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## **Project Description**

Economic development in West Virginia is critical, particularly in rural communities where traditional economic activities (principally coal and timber) have declined. Rural economic development has been a principal objective of the Aquaculture Food and Marketing Development Project (AFMDP). Strategies where aquaculture can impact economic development in West Virginia and adjacent states are: (1) development of mine water resources for commercial production of food fish, and (2) use of farm raised fish in recreation. Conducting work in both of these areas will encourage development of a dual market for growers in the region strengthening both components of the industry.

Five years of funding have been approved for this project and the work is in varying stages of completion. Work presented in this proposal is designed to complement the work which has been completed and presently underway.

### **Objectives (FY 2003)**

#### Development of Honeycomb Fiber Reinforced Polymer Tanks for Field Assembly from Flat Panels

The objective of this task is to develop the basic component for a modular raceway system assembled from flat panels. Modules of raceway systems previously developed with this material (Dogwood Lake and Reymann Memorial Farm) presently are shipped as one box shaped piece.

Development and Verification of a Model to Describe Production of Rainbow Trout in Raceway Systems The objective is to develop a model which adequately predicts physical, chemical and biological outputs based on major physical, chemical, and biological inputs and to verify the model with data from working raceways systems (Dogwood Lake and Reymann Memorial Farm).

#### Metabolic Aspects of Growth and Efficiency of Nutrient Use in Different Strains of Rainbow Trout.

The objective is to quantify production and metabolic (in vivo and in vitro) variables as well as product quality variables in three "purebred" and three "crossbred" strains of rainbow trout.

Development of Value-added Food Based on Proteins and Lipids Recovered from Trout Processing By-Products. Objectives are the development of a pH-driven protein and lipid recovery from the by-products, and value-added food from recovered proteins and lipids.

Omega-3 Fortified Rainbow Trout The objective is to develop omega-3 fortified Rainbow Trout that would allow WV aquaculture industry to market trout food products as functional foods with health benefits superior to other trout or equal to Salmon.

Production of All-Female Triploid Brook Trout. The objective is to produce all-female diploid and triploid brook trout, as well as normal mixed-sex diploids and triploids, and to conduct performance tests under commercial scale conditions in three different production environments. These trials will provide a range of baseline production data describing the relative advantages of

monosex female and/or triploid production, and will help trout growers throughout the region determine if culture of brook trout is an economical option for their farm.

#### Catch-Related Standards of Quality and Demand Behavior for Fee Fishing Stocking Projects

Objectives are to determine standards of quality for fishing programs that stock hybrid striped bass, determine the added benefits of the hybrid bluegill stocking project, and determine the stability of demand behavior for a changing fee fishing market following the hybrid bluegill stocking project

### **Progress Report**

#### *Developing the Mine Water Resource*

Much of the research supported by the Aquaculture Food and Marketing Development Project has involved production and processing of cool water food fish. A long term objective is to develop modifications to the proven raceway system design .

Production of rainbow trout in a modular raceway utilizing water from an Acid Mine Discharge Treatment Plant. Research was completed to determine the technical feasibility of raising rainbow trout at Dogwood Lake. A four step raceway system utilizing water from the polishing pond of an Acid Mine Discharge treatment plant has been installed about 15 miles west of Morgantown. The system was stocked with three strains of trout from the National Center for Cool and Cold Water Aquaculture (NCCCWA) in October of 2002. Various parameters including water quality, flow rate, fish growth, stress, metal content of the flesh, flesh quality, etc. are being tested. A control group of fish are maintained at NCCCWA.

These are the first raceways ever to be manufactured from Honeycomb Fiber-Reinforced Polymer (HFRP) sandwich panels. At this site, comparative studies are being carried out utilizing the side-by-side HFRP raceway construction, which has shown to be effective both for research and potentially for aquaculture producers in West Virginia. Lessons learned at Dogwood Lake has led to a modified design for a four-tank raceway system which is being installed at the WVU Reymann Memorial Farms in Wardensville, WV. This project was funded by the State's Competitive Research Challenge Grant. These two sites with HFRP raceway systems have offered the opportunity to focus research on utilization of mine-waters (Dogwood Lake) in relation to a bench mark using spring water, where both locations constitute large-scale field laboratories that provide data which will translate to commercial applications.

Studies are underway to optimize the composite material architecture (fibers and resins) to provide best performance in strength and stiffness for minimum weight, which is directly related to cost. We are conducting a survey of existing systems to have a better understanding of costs, including life-cycle, and explore market opportunities for HFRP raceways.

Effluent Characterization Sites involved in the West Virginia aquaculture effluent study were visited at approximately six- to eight-week intervals. Field measurements of flow, pH, conductivity, temperature, dissolved oxygen, and turbidity were made of both influent and effluent waters. Grab samples of influent and effluent waters were collected and analyzed to

determine settleable solids, total suspended solids, 5-day biochemical oxygen demand (BOD5), nitrogen species (NH<sub>3</sub>, NH<sub>4</sub>, NO<sub>2</sub>, NO<sub>3</sub>), and total phosphorus concentrations. Mass loadings of water quality constituents were calculated using flow and concentration data and were compared with current regulatory requirements. Data at two of the sites are being used to obtain an NPDES permit.

Assessing Yield. Production was measured in representative units at two commercial farms growing rainbow trout. The tank based system captured ground water as it flowed from a coal mine. Average exchange rates were 0.19 and 0.32 exchanges/hour. Production at this farm was destined for a processing plant. Management focused on growing as many pounds of fish as possible. The raceway based system captured spring water as it flowed from the base of a mountain. Average exchange rates were between 6 and 10 exchanges/hour. Production from this farm was destined primarily to the recreational market. Labor was a limiting commodity. Management focused on growing adequate volume to fill orders while minimizing the required labor. Fish in the raceway system were harvested or split when density approached 4 lb/cubic foot, whereas fish in the tank system were harvested as density approached 2 lb/cubic foot. Average production was 107 and 5.8 lb/gpm/yr for tank and raceway systems, respectively. Production rates were higher in the tank based system. This is consistent with management objectives at the two facilities.

Health Survey Current results complemented previous results in demonstrating that certifiable pathogens (e.g., IPNV, *Aeromonas salmonicida* and *Myxobolus cerebralis*) that are endemic in the northeastern U.S. are detectable at low prevalence proportions in the WV salmonid industry. These results present an opportunity to the industry to quickly benefit from the implementation of a state-wide biosecurity and fish health management program.

Resource Economics: According to the WV Department of Agriculture, commercial food fish sales (mostly sales of trout to processors) across the state amounted to over \$800 thousand in 2001, a 35% increase from the previous year. An additional \$1.4 million of trout was stocked for conservation and recreation, making the aquaculture production sector in WV an over \$2 million annual activity. By virtue of its linkages with other sectors of the economy, we find through an input-output analysis that a \$1 million increase in annual aquaculture production increases total output in the state by an estimated \$2 million annually, generates an additional \$1 million in income and business taxes, and adds 55 jobs.

Preliminary results from a national household survey of aquaculture consumption preferences reveal that: (a) almost half of all households consume fish or seafood at least once a week; among the 9% of households who do not consume fish, the main reason given was taste; (b) two-thirds prefer eating ocean-caught fish (c) 61% prefer eating fish or seafood products at home rather than in a restaurant; (d) over half of the respondents reported reading the package label when purchasing fish, with over 60% of them indicating that information on such attributes as nutrition, production and processing techniques, location of production, and freshness was important; (e) 70% felt that fish and seafood should also have a country of origin label; (f) over two-thirds prefer buying their fresh fish "unprepared"; (g) half of all households would not consume genetically-modified (GM) fish; the likelihood of consuming GM fish would increase if

it were found to be safer (52%), cheaper (37%), tastier (52%), environmentally friendlier (54%). These results are being used to formulate marketing strategies and guide industry development.

Food Science: Water velocity and harvest method studies have been completed and research summaries and peer-reviewed publications are forthcoming. Preliminary work to refine Vitamin E assay and establish a baseline for time and temperature effects on storage stability are complete. Vitamin E feeding trials will begin in March, 2003. Studies of the effects of transport, water treatment (Aqui-S(r), CO<sub>2</sub>, ice, and control), and harvest method (CO<sub>2</sub> and Manual) on stress response and fillet attributes of Arctic Char are completed. Data analyses are underway and results will be summarized for publication in peer-reviewed journals. We will be assessing the effect of feed on fillet attributes of 4 groups of rainbow trout produced in acid mine drainage following treatment. We anticipate beginning these analyses by February, 2003. Activa(r), a cold-set technology that uses transglutaminase to increase protein-protein interactions, has been used for manufacture of restructured rainbow trout product. Because frozen storage is a necessary component of this process, the utility of this restructuring technology in the presence of cryoprotectants (trehalose, sucrose, sorbitol combinations) is under evaluation.

### ***Use of Farm Raised Fish in Recreation***

Customer Satisfaction and Appropriate Fee Structure for Fee Fishing Enterprises. Phase one data were collected at three West Virginia fee fishing businesses during the summer of 2002. Analysis suggests that rainbow trout (catch-and-keep) and channel catfish (tournament) fishing formats attract different customers who demand different recreational experiences. Rainbow trout anglers tend to travel as a family seeking fishing opportunities for their children. They were highly satisfied with catching and keeping a minimum of five fish per trip. The suggested minimum size of rainbow trout is two pounds. Channel catfish anglers travel in a wide range of groups, and they seek to catch large fish. The suggested acceptable channel catfish size is 6 pounds per fish. Demand for trout fishing experiences is less sensitive to changes in price (i.e., price inelastic). Demand for catfish fishing experiences is price elastic. Phase two data collection will begin in the summer of 2003 to examine hybrid bluegill fishing opportunities at two West Virginia study sites. Standards of quality for catch related standards will be determined for this underutilized species.

Hybrid Bluegill Production A study was designed to determine the effects of strain and diet on growth and efficiency of nutrient retention in hybrid bluegill. A diet containing 42% crude protein and 16% fat was determined to be the best of the five diets tested for both strains used in the study. Moreover, it was determined that the strain of bluegill known as the Georgia Giant grew faster than a commercial strain of hybrid bluegill. With respect to feed efficiency, the commercial hybrid bluegill and the Georgia Giant were not different. However, there were differences in the efficiency of retention for some amino acids. This suggests that different strains of fish use different amino acids with different efficiencies and that amino acid requirements probably differ within a species depending on the strain. That the 42/16 diet was the optimal diet for growth of bluegill is very important because this is not the diet the feed manufacturer typically suggests for bluegill production. A pond based study of production at stocking densities of five thousand, ten thousand, and twenty thousand fingerlings/acre will be harvested in late spring, 2003.

Resource Economics: In terms of recreational impacts, our results suggest that every 20,000 additional anglers -- or additional visits by existing anglers - will increase statewide output by \$2.5 million, income by \$1.5 million thousand, and add 59 jobs.

### ***Technology Transfer***

Output from the Aquaculture Food and Marketing Development project has resulted in numerous presentations and publications ranging from extension bulletins to refereed manuscripts. Approximately thirty refereed publications or presentations at scientific meetings have been created to date. Each January we host a state wide aquaculture meeting featuring latest information from this research project and speakers from the commercial aquaculture industry. Demonstrations and workshops include a bioassay of Arctic Char at a mine water source in McDowell County, development of an abandoned Acid Mine Drainage treatment plant as a fee fishing venue, and marketing arctic char. Numerous presentations on commercial aquaculture were conducted in response to requests from county agents and other groups.

### **Procedures**

#### ***Commercial Production and Development of the Mine Water Resource***

The scope of work includes the following sub-tasks:

1. Development of concept (guided by the floating system under development)
2. Design of connections
3. Design of prototype system
4. Testing and modeling of component joints
5. Production, field installation and evaluation of a full-size tank
6. Development of draft assembly procedures and standardized sizes

Based on the above proposed work, the commercial feasibility of the proposed system will be evaluated in the future by seeking large-scale implementation opportunities in collaboration with producers. The above six sub-tasks will be accomplished as follows:

1. **Development of Concept:** In collaboration with the manufacturer and based on our current work for the in-pond floating system, we will develop a detailed concept for the field-assembled system. The design considerations will be guided primarily by practicality and costs.
2. **Design of Connections:** The furniture industry has developed interesting connector systems to join components and overcome the inherent weaknesses of wood, particularly reconstituted products, primarily low shear and compressive strengths, which are also critical design properties for HFRP. Thus, we will review the information on existing wood connectors to guide our thinking and either produce original or modified connector designs. The manufacturing feasibility will be considered in collaboration with Jerry Plunkett from KSCI.

3. **Design of Prototype System:** Following the necessary iterations and discussions with the manufacturer and opinions from producers, we will define the design of a prototype system, with details about connections, stiffeners, and other characteristics.
4. **Testing and Modeling of Component Joints:** The most important task is to evaluate the functionality and performance of panel-to-panel connections. Thus, we will produce typical joints to test the proposed connector, including elastomeric sealants or gaskets to prevent water leakage, moment-curvature response in relation to monolithic factory-produced joints, and failure modes. Based on the experimental results, we will develop Finite Element models to characterize the elastic and failure response of the joints. Production, Field Installation and Evaluation of a Full-Size Tank: To demonstrate and evaluate the concept, we will produce one 30-foot unit of the same dimensions as the current tanks at Dogwood Lake and Reymann Memorial. This effort will provide us the opportunity to evaluate the efficiency of manufacturing, transportation and installation. We propose to use this new tank as a replacement of one of the existing tanks at either location, in order to evaluate its field performance.
5. **Development of Draft Assembly Procedures and Standardized Sizes:** The experience obtained from the above five tasks will permit us to develop procedures for the assembly of the system by producers, who will be assumed to have no or little technical expertise. Similarly, we will develop plans for standardization of sizes and multiple uses of components to permit adding on additional raceway cells to the system.

#### *Development and Verification of a Model to Describe Production of Rainbow Trout in Raceway Systems*

Performance in flow-through raceway aquaculture systems is predictable. The relationships have been quantitatively described by a variety of authors (Klontz, Westers, Soderberg). For example, it is possible to construct budgets for important rate-limiting water quality parameters such as oxygen and ammonia, which can further be related to the volume of water flowing, the size of the vessel, the number of water exchanges per unit time; and the number of fish in the system, their size, feed requirements, and growth.

It follows that such relationships can be used as the basis for a computer model, which could be used to provide stakeholders useful insight regarding the potential profitability of a proposed system, as part of site selection. Similarly, potential benefits of operational changes to existing systems could be determined and assessed against alternatives.

In the initial development of the model, consideration will be limited to Rainbow Trout; however, in subsequent refinements of this approach, work can be conducted to extrapolate to other species. Similar additional work can also be conducted to ascertain the applicability of the model to various genetic strains of rainbow trout.

The following will be described using the proposed model: (1) Physical system design, layout, and operation, (2) fish-related parameters, and (3) water quality.

##### 1. Physical design, layout, and operation

- a. Input parameters related to the design and performance of the raceway system include:

- i. Water flow rate
  - ii. Raceway dimensions (length, width, height, water depth, freeboard)
  - iii. Number of raceway units
  - iv. Number of water exchanges per hour
  - v. Average monthly water temperature
  - vi. Headloss available for gravity reaeration ? reaeration efficiency =  $f(\text{headloss})$
  - vii. Oxygen supplementation
- b. Based on the parameters listed above, in conjunction with data related to the fish (2.a & 2.b), water quality parameters (3.b) can be predicted.
2. Fish
- a. Input parameters related to fish (verified at stock, harvest, and at select discrete intervals) include:
    - i. Number
    - ii. Size
    - iii. Size variation
    - iv. Feed composition (fat, carbohydrates, etc.)
    - v. Amount of feed fed (input data collected at day & time intervals)
    - vi. Appetite
  - b. Based on the parameters listed in 3.a, the following can be determined:
    - i. Density
    - ii. Loading rate
    - iii. Total weight of fish
    - iv. Ammonia released
    - v. Oxygen consumed
    - vi. Solid waste generated
    - vii. Theoretical condition factor
3. Water quality
- a. Input parameters related to water quality (verified going into and out of the system) include:
    - i. Water volume
    - ii. Water temperature
    - iii. Water pH
    - iv. Dissolved oxygen concentration
    - v. CO<sub>2</sub>(aq) concentration
    - vi. NH<sub>3</sub>/total ammonia nitrogen (TAN)
    - vii. Air temperature
    - viii. Reaeration efficiency
    - ix. Barometric pressure
  - b. The following effluent water quality parameters will be predicted based on data (verified through routine sampling - presuming volume, temperature, pH, and barometric pressure ~ throughout the system):
    - i. Dissolved oxygen concentration
    - ii. NH<sub>3</sub>/total ammonia nitrogen (TAN)
    - iii. Total suspended solids concentration

The proposed simulation will be developed in Microsoft Excel(r) as opposed to other software in order to create a tool that is both mathematically rigorous, while still being accessible to a wide

range of users. The model will consist of a series of algebraic equations, some of which are subordinate to others, in addition to correlations from the literature (Soderberg) and data from field studies. Data collected through the current efforts at Dogwood Lake will be used in the preliminary stages of model development. Harvest of the existing fish is planned for May 2003, at which point, data including all inputs and outputs will be available. As part of this work, it is proposed that the facility at Dogwood Lake be stocked for an additional season with 8,000 rainbow trout for grow-out in order to develop replicate supporting data for model development and calibration. Efforts will be made to coordinate the development of a second season of grow-out data with other investigators studying feeding practices, nutrition, fish stress, etc. to maximize the information which can be "mined" from the system.

Initially, the model will be developed and calibrated for rainbow trout, as data from the operations at both Dogwood Lake and Reymann Memorial Farm can be utilized. However, in future work, efforts will be made to build on other data, such as those collected through yield verification studies, to test the applicability of the model to other species, different feeds, etc. as well as to test calibration against other similar systems, species, etc.

*Metabolic aspects of growth and efficiency of nutrient use in different strains of rainbow trout.*

We will quantify production and metabolic variables in three purebred and three crossbred groups of rainbow trout. The proposed strains are TL, UW, and Shasta. A fourth strain, HC, will be crossed with TL, UW, and Shasta strains to generate three crossbred strains. In the first experiment, fingerlings, approximately 5 g apiece will be housed in 4L aquaria at the National Center for Cool and Cold Water Aquaculture (NCCCWA). Fish will be fed for 2 months at which time they will weigh approximately 40 g. At about 2 months the second experiment will begin when 30 of the fish from experiment one (5 of each strain) will be moved and singly housed in 25 gallon aquaria where they will be grown to a terminal weight of 500+10g. The third experiment is a replicate of the second experiment except different variables will be measured.

The efficiency of nutrient retention will be assessed in the first and second experiments. To measure efficiency of nutrient retention, a comparative slaughter approach will be taken as the Blemings' laboratory has done previously with fish (Stinefelt et al., 2003) and chickens (Engels et al., 2003). When fish are approximately 5 g, a sample will be taken for analysis of crude protein, fat (ether extract), glycogen, and amino acids from hydrolyzed protein. After fish are harvested, their gastrointestinal contents will be removed so as not to confound the analysis. Crude protein will be determined by the Kjeldahl procedure (AOAC, 1990). Fat will be determined by extraction with petroleum ether by a standard protocol (AOAC, 1990). Glycogen will be determined spectrophotometrically as glucose equivalents as originally described by Hassid and Abraham (1957) and modified by Goodwin and colleagues (2002). Amino acids will be determined via high performance liquid chromatography on acid hydrolyzed samples after derivatization with phenylisothiocyanate (Bidlingmeyer et al., 1987) as previously performed in the Blemings' laboratory (Engels et al., 2003; Stinefelt et al., 2003). The 5 g fish will be used to estimate initial body composition. Fish will then be processed for the same measurements at approximately 40 and 500 g. The analysis of the 40 g fish serves the dual purpose of the terminal point in experiment one and to estimate initial body composition of the fish grown to 500 g. Feed

intake will be monitored and diet analyzed for the same components as the fish. By knowing the initial and final composition as well as the composition and quantity of feed eaten, the efficiency of nutrient retention will be calculated. From this data set, it will be determined whether differences in growth are the result of decreased feed intake or decreased efficiency of nutrient retention. Additionally, the limiting amino acid in the diet will be identified because it will be the amino acid used with the highest efficiency. It will be interesting to learn if the same amino acid is limiting in all strains. Then, depending on the size of the operation it may be cost effective to supplement feed with that amino acid for that strain..

The second experiment will also be used to monitor non-invasive measures of metabolism. Specifically, metabolic status would be assessed through measurement of O<sub>2</sub> consumption rates, total ammonia nitrogen excretion rate, feed conversion efficiency, growth rates, oxygen/feed ratios, ammonia/feed ratios. Oxygen consumption rates and nitrogen excretion rates will be determined as described by Yager and Summerfelt (1997). Short-term feed intake may be quantified by use of x-ray technology to increase the precision of the estimate. The x-ray technology can then be used to compare short-term intake to the estimate of the daily intake from the normal feeding routine. The non-invasive metabolic data provides a powerful complement to the nutrient retention data. It will be of great interest to learn if the more efficient strains consume more or less oxygen per unit gain and how that impacts ammonia levels. This kind of data set will be useful in modeling of an aquaculture production system.

The third experiment will monitor invasive measures of metabolism as well as product quality. Additionally, the feed intake and growth data can be used to increase the replication, and thus statistical power of the second experiment. Fish will be grown as described in the second experiment. At harvest, blood samples would be obtained for physiological analyses. Variables to be measured are pH, hematocrit, plasma cortisol, and total antioxidant capacity. The hydrogen ion concentration (pH) will be measured with a probe type pH meter. Hematocrit will be measured using a microhematocrit reader (IEC, Meedham Heights, Massachusetts) by the method of Houston (1990). Plasma cortisol will be measured with a commercially available RIA kit (Coat-a-count, Diagnostic Product Corp., LA, CA) and quantified in a  $\gamma$ -counter. Total antioxidants will be measured with a commercial kit (Bioxytech AOP-490 from Oxis Research - Portland OR) and quantified spectrophotometrically. These measures will provide a picture of the overall health of the different strains of fish and will be useful to increase our understanding of the metabolism of the different strains and possible metabolic basis of their different growth rates and efficiencies. After blood has been drawn, fish will be filleted and the products analyzed for texture and color attributes. Fillet yield will be calculated for each fish, and fillets will be graded according to the Code of Federal Register (50 CFR, Ch. 11, Part 260). Texture attributes and color will be measured as previously described (Jittinandana et al., 2002; Jittinandana et al., 2003). These measures of final product quality are central to the ultimate application of what is learned from the overall project. We will determine if there are significant correlations of the final product with any of the metabolic and growth variables previously measured.

*Development of Value-added Food Based on Proteins and Lipids Recovered From Trout Processing By-products*

This project has two phases: (A) development of pH-driven protein recovery from the by-products, and (B) development of value-added food from recovered proteins and lipids. Trout by-products will be provided at no charge by our industry collaborator, High Appalachian, LLC (Sophia, WV).

During phase A, the following will be determined: (1) solubility of myofibrillar and sarcoplasmic proteins (Choi and Park 2000) from pH 2 to 11 using the Lowry method (Lowry et al. 1951) coupled with determination of isoelectric point (pI) of these proteins using isoelectric focusing (IEF) (Hsieh et al. 1997), and (2) degradation of myofibrillar and sarcoplasmic proteins at pH 2 to 11 using SDS-PAGE (Jaczynski et al. 2002).

During phase B, the following will be conducted: (1) protein and lipid recovery from the trout processing by-products (see below "Protein and lipid recovery from the by-products") based on the protein solubility and the pI determined in phase A, (2) development of surimi seafood product from recovered proteins (see below "Development of surimi seafood product from recovered proteins"), (3) texture and color determination of the surimi seafood product using torsion shear stress and strain, and color  $L^* a^* b^*$ , respectively (Jaczynski and others 2002), and (4) fatty acid profile (FAP) of recovered fish lipids using methyl ester derivatives and gas chromatography (GC) (Green and Selivonchick 1990).

Protein and lipid recovery from the by-products. Trout processing by-products will be minced with distilled deionized water (1:9 w:v) using a PowerGen homogenizer (Hultin and Kelleher 1999). The pH of the mince will be adjusted to the pH resulting in the highest muscle protein solubilization (as determined in protein solubility experiments, phase A 1) using 1 N cold NaOH or HCl. The mince will be centrifuged at 10000 x g at 4°C for 20 min. The top fraction containing lipids will be collected and analyzed for FAP (phase B 4). A middle fraction containing solubilized muscle proteins will be collected and the pH of this solution will be adjusted to the pI of muscle proteins (as determined in IEF experiment, phase A 1) using 1 N cold NaOH or HCl followed by centrifugation at 10000 x g at 4°C for 20 min. The precipitated myofibrillar proteins will be collected (Hultin and Kelleher 1999). Sarcoplasmic proteins will be salt (NaCl) precipitated (ionic strength = 0.6, Lanier 2000) and collected following third centrifugation at 10000 x g at 4°C for 20 min (Hultin and Kelleher 1999).

Development of surimi seafood product from recovered proteins. Surimi seafood (see below "Literature review") will be prepared according to Jaczynski and Park (2002). The myofibrillar and sarcoplasmic proteins recovered from the by-products as described above will be chopped in a universal food processor (Model UMC5, Stephan Machinery Corp., Columbus, OH) at low speed for 1 min. Salt (2 %, w/w) will be added to solubilize the myofibrillar proteins and the resultant paste will be chopped at low speed for 0.5 min. Final moisture content will be adjusted to 78% by adding ice to the paste, followed by chopping at low speed for 1 min. Vacuum (0.5 bar) and chopping at high speed will be applied for the last 3 min. During chopping, the temperature will be kept below 5°C. The paste will then be stuffed into torsion gel molds and cooked at 90°C for 15 min, resulting in hourglass-shaped surimi seafood gels (length = 2.9 cm, end diameter = 1.9 cm, and minimum diameter = 1.0 cm). The texture and color of the surimi seafood gels will be determined using torsion shear stress and strain, and color  $L^* a^* b^*$ , respectively (Jaczynski and others 2002).

### *Omega-3 Fortified Rainbow Trout*

Trout will be fed nine different diets for four months before slaughter. A control diet will be a grower-standard sinking diet (protein 42%, fat 16%, Zeigler Bros Inc., Gardners, PA). Diet 1 and 2 will be formulated by Ziegler and will use 15 and 30% of flaxseed oil, respectively, to replace fat in the control diet. Each diet will be supplemented with vit. E (dl-(-tocopheryl acetate) at 0, 300, and 1500 mg/kg feed. Each diet will be assigned to a separate fish tank. The experimental design will be randomized complete block. Facility in either Wardensville or Dogwood will be used.

The trout will be fed twice a day. Each tank will receive the same amount of feed, equal to the amount of least hungry tank would consume at feeding. If vit. E deficiency results in reduced appetite, the tanks with un-reduced appetite will be allowed to consume more. Following slaughter, trout fillets and stored at 4 and 10°C. To meet objectives the following will be conducted:

Objective (1). Fatty acid profile (FAP) will be determined using methyl esters and gas chromatography (GC) (Green and Selivonchick 1990). Fillets and feed will be analyzed for FAP at 0, 1, 2, 3, and 4 month of feeding. The FAP will be analyzed immediately following the slaughter, then once and three times a week for fillets stored at 4 and 10°C, respectively. Soxhlet extraction will be performed at the same time intervals as GC to determine lipid content in fillets and feed.

Objective (2) and (3). Lipid oxidation and the effect of antioxidants on inhibition of lipid oxidation will be determined using 2-thiobarbituric acid (TBA) assay (Yu and Sinnhuber 1957). The TBA will be performed immediately following the slaughter, then once and three times a week for fillets stored at 4 and 10°C, respectively. Concentration of vit. E (Liu and others 1996) in trout fillets will not be determined since it is currently investigated.

The fillets and feed will be homogenized prior to the GC and TBA analysis.

### *Use of Farm Raised fish in Recreation*

#### Production of All-Female (Triploid) Brook Trout

Eggs from female brook trout will be obtained in the fall of 2003 from Pisgah Forest State Fish Hatchery, Brevard, NC, and transported to facilities of the Mountain Aquaculture Research Center (MARC), Western Carolina University (WCU), Cullowhee, NC. There, the eggs will be pooled, and split in two portions. The first portion will be sub-lotted into approximately 20 equal groups. Each sub-lot will then be fertilized with the milt from one of approximately 20 3-year old sex-reversed XX "males" for the production of all-female fish. These broodstock are currently being reared at the Lonesome Valley Aquaculture Research Station, MARC; they were derived from the same Pisgah Forest hatchery brook trout strain. The other portion of the pooled eggs will be sub-lotted and fertilized with milt from approximately 20 normal males, also obtained from Pisgah Forest State Fish Hatchery, for the production of mixed-sex fish. The

fertilized eggs within groups, will immediately be repooled, and each will be divided into two sub-groups. One sub-group will be left untreated (diploids), and the other sub-group will be subjected to a thermal shock to induce triploidy: immersion in 28°C water for 10 min beginning 10 min post-fertilization (Galbreath and Samples 2000). Eggs will be incubated in separate trays within common vertical tray incubators. Upon initiation of feeding, the eggs will be transferred to separate tanks within a common recirculated-water rearing system. A sample of 40 fry per triploid group and 20 fry per diploid group will be sacrificed and examined by flow cytometry at WCU to confirm percent success of the triploidization treatment (Galbreath and Samples 2000).

At 2-3 months post-initiation of feeding, a portion of the fish from each of the four groups will be transported to the West Virginia University aquaculture demonstration facility at the Reymann Memorial Farm, Wardensville, WV for rearing for an additional 3 months. Rearing of the remaining fish in NC will continue at the Lonesome Valley Station. At each of these sites, parallel growth trials will be performed according to the design described below.

At 5-6 months post-initiation of feeding, fingerlings of each of the four types (all-female diploids and triploids, mixed-sex diploids and triploids) will be counted and weighed and stocked into a common tank(s) or raceway(s) at commercial scale densities. The fish will be fed with commercial trout diets at recommended rates for an additional 18-24 months. Growth will be monitored during rearing, by monthly sub-sampling, and feeding rates readjusted accordingly. Following harvest, data for growth, feed conversion, percent maturation, dress-out percentage, etc. will be measured and compared by analysis of variance for effects of ploidy and sex-ratio.

Each of the different facilities where the fish will be reared has its particular water source (spring, well, or surface water), water chemistry profile, and rearing system (circular tanks or concrete raceway, indoor or outdoor, etc.). As such, comparative data will be obtained over a range of environments, and will demonstrate the extent to which observations on relative performance of the brook trout might be generalized, or conversely, demonstrate an interaction relative to rearing environment that which should be taken into consideration. Additionally, enterprise budgets will be projected using the data, so as to provide growers with a means to estimate whether production of brook trout (all-female versus mixed-sex, and/or diploid versus triploid) might prove a viable option for their farm and their markets.

In addition to the rearing trials being conducted over this two-year period, work will continue at WCU to optimize the brook trout sex-reversal protocols. These experiments involve testing of MDHT immersion treatments of the brook trout fry, with or without additional application of the steroid via the feed. Various combinations of immersion dosage, duration and timing are being tested, in combination with different dosage and duration regimes for treatment of the steroid via the feed. The objective is to identify a protocol which maximizes production of effectively sex-reversed "XX" males - preferably ones from which the milt can be manually stripped, so that the broodfish need not be sacrificed and might be reused for multiple spawning seasons.

### Catch-Related Standards of Quality and Demand Behavior for Fee Fishing Stocking Projects

Determine standards of quality for fishing programs that stock hybrid striped bass.  
Catch-related factors such as catch rates and size of fish caught are important indicators of

quality for some recreational fishery settings and experiences. Logar, Ponzurick, and Semmens (2001) have determined that type and size of fish are the two most important features that a West Virginia recreational fee fishing operation could offer out-of-state and in-state anglers. Pierskalla and Schuett (2003) are completing a study that determines the customers' minimum acceptable catch size and rate for trout, catfish, and hybrid bluegill. Preliminary results suggest that anglers' motives and standards of quality can vary by the species sought (trout or catfish), fee structure (catching and/or keeping fish), and program format (competitive fishing tournaments or non-competitive fishing). Little is known about how these catch-related standards of quality vary for another popular, and perhaps underutilized recreational species-hybrid striped bass.

Determine the added benefits of the hybrid bluegill stocking project.

The number of recreational trips is a common measurement used to assess the value for natural resources, and it was used in a 2002 study conducted by Pierskalla and Schuett to measure the value of benefits provided by various West Virginia pay ponds. However, the added benefits (increased recreational trips) attributed to the hybrid bluegill stocking project over time requires additional research. This information can help pay fishing operators determine if stocking hybrid bluegill is a desirable project for their business as the market develops.

Determine the stability of demand behavior for a changing fee fishing market following the stocking of hybrid bluegill.

The profile of customers of a West Virginia pay pond has been developed in recent research. These customers participated in a study one year prior to the initiation of the hybrid bluegill stocking project. Information about hybrid bluegill collected during this pre-stocking time frame is based on potential customers' expectations regarding their values and preferences for this species. We propose re-contacting these customers one year after the stocking project to determine the association between expected values (pre-stocking) and actual values (post-stocking), and the stability of preferences for hybrid bluegill. Understanding these changes will help fishing operators market and manage new recreational opportunities created by additional stocking. A two-year study of fee fishing customers is proposed.

## **Year 1**

The first phase of this study will build on the work conducted in a previous aquaculture research project. The previous study examined customers of three West Virginia pay ponds. Mail-back questionnaires were developed and distributed to customers in 2002. Customers were asked to report their demographic information, level of satisfaction, and motivation for fishing. In addition, customer willingness to pay for hybrid bluegill fishing opportunities was determined. During the summer of 2003, a hybrid bluegill stocking project will be implemented at two of the three study sites to determine catch-related standards of quality. However, no additional research is proposed after 2003 to determine the added benefits provided by the stocking project. This study proposes to collect data in 2004 and compare them to 2002 data.

### *Study instrument*

We will mail questionnaires to customers one year after the initiation of the 2003 hybrid bluegill stocking project to determine the added benefits provided by the stocking project and the stability of demand behavior for this new market. The same questionnaire used in the 2002 study will be used again in this study to make comparisons between pre and post stocking. Several additional questions will be sent along with the questionnaire to determine the number of visits

each study participant made to the study site since the initiation of the hybrid bluegill stocking project. By comparing pre and post stocking responses (such as visitor motives, visitor satisfaction, visitor use level, and willingness to pay), we will be able to determine the changes in demand and demand behavior that results from implementing a hybrid bluegill stocking project.

#### *Sampling and analysis*

Questionnaires will be mailed to the same customers that participated in the 2002 study. (Only those 2002 study respondents that were contacted at study sites participating in the hybrid bluegill stocking project will be asked to participate). The responses from the 2004 mailing will be entered into a statistical database and analyzed. A comparison between sample mean scores for several demand related variables (e.g., number of visits made in last 12 months, willingness to pay for hybrid bluegill opportunities, and visitor motives) will be conducted.

## **Year 2**

Phase two of this study will determine catch-related standards of quality (e.g., minimum acceptable catch rate and fish size) for a fish species (hybrid striped bass) currently underutilized in West Virginia. Standards of quality will be compared for two types of fee fishing formats (catch-and-keep and catch-and-release). The relationship between hybrid striped bass catch rates and stock densities will be determined.

#### Study instruments

A short on-site interview and mail-back questionnaire will be developed. The on-site interview will include questions about customer and trip characteristics, and it can be used to help determine non-response biases. The questionnaire will measure each study participant's beliefs about minimum acceptable fish size and catch rates, and his/her level of satisfaction with hybrid striped bass fishing opportunities.

#### Sampling and analysis

On-site interviews will be conducted and questionnaires distributed to fee fishing customers during the summer of the second year. Hybrid striped bass will be stocked at various sizes at a pay pond every other week for six weeks in a catch-and-release managerial setting. This stocking program will allow fish population densities to gradually increase over time. A catch-and-keep fishing format will be implemented during the second half of the study until the fish population declines to minimal levels. Customers will be asked to report their beliefs regarding the acceptability of fish size and catch rates under both managerial settings.

Data will be entered into a statistical database. The mean fish size and catch rate and their associated standard of quality will be determined for both fee fishing formats. The correlation between catch rates and stocking densities also will be determined for both fee fishing formats.

#### ***Technology Transfer***

The need for technology transfer is increasing. Requests for literature and information have doubled since the inception of the project. There is greater demand for site visits to assess opportunities, and assist individuals growing or utilizing farm raised fish. Administrative duties increasingly require the attention of the Extension Specialist in his role as Principal Investigator

making it increasingly difficult to meet all requests and reach out to new stakeholder groups. We responded to this need by supporting two positions, each oriented to one area of project focus.

Development of the Mine Water Resource. Mine water has emerged as the most important source of water for commercial production of salmonids in West Virginia. Each site must be evaluated on its unique topography, water quality, water volume, etc. This proposal will support Dan Miller, Sr. Research Coordinator, in extension of this project area. We will continue to work with coal companies, individuals, and economic development agencies the step by step process of site assessment and education required for development of the mine water resource. We will also coordinate with processing plants and others to assist with determination production capacity and appropriate facility design once a site has been chosen for development. As part of this effort, we will continue to assess yield at two commercial trout production facilities in southern West Virginia. This will be a continuation of the yield verification project begun in FY 99 and will be coordinated by Julie Delabbio at Bluefield State College. Previous work with West Virginia State College and Bluefield State College collected data for production of foodfish at one facility growing foodfish utilizing mine water and another facility growing fish for the recreational market utilizing spring water. We will subcontract with Julie Delabbio at Bluefield State College for one year to collect data for fingerling production at the same two locations. This is essential for a complete data set regarding production efficiency from egg to market size trout.

Aquaculture and Recreation. Most fish farmers in West Virginia sell their fish in the recreational market. Approximately 300,000 lb is sold to fee fishing businesses around the state. In addition, live fish are sold to fishing clubs, large companies, housing associations, and private individuals for recreational use in both private and public waters. Building upon the data developed by the project, it is timely that this information be combined with the experiences of fish farmers serving the recreational industry and extend it to a wide variety of recreational stakeholders. Such stakeholders would include resorts, attractions, fishing clubs, communities developing strategies to enhance tourism, and the general public. There is a need to develop materials which describe how farm raised fish can be utilized in recreational activities. There is opportunity to develop partnerships between segments of the tourism industry, fish farmers, fisheries managers, and the resource base to respond to the recreational opportunities described by marketing research. As part of this effort, we propose to assess safety at fee fishing locations. Fee fishing operations must secure insurance coverage to conduct business. Both in-state and out-of-state visitors surveyed in recent studies by the College of Business and Economics ranked site safety as an important consideration in choosing a fee fishing facility. Patricia Miller will assess facilities and current practices regarding safety at fee fishing businesses. Following a review of facilities, we will work with the West Virginia Farm Bureau to develop design, best practices, and management recommendations. Safety training workshops and a self assessment tool will also be developed.

We will continue current technology transfer activities. The project web site featuring project output will be maintained (<http://www.caf.wvu.edu/afmdp/>) and the annual Aquaculture Forum targeting WV aquaculture producers will be held in January. We will continue to maintain the Extension web site for aquaculture (<http://www.wvu.edu/~agexten/aquaculture/index.htm>) and continue developing the new quarterly newsletter "Fishtales". Faculty will continue to make

presentations at regional and national meetings and develop publications for peer review. Extension efforts will include responding to specific inquiries, site assessments, demonstrations, and presentations at fish, recreation, and coal related meetings. We will disseminate information generated by this project to the aquaculture industry in Appalachia, to government agencies with aquaculture-related responsibilities, and to the general public. The investigators will continue to collaborate with the Freshwater Institute's Aquaculture Program, the National Center for Cool and Cold Water Aquaculture, the Canaan Valley Institute, the West Virginia Department of Agriculture, the West Virginia Aquaculture Association, and other organizations to deliver research findings in the most user friendly manner possible.

## **Justification**

Aquaculture has significant potential to improve economic conditions in West Virginia and surrounding areas in Appalachia. The supply-demand balance for seafood is, and promises to remain, favorable from the producer's viewpoint. Additionally, West Virginia and Central Appalachia possess water resources which confer meaningful competitive advantages in the practice of cool and cold water aquaculture by virtue of their being abundant, near constant temperature and gravity fed. It also is important that many of these water resources occur in areas most in need of economic development. Although growing, the aquaculture industry in Central Appalachia has limited infrastructure and is not well organized. There are a variety of constraints to industry growth and sustainability. Those targeted for remediation by this project include:

1. Need for information, methods, and systems appropriate for commercial production of foodfish appropriate to development of the mine water resource;
2. Absence of a strategic marketing plans for fee fishing operations and assessment of alternative species.
3. Need to increase product diversity appropriate for niche markets for edible aquaculture products.
4. Need for technical assistance in association with a working production facility to demonstrate appropriate application of current technology.

## **Literature Review**

### Development of HFRP tanks for Field Assembly from Flat Panels

Fiber-reinforced plastic (FRP) cellular panels have been increasingly used in civil infrastructure, particularly in the new and rehabilitative construction of highway bridge decks. This recent fast growing interest in composite materials provides an excellent opportunity for the development and implementation of honeycomb structures. The concept of a light-weight yet heavy-duty honeycomb structure with a core extending vertically from the face sheets was introduced by Plunkett (1997). Since then, design and optimization studies (Davalos et al., 2001), as well as material response studies (Wang et al., 2002, Chen et al., 2002), have been performed. Furthermore, studies regarding the use of honeycomb fiber-reinforced materials (HFRP) for aquaculture purposes, particularly modular raceway units (Vantaram et al., 2002), have been underway.

## Development and Verification of a Model to Describe Production of Rainbow Trout in Raceway Systems.

Performance in flow-through raceway aquaculture systems is predictable. The relationships have been quantitatively described by a variety of authors (Klontz 1991, Westers 1984, Soderberg 1995). For example, it is possible to construct budgets for important rate-limiting water quality parameters such as oxygen and ammonia, which can further be related to the volume of water flowing, the size of the vessel, the number of water exchanges per unit time; and the number of fish in the system, their size, feed requirements, and growth. It follows that such relationships can be used as the basis for a computer model, which could be used to provide stakeholders useful insight regarding the potential profitability of a proposed system, as part of site selection. Similarly, potential benefits of operational changes to existing systems could be determined and assessed against alternatives.

## Metabolic aspects of growth and efficiency of nutrient use in different strains of rainbow trout

From an economic perspective, rainbow trout are the primary species of interest to West Virginia. Two of the major considerations in any aquaculture enterprise are: what fish to grow and what to feed them. There are relatively few options available for fish farmers with respect to what strain will be grown as there are few commercial suppliers. Strain improvement is an important goal from a research perspective and there are only a few facilities dedicated to this task, among them is the National Center for Cool and Cold Water Aquaculture (NCCCWA). Work done by NCCCWA and WVU at the Dogwood Lake facility has shown differences in the growth rate of rainbow trout strains reared in treated mine waters. Certainly, that different strains within a species would differ in growth and other metabolic variables is not surprising. Aspects of metabolism that may contribute to the genetics of the growth differences between different strains include, but are not limited to, stress response, antioxidant status, feed intake, and efficiency of nutrient retention.

Strain differences are observed under the appropriate environmental conditions as there are interactions between strains and environments. For example, when comparing two strains of rainbow trout, the increased growth rate of one strain was not apparent unless the fish were fed ad libitum (Valente et al., 2001). Growth differences of at least two strains have been attributed to differences in the ability to recruit muscle fibers and diameter of white muscle fibers (Valente et al., 1999). The differences in percent of the different fiber types making up the fillet could impact other compositional variables as well as texture and organoleptic properties. However, a literature search only returned one report where different strains of rainbow trout were evaluated for these measures. In this study, strains varied in carcass composition but not in organoleptic qualities (Smith et al., 1988).

The stress response in fish is associated with increases in the circulating concentration of the hormone cortisol. Even within the species of rainbow trout, strains of fish that have the highest cortisol response to a stressor have decreased survivability (Fevolden et al., 2003). The high cortisol response to a stressor is also correlated with decreased performance in the different strains of rainbow trout (Fevolden et al., 2002) and the cortisol response was shown to have a very high heritability ( $h^2$ ). Some have suggested that the measure of greatest interest is not

necessarily the peak cortisol concentration but, how quickly does the cortisol return to baseline values (Weil et al., 2001). Given the heritability of the cortisol response and its effect on growth, more data on the cortisol responses of different strains of rainbow trout is warranted because the preponderance of evidence indicates "crossbreeding" of rainbow trout strains is advantageous from the viewpoint of hybrid vigor (Guo et al., 1990, Wangila et al., 1996)

With respect to antioxidant status, data in other species indicate antioxidant status can have a profound effect on overall health status and production efficiencies. In a study with chickens, addition of the uric acid precursor inosine to the diet increased plasma uric acid levels. The increased plasma uric acid levels was associated with a decline in whole body oxidative stress as assessed by leukocyte oxidative activity (Simoyi et al., 2002). This effect is not unique to chickens as similar results have been observed in rats treated with inosine and the uricase inhibitor oxonic acid where increased antioxidant status was associated with an increased growth rate (Kain et al., 2002). Investigations into the role of uric acid in fish are few (De La Higuera, 1981, Ciereszko et al., 1999, Rehulka 2000) but are consistent with uric acid serving as an antioxidant in fish.

Not surprisingly, feed intake and efficiency of nutrient retention are critical factors effecting growth and production efficiencies. Part of the differences in growth may be a function of differences in the ability to digest protein (Austreng and Refstie 1979, Valente et al., 1998). But, beyond differences of protein digestibility is the efficiency with which dietary amino acids are retained in protein. Fish have a high crude protein requirement compared to traditional livestock species and this high requirement is probably a function of their relatively efficient growth, and a low tolerance for diets high in carbohydrate. The low tolerance for carbohydrate suggests a heavy reliance on amino acid carbon skeletons for gluconeogenesis. The resultant  $\alpha$ -amino nitrogen released in the form of ammonia is an important environmental concern and not surprisingly the ratio of dietary protein/energy can impact nitrogen excretion in rainbow trout (Medale et al., 1995). The problem of ammonia excretion from aquaculture facilities is likely to be exacerbated as the industry begins to move to diets containing higher levels of cereal grain protein (Encarnacao and Bureau) which contain an amino acid pattern less optimal for growth of salmonid species.

Although few studies have addressed the retention of individual amino acids, considerable effort is going into increasing nitrogen retention while decreasing nitrogen outflow particularly in the form of ammonia (NH<sub>3</sub>). Diets that contain sufficient energy in the form of lipid decrease nitrogen waste (Green et al., 2002). Additionally, there is a benefit of adding phytase to diets high in soybean meal with respect to protein digestibility (Sugiura et al., 2001). Experiments with hybrid bluegill have already demonstrated interactions of strain with diet (Stinefelt et al., 2003) and baseline data in different rainbow trout strains is critical to development of optimal strains for foodfish production.

#### Development of Value-added Food Based on Proteins and Lipids Recovered from Trout Processing By-products

Surimi and surimi seafood. Surimi is the main ingredient used in manufacturing surimi seafood products (i.e., imitation crabmeat). Surimi contains fish muscle proteins (primarily salt-soluble

myofibrillar proteins) obtained by mincing various fish meat fractions followed by sequential water washings of this mince (Lanier 2000). Sarcoplasmic proteins are water-soluble, and therefore, are lost during the washings. However, several studies indicated that sarcoplasmic proteins strengthen gelation of myofibrillar proteins (Marioka and Shimizu 1990, Okazaki and others 1986). Therefore, the loss of the sarcoplasmic proteins is often considered a drawback of surimi production using the water washings. The pH-driven recovery will potentially recover both the sarcoplasmic and the myofibrillar proteins. During the manufacture of surimi seafood products, myofibrillar proteins primarily are solubilized using salt (NaCl), resulting in a paste. The paste is then heated, resulting in development of texture due to heat-induced protein gelation. Therefore, the gel-forming ability of recovered proteins is critical. Degraded proteins lose the gel-forming ability.

The pH-driven protein recovery. Protein solubility is lowest at a protein's isoelectric point (pI) (Srinivasan 1996). The pI is the pH at which the protein molecule's net electric charge is zero. As pH diverges from the pI, the increased protein-protein electrostatic repulsion facilitates protein-water interaction, resulting in protein solubilization (Srinivasan 1996). Conversely, as pH approaches pI, the decreased protein-protein electrostatic repulsion decreases protein-water interaction, allowing for protein-protein hydrophobic interactions, thereby leading to protein precipitation (Srinivasan 1996). Precipitated proteins can be separated from lipids, debris (bones, skin, etc.), and water by centrifugation. This pH-driven solubilization-precipitation cycle can be used to isolate proteins and lipids from the trout processing by-products.

Gel-forming ability. To be useful in making surimi seafood products, muscle proteins recovered from the by-products must retain their gel-forming ability (Lanier 2000). Heat-induced gelation is a result of protein denaturation, leading to inter- and intramolecular covalent and non-covalent interactions (Lee and Lanier 1995). Solubilized proteins undergo denaturation followed by an ordered aggregation to form a gel network (Srinivasan 1996). Therefore, solubility and denaturation are the critical prerequisites for heat-induced protein gelation.

Fish oil and omega-3 fatty acids. Fish oil contains high concentrations of omega-3 polyunsaturated fatty acids ( $n-3$  PUFA). In 2002, the Food and Drug Administration (FDA) approved a health claim for the  $n-3$  PUFA. According to the FDA, "consumption of  $n-3$  PUFA may reduce the risk of coronary heart disease". This approval created a niche market in the functional foods and dietary supplements arenas for fish oils. The concentrations of the  $n-3$  PUFA in a fatty acid profile (FAP) of fish oil vary depending on species, diet and processing. Therefore, determination of the FAP is critical.

Current research in the field. Hultin and Kelleher (1999) applied the pH-driven protein recovery to mackerel and demonstrated that solubilization of mackerel muscle proteins at pH 2-3, followed by precipitation at pH 5.5 and centrifugation, resulted in high recovery of muscle proteins and lipids. Choi and Park (2000) applied the pH-driven protein recovery to Pacific whiting (similar pH as Hultin and Kelleher), which increased muscle protein recovery by 80% as compared to conventional surimi production (sequential washings with water). However, the poor texture of the surimi seafood product prepared from Pacific whiting proteins recovered at acidic pH was attributed to activation of proteases at acidic pH. Yongsawatdigul and Park (2001)

further investigated protein recovery of fish muscle using an alkaline pH, which resulted in improved texture of the surimi seafood product.

There are no reports on application of the pH-driven protein recovery to the farm raised fish or farm raised fish processing by-products, including trout.

### Omega-3 Fortified Rainbow Trout

Castell and others (1972) showed that dietary linoleic (18:2(-6) and linolenic (18:3(-3) fatty acids increased the concentration of these fatty acid and that of the EPA and DHA in the phospholipids and neutral lipids in the muscle tissue of the fingerlings. Green and Selivonchick (1990) suggested that trout maintained concentration of the EPA (20:5(-3) and DHA (22:6(-3) at physiologically optimum level. The fish deposited 10-12% of lipids from the diet as the EPA and DHA despite the availability of the dietary precursor (linolenic, 18:3(-3) to further elongate and desaturate the precursor to the EPA (20:5(-3) and DHA (22:6(-3) (Green and Selivonchick 1990). Bharadwaj and others (2003) fed trout fourteen different diets, including a diet supplemented with flaxseed oil. They concluded that the flaxseed oil increased the (-3/(-6 ratio and the concentration of the (-3 PUFA in the muscle tissue. While these authors tested fourteen diets and fatty acid profile in the muscle and liver, they did not vary the concentrations of flaxseed oil in the diet to determine the relationship between increased concentration of flaxseed oil in the diet and the increase of (-3 PUFA in the muscle. The authors did not determine the effects of the increased (-3 PUFA in the muscle on lipid oxidation or human health.

The desaturation and elongation of linolenic fatty acid (18:3n-3) was significantly greater in Arctic charr, brown trout, and Atlantic salmon fed a diet containing vegetable oil (1:1 blend of flaxseed and rapeseed oils) as compared to a control diet containing fish oil (Tocher and others 2001). Rosenlund and others (2001) replaced 50% of the fish oil in a high-energy salmon diet with flaxseed oil. No significant effects of dietary lipid source were found on growth, survival, body traits, or fillet quality. The fatty acid profile results showed that high-energy diet where 50% of the fish oil was replaced with the flaxseed oil produced similar results to diets containing 100% fish oil in Atlantic salmon. This further confirms that the essential fatty acids in the flaxseed oil were used by Atlantic salmon to synthesize the omega-3 polyunsaturated fatty acids.

Chaiyapechara and others (2002) fed trout diets with lipids at 15 and 30%. The diets were supplemented with vitamin E at 300 and 1500 mg of dl-(-tocopheryl acetate per kg of feed. Sensory analysis of trout fillets obtained from fish fed diets with 30% of lipids and 300 mg/kg of vitamin E showed off-flavor, which was correlated with such flavor attributes as fishy, musty, sour, and bitter. The dl-(-tocopheryl acetate at 1500 mg/kg of feed reduced the rate of lipid oxidation and the formation of undesirable off-flavors.

Production of All-Female Triploid Brook Trout. Brook trout *Salvelinus fontinalis* is the only salmonid native to the Appalachian Mountains in the eastern United States (south of Vermont and New Hampshire). They are a colorful, popular species produced by both private and public growers for sport fishing. Non-native rainbow trout *Oncorhynchus mykiss* and brown trout *Salmo trutta* have also been stocked into the region's waters to supplement the sports fishery, where these larger, longer-lived species have displaced brook trout from much of its original

range (MacCrimmon and Campbell 1969, Flick 1991, Johnson 1991, Karas 1997). However, brook trout continue to add colorful diversity for recreational trout anglers. During the 1997-98 fiscal year, the West Virginia Department of Natural Resources stocked 76,435 fish weighing 40,272 lb or 6% of total weight stocked by the agency (WV DNR Annual Report). (NASS data doesn't distinguish among trout species.) In western North Carolina, a total of approximately 700,000 trout are stocked into streams in the region, of which 40% are brook trout (the remainder being 40% rainbow trout and 20% brown trout). Brook trout is generally considered the finest trout at the table." (McClane 1977).

Although brook trout are greatly appreciated, especially in the eastern United States, essentially all commercial food fish production of trout in the country is of rainbow trout. Brook trout, because of a reputation for slower growth and because of the relatively young age and small size at which they become sexually mature, have been considered less appropriate for commercial culture than rainbow trout. And, now that rainbow trout have undergone numerous generations of selective breeding for commercial production, this view may have been to some extent self-fulfilling. However, it should also be noted that at the Pisgah Forest State Fish Hatchery, Brevard, NC, growth to age 1 is similar between the two species.

Brook trout do possess several characteristics which would lend themselves to commercial food fish production. They are easy to propagate and possess a relatively high tolerance to crowding and are more tolerant than rainbow trout to the acid waters found in many Appalachian watersheds (Robinson et al. 1976; Power 1980). Brook trout are more resistant to enteric redmouth than rainbow trout, although brook trout are also more susceptible to furunculosis (Leitritz and Lewis 1980). Both of these pathogens can be problematic for commercial trout growers in the region. Brook trout does, however, have a reputation for being more sensitive than rainbow trout to thermal stress (Gabrielson 1963, Bardach et al 1972, Schwiebert 1978). The optimum temperature for brook trout is indicated as 45-55 °F and rainbow trout is 50-60 °F (Piper et al. 1982), and chronic thermal maximum for brook trout was slightly lower than for rainbow trout (Galbreath unpublished data). On the other hand, brook trout would possess certain attractive marketing characteristics as a food fish. Just as many regional fishermen hold the native brook trout in higher esteem than the introduced trout species, so might they possess a particular attraction in the white tablecloth market or as a "specialty" retail food item were they available commercially, particularly with relevant promotion.

However, culture of brook trout as a food fish is additionally constrained by the lack of commercially available all-female stocks. Somatic growth in salmonids slows dramatically with the onset of sexual maturity, as the fish divert metabolic resources for growth and maturation of the gonads (Lincoln and Scott 1984). Additionally, sexual maturation in salmonids is accompanied by acquisition of secondary sexual characteristics - decrease in growth rate, darkening, degradation of flesh quality, etc. - which significantly reduce their attractiveness and market value (Hunter and Donaldson 1983; Bye and Lincoln 1986). Although the incidence of maturation in rainbow trout is typically at age 2 or 3, early maturation (at age 1) occurs in a certain percentage of male rainbow trout, and this percentage tends to be higher in fast growing commercially reared fish. For this reason, food fish production of rainbow worldwide is predominantly of all-female stocks, generated by use of sex-reversed XX "male" broodstock (Galbreath and Stocks 1999). In salmonids, the homogametic sex is the female (XX), and the

heterogametic sex is the male (XY). All-female stocks may be produced by application of steroid hormone treatments to newly hatched trout fry - the period during which the gonadal tissue is undergoing differentiation - which induces sex-reversal of genetic females into functional phenotypic "males". When raised to maturity, however, these fish will produce sperm which contains only X chromosomes and 100% of their progeny, therefore, will be female (Johnstone et al. 1978, Hunter et al. 1983, Piferrer et al. 1993). Sexual maturation in brook trout occurs consistently at 2 years of age, and the problems associated with early maturation among male fish can only be expected to be worse for this species. Therefore, commercial rearing of brook trout as a food fish must necessarily be with all-female stocks if it is to be economically viable. However, the first published protocol for sex-reversal of brook trout (with which to produce the required XX "male" broodfish) is only now being published (Galbreath et al. in press), and no commercial supplier of all-female brook trout exists in the United States.

Inducing sex-reversal in brook trout has proven difficult. Sex-reversal protocols effective with rainbow trout had very limited success when applied to brook trout (Galbreath and Stocks 1999). However, continued efforts finally yielded limited numbers of XX sex-reversed brook trout "males" produced by treatment of fry with methylidihydrotestosterone (MDHT), as confirmed by progeny tests which yielded 100% females among the offspring (Galbreath et al. in press). Additional trials of the sex-reversal protocols conducted each fall are on-going, and will hopefully continue on a yearly basis, in order to identify a protocol with maximized efficacy - both in terms of percent masculinization, and in terms of producing males with intact sperm ducts such that the milt can be manually stripped and the fish retained alive for spawning in successive seasons.

With the availability now of XX sex-reversed brook trout "males", commercial production of all-female fish can be envisioned. Production of pan-sized all-female brook trout should be feasible within a two year production cycle, such that the fish reach market size prior to the onset of maturation. However, there is also an economical means to preclude sexual maturation entirely in female salmonids - by the production of triploid fish. Triploidy - the presence of three full sets of chromosomes per somatic cell - is induced in trout, and a wide variety of other fish species, by application of a thermal or hydrostatic pressure shock to newly fertilized eggs (see review by Ihssen et al. 1990). Triploidy in female salmonids causes ovarian development to arrest very early in embryonic development. Triploid females are therefore sterile, and they retain their commercially attractive silvery juvenile appearance all their lives. Also, as they approach two years of age, triploid females maintain a steady rate of somatic (muscle) growth. At this age, normal diploid females, however, become sexually mature and use valuable food energy for production of gonads, significantly reducing their dress-out percentage (Lincoln and Scott 1984). (Unfortunately, this is not the case with triploid males, which do develop testes and the accompanying undesirable secondary sexual characteristics.) Production of triploid all-female trout therefore expands the window for marketing of pan-sized fish, and also permits rearing of fish to ages and sizes greater than that achieved at two years, for use in markets requiring large sized fish, e.g., for sale of fillets. Triploid (all-female) rainbow trout eyed-eggs are commercially available to growers who wish to produce sterile fish. However, the advantages of all-female triploid production would be even more attractive with brook trout. Protocols to induce triploidy in brook trout are very similar to those used with other salmonids, and are easy to apply (Galbreath and Samples 2000), and production of all-female triploid brook trout is now possible.

It is of interest to note as well, that production of triploid trout also presents advantages in a sport fishing/fisheries management context. Triploids are functionally sterile, and therefore unable to successfully interbreed with native/wild salmonid populations. Therefore, stocking of hatchery-reared triploid fish will not compromise the genetic integrity of native/wild populations. They are also stocked to promote trophy fisheries, as triploid females are not susceptible to spawning - related mortality and tend to be longer-lived. Triploids are equally susceptible to capture by hook and line (Dillon et al. 2000), and triploid female salmonids do not undergo spawning migrations and will remain in the lake or stream into which they were stocked for the sport fishery (Warrillow et al. 1997, Benfey 1999).

Performance and physiology of triploid salmonids in culture has been examined primarily for rainbow trout and Atlantic salmon. Generally, results indicate little if any difference related to ploidy as juveniles, followed by improved performance of triploid females following sexual maturation (Pepper 1991, Galbreath et al. 1994, Benfey 1996, Benfey 1997, Sheehan et al. 1999, Cotter et al. 2002). Only a very few comparative growth trials of diploid and triploid brook trout have been published, and these studies indicate similar performance relative to ploidy (Boulanger 1991, O'Keefe and Benfey 1999). Anecdotal evidence has been described to indicate an apparent reduced stress tolerance of triploid salmonids in culture, however these observations have not been consistently supported by controlled experimentation (see review in Benfey 1997).

Recent efforts to commercially produce Arctic char *Salvelinus alpinus* in the eastern United States demonstrate the existence of markets for specialty salmonid production in addition to those typically filled by production of pan-sized rainbow trout. However, these efforts have met with significant problems of both a biological and economic nature. Production of the native brook trout (also a char) may be a more attractive alternative to meet this market, particularly if all-female and all-female triploid stocks can be reliably produced. Prior to promoting production of brook trout, however, demonstration of their growth and marketing potential in the region is needed.

#### Catch-related standards of quality and demand behavior for fee fishing stocking projects

There are over 30 pay fishing establishments in West Virginia that charge customers a fee for the privilege of fishing a privately owned pond or lake where fish populations are enhanced by stocking fish. These businesses purchase about 300,000 pounds of fish each year. Findings from recent studies conducted by West Virginia researchers suggest that there are opportunities for growth in this recreational market. Logar, Ponzurick, and Semmens (2001) reported that bluegill, bass, trout, and catfish are the most preferred species that anglers would like to catch. Trout and catfish are widely stocked in West Virginia pay ponds and have been examined in previous studies to help determine standards of quality (e.g., minimum acceptable size of fish and catch rates). However, bluegill and bass are not widely stocked in the state and the effect they have on demand behavior and their catch-related standards of quality are not understood.

#### **Current Work**

The progress report (pg. 5 ) briefly described current work that is funded by this project. In this section, we will emphasize work that is funded from other sources.

Lessons learned at Dogwood Lakes have served to produce a modified design of a four-tank raceway system that is being installed at the WVU Reymann Memorial Farms in Wardensville, WV; this project was funded by the State's Research Challenge Competitive Grant. These two sites with HFRP raceway systems have offered the opportunity to focus the research on utilization of mine-waters (Dogwood Lakes) in relation to a bench mark using spring water (Reymann Memorial), where both locations constitute large-scale field laboratories that can be translated to commercial applications. In addition, we are currently working on a floating raceway system, funded by the Northeast Regional Aquaculture Center (NRAC), of two tanks that will be assembled from flat panels, unlike the built-in construction of the two raceway systems, and installed in treated mine waters at Warwick Mine in Greene Co., Pennsylvania. These efforts have indicated that significant opportunities exist for future commercial aquaculture applications of HFRP tanks.

We are also working to optimize design of the quiescent zone; the detachable and replaceable end-unit of each raceway tank used for waste collection and subsequent treatment. Studies are underway to optimize the composite material architecture (fibers and resins) to provide best performance in strength and stiffness for minimum weight, which is directly related to cost. We are conducting a survey of existing systems to have a better understanding of costs, including life-cycle, and explore market opportunities for HFRP raceways.

Work is in progress to assess the effect of elevated levels of CO<sub>2</sub> on growth and metabolism in rainbow trout and feeding strategies to alleviate the effects of elevated CO<sub>2</sub>. Work is also underway to assess the effects of vitamin E on storage properties of rainbow trout fillets as are investigations as to the effects of different harvest methods on fillet attributes.

Additional work relating to this project includes the Trout Genome Project funded by USDA/ARS, a study conducted by Drs. Logar and Ponzurick regarding fee fishing at Ogleby Park funded by the Benedum Foundation, and a "Partnerships For Innovation" grant from the National Science Foundation to Dr. Davalos for development of novel HFRP materials for industrial applications such as bridge decks and aquaculture raceways.

## **Facilities and Equipment**

The physical facilities of West Virginia University will be utilized for developing, analyzing and reporting the study. Facilities of the Davis College of Agriculture, Forestry and Consumer Sciences will draw upon resources from the three divisions of Animal and Veterinary Science, Resource Management (Resource Economics), and Forestry (Recreation and Parks). The Department of Civil and Environmental Engineering, in the College of Engineering, the West Virginia Extension Service and the Cooperative Fishery Research Unit at WVU will provide additional resources. Personal computers and software provided by the institution will be used in implementing this project.

Proposed work will take place at two pilot scale raceway systems. Each system is composed of HFRP material with four levels of paired units, each 30 feet long capable of maintaining approximately 1000 lb of fish. Combined, the systems are supplied with about 1000 gallons/minute, and have a total of 16 experimental units. The facility at Dogwood Lake,

approximately 15 miles west of WVU main campus is supplied with treated mine water. The facility at Reymann Memorial Farm is part of the WV Agricultural Experiment Station near Wardensville, WV and is fed by spring water.

Rainbow trout utilized for the metabolic aspects of growth and efficiency will be maintained at the National Center for Cool and Cold Water Aquaculture (Kearneysville, WV). This facility is equipped to rear fish and the experienced staff will oversee care of the fish. The staff at the NCCCWA will collect the feed intake data.

Laboratory analyses for the metabolic experiments will predominately occur within the Division of Animal and Veterinary Sciences at West Virginia University. Measures related to the efficiency of nutrient retention will occur in the laboratory of Dr. Blemings. The Blemings laboratory is equipped with a spectrophotometer (Beckman DU640), a fluorometer (Turner Designs 700), thermal cyclers, scales, speed-vacuum, electrophoresis equipment, computers for data analysis, shaking water baths, tissue grinders, homogenizers, and a Waters Breeze HPLC system with UV-VIS and radiochemical detection. Within the Division, there is also access to ? (Beckmann LS6500) and ??(Wallac 1470 WizardEKG) -Counters, sonicating waterbaths, Spectrofluorometer (Cary Varian Eclipse), centrifuges (nano, micro, clinical, superspeed, ultraspeed), autoclave, water purification system, fume hoods, refrigerators, freezers, film developers, drying ovens, Tecator Kjeltac Auto 1030 Analyzer unit for Kjeldahl nitrogen determinations, ABI prism DNA sequencer, an AlphaInnoTech image analyzer, and a chloride analyzer.

Product quality analyses for the metabolic experiment will occur in the Kenney laboratory. The Kenney laboratory is equipped with a 1100 ft<sup>2</sup> meat processing facility, a 750 ft<sup>2</sup> wet lab as well as walk-in coolers (940 ft<sup>2</sup>) and freezers (150 ft<sup>2</sup>). The meat processing facility is equipped with a grinder, meat stuffer, band saws and mixers. Additionally, a Griffith mincemaster emulsion mill, Hobart bowl mixer and a microprocessor controlled smokehouse are available. The Instron Universal testing machine (Model TM) , interfaced with data acquisition hardware and signal processing software for texture analysis are housed in the Kenney laboratory. The wet lab houses vertical slab gel electrophoresis equipment, Minolta chromometer, Goldfish and Soxhlet fat extractors and an ashing oven.

The antioxidant status work for the metabolic experiments will be conducted in the laboratory of Dr. Klandorf. The Klandorf laboratory is equipped with centrifuges, scales, pH meters, microscopes, refrigerators, and freezers, computer for data analysis and a glucose analyzer. Additionally, Dr. Klandorf has access to a spectrophotometer and a Waters Alliance HPLC system with photodiode array and scanning fluorometric detection.

The omega 3 and protein recovery work will be conducted by Dr. Jaczynski. The fatty acid profile, Soxhlet extraction, and 2-thiobarbituric acid assay will be performed in the Division of Animal and Veterinary Sciences (A&VS). Dr. Jaczynski has a sufficient laboratory space in the A&VS and a direct access to the gas chromatograph (Varian CP-3800), Soxhlet extraction apparatus, and spectrophotometer (Beckman DU640) necessary to carry out the analyses. There is direct access to the spectrophotometer, SDS-PAGE cell, homogenizer, ultra-speed

refrigerated centrifuge, pH meter, universal food processor, water bath, colorimeter, torsion gelometer, texture analyzer, and gas chromatograph.

Engineering. Facilities of the College of Engineering and Mineral Resources contains in excess of 5000 ft<sup>2</sup> of laboratory space. Analytical equipment includes: one atomic absorption spectrophotometer; three gas chromatographs with multiple detector arrays (PID and FID) and a purge and trap apparatus; two total organic carbon analyzers; three automated pH titrators; one scanning electron microscope; and one UV-visual light absorbance spectrophotometer. The National Research Center for Coal and Energy (NRCCE) located adjacent to the engineering facilities will conduct water quality analyses. The laboratory is EPA certified for performing analysis under the National Pollutant Discharge Elimination System of the Clean Water Act. All analytical methods are EPA approved and have a standard Quality Assurance/Quality Control protocol.

### **Project Timetable**

Most components of this proposal will take a year to complete. Not all components will begin at the same time, however and may depend on work presently underway to be completed. Analysis and technology transfer efforts will require additional time such that projects are expected to be complete in two years. For example, experiments conducted to measure metabolic aspects of growth and efficiency of nutrient use in different strains of rainbow trout will require 14 months in the lab. The grow-out time for experiment 1 is about 2 months and the grow-out time for experiments 2 and 3 is 5 months. This gives a total time for fish growth of about 12 months. Analysis of experiments 1 and 2 can be completed while experiment 3 is in progress and the analyses of experiment 3 may take an additional 2 months. The triploid brook trout experiment will begin in fall of 2003 and require two years. The fish must be spawned, and the fingerlings grown during the first six months. Fingerlings will be distributed and grown out to a harvestable size during the next 14 months. We estimate all components of the project will begin after August, 2003 and be completed by September, 2005.

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## **Key Personnel**

Kenneth J. Semmens. Dr. Semmens, is State Extension Specialist for Aquaculture and specializes in aquaculture with twenty years of experience producing and marketing a wide variety of warm, cool and coldwater fish species. He holds a joint appointment with the Cooperative Extension Service and the West Virginia Agricultural and Forestry Experiment Station. He will oversee the execution of the project's activities, assist the co-investigators with problems and issues that arise, facilitate resource allocation and integrate project activities in support of the aquaculture industry in West Virginia.

Kenneth P. Blemings. Dr. Blemings is an assistant professor of nutritional biochemistry in the Division of Animal and Veterinary Sciences and a member of the interdepartmental program in Genetics and Developmental Biology. His research interests are in the efficiency of nutrient use, gene-nutrient interactions and regulation of growth. He will oversee and coordinate the metabolic part of the project.

Kelly S. Bricker, Ph.D., Assistant Professor in Recreation, Parks, and Tourism Resources, will work with graduate students in developing a comprehensive analysis program of data (SPSS or other), supervise publications or results in journals

Julio Davalos is C.W. Benedum Distinguished Teaching Professor, Department of Civil and Environmental Engineering, College of Engineering and Mineral Resources, West Virginia University. His primary research interests are analytical and applied mechanics, characterization of wood and fiber-reinforced polymer composites, structural and bridge engineering, and effective teaching methods. Dr. Davalos has produced design manuals and taught courses on composite materials. He will lead the task developing HFRP tanks for field assembly from flat panels.

Jacek Jaczynski is an assistant professor of food science in the Division of Animal and Veterinary Sciences at West Virginia University. His interests are aquatic foods and food safety. He will lead the two tasks, Omega-3 fortified Arctic Charr as functional food, and development of value-added food based on proteins and lipids recovered from trout processing by-products.

P. Brett Kenney. Dr. Kenney is an associate professor of muscle foods in the Division of Animal and Veterinary Sciences. His research interests are in the effect of management practices on quality of fresh and smoked products as well as food safety. He will oversee the texture and color analyses of the fillets.

Hillar Klandorf. Dr. Klandorf is a professor of physiology in the Division of Animal and Veterinary Sciences. His research interests are in antioxidants and their affect on tissue crosslinks. He will oversee the analyses of total antioxidant capacity.

Patricia M. Mazik. Dr. Mazik is an adjunct professor of fish and wildlife in the Division of Forestry. Her research interests is in fish response to stress and toxicology. She will supervise the graduate student who will do the pH, hematocrit, cortisol and in vivo metabolism work.

Daniel Miller, M.S., is Senior Research Coordinator working in the Animal and Veterinary Science and the Agricultural and Resource Economics Program. Mr. Miller's work experience emphasizes the biological and production aspects of aquaculture and fisheries. He has served as a manager of and consultant on several aquaculture-related projects in various parts of the U.S. and overseas. His work on the project will focus on technology transfer particularly development of the mine water resource for commercial production of salmonids.

Patricia Miller, Ph.D. Assistant Professor & Extension Specialist, Agricultural/Rural Safety, Health, & Environmental Health. She will assess facilities and current practices regarding safety at fee fishing businesses to develop design, best practices, and management recommendations.

Chad D. Pierskalla, Ph.D., Assistant Professor in Recreation, Parks and Tourism Resources, will coordinate the study of catch related standards for hybrid striped bass and will help coordinate the study of demand behavior for a hybrid bluegill stocking project. Dr. Pierskalla has research experience in the human dimensions of natural resources management.

Justin Robinson. Mr. Robinson is a Engineering Scientist working for the Department of Civil and Environmental Engineering under the direction of Dr. Julio Davalos. He will assist in the development and evaluation of the improvements to be made on the HFRP raceways from flat panels.

Randall S. Rosenberger, Ph.D., Assistant Professor in Agricultural and Resource Economics, will assist in the study of demand behavior and estimation of recreational values. Dr. Rosenberger has expertise in recreation demand and valuation estimation.

Richard Turton, Ph.D., Professor, Department of Chemical Engineering will collaborate with Dr. Viadero and Dr. Semmens on the development and verification of a model to describe production of rainbow trout in raceway systems. He has specific expertise in various forms of modeling and simulation.

Roger C. Viadero, Jr., Assistant Professor of Civil and Environmental Engineering will conduct task development and verification of a model to describe production of tainbow trout in raceway systems. Dr. Viadero has led research on engineering aspects of water treatment in recirculating aquaculture systems used to raise yellow perch.