University of Idaho
Aquaculture Research Institute
SPRING 2005

TEACHING

Although the Aquaculture Research Institute (ARI) is not an academic department, we provide educational and research opportunities to students at both the undergraduate and graduate level. Because of the diversity of aquaculture sciences, graduate degree programs with an aquaculture emphasis may be designed for post-baccalaureate students enrolled in fisheries, biology, animal sciences, agricultural economics and engineering programs.

ARI Director Ron Hardy is a Professor in the Department of Animal and Veterinary Sciences (College of Agricultural and Life Sciences) and an adjunct professor in the Department of Fish and Wildlife Resources (College of Natural Resources), Associate Director for campus programs Kenneth Cain is an Associate Professor in Fish and Wildlife Resources, and Associate Director for Hagerman Programs Matt Powell is an Assistant Professor in the Department of Animal and Veterinary Sciences. Each serves as major professor or committee members for graduate students enrolled in the two departments. They also offer special topic classes and independent study projects for graduate and undergraduate students.

Graduate Student Sean Wilson

Graduate student Sean Wilson holding a walleye at the ARI warmwater wet laboratory

Sean Wilson grew up with a love of the outdoors coupled with a deep interest in science that led him to pursue a career in natural resource management. Sean graduated from the University of Washington's School of Fisheries in 1993 and spent five years working as a seasonal technician for Washington Department of Fish and Wildlife. Sean left his last position with WDFW, working with a hatchery evaluations unit in Wenatchee, to pursue a M.S.degree under the direction of Dr. James Nagler in the Department of Biological Science. His research is aimed at producing sterile walleye.

The walleye (*Sander vitreus*) is a popular sport fish throughout North America. Since its introduction to the Pacific Northwest, the popularity of walleye fishing has grown in the region. A number of management agencies, including Montana Fish, Wildlife and Parks and Washington Department of Fish and Wildlife currently stock walleye to increase the angling opportunity for this species. However, due to their pivicerous nature, walleye introductions can lead to detrimental impacts on native fauna. To avoid problems, MFWP expressed interest in methods to produce sterile walleye for stocking purposes.

Currently, the only method that has been used to produce sterile walleye is triploidization. This technique uses a heat or pressure shock to cause recently fertilized eggs to retain an extra set of chromosomes. The fish that subsequently develop have three sets of chromosomes (triploid) rather than the normal two sets (diploid). Complications with meiosis in triploid fish typically cause them to be functionally infertile. However, triploidy in walleye is difficult to induce consistently. The only way to ensure that 100% triploids are stocked would be to individually screen each fish, a practice that would not be practical for a production facility.

(cont. on pg.2)

INSIDE

Aquaculture Research Institute

The ARI newsletter will be produced semi-annually and available online in Adobe Acrobat format through **www.webs.uidaho.edu/aquaculture/**. If you would like to be notified via email when the latest edition is available on our web page, please notify the editor at **aqua@uidaho.edu.**

We would be happy to in include appropriate contributions from those of you working in the field! Feedback and suggestions on how to improve this newsletter would also be appreciated.

This issue of the newsletter highlights various projects being conducted on the Moscow campus, the Hagerman Fish Experiment Station and includes various extension activities.

The ARI-2004 Annual Report is available on-line at www.webs.uidaho.edu/aquaculture/.

Dr. Ken Cain, Editor
Aquaculture Research Institute
PO Box 442260
University of Idaho
Moscow, ID 83844-2260

Email: aqua@uidaho.edu
Phone: (208) 885-5830
Fax: (208) 885-5968
Web site: www.webs.uidaho.edu/aquaculture/

(cont. from pg.1)

Because of this, MFWP funded research for Sean to develop a novel method of inducing sterility in walleye. After considering techniques that have been used in other fish species, Sean decided to concentrate his efforts on using heat applied after hatching at the fry stage to induce sterility. Although not widely known, it has been documented that germ cells can be destroyed at high rearing temperatures in the pejerry and tilapia. However, in order for Sean to reproduce the desired effect with walleye, proper temperature and timing of the treatment needed to be determined. He began by exposing groups of juvenile walleye to different temperatures, the highest (33°C) being just below lethal (34.1°C), for as long as the fish would tolerate them. This year, Sean has been exposing walleye to the highest temperature at different periods, starting with fish that had just undergone the larval/juvenile transition, and finishing with fish that had already began to differentiate sexually. Sean presented his results at Aquaculture America 2005 in New Orleans.

ONGOING RESEARCH PROJECTS (Hagerman)

Research programs at the Hagerman Fish Culture Experiment Station fall into three main areas: fish nutrition (alternate feed ingredients and low-pollution feeds); population genetics of anadromous and inland fish stocks; and selective breeding of rainbow trout using marker-assisted selection. The latter program includes sophisticated analysis of gene expression and metabolomics, which involves analysis of gene expression to identify metabolic pathways associated with differences in fish performance.

The fish nutrition program continues to focus on developing ways to increase the percentage of plant proteins in rainbow trout diets, thereby lowering the percentage of fish meal. Idaho grains, e.g. barley and oats, are the primary subjects of study, but other plant protein ingredients produced from soybeans, canola, flaxseed and others are also included. Soybean meal is widely available and less expensive than fish meal, but its use is limited by amino acid profile and the presence of constituents that lower feed intake, inhibit protein digestion, cause lesions in the gut, and increase water pollution by increasing the amount of indigestible material in the diet.

Research studies have been conducted over the past six months that address each of these problems. For example, the dietary requirement for essential amino acids of trout and salmon is similar to the amino acid profile of fish meal, whereas soybean protein is deficient in methionine, corn protein (gluten meal) is deficient in lysine, and wheat gluten meal comes up short in arginine. However, by blending the three protein sources together and supplementing with amino acids and components that stimulate feed intake, Hagerman scientists have been able to develop diets without fish meal that supported fish growth performance to about 90% of the growth of fish fed a conventional, fish meal-based-diet. What appears to prevent the experimental plant-based diet from performing as well as the fish meal-based diet is the presence of indigestible carbohydrates in the plant protein sources. In poultry and swine, researchers have demonstrated that adding supplemental enzymes facilitates the digestion of the plant carbohydrates, but studies at Hagerman showed that the enzyme supplements were not effective in rainbow trout. Enzyme activity increases with temperature, so the most likely reason that the enzyme supplements were not effective in

trout was related to their low body temperature compared to that of birds and mammals.

Today's fish feeds are mixtures of ingredients that are processed into pellets by a process called cooking-extrusion. Cooking-extrusion involves several steps that can be varied to modify the feed mixture, including pre-conditioning the mixture with steam for various lengths of time, varying the temperature in the barrel of the extruder itself, varying the amount of energy (or pressure) in the barrel of the extruder, and varying the length of time that the mixture is in the barrel of the extruder. In addition, the actual feed mixture can be adjusted to have more or less starch and more or less moisture. These factors affect pellet density so pellets float or sink in water, and also the degree of starch gelatinization that controls pellet hardness, water stability, and starch digestibility. As is always the case with processing, the goal is to find the right combination of variables to make pellets with the least possible energy and the highest possible nutritional value and stability. Often, these goals are mutually exclusive, so feed manufacturing is a compromise.

The ARI/Hagerman program has a cooperative agreement with the USDA Agricultural Research Service and the US Fish and Wildlife Service that provides access to a very well equipped feed manufacturing laboratory. Experimental feeds can be produced there using commercial-type equipment in which all variables can be changed and monitored with recording equipment. Using this laboratory, a series of studies are being conducted to determine the effects of feed manufacturing on the nutritional quality of plant protein-based feeds for trout to see if simple changes in production improves fish performance and nutrient retention, which means lower metabolic losses and less pollution from fish farms. Thus far, studies have shown that the digestibility of dry matter, lipid and energy from feeds can be improved by cooking-extrusion, but that protein and mineral digestibility is reduced under some conditions, most likely associated with the high temperatures used in extrusion. Further, studies with soybean meal-based diets showed that protein digestibility was not improved by subjecting the feed mixture to higher temperatures or pre-conditioning. We had hypothesized that such processing would lower the activity of trypsin inhibitors in commercial soybean meal and thus improve protein digestibility and fish performance. However, this was not the case, showing that conventional processing conditions used to make soybean meal were sufficient to inactivate this particular anti-nutrient. Studies also showed that the digestibility of non-starch polysaccharides was not improved by varying the feed processing conditions. Information was obtained that showed that high-temperature, short-time extrusion yielded the best product and is thus the best compromise for current trout feed formulations.

Research is continuing in the area of molecular biology, particularly in the area of the expression of genes associated with trout muscle cell proliferation at different stages of the life cycle of the fish. The ultimate aim of this work is to gain a basic understanding of muscle cell production and protein turnover in muscle, and to use this information to improve protein efficiency, that is, the retention of dietary protein as muscle in trout. Currently, about 40-45% of dietary protein intake is retained as trout tissue protein, up from about 25% a decade or so ago when feeds were not formulated or produced as well. Studies with salmonids and marine fish in Europe have demonstrated that high-performing families have lower rates of protein turnover in muscle, so our effort in molecular biology and gene expression is aimed at identifying the underlying mechanisms that are at work in such fish, and to exploit this information through genetic selection. This work is linked with nutrition in that high-performing fish need high-performing diets to allow full expression of their genetic potential.

Hagerman Construction Update

The UI and ARI have been working toward the goal of constructing a new building designed to meet the needs of the expanding program at Hagerman. The old building was built over decades by the US Fish & Wildlife Service, and housed a staff of less than ten people. Current staff at the Hagerman Station includes nine scientists, six laboratory technicians and several administrative staff, or slightly more than double the capacity of the old building. The old building had three analytical laboratories; the new building will have six laboratories. The new building will be about 14,000 ft^2 and be located where the old building, which will be demolished, once stood. About half of the building will be analytical laboratories, with the remaining space used for offices, meeting rooms, and a large conference room with video-conferencing capabilities. Demolition of the old building and construction of the new building is scheduled to begin in early spring, 2005, and the new building should be completed in about one year. There has been a huge effort made by the ARI and the university administration, not to mention our partners, the Columbia River Inter-tribal Fish Commission, to get this building designed, financed and constructed. We plan to have a large dedication ceremony and celebration when the building is completed in 2006.

ONGOING RESEARCH PROJECTS (Moscow)

Highlighted below are a few examples of current research projects that are being conducted by Dr. Cain's graduate students at the ARI in Moscow. Additional projects are ongoing at the ARI and are being directed by faculty in the Department of Biological Sciences and Department of Fish and Wildlife.

Tribal leaders want to apply Cain's findings to conserve and restore populations of burbot, a coldwater fish found in northern Idaho's Kootenai River.

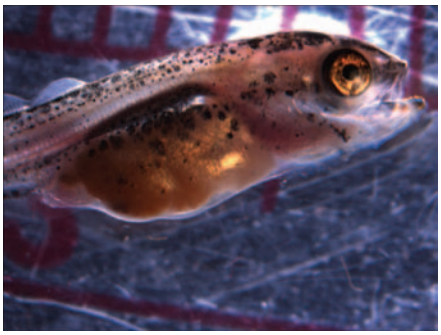
CONSERVATION AQUACULTURE
(Re-building small populations)

The story below first ran as a news release through the UI research office web page.

Burbot Aquaculture (Source: press release from UI research web page, www.uro.uidaho.edu/)/
Kootenai Tribe funds project to trace decline and help rebuild Idaho freshwater cod numbers
MOSCOW – The chips are down for burbot, a freshwater cod, in northern Idaho's Kootenai River.

Burbot protolarvae and developing embryo

University of Idaho aquaculture expert Ken Cain wants to learn how to raise burbot in captivity to help restore their fortunes through a project supported by the Kootenai Tribe of Idaho. Cain's team of researchers at the UI Aquaculture Research Institute and the Department of Fish and Wildlife received funding from the tribe to begin developing spawning and feeding techniques for the fish. Tribal leaders want to apply Cain's findings to conserve and restore populations of burbot, a coldwater fish found in northern Idaho's Kootenai River. The tribe helped lead efforts to restore the river's white sturgeon as well. Burbot once provided a valuable tribal, sport, and commercial fishery in the Kootenai River. In recent years biologists documented dramatic declines of the burbot population. Overfishing, habitat destruction, and altered water flows and temperatures following construction of Libby Dam in 1972 are implicated as factors. Resolution of the problem will take time, Cain said. "This research addresses many unknown questions," he said. "Nobody in the U.S. has cultured this species before and only limited information on the biology of this species is available." UI scientists are adapting techniques recently developed to culture marine cod. The Kootenai Tribe hopes to implement a conservation aquaculture program for burbot similar to the white sturgeon program begun in the early 1990s.



Burbot hatchling

In January of 2004, 20 adult burbot collected from Canada's Duncan River at the north end of Kootenay Lake, B.C., were transferred to UI tanks. The chilled 2-3^{O}C water is recirculated and mimics the burbot's natural spawning requirements. Lighting in the lab also mimics spawning-season day length. Ultrasound exams showed 13 were females, which were then divided into three treatment groups. Burbot in two groups were injected or implanted with small amounts of hormones. Male burbot play their part by fertilizing the eggs. Cain's team is also working to freeze sperm in case burbot numbers continue to dwindle and they are unable to catch adult

male fish in the future. Cain said all 13 females spawned during this first year, adding, "We have now met our first goal, which was to fertilize and incubate the eggs." The eggs are only about as thick as a grain of rice, and females can spawn up to three million of them. The team is testing several different types of incubators and will soon shift focus to rearing larval and juvenile fish.

Adult burbot from Duncan Reservoir, BC, Canada

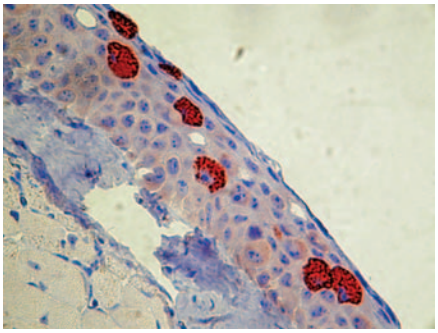
Our second goal is to begin feeding larval fish and show that we can get them to a size large enough for tagging, which would be important for a program that releases captive-reared fish into the wild," Cain said. The researchers hope to eventually write a technical manual about how to implement a burbot conservation program at tribal facilities in Bonners Ferry. "In addition to conservation aquaculture, the long-term implications for commercial foodfish aquaculture are pretty high," Cain said. "If we can successfully rear this freshwater cod, there is the potential to produce this species for food as well."

Preliminary efforts (described above), provided a strong foundation for us to build upon. Currently, burbot culture research is continuing. Nathan Jensen will focus on solving critical questions associated with spawning, egg incubation and larval rearing as part of his Master's program at UI.

FISH HEALTH RESEARCH

White Sturgeon Iridovirus (WSIV) in Kootenai River white sturgeon

Kootenai river white sturgeon are listed as endangered under the endangered species act and have not successfully reproduced in the Kootenai river in approximately 20 years.



Positive stain for WSIV infected cells in skin

Conservation aquaculture efforts to restore naturally producing populations to the Kootenai river are underway. Research collaborations between the Kootenai tribe and the ARI are focused on enhancing these efforts by addressing concerns over a viral disease, white sturgeon iridovirus (WSIV), that can affect these fish. Although WSIV is thought to be endemic in wild broodstock from this system, the impact this virus may have on natural recruitment in an environment highly altered from historic conditions is unknown. The conservation aquaculture program administered by the Kootenai Tribe of Idaho is proving successful, but it is generally agreed upon that all efforts should be made to minimize possible amplification of WSIV in the Kootenai system.

WSIV is believed to be passed vertically from adults to progeny and is periodically detected in juvenile fish following a stressful event. This indicates that progeny may harbor the virus throughout their life. Although the risk of this virus to native stocks is considered low, it would be highly desirable to eliminate vertical transmission of WSIV in the culture program. Surprisingly, work by John Drennan, a Ph.D. student of Dr. Ken Cain, has shown that vertical transmission may not occur in all cases. It appears that rearing sturgeon in a virus free water source could eliminate the disease in hatchery reared fish. This, along with iodine disinfection, would be beneficial to conservation and commercial white sturgeon hatchery programs.

Immunity in tilapia to Columnaris

Recent work conducted at the ARI by Leslie Grabowski, a former M.S. student of Dr. Ken Cain, has investigated how species such as tilapia respond to vaccination against Columnaris disease. This disease causes "Fin rot" in many cultured species along with valuable ornamental species. Vaccine development has met with limited success. In an effort to define immunity to this disease and determine if vaccination may be feasible in some species, studies in tilapia have been undertaken. It was found that tilapia are capable of mounting an immune response as indicated by the level of antibody present in their plasma (liquid portion of the blood). If fish are immunized by injection with a sonicated preparation of the bacteria, they are protected from disease. It was found that they develop antibodies following immunization, but it appears that antibodies may not be essential to provide protection. As part of this project, a number of tools were developed to help evaluate how tilapia respond to *Flavobacterium columnare*, the bacteria that causes causes Columnaris. One such tool was an assay (ELISA) that allows the detection of antibodies in the serum of fish either immunized or previously exposed to the bacteria. Another finding showed that bacterial strains varied in virulence and that exposing fish to a virulent strain by immersion resulted in significantly higher mortality then when fish were challenged by injection. The results of this project have been published in the Journal of Fish Diseases. (Grabowski, et. al., 2004: Vol 27, 573-581).

SCHEDULE OF EVENTS

May 9-13, 2005. World Aquaculture Annual Meeting. "International Peace and Development through Aquaculture." Bali, Indonesia. Website www.was.org.

The 46th Annual Meeting of the Western Fish Disease Workshop will be held on June 27 - 29, 2005 at the Double Tree Riverside in Boise, ID. Website: www.fisheries.org/fhs/

July 27-29, 2005, Annual Meeting of the Fish Health Section/ American Fisheries Society Minneapolis, MN. For more info contact: Joe Marcino (joe.marcino@dnr.state.mn.us

September 11-15, 2005. American Fisheries Society Annual Meeting. Anchorage, AK. "Creating A Mosiac: Connections Across Jurisdictions, Disciplines, and Cultures." Website: www.wdafs.org/anchorage2005/index.htm.

September 29-October 1, 2005, 2nd Coastal Cutthroat Trout Symposium, at Fort Worden State Park , near Port Townsend, Washington Web site: http://www.orafs.org/cutthroat.html.

February 13-16, 2006, Aquaculture America 2006, Las Vegas, Nevada USA. Website www.was.org.

UNIVERSITY OF IDAHO EXTENSION

News from Washington

EPA Rule Published in Federal Register

Yesterday, the EPA final rule for effluent limitations guidelines and new source performance standards for the concentrated aquatic animal production (aquaculture) point source category was published in the Federal Register. To review the final rule or print a copy go to EPA's website at: **www.epa.gov/guide/aquaculture.** This document is 40 pages and contains the preamble to the rule and the rule text. At this same website you can review and download a 1-page fact sheet that summarizes the rule, a press release and access to support and background materials. Other information relating to the proposed rule is also available.

EPA is preparing a BMP Guidance Document with their contractor, Tetra-Tech that should also be available soon. EPA is interested in organizing training workshops for permit writers and permitted facilities at a later time.

The Minor Use Minor Species Animal Health Act of 2004 (MUMS) was signed into law by President Bush on August 2, 2004.

While the MUMS legislation creates several new incentives, most prominent are the conditional drug approval and the index of legally marketed unapproved drugs (drug index). Each has certain advantages and limitations. Additional incentives include safeguards that have been created to protect existing New Animal Drug Approvals (NADA) from unwarranted scrutiny should a pharmaceutical company attempt to supplement the existing approval to address minor animal species or minor uses. The legislation also creates a new approach to facilitating drug development through a designated new animal drug classification system. Designated new animal drugs are eligible for grants for safety and efficacy testing, and for manufacturing process development. Designated drugs are also eligible for an exclusive seven year marketing time period. A new Office of Minor Use and Minor Animal Species Drug Development is created whose mission is to issue the grants, determine eligibility for listing on the drug index and for serving as a liaison amongst government agencies to improve opportunity for drug approvals.

The EPA final rule for effluent limitations guidelines and new source performance standards for the concentrated aquatic animal production (aquaculture) point source category was published in the Federal Register.

COLDWATER DISEASE WORKSHOP

The University of Idaho and College of Southern Idaho (CSI) held a one-day workshop on coldwater disease at the CSI fish hatchery on June 10, 2004. A University of Idaho/Washington State University Aquaculture Initiative Extension Products grant of $4,142 supported this workshop and will also be used for two additional workshops. Workshop instructors were Dr. Ken Cain, Ben LaFrentz, Terry Patterson (CSI), and Gary Fornshell.

Coldwater compression

Coldwater disease is caused by *Flavobacterium psychrophilum*, a gram-negative bacterium that produces an acute septicemic infection in salmonids and a few other fish species. *Flavobacterium psychrophilum,* considered one of the most important salmonid pathogens, can cause severe mortalities with subsequent economic loss among commercial and conservation hatcheries. In recent years the Idaho rainbow trout industry in south-central Idaho has experienced severe disease problems with this pathogen. Often major outbreaks are associated with co-infections of *F. psychrophilum* and infectious hematopoietic necrosis virus (IHNV).

The loss due to coldwater disease in Idaho is estimated to be 30% of total losses. Compounding the economic loss due to mortality is that up to 10% of survivors following an outbreak can have spinal deformities which decrease market value and processing efficiency. As such, coldwater disease

Typical growth of F. psychrophilum on tyes medica

is considered the most troublesome disease problem, and the industry places a high priority on research addressing potential treatments.

In addition to a basic introduction and general information about coldwater disease and its impacts on the Idaho trout industry; coldwater disease diagnostics, treatment and control options, and potential future control strategies were presented. Two hands-on activities, bacterial identification, and necropsy and bacterial plating, offered the participants practical and applied information and techniques to use for on-site presumptive diagnosis.

A pre- and post-test was administered to evaluate increase in knowledge due to the workshop. The class of 19 averaged 55% correct on the pre-test and 88% correct on the post-test, a 60% improvement.

Attendees said they gained ideas such as:

- "Trying everything else before trying to treat CWD, and especially if trying to prevent an outbreak"
- "Sample and examine sick fish. Keep complete records."
- "Look at fish more to detect CWD early instead of when when it has already gotten there and is killing lots of fish"
- "Better fish observation"

Two hands-on activities, bacterial identification, and necropsy and bacterial plating, offered the participants practical and applied information and techniques to use for on-site presumptive diagnosis.

AQUACULTURE 1
(Short Course Hagerman Fish Culture Experiment Station)

A six week introductory aquaculture short course for Native Americans entitled 'Aquaculture I' was held at the University of Idaho, Hagerman Fish Culture Experiment Station on June 15 through July 23, 2004. The course focused on aquaculture in the context of conservation fisheries where artificial propagation is integrated in fisheries management, and commercial propagation with captive populations. The primary course objectives were to provide students with an understanding of fish husbandry techniques, how to apply these techniques and where such practices were appropriate.

Subjects covered included: conservation, life history, anatomy, physiology, water quality, live hauling, carrying capacity, stock inventory, genetics, broodstock management, nutrition, and disease. The curriculum consisted of numerous lectures to introduce techniques and provide hands-on experience. Occasional field trips to commercial scale and private and government facilities were taken to observe how techniques could be applied. Job shadowing opportunities allowed students to practice husbandry skills in practical settings. To further reinforce these concepts, students also operated a mini-fish farm throughout the 6-week course where they were responsible for all management decisions and daily care of a population of fish. Five students from four different tribes participated in the short course. Shawn Wheeler and Arthur Broncheau, Nez Perce Tribe; Jerald Reed, Yakima Nation; Tyson Minthorn, Umatilla Tribe; and Preston Buckskin, Shoshone-Bannock Tribe. The course concluded with a luncheon banquet and ceremony during which the students were presented Certificates of Completion for their participation.

ARI Faculty and Staff lecturers participating included; Dr. Ernest Brannon-Project Director; Dr. Wendy Sealey-Aquaculture I class coordinator; Dr. Ronald Hardy, Dr. Madison Powell, Dr. Ken Cain, Dr. David Stone, and Mike Casten. Additional lecture support for the class was provided by USDA ARS staff; Dr. Rick Barrows, Dr. Gibson Gaylord, Dr. Ken Overturf and Dr. Katherine Johansen who are housed at Hagerman Fish Culture Experiment Station. Many additional Aquaculture producers and industry representatives participated in this course. Funding for the course was provided through an NSF EPScoR grant awarded in 2003 to Dr. Ernest Brannon to develop the American Indian Education in Science Program.

Proud graduates of the Aquaculture 1 course

* K I Y 0 1 0 *

University of Idaho
Aquaculture Research Insititute
PO Box 442260
Moscow, ID 83844-2260

Change Service Requested

Non-Profit Org
US Postage
PAID
University of Idaho