MANAGEMENT (BIOLOGICAL, CHEMICAL, PHYSICAL) OF SNAILS FOR GRUB CONTROL

Chairperson:	Gregory W. Whitledge, Southern Illinois University-Carbondale
Industry Advisory Council Liaison:	Rex Ostrum, McCook, Nebraska
Extension Liaison:	Joseph Morris, Iowa State University
Funding Request:	\$225,000
Duration:	2 Years (September 1, 2007 - August 31, 2009)

Objectives:

- Investigate one or more methods of potentially useful approaches to snail population management and/or grub control. The methods of greatest interest include those that will be effective, economical, and approvable by state and federal regulators at commercial production scale. These methods will include reviewing what has been done elsewhere and designing studies that will address the NCRAC conditions, especially in pond systems for the production of economically important food fish for the region. Attempts will be made to investigate and refine these methods.
- 2. Assemble an updatable snail management guide which includes a literature review of known control options, a method of determining snail infestation levels in any water system, and a set of standard operating procedures to reduce snail populations and trematode infestations based on the research cited in Objective 1.

Proposed Budgets:

Institution	Principal Investigator(s)	Objec- tive	Year 1	Year 2	Total
Southern Illinois University- Carbondale	Gregory W. Whitledge	1	\$59,680	\$61,660	\$121,340
University of Wisconsin- Stevens Point	Christopher F. Hartleb & Todd Huspeni	1	\$42,780	\$40,380	\$83,160
Iowa State University	Joseph E. Morris & Richard D. Clayton	2	\$11,678	\$8,822	\$20,500
		Totals	\$114,138	\$110,862	\$225,000

Non-funded Collaborators:

Facility	Collaborator
Blue Iris Fish Farm	Bill West
Wisconsin Department of Agriculture, Trade and Consumer Protection	Myron Kebus

TABLE OF CONTENTS

SUMMARY OVERVIEW (PARTICIPANTS, OBJECTIVES, AND PROPOSED BUDGETS)1
JUSTIFICATION
RELATED CURRENT AND PREVIOUS WORK4
ANTICIPATED BENEFITS
OBJECTIVES
PROCEDURES
FACILITIES
REFERENCES13
PROJECT LEADERS
PARTICIPATING INSTITUTIONS AND PRINCIPAL INVESTIGATORS
BUDGETS
BUDGET AND BUDGET EXPLANATION FOR EACH PARTICIPATING INSTITUTION 19 Southern Illinois University (Whitledge - Objective 1) 19 University of Wisconsin-Stevens Point (Hartleb and Huspeni - Objective 1) 22 Iowa State University (Morris and Clayton - Objective 2) 25
BUDGET SUMMARY FOR EACH YEAR FOR ALL PARTICIPATING INSTITUTIONS
SCHEDULE FOR COMPLETION OF OBJECTIVES
LIST OF PRINCIPAL INVESTIGATORS
CURRICULUM VITAE FOR PRINCIPAL INVESTIGATORS

JUSTIFICATION

Seafood consumption by the populace in the North Central Region (NCR) is estimated at approximately 453,592,370 kg/yr (1 million lb/yr). However, the aquaculture industry in this region supplies less than 2% of the amount consumed annually. To meet the demand for seafood, aquaculture production in the NCR of the United States has been gradually increasing over the past decade. The NCR produces a variety of aquatic species including yellow perch (*Perca flavescens*), hybrid striped bass (*Morone chrysops × M. saxatilis*), walleye (*Sander vitreus*), rainbow trout (*Oncorhynchus mykiss*), bluegill (*Lepomis macropterus*), largemouth bass (*Micropterus salmoides*), several baitfish species and freshwater shrimp (*Macrobrachium rosenbergii*). Many of these species are cultured in ponds. In the first census of aquaculture, the NCR region had 362 farms total, on which 81% of the producers use earthen ponds as the primary system for culture (USDA-NASS 2000).

Aquaculture producers have experienced significant monetary losses due to the infestation of digenetic trematodes, often referred to as grubs, in many commercially important food fish species, especially in yellow perch, bluegill, largemouth bass, hybrid striped bass, channel catfish, and others (Griffin et al. 2002; Overstreet and Curran 2004). These trematode species (*Posthodiplostonum minimum*, white grub; *Uvulifer ambloplitis*, black grub; and *Clinostomum complanatum*, yellow grub) naturally occur in many types of water systems, but are most troublesome in ponds utilized for commercial fish production. To decrease the number of infections, understanding the life cycle of these trematodes is important. The general life cycle of all three grub species includes a bird host (e.g., herons, kingfishers, and cormorants), and a snail intermediate host (*Physa* spp., white grub and *Helisoma* (= *Planorbella*) spp., black and yellow grub) (Figure 1). The parasites mature and sexually reproduce in the bird, releasing eggs that are voided in the bird's feces. In water, eggs embryonate and release a free-swimming miracidium that seeks out and penetrates the snail host. Once inside an appropriate snail host, the grub asexually reproduces, ultimately releasing swimming cercariae. Cercariae seek out and penetrate host fishes, developing into the characteristic "grubs" in the fish flesh. These grubs can sometimes cause significant losses in small

fish and baitfish, but are usually not detrimental to larger fish (Mitchell 1995). However, in larger fish they give the fish a "wormy" appearance, rendering it unmarketable, as most consumers do not find the wormy appearance aesthetically pleasing. It is important to note that the asexual multiplication of the worm inside the host snail greatly increases the risk of infection to the next host (i.e., the fish), and exacerbates the problem of control. For example, a given infected snail may release tens to hundreds of cercariae each day over a several month lifespan. This amounts to thousands (to

millions) of potential grubs in infected fish resulting from very

few infected snails.

One method to control grub infections is to disrupt the life cycle of the snail. This can be



accomplished by eliminating one or more of the grubs' host species, i.e., the bird or snail. However, bird control is very problematic because the major hosts are often federally protected migratory species. There are a variety of methods used to control bird populations on farms (Mott and Brunson 1997). These include the use of pyrotechnics, human effigies that inflate and deflate, and reflective objects. Unfortunately, birds quickly become acclimated to these types of devices and control is only temporary.



Bird exclusion devices such as wire, electric fences, and netting have also been used. Additional control methods used with the aforementioned include border collies, limited killing of migratory birds with a depredation permit, complete screened-in enclosures, manipulating pond shape, or stocking larger fish into ponds. Nevertheless, none of these methods used alone is cost effective. Moreover, the use of any of these methods does not prevent bird hosts from flying or roosting over the ponds.

Another method to control the grubs is a therapeutic treatment to either prevent the grubs from penetrating the fish or to eliminate grubs that have penetrated the flesh. Lorio (1989) found that administration of Droncit[®] (praziquantel) and Ivomec[®] (ivermectin) by injection reduced the number of yellow grub (*Clinostomum marginatum*) metacercariae penetrations in catfish. Expense and the need to inject each fish directly make this method of control impractical for fish producers. To date, there is no Food and Drug Administration (FDA) approved method for treating grubs once they are imbedded in the tissue of the fish. Therefore, the best way to control infestations is to control or limit the snail populations in ponds.

RELATED CURRENT AND PREVIOUS WORK

Chemical, Biological, and Mechanical Control of Snails (Objective 1)

Control efforts to date have largely focused on reducing snail populations in aquaculture ponds through chemical, physical, or biological means (Mitchell 1995). To date, copper sulfate and other chemicals appear to be the best means to control snail populations in ponds. Although copper sulfate alone or in combination with citric acid is successful in controlling rams-horn (*Helisoma*) snails, its applicability to the NCR is limited because its effectiveness is based on warm water temperatures (>26.5°C; 79.7°F), and a complex combination of proper pH, total hardness, and total alkalinity (Mitchell 2002). Also, copper sulfate control has never been totally effective because snails retreat into bottom substrate where copper sulfate cannot affect them. Copper controlled release glass was also employed as a treatment for reducing snail populations, but its application rate and stability were affected by wind, water movement, and pond management (Chandiwana et al. 1987). The long-term use of copper as a control method may result in an excessive accumulation of copper and thus decrease the natural food base of larval fish (Atchison et al. 1996). Additional concerns are related to the toxicity of these chemicals to cultured fish or their food resource. For instance, applications of copper sulfate directly impact zooplankton populations at less than 10% of application rates for algae control (Allen 1997) and suggested concentrations of copper sulfate for snail control are much higher (Mitchell 2002).

Treatment with lime or lime slurry along pond banks has also been used to control snails. Similar to copper treatments, snails can migrate to the bottom of the pond and avoid the effects of the lime. The major drawback to using lime to control snails is that it changes the alkalinity of the water, which can negatively affect the ecology of the pond.

Francis-Floyd et al. (1997) found that Bayluscide[™] (*Niclosamide*) effectively controlled snail populations in ornamental fish production ponds. Unfortunately, Bayluscide[™] has not been approved by the FDA for use in food fish. Additionally, discharge of treated water is prohibited until 1-week post application (Avery et al. 2001).

Vulgarone B, a sesquiterpene from the plant *Artemisia douglasiana* Besser (Asteraceae) was also found to be effective as a molluscicide. Unfortunately, this natural derivative has also not been approved by the FDA and is also extremely difficult to extract in large quantities (Meepagala et al. 2004).

Sodium chloride concentrations of 2.5 ppt have also been used to control snail populations in aquaculture ponds (Venable et al 2000). Increased salinity may also have beneficial effects on fish by reducing osmotic stress. However, despite the potential osmotic and snail control benefits of higher salinity, it is often prohibitively expensive to maintain ponds at 2.5 ppt.

Physical alterations of culture ponds by draining or scraping can be an effective snail control mechanism. Although winter draining often eliminates pond snail populations completely, many production ponds cannot be drained or scraped due to year-round production and the negative impact that scraped suspended particles have on pond nutrient dynamics, fish feeding, and gill obstruction (Mitchell 1995). One method that has been somewhat successful in controlling snail populations is the elimination of vegetation within the water around the shallow areas of the pond as snails use the vegetation as a refuge and foraging area.

Natural enemies have the potential to be applied to several stages in the digenean trematode life cycle, and that snail and grub control will most profitably be accomplished by directing novel biocontrol strategies at both snail populations and the larval trematodes within infected snails is asserted. With varying degrees of success, at least 25 species of mollusc-eating fish have been tested as snail control agents (Slootweg et al. 1994). These include black carp (*Mylopharyngodon piceus*; Thomforde 2000; Ledford and Kelly 2006), redear sunfish (*Lepomis microlophus*; Wang et al. 2003; Ledford and Kelly 2006), African cichlids (*Serranochromis* spp.), sheepshead (*Aplodinotus grunniens*), East African catfish (*Clarias gariepinus*), and eastern mudminnow (*Umbra pygmaea*). However, few of these molluscivores are native to the NCR and most, being nonnative, are not permitted in some of the NCR states. Redear sunfish is the most widely distributed native fish, but previous studies have concluded it is insufficiently voracious at eating snails to be effective (Mitchell 2002). While at this point in time, the applicability of molluscivores remains limited in the NCR, different molluscivore fish hybrid combinations have yet to be tested for efficacy, and may provide significant control options. Additionally, the use of molluscivores in combination with chemical treatments has not been reported. Combination of the two methods may eliminate some of the disadvantages of using only chemical or biological methods

Biological control of grub infections in snails may also be achieved by introduction of a molluscivore crustacean predator or via the introduction of competitively dominant larval trematodes that exclude subordinate grub infections from snails (Lie 1973). Below, each of these control strategies is introduced and experiments to test the efficacy of each are outlined. Development of a comprehensive guide to snail control is also proposed. This guide will fill the information gap between past and present research conducted on snail control and the current snail problems in the NCR. An interactive Web-based site and publication will be produced to facilitate easy access to the available snail control information.

ANTICIPATED BENEFITS

Grub infections in fish culture ponds are extremely relevant to the aquaculture industry in the NCR as the industry has experienced a loss of income in both commercially important food fish species and baitfish. These economic losses result both directly from fish mortality due to trematode infection, and indirectly because of unappealing visual presentation of food fish fillets containing grubs. As a result, the industry has requested increased research into chemical, physical, and biological methods to control grubs or the intermediate hosts that facilitate the grub's life-cycle. Though previous research has identified some chemical, physical, and biological control methods, few are universally applicable to all culture facilities in the NCR and most cannot be used to treat grub problems in ponds where food fish are raised.

From the proposed investigations, both chemical and biological control methods will be tested for their efficiency and applicability to control grubs and manage snail populations in fish ponds. By utilizing locally available biological control species, e.g., crayfish, and establishing a suitable competitively dominant non-infectious trematode that can both displace the digenean trematodes and potentially control snail populations through castration of male snails, an economically viable, adaptable, universally applied, and immediate method of snail and grub management can be developed. The proposed work will also permit further experimental testing and demonstration of the dominance hierarchy for intramolluscan competition in larval trematodes and demonstrate another control method which may also have relevance to other trematode infections of veterinarian and human importance.

OBJECTIVES

- Investigate one or more methods of potentially useful approaches to snail population management and/or grub control. The methods of greatest interest include those that will be effective, economical, and approvable by state and federal regulators at commercial production scale. These methods will include reviewing what has been done elsewhere and designing studies that will address the NCRAC conditions, especially in pond systems for the production of economically important food fish for the region. Attempts will be made to investigate and refine these methods.
- Assemble an updatable snail management guide which includes a literature review of known control
 options, a method of determining snail infestation levels in any water system, and a set of standard
 operating procedures to reduce snail populations and trematode infestations based on the research
 cited in Objective 1.

PROCEDURES

Chemical, Biological, and Mechanical Control (Objective 1)

Southern Illinois University-Carbondale (SIUC)

General

This study will be initiated with a thorough review of the literature, plus interviews with researchers currently conducting snail control research. This study proposes to investigate single and integrated management schemes in which biological and chemical controls are used in conjunction with one another, in an effort to reduce snail populations in ponds. Current native species that have been examined include redear sunfish, blue catfish, freshwater drum, river redhorse and a few crayfish species. Other potential species include pumpkinseed sunfish, freshwater crayfish, and freshwater prawn, hybrid crosses of redear × bluegill, redear × green sunfish, redear × pumpkinseed, and redear × warmouth. Some of the aforementioned species are being examined with respect to control of the ram's horn snails by SIUC researchers and other institutions outside of the NCR. The applicability of these species to the NCR will also depend on their legal status within each state.

The two most abundant species of snails in aquaculture ponds are *Physa* spp. and *Helisoma* spp., therefore, these will be the target snail populations for this study. The following species will be examined for biological control of snails, all of which will be compared to the redear sunfish, which will serve as the control; redear × bluegill (female × male), redear × green sunfish, redear × warmouth, and freshwater prawn. Methods that were developed by Wang et al. 2003 and Ledford and Kelly 2006 will be utilized. First, 10 fish of each species and cross (10.0-12.0 cm; 3.9-4.7 in total length [TL]) and 10 freshwater prawn (10.0-12.0 cm; 3.9-4.7 in TL) that have not been fed for 24 hours will be placed into 20 separate 37.8-L (10.0-gal) aquaria and exposed to known sizes and numbers of Heliosoma and Physa snails for 48 hours. Snail sizes will represent all sizes of snails found in aquaculture ponds. Water temperature in the aquaria will be maintained at 26.0 ± 1°C (78.8°F). Water flow in the aquaria will be 2.5 L/min (0.7 gal/min). Observations on the number, size and species of snail consumed will be made. Species or hybrids that consume at least 50% of the snails in the previous aquaria studies will be used in trials that will investigate maximum handling size of each species of snail. This will be done by using three size classes of fish and prawn, small (10.0-15.0 cm; 3.9-5.9 in), medium (15.0-20.0 cm; 5.9-7.9 in), and large (20.0-25.0 cm; 7.9–9.8 in). Ten fish of each size class from each of the hybrid crosses and 10 prawns will be placed in individual aguaria (i.e., 1 fish/aguarium), acclimated for 5 days, starved for 24 hours, and then offered one snail from each size range of 3.0-12.0 mm (0.1-0.5 in) for Physa and 3.0-18.0 mm (0.1-0.7 in) for Helisoma. Fish and prawn will have access to the snails for 48 hours. Uneaten snails will be measured to identify those sizes that were ingested. Next, consumption rates will be determined by housing 20 individuals of each species in individual aquaria (i.e., 1 fish/aquaria) and offered various size classes of snails for 7 days. Snails will be individually measured and weighed daily as a group and placed into aquaria. After 24 hours, uneaten snails will be removed, measured, and weighed as a group. The number and size of snails consumed will be recorded. Mean daily consumption rate for each fish and prawn will be calculated for each 7-day trial.

Fish and/or prawn that eat snails in the tank trials will be utilized in pond studies. Three size classes of each species, small (10.0-15.0 cm; 3.9-5.9 in), medium (15.0-20.0 cm; 5.9-7.9 in), and large (20.0-25.0 cm; 7.9–9.8 in), will be stocked individually into 3, 0.04 ha (0.1 acre) ponds containing Physa and Helisoma snails. Snail populations will be sampled prior to stocking fish. The number of fish to be stocked will be based on the results of the daily consumption rate experiments conducted in tanks. Control ponds will not be stocked with potential biological control species. Fish will be stocked into ponds mid-May and will remain in ponds until mid-October. Prawn will be stocked into ponds in early June and will remain in ponds until mid September. Monthly, snail concentrations in all chemically treated ponds will be determined. Snails will be collected using a randomized grid system where a 1.0×1.0 m (3.3×3.3 ft) grid is superimposed over pond diagrams and sampling locations are randomly selected in two zones. Three replicate samples will be collected in the open-water zone (lack of vegetation) and three replicate samples will be collected in the vegetation zone (located near shore) at each pond. A dip net (0.5 m³; 17.7 ft³) will be used to collect samples in the vegetation, by collecting both submergent and emergent vegetation and 5.0 cm (2.0 in) of substrate, while an Ekman dredge (0.3 m² [3.2 ft²] total area per sample) will be used to collect open-water benthic samples. By differentiating between the two zones and collecting three replicates in each zone, the samples of the size chosen should be large enough to provide reproducible results while minimizing the possible effects of a heterogeneous distribution of the snails. Samples containing substrate will be sorted through sieves and all snail specimens removed, while samples containing vegetation will be examined by hand and any remaining snails collected. Snails will be counted, measured, and weighed.

Chemical controls have also been shown to provide snail control in ponds. Previous studies on hydrated lime, copper sulfate and citric acid, and salt have utilized only one concentration to control snails. The use of hydrated lime, copper sulfate plus citric acid, and salt in various concentrations will be examined to determine the optimum range to use to control snails. Three 2.025 ha (5.0 acre) ponds will be used for each chemical at each concentration. The alkalinity of the ponds will be determined prior to treatment in an effort to duplicate ponds with similar alkalinities. Hydrated lime will be tested at 45.4 and 56.7 kg/30.5 linear m (100.0 and 125.0 lb/100 linear ft) of pond bank. A 0.9 m (3.0 ft) swath on each pond bank will be treated with the appropriate concentration of the chemical. Hydrated lime will be added only when wind speeds are less than 5 mph. Copper sulfate plus citric acid will be tested at 0.59 kg (1.3 lb) copper sulfate plus 0.056 kg (0.13 lb) citric acid/ 91.5 linear m (300.0 linear ft) of pond bank. Copper sulfate plus citric acid will be added in a 0.6 m (2.0 ft) swath around the edge of the pond. Copper sulfate without citric acid will be tested at 0.6 kg/91.5 linear m (1.3 lb/300.2 linear ft) of pond bank. Salt will be added to nine ponds (three ponds per salt concentration) at rate of 1, 2, and 3 ppt. Three ponds not receiving any chemical treatment will serve as the control. Weekly, for 8 weeks, snail concentrations in all chemically treated ponds will be determined as previously described. Snails will be counted, measured, and weighed.

An integrated approach in which biological and chemical controls are used will also be evaluated. Many snail species will occupy vegetation in the pond, consequently grass carp will also be used in some of the replicates of this study. In this study 3, 0.04 ha (0.1 acre) ponds without any treatment will serve as the control. All ponds are known to have problems with snail populations. Three 0.04 ha (0.1 acre) ponds will be treated with the chemical treatment found to best control snails. Three 0.04 ha (0.1 acre) ponds will be treated with the chemical treatment found to best control snails and stocked with the biological control agent that best controlled snails in previous experiments at rates determined in the consumption studies. Three 0.04 ha (0.1 acre) ponds will be chemically treated, stocked with biological control agent, and stocked with grass carp at the rate of 20 carp/ha (10 carp/acre). Weekly, for 8 weeks, snail concentrations in all ponds will be determined as previously described. Snails will be counted, measured, and weighed.

To determine the effectiveness of the treatment under production scenarios, the previous pond studies will be conducted but also include phase III hybrid striped bass stocked at 1,089 kg/ha (6,000 lb/acre). In addition to the transects for identification of snail abundance and numbers by species, ponds will be seined once a month and a sample of 100 fish will be randomly selected to undergo gross observation for the presence of grubs on gills or fins. Once fish have been examined, they will be returned to the pond.

At the end of the growing season, 100 fish from each pond will be examined in four locations, the gills, fins, fillets, and skin of fillets, to quantify the prevalence of grub infection.

All data will be analyzed by analysis of variance (ANOVA) through the General Linear Model (GLM) using the Statistical Analysis System version 9.1 software (SAS Institute, Inc., Cary, North Carolina). In cases where significant differences are noted, subsequent comparison of means by Tukey's post-hoc tests will be performed. All decisions on significance will be made at the P < 0.05 level.

University of Wisconsin-Stevens Point (UW-Stevens Point)

Biocontrol with Crayfish Predators

The use of natural enemies of snails, other than fish, as a control mechanism has been successfully tested for controlling schistosomiasis in humans by using crayfish as predators of the intermediate host (Mkoji et al. 1999). Crayfish are opportunistic feeders that frequently feed on the most abundant prey (Hofkin et al. 1992; Appleton et al. 2004). Where crayfish have been placed in ponds, they have significantly reduced the snail populations (Michelson 1957), and lowered infection rates of Schistosoma haematobium in humans (Mkoji et al. 1999). Crayfish require calcium for their exoskeleton and can be voracious predators of snails for calcium (Hofkin et al. 1992). The northern (fantail) crayfish (Orconectes virilis) is native to all states in the NCR and has been investigated for its culture and market potential (Brown and Gunderson 1997). This crayfish is common throughout the NCR, can be cultured in ponds, and reach market size in one growing season (Brown et al. 1990). It has high (75%) overwinter survival, becomes sexually mature after one growing season, and is relatively disease free. Sufficient numbers of northern crayfish can be captured from streams and lakes (three rivers in Portage County, Wisconsin contain reproducing populations of O. virilis) or mating pairs can be captured and cultured in tanks. To test the ability of a native crayfish to control pond snail populations and reduce grub infestations in fish, a field study will be employed where native crayfish are stocked into grub-infected fish ponds and the impact the crayfish have on both snails and grubs will be compared to control ponds where crayfish are absent.

Crayfish Collection (Year 1)

Northern fantail crayfish (*Orconectes virilis*) will be collected from local rivers in central Wisconsin (Plover River, Tomorrow River, and Wisconsin River) in late summer and early fall using methods described in Hobbs and Jass (1988). Briefly, a 0.64 cm (0.25 in) mesh seine will be used to drag the bottom of the river between two blocking nets placed 30 m (100 ft) apart. Also, baited wire (minnow) traps will be placed in pooled areas and collected every 12 hours. Adult northern fantail crayfish will be brought to the UW-Stevens Point aquaculture laboratory where they will be maintained until stocking into treatment ponds. Using a monosex, adult population of crayfish should help to ensure consistency in densities used in each pond, because reproduction should not occur, and adult crayfish should be large enough in size so as not to be consumed by fish predators and be able to exert predation pressure on a range of snail sizes.

Captive crayfish will be placed in 6, 830.0-L (219.3-gal) commercial flow-through rearing tanks ($4.5 \times 0.5 \times 0.4 \text{ m}$; $14.8 \times 1.6 \times 1.3 \text{ ft}$) each equipped with a biofilter, heating, and/or cooling capability, and an aeration head tank. Crayfish will be cultured in the flow-through tanks during the collection period, being fed *ad libitum* using Zeigler shrimp feed (Zeigler Bros., Gardners, Pennsylvania) and supplied with brick structures to be used as artificial burrows. Adult fantail crayfish will be size sorted and identified based on gender using methods described in Huner (1994). Only male crayfish with a carapace length 30.0–50.0 mm (1.2–2.0 in) will be retained and stocked into fish ponds. Selection of this size range is based on three factors: (1) Wisconsin water regulations restrict crayfish trap sizes to <6.35 cm (2.5 in) diagonal opening, (2) previous studies have shown that crayfish within this size range are capable of inducing significant mortality on pond snails (Olsen et al. 1991; Nystrom and Perez 1998), and (3) a limited size range for adult crayfish will help to minimize confounding effects on intra-crayfish predation.

Field Study (Years 1 and 2)

Investigations of biocontrol with crayfish predators will be conducted at three commercial fish farms, and the field study will be repeated over 2 years to provide for adequate replication and accommodate site specific and year-year variation. These procedures will insure maximum applicability of the studies to the broadest number of fish farms in the NCR. A total of six ponds will be used for both treatments of biocontrol with crayfish predators and control ponds. Treatments will be randomly assigned to triplicate

ponds (receiving crayfish) so that one pond at each farm serves as a treatment and one pond at each farm serves as a control.

The study locations for crayfish predation on snails include Northside Enterprises, Black Creek, Wisconsin that has 4, 0.13 ha (0.33 acre) ponds and raises yellow perch. The ponds are fed with groundwater and are aerated. BrookCrest Fisheries, Cedar Grove, Wisconsin has 3, 0.2 ha (0.5 acre) ponds and raises yellow perch. The ponds are fed with groundwater and are aerated when needed. Druckery Farms, Abrams, Wisconsin has three ponds of 0.05, 0.1, and 0.2 ha (0.125, 0.25, and 0.5 acre) that are fed by groundwater and aerated as needed. Ponds are used to raise yellow perch. Ponds at all three facilities are infected with yellow grubs.

Baseline data including measurements of snail size-frequency distributions (based on shell diameter or length), density, prevalence of grubs in snails, and prevalence and intensity (i.e., percent fish infected and number of grubs per infected fish) measurements on fishes reared in the ponds will be obtained prior to introduction of northern fantail crayfish.

Snails will be collected using a randomized grid system where a 1.0×1.0 m (3.3×3.3 ft) grid is superimposed over pond diagrams and sampling locations are randomly selected in two zones. Three replicate samples will be collected in the open-water zone (lack of vegetation) and three replicate samples will be collected in the vegetation zone (located near shore) at each pond. A dip net (0.5 m^3 ; 17.7 ft^3) will be used to collect samples in the vegetation, by collecting both submergent and emergent vegetation and 5.0 cm (2.0 in) of substrate, while an Ekman dredge (0.3 m^2 ; 3.2 ft^2 total area per sample) will be used to collected open-water benthic samples. By differentiating between the two zones and collecting three replicates in each zone, the samples of the size chosen should be large enough to provide reproducible results while minimizing the possible effects of a heterogeneous distribution of the snails. Samples containing substrate will be sorted through sieves and all snail specimens removed, while samples containing vegetation will be examined by hand and any remaining snails collected.

Collected snails will be returned to the lab, measured (aperture to apex length for Physa spp., total diameter for Helisoma spp.), and dissected. Trematode infections in snails, even early infections, are easily detected by the presence of sporocyst or rediae stages. Observed larval trematode infections in snails will be identified to the lowest taxonomic level possible. Infections will be identified by sporocysts and cercariae and compared to original life cycle descriptions as given in Miller (1954) and Hoffman (1958) (Posthodiplostomum minimum), Hunter and Hamilton (1941) and Hoffman and Putz (1965) (Uvulifer ambloplitis), and Hopkins (1933) and Hunter (1939) (Clinostomum complanatum) as well as other summary descriptions as given in Yamaguti (1975) and Schell (1985). To ensure accurate identification of grub species, and possible cryptic species, grubs procured from naturally infected pond fishes will be fed to lab-reared ducks. Adult worms will thereby be cultured for voucher identification by standard morphological measurements. Furthermore, both grub infections in fishes, as well as rediae/sporocysts and cercariae from infected snails will be sampled, and tissue cryopreserved for the extraction of total genomic DNA. Polymerase chain reaction techniques will be employed using available published primers (e.g., a 425 base pairs region of cytochrome oxidase 1) suitable for species and/or genotype identifications. UW-Stevens Point has a well-equipped fish molecular genetics facility and a DNA sequencing and fingerprinting facility which will readily permit these analyses.

Encysted metacercariae of echinostomes can serve as an important source of mortality in snails (Lie and Ow-Yang 1973; Kuris and Warren 1980), and seasonal and treatment effects on this variable will be monitored. The foot, mantle, and pericardial region of dissected snails will be examined for metacercarial stages. Prevalence (percent of snails infected) and average intensity (number of cysts infected snails) will be calculated.

Fish will be sampled using Fyke nets (0.64 cm; 0.25 in mesh) placed in each pond for 12 hours. A sample of 24 fish/visit will be euthanized using 250 mg/L (ppm) of Finquel, preserved, and taken to the Wisconsin Department of Agriculture-Division of Animal Health lab for dissection and analysis. Fish will be examined in four locations, the gills, fins, fillets, and skin of fillets, to quantify the prevalence of grub infection. Snail density and prevalence of grubs in snails and fish will be analyzed using pairwise t-tests for each location. All decisions on significance will be made at the P < 0.05 level.

Adult, male northern fantail crayfish will be added to experimental treatment ponds in mid-June (at densities \leq 5 m⁻²; 53.8 ft²) and measurements on the aforementioned variables will continue. Crayfish will be randomly sampled from treatment ponds once each month (July–October) using baited wire traps and measured for size (carapace length as an indicator of growth) and catch-per-unit effort as a proxy for crayfish density, and returned to their ponds. Crayfish growth rates will be compared by ANOVA using a repeated measures statistical design.

Control ponds will be monitored with an identical protocol, but no crayfish will be added. All ponds will be randomly sampled biweekly for crayfish, snail, and grub variables, with an initial pre-crayfish stocking sample collected after ice-out (April), and then regular biweekly samples collected from time of crayfish stocking until ice-over (November). Crayfish will be sampled and monitored in both control and treatment ponds, with control ponds being carefully monitored for unintentional migration by crayfish from the treatment ponds. An analysis of the efficacy of crayfish as biocontrol agents will be made by comparing snail populations, prevalence of infected snails, and prevalence and intensity of grubs in fishes for treatment and control ponds.

Sample and data collection will be continued in Year 2, with biweekly random sample collection beginning after ice-out (April). Crayfish stocked into treatment ponds in Year 1 will be sampled after ice-out in Year 2 with measurements of size and catch-per-unit effort calculated as an estimate of crayfish density in each pond. As necessary, at the beginning of Year 2 additional adult, male crayfish will be added to ponds to return densities to original levels established in Year 1. This will help minimize the effects of varying crayfish density that may have resulted from a loss of crayfish in treatment ponds during Year 1. Data collection in Year 2 will continue until the ponds freeze.

Biocontrol with Natural Dominant Trematodes

The grubs, as larval trematodes in host mollusks, are parasitic castrators (Kuris 1974), either hormonally manipulating the host reproductive system, or physically displacing or digesting host reproductive tissue (Adema and Loker 1997). The reproductive resources of a snail host are limited, and two trematode species co-occurring in the same mollusc host engage in well-documented deterministic hierarchical competitive interactions in which the subordinate species is excluded (reviewed in Lie 1973; Kuris and Lafferty 1994). The competitive exclusion of subordinate trematodes in double infections has been repeatedly observed in both lab and field, and its constancy permitted the construction of competitive dominance hierarchies for many species of larval trematodes in molluscan hosts (Kuris 1990; Kuris and Lafferty 1994). Larval trematodes infecting snails produce tailed cercariae via either sporocysts or rediae. Sporocysts are simple worm-like sacs whereas rediae possess a mouth, powerful pharynx, and a gut. Not surprisingly, trematode species with large rediae feed upon sporocysts and smaller rediae of other species, leading to the exclusion of the subordinate species (Lie 1973; Kuris 1990; Kuris and Lafferty 1994).

Two of the three grub species identified by NCRAC produce only sporocysts inside host snails (Uvilifer ambloplitis and Posthodiplostomum minimum) while a third (Clinostomum complanatum) produces cercariae via relatively small redial stages. Each grub species are very likely to be competitively subordinate to many large rediae-producing trematode species (e.g., Echinostoma spp.; Kuris 1990; Kuris and Lafferty 1994). Therefore, the culture and application of locally available competitively dominant trematodes as competitors (and excluders) of the grub species has great potential to reduce both grub and snail populations. Among the best candidates for locally available dominant trematodes are Echinostoma spp. The life cycles of many Echinostoma spp. use the same snails as the grub species, but once released, Echinostoma cercariae encyst in the pericardial regions of snails (not fish). It is important to note and understand that the cercariae of Echinostoma spp. will not in any way infect or affect fishes, but they generally have a significant negative effect on infected snails. The introduction of *Echinostoma* spp. eggs to a system with grub-infected snails can reduce grubs in fishes due to three effects. First, Echinostoma rediae can exclude grub (i.e., sporocyst) infections in snails. Second, snails infected with Echinostoma are castrated, reducing snail reproduction. Finally, cercariae of Echinostoma spp. encyst in the pericardial regions of snails inducing snail mortality in high intensity infections (Lie and Ow-Yang 1973; Kuris and Warren 1980). Several Echinostoma species are naturally occurring and commonly encountered in central North America (Schell 1985). These include Echinostoma trivolvis and E. revolutum which use Helisoma spp. as hosts, and several other Echinostoma spp. which infect Physa spp. To test the efficacy of dominant trematodes in controlling grubs, both lab and field tests will be employed.

Lab study (Year 1)

Snails will be collected from fish ponds that are naturally infected with the grubs affecting fishes and cultured. Patent grub infections in snails collected at ponds will be identified by shedding cercariae. Field collected snails will be placed in individual containers with 5.0 mL (0.17 oz) of filtered pond water and the water will be examined after six hours for any shed cercariae.

Echinostoma trivolvis infecting *Helisoma* snails is locally abundant in McDill Pond near the UW-Stevens Point campus. Additionally, other snails naturally infected with *Echinostoma* spp. will be collected from local habitats. To determine the most appropriate final host, the metacercariae of these *Echinostoma* species will be fed to ducks, chicks, mice, and rats either in food or introduced via oral lavage. Beginning at two weeks post infection, eggs from adult worms will be obtained from the feces of these experimentally infected hosts. Eggs will be incubated in filtered pond water at 28.0°C (82.4°F), hatched, and previously collected grub-infected *Helisoma* and *Physa* snails will each be exposed to 5 *Echinostoma* miracidia. *Echinostoma* exposed snails will be maintained in 3.8-L (1.0-gal) aquaria and fed *ad libitum*. Exposed snails will be monitored over several weeks, and observations made by dissection to determine whether the locally cultured *Echinostoma* spp. exclude grub infections from exposed snails. Evidence of exclusion includes observation of physical displacement of grub larval stages within gonads and digestive glands of infected snails, total exclusion of grub sporocysts or rediae by echinostome rediae, and predation upon grub larval stages by echinostome rediae. At the conclusion of this lab experiment, the potential for *Echinostoma* spp. to control grub infections in snails will be evaluated, and the feasibility of continuing with the field study proposed for Year 2 will be determined.

Field study (Year 2)

Three fish rearing ponds will be chosen to serve as treatment ponds to which echinostome eggs will be added, and three additional ponds will serve as controls. The locations for the natural dominant trematode study include Blue Iris Fish Farm, Black Creek, Wisconsin which has two ponds of 1.2 and 0.05 ha (3.0 and 0.125 acre) that are completely fed by controlled runoff. The ponds are supplemented with well water and aeration and are used to raise yellow perch. Pepco Aquaculture, Cecil, Wisconsin has two ponds of 0.2 and 0.1 ha (0.5 and 0.25 acre) that are fed by groundwater and used to raise yellow perch and sunfish. AquaPoint Fish Farm, LLC, Stevens Point, Wisconsin has 2, 0.05 ha (0.125 acre) ponds that are filled with groundwater and supplied with aeration. Ponds are used to raise yellow perch, golden shiner, bluegill, green sunfish, and hybrid sunfish. Ponds at all four facilities are infected with yellow grubs. Treatments will be randomly assigned to ponds (receiving the natural dominant trematode) so that one pond at each farm serves as a treatment and one pond at each farm serves as a control.

Baseline data including measurements of snail size-frequency distributions, prevalence of grub infection in snails, prevalence on intensity of echinostome metacercariae, and prevalence and intensity measurements on fishes reared in the ponds, will be obtained for three months prior to introduction of echinostome eggs. Eggs of lab-cultured Echinostoma spp. procured from experimentally infected mallards (or other suitable hosts) will then be added to experimental treatment ponds and measurements on the above variables will continue. Control ponds will be monitored with an identical protocol, but no echinostome eggs will be added. An analysis of the efficacy of echinostome trematodes as biocontrol agents will be made by comparing snail populations and prevalence of grub-infected snails for treatment and control ponds at the time of fish harvest. To estimate daily Echinostoma egg output from experimentally infected hosts, several infected hosts will be maintained in the laboratory and daily fecal egg counts will be performed with standard sedimentation techniques. This will provide an estimate for total daily egg production. To evaluate a possible increase in echinostomiasis in transient waterfowl using experimental ponds, monthly bird counts will be conducted at both control and experimental ponds. Where possible by permit, or during hunting season, waterfowl present on ponds will be collected, necropsied, and assessed for any increase in echinostome infections. Year 1 (before field introduction of Echinostoma eggs) will serve as a control for Year 2.

Fish will be sampled using Fyke nets (0.64 cm; 0.25 in mesh) placed in each pond for 12 hours. A sample of 24 fish per visit will be euthanized using 250 ppm of Finquel, preserved, and taken to the Wisconsin Department of Agriculture-Division of Animal Health lab for dissection and analysis. Fish will be examined in four locations, the gills, fins, fillets, and skin of fillets, to quantify the prevalence of grub infection.

Prevalence of grubs in snails and fish will be analyzed using pairwise t-tests for each location. All decisions on significance will be made at the P < 0.05 level.

Assemble an Updatable Snail Management Guide (Objective 2)

Iowa State University (ISU)

Each of these three control methods, biological, chemical and mechanical, has advantages and disadvantages. By conducting and examining the historical literature sources, and using the up-to-date research data (collected in Objective 1), a management tree will then be developed. This matrix of management options will consist of an interactive Web site for fish producers to access and obtain information potentially relevant to their snail problems. Among the various options, information regarding effectiveness, application costs, legal implications, and potential for impact on pond general ecology, e.g., zooplankton dynamics in fish fingerling ponds, will be listed. A publication could also be available with the complete list of references and a similar management tree. The Web site could be maintained and updated as needed.

Extension Plan

Outreach will be accomplished in a timely manner and under terms agreeable among research and extension scientists, and involve industry consultation to effectively fulfill the NCRAC program goal. The extension liaison will determine recommended mechanism(s) for information dissemination of research findings and/or outreach activities that facilitate information transference to producers of yellow perch and largemouth bass. Results of the experiments, where appropriate, will be presented at scientific meetings and extension workshops and may be published in scientific journals, extension bulletins, or NCRAC fact sheets and bulletins. Research results will also be disseminated through the NCRAC Annual Progress Reports. Annual Progress Reports are assembled and edited by the extension liaison with input by the Principal Investigators. These reports are available on the NCRAC Web site (http://www.ncrac.org).

FACILITIES

<u>SIUC</u>

The Fisheries and Illinois Aquaculture Center has over 1,394 m² (15,000 ft²) of floor space in the Life Science II and Life Science III buildings located on the campus of SIUC. The twelve research laboratories house modern instrumentation for research in nutrition, biochemistry, genetics, water quality, physiology, toxicology, etc. The Center is also equipped with extensive computer software and capabilities including SAS®, Ethernet connections, color scanners, laser printers as well as video capture and digital film transfer.

A 6,940 m² (8,300 ft²), temperature-controlled wet laboratory building houses more than 30, 2.000-L (528.3-gal) tanks, 15, 1,200-L (317-gal) tanks, and 18, 1,200-L (317-gal) raceways as well as approximately 100 flow-through aquaria varying in size. At least 15 recirculating systems are employed allowing for numerous studies to be conducted simultaneously. Four of the aforementioned recirculating systems have been equipped to manipulate temperature and photoperiod from ambient laboratory conditions, allowing conditioning of brood stock for spawning indoors. Another of the aforementioned recirculating systems was designed especially as a hatchery, consisting of over 24 hatching jars, 4 Heath Tray racks, and 12, 350.0-L (92.5-gal) tanks. In addition, this wet laboratory building houses feed storage and feed manufacturing rooms, a water chemistry laboratory, a large workshop, and a small toxicology laboratory.

The Fisheries and Illinois Aquaculture Center also has a 90-pond research facility located at Southern Illinois University-Carbondale. These ponds have a surface area of 0.04–0.05 ha (0.10–0.12 acre) and are equipped with electricity for aeration. A 10.0-ha (24.7-acre) reservoir serves as a water source. A temperature-controlled building is available for feed storage. Tractors, vehicles, and paddlewheels are available. Additionally, there is a full-time pond manager for this facility.

UW-Stevens Point

The UW-Stevens Point Biology Department has two aquaculture labs equipped with 6, 2,000-L (530-gal) recirculating aquaculture systems, 28, 114-L (30-gal) aquaria, and 6, 830-L (219-gal) commercial flow-through rearing tanks $(4.5 \times 0.5 \times 0.4 \text{ m}; 14.8 \times 1.6 \times 1.3 \text{ ft})$ each equipped with a biofilter, heating, and/or cooling capability, and an aeration head tank. The UW-Stevens Point campus also maintains a six room animal care facility. This facility will be used to house the necessary mice, hamsters, chicks, and ducks that will be required for procuring adult trematodes. Several animal care protocols permitting the use of the above-listed vertebrate experimental hosts have been approved by the UW-Stevens Point Institutional Animal Care and Use Committee (IACUC), and all research on experimental hosts will be conducted in accordance with humane animal care and methods outlined in these protocols. Other resources include an Olympus SZX12 research stereomicroscope and Olympus BX41 (phase contrast) and BX60 (fluorescence) compound research microscopes. Each of the above-listed microscopes can be fitted with an Olympus DP12 digital camera for photodocumentation.

Northside Enterprises, Black Creek, Wisconsin has 4, 0.3-ha (0.33-acre) ponds and raises yellow perch. The ponds are fed with groundwater and are aerated. BrookCrest Fisheries, Cedar Grove, Wisconsin has 3, 0.2-ha (0.5-acre) ponds and raises yellow perch. The ponds are fed with groundwater and are aerated when needed. Druckery Farms, Abrams, Wisconsin has three ponds of 0.05, 0.1, and 0.2 ha (0.125, 0.25, and 0.5 acre) that are fed by groundwater and aerated as needed. Ponds are used to raise yellow perch. Blue Iris Fish Farm, Black Creek, Wisconsin has two ponds of 1.2 and .05 ha (3.0 and 0.125 acre) that are completely fed by controlled runoff. The ponds are supplemented with well water and aeration and are used to raise yellow perch. Pepco Aquaculture, Cecil, Wisconsin has two ponds of 0.5 and 0.1 ha (0.5 and 0.25 acre) that are fed by groundwater and used to raise yellow perch and sunfish. AquaPoint Fish Farm, LLC, Stevens Point, Wisconsin has 2, 0.05-ha (0.125-acre) ponds that are filled with groundwater and supplied with aeration. Ponds are used to raise yellow perch, golden shiner, bluegill, green sunfish, and hybrid sunfish.

<u>ISU</u>

ISU extension has Web design capabilities and all other equipment and staff necessary to fulfill the obligations in Objective 2. In addition, materials generated from this objective will be placed on the newly developed NCRAC Web site hosted at ISU.

REFERENCES

- Adema, C.M., and E.S. Loker. 1997. Specificity and immunobiology of larval digenean-snail associations. Pages 229-263 in B. Fried and T.K. Graczyk, editors. Advances in trematode biology, CRC Press, Inc., Boca Raton, Florida.
- Allen, E.A. 1997. Effects of copper on zooplankton and larval fish in aquaculture ponds and pelagic mesocosms. Master's thesis, Iowa State University, Ames.
- Appleton, C.C., Hofkin, B.V. and A. Baijnath. 2004. Macro-invertebrate predators of freshwater pulmonate snails in Africa, with particular reference to *Appasus grassei* (Heteroptera) and *Procambarus clarkii* (Decapoda). African Journal of Aquatic Science 29:185-193.
- Atchison, G.J., M.B. Sandheinrich, and M.D. Bryan. 1996. Effects of environmental stressors on interspecific interactions of aquatic animals. *In* M.C. Newman and C.H. Jogoe, editors. Ecotoxicology: a hierarchical treatment. CRC Press, Boca Raton, Florida.
- Avery, J., D. Wise, J. Terhune, and L. Khoo. 2001. New trematode in channel catfish. Thad Cochran National Warmwater Aquaculture Center, Fact Sheet 004 (revision 8/2001).

- Brown, P.B., and J. Gunderson, editors. 1997. Culture potential of selected crayfishes in the North Central Region. NCRAC Technical Bulletin Series #112, NCRAC Publications Office, Iowa State University, Ames.
- Brown, P.B., M.L. Hooe, and W.G. Blythe. 1990. Preliminary evaluation of production systems and forages for cultures of *Orconectes virilis*, the northern or fantail crayfish. Journal of the World Aquaculture Society 21:53-58.
- Chandiwana, S.K., J. Ndamba, O. Makura, and P. Taylor. 1987. Field evaluation of controlled release copper glass as a molluscicide in snail control. Transactions of the Royal Society of Tropical Medicine and Hygiene 81:952-955.
- Griffin, H., A, Cooke, M. Levy, and J. Flowers. 2002. Development of methods for controlling parasitic grub infections utilizing pond modification in hybrid striped bass aquaculture. North Carolina Sea Grant Fisheries Research Project 99-AM-17. Final Report. 38 pp.
- Francis-Floyd, R., J. Gildea, P. Reed, and R. Klinger. 1997. Use of Bayluscide (Bayer 73) for snail control in fish ponds. Journal of Aquatic Animal Health 9:41-48.
- Hobbs, H.H., and J.P. Jass. 1988. The crayfishes and shrimp of Wisconsin (Cambaridae, Palaemonidae). Milwaukee Public Museum, Milwaukee, Wisconsin.
- Hoffman, G.L. 1958. Experimental studies on the cercaria and metacercaria of a strigeid trematode, *Posthodiplostomum minimum.* Experimental Parasitology 7:23-50.
- Hoffman, G.L., and R.E. Putz. 1965. The black spot *Uvilifer ambloplitis* (Strigeoida) of centrarchid fish. Transactions of the American Microscopical Society 94:143-151.
- Hofkin, B.V., D.M. Hofinger, D.K. Koech, and E.S. Loker. 1992. Predation of *Biomphalaria pfeifferi* and non-target mollusks by the crayfish *Procambarus clarkii*: implications for the biological control of schistosomiasis. Annals of Tropical Medicine and Parasitology 86:663-670.
- Hopkins, S.H. 1933. Note on the life history of *Clinostomum marginatum* (Trematoda). Transactions of the American Microscopical Society 52:147-149.
- Huner, J.V. 1994, editor. Cultivation of freshwater crayfishes in North America: Section 1-Freshwater crayfish culture. Pages 5-156 *in* Freshwater crayfish aquaculture in North America, Europe, and Australia: Families Astacidae, Cambaridae, and Parastacidae. Food Products Press & The Haworth Press, Inc., Binghamton, New York.
- Hunter, G.W. 1939. Studies on *Clinostomum*. V. The cyst of the yellow grub of fish (*Clinostomum marginatum*). Proceedings of the Helminthological Society of Washington 6:73-76.
- Hunter, G.W., and J.M. Hamilton. 1941. Studies on host parasite reactions to larval parasites. IV. The cyst of *Uvilifer ambloplitis* (Hughes). Transactions of the American Microscopical Society 60:498-507.
- Kuris, A.M. 1974. Trophic interactions: similarity of parasitic castrators to parasitoids. The Quarterly Review of Biology 49:129-148.
- Kuris, A.M. 1990. Guild structure of larval trematodes in molluscan hosts: prevalence, dominance and significance of competition. *In* Esch, G.W., A.O. Bush, and J.M. Aho, editors. Parasite communities: patterns and processes. Chapman and Hall, New York.
- Kuris, A.M., and K.D. Lafferty. 1994. Community structure: larval trematodes in snail hosts. Annual Review of Ecology and Systematics 25:189-217.

- Kuris, A.M., and J. Warren. 1980. Echinostome cercarial penetration and metacercarial encystment as mortality factors for a second intermediate host, *Biomphalaria glabrata*. Journal of Parasitology 66:630-635.
- Ledford, J.J., and A.M. Kelly. 2006. A Comparison of black carp, redear sunfish, and blue catfish as biological controls of snail populations. North American Journal of Aquaculture 68:339-347.
- Lie, K.J. 1973. Larval trematode antagonism: principles and possible application as a control method. Experimental Parasitology 33:343-349.
- Lie, K.J., and C.K. Ow-Yang. 1973. A field trial to control *Trichobilharzia brevis* by dispersing eggs of *Echinostoma audyi*. Southeast Asian Journal of Tropical Medicine and Public Health 4:208-217.
- Lorio, W.J. 1989. Experimental control of metacercaiae of the yellow grub *Clinostomum marginatum* in channel catfish. Journal of Aquatic Animal Health 1:269-271.
- Meepagala, K.M., G. Sturtz, C.C. Mischke, D. Wise, and S.O. Duke. 2004. Molluscicidal activity of vulgarone B against ram's horn snail (*Planorbella trivolvis*). Pest Management Science 60:479-482.
- Michelson, E.H. 1957. Studies on the biological control of Schistosome-bearing snails; predators and parasites of fresh-water mollusca: a review of the literature. Parasitology 47:413-426.
- Miller, J.H. 1954. Studies on the life history of *Posthodiplostomum minimum* (MacCallum 1921). Journal of Parasitology 40:255-270.
- Mitchell, A.J. 2002. A copper sulfate-citric acid pond shoreline treatment to control the rams-horn snail *Planorbella trivolvis*. North American Journal of Aquaculture 64:182-187.
- Mitchell, A.J. 1995. Yellow grubs and other problems associated with aquatic birds. Aquaculture Magazine, July/August:93-97.
- Mkoji, G.M., B.V. Hofkin, A.M. Kuris, A. Stewart-Oaten, B.N. Mungai, J.H. Kihara, F. Mungai, J. Yundu, J. Mbui, J.R. Rashid, C.H. Kariuki, J.H. Ouma, D.K. Koech, and E.S. Loker. 1999. Impact of the crayfish *Procambarus clarkii* on *Schistosoma haematobium* transmission in Kenya. American Journal of Tropical Medicine and Hygiene 61:751-759.
- Mott, D.F., and M.W. Brunson. 1997. A historical perspective of catfish production in the southeast in relation to avian predation. Proceedings of the Eastern Wildlife Damage Management Conference 7:23-30.
- Nystrom, P., and J.R. Perez. 1998. Crayfish predation on the common pond snail (*Lymnaea stagnalis*): the effect of habitat complexity and snail size on foraging efficiency. Hydrobiologia 368:201-208.
- Olsen, T.M., D.M. Lodge, G.M. Capelli, and R.J. Houlihan. 1991. Mechanisms of impact of and introduced crayfish (*Oreconectes rusticus*) on littoral congeners, snails, and macrophytes 48:1853-1861.
- Overstreet, R.M., and S.S. Curran. 2004. Defeating diplostomoid dangers in USA catfish aquaculture. Folia Parisitologica 51:153-165.
- Schell, S.C. 1985. Handbook of trematodes of North America north of Mexico. University of Idaho Press. Moscow, Idaho.
- Slootweg, R.E., A. Malek, and F.S. McCullough. 1994. The biological control of snail intermediate hosts of schistosomiasis by fish control. Reviews in Fish Biology and Fisheries 4:67-90.
- Thomforde, H. 2000. Black carp control grub in fathead minnows. Aquaculture Magazine, March/April:23-24.

- USDA-NASS (United States Department of Agriculture-National Statistics Service). 2000. 1998 Census of Aquaculture. USDA, Washington, D.C.
- Venable, D.L., A.P. Gaude, III, and P.L. Klerks. 2000. Control of the trematode *Bolbophorus confusus* in ponds using salinity manipulation and polyculture with black carp *Mylopharyngodon piceus*. Journal of the World Aquaculture Society 31:158-166.
- Wang, H.P. R.S. Hayward, G.W. Whitledge, and S.A. Fischer. 2003. Prey-size preference, maximum size and consumption rates of redear sunfish feeding on two gastropods common to aquaculture ponds. Journal of the World Aquaculture Society 34:379-386.
- Yamaguti, S. 1975. A synoptical review of life histories of digenetic trematodes of vertebrates with special reference to the morphology of their larval forms. Keigaku Publishing Company, Tokyo.

PROJECT LEADERS

<u>State</u>	Name/Institution	Area of Specialization
Illinois	Gregory W. Whitledge Southern Illinois University-Carbondale	Aquaculture/Physiology
lowa	Joseph E. Morris Iowa State University	Aquaculture/Fisheries
	Richard D. Clayton Iowa State University	Aquaculture
Wisconsin	Christopher F. Hartleb University of Wisconsin- Stevens Point	Aquaculture/Fish Biology
	Todd Huspeni University of Wisconsin- Stevens Point	Parisitology

PARTICIPATING INSTITUTIONS AND PRINCIPAL INVESTIGATORS

Southern Illinois University-Carbondale (SIUC) Gregory W. Whitledge

University of Wisconsin-Stevens Point (UW-Stevens Point) Christopher F. Hartleb Todd Huspeni

Iowa State University University (ISU) Joseph E. Morris Richard D. Clayton

		DU	DOLI				
ORGANIZATION AND ADDRESS				USDA AWARD NO	 Year 1: Object 	ve 2	
Board of Trustees Southern Illinois University-Carbondale Carbondale, IL 62901	Duration Proposed Months: <u>12</u> _	Duration Proposed Months:	Non-Federal Proposed Cost- Sharing/	Non-federal Cost-Sharing/ Matching Funds			
PROJECT DIRECTOR(S) Gregory W. Whitledge	Funds Requested by Proposer	Funds Approved by CSREES (If different)	Matching Funds (If required)	Approved by CSREES (If Different)			
A. Salaries and Wages	CSREES FU	UNDED WORK	MONTHS				
1. No. of Senior Personnel	Calendar	Academic	Summer				
a (Co)-PD(s)							
b Senior Associates							
2. No. of Other Personnel (Non-Faculty)							
a Research Associates-Postdoctorates							
C Paraprofessionals							
d 2 Graduate Students				\$34 272			
				¢7,272			
e. <u>1</u> Prebaccalaureate Students				\$7,200			
f Secretarial-Clerical							
g Technical, Shop and Other							
Total Salaries and Wages \rightarrow				\$41,472			
B. Fringe Benefits (If charged as Direct Costs)							
C.Total Salaries, Wages, and Fringe Benefits (A plus	s B) →			\$41,472			
D. Nonexpendable Equipment (Attach supporting data for each item.)	a. List item	s and dollar	amounts				
E. Materials and Supplies		\$9,708					
F. Travel				\$2,500			
G. Publication Costs/Page Charges							
H. Computer (ADPE) Costs							
I. Student Assistance/Support (Scholarships/fellowsh education, etc. Attach list of items and dollar amou	nips, stipeno unts for eac	ds/tuition, co h item.)	ost of				
J. All Other Direct Costs (In budget narrative, list item provide supporting data for each item.)	is and dolla	ar amounts a	and	\$6,000			
KTotal Direct Costs (C through I)	→			\$59,680			
L. F&A/Indirect Costs. (If applicable, specify rate(s) activity. Where both are involved, identify itemized	and base(s costs in or	s) for on/off n/off campus	campus s bases.)				
MTotal Direct and F&A/Indirect Costs (J plus K)	. →						
NOther	→						
O Total Amount of This Request	→			\$59 680			
P. Carryover (If Applicable) Federal	Funds: \$		N	on-Federal funds	: \$	Total \$	
Q Cost Sharing/Matching (Breakdown of total am	ounte chou	wn in line C))				
 Q. Cost Snaring/Matching (Breakdown of total amounts shown in line O) Cash (both Applicant and Third Party)→ Non-Cash Contributions (both Applicant and Third Party) → 							
NAME AND TITLE (Type or print) SIGNATURE (required for revised budget only)							DATE
Project Director							DAIL
Authorized Organizational Representative							
Signature (for optional use)							

According to the Paperwork Reduction Act of 1995, an agency may not conduct or sponsor, and a person is not required to respond to a collection of information unless it displays a valid OMB control number. The valid OMB control number for this information collection is 0524-0039. The time required to complete this information collection is estimated to average 1.00 hour per response, including the time for reviewing instructions, searching existing data sources, gathering and maintaining the data needed, and completing the reviewing the collection of information.

		БО	DGET	· · · · · · · · · · · · · · · · · · ·			
ORGANIZATION AND ADDRESS Board of Trustees	USDA AWARD NO	D. Year 2: Object	ve 2				
Southern Illinois University-Carbondale Carbondale, IL 62901	Duration Proposed Months: _12_	Duration Proposed Months:	Non-Federal Proposed Cost- Sharing/	Non-federal Cost-Sharing/ Matching Funds			
PROJECT DIRECTOR(S) Gregory W. Whitledge				Year 2 Funds Requested by Proposer	Funds Approved by CSREES (If different)	Matching Funds (If required)	Approved by CSREES (If Different)
A. Salaries and Wages	CSREES FU	JNDED WORK	MONTHS				
1. No. of Senior Personnel	Calendar	Academic	Summer				
a (Co)-PD(s)							
b Senior Associates							
2. No. of Other Personnel (Non-Faculty)							
a Research Associates-Postdoctorates							
c Paraprofessionals							
d. <u>2</u> Graduate Students				\$35,986			
e1_ Prebaccalaureate Students				\$7,560			
f Secretarial-Clerical							
g Technical, Shop and Other							
Total Salaries and Wages→				\$43,546			
B. Fringe Benefits (If charged as Direct Costs)							
C.Total Salaries, Wages, and Fringe Benefits (A plus	sB) →			\$43,546			
D. Nonexpendable Equipment (Attach supporting data for each item.)	a. List item	s and dollar	amounts				
E. Materials and Supplies				\$9,114			
F. Travel				\$3,000			
G. Publication Costs/Page Charges							
H. Computer (ADPE) Costs							
I. Student Assistance/Support (Scholarships/fellowsh education, etc. Attach list of items and dollar amou	nips, stipeno unts for eac	ds/tuition, co	ost of				
J. All Other Direct Costs (In budget narrative, list item provide supporting data for each item.)	is and dolla	ir amounts a	and	\$6,000			
KTotal Direct Costs (C through I)	→						
L. F&A/Indirect Costs. (If applicable, specify rate(s) activity. Where both are involved, identify itemized	and base(s l costs in or	s) for on/off n/off campus	campus s bases.)				
MTotal Direct and F&A/Indirect Costs (J plus K)	.→	·	,				
NOther	→						
O Total Amount of This Request	\rightarrow			\$61,660			
P. Carryover (If Applicable)						Total \$	
Q. Cost Sharing/Matching (Breakdown of total amounts shown in line O) Cash (both Applicant and Third Party)→ Non-Cash Contributions (both Applicant and Third Party)							
NAME AND TITLE (Type or print)	(required for revise	ed budget only)		DATE			
Project Director							
Authorized Organizational Representative							
Signature (for optional use)							

According to the Paperwork Reduction Act of 1995, an agency may not conduct or sponsor, and a person is not required to respond to a collection of information unless it displays a valid OMB control number. The valid OMB control number for this information collection is 0524-0039. The time required to complete this information collection is estimated to average 1.00 hour per response, including the time for reviewing instructions, searching existing data sources, gathering and maintaining the data needed, and completing the reviewing the collection of information.

BUDGET EXPLANATION FOR SOUTHERN ILLINIOS UNIVERSITY-CARBONDALE

(Whitledge)

Objective 1

- A. Salaries and Wages. Year 1: Salaries are requested for two 50% FTE graduate students to spawn fish to make the hybrids, collect, and collate data. Additionally, funding is requested for one undergraduate student (20 hours a week for 45 weeks @ \$8.00/hr) to help in spawning fish, fertilizing larval fish ponds, and collecting data for the study. Year 2: Salaries are requested for two 50% FTE graduate students to spawn fish to make the hybrids, collect, and collate data. Additionally, funding is requested for one undergraduate student (20 hours a week for 45 weeks @ \$8.40/hour) to help in spawning fish, fertilizing larval fish ponds, and collecting data for the study.
- E. Materials and Supplies. Year 1: Supplies needed include: white bass and striped bass brood stock (hybrid × will serve as the production species; \$2,400), freshwater shrimp (\$1,500), fish food (\$1,870), general wet-laboratory, and office and record keeping supplies (\$300), air fills for scuba tanks (\$10/month = \$120), and chemicals including salt (\$2,592), copper sulfate (\$324), and citric acid (\$225). Supplies will also be needed to cross the parental species to make hybrid crosses including nets (\$47), buckets (\$30), spawning pans (\$100), LHRHa (\$100), and human chorionic gonadotropin (\$100). Year 2: Supplies needed include: white bass and striped bass brood stock to replace those lost in year 1 (hybrid × will serve as the production species; \$2,100), freshwater shrimp (\$1,500), fish food (\$1,870), general wet-laboratory, and office and record keeping supplies (\$300), air fills for scuba tanks (\$10/month = \$120), chemicals including salt (\$2,592), copper sulfate (\$324), and citric acid (\$225), and nets (\$53) and buckets (\$30) for general fish husbandry.
- F. Travel. Year 1: \$150 is requested for gasoline and meal expenses to collect brood fish from the wild and \$2,350 is requested for transportation, lodging, and meal expenses for the PI and graduate students to attend and present results at a multi-day conference in the U.S. at a location to be determined. Year 2: \$250 is requested for gasoline and meal expenses to collect brood fish from the wild and \$2,850 is requested for transportation, lodging, and meal expenses for the PI and graduate students to attend and present results at a multi-day conference in the U.S. at a location to be determined.
- I. All Other Direct Costs. Annual costs: vehicle lease (\$6,000).

UNITED STATES DEPARTMENT OF AGRICULTURE COOPERATIVE STATE RESEARCH, EDUCATION, AND EXTENSION SERVICE

		BU	DGEI				
ORGANIZATION AND ADDRESS	USDA AWARD NO. Year 1: Objective 1						
Northern Aquaculture Demonstration Facility & Department of E University of Wisconsin-Stevens Point 800 Reserve Street, Stevens Point, WI 54481	Duration Proposed Months: <u>12</u>	Duration Proposed Months:	Non-Federal Proposed Cost- Sharing/	Non-federal Cost-Sharing/ Matching Funds			
PROJECT DIRECTOR(S) Christopher F. Hartleb/Todd Huspeni	Funds Requested by Proposer	Funds Approved by CSREES (If different)	Matching Funds (If required)	Approved by CSREES (If Different)			
A. Salaries and Wages	CSREES FL	JNDED WORK	MONTHS				
1. No. of Senior Personnel	Calendar	Academic	Summer				
a (Co)-PD(s)							
b Senior Associates							
2. No. of Other Personnel (Non-Faculty) a Research Associates-Postdoctorates b Other Professionals							
c Paraprofessionals							
d. <u>1</u> Graduate Students				\$14,000			
e. <u>2</u> Prebaccalaureate Students				\$11,600			
f Secretarial-Clerical							
g Technical, Shop and Other							
Total Salaries and Wages			→	\$25.600			
B. Fringe Benefits (If charged as Direct Costs)				\$5.080			
C. Total Salaries, Wages, and Fringe Benefits (A p	lus B)		→	\$30,680			
 D. Nonexpendable Equipment (Attach supporting data for each item.) 	a. List item	s and dollar	amounts	\$00,000			
E. Materials and Supplies				\$8,100			
F. Travel				\$4,000			
G. Publication Costs/Page Charges							
H. Computer (ADPE) Costs							
I. Student Assistance/Support (Scholarships/fellowsh education, etc. Attach list of items and dollar amou	nips, stipend unts for eac	ds/tuition, co h item.)	ost of				
 All Other Direct Costs (In budget narrative, list item provide supporting data for each item.) 	ns and dolla	r amounts a	Ind				
K. Total Direct Costs (C through I)			→				
L. F&A/Indirect Costs. (If applicable, specify rate(s) activity. Where both are involved, identify itemized	and base(s I costs in or	s) for on/off h/off campus	campus s bases.)				
M. Total Direct and F&A/Indirect Costs (J plus K)			→	\$42,780			
N. Other			→				
O Total Amount of This Request			→	\$42 780			
P. Carryover (If Applicable)	l Funds: \$		N	on-Federal funds:	\$	Total \$	
 Q. Cost Sharing/Matching (Breakdown of total amounts shown in line O) Cash (both Applicant and Third Party) → Non-Cash Contributions (both Applicant and Third Party) 							
NAME AND TITLE (Type or print)	GNATURE	(required for revise	ed budget only)		DATE		
Project Director							
Authorized Organizational Representative							
Signature (for optional use)							

According to the Paperwork Reduction Act of 1995, an agency may not conduct or sponsor, and a person is not required to respond to a collection of information unless it displays a valid OMB control number. The valid OMB control number for this information collection is 0524-0039. The time required to complete this information collection is estimated to average 1.00 hour per response, including the time for reviewing instructions, searching existing data sources, gathering and maintaining the data needed, and completing the reviewing the collection of information.

UNITED STATES DEPARTMENT OF AGRICULTURE COOPERATIVE STATE RESEARCH, EDUCATION, AND EXTENSION SERVICE

		DU	JGLI				
ORGANIZATION AND ADDRESS	liology			USDA AWARD NO). Year 2: Objecti	ve 1	
University of Wisconsin-Stephens Point 800 Reserve Street, Stevens Point, WI 54481	Duration Proposed Months: <u>12</u>	Duration Proposed Months:	Non-Federal Proposed Cost- Sharing/	Non-federal Cost-Sharing/ Matching Funds			
PROJECT DIRECTOR(S) Christopher F. Hartleb/Todd Huspeni	Funds Requested by Proposer	Funds Approved by CSREES (If different)	Matching Funds (If required)	Approved by CSREES (If Different)			
A. Salaries and Wages	CSREES FL	JNDED WORK	MONTHS				
1. No. of Senior Personnel	Calendar	Academic	Summer				
a (Co)-PD(s)							
b Senior Associates							
2. No. of Other Personnel (Non-Faculty) a Research Associates-Postdoctorates b Other Professionals							
c Paraprofessionals							
d. <u>1</u> Graduate Students				\$14,000			
e2_ Prebaccalaureate Students				\$11,600			
f Secretarial-Clerical							
g Technical, Shop and Other							
Total Salaries and Wages			→	\$25.600			
B Fringe Benefits (If charged as Direct Costs)				\$5,080			
C. Total Salaries, Wages, and Fringe Benefits (A p	lus B)		→	\$30,680			
 Nonexpendable Equipment (Attach supporting data for each item.) 	a. List items	s and dollar	amounts	\$30,000			
E. Materials and Supplies		\$4,700					
F. Travel	\$5,000						
G. Publication Costs/Page Charges							
H. Computer (ADPE) Costs							
I. Student Assistance/Support (Scholarships/fellowsh education, etc. Attach list of items and dollar amou	nips, stipend unts for eac	ds/tuition, co h item.)	ost of				
 J. All Other Direct Costs (In budget narrative, list item provide supporting data for each item.) 	is and dolla	r amounts a	Ind				
K. Total Direct Costs (C through I)			→				
L. F&A/Indirect Costs. (If applicable, specify rate(s) activity. Where both are involved, identify itemized	and base(s costs in on	s) for on/off h/off campus	campus s bases.)				
M. Total Direct and F&A/Indirect Costs (J plus K)				\$40,380			
N. Other			→				
O. Total Amount of This Request			→	\$40,380			
P. Carryover (If Applicable)						Total \$	
Q. Cost Sharing/Matching (Breakdown of total amounts shown in line O) Cash (both Applicant and Third Party) Non-Cash Contributions (both Applicant and Third Party)							
NAME AND TITLE (Type or print)		SI	GNATURE	(required for revise	ed budget only)		DATE
Project Director							
Authorized Organizational Representative							
Signature (for optional use)							

According to the Paperwork Reduction Act of 1995, an agency may not conduct or sponsor, and a person is not required to respond to a collection of information unless it displays a valid OMB control number. The valid OMB control number for this information collection is 0524-0039. The time required to complete this information collection is estimated to average 1.00 hour per response, including the time for reviewing instructions, searching existing data sources, gathering and maintaining the data needed, and completing the reviewing the collection of information.

BUDGET EXPLANATION FOR UNIVERSITY OF WISCONSIN-STEVENS POINT

(Hartleb and Huspeni)

Objective 1

- A. Salaries and Wages. Year 1: Salary for graduate student to assist PIs in research (\$14,000) and wages for undergraduate students to assist PIs and graduate student in the study (580 hours@\$10/hour for 2 students for a total of \$11,600). Year 2: Salary for graduate student to assist PIs in research (\$14,000) and wages for undergraduate students to assist PIs and graduate student in the study (580 hours@\$10/hour for 2 students for a total of \$11,600).
- **B.** Fringe Benefits. Annual costs: 30% for graduate student (\$4,200) and 7.59% for undergraduate student (\$440/student for a total of \$880).
- E. Materials and Supplies. Year 1: Purchase crayfish traps and feed (\$2,000), laboratory supplies for culturing competitive trematodes (\$2,100), snail collection traps/plates, and hosts for infection with metacercariae (including feed, bedding, and animal care facility charges; \$4,000). Year 2: \$4,700 for costs associated with feed for crayfish and snail hosts and additional traps for crayfish and snails.
- F. Travel. Year 1: \$4,000 is requested for vehicle charges to travel from Stevens Point to fish farm pond field sites, local ponds/lakes for trematodes collection, and crayfish collection at rivers. Also, travel from Madison, Wisconsin to fish farm pond field sites for fish health inspection/grub data collection. Year 2: \$5,000 is requested for vehicle charges to travel from Stevens Point to fish farm pond field sites and from Madison, Wisconsin to pond sites for fish health inspection/grub data collection.

		50	JGLI		Veer 1. Ohieeti		
Department of Natural Resource Ecology and Management	USDA AWARD NO	 Year 1: Objecti 	ve 2	Nov fodovol			
lova State University Ames, IA 50011-3221	Proposed Months: 12	Proposed Months:	Proposed Cost- Sharing/	Cost-Sharing/			
PROJECT DIRECTOR(S) Joseph E. Morris	Funds Requested by Proposer	Funds Approved by CSREES (If different)	Matching Funds (If required)	Approved by CSREES (If Different)			
A. Salaries and Wages	CSREES FU	JNDED WORK	MONTHS				
1. No. of Senior Personnel	Calendar	Academic	Summer				
a (Co)-PD(s)							
b Senior Associates							
2. No. of Other Personnel (Non-Faculty) a Research Associates-Postdoctorates b. Other Professionals							
c Paraprofessionals							
d. <u>1</u> Graduate Students				\$7,500			
e1_ Prebaccalaureate Students				\$1,000			
f Secretarial-Clerical							
g Technical, Shop and Other							
Total Salaries and Wages			→	\$8,500			
B. Fringe Benefits (If charged as Direct Costs)				\$ 962			
C. Total Salaries, Wages, and Fringe Benefits (A	olus B)		→	\$9,462			
 Nonexpendable Equipment (Attach supporting da for each item.) 	ta. List item	s and dollar	amounts				
E. Materials and Supplies				\$ 716			
F. Travel				\$1,500			
G. Publication Costs/Page Charges							
H. Computer (ADPE) Costs							
I. Student Assistance/Support (Scholarships/fellows education, etc. Attach list of items and dollar amo	hips, stipeno ounts for eac	ds/tuition, co h item.)	ost of				
J. All Other Direct Costs (In budget narrative, list iter provide supporting data for each item.)	ms and dolla	ir amounts a	ind				
K. Total Direct Costs (C through I)			→				
L. F&A/Indirect Costs. (If applicable, specify rate(s activity. Where both are involved, identify itemize) and base(s d costs in or	s) for on/off n/off campus	campus bases.)				
M. Total Direct and F&A/Indirect Costs (J plus K)				\$11,678			
N. Other			→				
O. Total Amount of This Request			→	\$11,678			
P. Carryover (If Applicable)	al Funds: \$		N	on-Federal funds	\$	Total \$	
Q. Cost Sharing/Matching (Breakdown of total an Cash (both Applicant and Third Party)	Third Port	wn in line O)		→ _→		
NAME AND TITLE (Type or print) SIGNATURE (required for revised budget only)							DATE
Authorized Organizational Representative							
Signature (for optional use)							

According to the Paperwork Reduction Act of 1995, an agency may not conduct or sponsor, and a person is not required to respond to a collection of information unless it displays a valid OMB control number. The valid OMB control number for this information collection is 0524-0039. The time required to complete this information collection is estimated to average 1.00 hour per response, including the time for reviewing instructions, searching existing data sources, gathering and maintaining the data needed, and completing the reviewing the collection of information.

		DU	DGET	1			
ORGANIZATION AND ADDRESS				USDA AWARD NO). Year 2: Objecti	ve 2	
Iowa State University Ames, IA 50011-3221	Duration Proposed Months: _12_	Duration Proposed Months:	Non-Federal Proposed Cost- Sharing/	Non-federal Cost-Sharing/ Matching Funds			
PROJECT DIRECTOR(S) Joseph E. Morris				Funds Requested by Proposer	Funds Approved by CSREES (If different)	Matching Funds (If required)	Approved by CSREES (If Different)
A. Salaries and Wages	CSREES FL	JNDED WORK	MONTHS				
1. No. of Senior Personnel	Calendar	Academic	Summer				
a (Co)-PD(s)							
2. No. of Other Personnel (Non-Faculty) a Research Associates-Postdoctorates b. Other Professionals							
c Paraprofessionals							
d. <u>1</u> Graduate Students				\$4,500			
e. <u>1</u> Prebaccalaureate Students				\$1,500			
f Secretarial-Clerical							
g Technical, Shop and Other							
Total Salaries and Wages			→	\$6.000			
B. Fringe Benefits (If charged as Direct Costs)				\$ 668			
C. Total Salaries, Wages, and Fringe Benefits (A p	lus B)		→	\$6.668			
D. Nonexpendable Equipment (Attach supporting dat for each item.)	a. List item	s and dollar	amounts	+ ,			
E. Materials and Supplies							
F. Travel	\$2,154						
G. Publication Costs/Page Charges							
H. Computer (ADPE) Costs							
I. Student Assistance/Support (Scholarships/fellowsl education, etc. Attach list of items and dollar amo	hips, stipend unts for eac	ds/tuition, co h item.)	ost of				
J. All Other Direct Costs (In budget narrative, list iten provide supporting data for each item.)	ns and dolla	r amounts a	Ind				
K. Total Direct Costs (C through I)			→				
L. F&A/Indirect Costs. (If applicable, specify rate(s) activity. Where both are involved, identify itemized	and base(s d costs in or	s) for on/off n/off campus	campus s bases.)				
M. Total Direct and F&A/Indirect Costs (J plus K)			→	\$8,822			
N. Other			→				
O Total Amount of This Request			→	\$8 822			
P. Carryover (If Applicable)							
Q. Cost Sharing/Matching (Breakdown of total amounts shown in line O) Cash (both Applicant and Third Party) Non-Cash Contributions (both Applicant and Third Party)							
NAME AND TITLE (Type or print) SIGNATURE (required for revised budget only)						DATE	
Project Director				•			
Authorized Organizational Representative							
Signature (for optional use)							

According to the Paperwork Reduction Act of 1995, an agency may not conduct or sponsor, and a person is not required to respond to a collection of information unless it displays a valid OMB control number. The valid OMB control number for this information collection is 0524-0039. The time required to complete this information collection is estimated to average 1.00 hour per response, including the time for reviewing instructions, searching existing data sources, gathering and maintaining the data needed, and completing the reviewing the collection of information.

BUDGET EXPLANATION FOR IOWA STATE UNIVERSITY (Morris and Clayton)

Objective 2

- A. Salaries and Wages. Year 1: Salary for graduate student to assist PIs in developing extension project (5 months@\$1,500/month = \$7,500) and wages for undergraduate student to assist PIs and graduate student in the study (125 hours@\$8/hour = \$1,000). Year 2: Salary for graduate student to assist PIs in developing extension project (3 months@\$1,500/month = \$4,500) and wages for undergraduate to assist PI and graduate student in the study (176 hours@\$8.50/hour = \$1,500).
- **B.** Fringe Benefits. Year 1: 11.5% for graduate student (\$862) and 10% for undergraduate worker (\$100). Year 2: 11.5% for graduate student (\$518) and 10% for undergraduate worker (\$150).
- **E. Material and Supplies.** Year 1: Purchase Dream Weaver software for Web site development (\$400) and cost of materials collection and analyses (\$316).
- F. Travel. Year 1: Transportation, lodging, and meal expenses for the PIs and graduate student to meet with other project PIs to collect references and input into the snail management tree (\$1,500). Year 2: Transportation, lodging, and meal expenses for the PI to consult with PIs funded in Objective #1 concerning their respective results as well present grub management tree at a regional aquaculture workshop (\$2,154).

BUDGET SUMMARY FOR EACH PARTICIPATING INSTITUTION

	SIUC	UW- Stevens Point	ISU	TOTALS
Salaries and Wages	\$41,472	\$25,600	\$8,500	\$75,572
Fringe Benefits	\$ O	\$5,080	\$ 962	\$6,042
Total Salaries, Wages, and Fringe Benefits	\$41,472	\$30,680	\$9,462	\$81,614
Nonexpendable Equipment	\$ O	\$ 0	\$ O	\$ O
Materials and Supplies	\$9,708	\$8,100	\$ 716	\$18,524
Travel	\$2,500	\$4,000	\$1,500	\$8,000
All Other Direct Costs	\$6,000	\$ O	\$ O	\$6,000
TOTAL PROJECT COSTS	\$59,680	\$42,780	\$11,678	\$114,138

Year 1

Year 2

	SIUC	UW-Stevens Point	ISU	TOTALS
Salaries and Wages	\$43,546	\$25,600	\$6,000	\$75,146
Fringe Benefits	\$ 0	\$5,080	\$ 668	\$5,748
Total Salaries, Wages, and Fringe Benefits	\$43,546	\$30,680	\$6,668	\$80,894
Nonexpendable Equipment	\$ 0	\$ 0	\$ O	\$ O
Materials and Supplies	\$9,114	\$4,700	\$ O	\$13,814
Travel	\$3,000	\$5,000	\$2,154	\$10,154
All Other Direct Costs	\$6,000	\$ O	\$ O	\$6,000
TOTAL PROJECT COSTS	\$61,660	\$40,380	\$8,822	\$110,862

SCHEDULE FOR COMPLETION OF OBJECTIVES

Objective 1: Initiated in Year 1 completed in Year 2.

Objective 2: Initiated in Year 1 completed in Year 2.

LIST OF PRINCIPAL INVESTIGATORS

Richard D. Clayton, Iowa State University Christopher F. Hartleb, University of Wisconsin-Stevens Point Todd Huspeni, University of Wisconsin-Stevens Point Joseph E. Morris, Iowa State University Gregory W. Whitledge, Southern Illinois University-Carbondale

Richard D. Clayton Department of Natural Resource Ecology and Management Iowa State University 339 Science II Ames, Iowa 50011-3221 Phone: (515) 294-8616 Fax: (515) 294-7874 E-mail: rclayton@iastate.edu

EDUCATION

B.S. Iowa State University, 1992, Fisheries and Wildlife Biology

M.S. Iowa State University, 2007, Fisheries Biology

POSITIONS

Extension Aquaculture Specialist/Ag Specialist II (2004-present), Research Associate I (2003-2004), Research Associate I (1997-2003), and Research Associate (1992-1997), Iowa State University Department of Natural Resource Ecology and Management

SCIENTIFIC AND PROFESSIONAL ORGANIZATIONS

American Fisheries Society Iowa Chapter of the American Fisheries Society Iowa Aquaculture Association Sigma Xi

SELECTED PUBLICATIONS

- Summerfelt, R.C., and R.D. Clayton. 2007. Evaluation of open-formula grower diets for juvenile walleye. North American Journal of Aquaculture 69:53:58.
- Summerfelt, R.C., J.A. Forsberg, R.D. Clayton, and B. Barton. 1999. Low stress procedures for transporting juvenile walleye. Completion Report for the Fisheries Bureau, Fish and Wildlife Division, Iowa Department of Natural Resources, Des Moines. 70 pp.
- Clayton, R.D., T.L. Stevenson, and R.C. Summerfelt. 1998. Fin erosion in intensively cultured walleyes and hybrid walleyes. Progressive Fish-Culturist 60:114-118.
- Phillips, T.A., R.C. Summerfelt, and R.D. Clayton. 1998. Feeding frequency effects on water quality and growth of walleye fingerlings in intensive culture. Progressive Fish-Culturist 60:1-8.
- Summerfelt, R.C., A. Moore, B.T. Bristow, and R.D. Clayton. 1997. Culture of advanced walleye fingerlings. Iowa Department of Natural Resources, Federal Aid to Fish Restoration Project F-151-R, Completion Report, Des Moines. 91 pp.

Bristow, B.T., R.C. Summerfelt, and R.D. Clayton. 1996. Comparative performance of intensively cultured larval walleye in clear, turbid, and colored water. Progressive Fish-Culturist 58:1-10.

- Clayton, R.D., and R.C. Summerfelt. 1996. Toxicity of hydrogen peroxide to fingerling walleyes. Journal of Applied Aquaculture 6:39-49.
- Summerfelt, R.C., R.D. Clayton, T.K. Yager, S.T. Summerfelt, and K.L. Kuipers. 1996. Live weightdressed weight relationships of walleye and hybrid walleye. Pages 241-250 in R.C. Summerfelt, editor. Walleye Culture Manual. NCRAC Culture Series #101. NCRAC Publications Office, Iowa State University, Ames.

Christopher F. Hartleb Department of Biology University of Wisconsin-Stevens Point 800 Reserve Street Stevens Point, WI 54481 Phone: (715) 346-3228 Fax: (715) 346-3624 E-Mail: chartleb@uwsp.edu

EDUCATION

B.S. Rensselaer Polytechnic Institute, 1990, Biology

M.S. University of New Hampshire, 1992, Zoology (Limnology)

Ph.D. University of Maine, Maine Cooperative Fish & Wildlife Research Unit, 1996, Fisheries Biology

POSITIONS

Co-Director, Northern Aquaculture Demonstration Facility (2006-present), and Associate Professor of Fisheries Biology and Aquaculture (2002-present), and Assistant Professor of Biology & Water Resources

(1996-2002), Department of Biology, and University of Wisconsin-Stevens Point Researcher Assistant (1992-1996), Maine Cooperative Fish and Wildlife Research Unit, University of Maine Research Assistant (1990-1992), Lakes Fish Condition Program, University of New Hampshire Research Assistant (1988-1990), Rensselaer Fresh Water Institute, Rensselaer Polytechnic Institute

SCIENTIFIC AND PROFESSIONAL ORGANIZATIONS

American Fisheries Society, Fish Culture Section North American Benthological Society Wisconsin Aquaculture Industry Advisory Council World Aquaculture Society / U.S. Aquaculture Society

- Malison, J.A., and C.F. Hartleb. 2005. A manual of best management practices for aquaculture in Wisconsin and the Great Lakes region. University of Wisconsin Sea Grant Institute, Madison.
- Hartleb, C.F. 2004. Floating raceways to raise yellow perch at cranberry farms. Aquaculture Magazine Jan./Feb.:18-24.
- Hartleb. C.F. 2003. Food chain dynamics and diets of larval and post-larval yellow perch in culture ponds. Pages 31-32 *in* T.P. Barry and J.A. Malison, editors. Proceedings of Percis III: The Third International Percid Fish Symposium. University of Wisconsin Sea Grant Institute, Madison, Wisconsin.
- Hartleb, C.F., and S.A. Timm. 2000. Survival and hatching success of stonefly eggs (*Paragnetina media*) following ingestion by three stream fishes. Journal of Freshwater Ecology 15:107-114.
- Hartleb, C.F., and J.F. Haney. 1998. Use of a thermal and light refugium by *Daphnia* and its effects on foraging pumpkinseeds. Environmental Biology of Fishes 51:339-349.
- Hartleb, C.F. 1996. Ecology and performance of stocked brook trout (*Salvelinus fontinalis*) in two Maine ponds. Doctoral dissertation. University of Maine, Orono, Maine.
- Hartleb, C.F., and J.R. Moring. 1995. An improved gastric lavage device for removing stomach contents from live fish. Journal of Fisheries Research 24:261-266.
- Hartleb, C.F., J.D. Madsen, and C.W. Boylen. 1993. Environmental factors affecting seed germination in *Myriophyllum spicatum* L. Aquatic Botany 45:15-25.

Todd Huspeni Department of Biology University of Wisconsin-Stevens Point 800 Reserve Street Stevens Point, WI 54481 Phone: (715) 346-4250 Fax: (715) 346-3624 E-mail: thuspeni@uwsp.edu

EDUCATION

- B.A. University of Minnesota, Twin Cities, 1990, Biology and History, *summa cum laude*
- M.A. University of California, Santa Barbara, 1997, Population Biology and Invertebrate Zoology

Ph.D. University of California, Santa Barbara, 2000, Parasite Ecology

POSITIONS

Assistant Professor of Parasitology (2004-present), Department of Biology, University of Wisconsin-Stevens Point

Assistant Research Biologist (2001-2004), Marine Science Institute, and Lecturer (2001-2004), Department of Ecology, Evolution and Marine Biology, University of California-Santa Barbara

SCIENTIFIC AND PROFESSIONAL ORGANIZATIONS

American Malacological Union American Society of Parasitologists The Helminthological Society of Washington Western Society of Naturalists

- Hechinger, R.F., K.D. Lafferty, T.C. Huspeni, A.J. Brooks, and A.M. Kuris. 2007. Can parasites indicate free living diversity? Relationships between the species richness and abundance of larval trematodes and local benthos and fishes. Oecologia 151:82-92
- Huspeni, T.C., Hechinger, R.F., and K.D. Lafferty. 2005. Trematode parasites as estuarine indicators:
 Opportunities, applications and comparisons with conventional community approaches. Pages 297-314 in
 S. Bortone, editor. Estuarine indicators. CRC Press, Boca Raton, Florida.
- Torchin, M.E., J.E. Byers, and T.C. Huspeni. 2005. Differential parasitism of native and introduced snails: Replacement of a parasite fauna. Biological Invasions 7: 885-894.
- Torchin, M.E., R.F. Hechinger, T.C. Huspeni, K. Whitney, and K.D. Lafferty. 2005. Ecology of the introduced ribbed mussel (*Geukensia demissa*) in Estero de Punta Banda, Mexico: interactions with the native cord grass, *Spartina foliosa*. Biological Invasions 7:607-614.
- Huspeni, T.C., and K.D. Lafferty. 2004. Using larval trematodes that parasitize snails to evaluate a saltmarsh restoration project. Ecological Applications 14:795-804.
- Huspeni, T.C. 2000. A molecular genetic analysis of host specificity, continental geography, and recruitment dynamics of a larval trematode in a salt marsh snail. Doctoral dissertation. University of California, Santa Barbara.

Joseph E. Morris Department of Natural Resource Ecology and Management Iowa State University 339 Science II Ames, IA 50011-3221 Phone: (515) 294-4622 Fax: (515) 294-7874 E-mail: jemorris@iastate.edu

EDUCATION

- B.S. Iowa State University, 1979, Fisheries and Wildlife Biology
- M.S. Texas A&M University, 1982, Wildlife and Fisheries Sciences
- Ph.D. Mississippi State University, 1988, Fisheries and Wildlife

POSITIONS

Fisheries and Aquaculture Specialist/Associate Professor (1995-present), Specialist/Assistant Professor (1988-present), Department of Natural Resource Ecology and Management, Iowa State University and Associate Director, North Central Regional Aquaculture Center (1990-present)
Graduate Research Assistant (1986-1988), Mississippi State University
Aquaculture Manager (1982-1986), Stiles Farm Foundation
Graduate Research Assistant (1981-1982), Texas A&M University
Research Technician I (1980-1981), Texas A&M University
Fisheries Biologist Aide (1979), Indiana Department of Natural Resources

SCIENTIFIC AND PROFESSIONAL ORGANIZATIONS

American Fisheries Society: Iowa Chapter; Education, Fish Culture, Early Life History, and Fish Management Sections Iowa Aquaculture Association World Aquaculture Society Phi Kappa Phi Sigma Xi

- Rogge, M.L., A.A. Moore, and J.E. Morris. 2003. Organic and mixed organic-inorganic fertilization of plastic-lined ponds for fingerling walleye culture. North American Journal of Aquaculture 65:179-190.
- Boylan, J.D., and J.E. Morris. 2003. Limited effects of barley straw on algae and zooplankton in a midwestern pond. Lake and Reservoir Management 19(3):265-271.
- Lane, R.L., and J.E. Morris. 2002. Comparison of prepared feed versus natural food ingestion between pond-cultured bluegill and hybrid sunfish. Journal of the World Aquaculture Society 33: 517-519.
- Mischke, C.C., G. Dvorak, and J.E. Morris. 2001. Growth and survival of hybrid sunfish larvae in the laboratory under different feeding and temperature regimes. North American Journal of Aquaculture 63:265-271.
- Lane, R.L., and J.E. Morris. 2000. Biology, prevention, and effects if common grubs (Digenetic trematodes) in freshwater fish. NCRAC Technical Bulletin Series #115, NCRAC Publications Office, lowa State University, Ames.
- Morris, J.E., and C.C. Mischke. 1999. Plankton management for fish culture ponds. NCRAC Technical Bulletin Series, #114, NCRAC Publications Office, Iowa State University, Ames.

Gregory W. Whitledge Fisheries and Illinois Aquaculture Center and Center for Ecology Department of Zoology Southern Illinois University Carbondale, IL 62901-6511 Phone: (618) 453-6089 Fax: (618) 453-6095 E-mail: gwhit@siu.edu

EDUCATION

- B.S. The University of Texas at Austin, 1993, Aquatic Biology
- M.S. University of Missouri-Columbia, 1996, Fisheries
- Ph.D. University of Missouri-Columbia, 2001, Fisheries

POSITIONS

Assistant Professor (2005-present), Department of Zoology and Fisheries and Illinois Aquaculture Center, Southern Illinois University, Carbondale

- Postdoctoral Fellow (2003-2005), Department of Fishery and Wildlife Biology, Colorado State University, Fort Collins
- Teaching Assistant (January-May 1996 and January-May 1997), Graduate Research Assistant (1993-2001), and Postdoctoral Research Associate (2002-2003), Department of Fisheries and Wildlife Sciences, University of Missouri-Columbia

Research Assistant (summers 1987-1993), University of Texas Marine Science Institute, Port Aransas

SCIENTIFIC AND PROFESSIONAL ORGANIZATIONS

American Fisheries Society

- Bajer, P.G., R.S. Hayward, G.W. Whitledge, and R.D. Zweifel. 2004. Simultaneous identification and correction of systematic error in bioenergetics models: demonstration with a white crappie (*Pomoxis annularis*) model. Canadian Journal of Fisheries and Aquatic Sciences 61:2168-2182.
- Whitledge, G.W., R.S. Hayward, R.D. Zweifel, and C.F. Rabeni. 2003. Development and laboratory evaluation of a bioenergetics model for sub-adult and adult smallmouth bass. Transactions of the American Fisheries Society 132:316-325.
- Bajer, P.G., G.W. Whitledge, R.S. Hayward, and R.D. Zweifel. 2003. Laboratory evaluation of two bioenergetics models applied to yellow perch (*Perca flavescens*): identification of a major source of systematic error. Journal of Fish Biology 62:436-454.
- Wang, H.P., R.S. Hayward, G.W. Whitledge, and S.A. Fischer. 2003. Prey-size selection, maximum handling size, and consumption rates for redear sunfish *Lepomis microlophus* feeding on two gastropods common to aquaculture ponds. Journal of the World Aquaculture Society 34:379-386.
- Whitledge, G.W., R.S. Hayward, and C.F. Rabeni. 2002. Effects of temperature on specific daily metabolic demand and growth scope of sub-adult and adult smallmouth bass. Journal of Freshwater Ecology 17:353-361.
- Whitledge, G.W. and C.F. Rabeni. 2002. Maximum daily consumption and respiration rates at four temperatures for five species of crayfish from Missouri, U.S.A. (Decapoda, Orconectes spp.). Crustaceana 75:1119-1132.