SALMONIDS

Project Component Termination Report for the Period June 1, 1990 to August 31, 1996

NCRAC FUNDING LEVEL: \$479,796 (June 1, 1990 to August 31, 1996)

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REASON FOR TERMINATION

The objectives for this work on Salmonids were completed.

PROJECT OBJECTIVES

- (1) Develop less-polluting diets. Develop practical rainbow trout diets using regionally available feed ingredients, including fish meal analogs.
- (2) Determine the practical limits on rearing density of juvenile rainbow trout by examining the effects of selected high rearing densities on trout stress responses, survival and growth. Use stress and performance responses in trout to evaluate culture system design and operation under

practical conditions. Use the stress response as a selection tool for developing strains of trout having improved performance under conditions found in the North Central Region (NCR).

PRINCIPAL ACCOMPLISHMENTS

OBJECTIVE 1

Investigators at Michigan State University constructed a nutritional phosphorus (P) mass balance model for coho and chinook salmon as a method of estimating P losses from the Platte River Anadromous State Fish Hatchery. For the production period of January 1993 through May 1994, the P mass balance model indicated that 37.7% of P fed was retained by fish, 21.0% was discharged in the feces, and 41.3% was discharged in dissolved form. Without any raceway solids removal, a maximum of 2.8 kg/metric ton (MT; 5.6 lb P/ton) of fish produced was discharged into the hatchery's stabilization pond. This loss rate was the lowest reported for a salmonid hatchery. Efficient removal of raceway solids could have reduced hatchery P losses to 1.8 kg P/MT (3.6 lb P/ton) of fish produced.

Work was completed on improving the digestibility and utilization of nutrients in feeds which should also reduce nutrients in aquaculture waste water. Rainbow trout were fed diets containing untreated or dephytinized soybean meal and corn gluten meal with or without supplemental zinc (Zn) to determine if fish dietary phytate impairs Zn bioavailability in fish. Fish fed diets containing untreated soybean meal and corn gluten meal without supplemental Zn (basal diet) were not Zn deficient after 170 days based on growth, whole fish Zn, P and protein content, total bone Zn, and the activity of alkaline phosphatase and carboxypeptidase B. Although bone Zn concentrations were reduced in fish fed the basal diet, total bone Zn increased in all fish regardless of dietary treatment. The basal diet contained enough available Zn to offset any negative effects of phytate on Zn bioavailability. Future assessments of Zn status in fish should be based on changes in Zndependent metabolism or total bone Zn rather than bone Zn concentration.

Researchers at Ohio State University compared the growth rates of rainbow trout fed five different diets in which fish meal protein was replaced by an animal by-product mixture (i.e., replacement of 0, 25, 50, 75 and 100%). No differences were found among the five treatment groups in fish growth, dressing percentage, fillet quality, or gamete quality. Analysis of P levels in fecal samples, however, indicated that diets containing animal by-products were not less-polluting.

Research at Purdue University (Purdue) first defined the methods for precisely determining available P from feedstuffs. Fecal collection methods were evaluated, and then a new indicator of nutrient availability was developed. After defining the appropriate methods, available P was determined from single ingredients in diets, as well as multiple ingredients fed to both rainbow trout and Atlantic salmon.

Purdue researchers found that a fish meal-free diet containing soybean meal, corn gluten meal, and corn grain as the predominate ingredients could promote weight gains in rainbow trout within 90% of fish fed a control diet. Improvements were made in this diet. Lysine was identified as the first-limiting essential amino acid, meat meal was successfully incorporated into the diet, and a combination of canola and fish oils were found to be better than either lipid source alone. A commercial astaxanthin product successfully masked the yellow pigmentation in the muscle of trout. Fish fed any of the fish meal-free diets were preferred by a trained taste panel over filets from fish fed a commercial diet. Two graduate students were trained during this project.

OBJECTIVE 2

University of Wisconsin-Madison (UW-Madison) investigators conducted laboratory studies to compare the performance of juvenile rainbow trout stocked at density indices (pounds of

fish/[cubic feet water x average fish length in inches]) of 0.45 (generally recommended maximum rearing density), 0.90 and 1.35 at loading rates ranging from 491.3 to 982.6 g/(Lmin) [4.1 to 8.2 lb/(galmin)]. Fish were reared at 15C (59.0F) for four weeks at the end of which time the fish were weighed, measured, and subjected to an acute stress challenge test to evaluate treatment effects on fish growth and specific physiological stress responses. High mortality occurred at a loading rate of 982.6 g/(Lmin) [8.2 lb/(galmin)], probably due to elevated unionized ammonia levels. Fish reared at densities of 5.68 and 8.51 g/(Lcm) [0.90 and 1.35 lb/(ft³in)] grew significantly less than fish reared at 2.84 g/(Lcm) [0.45 lb/(ft³in)] at loading rates of either 491.3 to 719.0 g/(Lmin) [4.1 and 6.0 lb/(galmin)]. Fish reared at densities of 5.68 and 8.51 g/(Lcm) [0.90 and 1.35 lb/(ft³in)] gained 78% and 62% of the weight of the fish reared at 2.84 g/(Lcm) [0.45 lb/(ft³in)], respectively. These results contrast with earlier studies conducted at UW-Madison showing that similar high rearing densities had no effect on the growth or feed conversion of juvenile rainbow trout reared at 9.8-11.9C (49.6-53.4F) at a loading of 299.6 g/(Lmin) [2.5 lb/(galmin)]. Cortisol levels were higher at 24 h post-stress in fish reared at low density compared to fish at medium and high density. This difference was attributed to the establishment of stressful social hierarchies in the low-density tanks (which break down under crowded conditions). Fish reared at high density had higher baseline cortisol levels and longer (though not higher) increases in cortisol following an acute stressor relative to fish reared at low density with corresponding changes in glucose and chloride levels. The data indicate that high rearing density can have a negative impact on fish growth and food conversion if loading rates are between 299.6 and 491.3 g/(Lmin) [2.5 and 4.1 lb/(galmin)] and/or rearing temperature is greater than approximately 12C (53.6F). The negative consequences of high density/loading appear to be at least partially mediated by physiological stress responses.

A 10-week production-scale field trail was performed at the Calamus State Fish Hatchery by University of Nebraska-Lincoln researchers with help from personnel of the Nebraska Game and Parks Commission comparing the growth, performance, mortality rates, health, and stress responses of rainbow trout in raceways versus oxygen-supplemented cylindrical tanks. Six of the latter were each equipped with a sealed packed column supplied with oxygen, and assigned fingerling trout at a rearing density of 2.84 and 5.68 g/(Lcm) [0.45 and 0.90 lb/(ft³in)] (three tanks per treatment). Six raceways equipped with conventional packed columns were also each assigned fish at a density of 2.84 and 5.68 g/(Lcm) [0.45 and 0.90 lb/(ft³in)] (three raceways per treatment). Turnover rates were kept constant between all four treatment groups. Parameters measured during the course of the study were dissolved oxygen, carbon dioxide, ammonianitrogen, pH, total dissolved gas pressure, P, and temperature. At the conclusion of the study, a stress challenge test and Goede health assessment were performed. The results indicated that juvenile rainbow trout could be reared at twice the generally recommended density with no negative impact on growth, food conversion or health. Similar results were found in both tanks and raceways.

Investigators at UW-Madison identified a physiological measure of stress that was well correlated with growth in rainbow trout -- serum cortisol levels 3 h following an acute handling stressor. Individual fish that consistently showed low 3 h post-stress cortisol levels (i.e., fish that recovered rapidly from stress, defined as "low" fish) had a mean specific growth rate (SGR; % weight gain per day) of 0.54, compared to a mean SGR of 0.41 in unselected fish. Fish with consistently high cortisol levels at 3 h post-stress and a low SGR ("high" fish) were also identified. A selection process was initiated in December 1995 to identify "low" and "high" fish for subsequent breeding. Starting with 160 two-year-old fish obtained from Seven Pines Trout Hatchery, five female and five male "low" fish, and nine female and five male "high" fish were selected. The fish were bred in the autumn of 1996 but there was poor fertilization success and very low larval survival due to a system-wide water quality problem. The selected fish were rebred in the autumn of 1997. Sperm from each selected "low" male was used to fertilize a subgroup of eggs from each selected "low" female, and likewise for the "high" fish and randomly selected controls. Groups of larval fish from each population ("low," "high," and control) are currently being reared for subsequent performance evaluations.

IMPACTS

The less-polluting diets developed during this project will benefit aquaculturists facing strict regulatory pressures to reduce waste nutrients in effluents.

The use of regionally available plant and animal by-product protein sources as substitutes for fish meal will reduce the cost of trout feed manufacture by at least 15% by reducing both ingredient and transportation costs. Diets formulated with fish meal analogs were found to have little or no impact on fish growth, feed conversion or flesh quality, and were also less polluting.

The results demonstrated that fish farmers may be able to at least double rainbow trout production from their *existing* facilities if loading rates are kept low by supplemental oxygenation and/or increased water flow rates. These findings are particularly important to trout farmers in the NCR who are generally constrained by limitations in water and rearing space.

An increased understanding of how rearing density, loading, and water turnover rates influence fish growth, feed conversion, and disease resistance will improve overall production efficiency and help reduce effluent wastes.

The physiological indicator of stress resistance and superior growth identified in this project will be invaluable for developing fast-growing, stress-resistant trout strains for use under the distinctive aquaculture conditions found in the NCR (i.e., relatively small-sized farms, low water flows, and variable water temperatures).

RECOMMENDED FOLLOW-UP ACTIVITIES

Factors that impact available P have not been determined. The results indicate that mineral interactions have the most profound effect on available P in diets fed to fish. Of the 20-24 essential minerals required by vertebrates, specific levels and forms of minerals that impact available P in fish have not been determined.

Further explore available animal by-product utilization as replacement of fish meal in combination with plant proteins available within (canola, soybean) or outside (cottonseed) of the region.

Evaluate brood stock diets prepared with fish meal analogs.

Evaluate the effects of diet formulations devoid of fish meal on fillet quality.

Evaluate diets prepared with fish meal analogs with other salmonid species.

Evaluate the stress responses of the offspring of "low" and "high" rainbow trout to determine the heritability of the stress response. Evaluate the performance of the offspring of selected "low" fish under commercial aquaculture conditions. Develop a stress-resistant, fast-growing strain of rainbow trout for the NCR using the stress response as a selection criterion. Determine if the stress hyperresponsive "high" fish identified in this study (which have stress and growth characteristics typical of "wild" trout) will have advantages for stocking recreational fisheries.

Conduct integrative research to evaluate the combined effects of the improved feeds, rearing strategies (e.g., high density), and fish strains identified or developed during this project on the salmonid aquaculture industry in the NCR.

Extend the insights and accomplishments of the completed salmonid project to other aquaculture species in the region, including yellow perch and walleye. It may be possible, for example, to

develop less-polluting yellow perch diets made with animal by-product or plant protein sources, or produce a fast-growing, "domesticated" strain of yellow perch by selecting for and breeding stress-resistant fish.

PUBLICATIONS, MANUSCRIPTS, AND PAPERS PRESENTED

See Appendix for a cumulative output for all NCRAC-funded Salmonid activities.

TOTAL SUPPORT FOR THE THREE PROJECTS

	NCRAC-USDA FUNDING	OTHER SUPPORT					TOTAL
YEARS		UNIVERSITY		OTHER FEDERAL	OTHER	TOTAL	SUPPORT
1990- 92	\$129,799	\$184,843				\$184,843	\$314,642
1992- 94	\$149,997	\$237,493			\$66,700 ^a	\$304,193	\$454,190
1994- 96	\$200,000	\$208,083		\$18,590 ^b	\$15,000°	\$241,673	\$441,673
TOTAL	\$479,796	\$630,419		\$18,590	\$81,700	\$730,709	\$1,210,505

^aNebraska Game and Parks Commission

^bUniversity of Wisconsin Sea Grant

^cInternational Collaborative Program for OSU to work jointly with the National Fisheries University of Pusan, Korea