

Caryn C Vaughn; U of Oklahoma

Biodiversity of Freshwater Mussel Assemblages and their Associated Macroinvertebrate Fauna in Streams of the Ouachita Uplands

Participant Individuals:

Graduate student(s): Daniel E Spooner; Melissa Moore

Research Experience for Undergraduates(s): Chad Kolkmann; Adam Richardson

Undergraduate student(s): Jennifer Johnson; Sarah Maddux; Joy Boggs; William Mahoney

Research Experience for Undergraduates(s): Heather Basara

Participants' Detail

Partner Organizations:

Other collaborators:

We collaborated with Dr. David J. Berg, Miami University of Ohio, on a population genetics study of several mussel species within the rivers we surveyed. We supplied Dr. Berg with mussel tissue and he performed the genetic analyses.

We also supplied tissue of numerous species to the U.S. Department of the Interior Freshwater Mussel Genetics Tissue Repository, Leetown Science Center, Kearneyville, West Virginia.

The following persons performed taxonomic identifications of specimens we collected either under contract or gratis:

Dr. Cheryl Barr : Elmidae

David Baumgardner: Ephemeroptera Dr. Elizabeth Bergey: Lepidoptera

Dr. Arthur Bogan: Mollusks
Dr. Harley Brown: Coleoptera
Heidi Dunn: Oligochaeta
Dr. Donald Klemm: Hirudinea

Dr. Boris Kondratieff: Megaloptera and Plecoptera

Dr. William Matthews: Fishes Dr. Dave Ruiter: Trichoptera

Activities and findings:

Research and Education Activities:

See attached pdf file

Findings:

See attached pdf file

Training and Development:

This project trained three graduate students and six undergraduate students. Graduate students learned how to design and carry out a biodiversity inventory. Two graduate students obtained their MS degrees in Zoology while working on this project, and one (Spooner) did a thesis that was an outgrowth of this project (see Findings section). Students who participated in field work learned how to navigate a river, take numerous kinds of environmental measurements, as well as how to collect benthic organisms. Graduate students became taxonomic experts on the groups that they were reponsible for (macroinvertebrates and mussels). They developed their communication skills in preparing and presenting talks over the project. Undergraduate students learned many valuable skills including good microscope skills, data entry and web page design.

Outreach Activities:

The primary outreach product from this project is a web site that presents species lists and information on the rivers we sampled and selected species:

http://www.biosurvey.ou.edu/Biodiversity Web Site/main.htm

Journal Publications:

Vaughn, Caryn C., "Life history attributes and patterns of extinction in freshwater mussels", Conservation Biology, vol., (), p. . Submitted

Vaughn, Caryn C., Daniel E. Spooner and Bruce W. Hoagland, "River weed growing epizoically on freshwater mussels.", *Southwestern Naturalist*, vol. 47, (2002), p. 604. Published

Vaughn, Caryn C., "The mussel fauna of the Glover River, Oklahoma", *Proceedings of the Oklahoma Academy of Science*, vol., (), p. . Accepted

Vaughn, Caryn C. and Christine C. Hakenkamp, "The functional role of burrowing bivalves in freshwater ecosystems.", *Freshwater Biology*, vol. 46, (2001), p. 1431. Published

Spooner, Daniel E. and Caryn C. Vaughn, "Impact of drought conditions on a mussel bed in the Kiamichi River, southeastern Oklahoma.", *Ellipsaria*, vol. 2000, (2000), p. 10. Published

Vaughn, Caryn C. and Daniel E. Spooner, "Freshwater mussels influence the distribution of co-ocurring benthic invertebrates across multiple spatial scales.", *Ecology*, vol., (), p. . In preparation

Vaughn, Caryn C. and Daniel E. Spooner, "Mussels of the Poteau River: Distribution, abundance and implications for future commercial harvest", *American Midland Naturalist*, vol., (), p. . Submitted Spooner, Daniel E. and Caryn C. Vaughn, "The mussels of the Mountain Fork River", *Southwestern Naturalist*, vol., (), p. . Submitted

Spooner, Daniel E. and Caryn C. Vaughn, "A field experiment examining the effects of freshwater

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mussels on sediment ecosystem function", Ecology, vol., (), p. . Submitted

Vaughn, Caryn C., Keith B. Gido and Daniel E. Spooner, "Ecosystem processes performed by unionid mussels in stream mesocosms: species roles and effects of abundance.", *Hydrobiologia*, vol., (), p. . Submitted

Vaughn, C.C., M. Moore and D.E. Spooner, "Biodiversity of freshwater mussel assemblages and associated invertebrates in streams of the Ouachita Uplands", *Bulletin of the North American Benthological Society (abstract)*, vol. 19, (2002), p. 275. Published

Berg, D.J., C.L. Elderkin, A. Christian, J.L. Metcalfe-Smith, C.C. Vaughn and S.I. Guttman, "Hierarchical analysis of genetic variation in freshwater mussels: comparsion of allozymes and DNA sequences", *Bulletin of the North American Benthological Society*, vol. 19, (2002), p. 182. Published

Spooner, D.E. and C.C. Vaughn, "Small-scale ecological function of the freshwater mussels in the Kiamichi River, Oklahoma.", *Bulletin of the North American Benthological Society*, vol. 19, (2002), p. 175. Published

Vaughn, C.C., D.E. Spooner, M. Moore and K. Gido, "The functional role of unionids in streams: comparative and experimental evidence from Ouachita Mountain Rivers", *Bulletin of the North American Benthological Society*, vol. 18, (2001), p. 187. Published

Berg, D.J., A.D. Christian, J.L. Metcalfe-Smith, C.C. Vaughn and S.I. Guttman, "Hierarchical analyses of genetic variation in freshwater mussels: differentiation at multiple spatial scales", *Bulletin of the North American Benthological Society*, vol. 18, (2001), p. 189. Published

Moore, M., C.C. Vaughn and D.E. Spooner, "Ephemeroptera associated with mussel beds in rivers of the Ouachita Uplands of Oklahoma and Arkansas", *Bulletin of the North American Benthological Society*, vol. 18, (2001), p. 263. Published

Spooner, D.E. and C.C. Vaughn, "Epilithic communities associated with two freshwater mussel species in the Kiamichi River of southeastern Oklahoma", *Bulletin of the North American Benthological Society*, vol. 18, (2001), p. 266. Published

Vaughn, C.C., "Relationship between the population structure of a dominant mussel species and mussel assemblage structure in five Oklahoma rivers", *Bulletin of the North American Benthological Society*, vol. 17, (2000), p. 188. Published

Spooner, D.E., M. Moore and C.C. Vaughn, "The mussel fauna of the Mountain Fork River, Oklahoma and Arkansas", *Bulletin of the North American Benthological Society*, vol. 17, (2000), p. 242. Published Vaughn, C.C. and C.C. Hakenkamp, "The functional role of bivalves in streams", *Bulletin of the North*

Vaughn, Caryn C. and Christopher M. Taylor, "Macroecology of a host-parasite relationship", *Ecography*, vol. 23, (2000), p. 11. Published

American Benthological Society, vol. 16, (1999), p. 174. Published

Vaughn, Caryn C. and Christopher M. Taylor, "Impoundments and the decline of freshwater mussels: a case study of an extinction gradient", *Conservation Biology*, vol. 13, (1999), p. 911. Published

Book(s) of other one-time publications(s):

Matthews, William J., Caryn C. Vaughn, Keith B. Gido and Edie Marsh-Matthews, "Southern Plains Rivers", bibl. Book to be published by Academic Press, (2003). *Book* Accepted of Collection: Benke, A. and Cushing, C., "Rivers of North America"

Vaughn, Caryn C., "Changes in the mussel fauna of the middle Red River drainage: 1910-present.", bibl. Ohio Biological Survey, (2000). *Book* Published

of Collection: Tankerlsey, R.A., Warmolts, D.I., Watters, G.T., Armitage, B.J., Johnson, P.D., and R.S. Butler., "Freshwater Mollusk Symposium Proceedings"

Spooner, Daniel E., "A field experiment examining the effects of freshwater mussels (family Unionidae) on sediment ecosystem function", bibl. University of Oklahoma, (2002). *Thesis* Published

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of Collection: , "MS thesis"

Other Specific Products:

Internet Dissemination:

http://www.biosurvey.ou.edu/Biodiversity Web Site/main.htm

This site presents results of the project to a lay audience.

Contributions:

Contributions within Discipline:

We have expanded our baseline knowledge of the distribution and abundance of three ecologically important groups of organisms, freshwater mussels, benthic macroinvertebrates and invertebrate meiofauna in a unique and understudied geographic region, streams of the Ouachita Uplands. As these areas undergo changes in the future from altered landuse, invasions by exotics (the probable invasion of the zebra mussel) and global climate change, we will have the information necessary for monitoring and to determine if changes in ecological function are occurring. We have discovered a new species of mussel and probable new species of benthic macroinvertebrates and meiofaunal invertebrates. Through both statistical analyses of our field data and experiments we have shown that mussels influence the local distribution of co-occurring benthic invertebrates, indicating that current declines in mussels may also be impact the rest of the benthic community. Finally, extrapolations of our quantitative data from our field survey combined with laboratory data on mussel function indicate that declines in mussel populations may impact stream ecosystem function.

Contributions to Other Disciplines:

The results of this project have many implications for conservation and environmental planning, some of which are already occurring.

The Oklahoma Chapter of The Nature Conservancy has made several rivers in southeastern Oklahoma (the Kiamichi, Glover, Mountain Fork and Little Rivers) its top priority in Oklahoma, based largely on the results of our surveys of these rivers. I am an active member of a team working with the public sector to derive conservation strategies for these rivers. I am participating in a process called Ecologically Sustainable Water Management (ESWM) which seeks to find solutions for maintaining natural flow regimes and meeting the public's need for clean water. Much of the data being used in the ESWM process comes from our project. Related to this process, I have given presentations to the Environment and Natural Resources Committee of the Oklahoma State Legislature (Oct. 2001) and at an ESWM workshop (Feb. 2003).

In August 2000, based on our field observations of drought-related mussel mortality, we were able to convince the Corps of Engineers to

perform an immediate, emergency release of water from Sardis Reservoir into the Kiamichi River. This action probably prevented the deaths of 1000's of mussels.

Freshwater mussels in one of the rivers we surveyed (Poteau River) are threatened by extensive harvest by companies from out of state (primarily Tennessee and Arkansas). At the request of the Oklahoma Department of Wildlife Conservation we provided distribution, abundance and demographic information on populations from this river. I attended two Oklahoma Department of Wildlife Conservation commission meetings over this matter as an invited expert, and gave a presentation at one of these meetings.

The information we collected was used in the critical review of the mussel bioassessment plan for the U.S. Department of Interior, Forest Service, Ouachita National Forest.

Contributions to Education and Human Resources:

Numerous students were trained by this project. See Training and Development under 'Activities'.

Contributions to Resources for Science and Technology:

We have created two important collections and associated databases, one of freshwater mussels and the other of benthic invertebrates, for rivers of the Ouachita Uplands.

Contributions Beyond Science and Engineering:

Our results affect public welfare because our data is already being used in making water resource decisions (See Contributions to other Disciplines in Science and Engineering)

Categories for which nothing is reported:

Participants: Partner organizations Products: Other Specific Product

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Project Participants

Vaughn C Caryn: Principal Investigator Has worked for more than 160 hours: Yes

Contribution to project: Caryn Vaughn was the Principal Investigator on this project. She designed the project and supervised and participated in all aspects of data collection and synthesis.

Spooner E Daniel: Graduate student

Has worked for more than 160 hours: Yes

Contribution to project: Daniel was supported by the grant as a graduate research assistant for 20 hours a week, 12 months a year. Daniel participated in field work and had primary responsibility for identifying and cataloging mussels. Daniel also processed meiofauna samples and entered data. Daniel finished his MS in June, 2002, and is now working on a PhD at the University of Oklahoma.

Moore Melissa: Graduate student

Has worked for more than 160 hours: Yes

Contribution to project: Melissa was employed as a graduate research assistant on this project for 20 hours a week, 12 months per year. Melissa participated in field work and had primary responsibility for initial identifications of benthic macroinvertebrates. Melissa developed a voucher collection and sent specimens to experts for further identification and verification. Melissa developed an excel spreadsheet system for keeping track of the macroinvertebrate data. Melissa completed her MS in July, 2002, and is now teaching community college in Alabama.

Johnson Jennifer: Undergraduate student Has worked for more than 160 hours: Yes

Contribution to project: Jennifer worked as a student technician on the project. Jennifer assisted with field collections, processed macroinvertebrate and meiofaunal samples, and entered data. Jennifer is currently employed as a technician with the Sevilleta LTER and is finishing her MS in Botany at the University of Oklahoma.

Maddux Sarah: Undergraduate student Has worked for more than 160 hours: Yes

Contribution to project: Sarah processed meiofaunal samples in the laboratory and performed data entry.

Boggs Joy: Undergraduate student

Has worked for more than 160 hours: Yes

Contribution to project: Joy processed macroinvertebrate and meiofaunal samples in the

laboratory and performed data entry.

Mahoney William: Undergraduate student Has worked for more than 160 hours: Yes

Contribution to project: William processed benthic macroinvertebrate samples in the laboratory.

Kolkmann Chad: Research Experience for Undergraduates

Has worked for more than 160 hours: Yes

Contribution to project: Chad was employed as a summer REU student. The REU was a supplement to the project to add a meiofaunal component. Chad sampled meiofauna and participated in other field work. Chad continued his research with the meiofauna after his REU ended, examining bacterial populations. He has since left the University.

Years of schooling completed :Junior Home Institution: Same as Research Site

Home Institution if Other:

Home Institution Highest Degree Granted (in fields supported by NSF): Doctoral Degree

Fiscal year(s) REU Participant supported: 2000

REU Funding: REU supplement

Richardson Adam: Research Experience for Undergraduates

Has worked for more than 160 hours: Yes

Contribution to project: Adam was employed as an REU student for two summers (1999 and 2000). Adam's principal responsibility was meiofaunal sampling, but he also assisted with all other field work. Adam graudated with his BS and is now a laboratory technician at the University of South Florida and is applying to graduate school.

Years of schooling completed :Junior

Home Institution: Other than Research Site

Home Institution if Other: Southwestern Oklahoma State University

Home Institution Highest Degree Granted (in fields supported by NSF):Bachelor's Degree

Fiscal year(s) REU Participant supported: 2001 2000

REU Funding: REU supplement

Basara Heather: Research Experience for Undergraduates

Has worked for more than 160 hours: Yes

Contribution to project: Heather developed the project web site.

Years of schooling completed :Junior Home Institution: Same as Research Site

Home Institution if Other:

Home Institution Highest Degree Granted (in fields supported by NSF):Doctoral Degree

Fiscal year(s) REU Participant supported: 2001

REU Funding: REU supplement

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System We welcome comments on this system

The freshwater mussel (Bivalvia: Unionidae) fauna of North American streams is the most diverse in the world, but is highly threatened and declining at an alarming rate. The consequences of this catastrophic decline of an entire family go beyond the loss of species. Mussels serve critical trophic and non-trophic roles in the functioning of river ecosystems. Because mussels can achieve very high densities and are filter feeders, they are important seston consumers and play a significant role in both energetics and particle dynamics in rivers. Because of their dependence on appropriate substrate and flow conditions, mussels are naturally patchily distributed in many rivers, occurring in densely aggregated multi-species "beds". The living mussels and their spent shells in these beds provide habitat for other benthic macroinvertebrates, alter hydraulic and sediment dynamics at the benthic-water interface, and stabilize the substrate. Through their feeding activities and production of pseudofeces, mussels provide nutrients to the remainder of the benthos. Thus, the current decline in mussel species also represents a loss of critical habitat and food resources for the remaining aquatic fauna, and may alter the ecosystem functioning in many North American rivers. In addition, there are likely benthic macroinvertebrates and meiofaunal invertebrates that have co-evolved with mussel assemblages and are specifically dependent on them. No one has ever surveyed the invertebrates specifically associated with mussel assemblages. As mussel populations decline these other invertebrates species are also being lost.

The Ouachita Uplands represent one of the last strongholds of freshwater biodiversity in North America, and may represent one of the last strongholds of freshwater mussel biodiversity in the world. The Ouachitas are unglaciated and have been isolated from other mountain systems for 225 million years. They are a center of speciation for both terrestrial and aquatic organisms, with a high number of endemic species. The rivers have been far less impacted by human disturbance than faunas of more eastern and western U.S. rivers, largely because the area is relatively unpopulated and undeveloped. Thus, the rivers of the Ouachita Uplands may represent one of the few areas in North America where conservation of an intact mussel fauna may be possible.

Biodiversity Survey

The primary goals of this project were to collect specimens, tissues and locality information for a wide variety of taxa to serve as samples and baseline information for a geographic region, development of electronic media to disseminate the information collected, and production of publications and presentations documenting the biodiversity.

Freshwater mussels, and the invertebrate fauna associated with assemblages of mussels, were surveyed in nine rivers of the Ouachita Mountains Physiographic Subprovince in central and western Arkansas and southeastern Oklahoma, U.S. during the summers of 1999 - 2001. The surveyed rivers were the Kiamichi, Little, Glover, Mountain Fork, Poteau, Little Missouri, Caddo, Ouachita and Saline. For NSF budgetary the start date for this project was August, 1998, but for logistical reasons we were unable to begin field work until June, 1999.

The Kiamichi River, a major tributary of the Red River, flows through the Ouachita Mountains and Gulf Coastal Plain of southeastern Oklahoma. The river is 272 km long and has a drainage area of 4,800 km². The Kiamichi is known for its clean water and high aquatic biodiversity, including more than 100 fish species.

The Mountain Fork River, a tributary of the Little River, has a drainage area of 2240 km². The lower Mountain Fork River is impounded by Broken Bow Reservoir which is used for hydroelectric power among other uses.

The Glover River is the only remaining unimpounded tributary of the Little River. This highly pristine river is 48 km long, has a drainage area of 876 km², and is known for its scenic value and fish biodiversity. The river contains the largest known population of the federally threatened leopard darter, *Percina pantherina*. The upper two thirds of the river are high gradient (average slope 19 m/km) and characterized by low bedrock and boulder falls, chutes and riffles. Below the fall line, the gradient decreases to 1 m/km as the river flows through the Gulf Coastal Plain before entering the Little River. Here stream habitat is characterized by deep, long pools separated by shallow riffles. Riparian areas are mixed pine and oak forest historically harvested for timber by private companies. Since 1996, much of the watershed has come under the management of the U.S. Forest Service and timber harvest has decreased

The Little River, a major tributary of the Red River, drains 5,700 km² in southeastern Oklahoma and southwestern Arkansas. The river is 349 km in length. The upper reaches of the river flow through the Ouachita Mountains and the upper watershed is heavily forested and used primarily for silviculture. Lower reaches of the river flow through the fertile bottomlands of the Gulf Coastal Plain. The Little River headwaters are boulder-cobble and gravel with well-developed pools and swift rapids until the mainstem forms Pine Creek Reservoir. Below the reservoir the river retains pool-riffle structure over a mostly gravel bed, with water willow a common structural feature in the shallows. The Little River is influenced by two impoundments. The mainstem Little River is impounded by Pine Creek Reservoir, used for flood control, water supply and recreation. Approximately 30 miles below Pine Creek Reservoir, a sixteen mile section of the river flows through the Little River National Wildlife Refuge (15,000 acres). Sixty-four km downstream of Pine Creek dam, the Mountain Fork River enters the Little River.

The Poteau River arises in the Ouachita Mountains of western Arkansas, flows westward into Oklahoma, then flows north to join the Arkansas River at Fort Smith. The Poteau has a drainage area of 4895 km²

The Ouachita River originates in the Ouachita Mountains northwest of Mena, Arkansas, and meanders eastward through the heart of the Ouachita National Forest to just west of Hot Springs where it forms three impoundments. The river then flows southward across the Gulf Coastal Plain into Louisiana where it enters the Red River. The Ouachita River is the largest river in the Ouachita Highlands with a drainage area of 67,337 km² and a total length of 872 km. Major tributaries originating in the Ouachita Highlands include the Caddo, Little Missouri and Saline rivers.

The Little Missouri River is a major tributary of the Ouachita River. The headwater area is known for its scenic pine-forested ridges and valleys. The river cascades through a series of white-water gorges, including Little Missouri Falls, before leveling off as it flows across the Arkansas Piedmont. The river has a drainage area of 5447 km².

The Saline River originates in the eastern Ouachita Mountains in central Arkansas and flows southward 328 km to converge with the Ouachita River. The Saline River has a drainage area of 8417 km². The upper, mountainous reaches of the Saline consist of a series of cobble riffles and pools. In lower reaches where the river flows through Gulf Coastal Plain the channel consists of sand and mud.

The Caddo River is a major tributary of the Saline River. The Caddo arises in the Ouachita Mountains of western Arkansas. The Caddo River has a drainage area of $1235~\rm km^2$.

Streams were traversed by canoe and reconnaissance snorkel searches were conducted to locate mussel beds. Reconnaissance snorkel searches were conducted in areas where dead shells were observed and in areas where the habitat looked appropriated for mussels. Location of each site was recorded with GPS. SCUBA was used where water depths required it. Mussels were quantitatively sampled with timed searches and quadrats, and demographic data were taken.

Timed searches were performed at 81 sites (stream reaches). At these sites we recorded mussel species, abundance, and the size distribution of mussel species. Timed searches were conducted visually and by hand searching (grubbing). The stream reach was searched by at least two people for periods ranging from 15 minutes to an hour, depending on reach size. All mussels encountered were removed to shore, identified, and their shell length was recorded. After all sampling was completed, mussels were returned alive to the streambed as close to where they were captured as possible. Timed searches were performed subsequent to quadrat sampling.

For each stream reach we also qualitatively sampled benthic and stream margin invertebrates with dipnets and by hand picking substrate and vegetation. These organisms were preserved in 70% ethanol. At the stream reach scale we recorded a suite of environmental variables. We recorded flow and depth at 1 m intervals for several transects across each reach, allowing us to calculate discharge. We measured dissolved oxygen, conductivity and pH with electronic meters. We took two water samples, stored them on ice, froze them in the laboratory, and later sent them to a professional water quality laboratory for analysis of 20 elements, including calcium, a limiting factor for mussels. We recorded bank and streambed slope and gradient and riparian characteristics.

For 30 stream reaches, we also performed extensive quantitative sampling in the following manner. Fifteen 0.25 m² quadrats were randomly laid out in the stream reach using a stratified sampling protocol to encompass all microhabitats and areas with and without aggregations of mussels. Within each quadrat we sampled mussels, and within 10 quadrats we sampled benthic macroinvertebrates, meiofauna and local environmental parameters in addition to mussels. Sampling within each quadrat occurred in the following order: environmental parameters, benthic macroinvertebrates, meiofauna, and mussels.

Mussels were sampled visually and by sieving the substrate to a depth of 30 cm with the fingers. All mussels were measured (shell height) and returned alive to the streambed following sampling.

Benthic macroinvertebrates were quantitatively sampled within each quadrat with a Hess-type sampler modified to vacuum the streambed, allowing comparable sampling in areas with substantial vs. no flow. The substrate was vacuumed for a period of five minutes. Samples were stored in 70% ethanol.

Two meiofauna cores were taken in each quadrat. Samples were placed in ziplock bags with half of an alka seltzer tablet to relax the organisms. Samples were then preserved in 10% buffered formalin stained with rose bengal.

The following environmental variables were recorded from each quadrat: % green algae, diatoms, and detritus; % shade; substrate stability (as resistence to a handheld penetrometer); % bedrock, boulder, cobble, gravel, sand and silt.

Adult aquatic insects were sampled with light traps. Invertebrate meiofauna were sampled by taking sediment cores. Environmental data were measured at each site.

Vouchers and databases

Limited mussel vouchers were taken for all mussel species except state and federally **protected** species. We removed a small clipping of mantle tissue (approximately 1 cm²) from each voucher specimen and stored it in 95% ethanol so that it will be available for future DNA analysis. Remaining tissue was preserved in 70% ethanol. Shells were cleaned and stored dry. All voucher specimens have been cataloged into the mollusk collection of the Oklahoma Biological Survey. A searchable Access database was created to store the mussel catalog. Species lists of mussels for each river that we surveyed have been posted on the web (http://www.biosurvey.ou.edu/Biodiversity Web Site/Mussels.htm).

Mantle tissue clippings of several common species were collected from multiple sites, stored in the field in liquid nitrogen, and shipped to Dr. David J. Berg, Miami University of Ohio. Dr. Berg is using this tissue in a study of population genetics of North American mussels. Whole individuals of some species were shipped alive to the Freshwater Mussel Tissue Repository run by the USGS at the Leetown Science Center in West Virginia.

Following preliminary identification in our laboratory, benthic and meiofaunal invertebrates were sent to experts for verification and/or further identification. We allowed experts to keep some specimens for their taxonomic studies, however we retained several voucher specimens of each benthic macroinvertebrate and meiofaunal species. These are cataloged in an Excel database and stored in the collections of the Oklahoma Biological Survey. We intend to convert the Excel database to a more user-friendly Access database. Many invertebrate specimens are still being identified by experts and we do not yet know how many species we collected. Species lists of benthic macroinvertebrates identified to date have been posted on the web (http://www.biosurvey.ou.edu/Biodiversity_Web_Site/benthictaxa.htm). Meiofaunal samples are still being processed and identified.

Ecological experiments and analyses

A secondary goal of this project was to use the collected biodiversity information to address conceptual issues in conservation biology and ecology. We wanted to determine if there is

streambed invertebrate fauna associated with and perhaps dependent on the mussel fauna. If there is, then the current decline in freshwater mussels could also mean a decline in co-occurring invertebrates. We also wanted to examine some of the functions that freshwater mussels perform in ecosystems, as these functions may change as mussel assemblages change and decline over time. Once we have measured these functions, we can use the quantitative data that we collected on mussel assemblages to predict ecological function for actual stream reaches. To address these questions we did the following. First, we examined the relationship between the distribution and abundance of mussels and co-occurring benthic invertebrates using various statistical techniques. We did this using the quantitative data that we collected for 30 sites. Second, we performed some laboratory experiments examining the function of two common mussel species across a gradient of mussel biomass. Third, we performed a field experiment examining colonization of benthic macroinvertebrates in treatments with two common mussel species, vs. shells alone, vs. streambed sediment alone.

Dissemination of Collected Information

We developed a website describing the Ouachita Uplands and presenting the results of the project (http://www.biosurvey.ou.edu/Biodiversity_Web_Site/main.htm). This web site is updated frequently as we get new information. We published journal articles and abstracts. These are listed in the Publications section of this report. Unpublished presentations are listed below:

Presentations (presentations with published abstracts were listed under Journals)

Vaughn, C.C., D.E. Spooner, M. Moore and K.B. Gido. Comparative and experimental evidence for the functional role of freshwater mussels in streams. Annual Meeting of the National Shellfisheries Association, April 14 - 18, 2002, Mystic, CT. (*Invited talk in special session*).

Elderkin, C.L., D.J. Berg, J.L. Metcalfe-Smith, C.C. Vaughn, A.D. Christian, and S.I. Guttman. Hierarchical analysis of mtDNA variation in *Amblema plicata*, a widespread mussel species. Annual Meeting of the National Shellfisheries Association, April 14 - 18, 2002, Mystic, CT. (*Invited talk in special session*).

Vaughn, C.C. and D.E. Spooner. The functional role of mussels in streams: species roles and environmental context. 1st Oklahoma-Texas Aquatics Research Group Meeting and 29th Annual Great Plains Limnology Conference, October 24 - 27, 2002, University of Oklahoma Biological Station. (*Contributed*)

Matthews, W.J., C.C. Vaughn, K.B. Gido and E. Marsh-Matthews. Rivers of the southern plains. Plenary lecture. 1st Oklahoma-Texas Aquatics Research Group Meeting and 29th Annual Great Plains Limnology Conference, October 24 - 27, 2002, University of Oklahoma Biological Station. (*Invited*)

Spooner, D.E. and C.C. Vaughn. Small-scale ecological function of freshwater mussels in the Kiamichi River, Oklahoma. Annual Meeting of the National Shellfisheries Association, April 14 - 18, 2002, Mystic, CT. (Invited talk in special session).

- Spooner, D.E. and C.C. Vaughn. Small-scale ecological function of the freshwater mussels in the Kiamichi River, Oklahoma. Annual meeting of the North American Benthological Society, May 26 June 1, 2002, Pittsburgh, PA. (Contributed)
- Vaughn, C.C., K.B. Gido and D.E. Spooner. The functional role of mussels in streams: effects of biomass and species. Freshwater Mollusk Conservation Society, 2nd Symposium, March 12 14, 2001, Pittsburgh, PA. (Contributed)
- Vaughn, C.C. and D.E. Spooner. Mussels of the Poteau River: Distribution, abundance and implications for future commercial harvest. Oklahoma Academy of Science, Lawton, OK, November 2, 2001. (Contributed)
- Spooner, D.E. and C.C. Vaughn. Epilithic communities associated with two different species of freshwater mussels in the Kiamichi River of southeastern Oklahoma. Freshwater Mollusk Conservation Society, 2nd Symposium, March 12 14, 2001, Pittsburgh, PA. *(Contributed)*
- Spooner, D.E. and C.C. Vaughn. Ecosystem effects of native freshwater mussels (family Unionidae). Oklahoma Academy of Science, Lawton, OK, November 2, 2001 (Contributed)
- Spooner, D.E. and C.C. Vaughn. Impact of drought conditions on a mussel bed in the Kiamichi River, southeastern Oklahoma. 48th annual meeting of the Southwestern Association of Naturalists, Hayes, Kansas, April 19 -21, 2001. (*Contributed*)
- Vaughn, C.C., K.B. Gido and D.E. Spooner. The functional role of biodiversity: are bivalves redundant? Annual Meeting of the Southwestern Association of Naturalists, Denton, Texas, April, 2000. (Contributed)
- Vaughn, C.C., D.E. Spooner, M. Moore and J.E. Johnson. Biodiversity of freshwater mussel assemblages and associated invertebrate fauna in the streams of the Ouachita Uplands, US. National Science Foundation Workshop on Biotic Surveys and Inventories, Orcas Island, Washington, May 18-21, 2000. (*Invited*)
- Spooner, D.E. and C.C. Vaughn. Impact of drought conditions on a mussel bed in the Kiamichi River. Great Plains Limnology Meeting, Lawrence, Kansas, November 4, 2000. (Contributed)
- Moore, M., C.C. Vaughn, D.E. Spooner, and J.E. Johnson. Biodiversity of Ouachita Upland rivers. Annual meeting of the Southwestern Association of Naturalists, Denton, TX, April, 2000. (Contributed)
- Vaughn, C.C. "Biodiversity surveys in rivers", National Biological Information Infrastructure conference for NSF P.I.s sponsored by the USGS, March 15 16, Denver, CO. (Invited)
- Vaughn, C.C. Changes in the mussel fauna of the Red River drainage: 1910 present", First Symposium of the Freshwater Mollusk Conservation Society, March 17 19, 1999, Chattanooga, TN. (Contributed)

Vaughn, C.C. Effects of reproductive strategy on extinction rates of freshwater mussels, Annual meeting of the Southwestern Association of Naturalists, April, 1999, Monterrey, Mexico. (Contributed)

Vaughn, C.C. "Macroecology of a host-parasite relationship: mussels, fishes and extinction". February 1999. Institute of Ecosystem Studies, Millbrook, NY. (Invited)

Consistent with the goals of the Biotic Surveys and Inventories Program, the primary goals of this project were to collect specimens, tissue and locality information for a wide variety of taxa to serve as samples and baseline information for a geographic region, development of electronic media to disseminate the collected information, and production of publications and presentations documenting the biodiversity information. Secondary goals included using the collected biodiversity information to address conceptual issues in biology and ecology. Both primary and secondary goals have been achieved.

Biodiversity Information

We documented the occurrence of 54 species of mussels in the nine surveyed rivers. These records include the discovery of many new mussel beds for most rivers, range expansions for multiple species, and the discovery of a new mussel species. The majority of these rivers had been largely unexplored for mussel except at areas near bridge crossings, which are notoriously poor habitat for mussels because of human disturbance. Because we canoed rivers we were able to find and map previously undiscovered mussel beds. Because we recorded not only the presence of species, but also collected density, demographic information, and mapped bed locations with GPS, our data will serve as important baseline information that can be used to monitor the success of these mussel assemblages in the future.

Mussel range expansions included our discovery of *Quadrula nobilis* (Gulf mapleleaf) in the Poteau River, *Quadrula cylindrica* (Little spectaclecase) in the Glover river, and *Arkansia wheeleri* (Ouachita rock pocketbook, a federally endangered species) in the Little River. We also rediscovered *Leptodea leptodon* (Scaleshell), a federally endangered species, in the Kiamichi River. This species was thought to be extirpated.

In the Ouachita River we discovered a mussel species that has similarities with both *Alasmidonta marginata* and *Lasmigona costata*. This species was actually fairly abundant at several sites. We provided whole specimens and tissue to Drs. Arthur Bogan and John Harris who are determining if this is a new species.

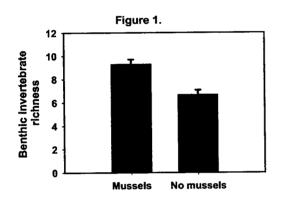
Mussel species list for the entire project and by river are provided on the web (http://www.biosurvey.ou.edu/Biodiversity_Web_Site/Mussels.htm).

From our benthic invertebrate samples we have identified 64 species of mayflies, 40 species of caddisflies, 16 species of gastropods, 14 species of odonates, 20 species of beetles, 8 species of lecches, four families of mites, 14 species on non-chironomid diptera, and 15 genera of chironomids. Many benthic invertebrate specimens are still being identified by experts, and we expect these numbers to increase substantially. We also anticipate the description of new species. Benthic invertebrate species lists to date are posted on the web (http://www.biosurvey.ou.edu/Biodiversity Web Site/benthictaxa.htm).

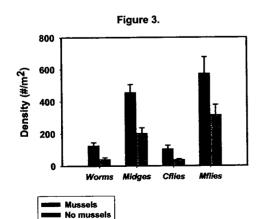
The original scope of work for this project involved sampling only mussels and benthic macroinvertebrates. In the summers of 1999 and 2000, REU funding was used to hire undergraduate students to collect meiofauna samples. An additional supplement was obtained in 2001 for laboratory processing of meiofaunal samples. However, the meiofaunal component of this project has turned out to be more labor-intensive than we had anticipated. We collected 300

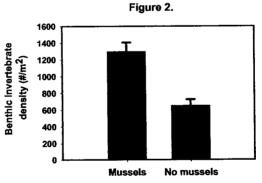
meiofauna cores from eight rivers from 1999 - 2001. We are still processing and identifying these samples (using non-NSF funding since the project has expired). The samples that have been processed so far contain a diversity of microinvertebrates including oligochaetes, nematodes, chironomids, mites, juvenile bivalves, and micro crustaceans such as ostracods and copepods. Because meiofauna has never before been examined in any of the rivers we sampled, it is highly likely that we will discover new species, particularly of oligochaetes and micro crustaceans. Thus, it is important that we process all of the samples we collected. In addition, recent articles in journals such as *BioScience* indicate that meiofauna are critical to stream ecosystem function.

Statistical Examination of the relationship between mussels and co-occurring benthic macroinvertebrates.



Both densities and richness of benthic macroinvertebrates were higher in quadrats that contained mussels (Figure 1, Figure 2) and this relationship held across different functional groups of benthic invertebrates (Figure 3).





We used canonical ordination to partition the variation in benthic macroinvertebrate assemblages explained by environmental factors, mussel distribution and abundance, and shared variation between mussel assemblages and environmental factors. We did this for a local scale using quadrat data and at a regional scale using stream-reach data. At the local scale, we explained 30% of the variation in invertebrate assemblages, and mussels accounted for approximately 70% of the explained variation (Figure 4). At a regional scale we explained 93% of the variation in mussel assemblages, but mussels accounted for less variation. At the regional scale effects of mussels on co-occurring benthos appear to be dampened by regional gradients in environmental factors and the influence of dispersal and biogeographic history (Figure 5). We are still working on these analyses and plan to analyze these data on a biomass basis and also incorporate meiofauna once we have finished processing those samples. Nevertheless, these analyses indicate that at a local scale mussels have a strong influence on the distribution and abundance of co-occurring benthic macroinvertebrates.

Figure 4.

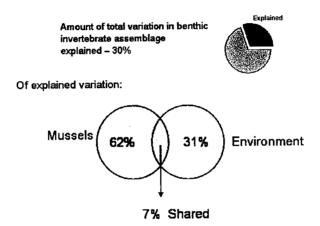
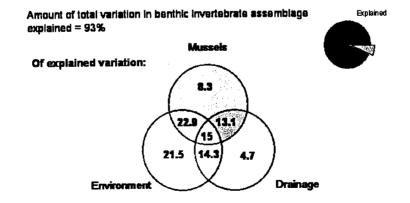


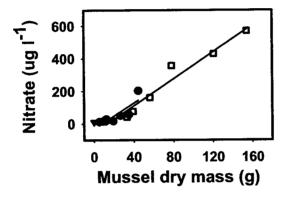
Figure 5.



Laboratory Experiments

Ecosystem process rates were compared for two species, *Actinonaias ligamentina* and *Amblema plicata*, under two sets of background environmental conditions (high and low algal productivity, both at low flow), and across a range of densities (0, 3, 7, 10, 14, 20, 27, and 34 mussels m⁻²). Each experimental stream contained only a single species of mussel, and each species × density treatment was replicated twice. Densities were converted to biomass for analyses. Data were analyzed with analysis of covariance to test for the effects of biomass (the covariate) and species on ecosystem properties.

FIGURE 6. Water column nitrate for day 4 at low productivity. *Actinonaias* = squares, *Amblema* = circles.



For these two species, mussels had strong effects on measured ecosystem processes, effects were linearly related to biomass regardless of species, and mussels influenced these processes even at low biomass (relative to controls).

Figure 6 and Table 1 are examples of typical results. In addition, effects were observed in both low and high productivity treatments, suggesting that mussels influence ecosystem processes across a range of stream trophic states. Analyses corrected for biomass, so any significant species effects are due to differences between species (i.e., the slope of the relationship between biomass and the response variable), and not just differences in average size. Strong biomass effects, and no species effects, were observed for most variables, in both low and high productivity experiments, and across time periods in this experiment (Table 1). This experiment demonstrated that effects of mussel biomass are linear (at least in single-species treatments) and that the processes performed by mussels are similar under low and high productivity conditions (although the magnitude differs).

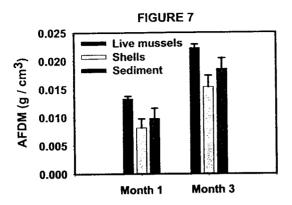
Table 1. F-statistics and associated P-values (in parentheses) derived from ANCOVAs that tested for the effects of body mass (covariate) and species on the various ecosystem properties of experimental streams. Results presented here are for only two of the response variables and three of the time periods, because of proposal space limitations, but similar results were obtained for the other response variables, and across all time periods.

Response	Productivity	Day 1	Day 2	Day 4
Respiration	-	•		
Biomass	high	0.20 (0.664)	66.09 (< 0.001)	187.68 (< 0.001)
	low	23.42 (0.001)	8.40 (0.014)	13.14 (0.004)
Species	high	5.35 (0.041)	0.07 (0.793)	0.140 (0.715)
	low	0.05 (0.826)	0.88 (0.369)	1.11 (0.315)
NH_3 - N		•		
Biomass	high	25.14 (< 0.001)	249.07 (< 0.001)	87.40 (< 0.001)
	low	40.49 (< 0.001)	29.32 (< 0.001)	28.16 (< 0.001)
Species	high	0.353 (0.565)	4.66 (0.054)	0.861 (0.373)
	low	1.568 (0.236)	0.06 (0.817)	0.05 (0.829)

Biodeposition rates were measured for Amblema plicata and Actinonaias ligamentina in 1 liter beakers at three algal food concentrations and 21° C, replicated 8 times. The amount of organic material deposited on the sediment was significantly higher in the mussel treatments than the control (F = 38.65, P < 0.001; mean AFDM for A. plicata treatments = 2.15 mg, A. ligamentina treatments = 6.81 mg, and control = 0.35 mg). Biomass-corrected biodeposition significantly increased with increasing food concentration (F = 50.01, P < 0.001), but was only marginally different between species (F = 3.38, P = 0.087), probably as a result of small sample size.

Field Experiment

This experiment was the M.S. thesis project of Daniel Spooner, a Graduate Research Assistant on the project. The experiment, performed in 0.25 m² enclosures in the Kiamichi River, examined the effects of live mussels, shells, and sediment on organic matter accumulation, and algal and invertebrate colonization. Treatments were live mussels (Amblema plicata and Actinonaias ligamentina), shells (weighted with sand and glued together), and mussel-free sediment. Each treatment was replicated 15 times, with 5 replicates of each treatment pulled after one, three and twelve months. Prior to the experiment, Kiamichi River sediment was homogenized and divided among the treatments. Enclosures were buried 15 cm deep in the riverbed, with an additional 15 cm extending above the surface. The experiment ran from August 2000 - August 2001, and we were able to recover all of our replicates for each time period (despite several large winter floods), demonstrating that long-term experiments can be performed with these enclosures and at this site. Response variables measured for the three time periods were AFDM and invertebrate composition (richness and density) in the sediment of the enclosures, chlorophyll and AFDM on glass slides placed in the enclosures, and AFDM, chlorophyll and algal and invertebrate abundance and composition on shells. We found significant differences between live mussels and dead shells or sediment, with live mussel treatments having more organic matter in surrounding sediment (month one: t = 2.14, P < 0.05; month three: t = 1.93, P < 0.05; Figure 7) and higher sediment chlorophyll concentrations (month one: t = 2.09, P < 0.05; month three: t = 2.81, P <



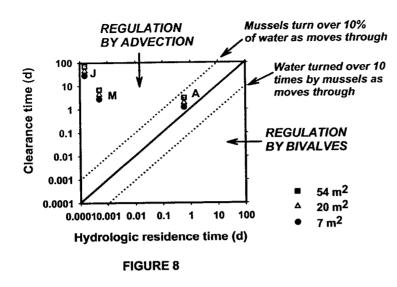
0.05). Results also indicated that effects of mussels vary with environmental context. For example, chlorophyll a on shells was significantly higher on live compared to dead shells for month one (t = 2.11, P < 0.05), but not for month three. During the first month of the experiment flow and water volume were low, but they increased in month three. Lower flows should allow nutrients excreted by mussels to be taken up by algae and would also allow more algae to remain attached to shells,

while higher flows would move both nutrients and algae downstream.

Scaling up and River Comparisons

For each of our stream reaches that were quantitatively sampled we know the areal extent of the mussel bed, mussel species composition, density, size and biomass, and composition and density of benthic invertebrates and meiofauna. In addition, we know the distribution of the mussel beds throughout these streams, and can thus estimate the overall mussel biomass for each stream. We measured the bank slope, thalweg, and summer discharge, and can combine this with hydrologic and temperature information available on the internet from the USGS to estimate year round stream conditions. We plan to combine these data with the quantitative information on mussel functional roles obtained in a new research project we have just begun (NSF DEB-0211010) to make predictions about overall mussel function in streams that vary in overall mussel biomass, species composition, size, and hydrologic and temperature regimes. An example of one such extrapolation from the data collected in the laboratory experiment described above is shown below.

We used filtering rates (estimated from water column chlorophyll concentrations) measured in our laboratory experiment to estimate clearance time in days for an existing mussel bed in the Kiamichi River under three hydrologic regimes and three mussel densities. The natural mussel bed in the Kiamichi River modeled was 1300 m² with an average mussel density of 21 individuals m² (based on sampling from this project). Hydrologic residence times in days were estimated from field measurements of depth and discharge collected from this site in August (low flow), May, and January (peak flow). In August, water volume in the pool containing the mussel bed is reduced and flows are very low, thus it takes almost a day for water to move across the mussel bed (Figure 8). Under these conditions, mussels can turnover a substantial proportion of the water column at even relatively low densities. Consequently, at low flows mussels should be affecting most ecosystem processes in the vicinity of the mussel bed, and the magnitude of these effects will increase with increased biomass. At higher flows and water volumes during May and January, only a small fraction of the water column is filtered by mussels before it flows across the bed (Figure 8). Under these conditions, small-scale effects of mussel activity are likely



overridden by advective forces. This extrapolation is for a mussel bed contained in one stream reach: the degree to which mussels may impact ecosystem processes in an entire stream will depend on the number and spatial distribution of mussel beds, the biomass of mussels in those beds, and overall stream hydrologic state. However, this extrapolation demonstrates that effects of mussels on stream ecosystems should be context dependent and

will vary with system size, system stability, and the distribution and abundance of mussels.

Findings by collaborators using our specimens

Genetic structure of freshwater mussel populations is highly variable. Headwater species show low within-population variation and high among-population variation, while some large river species exhibit the opposite pattern. Dr. David J. Berg, Miami University of Ohio, performed a hierarchical analysis of genetic variation to test the hypothesis that species with different genetic structures will exhibit different biogeographic patterns. He examined the partitioning of allozyme variation at spatial scales ranging from local (within stream) to interregional (Great Lakes vs. Ohio basin vs. Ouachita Uplands) for three common species of mussels. Mussel tissue from the Ouachita Uplands was provided from our collections. Variation within populations was highest in Quadrula quadrula, intermediate in Amblema plicata, and lowest in Elliptio dilatata. The order of species was reversed for variation within rivers, except that Q. quadrula in Great Lakes tributaries showed the highest levels of diversity. Variation was higher in the Ouachita Uplands than in the Ohio basin for A. plicata, but regional variation was no higher than variation among rivers within a region for all three species. His results suggest that the greatest proportion of variation is contained among basins for large river species, while for species from smaller rivers and headwater streams, both within river and among basin variation is significant. Such differences might be explained by demographic or historical processes.

Knowledge of genetic structure of target species is essential for the development of effective conservation plans. Amblema plicata and Eliptio dilatata are common, widespread freshwater mussel species. However, A. plicata are habitat generalists, whereas E. dilatata are generally restricted to headwater habitats. Drs. Curt Elderkin and David J. Berg, Miami University of Ohio, sequenced mtDNA from ~10 individuals in two populations, from two rivers in each of three distinct drainages: Northern Lake Erie, Ohio River, and Ouachita River (for a total of twelve

populations). They used this hierarchical design to estimate percent variation at four levels: within populations, among populations within river, among rivers within drainage, and among drainages. They then sequenced a 652-base portion of the mitochondrial cytochrome oxidase I (COI) gene. Multiple haplotypes were identified for both species, and unique sequences were found in southern populations, indicating that these populations are much older. Also, for *E. dilatata* they found 4X the variation among rivers and among drainages than for *A. plicata*. These results suggest that genetic drift has had a more profound effect on *E. dilatata* than on *A. plicata* populations. Overall, management decisions for habitat specialists may involve a much larger geographic scale than for generalists, in order to preserve the genetic variation present in these species.