water Snake				X	
<i>Nerodia sipedon</i>	Midland Water Snake			X		
<i>Storeria occipitomaculata</i>	Red-bellied Snake	X				X
Viperidae						
<i>Agkistrodon contortrix</i>	Copperhead				X	
<i>Agkistrodon piscivorus</i>	Cottonmouth		X	X		

* Denotes species not found during designated study survey dates.

Appendix E. Presence and characteristics of vascular plant species found at study sites in Early County, Georgia. An ‘X’ denotes presence of a particular species at the respective site.

Botanical name	Family	Growth Habit	Common name	U.S. nativity	Duration	UF1	UF2	UF3	F1	F2
<i>Acalypha sp.</i>	Euphorbiaceae	Forb/Herb			Annual			X		
<i>Acer rubrum</i>	Aceraceae	Tree	Red maple	Native	Perennial	X	X	X	X	X
<i>Alnus serrulata</i>	Betulaceae	Tree/Shrub	Hazel alder	Native	Perennial	X				
<i>Ambrosia artemisiifolia</i>	Asteraceae	Forb/herb	Ragweed	Native	Annual			X		X
<i>Arundinaria gigantea</i>	Poaceae	Subshrub/shrub	Giant cane	Native	Perennial	X			X	X
<i>Asimina parviflora</i>	Annonaceae	Tree/shrub	Smallflower pawpaw	Native	Perennial				X	
<i>Aster sp.</i>	Asteraceae	Forb/herb			Perennial	X		X		
<i>Bignonia capreolata</i>	Bignoniaceae	Vine	Crossvine	Native	Perennial				X	X
<i>Boehmeria cylindrica</i>	Urticaceae	Forb/herb	Smallspike false nettle	Native	Perennial					X
<i>Callicarpa americana</i>	Verbenaceae	Shrub	American beautyberry	Native	Perennial	X			X	X
<i>Carex sp.</i>	Cyperaceae	Graminoid	Sedge		Perennial	X				
<i>Carya glabra</i>	Juglandaceae	Tree	Pignut hickory	Native	Perennial	X				X
<i>Carya pallida</i>	Juglandaceae	Tree	Sand hickory	Native	Perennial				X	
<i>Celtis laevigata</i>	Ulmaceae	Tree/shrub	Sugarberry	Native	Perennial			X	X	
<i>Clematis virginiana</i>	Ranunculaceae	Vine/subshrub	Devil’s darning needles	Native	Perennial	X				X
<i>Clematis sp.</i>	Ranunculaceae	Vine/subshrub			Perennial					X
<i>Cornus florida</i>	Cornaceae	Tree/shrub	Flowering dogwood	Native	Perennial				X	X
<i>Cuphea carthagenensis</i>	Lythraceae	Forb/herb	Columbian waxweed	Introduced	A/P*	X				
<i>Cyperus sesquiflorus</i>	Cyperaceae	Graminoid	Fragrant spikesedge	Native	A/Bi/P*	X				
<i>Decumaria barbara</i>	Hydrangeaceae	Vine	Woodvamp	Native	Perennial					X
<i>Dichanthelium dichotomum</i>	Poaceae	Graminoid	Cypress panicgrass	Native	Perennial	X			X	X
<i>Dichondra carolinensis</i>	Convolvulaceae	Forb/herb	Carolina ponysfoot	Native	Perennial	X				
<i>Digitaria ciliaris</i>	Poaceae	Graminoid	Southern crabgrass	Introduced	Annual	X				
<i>Diospyros virginiana.</i>	Ebenaceae	Tree	Common persimmon	Native	Perennial				X	X
<i>Eupatorium capillifolium</i>	Asteraceae	Forb/herb	Dogfennel	Native	Perennial	X				
<i>Fagus grandifolia</i>	Fagaceae	Tree	American beech	Native	Perennial				X	X
<i>Halesia diptera</i>	Styracaceae	Tree/shrub	Two-wing silverbell	Native	Perennial				X	X
<i>Hamamelis virginiana</i>	Hamamelidaceae	Tree/shrub	American witchhazel	Native	Perennial					X
<i>Hypericum mutilum</i>	Clusiaceae	Forb/herb	Dwarf St.Johnswort	Native	A/P*	X				

* A=annual; P=perennial, Bi=Biennial

Appendix E. Cont'd

Botanical name	Family	Growth Habit	Common name	U.S. nativity	Duration	UF1	UF2	UF3	F1	F2
<i>Ilex opaca</i>	Aquifoliaceae	Tree/shrub	American holly	Native	Perennial				X	X
<i>Itea virginica</i>	Grossulariaceae	Shrub	Virginia sweetspire	Native	Perennial				X	X
<i>Juglans nigra</i>	Juglandaceae	Tree	Black walnut	Native	Perennial			X		
<i>Juncus coriaceus</i>	Juncaceae	Graminoid	Leathery rush	Native	Perennial	X				
<i>Juncus sp.</i>	Juncaceae	Graminoid			Perennial	X				
<i>Juniperus virginiana</i>	Cupressaceae	Tree	Easter cedar	Native	Perennial					X
<i>Leersia virginica</i>	Poaceae	Graminoid	Whitegrass	Native	Perennial				X	
<i>Lespedeza angustifolia</i>	Fabaceae	Forb/herb	Narrowleaf lespedeza	Native	Perennial	X				
<i>Ligustrum sinense</i>	Oleaceae	Tree/shrub	Chinese privet	Introduced	Perennial	X	X	X	X	X
<i>Liquidambar styraciflua</i>	Hammamelidaceae	Tree	Sweetgum	Native	Perennial	X	X		X	
<i>Liriodendron tulipifera</i>	Magnoliaceae	Tree	Tuliptree	Native	Perennial	X	X	X	X	X
<i>Lonicera japonica</i>	Caprifoliaceae	Vine	Japanese honeysuckle	Introduced	Perennial	X			X	
<i>Lonicera sempervirens</i>	Caprifoliaceae	Vine	Trumpet honeysuckle	Native	Perennial				X	
<i>Ludwigia decurrens</i>	Onagraceae	Forb/herb	Wingleaf primrose-willow	Native	A/P*	X				
<i>Lycopus virginicus</i>	Lamiaceae	Forb/herb	Virginia water horehound	Native	Perennial	X				
<i>Magnolia grandiflora</i>	Magnoliaceae	Tree	Southern magnolia	Native	Perennial		X			X
<i>Magnolia virginiana</i>	Magnoliaceae	Tree	Sweetbay	Native	Perennial	X		X	X	X
<i>Mitchella repens</i>	Rubiaceae	Shrub/Forb/herb	Partridgeberry	Native	Perennial					X
<i>Muhlenbergia schreberi</i>	Poaceae	Graminoid	Nimblewill	Native	Perennial	X				
<i>Nyssa biflora</i>	Nyssaceae	Tree	Swamp tupelo	Native	Perennial	X	X	X	X	X
<i>Oxydendrum arboreum</i>	Ericaceae	Tree/shrub	Sourwood	Native	Perennial			X		
<i>Phytolacca americana</i>	Phytolaccaceae	Forb/herb	American Pokeweed	Native	Perennial					X
<i>Pinus echinata</i>	Pinaceae	Tree	Shortleaf pine	Native	Perennial				X	
<i>Pinus taeda</i>	Pinaceae	Tree	Loblolly pine	Native	Perennial				X	
<i>Prunus caroliniana</i>	Rosaceae	Tree/shrub	Carolina laurelcherry	Native	Perennial				X	
<i>Prunus serotina</i>	Rosaceae	Tree/shrub	Black cherry	Native	Perennial			X	X	X
<i>Quercus alba</i>	Fagaceae	Tree	White oak	Native	Perennial			X	X	X

* A=annual; P=perennial, Bi=Biennial

Appendix E. Cont'd

Botanical name	Family	Growth Habit	Common name	U.S. nativity	Duration	UF1	UF2	UF3	F1	F2
<i>Quercus hemisphaerica</i>	Fagaceae	Tree	Darlington oak	Native	Perennial					X
<i>Quercus laurifolia</i>	Fagaceae	Tree	Laurel oak	Native	Perennial	X			X	
<i>Quercus nigra</i>	Fagaceae	Tree	Water oak	Native	Perennial		X	X		X
<i>Quercus phellos</i>	Fagaceae	Tree	Willow oak	Native	Perennial				X	X
<i>Rubus argutus</i>	Rosaceae	Shrub	Sawtooth blackberry	Native	Perennial				X	X
<i>Salix nigra</i>	Salicaceae	Tree	Black willow	Native	Perennial	X				
<i>Sassafras albidum</i>	Lauraceae	Tree/shrub	Sassafrass	Native	Perennial				X	
<i>Scleria sp.</i>	Cypereaceae	Graminoid			Perennial				X	
<i>Sedge sp.</i>	Cyperaceae	Graminoid			Perennial		X			
<i>Sideroxylon sp.</i>	Sapotaceae	Shrub			Perennial	X				
<i>Smilax bona-nox</i>	Smilacaceae	Subshrub/vine	Saw greenbrier	Native	Perennial			X		
<i>Smilax glauca</i>	Smilacaceae	Subshrub/vine	Cat greenbrier	Native	Perennial				X	X
<i>Smilax laurifolia</i>	Smilacaceae	Subshrub/vine	Laurel greenbrier	Native	Perennial					X
<i>Smilax pumila</i>	Smilacaceae	Subshrub/vine	Sarsparilla vine	Native	Perennial				X	X
<i>Smilax smallii</i>	Smilacaceae	Subshrub/vine	Lanceleaf greenbrier	Native	Perennial				X	
<i>Smilax tamnoides</i>	Smilacaceae	Subshrub/vine	Bristly greenbrier	Native	Perennial				X	X
<i>Smilax walteri</i>	Smilacaceae	Subshrub/vine	Coral greenbrier	Native	Perennial	X			X	X
<i>Solidago caesia</i>	Asteraceae	Forb/herb	Wreath goldenrod	Native	Perennial				X	
<i>Solidago sp.</i>	Asteraceae	Forb/herb			Perennial				X	X
<i>Symplocos tinctoria</i>	Symplocaceae	Tree/shrub	Common sweetleaf	Native	Perennial				X	X
<i>Thelypteris quadrangularis</i>	Thelypteridaceae	Forb/herb	Maiden fern	Native	Perennial	X				
<i>Toxicodendron radicans</i>	Anacardiaceae	Subshrub/vine	Eastern poison ivy	Native	Perennial	X	X			X
<i>Trachelospermum sp.</i>	Apocynaceae	Vine/subshrub		Native	Perennial				X	X
<i>Vaccinium sp.</i>	Ericaceae	Subshrub/shrub		Native	Perennial				X	
<i>Viburnum nudum</i>	Caprifoliaceae	Tree/shrub	Possumhaw	Native	Perennial	X				
<i>Viola sp.</i>	Violaceae	Forb/herb			Perennial		X			
<i>Vitis rotundifolia</i>	Vitaceae	Vine	Muscadine	Native	Perennial				X	X
<i>Woodwardia areolata</i>	Blechnaceae	Forb/herb	Netted chainfern	Native	Perennial	X			X	X

A=annual; P=perennial, Bi=Biennial

Appendix F. Images of amphibian species found at study sites in Early County Georgia.

I. Treefrogs (Hylidae)

Cope's Gray Treefrog, *Hyla chrysoscelis*



Green Treefrog, *Hyla cinerea*



Barking Treefrog, *Hyla gratiosa*

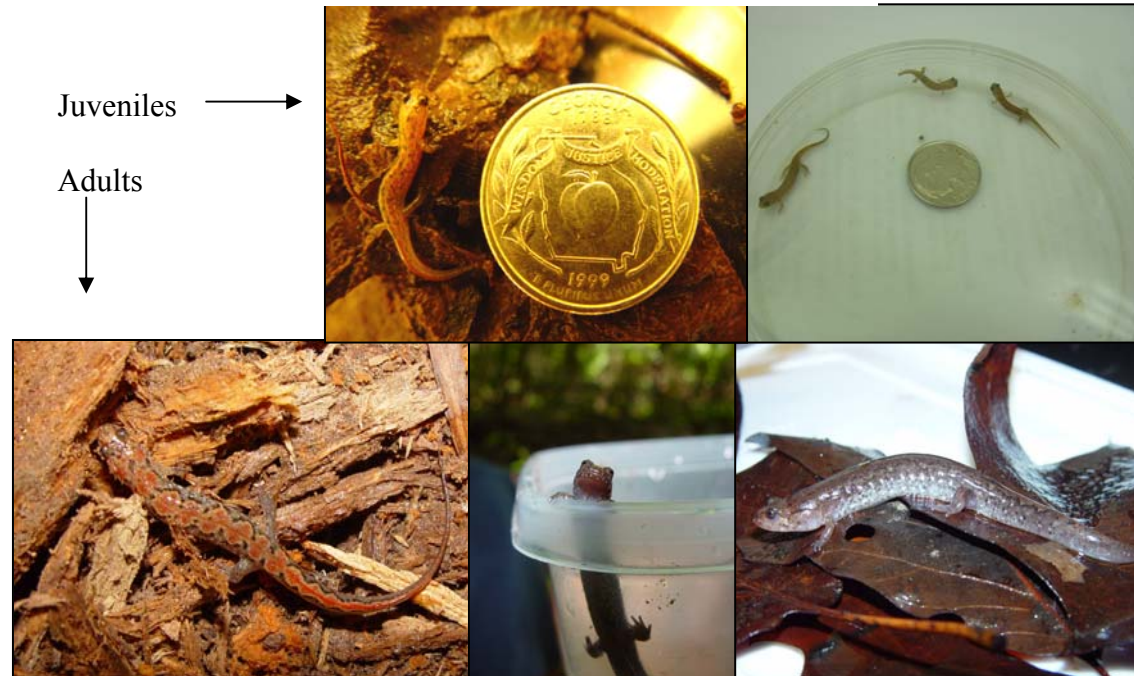


Squirrel Treefrog, *Hyla squirella*



II. Salamanders (all Plethodontidae)

Apalachicola Dusky Salamander, *Desmognathus apalachicola*



Southern Two-lined Salamander, *Eurycea cirrigera*



Adult

Larva

Southeastern Slimy Salamander, *Plethodon grobmani*



Red Salamander, *Pseudotriton ruber*

Adults

Larva

