Wallers 1994

The Nature Conservancy

PROPOSAL EVALUATION FORM
Please return by March 31, 1994

Proposal Title:

Summary Rating: ❑ Excellent ❑ Very Good ❑ Good ❑ Fair ❑ Poor

Reviewer's Signature : R J Neves Reviewer's name (printed): Richard J. Neves

Please evaluate this proposal using the criteria presented in the attached information sheet. Use additional sheets as necessary.

1. Intrinsic merit of the research

The proposed work will undoubtedly provide new information on the diel movement patterns of stream fishes (host-fish dynamics), but the linkage between these movements and "determining mussel distributions" is very speculative. The researchers have taken the somewhat myopic view that fish hosts in essence determine the location and ranges of mussel species in Big Darby Creek and elsewhere. They have seemingly discounted physical habitat heterogeneity, water chemistry, and other factors that could be important in describing the specific patterns of distribution within a river system. This assumption is particularly disconcerting when the authors admit that the "fish hosts are either partially or completely unknown for at least half of them," and they commit to determining hosts for only the 2 federally endangered mussels (*P. clava* and *E. t. rangiana*). Without a fairly complete data set of mussel species and fish hosts, the results will be inconclusive and speculative. The predictive model of mussel and fish distributions may consist of papier-mache rather than concrete. Similarly, the authors speculate that results can be applied to other midwestern stream systems, yet they do not intend to test their model in another system. Because Big Darby Creek system in Ohio "has perhaps the greatest diversity of fishes and freshwater mussels of any system its size in North America," why would results from this superlative system apply to other systems of equal or unequal size? They've selected a Mercedes and presume that the functions and parts are transferable to Hyundai's!

2. Utility or relevance of the research

In my opinion, the most useful data to be collected will be any new host fish identifications that come from this work. In one sense, focusing only on fishes as the regulatory mechanism for mussel distributions and diversity may promote a further bias in mussel ecology, particularly if most mussel species do not have host fishes identified. I am particularly concerned about using host fish determinations from other river systems and applying them throughout the range of a mussel species. As intimated in the literature, additional host species likely occur for mussel populations in geographically or geologically disjunct rivers. The assumption of 3 known hosts for mussel species X in the Missouri River does not translate to 3 hosts in the Ohio River. Our knowledge of fish hosts is so rudimentary that the application of this paucity of data to explain watershed-level distribution patterns of mussels is highly speculative.

The author makes a point of dams as barriers and the opportunity presented by dam removal at 2 sites on the creek. There are numerous analogous studies in the literature on recolonization of fishes into stream reaches previously affected by barriers such as water pollution, toxic spills, etc. The opportunity to assess fish movements upstream of these old barriers is really not new or a "rare opportunity."

3. Research performance and competence

The investigator is well qualified to conduct the proposed research, and facilities at Ohio State should be adequate to support the proposed work. There seems to be too much effort and cost to determine the movement direction of fishes and no effort to define habitats used by host fishes. Both of these are needed if one expects to model the patchy distribution of mussels.

I feel obligated to comment on the hefty budget of this proposal, as a reviewer and as a TNC member. The request for $288,600 for nearly 3 years is excessive for the products that are offered. Two full time GRAs, a full-time technician, 2 half-time technicians, and the involvement of 3 faculty is excessive for this type of project. In my opinion, this heavy cost in labor could be significantly cut by reducing the emphasis on fish trapping, which again is only a piece of the information needed on fishes. Why are 75 aquaria needed?

I noted under Travel (p. 20) that 2 trips to PA are planned to collect gravid female mussels. Are these the 2 federally endangered species? This gets back to one of my original questions: do the populations in PA use the same hosts as the populations in Big Darby Creek? If so much is to be made of this study, then all mussels and host fish testing should occur within the Darby Creek system.

The Nature Conservancy®

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February 4, 1994

Dr. Richard Neves
VA Co-op Fish & Wildlife Research Unit
106 Cheatham Hall
Virginia Tech
Blacksburg, VA 24061-0321

Dear Dr. Neves:

Thank you for agreeing to review this proposal. I realize you have a busy schedule but I would appreciate having it back from you by March 31, 1994. If you have any questions about the please call me at (703) 841-5386.

Sincerely,

Bob Unnasch

The Nature Conservancy®

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Information for Peer Reviewers

The Nature Conservancy is seeking to support the most meritorious research through it Ecosystem Research Program. Your review is essential in our meeting this goal. Please focus your evaluation on the scientific merit of the attached research proposal . Please provide both written comments that may be shared with the researcher(s) and a summary rating on the attached evaluation form.

Please try to address each of the following *proposal evaluation criteria*

1. Intrinsic merit of the research -- Address the likelihood that the proposed research will lead to new discoveries of fundamental advances within its field of science or have substantial impact on the science of Ecology.

2. Utility or relevance of the research -- Address the likelihood that the proposed research can contribute to the achievement of the Conservancy's goal of protecting biodiversity, especially how the expected results may serve as the basis for new or improved conservation technologies.

3. Research performance and competence -- Comment, to the best of your ability, on the capability of the investigator(s), the technical soundness of the proposed approach, and the adequacy of the institutional resources available. Please include comments on the proposer's recent research performance.

Summary ratings:

Excellent: Probably will fall among the top 10% of the proposals; highest priority for support. This category should be used only for truly outstanding proposals.

Very Good: Probably will fall among the top one third of the proposals submitted; should be supported.

x **Good:** Probably will fall among the middle on third of the proposals submitted; worthy of support.

Fair: Probably will fall among the lowest one third of the proposals submitted.

Poor: Proposal has serious deficiencies and should not be supported.

Conflict of interest

If you have an affiliation or financial connection with the institutions or persons submitting this proposal that might be construed as creating a conflict of interest, please describe those affiliations on a separate page and attach it to your review. Regardless of any such affiliations or interests, unless you believe that you cannot be objective, we would like to have your review. If you do not attach a statement we will assume that you have no conflicting interests.

Confidentiality of proposals and peer reviews

The Nature Conservancy has received these proposals in confidence and is responsible for protecting the confidentiality of their contents. In addition, the identity of reviewers will be kept confidential to the maximum extent possible. For this reason, please do not copy, quote, or otherwise use material from this proposal. If you believe that a colleague can make a substantial contribution to the review, please consult Dr. Robert Unnasch, (703)841-5386, before disclosing either the contents of the proposal or the applicants name. When you have completed your review, please destroy the proposal.

Please mail your reviews to:

Robert S. Unnasch, Ph.D.
The Nature Conservancy
1815 North Lynn Street
Arlington, VA 22209

Proposal Summary

For Funding Through The Nature Conservancy's Ecosystem Research Program

Proposal Title
Importance of Host-fish Dynamics and Ecosystem Characteristics in Determining Mussel Distributions

Date Submitted	Date Received	Number of Copies
October 29, 1993		

PI / Project Director	Address	Telephone
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Ohio State University Research Foundation	1960 Kenny Road Columbus, OH 43210-1063

Employer Identification Number(EIN) or Taxpayer Identification Number(TIN)
31-6401599

Requested Amount	Proposed Duration	Proposed Starting Date
$288,600	2.75 years	March 1, 1994

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Project Summary

The freshwater mussel diversity of North America is declining rapidly. Impoundment, toxins, and over-harvesting have driven over a dozen species to extinction, and at least twice that number face the same fate in the next decade. Currently 42 species of mussels are considered federally endangered. The conservation of these species requires the establishment and maintenance of suitable habitat and water quality. However, this is not sufficient in itself to manage a mussel population. Freshwater mussels use fishes as hosts for their parasitic larvae, the glochidia. The relationship is obligatory, and even healthy mussel populations will eventually die out without recruitment if the host fish is absent. Unfortunately, the host or hosts for most mussels are not known. This is particularly true of rare and endangered species.

The Big Darby Creek system in Ohio, a Last Great Place, has perhaps the greatest diversity of fishes and freshwater mussels of any system its size in North America. To adequately understand the distribution and causes of decline of mussels in the Big Darby Creek, our knowledge must go beyond microhabitat characteristics and include information about the larger ecosystem. We propose to take a broad perspective in studying the distribution of mussels in the Big Darby Creek system by first looking at whole-ecosystem patterns, next using experiments to evaluate the mechanism leading to these patterns, and finally, using this information to generate expectations about mussel species abundance in other systems. Specifically, we will address the system at four levels. 1) Synthesizing existing information on fish distribution, water quality characteristics, and land use, we will take an ecosystem perspective in predicting mussel distributions. 2) At a more mechanistic level, we will study species-specific patterns of large-scale fish movements and their temporal and spatial overlap with spawning mussels. This will also include a study of the impact of dams as barriers to fish movement, and consequently, to mussel distributions. 3) Concurrent with the above study, we will use laboratory experiments to identify fish species that can serve as hosts to these species of mussels. 4) Using information from our studies of mussel-fish relationships, we will build a simulation model to help understand the previously observed patterns in mussel/fish species abundances and predict baseline abundances for other systems..

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Introduction

The Big Darby Creek system in Ohio, a Nature Conservancy Last Great Place, has perhaps the greatest diversity of fishes and freshwater mussels of any system its size in North America (Watters, in press a & b). These faunae have been intensively sampled and the distributions of both are fairly weii understood, again, to a level not found in any other system of its size. In addition, data on water quality, stream morphology, siltation, and land use are available for much of the system. These data offer a unique opportunity to study the relationships between mussels, their fish hosts, and their physical environment in a manner that could not be attained in more degraded or less surveyed systems. Basic insights, applicable to any river system, may be derived from such a study. These insights will be important in allowing us to identify habitat requirements of endangered mussel species, to characterize potential hosts by ecology and availability, to distinguish land use practices that adversely affect mussels and aquatic organisms in general, and recognize potential translocation sites based on biological and physical parameters.

The freshwater mussel diversity of North America is declining rapidly. Impoundments, toxins, and over-harvesting have driven over a dozen species to extinction, and at least twice that number face the same fate in the next decade. The recent introduction of the zebra mussel is believed to be a grave threat to native species, the impact of which yet has scarcely been felt. Currently 42 species of North American mussels are considered federally endangered. The conservation of these species requires the establishment and maintenance of suitable habitat and water quality. This is a difficult and complex task involving large scale land management, cooperation between landowners and conservationists, and continuous monitoring. However, this is not sufficient in itself to manage a mussel population.

Freshwater mussels use fishes as hosts for their parasitic larvae, the glochidia. This relationship is obligatory, and even healthy mussel populations will eventually die out without recruitment if the host fish is absent. Unfortunately, the host or hosts for most mussels are not known. This is particularly true of rare and endangered species. Research indicates that some mussels may use numerous, widespread hosts, while others are confined to parasitizing a single species or a small, ecologically similar group of fishes. However, our knowledge of this relationship is largely fragmentary.

The distribution of mussels is a product of their environment, including both biotic and abiotic factors. In theory, if we know all of the ecological requirements of a species, we could predict where that species occurs. The zoogeography of mussels is complicated by the fact that their overall distribution depends not only on appropriate habitat, but also on the movement of their host fishes. The zoogeography of fishes is likewise complicated by the migration habits of many species. Habitat data thus have both temporal and spatial components. Because species distributions have been measured for fishes and mussels in the Big Darby Creek system, it is possible to compare zoogeographic ranges within a single system on a scale not possible in other drainages. Basic data, previously unknown, can be gathered on the relationship between these hosts and parasites, such as the importance of fish migration, mussel spawning times, ecological similarity, and strategies used to infest hosts.

Approach

To adequately understand the distribution and causes of decline of mussels in the Big Darby Creek, our knowledge must go beyond microhabitat characteristics and include information about the larger ecosystem. We propose to take a broad perspective in studying the distribution of mussels in the Big Darby Creek system by first looking at whole-ecosystem patterns, next using experiments to evaluate the mechanism leading to these patterns, and finally, using this information to generate expectations about mussel species abundance in other systems. Specifically, we will address the system at four levels: 1) synthesizing existing information on fish distribution, stream bed characteristics, water quality data, and land use patterns, we will take an ecosystem perspective in predicting mussel distributions, 2) at a more mechanistic level, we will study species-specific patterns of fish movements and their temporal and spatial overlap with spawning mussels (this will also include a study of the impact of dams as barriers to fish movement, and consequently, to mussel distributions), 3) concurrent with the above study, we will use laboratory experiments to identify fish species that can serve as hosts to these species of mussels, 4) using information from our studies of mussel-fish relationships, we will build a simulation model to help understand the previously observed patterns in mussel/fish species abundances and predict baseline abundances for other systems.

Background and Methods

Choice of Species

Although the Big Darby Creek system has 40 species of mussels recorded from it, the hosts are either partially or completely unknown for at least half of them. Many of these species are uncommon or rare elsewhere, and occur only in high quality water streams such as Big Darby. These include seven Ohio endangered and two federally endangered taxa. The declining numbers of these (and most) mussel species point out the need to understand their ecology and life history before they have vanished. This study will focus on these rare species, particularly *Epioblasma torulosa rangiana* (northern riffle shell) and *Pleurobema clava* (northern club shell), which were listed as federally endangered on 22 February 1993 (50 CFR 17), and their surrogates. If time permits, the study also will include additional Ohio endangered species. This study will address directly the Federal Recovery Tasks 1.41, 1.42, 2.1, and 4.1 towards down- or delisting. These tasks have priority 1 and 2 listings, and The Nature Conservancy has been given as a responsible implementing agency for several of these tasks.

Ecosystem Synthesis

Because the Big Darby has been extensively studied for decades, habitat data for Big Darby Creek probably exceed that of any other midwestern system. We are taking advantage of the existence of data from a number of systematic surveys (fish, insects, mussels, water quality) as well as a more varied, but long-term data set (fish) to help explain and predict mussel and fish distributions (Table 1). We will use these data to answer a hierarchy of questions: 1) Can we correlate limits of mussel and fish species distributions with water quality and physical habitat characteristics? 2) Do species distributions correlate better with indices of aquatic community health (e.g., Index of Biotic Integrity (IBI)) which may better represent the history of water and habitat quality at a site than one-time physical measurements? 3) Can we correlate limits of mussel species distributions with distributions of certain fish species during periods of larval mussel release and use this data to help identify potential hosts of the mussel glochidia? 4) When we combine fish distributional data, water and habitat quality data, and community health data with land-use data into a picture of the whole ecosystem, can we generate a predictive model of mussel and fish distributions? A predictive model such as this would allow us to take advantage of the extensive data from the Big Darby system and apply it to other midwestern stream systems. Such

Table 1. Description of historical survey data available to be used in the ecosystem synthesis.

Data	Years	Source
Mussel distribution, 76 sites on Little Darby and Big Darby Creeks	1986	Watters 1986
Mussel distribution, 100 sites on Little Darby and Big Darby Creeks	1990	Watters 1990
Fish (voucher material) various sites on Big and Little Darby Creeks	1939-1993	OSU MBD[1]
Fish (presence /absence) various sites on Big and Little Darby Creeks	1963-1993	OSU MBD
Invertebrate community indices	1983, 1986	OEPA[2]
Physical habitat characteristics[3]	1980's-90's	OEPA
Fish distributions	1980's-90's	OEPA
Physical characteristics of the watershed, including information from Landsat[4]	1987	TNC[5], ODNR[6], DMA[7]

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[1]The Museum of Biological Diversity at The Ohio State University

[2]Ohio Environmental Protection Agency

[3]qualitative measures of flow conditions,current velocity, channel morphology, bank erosion, riffle development, riffle quality, water clarity, water color, canopy cover, substrate characteristics, riparian vegetation, margin habitat, and land-use.

[4]soil texture, soil erodability, total N and P in sediment, topographic characteristics, cropping and practice factors, Mannings roughness coefficient

[5]The Nature Conservancy

[6]Ohio Department of Natural Resources

[7]Defense Mapping Agency

maps would also be useful in the Big Darby system itself, to identify biologically significant areas along the stream for protection, and to identify potential translocation areas within the system, as called for by the Federal Recovery Plans for most endangered mussel taxa.

The compilation of such diverse data presents computational problems. Recently, geographical information systems (GIS) have been developed to map complex geographical data sets. We propose to use the GIS to overlay the available data and integrate spatial data for the system. The relationships developed between organisms and their habitat will be species-specific, with resulting GIS maps showing areas of the system having a particular set of habitat parameters most correlated with that species. Site specific data for IBI, substrate, water quality, and land use will be coded or ranked and entered into the GIS. The result will be a site-specific map having composite scores for environmental parameters. Distributions of fish (by season) and mussels will be overlain to determine if particular species of mussels and fishes occur at a certain range of scores. Regions having similar scores, but lacking the species, may be potential translocation sites. Because not every mussel species is found in the same habitat, the equations used to generate the GIS map will be taxon specific.

To reach this end, we will retrieve the historical data from the various sources (e.g., OEPA, ODNR, OSU-MBD, TNC; see Table 1). Because data from each source will be in a different form, we must next enter all data into a common database. All data, including species occurrences, physical habitat measurements, water quality information, land-use, etc. will be entered into the database with time and location information. As we collect new data during the duration of this study, we will include it in this form.

Fish Movement Patterns

Long-distance seasonal movements by non-salmonid fishes are poorly documented in the ecological literature. Most evidence is anecdotal. Trautman (1981) interpreted many of his observations of Ohio stream fish as indications of seasonal migrations: upstream movements before spawning, downstream movements before winter. Of the darters found in the Big Darby Creek system, Trautman (1981) indicates that the greenside darter (*Etheostoma blennioides)*, banded darter (*E. zonale)*, variegate darter (*E. variatum*), bluebreast darter (*E. camurum*), and fantail darter (*E. flabellare*) all move upstream to spawn and downstream to overwinter. He describes the same pattern of seasonal movements for at least five species of dace and shiners found in the Big Darby system (Trautman 1981). There is some evidence of short pre-winter movements by Johnny darters (*E. nigrum*) and fantail darters in Ohio streams (Mundahl & Ingersoll, 1983), as well as short upstream spring movements by creek chubs (*Semotilus atromaculatus*) (Storck & Momot, 1981).

An understanding of seasonal movement patterns in non-salmonid stream fish is essential to understanding the dynamics of mid-western stream fish and mussel communities. Without clear knowledge of this spatial and temporal variation in fish distributions, we cannot hope to understand how impacts at specific locations along a stream may affect fish and mussel populations not presently at those sites. Historically, we have had a tendency to preserve a particular type of habitat for a specific fish species. This will not be sufficient. We actually may need to ensure the availability of season-specific habitat as well as the availability of the corridor for movement between these habitats.

If mussels are relying on seasonal fish movements for dispersal of their larvae, then anything that interferes with the fish movements will also interfere with future mussel distributions. There are several factors that might reduce the pool of available fish hosts: overall reduction in numbers of fish of the appropriate species, placement of barriers that block fish movement, existence of inhospitable habitat between fish winter locations and spawning locations, and water quality changes (e.g., anthropogenic temperature changes) that mask the cues for seasonal spawning movements. In this study, we will be measuring the timing and location of fish movements during mussel spawning times, and addressing the impact of barriers as one mechanism that may affect these movements.

Temporal and spatial patterns. We will use portable directional weirs to measure fish movements. A portable weir consists of a small plexiglas and mesh fish trap (25 mm x 25 mm x 50 mm) with mesh wings (6 m long). Traps will be set in pairs, with one trap in each pair open toward upstream and the other trap open toward downstream. Although traps will capture local fishes in addition to fishes moving upstream or downstream, by setting traps in directional pairs, we can control for this and identify trends in the direction of movement of the captured fish. Traps similar to these have been used to assess stream fish and crayfish movement in previous studies (Marschall, unpublished data). At each site, traps will be set across the entire stream, both at the substrate level (to capture benthic fish) and up to the surface (to capture fish swimming through the water column). Traps will be set twice each sample day: during a 6-h daylight interval (late morning to late afternoon) and during a crepuscular/dark interval (evening through morning). If we find that rate of fish capture is much higher during the crepuscular/dark hours, we will further divide this interval into separate dusk and dawn intervals. Captured fish will be identified, measured (total length), and released at the site of capture.

The timing of our measurements of fish movement should correspond to the release of larvae of the two federally endangered mussel species. Although we do not know when either riffleshells or clubshells release larvae in this system, we have some historical information from another system. Ortmann (1919), in Pennsylvania, reported *P. clava* as gravid from May to July, and *E. rangiana* from August through September. We will begin sampling for fish in late April, and will sample weekly until both species have released larvae.

In the first year, we will concentrate our efforts on areas vital to the riffleshell and clubshell and the two locations at which dams have been recently removed. Traps will be set across the stream in six locations: 1) at the site of the former dam at the confluence of Little Darby Creek and Big Darby Creek, 2) at the site of the former dam on the Big Darby, 3) on the Little Darby above the confluence of Spring Run (1990 location of *P. clava*), 4) on the Little Darby below Spring Run (historical site of *P. clava*), 5) on the Big Darby below the confluence with Hellbranch Run (1990 location of *E. rangiana*), and 6) on the Big Darby above the confluence with the Little Darby (historical site of *E. rangiana*).

In the second year, if we have successfully identified hosts of the two federally endangered mussel species, we will concentrate our trapping efforts more intensively on areas relevant to these host species. In addition, we will continue to sample the sites of the former dams.

Although there are several ways to measure fish movement, this passive directional trapping method is the only method that would be feasible for a study area of this size, given what little we know of non-salmonid stream fish migrations. An alternative to this method would be to capture fish, mark individuals or batches, release them, and try to recapture them later to see where they had traveled. Although the number of marked fish and the sample effort required to allow us to rely entirely on this method would be prohibitive, we believe the potential exists to gain some information at a low cost with this method. Consequently, as we sample fish in traps, we will give them individual tags before releasing them. As we sample throughout the stream, over the course of this study, we will record timing and location of any recaptures. Because other biologists also capture fish in this system, (e.g., OEPA, ODNR), we increase the probability of tagged fish being recaptured. To take advantage of the possibility of other agencies reporting recapture information, we must use an easily visible external tag. Although tagging techniques are well-developed for large fish, fisheries biologists are still experimenting with techniques for small fish. We will work with two types of tags, experimenting with them in the laboratory before using them in the field. First, we will use traditional individually numbered anchor tags through the dorsal musculature (Wydosky & Emery, 1983). Next, we will experiment with a new technique, monofilament :spaghetti" tags, each with a unique combination of colored beads (made of drops of fingernail polish) inserted through the dorsal musculature. This method has been used successfully on other small fish (*Poecilia gillii*, Chapman & Bevan, 1990).

In addition to trapping fish that are moving through the stream, we will also sample these sites using seines and electrofishing. As with trapping, this sample effort will occur near the time of larval release by these two species of mussels. Sample sites will coincide with present distributions of adults of the two mussel species.

Effect of dams as barriers. The stream ecology and physical regime of a river are altered by impoundment, often resulting in a loss of original species, and the subsequent colonization of very different ones (Ellis, 1942; Bates, 1962; Neel, 1963; Harman, 1974; Ridley & Steel, 1975; Baxter, 1977; Williams et al., 1992; Parmalee & Hughes, 1993). Aside from these considerations of habitat modification, the importance of dams as distributional barriers to fishes and, indirectly, to mussels has been underestimated. There is circumstantial evidence that dams directly limit the upstream boundaries of several mussel species' ranges (Watters, 1988), including rare and endangered taxa. Big Darby Creek has recently had two dams removed that formed such barriers. A dam at the confluence of Big and Little Darby Creeks (installed in 1950s) may have limited the ranges of both the clubshell and riffleshell. Since its removal, a mussel species not previously recorded from Little Darby has been found above the dam. Several fish species also appear to have moved into Little Darby (D. Rice, ODNR, pers. comm. 21 Oct. 1993). A dam in the middle reach of Big Darby (installed in 1940s) limited the upstream range of riffleshell. The strategic placement of these dams and the timing of their removal make this a rare opportunity to gather important information on the potential for recovery of dam-impacted rivers. We now have the opportunity to test whether changes in species distributions and relative abundances will change with removal of a barrier to fish movement. Such observations would have wide applicability to other systems.

To quantify the effect of dam removal on fish and mussel distributions, we will measure fish movements through the area formerly housing the dams (during the mussel brooding season), and sample fish and mussel species diversity above and below the former dam locations. We will compare fish and mussel distributions to data from before dam placement and before dam removal (previous data from OEPA and OSU-MBD). Samples of fish movements will be incorporated into the study described above (in *Temporal and spatial patterns*).

Fish species diversity will be sampled using seines and electrofishing. Each sample site will consist of a 50-meter section of stream. We will sample two sites above each dam location and two sites below each dam location. Samples will be taken during spring, late-summer to early-autumn, and winter, to ensure inclusion of any seasonal inhabitants. For each species, we will have presence/absence data that can be classified according to three factors: season (spring, summer-autumn, and winter), location (above dam and below dam), and time (pre-dam, dam, and post-dam). By analyzing all three time periods, we can separate the effect of the presence of the dam from the effect of temporal trends in the fish community as a whole.

Both dam locations will be intensively sampled by hand and sieve during favorable conditions for mussels. In choosing sample times and methods, emphasis will be put on collecting juveniles, since this is the stage at which we will see the first signs of changes in distribution. For each species of mussel, we will have presence/absence data for each sample. Each sample will be classified according to time (dam and post-dam) and location (above dam and below dam). We do not have to consider season in this analysis since individual mussel locations are not changing. No comprehensive pre-dam data exist for mussels.

For both fish and mussel samples, we will be dealing with presence/absence data. As with all presence/absence data, presence in a sample ensures presence in the system, but absence in a sample does not ensure absence in the system. We will handle this statistically by using data from different sample sites and years to estimate the probability of absence from a site given known presence in the system. In this way, we can get a measure of confidence in our conclusions of "absence at a site" at any sample time.

Determination of Mussel Host Species

When the hosts of mussel species are unknown, the selection of candidates to test as potential hosts can be labor and time intensive. Any species of fish that occurs with a mussel may potentially be a host, although most may not be suitable. However, the identification of narrowly co-occurring distributions based on the copious data available for Big Darby Creek could limit the search to a few likely taxa (see *Ecosystem synthesis* above). Identified hosts of closely related mussels from other drainages have counterparts in the Big Darby drainage, further limiting the scope of the search (Neves, 1983; Yeager, 1986; Weaver et al., 1991). Once implicated, fishes will be tested in the laboratory for their suitability as a host. Because so many species of fishes and mussels found in this system also are found elsewhere, these host identifications would be useful throughout much of the Ohio River drainage.

During their gravid season, current ranges of both federally endangered mussels will be sampled for fishes (see *Fish Movement Patterns* above). A subset of these fishes will be used for artificial infestation of glochidia in the laboratory. The minimum subset will include phylogenetically and ecologically similar fish species to those hosts identified for Cumberlandian mussel analogs. The clubshell analog, *Pleurobema oviforme*, has been shown to parasitize central stoneroller, common shiner, fantail darter, river chub, and whitetail shiner (Neves 1983; Weaver et al. 1991). The northern riffleshell analog, *Epioblasma capsaeformis*, uses primarily benthic species, including banded sculpin, and dusky, redline, and spotted darters (Yeager, 1986). Several of these fishes occur in the Big Darby drainage (Trautman, 1981). Additional fish species occupying similar habitats will be used as warranted. Because of space limitations and the need to replicate the experiments, we will choose only the 12 most likely fish species for each of the two endangered mussel species during each year of the study.

The fish will be artificially infested in the laboratory and maintained in aquaria to determine their suitability as hosts. Fishes may have either a natural or an acquired immunity to infestation (Bauer & Vogel, 1987; Reuling, 1919; Arey, 1924, Bauer, 1987). It is not clear if immunity to one mussel species produces immunity to all others (Reuling, 1919), or if adult hosts are more susceptible than juveniles. However, because glochidial infestations are overdispersed (Weir, 1977; Dartnall & Walkey, 1979; Neves & Widlak, 1988) and immunity is lost if the host is not exposed within a year, the great majority of fishes at any given time and location are uninfested. If possible, individuals from areas devoid of mussels will be used.

Procedures to artificially infest fishes with glochidia are modified from Yeager (1986). Fishes will be checked for natural infestation under anesthesia (MS-222), isolated by species in aquaria, and given a period of acclimatization. Fishes captured that are already infested will not be used. The marsupial region of a gravid mussel will be penetrated by a sterile pipette and the glochidia flushed into a petri dish. A subsample of the glochidia will be exposed to a saline solution to determine if they are mature and living (Zale & Neves, 1982). Anesthetized fishes will be exposed by pipetting several hundred glochidia directly onto the gills and fins. When possible, for each fish/mussel species pair, we will infest three individual fish with glochidia from three individual mussels. Each fish will be maintained in an aquarium in an environmentally controlled room. We will maintain the water temperature at the concurrent average stream temperature. Fish will be fed live food at a daily maintenance ration. Photoperiod will be set to mimic the concurrent natural photoperiod.

Beginning at 10 days post infestation, material from the bottom of the aquaria will be siphoned through a 35 µm mesh sieve. Fishes are considered hosts only if the glochidia live to excyst from the host and metamorphose. If an individual fish has developed an immunity, the glochidia will have sloughed off and died within five days (Fustish & Millemann, 1978; Waller & Mitchell, 1989). We will first concentrate our efforts on the two federally endangered mussels. If we successfully identify hosts in the first year, we will add additional mussel species the second year.

Explaining Patterns in Species Abundances

Ecologists have long been interested in the relationship between number of species and area sampled (or number of individuals sampled) as a general pattern in communities (e.g., Fisher, Corbet, & Williams, 1943; Preston, 1962; May, 1975). Watters (1992) has measured this relationship for mussels and fish in many Ohio drainages, including the Scioto, which contains the Big Darby system. The patterns reported in that paper suggests that the log of mussel species number exactly tracks the log of fish species number over a wide range of drainage sizes (Watters, 1992, figure 1). This will have important implications for the preservation of whole ecosystems. If mussel species diversity is so tightly linked with fish species diversity, then limitation of fishes in a system will have a *predictable* impact on mussel diversity. The pattern suggests that a certain proportional increase in fish species numbers will result in a certain proportional increase in mussel species number. We do not however, understand the mechanism that produces this tight link.

To gain a mechanistic understanding of this link in species abundances, we will simulate the relationships between mussel species and fish species, varying the distribution of degree of host-specificity. For example, at one extreme we have only one mussel species associated with any fish species. Nearing the other end of the continuum, we have great generality in mussel/fish associations. In addition to varying the degree of host specificity, we will vary the degree of dispersion of glochidia among fish individuals. We believe that, in nature, there is a high degree of contagion in infestation; i.e., few fish have glochidia, but those that do are very likely to have high concentrations of them.

Given these rules of association between mussels and fish, we will construct hypothetical mussel/fish communities over a range of drainage sizes. For each constructed community, we will analyze the resulting relationship between mussel species abundance and fish species abundance. We are seeking answers to two questions: 1) what types of mussel/fish associations produce the patterns of species abundances observed (Watters 1992) for this drainage system? and 2) how do patterns of host-specificity observed during the course of the present study of mussel and fish associations fit into the predictions from the simulated communities?

Results

The unprecedented distributional data available for Big Darby Creek's fishes and mussels will allow the first system-wide study of the relationships between these two groups - how diversity is distributed throughout the system, how dependent is mussel diversity on fish diversity, and patterns of parasitism. The results can be applied to less studied systems to gauge the effects of habitat modification, to determine diversity estimates, and to aid conservation methods.

The abundant data on Big Darby Creek allow us to implement a new technology (GIS) in the characterization of habitats for the first time. The interactions between mussels and their hosts, in terms of seasonal migrations of fishes, have rarely been addressed although crucial to mussel reproduction. The temporal data on fishes in the system may give us opportunity to more fully understand these interactions. Overlain with multiple sets of physical and IBI data, we may get the most complete picture to date of the mussels' place in the ecosystem.

The identification of hosts is critical to any effort to conserve and manage a mussel species. The results of the above study would be used to identify potential hosts based on proximity to the mussel, presence during larval release, and ecological similarities. This will greatly simplify the search for hosts for future studies. The results of this portion of the study will be essential in fulfilling Recovery Task 1.41 and 1.42 of the Federal Recovery Plan for the clubshell and northern riffleshell (USFWS, 1993).

In addition to identifying host fish species, it is also important to recognize that these fish can move throughout the stream. Our measurements of timing and location of species-specific patterns of movement will be important in understanding what limits the distributions of mussels. This information will be critical to our future ability to make recommendations about preserving corridors of movement for these fish. The fortuitous timing of dam removals provide us with the opportunity to test these ideas.

And finally, with a community model of species abundances, we can test how different patterns in mussel-fish associations lead to different patterns in relative species abundances. With a mechanistic model such as the one proposed, we will be able to generate expectations of baseline abundances of mussel species in different systems. Deviations from these expectations, then, will help us assess the relative richness of mussel species diversity in less-studied stream systems.

Work Plan

Spring 1994: Set up laboratory for host-specificity experiments; build fish sampling gear; locate specific sample sites; begin passive directional fish trapping.

Summer 1994: Begin retrieval of historical data; continue directional trapping; sampling for gravid mussels, seine/electrofish sampling for fish in areas with gravid mussels; collect fish for host-specificity experiments; collect mussels for experiments; run experiments.

Autumn 1994: Dam samples of fish and mussels; continue retrieval of historical data.

Winter 1995: Analyze data from experiments, traps; maintain, repair, make adjustments to laboratory and field gear; sample dam sites for fish; begin synthesis of historical data.

Spring 1995: Sample dam sites for fish; begin directional trapping at mussel sites; begin collecting fish for host-specificity experiments; continue synthesis of historical data.

Summer 1995: Collect mussels for host-specificity experiments; sample mussel distributions; run lab experiments; seine/electrofish at sites of gravid mussels; continue directional trapping of fish;

Autumn 1995: Dam samples of fish and mussels; begin development of species abundance model.

Winter 1996: Analyze data; submit results for publication; sample dam sites for fish; based on two year's of data, determine which of the field sampling and laboratory experiments need to be continued or altered; continue development and begin analysis of species abundance model.

Spring 1996: Continue trapping and sampling as necessary; prepare for host-specificity experiments.

Summer 1996: Complete model; present results of first two years at scientific meetings; run host-specificity experiments; continue field sampling as necessary.

Autumn 1996: Continue field sampling as necessary; analyze data from final year; write-up model results.

Budget

Budget: March – December 1994			
Item	Total Request	Waived or Provided by OSU or ODNR	Request from TNC
Salaries, Wages			
G.T. Watters, 50%	14,625	14,625	0
E. A. Marschall, 2 months	9,120	4,560	4,560
T. Cavender, 1 month	4,560	0	4,560
Graduate Research Associate (salary, tuition, fees), 3 quarters	13,050		13,050
Graduate Research Associate (salary, tuition, fees), 3 quarters	13,050		13,050
Technical help: hourly (6 months @ $6.67/h)	6,936		6,936
Technical help: hourly (6 months @ $6.67/h)	6,936		6,936
Technical help: full-time permanent	14,250		14,250
Fringe Benefits	10,869	1,186	9,683
Permanent Equipment			
Fish traps (100 x $65/trap)	6,500		6,500
Fish holding tank (living stream $1375, water chiller $625)	2,000		2,000
Travel			
Travel to field sites	4,650		4,650
Travel to meetings	0		0
Materials and Supplies			
Aquaria (75 x $20)	1,500		1,500
Aquarium supplies and lab experiment supplies and maintenance	2,500		2,500
GIS materials	500		500
Publication Costs	0		0
Total Direct Costs	111,046	20,371	90,675
Indirect Costs	18,903	18,903	0
Total Direct and Indirect Costs	129,948	39,274	90,675

Budget: January – December 1995			
Item	Total Request	Waived or Provided by OSU or ODNR	Request from TNC
Salaries, Wages			
G.T. Watters, 50%	22,250	22,250	0
E. A. Marschall, 2 months	9,348	4,674	4,674
T. Cavender, 1 month	4,674	0	4,674
Graduate Research Associate (salary, tuition, fees), 4 quarters	17,835		17,835
Graduate Research Associate (salary, tuition, fees), 4 quarters	17,835		17,835
Technical help: hourly (6 months @ $6.85/h)	7,124		7,124
Technical help: hourly (6 months @ $6.85/h)	7,124		7,124
Technical help: full-time permanent	19,475		19,475
Fringe Benefits	12,414	1,215	11,198
Permanent Equipment	0	0	0
Travel			
Travel to field sites	4,650		4,650
Travel to meetings	1,000		1,000
Materials and Supplies			
Laboratory	1,250		1,250
GIS materials	500		500
Publication Costs	500		500
Total Direct Costs	125,979	28,139	97,839
Indirect Costs	22,302	22,302	0
Total Direct and Indirect Costs	148,280	50,441	97,839

Budget: January - December 1996			
Item	Total Request	Waived or Provided by OSU or ODNR	Request from TNC
Salaries, Wages			
G.T. Watters, 50%	22,750	22,750	0
E. A. Marschall, 2 months	9,582	4,791	4,791
T. Cavender, 1 month	4,791	0	4,791
Graduate Research Associate (salary, tuition, fees), 3 quarters	18,281		18,281
Graduate Research Associate (salary, tuition, fees), 3 quarters	18,281		18,281
Technical help: hourly (6 months @ $7.02/h)	7,301		7,301
Technical help: hourly (6 months @ $7.02/h)	7,301		7,301
Technical help: full-time permanent	19,962		19,962
Fringe Benefits	12,724	1,246	11,478
Permanent Equipment	0	0	0
Travel			
Travel to field sites	4,650		4,650
Travel to meetings	1,000		1,000
Materials and Supplies			
Laboratory	1,250		1,250
GIS materials	500		500
Publication Costs	500		500
Total Direct Costs	128,873	28,787	100,086
Indirect Costs	22,816	22,816	0
Total Direct and Indirect Costs	151,689	51,603	100,086

Budget

Budget: Summary			
Item	Total Request	Waived or Provided by OSU or ODNR	Request from TNC
Salaries, Wages			
G.T. Watters, 50%	59,625	59,625	0
E. A. Marschall	28,050	14,025	14,025
T. Cavender	14,025	0	14,025
Graduate Research Associate (salary, tuition, fees)	49,166		49,166
Graduate Research Associate (salary, tuition, fees)	49,166		49,166
Technical help: hourly	21,361		21,361
Technical help: hourly	21,361		21,361
Technical help: full-time permanent	53,687		53,687
Fringe Benefits	36,006	3,647	32,359
Permanent Equipment	8,500	0	8,500
Travel	15,950		15,950
Materials and Supplies	8,000		8,000
Publication Costs	1,000		1,000
Total Direct Costs	365,897	77,297	288,600
Indirect Costs	64,021	64,021	0
Total Direct and Indirect Costs	429,918	141,317	288,600

Budget Justification

Many of our costs will be for personnel and travel. Because we are attempting to tie together processes (fish movement, larval mussel release, land-use) at multiple locations over a large part of the Big Darby Creek system, we will be using methods that are at times quite labor intensive. However, we believe these costs are justified. Below, we itemize each amount listed on the previous budget pages.

Salaries, wages

In all years, Watters will be dedicating 50% of his time to this project. In the first year, he will be doing the mussel distribution sampling, the mussel collection for host specificity experiments, developing the standardized qualitative mussel sampling technique, working out methods for the host-specificity experiments, and supervising graduate students and technicians. In the last two years, he will continue his sampling and collection work, but also be concentrating on analysis of data collected. In each year, Marschall will be dedicating 2 months of her time to this project. In the first year, she will be developing the fish sampling techniques (including choosing exact sample sites, modifying gear to be appropriate for the Big Darby Creek system, and developing the sample design) and supervising graduate students and technical help. In the last two years, she will concentrate on doing statistical analyses of the data and supervising graduate students and technicians on the project. Both Marschall and Watters will dedicate time in the third year of the project to develop the general species-abundance simulation model and write up results for publication. In each year, Cavender will be giving one month of his time to this project. He will be supervising the retrieval of data from the OSU Museum and the identification of fishes in all fish samples.

Watters' entire salary and benefits are being contributed by the Ohio Department of Natural Resources. We are requesting funds for only one half of Marschall's time on this project.

The time for GIS efforts will be donated by the U. S. Forest Service, Northeastern Forest Experiment Station, in a spirit of cooperation with OSU and TNC. These efforts will be conducted by Dr. Louis Iverson, Research Landscape Ecologist and Mr. Anantha Prashad, GIS specialist. Some minimal funding ($500/yr) is needed to recover costs for GIS supplies.

We are requesting two graduate students be involved in this project. Because we are an academic institution, one of our missions in research is the training of graduate students. We believe this project provides an excellent opportunity to involve graduate students in the design, implementation , and analysis of solid research in conservation biology. This proposal can be divided into two discrete pieces. One graduate student will have responsibility for each of the pieces: host specificity and fish movement. In addition, depending on the interest and level of the graduate student (M.S. or Ph.D.), one will take responsibility for accumulating, organizing, and synthesizing the data from the variety of sources (e.g., from OSU-MBD, OPEA) to go into the ecosystem information base.

We are requesting funding for technical help for the duration of the project. Hourly help during the field season will be essential to complete the intensive field portion of the work (see *Work Schedule).* One full-time permanent technician will, among other things, orchestrate field work during the field season, retrieve historical data, assist in generating and maintaining the ecosystem database, and maintain the animals in the laboratory. We have budgeted for a person with some expertise in fish identification in this position.

On all salaries and wages, we have budgeted for a 2.5% increase per year.

Travel

Travel costs are budgeted at a rate of $0.28/mile for personal cars and $0.40/mile for trucks maintained by the Aquatic Ecology Laboratory at OSU. For truck trips greater than 140 miles, it becomes more economical to use OSU Motor Pool vehicles - in those cases we budgeted for that. Trucks are necessary for fish trapping and sampling trips. Annual budgeted travel miles include: 2 trips to Pennsylvania to collect gravid female mussels,* 4 trips to the big Darby system to collect fish and mussels for host-specificity experiments, 3 trips to sample the dam sites for mussels, 6 trips to sample dam sites for fish, and 20 trips/month for 4 months to sample fish with traps and seines.

In the final two years of the project, we have budgeted for travel money to allow us to travel to meetings to report our results to the scientific and conservation communities.

Equipment

We are requesting $6500 for construction of passive directional traps. We estimate that we can construct these for approximately $65 each. Clearly, this research can not be done without this set of sampling gear. We also request $2000 for a fish holding tank with chiller. The OSU Aquatic Ecology Laboratory will be providing the environmental control room, but we will need additional space to hold the pool of fish as we collect them before the experiment.

Facilities

Host specificity experiments will be conducted at the Aquatic Ecology Laboratory at OSU. This facility (where both Marschall and Watters are located) has a wet laboratory, environmental control rooms, a supply of treated well water, a dry laboratory, and access to an array of computers. We will also base our field work from this facility. Cavender will be located just across the street at the OSU Museum of Biological Diversity.

The GIS work will be done at the US Forest Service Experimental Station in Delaware, Ohio. The GIS facilities include ARC/INFO, GRASS, and ERDAS running on a HP 9000 series workstation. The GIS lab is also equipped with a digitizer for data input and an electrostatic plotter for map production.

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