

LIFE HISTORY OF THE ENDANGERED SHINY PIGTOE  
PEARLY MUSSEL, FUSCONAIA EDGARIANA,  
IN THE NORTH FORK HOLSTON RIVER, VIRGINIA

by

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TABLE OF CONTENTS

	<u>Page</u>
ACKNOWLEDGEMENTS.....	ii
TABLE OF CONTENTS.....	iv
LIST OF FIGURES.....	vi
LIST OF TABLES.....	vii
LIST OF APPENDIX TABLES.....	ix
INTRODUCTION.....	1
Decline of Mussels.....	2
Endangered Species.....	3
Life History of Mussels.....	4
Age and Growth of Mussels.....	7
Shiny Pigtoe Pearly Mussel.....	8
Study Objectives.....	10
MATERIALS AND METHODS.....	11
Study Site.....	11
Mussel Composition.....	17
Stream Substrate.....	23
Drift of Glochidia.....	24
Differentiation of Glochidia.....	26
Natural Infestations on Fish.....	28
Laboratory Fish Infestations.....	29
Age and Growth.....	32
RESULTS.....	35
Mussel Assemblage at North Holston Ford.....	35

Substrate Analysis at North Holston Ford.....	39
Description of Glochidia.....	43
Stream Drift.....	45
Natural Infestations on Fish.....	55
Laboratory Induced Infestations.....	63
Age and Growth.....	69
DISCUSSION.....	74
Mussel Assemblage .....	72
Substrate Composition.....	78
Stream Drift.....	83
Differentiation of Glochidia.....	88
Determination of Fish Host.....	90
Natural Infestations on Fish.....	90
Laboratory Induced Infestations.....	96
Age and Growth.....	106
CONCLUSIONS.....	111
LITERATURE CITED.....	112
APPENDIX .....	117
VITA.....	120
ABSTRACT	

LIST OF FIGURES

<u>Figure</u>	<u>Page</u>
1 Map of southwest Virginia showing study site location on the North Fork Holston River.....	12
2 Diagram of the study site showing sampling sections, drift net stations, habitat types, and islands....	16
3 Weekly range of water temperatures at North Holston Ford from March 1981 to July 1982.....	19
4 Map of mussel densities (no./m <sup>2</sup> ) from quadrat samples at North Holston Ford, with low (0-5), moderate(6-10), and high(10+) densities indicated...	41
5 Mean densities of glochidia in drift samples from North Holston Ford for left, center, and right sampling locations, 1981-1982.....	46
6 Mean densities of glochidia of <u>F. edgariana</u> , <u>L. dolabelloides</u> , and <u>P. oviforme</u> in drift samples from North Holston Ford, 1981-1982.....	48
7 Mean observed (*) and predicted (') length-at-annuli for <u>F. edgariana</u> from North Holston Ford.....	72

LIST OF TABLES

<u>Table</u>	<u>Page</u>
1 Mussel species collected at North Holston Ford from 1978 to 1983.....	13
2 Fish species collected at North Holston Ford from 1981 to 1982.....	14
3 Mean and range of monthly water quality parameters at North Holston Ford from January 1981 to March 1982.....	18
4 Discharge data from USGS gaging station on the North Fork Holston River (NFHR 85.0) June 1981-June 1982.....	20
5 Density and population estimates (No./2930 m <sup>2</sup> ) of mussel species at North Holston Ford, based on sixty 0.5 m <sup>2</sup> quadrat samples.....	36
6 Percent composition of surface substrate at North Holston Ford, based on thirty 0.5m <sup>2</sup> quadrats.....	40
7 Dimensions of mature glochidia of <u>Fusconaia edgariana</u> , <u>F. barnesiana</u> , <u>Lexingtonia dolabelloides</u> , and <u>Pleurobema oviforme</u> .....	44
8 Number and density (No. x 10/m <sup>3</sup> ) of glochidia in drift samples from North Holston Ford, June-August 1981 and June-September 1982 .....	49
9 Mean number of viable glochidia and unfertilized eggs per conglomerate (N) for <u>Fusconaia edgariana</u> , <u>F. barnesiana</u> , <u>Pleurobema oviforme</u> , and <u>Lexingtonia dolabelloides</u> .....	54
10 Abundance and incidence of glochidial infestations on cyprinid species at North Holston Ford, June 1981-June 1982.....	56
11 Number and percent of fish infested with glochidia of <u>Fusconaia edgariana</u> , <u>F. barnesiana</u> , <u>Pleurobema oviforme</u> , and <u>Lexingtonia dolabelloides</u> at North Holston Ford, June-August 1981 and June 1982.....	58

<u>Table</u>	<u>Page</u>
12 Degree of infestation by glochidia on cyprinids at North Holston Ford, June-August 1981 and June 1982.....	61
13 Seasonal incidence of infestation by glochidia on cyprinid species at North Holston Ford, June-August 1981 and June 1982.....	64
14 Fish species artificially infested with glochidia of <u>Fusconaia edgariana</u> , July 1981.....	66
15 Cyprinid species artificially infested with glochidia of <u>Fusconaia edgariana</u> , July-August 1982.....	67
16 Observed and predicted lengths-at-annuli (mm) for <u>Fusconaia edgariana</u> from North Holston Ford, 1981-1982.....	70
17 A comparison of ages for <u>Fusconaia edgariana</u> as determined by external growth rings and a thin section technique.....	72

LIST OF APPENDIX TABLES

<u>TABLE</u>		<u>PAGE</u>
1	Water quality parameters reported by TVA at NFHRM 85.9.....	117
2	Length measurements of all mussel species taken in 60 quadrat samples at North Holston Ford.....	118

## INTRODUCTION

North America has the richest freshwater mussel (naiad) fauna in the world (Stansbery 1979). The fast flowing riffle-pool reaches of the upper Cumberland and Tennessee Rivers contain the greatest diversity of mussels in the United States (van der Schalie and van der Schalie 1950). The 45 mussel species endemic to this region are collectively termed the Cumberlandian fauna (Ortmann 1924).

The contribution of mussels to freshwater ecosystems is often underestimated. As filter feeders, freshwater mussels convert suspended particulate matter into pseudofeces readily utilized by fish and stream invertebrates (Fuller 1979, Perry 1979). Filter feeding also acts to cleanse the water (Stansbery 1979).

Mussels have been used as indicators of stream health (Imlay 1977). Their wide distribution and sensitivity to pollution has led to the use of mussels as monitors of water quality (Bedford et al. 1968, Fike and Tubb 1972, Foster and Bates 1978). Declining water quality has resulted in declining mussel populations and the extinction of some species. Pollutants account for some of this

decline (Cairns et al. 1971), but alteration and destruction of mussel habitat has been the major contributor to the loss of mussel populations during this century (van der Schalie and Locke 1941, Fuller 1979, Havlik 1979).

### Decline of Mussels

The impounding, dredging, and channelizing of rivers with diverse freshwater mussel faunas have been the most damaging activities to mussel resources (Fuller 1979). Impoundment of mussel habitats has significantly changed species density and diversity (Bates 1962). Impoundments reduce water velocity which in turn decreases currents necessary for feeding and respiration of freshwater mussels (Hynes 1970). Increased siltation, as a result of reduced velocity, impairs feeding, respiration, reproduction, and ultimately survival of mussels (Williams 1969, Fuller 1974). Stream channelization and dredging contribute to the decline of mussels by disrupting and changing substrate composition, and physically dislodging mussels (Stein 1972, Yokley 1976). These activities also eliminate lotic fish species which may serve as hosts to many mussel species (Fuller 1974).

High species diversity and degree of endemism in the Cumberlandian region makes this geographic area of primary concern in the preservation of endangered species (Dennis 1977). These mussel populations have suffered the greatest losses from habitat modification and degraded water quality (Stansbery 1979). Of the 63 mussel species recorded by Ortman (1925) on the Tennessee River prior to the Wilson Dam impoundment, only 30 species have been found in recent surveys (Stansbery 1964). The greatest loss in diversity at the site was in the Cumberlandian mussels, with all but two of the original 22 species eliminated by construction of the dam. Stansbery (1979) noted that all Cumberlandian species are in danger of extinction due to declining water quality from agricultural, municipal, and industrial pollution, and loss of habitat from further dam building. At least 14 mussel species of the Cumberland Plateau are known to be extinct (Stansbery 1979).

#### Endangered Species

Widespread recognition of habitat loss and growing appreciation for the value of species diversity eventually lead to the passage of the Endangered Species Act of 1973 [P.L. 93-205]. The purpose of this act was to conserve

species, preserve habitats, and promote long range planning to ensure the continued existence of species (Bean 1977). The Endangered Species Act and subsequent amendments provide a means by which species, such as rare mussels, are granted protection and given status under the law when listed as endangered or threatened. Such species are protected by regulations which limit activities that could jeopardize their survival or adversely affect mussel habitats (Bean 1977).

In 1973, there were 23 mussel species listed federally as endangered and 58 others were proposed for listing (Imley 1977). At present, there are 22 species of freshwater mussels within the United States listed as endangered by the Department of the Interior. Fourteen of the federally listed species are Cumberlandian mussels. Nine of these occur in Virginia (Dennis 1977), including the shiny pigtoe pearly mussel, Fusconaia edgariana, listed as endangered on June 14, 1976 (Federal Register 41:24062-24067).

#### Life History of Mussels

Long range planning for the recovery of endangered mussel species is limited by lack of biological

information. Although the Cumberlandian mussel assemblage has been cataloged for nearly a century, only recently have studies been undertaken to determine life histories of the unique mussel fauna of this region (Zale 1980, Weaver 1981). No life history studies have been published on any of the endangered Cumberlandian species.

Ortmann (1911) briefly described the reproductive processes of many freshwater mussel species and categorized them as either long-term or short-term breeders. Fertilization occurs in the spring in short-term breeders, and glochidia are released in late spring or summer. Long-term breeders overwinter their glochidia, which begin develop in late summer or fall, and are released the next spring or summer. Zale (1980) noted that some long-term breeders also released glochidia in fall and early winter. Glochidia can develop separately within the gill marsupia or form a mass of glochidia that is encased in a matrix, forming an ovisac. The ovisacs, or conglutinates, are characteristic of short-term breeders. The size and shape of these conglutinal masses has become a taxonomically important species character (Stein 1971).

Leydig discovered in 1866 that glochidia require a period of encystment on the gills of fish to complete their life cycle (Howard 1922). Glochidia released from gravid

females, in conglomerates or singly, contact fish either passively through drift or actively by feeding fish (Weaver 1980). Upon contact, glochidia attach and encyst on fins or gill filaments. The duration of encystment depends on the fish and mussel species, water temperature, and other environmental and biological factors (Stein 1971). If glochidia attach to a suitable host fish, they undergo metamorphosis to the juvenile stage. If the fish species is unsuitable, glochidia will be sloughed off within a short period of time. Encystment is apparently necessary for all freshwater mussels in North America (Bingham 1968).

The few life history descriptions of North American freshwater mussels have focused on the reproductive biology and identification of the critical fish host(s) species (Lefevre and Curtis 1910, Surber 1912, Coker et al. 1921, Matteson 1948). Identification of apparent host fish species had been determined by observation of natural glochidial infestations (Tedla and Fernando 1969, Stern and Fedler 1978, Zale 1980, Weaver 1981), as well as by induced infestations (Matteson 1948, Stein 1968, Yokley 1972, Smith 1976, Zale 1980, Weaver 1981).

The critical role of a glochidial fish host in the success or failure of a mussel species is exemplified by Fusconaia ebena. Construction of dams on the Mississippi

River in the 1930's blocked upstream migration of its host fish, the skipjack herring (Alosa chrysochloris). Unable to complete its reproductive cycle, E. ebena is proceeding toward extinction in the upper Mississippi River (Fuller 1979). Had knowledge of the host fish been available, loss of this once commercially preferred mussel species may have been prevented.

#### Age and Growth of Mussels

Determination of age and growth characteristics of mussel species is essential to understanding the population dynamics and cohort composition of a population. Of all the freshwater invertebrates, mollusks have the longest natural life spans (Stein 1971). Fifteen and twenty-year-old mussels are frequently encountered for many species (Fuller 1979), and Moyer (1984) found specimens with maximum ages that ranged from 22 to 56 years, depending on the species.

Age determination by counting external rings on the shells is the most common aging technique used on freshwater mussels (Coker et al. 1921). Although external tabulation is not always reliable, it is the most appropriate aging technique for live specimens (Imlay

1977). Thin-sectioning of shells, a technique used for aging marine mollusks, has recently been investigated for use on freshwater mussels. This technique yields the most accurate values of age, especially for older mussels, but cannot be used on live mussels (Moyer 1984). To assist in management and recovery of endangered mussels, the age and growth patterns of the species need to be investigated.

#### Shiny Pigtoe Pearly Mussel

The shiny pigtoe pearly mussel was described by Lea (1840) as Unio edgarianus from the Holston River in Tennessee. A description by Conrad (1834) of Unio cor from the Elk and Flint Rivers of Alabama is believed to be the same species. Ortmann (1918) described Fusconaia edgariana analoga as a compressed headwater form of the species from the Clinch River, Virginia. A detailed taxonomic description of this species is found in the Recovery Plan for F. edgariana (Neves 1983). Color photographs of the shiny pigtoe can be found in Endangered and Threatened Wildlife of the Chesapeake Bay: Delaware, Maryland, and Virginia (White 1982).

Ortmann (1921) classified the shiny pigtoe as a short-term breeding mussel in the subfamily Unioninae. More

recent taxonomic considerations have placed the genus Fusconaia in the subfamily Ambleminae, based on the use of all four gills as marsupia (Burch 1975). The subfamily Unioninae is considered to include those short-term breeders that use only the outer gills as marsupia, such as the genera Pleurobema and Lexingtonia.

Taxonomic descriptions and historical distribution records coupled with recent surveys comprise the majority of information available on this species (Neves 1983). The only biological information known for the shiny pigtoe pearly mussel was reported by Ortmann (1921), who collected gravid females in May and July. He described it as a short-term breeder, noting that all four gills served as marsupia. He also noted the gonads were principally crimson in color (Ortmann 1921).

The shiny pigtoe is considered a riffle species. It is usually found in shoals of stable substrate in moderate to fast flowing streams (Bogan and Parmalee 1983). In Virginia, Fusconaia edgariana is the most widely distributed endangered mussel, occurring in at least 30 sites in the Clinch, Powell, and North Fork Holston Rivers of southwestern Virginia (Stansbery 1972, Stansbery and Clench 1974, Neves et al. 1980). Recent surveys indicate that a large population of shiny pigtoes occurs at North

Holston Ford near McCrady, Virginia. Interest in the recovery of endangered mussel species lead to the selection of this site to conduct studies on the life history of the shiny pigtoe pearly mussel. The goal of this research was to determine the life history of a population of Fusconaia edgariana in the upper North Fork Holston River, at North Holston Ford in southwest Virginia.

Specific objectives at the North Holston Ford study site were:

1. To describe the reproductive cycle of the shiny pigtoe.
2. To determine the required fish host(s) of the shiny pigtoe.
3. To determine growth rate, age composition, density, and preferred habitat of the shiny pigtoe at this site.

## MATERIALS AND METHODS

### STUDY SITE

The North Fork of the Holston River (NFHR) is a fourth-order stream which drains an area of approximately 1888km<sup>2</sup> (730mi<sup>2</sup>) in Bland, Smyth, and Washington Counties of southwestern Virginia (Ross and Carico 1963). The study site at North Holston Ford [36 55'N, 81 40'W] was located at river mile 86.9 in Washington County upstream of Saltville on state route 42 near McCrady, Virginia (Figure 1). Recent surveys indicated that the faunal assemblage at this location consisted of 14 species of mussels (Table 1) and 41 species of fish (Table 2) (Widlak 1982). Although the shiny pigtoe occurs in the Clinch and Powell Rivers, as well as other portions of the upper North Fork Holston River, it appears to be most abundant at North Holston Ford, contributing slightly less than 5% of the mussel assemblage (Neves et al. 1980). This location was chosen as the study site because of its accessibility and the relative abundance of Fusconaia edgariana.

The study section consisted of a 100-meter section of river upstream from North Holson Ford, with an additional 250-meter section downstream for fish collections (Figure 2). The 100 m study reach encompassed an extensive riffle

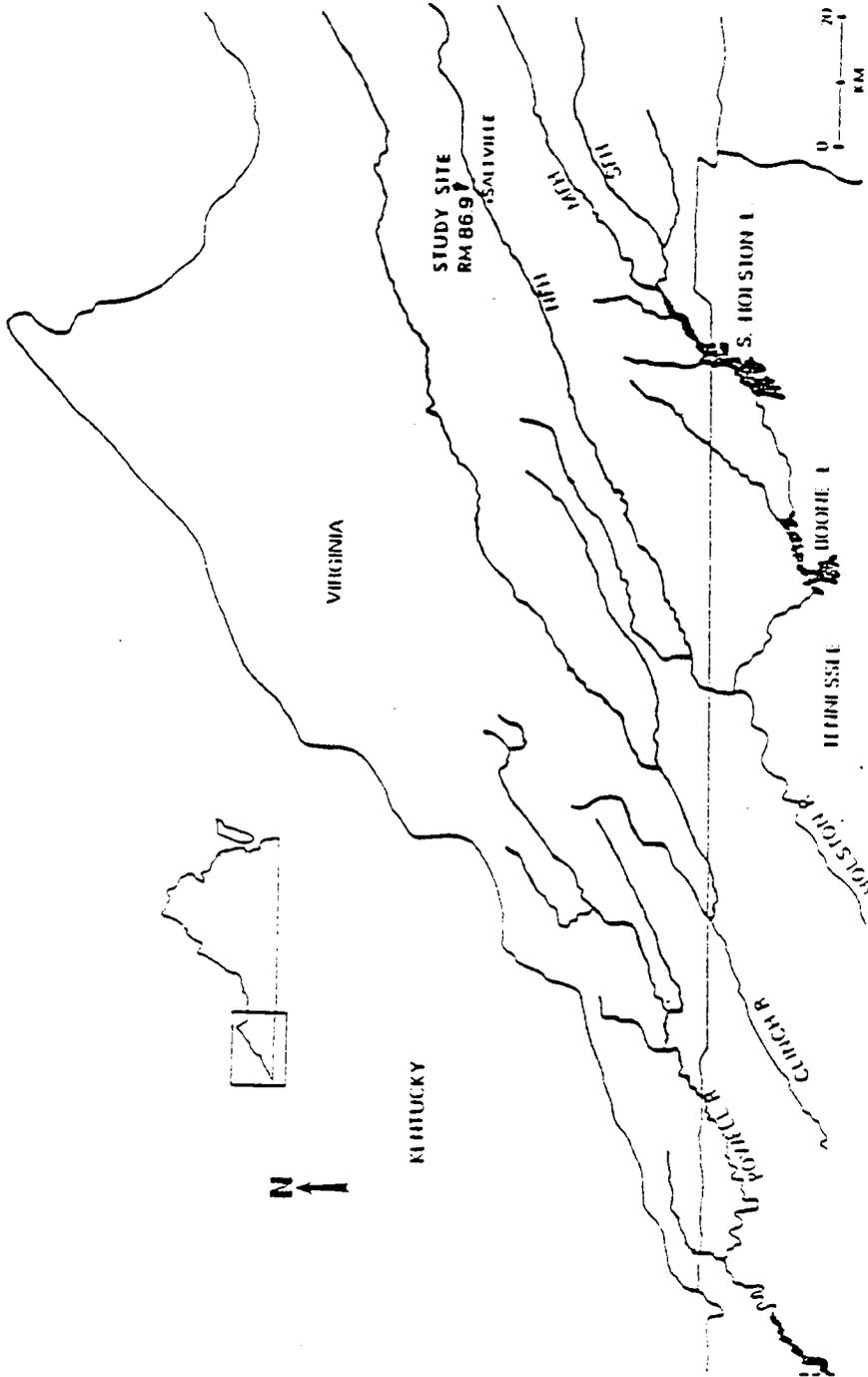


Figure 1. Map of southwest Virginia showing study site location on the North Fork Holston River.

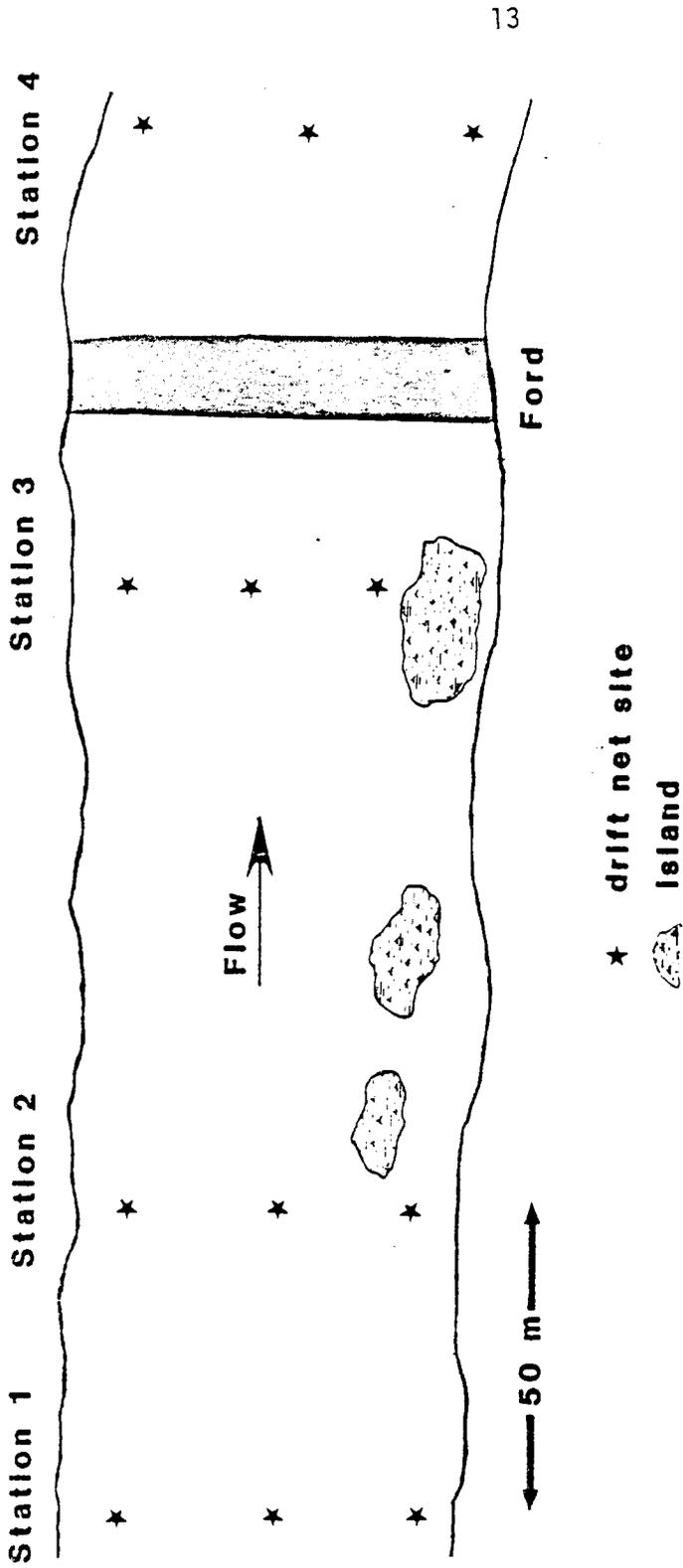


Figure 2. Diagram of the study site showing sampling section and drift net locations.

Table 1. Mussel species collected at North Holston Ford  
from June 1981 to June 1982.

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Anodontinae

- Alasmidonta marginata (Say 1819)  
Alasmidonta minor (Lea 1845)  
Lasmigona costata (Rafinesque 1820)

Ambleminae

- Fusconaia barnesiana (Lea 1838)  
Fusconaia edgariana (Lea 1840)

Unioninae

- Lexingtonia dolabelloides (Lea 1840)  
Pleurobema oviforme (Conrad 1834)

Lampsilinae

- Actinonaias pectorosa (Conrad 1834)  
Lampsilis fasciola (Rafinesque 1820)  
Lampsilis ovata (Say 1877)  
Medionidus conradicus (Lea 1834)  
Ptychobranhus fasciolaris (Rafinesque 1820)  
Ptychobranhus subtentum (Say 1824)  
Villosa nebulosa (Conrad 1834)  
Villosa yanuxemi (Lea 1834)  
Toxolasma lividus (Rafinesque 1831)
-

Table 2. Fish species collected at North Holston Ford  
from June 1981 to June 1982.

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CATOSTOMIDAE

<u>Catostomus commersoni</u>	White sucker
<u>Hypentelium nigricans</u>	Northern hogsucker
<u>Moxostoma erythrurum</u>	Golden redhorse

CENTRARCHIDAE

<u>Ambloplites rupestris</u>	Rock bass
<u>Lepomis auritus</u>	Redbreast sunfish
<u>Lepomis macrochirus</u>	Bluegill
<u>Lepomis megalotis</u>	Longear sunfish
<u>Micropterus dolomieu</u>	Smallmouth bass
<u>Micropterus salmoides</u>	Largemouth bass
<u>Pomoxis nigromaculatus</u>	Black crappie

COTTIDAE

<u>Cottus baileyi</u>	Black sculpin
<u>Cottus carolinae</u>	Banded sculpin

CYPRINIDAE

<u>Campostoma anomalum</u>	Stoneroller
<u>Hyboopsis dissimilis</u>	Streamline chub
<u>Nocomis micropogon</u>	River chub
<u>Notropis ariommus</u>	Popeye shiner
<u>Notropis coccogenis</u>	Warpaint shiner
<u>Notropis cornutus</u>	Common shiner
<u>Notropis galacturus</u>	Whitetail shiner
<u>Notropis leuciodus</u>	Tennessee shiner
<u>Notropis photogenus</u>	Silver shiner
<u>Notropis rubellus</u>	Rosyface shiner
<u>Notropis rubricroceus</u>	Saffron shiner
<u>Notropis spectrunculus</u>	Mirror shiner
<u>Notropis telescopus</u>	Telescope shiner
<u>Phenacobius uranops</u>	Stargazing minnow
<u>Rhinichthys atratulus</u>	Blacknose dace
<u>Semotilus atromaculatus</u>	Creek chub

Table 2 (continued)

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## ICTALURIDAE

<u>Ictalurus natalis</u>	Yellow bullhead
<u>Ictalurus nebulosus</u>	Brown bullhead
<u>Noturus insignis</u>	Margined madtom

## PERCIDAE

<u>Etheostoma blennioides</u>	Greenside darter
<u>Etheostoma camurum</u>	Bluebreast darter
<u>Etheostoma maculatum</u>	Spotted darter
<u>Etheostoma rufilineatum</u>	Redline darter
<u>Etheostoma simoterum</u>	Tennessee snubnose darter
<u>Etheostoma zonale</u>	Banded darter
<u>Percina aurantiaca</u>	Tangerine darter
<u>Percina caprodes</u>	Logperch
<u>Percina macrocephala</u>	Longhead darter

## PETROMYZONTIDAE

<u>Ichthyomyzon bdellium</u>	Ohio lamprey
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of moderate depth and current, and averaged 29.3 meters in width. Islands were present seasonally along the left ascending bank, but were inundated during high flow conditions. Substrate composition was predominantly coarse gravel and cobbles, with some boulders. Sand was common around the islands and along the river banks.

Water quality was measured at the site from June 1981 to June 1982 using a Hach (DR EL/1) water chemistry kit. Dissolved oxygen, pH, conductivity, alkalinity, hardness, and turbidity were monitored monthly (Table 3). Water chemistry analysis revealed the river water was moderate in hardness and had some buffering capacity. Dissolved oxygen and pH levels were good, and conductivity and turbidity were relatively low. Water temperature was monitored daily during the study period using a continuous recording thermograph (Ryan Instrument Co, Kirkland, Washington) (Figure 3). Temperature extremes were 0.5 C and 29 C, but averaged 24 C in the summer, and 3 C in the winter. Discharge information was obtained from a USGS gauging station approximately one mile downstream (Table 4). Detailed water quality data involving 41 variables were compiled by Poppe (1982) at NFHRM 85.2 (Appendix Table 1), approximately 1.5 river miles downstream of the study site.

#### MUSSEL COMPOSITION

Table 3. Mean and range of monthly water quality parameters at North Holston Ford from January 1981 to March 1982.

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Parameter	Mean	(Range)
Alkalinity (mg/l)	116	(90-175)
Conductivity (umhos)	143	(81-218)
DO (mg/l)	9	(7-12)
Hardness (mg/l)	130	(90-175)
pH	7.1	(6.6-8.3)
Turbidity (FTU)	14	(81-218)

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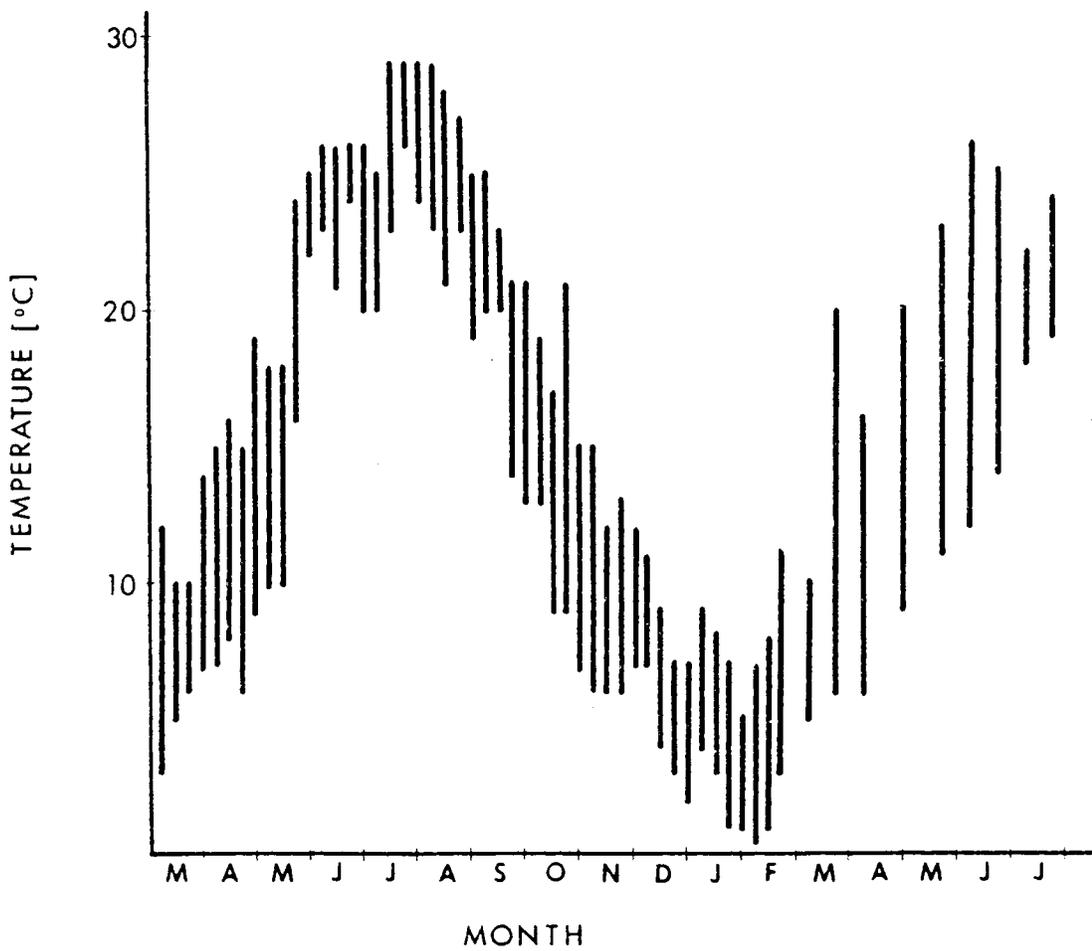


Figure 3. Weekly range of water temperatures at North Holston Ford from March 1981 to July 1982.

Table 4. Discharge data from the USGS gauging station on the North Fork Holston River (NEHRM 85.0), June 1981-June 1982.

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Month	Mean (cfs)	Maximum (cfs)	Minimum (cfs)
June 1981	385.0	1210	84
July	81.1	161	53
August	50.7	114	33
September	83.8	217	35
October	58.7	205	37
November	59.7	103	44
December	217.0	1090	63
January 1982	678.0	3490	225
February	915.0	2820	308
March	677.0	1710	249
April	214.0	507	121
May	125.3	259	83
June	380.3	1240	100

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Sixty 0.5m<sup>2</sup> quadrats were taken at the study site to determine mussel species composition, abundance, and density. A stratified transect sampling scheme was implemented to represent the diversity of habitat available to mussels. Six transects were established at 10 m intervals across the length of the study section. Ten random locations were chosen for quadrats (0.5m<sup>2</sup>) along each transect. Quadrats were sampled by removal and examination of the substrate to a depth of approximately 10 centimeters. All mussels collected in a quadrat were retained in muslin bags for later species identification.

Total length (greatest antero-posterior dimension) of each specimen was measured with vernier calipers. Gender was recorded if the species was sexually dimorphic, and water depth and substrate composition at each location were recorded. Any specimens of Fusconaia edgariana collected in the quadrats were retained for further study, and all other mussel species were returned to the sample location after replacement of the substrate. Sizes and numbers of each species were tallied, and mean density was computed. Population abundances were estimated by extrapolation of quadrat densities to total area of the study site. Total area of the site was computed by averaging midsummer stream widths measured at 10m intervals along the river section.

In addition to the quadrats, separate collections of

live F. edgariana were made throughout the year by handpicking and use of water scopes. All specimens were placed in muslin bags and held for further examination. Total width (greatest dorso-ventral dimension), total length, and age were determined for live specimens from measurement of the valves. Since sexual dimorphism is not exhibited in the shells of F. edgariana, gender of living specimens was determined when possible by visual examination of the gills. Internal parts were viewed by inserting reverse-pull pliers between the valve edges and opening them slightly so as not to harm the mussel. Gravid females were identified by intensely pink and swollen marsupial gills. Extent of gravidity was judged by the degree of distendedness of gills used as marsupia. Normal respiratory gills appeared flat and opaque in nongravid females and males. All mussels were categorized as either gravid females or nongravid individuals. Males could not be sexed by visual inspection of live specimens.

Gravid females were transported in muslin bags on ice to the laboratory for use in fish host infestations. All other F. edgariana collected at the study site were placed in the substrate under one meter square exclosures of one inch mesh hardware cloth. The exclosures permitted normal water flow, prevented muskrat predation, and allowed periodic monitoring of mussels. Observations indicated

that transplanted mussels did not appear to be affected by being placed under the exclosures. Gravid individuals were encountered only two months of the year, thereby limiting gender identification to those months. Individuals collected at other times were measured and placed under the exclosures for subsequent examination.

#### STREAM SUBSTRATE

A total of 50 substrate samples was collected in the 100 meter study section at North Holston Ford. Thirty 0.25 m<sup>2</sup> samples were taken using half of a 0.5m<sup>2</sup> quadrat sampler. A grid of six transects was established at 10 m intervals along the study reach, and five samples per transect were collected at equally-spaced intervals. Substrate within these samples was sorted through a series of U. S. Standard sieves or metal frames for the larger components. Five particle size classes were established, as modified from Cummins (1962); silt (less than 0.125mm), sand (0.125 to 2.0mm) gravel (2.0 to 16.0mm), pebble (16 to 64mm), cobble (64 to 256mm), and boulder (larger than 256mm). The percent composition of each substrate sample was determined by weight of each particle size class.

Twenty qualitative estimates of substrate composition were conducted at intervals between quantitative samples.

These estimates were based on percent composition of each substrate type as a portion of total surface area examined. A substrate map of the site was constructed using both quantitative and qualitative measurements. Percent composition of the substrate was indicated, and areas of similar composition were noted on the map.

Substrate composition was analyzed according to percent composition of each of the six substrate categories; only quantitative samples were used for this analysis. Differences in substrate composition were compared to mussel diversity and density from quadrat samples.

#### DRIIFT OF GLOCHIDIA

Occurrence and density of glochidia from the four species of short-term breeders (Ambleminae and Unioninae) at the study site were determined. Stream drift was sampled to determine the period of glochidial release by gravid females of these species. Drift nets consisted of square (30 x 30cm or 45 x 45cm) metal frames covered with 130u mesh netting and removable cod ends. Nets were placed on the river bottom and secured with metal rods. Time of drift collection and water depth at the mouth of each net were recorded. Water velocity at time of set was measured with a Gurley pygmy flow meter (Teledyne Gurley Inc., Troy,

New York) positioned midway in the water column at the mouth of the net.

Three nets were set between mid-morning and mid-afternoon for 2 hours at each of four stations to represent the left, center, and right ascending sides of the river. Station 1 was at the top of the study section, Stations 2, 3, and 4 were located at 50, 150, and 250 meters downstream of Station 1, respectively (Figure 2). During June, July and August of 1981, samples were collected weekly at Stations 2 and 3 and bimonthly at Stations 1 and 4. All stations were sampled monthly from September 1981 through April 1982 and bimonthly from May 1982 through August 1982.

Drift samples were washed from the nets into jars and preserved in 10% buffered formalin. Rose bengal was added as a protein stain to facilitate sorting in the laboratory. Prior to examination, the samples were filtered through a 0.5mm mesh screen to remove large particulate matter. Samples were then filtered through a 130u mesh screen to remove silt and other fine sediment. Glochidia and other material retained on the screen were then backwashed into a jar. Aliquots of the drift sample were placed in a gridded petri dish and examined under a dissecting microscope (30X). Glochidia were counted and transferred, with a capillary tube, to vials according to taxonomic subfamily. The process was repeated until the entire sample was

examined.

Diel periodicity of glochidial release was examined at the study site for a 24-hour period from 28 to 29 July, 1982. Nets were set at four hour intervals, for one hour duration, along the left ascending side of the stream at Stations 2 and 3. These locations were chosen due to the high densities and diversity of mussels immediately upstream. Samples were processed following the same procedures previously described for other drift samples.

#### DIFFERENTIATION OF GLOCHIDIA

Glochidia of the subfamilies Ambleminae and Unioninae were removed from drift samples and further sorted and identified to species. The number and density of drifting glochidia for each species were calculated, and release periods of the species were determined. To assist in identification of these glochidia at North Holston Ford, gravid females of the four species; E. edgariana, E. barnesiana, P. oviforme, L. dolabelloides were collected, placed in muslin bags and transported to the laboratory on ice. In the laboratory, mussels were separated by species and placed in aerated aquaria with recirculating flow. Females were induced to release conglutinates by either restricting flow or increasing water temperature. Emitted

conglutinates were collected from the water column with a fine mesh dip net and transferred to watch glasses of distilled water, according to species. The characteristic size, shape and color of conglutinates were noted for each species and documented with photographs.

Numbers of glochidia in each of 5 to 10 conglutinates were counted for each species. The proportion of viable glochidia to unfertilized eggs was determined, and percent viability of glochidia in conglutinates was calculated for each species.

Size and shape of glochidial valves was recorded and photographed for each species. Total length (maximum antero-posterior dimension parallel to hinge), width (maximum dorso-ventral dimension perpendicular to hinge) and hinge length (maximum length from tip to tip along hinge line) of glochidial valves were measured on a sample of 50 glochidia of each species under a compound microscope (100X). Glochidia were obtained from at least 2 to 5 different females per species for these measurements.

Measurements of glochidial valve dimensions were statistically compared among species using a Duncans Multiple Comparison test and analyzed using discriminate multivariate analysis.

#### NATURAL INFESTATIONS ON FISH

Natural infestations of glochidia on stream fishes were recorded at North Holston Ford from June 1981 to June 1982. Fish samples were collected twice monthly from six 50m sections during June, July and August of 1981, and monthly from September 1981 through April 1982. Sections 2 and 3 were sampled weekly from June through August 1981 and monthly from September 1981 through June 1982, because of higher mussel densities in these sections.

Fish were collected using a backpack electroshocking unit (Coffelt BP-1C). River sections were sampled separately to obtain a representative sample of the diversity and abundance of fish species present. Collected fish were anesthetized with tricaine methanesulfonate (MS-222), and the gills and fins were inspected for glochidia. All fish were identified to species, tallied, and presence or absence of glochidia was noted. Fish too small to adequately check for attached glochidia in the field were preserved in 10% buffered formalin. Fish with encysted glochidia or specimens of questionable identification were also placed in formalin for further examination. All fish free of glochidia were released at the site.

In the laboratory, glochidial infestations on preserved fish were examined using a dissecting microscope

(30X). Glochidia on the gills or fins of fish were counted and identified to subfamily. Fish species with encysted glochidia of the subfamilies Ambleminae and Unioninae were examined further. Glochidia were identified to species by morphometric comparison with glochidia previously identified and described. A phase contrast microscope (40x) facilitated delineation of glochidial size and shape for identification purposes. Measurements were taken to verify questionable specimens. Incidence and degree of glochidial infestation were calculated for each fish species.

#### LABORATORY INFESTATION OF FISH

Laboratory techniques developed by Zale (1980) and Weaver (1981) were followed for induced infestation experiments. Fish used in induced infestation experiments were collected by electrofishing from areas without mussels to eliminate potential immunization problems from previous exposure to mussel glochidia. Fish were transported to the laboratory in coolers or live tanks with aerators. A dilute solution of salt was sometimes added to the water to help alleviate handling stress. In the laboratory, fish were allowed to acclimate to the aquaria and experimental water temperatures of 16 to 18 C. They were fed a variety

of foods, including mealworms, brine shrimp, and trout chow.

Mussels providing glochidia for infestation trials were collected at the study site, transported to the laboratory, and maintained in laboratory aquaria containing natural substrate to a depth of 10 cm. Flow was maintained by a submersible recirculating pump (Little Giant Pump Co., Oklahoma City, Oklahoma). Mussels were fed Marine Invertebrate Diet (Marine Imports, Inc., Houston, Texas) mussel chow, and algae naturally occurring in the aquarium.

Gravid females were induced to release conglomerates by removing individual mussels from holding aquaria and placing them in pans or jars of aquarium water. The pans of water were allowed to warm to room temperature. The jars were returned to holding aquaria to maintain water temperature, but eliminate flow over the mussel. Since the first method involved changing flow and temperature conditions, it is not known whether release occurred because of temperature stress or lack of flow. Some gravid females kept in the holding aquaria released conglomerates naturally.

Expelled conglomerates were collected with a fine mesh dip net and placed in a watch glass of distilled water. Glochidia were separated from the conglomeral mass by physical agitation. A few glochidia were removed to test

viability. Good viability was indicated by rapid snapping of the valves when a salt crystal was added to the water. Poor viability was indicated by slow or no closure of the valves. If the sample contained viable glochidia, the remaining glochidia were used for induced infestations.

Glochidia were pipetted directly onto the gills of fish that had been anaesthetized with tricaine methanesulfate. Infested fish were allowed to recover and then placed in separate aquaria (40L) according to species. Aquaria were set in Living Streams (Frigid Units, Inc., Toledo, Ohio) that served as water baths, and water temperature was maintained at approximately 18 C. Several hours after infestation, one individual of each fish species was anaesthetized and examined for glochidial attachment. Thereafter one fish per species was examined for encystment on a daily basis for one week, and at weekly intervals thereafter.

Rejection of glochidia by the fish species was assumed if the presence of glochidia on the fish decreased rapidly during the week following infestation, particularly the first few days. If glochidia remained attached after one week, daily siphoning of the tanks was initiated. The bottom, sides, and surfaces of objects in the aquaria were siphoned with a flexible hose (25 mm diameter) through a 130u mesh screen. The siphonate was transferred to a

gridded petri dish and examined under a dissecting microscope (30X) for metamorphosed juvenile mussels and glochidial valves. Siphoning was continued daily until no glochidia were present on fish, determined by the weekly examinations.

#### AGE AND GROWTH

Growth rate and ages of specimens of E. edgariana at North Holston Ford were determined by tabulation and measurement of external growth rings as described by Isely (1911). Total length of valves was measured for all mussels to the nearest 0.1mm with vernier calipers. Shells were measured with the hinge perpendicular to the long axis of the calipers. Fifty shells of shiny pigtoe mussels, collected live or from muskrat middens, were measured for length-at-age using this method for each external growth ring. Only specimens in good condition and with clear annuli could be measured. However, most shells had badly eroded umbo regions, and measurement of first and second growth rings was frequently not possible.

A thin-sectioning technique for aging shells was used to verify the accuracy of the external shell aging technique. A subsample of the shells previously measured and aged externally were sectioned along a line perpendicular to the umbo, using a Buehler Isomet low speed

diamond saw (Buehler Ltd., Evanston, Illinois). A second parallel cut created a thin-section of shell which was mounted on a microscope slide. Growth rings in the cross section were counted. Ages of the shells, determined by the two techniques, were compared. Shells which had inconsistent growth patterns were difficult to age by either technique.

Logistic, Gompertz, and von Bertalanffy growth equations were used to fit the length-age relationship of F. edgariana at North Holston Ford. Maximum length ( $L_{\infty}$ ) predicted by the von Bertalanffy was compared to predicted maximum length from a Ford-Walford plot.

Measurements from valves of 24 gravid females were initially analyzed separately from those of 22 nongravid females or males to determine possible differences in growth rate according to gender. However, a Wilcoxon Rank Sum indicated the two groups had comparable growth rates, and their measurements were combined for all other statistical analyses.

## Results

### Mussel Assemblage at North Holston Ford

Earlier surveys indicated that the mussel assemblage at North Holston Ford included four subfamilies. The long-term breeders were represented by the subfamilies Anodontinae and Lampsilinae, and the short-term breeders belonged to the subfamilies Unioninae and Ambleminae. A total of 16 species in these four subfamilies were collected at North Holston Ford during the study period (Table 5). Ten species were collected and quantified in quadrat samples. Six species were collected only by handpicking or in muskrat middens, and were not quantified at the site.

Although four species of short-term breeding mussels occurred at North Holston Ford, only three were collected in quadrats. The subfamily Ambleminae was represented by two species at the site; Fusconaia barnesiana and F. edgariana. No specimens of F. barnesiana were collected in the quadrats, but live specimens were encountered by handpicking and were found freshly dead in muskrat middens. Fifteen specimens of F. edgariana were collected in quadrats, contributing 4.7% of the mussels collected. Sixty-seven additional specimens of F. edgariana were

Table 5. Numbers, densities (No./m<sup>2</sup>±1 S.D.), and population estimates (No./2930 m<sup>2</sup>), with 95% C.I., of mussel species at North Holston Ford, based on sixty 0.5 m<sup>2</sup> quadrat samples.

Species	Number collected	Density (No./m <sup>2</sup> )	Population estimate
<b>Anodontinae</b>			
<u>Alasmidonta marginata</u>	0	0	*
<u>Alasmidonta minor</u>	0	0	*
<u>Lasmigona costata</u>	0	0	*
<b>Ambleminae</b>			
<u>Fusconaia edgariana</u>	15	0.5±1.4	1,465±374
<u>Fusconaia barnesiana</u>	0	0	*
<b>Unioninae</b>			
<u>Pleurobema oviforme</u>	30	1.0±2.2	2,960±556
<u>Lexingtonia dolabelloides</u>	16	0.5±1.8	1,562±468
<b>Lampsilinae</b>			
<u>Medionidus conradicus</u>	106	3.5±3.1	10,353±779
<u>Villosa nebulosa</u>	59	2.0±2.3	5,762±589
<u>Villosa vanuxemi</u>	36	1.2±2.2	3,516±559
<u>Ptychobranchus subtentum</u>	21	0.7±1.9	2,051±480
<u>Ptychobranchus fasciolaris</u>	0	0.3±1.2	977±314
<u>Actinonaias pectorosa</u>	4	0.1±0.8	391±217
<u>Lampsilis fasciola</u>	4	0.1±0.8	391±217
<u>Lampsilis ovata</u>	0	0	*
<u>Toxolasma lividus</u>	0	0	*
Juveniles (unidentified)	16	0.5±0.9	1,562±242
Total	317	10.6±7.4	30,960±7,031

\*Species noted in surveys but not quantitatively sampled.

collected by handpicking. Numerous specimens of P. oviforme and L. dolabelloides, both of the subfamily Unioninae, were also collected by handpicking. Pleurobema oviforme was twice as abundant as Lexingtonia dolabelloides and collectively these two species together contributed 14.4% of the mussels collected in quadrat samples. The three species of short-term breeding mussels taken in quadrat samples comprised nearly 20% of the mussel assemblage at North Holston Ford.

Alasmidonta marginata, A. minor, and Lasmigona costata, three species of anodontine mussels, were collected only by handpicking at North Holston Ford. These species were rarely encountered at the study site. They were not collected in quadrat samples or in muskrat middens.

Of the nine species of lampsiline mussels at the site, only two were not collected in the quadrats. Lampsilis ovata was not uncommon at the site, but was collected only by handpicking. Only one specimen of Toxolasma lividus was found during the entire study period, and it was found freshly dead in a muskrat midden. The other seven lampsiline species contributed the greatest abundance of mussels at North Holston Ford (75.6% of the total number collected). Medionidus conradicus was by far the most abundant mussel at the site, contributing over

one-third (33.2%) of the mussels collected in the quadrats. Villosa nebulosa and V. vanuxemi were next in abundance, and together contributed an additional 30% of the mussel assemblage. The two species, Ptychobranthus subtentum and P. fasciolaris, followed in abundance and comprised 6.6% and 3.1% of the mussels, respectively. The least commonly collected lampsiline mussels were Actinonaias pectorosa and Lampsilis fasciola, which each contributed 1.3% of the mussel assemblage.

Density of mussels ranged from 3.5/m<sup>2</sup> to 0.1/m<sup>2</sup>, according to species. Six mussel species quantified in the quadrat samples all had densities of less than 1 mussel/m<sup>2</sup>. Both F. edgariana and L. dolabelloides had densities of 0.5/m<sup>2</sup>, and estimated populations of between 1,091 to 1,839 adult mussels per species in the 2930m<sup>2</sup> study area. P. oviforme had the highest density of the short term breeding mussel species with 1/m<sup>2</sup>, and a population estimate that ranged from 2,404 to 3,516 mussels at North Holston Ford. M. conradicus occurred in the highest density (3.5/m<sup>2</sup>), with an estimated population of between 9,574 and 11,132 adults for the study area. Villosa nebulosa and V. vanuxemi had mean densities of 2.0/m<sup>2</sup> and 1.2 specimens/m<sup>2</sup>, with adult populations of 5,173 to 6,351 and 2,957 to 4,075, respectively.

Ptychobranthus subtentum was twice as abundant as P.

fasciolaris with 0.7/m<sup>2</sup> and 0.3 mussels/m<sup>2</sup>, and population estimates that ranged from 663 to 2,531 mussels for the two species at the site. The remaining two species, Actinonaias pectorosa and Lampsilis fasciola, each had densities of 0.1/m<sup>2</sup>, and an estimated population of 174 to 608 adult mussels for each of the species.

The density of juvenile mussels at North Holston Ford was estimated at 1 juvenile for every 2 square meters. Juvenile mussels could not be identified to species, so population estimates ranged from 1,320 to 1,804 for all species of juveniles.

#### Mussel Assemblage

The mussel assemblage at North Holston Ford consists of sixteen species representing four subfamilies; Anodontinae, Ambleminae, Unioninae, and Lampsilinae. Neves et al. (1980) found fourteen species of mussels at North Holston Ford (NEHRM 86.9), with a mean density of 9.6 mussels/m<sup>2</sup>. This density was comparable to the 10.6 mussels/m<sup>2</sup> reported at North Holston Ford in this study. A survey at NEHRM 85.2 reported ten mussel species (Barr et al. 1982). Estimates of abundance were comparable for most of the species found at both sites.

The highest mussel densities at North Holston Ford

occurred in areas around the islands and along the left ascending bank (Figure 4). The left half of the midstream section also had high mussel densities, but the densities decreased rapidly in midstream and along the right ascending bank.

The lampsiline mussels were by far the most abundant and diverse subfamily at North Holston Ford, composing the majority of the mussel assemblage. The anodontine mussels were rarely encountered and contributed only a little diversity to the mussel assemblage. Mussels in the subfamilies of Unioninae and Ambleminae were common but not abundant, and provided approximately one fifth of the adult mussels at the study site.

The river bottom at North Holston Ford contained a variety of habitats. The greatest number of mussels was found along the left ascending bank, with mussels decreasing numerically toward the right bank. An exception to this was a shallow riffle in the lower portion of the study section, that contained many mussels across the stream width. Mussel distribution was quite clumped throughout the study section.

The habitat in which F. edgariana was most commonly encountered was along the left ascending bank, along with the majority of mussel species at the site (Figure 4). No specimens of F. edgariana were found along the right bank.

F. edgariana was found in areas of moderate to low flows, particularly low in the summer, and seasonally quite shallow. The substrate most commonly associated was sand, gravel, and either silt or pebble depending on proximity to the left bank. Quadrats in which F. edgariana were found included a fairly high number and diversity of other mussels. No more than three F. edgariana were collected in a single 0.5m<sup>2</sup> quadrat sample.

#### Substrate Analysis at North Holston Ford

Substrate samples collected at North Holston Ford included five particle categories ranging from silt to boulder, but consisted primarily of sand, gravel, and cobble (Table 6). Both quantitative samples and visual estimates at the study site indicated five longitudinal sections of similar substrate composition. Substrate sections were distinguished by changes in the percent composition of the substrate components. Proceeding from left to right ascending bank, the substrate changed from predominantly sand and gravel to predominantly pebble, and then cobble. Boulders occurred along the right bank, but were never a significant portion of the substrate. Silt occurred along the left bank, in areas around and between the islands. No large substrate sizes (cobble and boulder)

Table 6. Percent composition of surface substrate at North Holston Ford, based on thirty (0.25m<sup>2</sup>) quadrats.

Substrate Composition (%)						
Location	Silt	Sand	Gravel	Pebble	Cobble	Boulder
Left bank	14.5	45.0	38.4	2.1	-	-
Island transect	*	26.9	37.8	35.3	*	-
Midstream	-	23.3	27.1	49.6	*	-
Midstream	-	5.0	40.5	31.5	23.0	*
Right bank	-	4.0	31.0	31.0	32.2	1.8

\*indicates < 1% of that particle size encountered

[silt < .125mm, sand = .125-2mm, gravel = 2-16mm, pebble = 16-64mm, cobble = 64-256mm, boulder > 256mm]

were noted around the islands or in areas overlain with silt.

Flow conditions were quite variable weekly and seasonally, and ranged from a summer low flow average of 0.1m/second to an early summer high flow average of 0.6m/second. In general, velocities were greater along the right bank and midstream, and lower along the left bank. Water depths, on the average, increased from left to right bank, and were shallow throughout the ford area. Water depth ranged from 10 to 60 centimeters during the shallow summer period. Spring floods brought water levels up 2 to 3 meters or more. Although water depth was quite variable seasonally, the right ascending bank always had deeper water than the left bank.

Aquatic vegetation occurred only on the islands and in a small section directly upstream of the ford. Islands were visible through summer, fall and winter, and were covered during high water conditions in spring. Water willow (Justicia americana) grew on the islands throughout the summer and into the fall, but was not present in the winter. The other areas with vegetation were shallow portions of the river, adjacent to the ford, which became dry during low flow conditions, and supported seasonal growth of water willow.

Description of Glochidia

To differentiate the glochidia of the four species of short-term breeders at North Holston Ford, principle component and discriminant analyses were used to evaluate whether any glochidial valve measurement, or combination of measurements, differed significantly among species so that glochidia of F. edgariana could be distinguished (Table 7). Tests of homogeneity between matrices indicated that overlap existed within and among measurements of shell width, length, and hinge length, and no single measurement proved diagnostic for a particular species. However, multiple comparison statistical analysis of the measurements indicated that glochidial valves could be differentiated to species by comparing two or more of the glochidial shell measurements (Table 8).

F. edgariana glochidia were comparable to L. dolabelloides in length, and both were significantly shorter than F. barnesiana or P. oviforme. In width, glochidia of F. edgariana were comparable to F. barnesiana, larger than P. oviforme, and smaller than L. dolabelloides. Hinge length measurements were comparable for F. edgariana, L. dolabelloides and P. oviforme, but differed significantly from the longer hinge length of F. barnesiana.

Although overall shape of the glochidia of F.

Table 7. Dimensions of mature glochidia of Fusconaia edgariana, F. barnesiana, Pleurobema oviforme and Lexingtonia dolabelloides

Species	Mean $\pm$ SD ( $\mu\text{m}$ )	Range ( $\mu\text{m}$ )
		Width
<u>F. edgariana</u>	172.50 $\pm$ 7.95	156.9 - 185.0
<u>F. barnesiana</u>	176.05 $\pm$ 7.59	162.9 - 185.0
<u>L. dolabelloides</u>	194.71 $\pm$ 6.62	181.3 - 203.5
<u>P. oviforme</u>	166.68 $\pm$ 4.32	155.4 - 170.8
		Length
<u>F. edgariana</u>	143.06 $\pm$ 7.95	133.2 - 155.4
<u>F. barnesiana</u>	174.16 $\pm$ 7.47	162.8 - 185.0
<u>L. dolabelloides</u>	151.24 $\pm$ 4.57	140.6 - 159.1
<u>P. oviforme</u>	173.89 $\pm$ 4.43	162.8 - 179.2
		Hinge Length
<u>F. edgariana</u>	116.18 $\pm$ 7.43	103.6 - 129.5
<u>F. barnesiana</u>	144.19 $\pm$ 7.72	133.2 - 155.4
<u>L. dolabelloides</u>	127.28 $\pm$ 4.96	118.4 - 133.2
<u>P. oviforme</u>	126.47 $\pm$ 6.19	111.0 - 133.2

Table 8. Duncans Multiple Comparison Test among species, for glochidial shell measurements of width, length, and hinge length. Species underscored indicate no significant difference in specified measurement ( $p=0.05$ ).

Glochidial Dimension	Species			
Width	Fusconaia barnesiana -----	Fusconaia edgariana -----	Pleurobema oviforme -----	Lexingtonia dolabelloides -----
Length	Fusconaia barnesiana -----	Pleurobema oviforme -----	Fusconaia edgariana -----	Lexingtonia dolabelloides -----
Hinge	Fusconaia barnesiana -----	Fusconaia edgariana -----	Pleurobema oviforme -----	Lexingtonia dolabelloides -----

edgariana was similar to that of L. dolabelloides, it could be differentiated by its smaller width. Glochidia of P. oviforme and F. barnesiana were similar to each other in overall shape, and could be readily distinguished from F. edgariana by their greater length.

### Drift

Glochidia of the short-term breeding mussel species were present in drift samples from June 10 to August 18, 1981, and from June 23 to August 26, 1982. Results of the 24-hour drift study indicated the density of glochidia of the short-term breeding mussel species was greatest from mid to late afternoon (Figure 5). Density of glochidia increased rapidly from mid-morning (1000hrs) to a peak in early afternoon (1400hrs) that apparently continued until early evening (1800hrs). Glochidial density appeared to decline rapidly through the evening hours (2200hrs) and remained low throughout the night (0200hrs). Density of glochidia in the drift was lowest in early morning hours (0600hrs).

Samples collected from drift nets on the left ascending side of the river at all stations contained more glochidia than samples from either the center or right locations (Figure 6). Glochidial densities in the left net

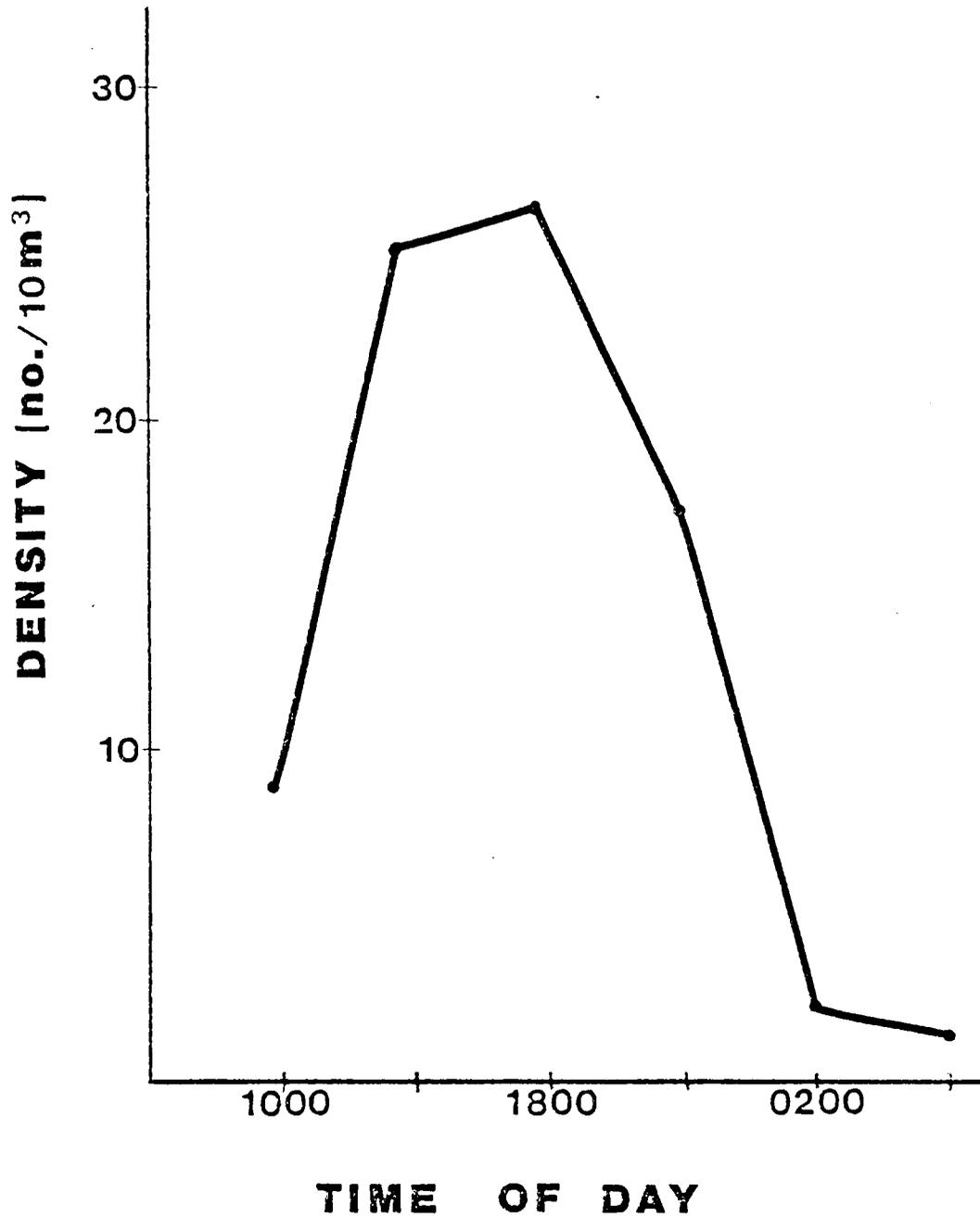


Figure 5. Diel densities of drifting glochidia (no./10 m<sup>3</sup>) from North Holston Ford at left sampling locations Stations 2 and 3 during 28-29 July 1982.

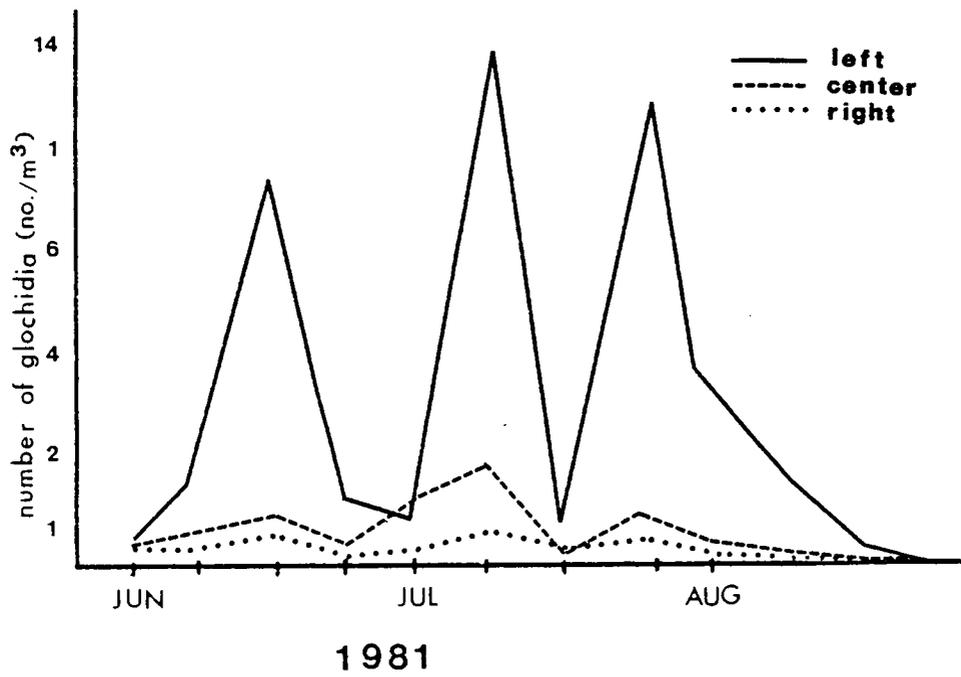


Figure 5. Mean densities of glochidia in drift samples from North Holston Ford for left, center, and right sampling locations, 1981-1982.

at Stations 2 and 3 greatly influenced estimates of drift densities at North Holston Ford. From samples collected in 1981, it was apparent that these two stations were best for monitoring glochidial drift at the site. In 1982, only these two locations were sampled.

Of the four species of short-term breeders, only glochidia of three were collected in the drift at the site; Fusconaia edgariana, Pleurobema oviforme and Lexingtonia dolabelloides. No glochidia of Fusconaia barnesiana were identified in drift samples, although adults of this species were found to occur at the site. P. oviforme glochidia were by far the most abundant of the three species in the drift, comprising 55.7% of the number collected. F. edgariana glochidia were less abundant, contributing 24.5% of the drift, and L. dolabelloides was the least abundant with only 20.1%.

Glochidia of Fusconaia edgariana were first present in drift samples on June 23, 1981 (Table 9). The numbers slowly increased to a relative low peak on July 2 (0.21 glochidia/m<sup>3</sup>). A second peak of higher density appeared in mid-July (0.51 glochidia/m<sup>3</sup>), which coincided with the major release period for both P. oviforme and L. dolabelloides (Figure 7). The third and major peak in glochidial density of F. edgariana occurred in late July, with an average of 1.12 glochidia/m<sup>3</sup> in drift samples. The

Table 9. Number and density (No.x10/m<sup>3</sup>) of glochidia in drift samples from North Holston Ford, June-August 1981 and June-September 1982.

Date	<u>F. edgariana</u>	<u>P. oviforme</u>	<u>L. dolabelloides</u>
1981			
10 June	0 (0)	73 (0.77)	10 (0.11)
15 June	0 (0)	1345 (5.63)	224 (0.94)
23 June	380 (1.62)	91 (3.87)	538 (2.28)
2 July	548 (2.13)	239 (0.93)	130 (0.51)
8 July	71 (1.07)	146 (2.21)	49 (0.74)
14 July	701 (5.12)	5499 (40.29)	743 (5.42)
21 July	157 (1.72)	114 (1.24)	30 (0.33)
28 July	1055 (11.18)	1907 (20.20)	265 (2.79)
5 August	51 (1.48)	58 (1.69)	79 (2.35)
12 August	34 (0.57)	121 (1.51)	80 (0.86)
18 August	1 (0.01)	20 (0.39)	2 (0.01)
26 August	0 (0)	0 (0)	0 (0)
1982			
24 June	2 (0.04)	190 (5.82)	27 (0.86)
5 July	33 (1.36)	103 (4.33)	32 (1.34)
13 July	568 (16.50)	149 (4.33)	450 (13.05)
28 July	84 (1.90)	595 (13.50)	276 (6.28)
10 August	107 (4.46)	141 (5.88)	70 (2.92)
18 August	16 (0.13)	110 (0.92)	27 (0.23)
26 August	3 (0.03)	29 (0.30)	4 (0.04)
2 September	0 (0)	0 (0)	0 (0)

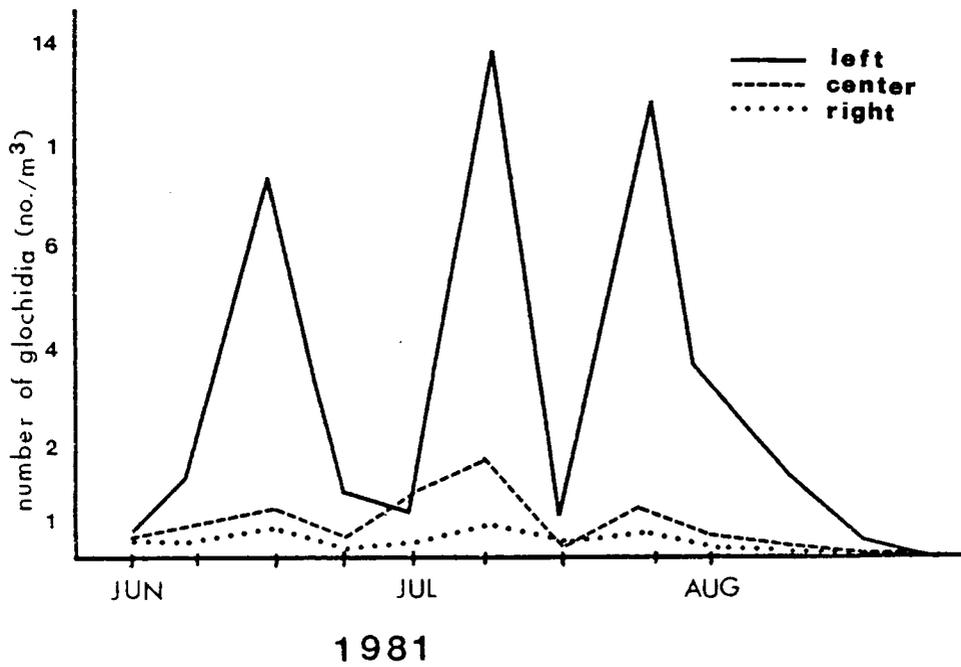


Figure 5. Mean densities of glochidia in drift samples from North Holston Ford for left, center, and right sampling locations, 1981-1982.

density of F. edgariana dropped quite rapidly in August, and only one glochidium was found in the August 18 sample of 1981.

In 1982, glochidia of F. edgariana were present in the drift from June 24 until August 26. Two peaks of high density occurred during the 1982 release period. The first peak occurred in mid-July and had the highest density recorded for F. edgariana, 3.39 glochidia/m<sup>3</sup>. The second peak occurred in mid-August with a density of 1.33 glochidia/m<sup>3</sup>. The density of F. edgariana in the drift declined rapidly in August and only three glochidia were found in the sample of August 26.

P. oviforme glochidia were found in drift samples from June through August. Three peaks of P. oviforme glochidia were noted in the drift in 1981 (Figure 7). Highest densities were noted in mid-July with 5.08 glochidia/m<sup>3</sup>, which coincided with high numbers of glochidia of the other two short-term breeding species. After a small peak in mid-August, no glochidia of P. oviforme were collected. In 1982, two peaks in glochidial density were noted for P. oviforme. The first occurred in late June and the second extended from late July until mid-August. Since samples were taken twice monthly in 1982, as compared to weekly in 1981, this extended peak may be an artifact of less intensive sampling.

Glochidia of L. dolabelloides were present in drift samples from June through early August of 1981 and 1982. Three peaks in glochidial density was noted in 1981, and the highest density recorded were 4.03 glochidia/m<sup>3</sup>. After the first week in August, the number of L. dolabelloides in the drift decreased rapidly until only two glochidia were found in the mid-August sample of 1981. In 1982 a single extended peak was indicated from mid to late July. As noted with P. oviforme, this extended peak may have resulted from a lack of samples taken during the intervening period. The average density of glochidia of L. dolabelloides in the drift varied from 0.15 glochidia/m<sup>3</sup> in 1981 to 0.33 glochidia/m<sup>3</sup> in 1982.

The densities of drifting glochidia for each of the three species were found to approximate the percent of adults of those species in the assemblage at North Holston Ford. The percent of E. edgariana at North Holston Ford (24.5%) was very similar to its abundance in the drift (24.4%). Quadrat samples at the site indicated that P. oviforme was the most abundant of the four short term breeders at the site. It contributed slightly over half (55.7%) of the glochidia in the drift samples of 1981 and 1982, and comprised approximately half (49.2%) of the short term breeders. The density of glochidia of L. dolabelloides in the drift doubled between 1981 and 1982,

with an average density for both years of 20.1%. This percentage was similar to the 26.2% that L. dolabelloides contributed to the mussel assemblage.

Gravid females of Fusconaia edgariana were first collected at North Holston Ford on June 16, 1981. No specimens collected at the site appeared to be releasing conglutinates until June 25, 1981. Female F. edgariana were found to release glochidia at North Holston Ford from June 25 to July 22, 1981. Gravid females brought to the laboratory released glochidia from June 25 to July 30. This extra week of glochidial release in the laboratory may have resulted from temperatures slightly lower than stream temperatures.

In 1982, the first gravid female F. edgariana was collected on June 25. Females were found to release glochidia at North Holston Ford from July 5 to August 10, 1982. Gravid females brought to the laboratory also released glochidia during this same period, with the exception of one specimen which retained conglutinates until August 16. Individual females released glochidia over a period of 3 to 7 days. Some mussels were observed to expel all conglutinates within their gills in the laboratory, while others did not.

#### Conglutinates

Counts of glochidia within conglutinates from each of the species are summarized in Table 10. Conglutinates collected from gravid *F. edgariana* contained a low percentage of mature glochidia. An average of 30% successful fertilization occurred within the conglutinates examined, ranging from 11% to 42% viable glochidia per conglutinate. The remaining portion of these conglutinates consisted of unfertilized eggs. Unfertilized eggs were not found in drift samples for any species, but were observed in conglutinates expelled in the laboratory that also contained fully developed glochidia. Glochidial counts from conglutinates for the other species indicated that unfertilized eggs comprised only 2% to 5% of the conglutinates of *P. oviforme* and *L. dolabelloides*. The only conglutinates available from *F. barnesiana* were not fully developed; however, developing glochidia were counted along with unfertilized eggs to determine numbers of potential glochidia per conglutinate.

#### Natural Infestations on Fish

From June 1981 through June 1982 a total of 4,800 fish were collected at North Holston Ford and inspected for glochidial attachment. Of the 41 species sampled at the site, 12 of the 16 cyprinid species were found to carry

Table 10. Mean number of unfertilized eggs and viable glochidia per conglutinate in Fusconaia edgariana, F. barnesiana, Pleurobema oviforme, and Lexingtonia dolabelloides.

Species	Eggs	Glochidia	Total
<u>F. edgariana</u> (1981) (n=10)	267.2±53.1	112.0±39.1	379.2±31.7
<u>F. edgariana</u> (1982) (n=10)	243.4±47.4	121.0±35.1	364.4±26.8
<u>F. barnesiana</u> (n=5)	6.4±1.8	212.0±58.8	219.2±58.5
<u>P. oviforme</u> (n=5)	5.0±5.5	385.0±45.6	400.0±50.6
<u>L. dolabelloides</u> (n=5)	9.2±4.2	208.2±66.5	217.8±70.9

glochidia of the subfamilies Unioninae and Ambleminae (Table 11). No other fish species were found to carry glochidia of the short-term breeding mussel species. Glochidia were found attached to fish from June to August of 1981 and were present again in June 1982. A total of 1,442 cyprinids were examined during this period of glochidial attachment, of which 23.2% were infested with glochidia of these two subfamilies.

Further examination of the gills of these fish indicated that glochidia of three of the four mussel species were attached. Of the total number of fish parasitized by glochidia of the short-term breeding mussels, 6.6% were infested with glochidia of E. edgariana, as compared to 57.4% infested with P. oviforme and 36% with L. dolabelloides. Glochidia of F. barnesiana, could not be confirmed on any fish, although adults of this species occurred at the site. Number and percent of cyprinids infested by each of the short-term breeding mussel species is summarized in Table 12. Some of the fish species were infested exclusively with one species of glochidia, whereas others had two species of glochidia on their gills, and one species, the telescope shiner, was found to have glochidia of three mussel species attached to some of the specimens.

P. oviforme was found on 11.7% of the total number of cyprinids examined. Nine cyprinid species bore glochidia of

Table 11. Incidence of glochidial infestations on cyprinid species at North Holston Ford, June 1981-June 1982.

Species	Number examined	Number infested	Percent infested
Telescope shiner	499	59	11.8
Stoneroller	442	10	2.3
Tennessee shiner	273	139	50.9
Common shiner	157	23	14.6
River chub	144	11	7.6
Warpaint shiner	102	16	15.7
Whitetail shiner	73	35	48.0
Popeye shiner	51	12	23.5
Saffron shiner	47	25	53.2
Streamline chub	29	0	0
Silver shiner	28	3	10.7
Rosyface shiner	15	1	6.7
Stargazing minnow	10	0	0
Blacknose dace	6	0	0
Creek chub	1	0	0
Mirror shiner	1	1	-

Table 12. Number and percent (%) of fish infested with glochidia of E. edgariana, P. oviforme, and L. dolabelloides at North Holston Ford, June-August 1981 and June 1982.

Fish species	Number examined	Mussel Species		
		<u>Fusconaia edgariana</u>	<u>Pleurobema oviforme</u>	<u>Lexingtonia dolabelloides</u>
<u>Telescope shiner</u>	499	6 (1.2)	43 (8.6)	21 (4.2)
<u>Stoneroller</u>	442	-	9 (2.0)	-
<u>Tennessee shiner</u>	273	-	56(20.5)	88(32.2)
<u>Common shiner</u>	157	5 (3.9)	23(14.6)	-
<u>River chub</u>	144	-	11 (7.6)	-
<u>Warpaint shiner</u>	102	3 (2.9)	14(13.7)	-
<u>Whitetail shiner</u>	73	13(17.8)	34(46.6)	-
<u>Popeye shiner</u>	51	-	-	12(23.5)
<u>Saffron shiner</u>	47	-	23(48.9)	4 (8.5)
<u>Silver shiner</u>	28	-	-	3(10.7)
<u>Rosyface shiner</u>	15	-	-	1 (6.7)
<u>Mirror shiner</u>	1	-	1 (100)	-
Total	1832	27 (1.5)	214(11.7)	129 (7.0)

P. oviforme, the river chub, stoneroller, and Tennessee, telescope, common, warpaint, whitetail, saffron and mirror shiners. Three species, the stoneroller, river chub and mirror shiner, were found to carry only glochidia of P. oviforme. The Tennessee and saffron shiners harbored glochidia of both P. oviforme and L. dolabelloides. Six telescope shiners had glochidia of these two species and F. edgariana. L. dollabelloides was found on 7% of the cyprinids examined, representing six species; the popeye, rosyface, saffron, silver, telescope, and Tennessee shiners. The three species which carried only L. dolabelloides glochidia were the popeye, silver and rosyface shiners. Tennessee and saffron shiners also carried P. oviforme glochidia.

Glochidia of Fusconaia edgariana were found on 1.5% of the cyprinids examined and, with one exception, were always found on fish which also had P. oviforme glochidia attached to the gills. One warpaint shiner was found to have only glochidia of Fusconaia edgariana attached to its gills, and was the only specimen found exclusively with F. edgariana. Thirteen whitetail shiners were observed with F. edgariana glochidia, constituting 17.8% of the total number of whitetail shiners collected during the summer period. The telescope shiner had the lowest percentage of encystment by F. edgariana, with only 1.2% (6 specimens) of that species

found to carry shiny pigtoe glochidia. The common and warpaint shiner were roughly comparable in occurrence of F. edgariana with approximately 4% (5 specimens) and 3% (3 specimens) of each of the species encysted by F. edgariana. Averaging all four species, the infestation rate for F. edgariana on cyprinids at North Holston Ford was 6.45%.

Glochidia of F. edgariana were present on fish from July 1 through August 6 of 1981. They were most abundant on fishes of the July 21 sample; 12 fish representing 4 species were found with F. edgariana glochidia (Table 12). This also coincided with peak numbers of fish infested with other glochidia as well. The July 1 sample had three fish species encysted with glochidia of F. edgariana, the telescope, warpaint and whitetail shiners each had one fish infested. The next week (July 7) there were two fish, both common shiners, with F. edgariana on them. On July 14 only one fish, a warpaint shiner, had glochidia of F. edgariana. As mentioned previously, the greatest number of encysted glochidia occurred on July 21; 1 warpaint, 2 telescope, 3 common and 6 whitetail shiners were infested with glochidia of F. edgariana. On July 28 only 1 fish, a telescope shiner, was collected with F. edgariana glochidia. Two specimens each of the telescope and whitetail shiners carried glochidia of F. edgariana on August 5, which was the last sample date in which F. edgariana glochidia were found

encysted in 1981. In 1982, four whitetail shiners were the only fish collected with glochidia of F. edgariana encysted.

The degree of glochidial infestation and percent of the total number per species for F. edgariana, P. oviforme, and L. dolabelloides is summarized in Table 13. The degree of infestation was low for F. edgariana, ranging from 1 to 3 glochidia per fish. No more than 3 glochidia occurred on any of the 27 fish encysted with glochidia of F. edgariana.

Infestation rates for P. oviforme ranged from 1 to 140 glochidia for the 216 infested fish. Of those fish, 77.8% carried 1 to 5 glochidia (51% carried only 1 glochidium), 8.8% carried 6 to 10 glochidia, 7.4% carried 11 to 40 glochidia, and 6.0% were found with over 40 glochidia per fish. Of the 137 fish encysted with glochidia of L. dolabelloides, slightly over half (54.0%) had 1 to 5 glochidia per fish. The remaining fish had 6 to 10 (17.5%) and 11 to 20 (21.9%) encysted glochidia per fish, and 5.8% had 21 to 40 glochidia per fish. Only one fish (0.8%) had over 50 glochidia encysted, which was the maximum number of glochidia of L. dolabelloides found on an individual fish.

Degree of infestation was categorized as low (1 to 5 glochidia per fish), moderate (6 to 20 glochidia per fish), and high (over 20 glochidia per fish). F. edgariana was characterized by very low infestation rates on all fish. The vast majority of fish encysted by P. oviforme also had

Table 13. Degree of infestation and percent (%) of F. edgariana, P. oviforme, and L. dolabelloides glochidia on fishes at North Holston Ford from June-August 1981 and June 1982.

Number glochidia	<u>F. edgariana</u>	<u>P. oviforme</u>	<u>L. dolabelloides</u>
	No. fish (%)	No. fish (%)	No. fish (%)
1 - 5	27 (100)	168 (77.8)	74 (54.0)
6 - 10	-	19 (8.8)	24 (17.5)
11 - 20	-	5 (2.3)	30 (21.9)
21 - 40	-	11 (5.1)	8 (5.8)
40+	-	13 (6.0)	1 (0.8)
Range	1 to 3	1 to 140	1 to 50

low infestation rates; only 11.1% of the fish carried moderate and high infestations, respectively. Of the fish encysted with L. dolabelloides, approximately half (54.0%) had low infestations and slightly fewer (39.4%) had moderate infestations. Very few fish (6.6%) had high rates of infestation with glochidia of L. dolabelloides.

#### Laboratory Induced Infestations

A total of 53 fish specimens representing the families Catostomidae, Cyprinidae, Ictaluridae, Cottidae, Percidae and Centrarchidae were infested with glochidia of Fusconaia edgariana between July 6 and July 27, 1981 (Table 14). At least one species of each family was chosen for initial laboratory trials in 1981. Additional species of percids and centrarchids were infested because of their greater abundance and availability at the study site. Five fish per species were infested, except the catostomid and ictalurid species of which three individuals were infested. Two infestation trials were conducted on the cyprinid species due to mortality of all specimens from the first infestation. All other infestations were conducted only once, and no mortalities occurred during the infestation period.

Non-cyprinid species sloughed all glochidia within 5 days of initial infestation. No metamorphosis was noted in

Table 14. Fish species artificially infested with glochidia of Fusconaia edgariana in July 1981. All glochidia were sloughed within 5 days post-infestation.

Fish species	Number infested
<b>Centrarchidae</b>	
Smallmouth bass	5
Rock bass	5
Bluegill sunfish	5
Longear sunfish	5
<b>Percidae</b>	
Redline darter	5
Greenside darter	5
Banded darter	5
<b>Catostomidae</b>	
Northern hogsucker	3
<b>Cottidae</b>	
Banded sculpin	5
<b>Ictaluridae</b>	
Margined madtom	3
<b>Cyprinidae</b>	
Stoneroller	8*

\*All specimens died within three days of infestation.

glochidia collected from the aquarium siphonate, and no glochidia remained on the gills of fish for longer than 5 days. These laboratory results corroborated the field observations that non-cyprinids did not harbor glochidia of Fusconaia edgariana at North Holston Ford.

Because only cyprinid species harbored glochidia of F. edgariana at North Holston Ford in 1981, seventy-four cyprinids representing 11 species were infested with glochidia of F. edgariana between July 15 and August 5, 1982 (Table 15). Due to high mortality of many of the fish species, seven of the eleven infestation experiments were repeated. A minimum of three fish per species was infested, and up to 8 fish were infested if glochidia were available. With the exception of the common and whitetail shiners, all cyprinid species sloughed glochidia by 14 days post-infestation. Encystment periods ranged from 4 to 14 days, with all glochidia remaining attached for at least 4 days. The encystment period was quite variable, and often occurred in two phases. Some fish initially sloughed glochidia after 4 to 6 days, while other individuals of the same species sloughed glochidia after 9 to 11 days. This was noted in the river chub, stoneroller, and in the Tennessee, warpaint, popeye, silver and rosyface shiners. A few specimens sloughed glochidia throughout the infestation period. The saffron and telescope shiners retained glochidia for up to

Table 15. Cyprinid species artificially infested with glochidia of Fusconaia edgariana, July-August 1982.

Fish Species	Number infested		Period of encystment (days)
	Trial 1	Trial 2	
Popeye shiner	4	3	5 - 11
River chub	5	-	5 - 11
Rosyface shiner	3	-	5 - 8
Saffron shiner	3	-	6 - 14
Silver shiner	4	3	4 - 9
Stoneroller	5	3	5 - 8
Telescope shiner	6	5	7 - 14
Tennessee shiner	8	4	4 - 10
Warpaint shiner	3	4	5 - 10
Whitetail shiner	3	3	38
Common shiner	6	-	38

14 days; this unusually long encystment period may have resulted from the cooler temperatures (16 C) of the aquaria. No evidence of metamorphosis was noted in glochidia collected in the siphonate of any of these species.

The common and whitetail shiners retained glochidia of F. edgariana for up to 38 days, at which time pre-metamorphosed juveniles were found in the siphonate of aquaria. Two specimens were found in the siphonate of the common shiner, and one in the siphonate of the whitetail shiner. Three days later, another pre-metamorphosed juvenile was collected in the common shiner tank. These pre-juveniles have been noted by other researchers, just prior to fully metamorphosed juvenile excystment (Lynn Weaver, pers. comm.). The pre-metamorphosed juveniles appeared darker than sloughed glochidia. The single adductor muscle, indicative of the glochidial stage, was still present but less pronounced. The two adductor-abductor muscles, typical of a fully developed juvenile, were not visible, but that area of the glochidial valve was darkened.

The last whitetail shiner died after 45 days, and was examined; no glochidia were found on its gills. After 48 days, the three remaining common shiners were sacrificed and no glochidia were noted on their gills. No free-living juveniles were found in the aquaria of the two species, and

all glochidia apparently dropped off prior to completion of metamorphosis. Water temperatures of the aquaria were maintained at 16 to 18 C during the infestation periods, but a power failure caused a 6 to 8 hour increase in water temperature to 22 C. The pre-metamorphosed juveniles were found in the siphonate two days after this increase in temperature. Since no other cyprinid species were infested with glochidia at the time, the influence of this abrupt temperature increase on glochidial excystment is not known.

#### Age and Growth of *Fusconaia edgariana*

Fifty-eight specimens of *F. edgariana* were aged and measured to the nearest 0.1mm for total length (maximum antero-posterior dimension) and length to each annulus on the valves. The youngest specimen collected was age 3 and measured 12.8mm in length. This was the smallest shiny pigtoe mussel found at North Holston Ford. Only four specimens less than 6 years of age were encountered, and no age 6 specimens were encountered. The majority of mussels were age 7 and older. Few mussels older than age 10 were used for age analysis because of poor shell condition. Age 20 was the maximum age estimated for a specimen with discernible annuli at North Holston Ford. Average lengths-at-annuli and size ranges of observed lengths are summarized in Table 16. Measurements of first and second

Table 16. Observed and predicted lengths-at-annuli (mm) for Fusconaia edgariana from North Holston Ford, 1981-1982.

Annulus	Observed lengths		Predicted lengths	N*
	Mean	Range	(von Bertalanffy)	
1	5.3	4.2 - 6.5	7.8	4
2	9.8	7.3 - 14.6	13.0	6
3	14.9	12.6 - 19.0	18.8	30
4	21.4	17.8 - 24.8	22.2	35
5	27.3	23.0 - 30.1	25.9	35
6	32.5	27.8 - 35.6	29.6	35
7	36.3	31.6 - 39.0	33.2	35
8	38.6	34.4 - 41.3	36.7	35
9	41.1	36.3 - 44.6	39.9	35
10	43.6	39.2 - 46.9	42.8	35
11	45.5	40.1 - 48.2	45.8	7
12	46.5	42.5 - 49.8	47.5	6
13	47.6	44.7 - 51.1	49.3	5
14	49.3	46.0 - 52.6	50.8	5
15	53.6	52.0 - 55.6	51.9	2
16	55.0	52.8 - 56.2	52.9	2
17	56.2	53.6 - 58.7	53.7	2
18	56.9	54.9 - 59.4	54.3	2
19	57.3	54.4 - 58.3	54.8	2
20	58.8	57.5 - 59.8	55.4	2
Total =				58

\*Total number of measurements to that annulus.

annuli were limited to four and six observations, respectively, due to erosion of the umbo region on most valves.

The von Bertalanffy equation provided the best description of growth for Fusconaia edgariana at North Holston Ford ( $r=0.89$ ), indicating a predicted maximum length of 58.5mm, while the Ford-Walford plot predicted 61.8mm as the maximum length. Observed and predicted lengths-at-age are given in Table 16 and depicted graphically in Figure 8. Predicted lengths corresponded closely to measured lengths at 7 to 14 years of age. Predicted lengths were overestimated in ages less than 7 years and underestimated in ages greater than 14 years. Small sample sizes for both older and younger age classes may have caused the poor predicted versus observed values in these age groups.

Age determination by external growth rings was compared to aging by the shell thin-sectioning technique (Table 17). Ages were consistent through age 8 using both techniques. Thereafter, ages based on thin-sectioning indicated additional annuli that were not externally discernible. The number of annuli indicated by thin-section aging was 2 to 4 annuli greater than external ring aging for ages beyond 12. Only two shells aged by thin-sectioning had fewer number of annuli than the number

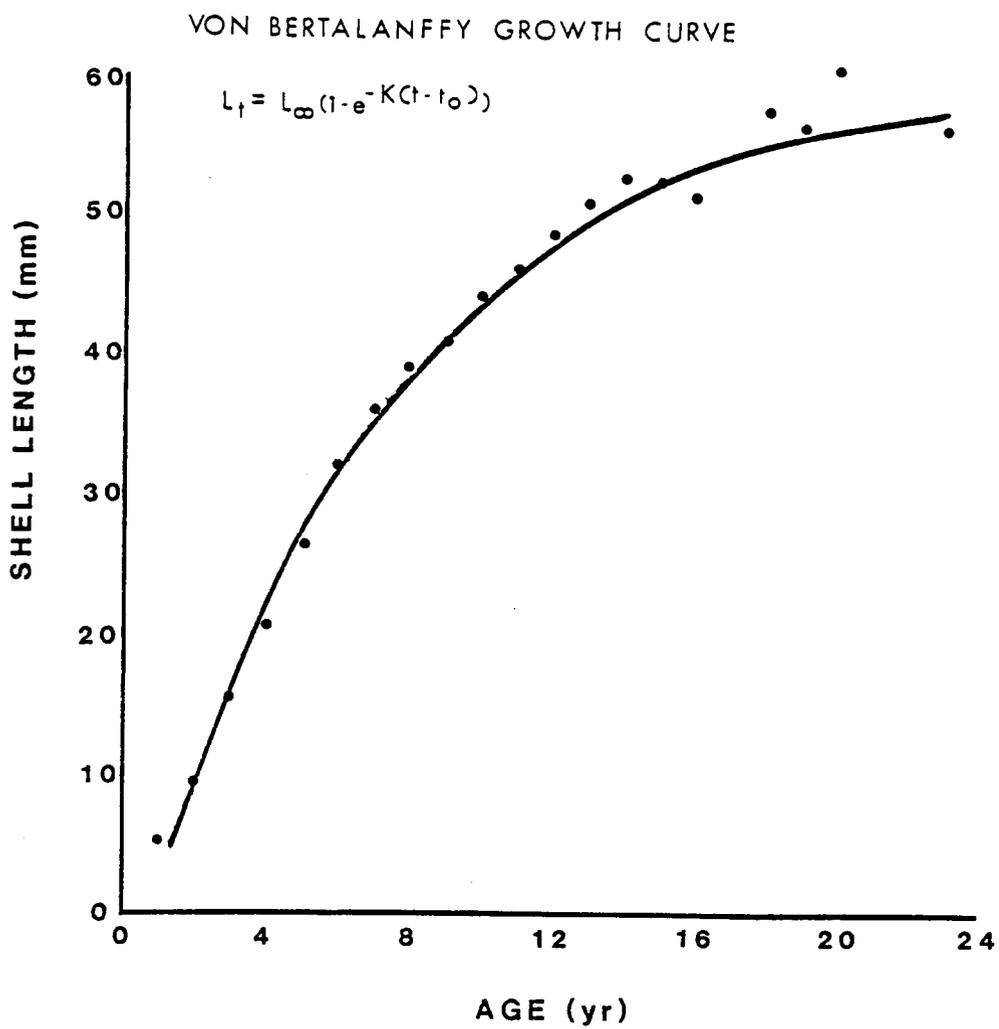


Figure 7. Mean observed (●) and predicted lengths-at-annuli for E. edgariana from North Holston Ford.

Table 17. A comparison of ages for Fusconaia edgariana as determined by external growth rings and shell thin-sectioning.

Valve length (mm)	Age (years)	
	External rings	Internal rings
38.3	7	7
38.7	7	7
37.7	8	8
40.0	8	8
41.0	8	8
38.0	9	9
36.4	9	10
40.6	9	10
41.2	9	10
39.4	10	10
37.6	10	11
43.8	10	11
45.0	10	11
43.1	10	8
46.4	11	9
44.2	11	11
47.0	11	12
47.4	11	12
45.2	12	12
48.2	13	15
48.4	13	15
48.4	13	15
50.1	14	16
52.6	14	16
52.8	16	18
52.9	16	20
57.2	16	20

indicated by external growth rings.

## DISCUSSION

### Mussel Assemblage

The mussel assemblage at North Holston Ford consists of sixteen species representing four subfamilies; the Anodontinae, Ambleminae, Unioninae, and Lampsilinae. Neves et al. (1980) found fourteen species of mussels at North Holston Ford (NFHRM 86.9), with a mean density of 9.6 mussels/m<sup>2</sup>. This density was comparable to the 10.6 mussels/m<sup>2</sup> reported at North Holston Ford in this study. A survey at NFHRM 85.2 reported ten mussel species (Barr et al. 1982). Estimates of abundance were comparable for most of the species found at both sites.

The highest mussel densities at North Holston Ford occurred in areas around the islands and along the left ascending bank. The left half of the midstream section also had high mussel densities, but the densities decreased rapidly in midstream and along the right ascending bank.

The lampsiline mussels were by far the most abundant and diverse subfamily at North Holston Ford, comprising the majority of the mussel assemblage. The Anodontinae were rarely encountered and contributed only a little diversity to the mussel assemblage. Mussels in the subfamilies of Unioninae and Ambleminae were common but not

abundant, and composed approximately one fifth of the adult mussels at the study site.

The river bottom at North Holston Ford contained a variety of habitats. The greatest number of mussels was found along the left ascending bank, with mussels decreasing numerically toward the right bank. An exception to this was a shallow riffle, in the lower portion of the study section that contained many mussels across the stream width. Mussel distribution was quite clumped throughout the study section. The highest densities of mussels were found around islands of water willow (Justicia americana) along the left bank.

#### Substrate Composition

Harman (1972) indicated a strong relationship between substrate and distribution of mussels. Sickel (1980) also considered substrate to be one of the major factors in the occurrence of mussels. The substrate at North Holston Ford is characterized by roughly longitudinal zones that increase in substrate particle size from left to right ascending bank. These zones were apparently affected by the presence of two islands and the intrusion of bank features.

The composition of stream bed is heavily dependent on stream flow. Stream velocities at North Holston Ford generally increased from left to right ascending bank, following patterns that may account for the substrate zones. The velocities were also dependent on water depth, which was shallower on the left bank and deeper on the right. The exception to this was a riffle area that was shallow across the stream width and maintained a moderately high velocity. Flow characteristics are also influenced by streambed features, such as islands. Silt was found only along the left bank, especially on the leeward side of the islands. The areas between the islands and the bank were shallow and had reduced flows.

Distribution of mussels has been attributed to various chemical, physical and biological characteristics (Williams 1969). Results of water quality measurements at North Holston Ford indicated average to good water quality at the study site and did not show any contaminants that might affect mussel distribution. The physical characteristics examined in this study, substrate composition and water velocity, appear to change in a manner similar to changes in mussel densities. Areas of highest mussel densities at North Holston Ford were characterized by low to moderate velocities, shallow depths, and sand, gravel, or pebble substrates. Most aquatic invertebrates have an inherent

need for current for feeding or respiration (Hynes 1970), and the low flows observed are apparently adequate for continued existence of the mussels.

Substrate composition was consistent throughout the study site along a gradient from left to right bank. Mussel densities generally decreased with increasing substrate particle size. The riffle zone had fairly uniform substrate across the width of the river, and high mussel densities throughout. Water depth and flow also followed the left to right bank phenomenon, except in the riffle zone. Higher velocity conditions in the riffle may account for higher densities of mussels observed there. The deep water zones at North Holston Ford had low numbers of mussels. The mussels at North Holston Ford appeared to prefer shallow water conditions.

The distribution of mussels relative to substrate has been investigated by many authors. Coker et al. (1921) indicated the importance of bottom sediments to the occurrence of mussels. Sickel (1980) could correlate at least some mussel species with bottom sediments, as did Harman (1972). Porter and Horn (1983) found no differences in distribution patterns of mussel species, but did note consistent trends in the location of certain species.

As judged by substrate and water velocity conditions at North Holston Ford, there was no single physical

characteristic that appeared to account for the distribution of mussels. Mussels were abundant in areas of low flows and high flows and occurred in a variety of substrate types. Distribution of mussels may rather be a tradeoff of characteristics, such as flow, water depth, and substrate. Preferred habitat may be the optimum of these environmental influences. Strayer (1981), in finding no significant difference in the distribution patterns of mussel species, attributed the wide dispersal of mussels to a heterogeneous stream environment.

F. edgariana was found throughout at least one third of the study area, along the left bank and close to the islands. The shiny pigtoes were distributed over a fairly large area of approximately 1000m<sup>2</sup>, indicating a dispersed use of habitat, not limited in its ability to use a variety of similar substrates.

The habitat in which F. edgariana was most commonly encountered was along the left ascending bank, along with the majority of mussels at the site. No specimens of F. edgariana were found along the right bank. F. edgariana was found in areas of moderate to low flows, particularly low in the summer, and seasonally quite shallow. The substrate most commonly associated was sand, gravel, and either silt or pebble depending on proximity to the left bank. Quadrats in which F. edgariana were found included a

fairly high number and diversity of other mussels. No more than three F. edgariana were collected in a single 0.5m<sup>2</sup> quadrat sample. Specimens were collected along the entire length of the left bank, an area of approximately 1000m<sup>2</sup>.

### Stream Drift

Freshwater mussels of the subfamilies Ambleminae and Unioninae are short term breeders, typically releasing glochidia and completing their metamorphosis on host fish during spring or summer (Lefevre & Curtis 1910). Ortmann (1921) noted that several short-term breeding mussels, including many Fusconaia species, were gravid from May to July in the upper Tennessee River drainage. Yokley (1972) found F. undata and P. cordatum gravid in July in the upper Tennessee River. Sickel and Chandler (1982) found gravid F. undata in March and April, and in June through August in Kentucky Lake. Fusconaia ebena was also present in Kentucky Lake, but no gravid individuals were collected. Ortmann (1914) reported F. undata gravid in September and March in Arkansas.

At North Holston Ford, gravid F. edgariana females were first collected in early June and through July. During this period all four gills were observed to carry glochidia, a characteristic of the genus Fusconaia (Ortmann 1921). Gravid female P. oviforme and L. dolabelloides were

collected at North Holston Ford from early June through July, but only the outer gills carried glochidia. Weaver (1981) reported gravid P. oviforme, with only the outer gills charged, in May and June at Big Moccasin Creek. Only one gravid female F. barnesiana was collected at North Holston Ford, which released conglomerates in the laboratory aquaria, and was not examined internally. At North Holston Ford, the species of short-term breeders, F. edgariana, P. oviforme, and L. dolabelloides, were found to be gravid for two to three months in late spring and early summer. F. barnesiana may also be gravid during this time, but not enough specimens were collected to indicate the gravid period for this species at North Holston Ford.

The youngest gravid specimen of F. edgariana was aged at six years, and the oldest gravid specimen was over twenty years old. The exact age of the oldest gravid female could not be readily determined on the live mussel due to the eroded condition of the valves. Sickel and Chandler (1982) found gravid F. undata from five to thirteen years of age; most mussel species in Kentucky Lake were gravid at five years of age. Sexual maturity for Pleurobema cordatum (Matteson 1948), Pleurobema oviforme (Weaver 1981), and Amblema plicata (Stein 1971) occurs at four years of age.

Conglomerates were collected from all four species of

short-term breeders at North Holston Ford, the size, shape and color of which were species specific. Ortmann (1911) stated that some mussel species released glochidia in conglutinates that maintained the shape and size of the water tubes in which they developed. Although identification of live specimens based on shell morphology is often difficult in the Fusconaia/Pleurobema/Lexingtonia complex, species identification based on conglutinal masses is quite obvious and definitive.

Conglutinates of F. edgariana were colored pink and of a subcylindrical shape. Ortmann (1921) described the glochidia of F. edgariana as pink to red, but no specimens darker than pink were observed at North Holston Ford. The conglutinal shape of all Fusconaia species was described by Ortmann (1921) to be subcylindrical, but differed in color and size according to species. He described the conglutinates of F. barnesiana as being dark purple, yet those collected from the specimen at North Holston Ford were yellow to brown. Ortmann's description of P. oyiforme conglutinates as lanceolate and compressed agreed with observations of conglutinal shape from the study site and Big Moccasin Creek (Weaver 1981). Ortmann indicated the color of P. oyiforme conglutinates as cream to pale orange, but the conglutinates of P. oyiforme from North Holston Ford and Big Moccasin Creek (Weaver 1981) were always

white. The shape of L. dolabelloides conglomerates was noted as cylindrical by Ortmann (1921). The shape was confirmed in this study, but the red to orange color indicated by Ortmann was not the pink noted at North Holston Ford. It would seem that although the size and shape of conglomerates are indicative of a species, the color is variable and not a reliable distinguishing characteristic.

Glochidia of the four species at North Holston Ford were identified to species by differences in glochidial valve dimensions. Length, width, and hinge length measurements of each of the species indicated the species could be differentiated by comparing relative sizes of one or more of these dimensions. Length and breadth measurements of this study were greater than those reported by Ortmann (1921). Measurements of P. oviforme from North Fork Holston River (length=167um, breadth=164um) were comparable to those from P. oviforme (length=168um, breadth=161um) from Big Moccasin Creek (Weaver 1981). These measurements are definitely larger than those reported by Ortmann (150um; 170um). Differences in glochidial measurements probably resulted from differences in measuring accuracy.

Stream temperature has been implicated as a cue for the release of glochidia (Matteson 1955, Zale 1980, Weaver

1981), and may be the controlling factor. Peak densities of glochidia were found at North Holston Ford after stream temperature reached 20 C. F. edgariana was most abundant in the drift after water temperatures reached 26 C. Weaver (1981) noted a similar response by P. oviforme at 24 C in Big Moccasin Creek. Maximum water temperatures of 29 C were noted during the two periods of peak glochidial release by F. edgariana in late July and early August. P. oviforme and L. dolabelloides also reached peak densities during this period of high water temperature.

Diel glochidial release may also be affected by water temperature. Peak numbers of drifting glochidia occurred during the early to late afternoon, which coincided with the period in which the stream was the warmest. Stream temperature also was coolest at the onset of morning, which was the period when glochidial density in the drift was lowest. Abundance of glochidia in the drift appears to follow a similar pattern as the changing water temperature through the day. Although glochidia were present in the water column during the entire sample period, a substantial increase in the density of drifting glochidia from mid-morning through late afternoon appears to indicate periods of peak glochidial release occur during a 24-hour period, rather than constant glochidial release.

Slight differences in seasonal trends for glochidial

release were indicated for the short-term breeding species between the two years of this study. Peak releases were observed to occur later during the summer of 1982, and total densities of drifting glochidia were less. Water temperature was slower to warm in 1982 and may account for later peak releases the second summer. Stream discharge and maximum water temperature also differed between years at North Holston Ford. Discharge was below normal in 1981 and above normal in 1982 (Virginia State Water Control Board 1983). Whether stream flows or water temperature influenced the abundance of glochidia in the drift at North Holston Ford is unknown. Peak releases of glochidia appeared to shift in accordance with shifts in peak water temperatures.

Individual females were observed to release glochidia in the laboratory over a three to seven day period. Matteson (1955) also observed that stream mussels commonly expel glochidia over a period of one to three days or longer. Gravid females in the laboratory were maintained at temperatures cooler than those in the stream, which could account for the longer release period observed in the laboratory. Matteson (1955) stated that completion of the reproductive cycle of mussels is highly dependent on a favorable temperature cycle. Fisher and Tevesz (1976) found that changing temperature and changing dissolved

oxygen levels influenced the development and expulsion of glochidia in the marsupia of gravid females. Each step along the cycle from initial fertilization to completion of glochidial metamorphosis appears to be influenced by water temperatures and other environmental conditions (Sickel and Chandler 1982).

#### Determination of Fish Host

##### Natural infestations on fish

F. edgariana was found encysted on one-third (four species) of the infested cyprinids at North Holston Ford. All but one of these species were also infested with glochidia of P. oviforme and L. dolabelloides. Averaging all four species, the infestation rate for F. edgariana on cyprinids at North Holston Ford was 6.45%. This is comparable to the rate of 6.2% reported by Weaver (1981) for P. oviforme on cyprinids at Big Moccasin Creek.

Numbers of fish observed with F. edgariana glochidia appears to be a better indicator of potential host species for this study. The whitetail shiner had the greatest number of fish infested, as well as the highest percentage of F. edgariana encystment. In addition, this was the only

species collected with a specimen infested only with the glochidia of F. edgariana. These factors would seem to indicate a higher probability of the whitetail shiner being a host fish than other implicated species at the site. The telescope shiner would seem to be a less probable host because of the few specimens collected with F. edgariana on the gills, and the presence of multi-species infestations. The probability of the common and warpaint shiners as host species is not well indicated. The number of infested fish of these two species was low compared to the other infested cyprinids at the site, but the infestation rate of F. edgariana at North Holston Ford appears low on all fish, whether host or nonhost.

Glochidia of P. oviforme occurred on 75% of the cyprinids infested at North Holston Ford. Weaver (1981) found P. oviforme encysted on 55% of the cyprinid species in Big Moccasin Creek, a tributary of the North Fork Holston River. P. oviforme was found encysted on the whitetail, common, and warpaint shiners, and stoneroller, bluntnose minnow, and river chub in Big Moccasin Creek. Of the six fish species encysted with P. oviforme glochidia in Big Moccasin Creek, and nine species in North Holston Ford, five species, the stoneroller, river chub, and whitetail, common, and warpaint shiners carried glochidia of P. oviforme at both locations. Since over 50% of the cyprinid

species at two different locations were implicated as potential fish hosts, the field data appear to indicate multiple host specificity.

In addition to the five cyprinid species common to both streams, four other cyprinid species at North Holston Ford and one additional cyprinid in Big Moccasin Creek also carried glochidia of P. oviforme. These infestation data indicate that P. oviforme can utilize a variety of similar hosts, making it adaptive to similar systems, as well as a variety of hosts within a system. Weaver (1981) postulated that P. oviforme could infest several fish species to insure adequate reproductive success, therefore increasing the potential of the species to survive. The ability of P. oviforme to use a number of similar fish species in various river systems would also contribute to increasing the reproductive success of the species.

It is not known whether all of the species naturally infested would have successfully served as hosts. The infestation of a majority of the cyprinid species in Big Moccasin Creek and the North Fork Holston River may not be indicative of the extent to which successful metamorphosis can occur. However, large numbers of infested fish, of several species, provides evidence for probable hosts.

Glochidia of Lexingtonia dolabelloides were found encysted on approximately 64% of the encysted shiners at

North Holston Ford. Three of the six species harbored only glochidia of L. dolabelloides, while the others carried glochidia of both L. dolabelloides and P. oviforme. It is not known whether these species could actually host both P. oviforme and L. dolabelloides or would have sloughed the glochidia of one or both species. No fish host information is available for L. dolabelloides \ therefore, comparisons with fish species from other studies and implicated hosts from this study are not possible.

Host-mussel specificity by the short-term breeding mussel species appears to be exhibited at North Holston Ford within a group (Cyprinidae) of similar host species. The use of a variety of fish hosts to increase reproductive success in a river or to ensure success in a variety of river systems may result in overlap of the fish species encysted within and between river locations.

Encystment rates of glochidia may be another factor contributing to the survival of a mussel species. Fusconaia edgariana had very few encysted glochidia on the fish gills, ranging from one to three glochidia per fish. Infestations approaching 3000 glochidia on an individual fish have been reported for the freshwater drum (Surber 1912), and numbers over 1,000 have been observed on trout (Murphy 1942) and yellow perch (Tedla & Fernando 1969).

Although encystment for P. oviforme at the site ranged

from low to high, over half of the fish carrying glochidia of P. oviforme were encysted by only one glochidium. Number of encysted glochidia varied from low to moderate for L. dolabelloides, with one-fourth of the encysted fish carrying only one glochidium. Fish that carried glochidia of both of these species had encystment rates which ranged from low to moderate, and were quite variable in the number and type of glochidia on individual fish. High encystment rates on a single fish were usually represented by a single mussel species.

Number of encysted glochidia, numbers of fish infested, or specificity of infested fish can be used to implicate fish host species. A low encystment could represent incidental infestation on a fish species, while higher encystment may indicate actual hosts. E. edgariana was always observed in low numbers on potential hosts. One to three glochidia per fish can be considered an incidental infestation, or low natural occurrences on host species. Widlak (1982) attributed infestations of up to three lampsiline glochidia as incidental on cyprinids. Since E. edgariana has reproduced within the last three years at North Holston Ford, all of the observed infestations are probably not incidental. It may be that E. edgariana has an intrinsically low degree of encystment, and the number of encysted glochidia may not be a good indicator of

potential host species for F. edgariana.

In the laboratory, low numbers of viable glochidia were released from adult F. edgariana relative to numbers of viable glochidia from P. oviforme and L. dolabelloides. On the average, 70% of the shiny pigtoe glochidia were not viable; only 30% of those released had the capability of encystment. This could account for a much lower encystment by F. edgariana as compared to the range of encystment for the other two species found on cyprinids at North Holston Ford.

#### Laboratory Induced Infestations

Laboratory techniques used in the infestation experiments were those developed and successfully used by Zale (1980) and Weaver (1981) for other Cumberlandian mussels. The primary modification of these procedures was that gravid females were not sacrificed to procure glochidia. The endangered species permit to handle F. edgariana did not allow sacrifice of specimens to obtain glochidia.

Mussels were induced to release glochidia by stressing them under conditions of altered water temperature or flow conditions. The exact stimulus for glochidial release was not determined in this study, but water temperature, flow rates, and photoperiod may be factors influencing glochidial release in mussels (Zale 1980). Matteson (1955)

indicated that during the period when gills act as marsupia, females are more sensitive to lowered oxygen content and elevated water temperature. Gravid females were also reported to abort conglomerates when stressed, especially with increasing temperature (Matteson 1955).

It is not known whether mussels in the lab aborted glochidia because of stressful conditions, or whether manipulation of temperature and flow triggered natural release of glochidia. Some gravid females were observed to release conglomerates in laboratory aquaria without manipulating flow or temperature, whereas other mussels released glochidia when water temperature was elevated or flow rate decreased. The number of conglomerates released seemed to vary between situations. Mussels under "natural" laboratory conditions seemed to release conglutinal masses continually one or two at a time, over a period of hours to days. Conversely, females placed in jars of warm water with restricted flow released large numbers of conglomerates in a short period of time.

In the field, females were never observed releasing conglomerates, although a few gravid females expelled conglomerates upon removal from the substrate. This response was attributed to handling stress. Other mussels released conglomerates after being placed on ice for transport to the laboratory. It appears that various

factors can produce glochidial release, either singularly or in concert, although exact stimuli are unknown. There may be an adaptive value in mussels relying on a variety of stimuli to initiate glochidial release when considering the variable environmental conditions of flowing water systems.

A major problem encountered after conglomerates of F. edgariana glochidia had been collected was low numbers of viable glochidia. Conglomerates of F. edgariana consisted primarily of unfertilized eggs and relatively few viable glochidia, attributed to low fertilization. Matteson (1948) observed very few unfertilized ova in the marsupia of Anodontoides ferussianus following fertilization. Zale (1980) and Weaver (1981) noted almost complete fertilization in conglomerates of the mussels they studied.

A comparison of the number of glochidia to unfertilized eggs for the four species studied at North Holston Ford indicated that F. edgariana had a much lower percentage of viable glochidia than the other three species. Fusconaia barnesiana, Lexingtonia dolabelloides, and Pleurobema oviforme approached 100% viable glochidia per conglomerate, while F. edgariana averaged only 30% viability. Even the highest fertilization in some specimens of F. edgariana was less than half the rate of the other three species at the site.

Low numbers of viable glochidia limited the number of

fish that could be infected, and the number of glochidia available per fish. Weaver (1981) indicated that 15 to 20 fish could be infested with glochidia from one P. oviforme, allowing hundreds of glochidia per fish. Due to the limited number of glochidia, it was decided that fewer fish of each species would be infested, so that more species could be tested as hosts.

Although fish host identification studies have been conducted since the beginning of the century, the emphasis historically has been on commercially important mussels. Howard (1914) identified three centrarchid species, white crappie, black crappie and largemouth bass, as probable hosts for Fusconaia ebena, a commercial mussel in the upper Mississippi River. Coker et al. (1921) added another centrarchid, the green sunfish, and a clupeid, the skipjack herring, as host species for F. ebena. Fusconaia flava, another upper Mississippi River species, also has three centrarchid species listed as hosts; bluegill, white crappie, black crappie (Surber 1913, Howard 1914, Coker et al. 1921).

Four centrarchid species, smallmouth bass, rockbass, bluegill and longear sunfish, were experimentally infested with F. edgariana glochidia. Rejection of glochidia occurred within five days of infestation, indicating that none of these were host fishes. This is consistent with

the observation that on nonhost fish, the rejection of glochidia is rapid and usually occurs within a few days (Zale 1980). The same prompt rejection of F. edgariana glochidia was observed on all other fish species infested in the lab, except the cyprinids.

Lab infestations of cyprinids did not exhibit a rapid rejection period as with other fish species. Periods of encystment ranged from 4 to 14 days for most cyprinid species. Water temperature, health of specimens, and species of mussel or fish are factors which have been suggested as influencing length of encystment (Zale 1980). Howard and Anson (1922), Zale (1980), and Weaver (1981) indicated that water temperature affected the rate of glochidial metamorphosis. Increased water temperatures shortened encystment periods.

Coker et al. (1921) considered the condition of host fish to be a determining factor in the period of glochidial metamorphosis. Howard and Anson (1922) noted that fish succumbing to fungal infections sloughed encysted glochidia. This was also observed by Weaver (1981) and proved to be a complication in her laboratory infestations. An attempt was made to prevent fungal infections in this study by minimizing handling, bathing the fish periodically in antifungal agents, and maintaining aquaria water at lowered temperatures. The incidence of fungal infestation

and mortality due to stress were important factors in the number of fish per species which survived through glochidial encystment. Often only one or two fish survived initial infestations, resulting in second trials for seven of the eleven species. It is not known to what extent glochidial sloughing was influenced by the state of health of the fish species infected. The periods of encystment were approximately the same for both trials of a species, indicating a consistency of response.

Several investigators (Corwin 1920, Tedla and Fernando 1969, Zale 1980) reported that the length of time glochidia are retained in the marsupia affected the length of encystment. All of the mussel species in these studies were long-term breeders of the subfamily Lampsilinae, and no such information is available for short-term breeders. Since F. edgariana is a short-term breeder, the four to six week age of glochidia in the marsupia is probably of little consequence compared to the age of overwintered glochidia of long-term breeders.

Encystment periods have also been noted to vary with the species of mussel (Zale 1980); presumably glochidia exhibit an encystment time characteristic of that species. Encystment periods are not known for other Fusconaia species, and similarities are not obvious when compared to related species. Weaver (1981) found rejection of

glochidia from nonhost species, both cyprinids and others, within a few days of infection. This rapid rejection was observed for F. edgariana on noncyprinid species, but not on nonhost cyprinids. All cyprinids infested with glochidia of F. edgariana carried glochidia at least 4 days. Water temperature may have contributed to some of the differences noted between the studies. Fish with P. oviforme glochidia were maintained in aquaria approximately 5 C warmer than those of F. edgariana. Since temperature influences length of encystment, the faster rejection of P. oviforme glochidia may have resulted from the warmer water temperatures. Shorter periods of metamorphosis were noted by Weaver (1981) for P. oviforme than for any of the fishes infested with F. edgariana. However, these apparent species differences may also be an artifact of water temperature. The experimental evidence is too cursory to indicate whether the variation between encystment periods of P. oviforme and F. edgariana is attributable to differences in environmental conditions.

The species of fish infested apparently can influence the length of time of encystment. Zale (1980) noted longer encystment periods before rejection by fish species closely related to host species. This may account for the longer rejection periods by cyprinid species and more sloughing of glochidia by noncyprinids in this study. Several mussel

species listed by Fuller (1974) were hosted by a number of related fish species. Host specificity may be associated with a group of closely related species as potential hosts, and the actual species utilized dictated by availability of that species at a site, particularly at the time of glochidial release.

There would seem to be an adaptive value for mussels to have a variety of potential hosts rather than single host specificity. The implications are obvious in terms of reproduction and perpetuation of the species. Fuller (1974) listed 46 mussel species and their known or implicated host fishes, and only eight species had only one host listed. So little is known on life histories of freshwater mussels that other host fish probably exist for the majority of these mussels, but have yet to be ascertained. It would appear that single host specificity is rare, and that a mussel species may use a variety of hosts determined by factors such as habitat, behavior, or availability of the fish species.

Laboratory infestations indicated two species as potential hosts for F. edgariana, the whitetail and common shiners. Although no completely metamorphosed juveniles were found, these fish retained glochidia for an extended period. The two species released what has been referred to as pre-metamorphosed juveniles. These embryonic mussels

had the abductor-adductor muscle scars of a juvenile as well as the single muscle scar of a glochidium. Weaver (pers. comm.) indicated she had also observed the occurrence of such prejuvenile mussels just prior to the collection of fully metamorphosed juveniles.

The lack of metamorphosed juveniles may be attributed to the low degree of infestations per fish such that juveniles were missed in the siphoning process. Fully developed glochidia of E. edgariana for use in infections were in low numbers. This number is very low compared to the hundreds and even thousands of glochidia used in the laboratory infestation reported by Zale (1980) and Weaver (1981). The number of glochidia attached to the gills of recently infested fish was also low compared to other studies (Zale 1980, Weaver 1981). Actual glochidial counts were not made due to the stress involved with extensive gill filament examination, and subsequent high mortality.

Fish were monitored periodically for presence or absence of attached glochidia and sacrificed a few days after glochidia no longer appeared in the siphonate to verify absence of glochidia. Fish were also sacrificed to verify species identification. A few of the cyprinid species were not readily distinguishable and required extensive examination to verify the species. Therefore, fish were separated to species, prior to laboratory

experiments, and those in doubt were verified at the termination of the infestation experiments. The majority of the cyprinids were easily identified to species. Problems with maintaining healthy fish and limited numbers of fish were more consequential than taxonomic considerations at the time of laboratory infestations.

Field studies showed the presence of F. edgariana glochidia on two of the species implicated in the laboratory infestations. The lab and field components of this study indicate that cyprinids are important host species for short-term breeding mussels. Whether the other two species implicated from the field studies are also hosts was not indicated by laboratory results, but does not exclude them from consideration as potential hosts.

The large number of centrarchids listed by Fuller (1974) as hosts for Fusconaia and other related species would seem to indicate that in the Mississippi River drainage, centrarchids serve as important host species for mussels. The results of this study and those of Weaver (1981) showed that cyprinids were important host species for short-term breeders of the Tennessee River drainage. Differences noted in host preference between the Mississippi and Tennessee Rivers may be a function of habitat availability and utilization by fish and mussels. Cumberlandian mussels are characteristically headwater

species, while those in the Mississippian fauna are large river species. Zale (1980) postulated that mussel and fish host relationships have evolved through mutual habitat preference or habitat use by host fish. Whether dictated by mutual preference or limited by use of habitat, it seems that habitat considerations are important in determining mussel-fish host interactions.

#### Age and Growth

Two techniques were used to age shells of F. edgariana, measurement of external growth rings and examination of internal rings via shell thin-sectioning. The two methods were in agreement for mussels from six to twelve years of age, which comprised the majority of specimens at North Holston Ford. Few old specimens and even fewer young specimens were found during this study. Most mussels were collected by waterscopes and hand picking, which may account for the paucity of young mussels collected (Haukioja and Hakala 1978). Fewer than 1% of the F. edgariana collected were less than three years of age, and only one of these was located using a waterscope. Fisher and Tevesz (1976) also reported low numbers of mussels in the less-than-three year age class for Elliptio complanata.

Older mussels were consistently underestimated in age

using external growth rings when compared with ages based on thin-sectioning. Haukioja and Hakala (1978) noted difficulty in aging older specimens externally due to proximity of external rings. Growth may become imperceptible in older mussels, due to a dramatic decline in growth rate (Lutz and Rhoads 1982). Inability to distinguish rings externally may account for the underestimated ages of older mussels. Also, the reduced growth rate of older mussels cause the age classes to overlap in sizes, thereby making the older age classes harder to distinguish by using age-length relationships to predict age (Lutz and Rhoads 1982).

Internal sections of shells appear to yield more accurate values of mussel age than external ring counts (Moyer 1984). The projection and magnification of shell sections increased the ability to distinguish and differentiate annuli and eliminate externally confusing growth rings. It was noted that some mussels which were difficult to age externally were also difficult to age by thin sectioning.

The von Bertalanffy growth model was found to best describe the length-age relationship of E. edgariana. Lengths of mussels in the middle size range were predicted quite accurately, while younger and older mussels were either under or overpredicted in length. These

discrepancies may be attributed to large sample sizes for mussels in the middle range, and few individuals of the young and old age classes. Small sample size could easily cause predictability to either under or over estimate the length-at-age.

A Ford-Walford plot provided a predicted maximum length (L) of 61.8 mm, and the von Bertalanffy growth equation predicted a value of 58.5 mm for F. edgariana. The largest mussel collected was 59.8mm, a value which fell between the two predicted lengths. Moyer (1984), in studying the age and growth of F. edgariana at North Holston Ford, used the von Bertalanffy growth equation and predicted a maximum length of 62.5 mm from thin-sectioned shells. This again indicates an underestimation of age using external aging techniques. Since no age and growth information is available for other populations of F. edgariana, comparisons of maximum age, length at age, and growth rates are not possible at this time.

Sexual dimorphism was not observed in F. edgariana, and no differences were noted between sexes in shell length, width, or overall size; shell length was not statistically different between genders. Female mussels have often been described as having shells more inflated than those of males (Stein 1971). Although shell depth (thickness) was not measured, inflated valves were not

observed in the shiny pigtoe. Gender of the shiny pigtoe could only be determined when the females were gravid, and all nongravid mussels at this time were considered to be males. Since gravid mussels are females, and nongravid mussels were presumed males, then based on the total number of specimens collected during the period of gravidity at North Holston Ford, the sex ratio was roughly equal for the shiny pigtoe during 1981 and 1982.

## CONCLUSIONS

1) Fusconaia edgariana is a short-term breeder with gravid females occurring in June until August at North Holston Ford.

2) F. edgariana comprised 4.7% of the mussels collected at North Holston Ford, indicating a density of 0.5/m<sup>2</sup> and an estimated population of 1,465 adult mussels.

3) F. edgariana was found in shallow water with low to moderate water velocities, in areas of mixed sand, gravel, and pebble substrate.

4) Glochidia were released into the water column from mid-June to early August, with peak expulsion in July.

5) Glochidial dimensions (um) for F. edgariana were 172.5±7.95 (width), 143.06±7.95 (length), 116.18±7.43 (hinge length), and could be differentiated from other species by differences in shell dimensions.

6) Naturally encysted glochidia of F. edgariana were found only on cyprinid species at North Holston Ford; 27 fish were found with F. edgariana encysted.

7) The whitetail, warpaint, common, and telescope shiners were found to carry 1 to 3 glochidia of F. edgariana per fish.

8) Laboratory infestations implicated the common and whitetail shiners as probable hosts.

9) Low fertilization of eggs was observed in females of F. edgariana, averaging 30% viable glochidia per conglutinate.

10) The low fertilization may account for the low incidence of encystment on fishes at North Holston Ford.

11) Specimens of F. edgariana collected at North Holston Ford ranged from age 3 (12.8mm) to age 24 (59.5mm).

12) Predicted maximum length was 58.5mm and 61.8mm using the von Bertalanffy growth equation and a Ford-Walford plot, respectively; the largest specimen at the site was 59.8mm in length.

12) Age estimates by two aging techniques (external shell ring counts and shell thin-sectioning) indicated that the external aging method underestimated ages of older specimens (over age 15).

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APPENDIX TABLES

Table 1. Summation of water quality data North Fork Holston River mile 85.2 (Poppe 1982).

Variable	N	Mean	Standard deviation	Minimum value	Maximum value	Std error of mean	Sum	Variance	Coefficient of variation
Alkalinity (phenolphthalein)	0								
Alkalinity (total)	17	114.9	35.0	56.0	150.0	8.5	1,953.0	1,226.5	30.5
Aluminum	10	146.8	70.0	70.0	300.0	22.1	1,468.0	4,898.0	47.7
Ammonia N	18	0.0	0.0	0.0	0.1	0.0	0.7	0.0	46.8
BOD (5-day)	14	1.5	0.9	1.0	3.8	0.2	21.3	0.8	58.7
Boron	0								
Cadmium	10	0.1	0.1	0.1	0.3	0.0	1.3	0.0	51.9
Calcium (total)	10	32.6	11.9	16.7	58.0	3.8	326.0	142.7	36.6
Chloride	18	3.9	2.2	2.0	12.0	0.5	71.0	4.6	54.6
Chromium	10	1.1	0.3	1.0	2.0	0.1	11.0	0.1	28.7
Conductivity	18	246.1	67.2	140.0	320.0	15.8	4,430.0	4,519.3	27.3
Cobalt	0								
COD	18	6.2	2.7	2.0	13.0	0.6	111.0	7.2	43.0
Copper	10	10.3	9.1	1.0	30.0	2.9	103.0	82.5	88.2
Dissolved oxygen (mg/L)	16	9.2	2.4	7.0	14.0	0.6	147.1	5.8	26.2
Hardness	18	126.4	34.6	69.0	160.0	8.2	2,276.0	1,195.7	27.3
Iron (dissolved)	10	85.0	96.7	13.0	350.0	30.6	850.0	9,358.7	113.8
Iron (total)	10	231.0	106.5	70.0	440.0	33.7	2,310.0	11,337.8	46.1
Lead	10	3.2	3.6	1.0	13.0	1.1	32.0	12.8	112.0
Lithium	0								
Magnesium (total)	0								
Manganese (dissolved)	10	36.3	64.9	8.0	220.0	20.5	363.0	4,212.5	178.8
Manganese (total)	10	28.3	19.6	10.0	60.0	6.2	283.0	384.5	69.3
Mercury	10	0.2	0.1	0.2	0.6	0.0	2.4	0.0	52.7
Nickel	0								
Nitrogen (organic)	18	0.1	0.1	0.0	0.4	0.0	1.9	0.0	97.6
NO <sub>2</sub> + NO <sub>3</sub>	18	0.6	0.2	0.3	1.4	0.1	10.8	0.1	39.7
pH	17	7.6	0.3	6.8	8.1	0.1	128.9	0.1	3.9
Phosphorus (dissolved)	0								
Phosphorus (total)	18	0.0	0.0	0.0	0.0	0.0	0.3	0.0	49.4
Residue (total)	18	135.0	32.0	80.0	180.0	7.6	2,430.0	1,026.5	23.7
Residue (dissolved)	18	6.9	4.0	1.0	13.0	0.5	124.0	16.0	58.0
Selenium	10	1.0	0.0	1.0	1.0	0.0	10.0	0.0	0.0
Silver	0								
Sulfur sulfate	10	1.6	0.4	1.0	2.0	0.1	15.9	0.1	23.0
Temperature	18	10.5	2.9	4.0	15.0	0.7	189.2	8.3	27.5
Titanium	16	16.5	7.0	3.1	24.0	1.7	263.4	48.8	42.5
TCC	0								
Turbidity	18	2.8	1.3	1.7	7.8	0.3	51.2	1.8	47.0
Zinc	10	22.9	41.3	5.0	140.0	13.1	229.0	1,709.7	180.6

Appendix Table 2. Species and number of mussels collected in quadrat samples at North Holston Ford. (\* denotes species not found in quadrat sample)

Species	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	
<b>Ambleminae</b>																					
<u>Fusconaia barnesianax</u> *				1			1														
<u>Fusconala edgariana</u>																					
<b>Lampsilinae</b>																					
<u>Actinonaias pectorosa</u>																					
<u>Lampsilis fasciola</u>				1																	
<u>Lampsilis ovatax</u>								1													
<u>Medionidus conradicus</u>	2		3	1	1	1	1	2		4	6	2	4	4	2	1	1	3	1		
<u>Ptychobranchus fasciolaris</u>	1				1			1													
<u>Ptychobranchus subtentum</u>			1	3		1	4	1		3	2		1	2		2		1		1	
<u>Villosa nebulosa</u>			3	1			2			4	1	1	2								
<u>Villosa vanuxemi</u>																					
<b>Unioninae</b>																					
<u>Lexingtonia dolabelloides</u>					1	1															
<u>Pleurobema oviforme</u>	1	3	2		1	1	1	3	1												
Juveniles (unidentified)				2	1	1		2		2				1							
Total	4	0	12	11	4	2	12	5	6	7	14	2	9	1	8	2	3	2	3	2	

Appendix Table 2 (continued). Species and number of mussels collected in quadrat samples at North Holston Ford. (\* denotes species not found in quadrat sample)

Species	Quadrat Number																			
	21	22	23	24	25	26	27	28	29	30	31	32	33	34	35	36	37	38	39	40
Ambleminae																				
<u>Fusconaia barnesiana*</u>																				
<u>Fusconaia edgariana</u>	1	2	1	1	1	1	2	1	2										1	3
Lampsilinae																				
<u>Actinonaias pectorosa</u>	1																			
<u>Lampsilis fasciola</u>																				
<u>Lampsilis ovatax</u>																				
<u>Medionidus conradicus</u>	5	1	1	1	1	1	4	2	1	1	1	4	2	3	3	1	8	1	3	
<u>Ptychobranchus fasciolaris</u>	1	2		1	2	2				3			1				1			
<u>Ptychobranchus subtentum</u>	3	1	2	4	1	1	2	3	1	3	2	3	2	3	2	1	1	1	1	2
<u>Villosa nebulosa</u>	1	1	3			1							4	1	3	2				
<u>Villosa vanuxemi</u>																				
Unioninae																				
<u>Lexingtonia dolabelloides</u>		4													3					1
<u>Pleurobema oviforme</u>		4	1	1	1					2	1						1	1		
Juveniles (unidentified)	2	1										2	1							
Total	12	5	16	5	7	4	6	5	4	9	5	5	12	6	9	10	3	13	3	8

Appendix Table 2 (continued). Species and number of mussels collected in quadrat samples at North Holston Ford. (\* denotes species not found in quadrats)

Species	41	42	43	44	45	46	47	48	49	50	51	52	54	55	56	57	58	59	60	
<b>Ambleminae</b>																				
<u>Fusconaia barnesianax</u>																				
<u>Fusconaia edgariana</u>																				
<b>Lampsilinae</b>																				
<u>Actinonaias pectorosa</u>										1									1	
<u>Lampsilis fasciola</u>																1				
<u>Lampsilis ovatax</u>																				
<u>Medionidus conradicus</u>	4		2	2	4	4	1	4					2	3	1	2	1	1		
<u>Ptychobranchus fasciolaris</u>					1															
<u>Ptychobranchus subtentum</u>	2					1						2						2		
<u>Villosa nebulosa</u>	1		1		1		1					1	1					1	1	
<u>Villosa vanuxemi</u>	2		1					1					1							
<b>Unioninae</b>																				
<u>Lexingtonia dolabelloides</u>																				
<u>Pleurobema oviforme</u>	1												2	1	2				2	
Juveniles (unidentified)																			1	
Total	10	2	4	2	6	5	2	5	1	3	1	4	8	3	4	4	6	2	1	2

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LIFE HISTORY OF THE ENDANGERED SHINY PIGTOE

PEARLY MUSSEL, FUSCONAIA EDGARIANA,

IN THE NORTH FORK HOLSTON RIVER, VIRGINIA

by

Helen Elise Kitchel

ABSTRACT

The life history of Fusconaia edgariana, the endangered shiny pigtoe pearly mussel, was determined in a two year study at North Holston Ford, North Fork Holston River (NEHRM 86.9) above Saltville, Virginia. Sixty 0.5m<sup>2</sup> quadrats indicated a mean density of 10.6 mussels/m<sup>2</sup>, representing 10 species. Six additional species were collected by handpicking or in muskrat middens. The density of F. edgariana was 1 adult/2 m<sup>2</sup>. Distribution of the shiny pigtoe was restricted to shallow areas of mixed sand, gravel, and pebble substrate in low to moderate water velocities.

Drift nets (130um mesh) indicated glochidia of F. edgariana were released into the water column from 23 June

to 18 August, 1981 and 24 June to 28 August, 1982. Of the 4,800 fish examined from June 1981 to June 1982, 1.5% carried shiny pigtoe glochidia. The telescope, common, warpaint, and whitetail shiners were found naturally encysted by 1 to 3 glochidia of E. edgariana. Laboratory induced infestations were conducted on twenty-two species of fish, and all but two species sloughed the glochidia within 4 to 14 days. The whitetail and common shiners retained glochidia for 38 days, indicating these two fish species to be probable hosts for the shiny pigtoe.