**ATTACHMENT A**

**PROJECT NAME:** Culture Technology of Walleye

**FUNDING LEVEL:** Year 1 - $58,614
Year 2 - $53,043

**DURATION:** 2 Years

**ADMINISTRATIVE ADVISOR:** Dr. Donald L. Garling, Department of Fisheries and Wildlife, Michigan State University, East Lansing, MI 48824-1222

**TABLE OF CONTENTS**

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## JUSTIFICATION

The walleye (*Stizostedion vitreum*) and the walleye (-sauger) hybrid (*S. vitreum* female x *S. canadense* male) have been designated priority candidates for commercial aquaculture development in the North Central Region (North Central Regional Aquaculture Center FY90 Program Development Meeting, 20-23 May 1989, Des Moines, Iowa). Traditionally, the walleye has been one of the most exploited coolwater species in North American commercial and recreational fisheries (Kendall 1978). In recent years, the commercial harvest of walleye in the U.S., except for a few tribal fisheries, has essentially been eliminated in favor of sport fishing. To support walleye fisheries, state, federal, and provincial fisheries management agencies in North America currently stock more than one billion walleye fry and fingerlings annually (Conover 1986). The walleye hybrid has also been propagated and stocked in many parts of the North Central Region, using procedures almost identical to those employed with purebred walleye (see references in Colby et al. 1979; Ebbers et al. 1988).

Given the large number of walleye fry and fingerlings reared for stocking, various fisheries management agencies in the U.S. and Canada have conducted applied research on various phases of walleye aquaculture for many years (Coolwater Culture Workshop 1985, 1986, 1987, 1988, 1989). Fish culture activities traditionally associated with producing walleye include spawning of wild broodstock, hatching fry, and rearing small fingerlings in ponds. Numerically, fry comprise 98% of walleye stockings in the U.S. and Canada (Conover 1986). However, the relative survival of fingerling walleye after stocking is 16 to 60 times greater than with fry (Heidinger et al. 1985). Thus most fisheries management agencies prefer fingerling stockings; and large numbers of fingerlings can be produced to a size of 35-50 mm total length (TL) by traditional (pond culture) methods.

Increasingly, the focus of regional research is being directed toward the development of techniques for rearing small (35-50 mm TL) pond-reared walleye fingerlings to a larger size (100-150 mm TL). Such techniques involve habituating pond-reared fingerlings to intensive culture conditions and formulated diets. However, success is variable and affected by many factors, including temperature, light, size and condition of the fingerling, harvest and handling stress, diet, and feeding methods (Nickum 1978, 1986).

At present, commercial walleye aquaculture is also mainly geared to the production of eggs, fry and pond-reared fingerlings. Over the past 27 years, the Lac du Flambeau Tribal Fish Hatchery, Lac du Flambeau, Wisconsin, has produced 373 million walleye fry for stocking in tribal waters and for sale. Commercially, walleye fry and fingerlings are sold to lake associations, sports clubs and individual lake and pond owners for stocking. Given the incentive of market prices for individual fry of 1 to 1.5 cents and $0.25 to $0.75 for fish 35 to 100 mm TL, respectively, commercial walleye production has expanded rapidly in the past 5 years. The growth of private-sector pond production has been particularly marked in Minnesota, Nebraska, Wisconsin, Iowa, and Michigan.

Experience with rearing walleye to food-size has been largely limited to a few researchers in the region (primarily at Iowa State University, and Southern Illinois University). However, the high retail prices of walleye fillets that have prevailed throughout the region ($16.09-25.35/kg) have been a strong stimulus to private sector interest in the production of food-size fish. Although total production has been small, some commercial culture of food-size walleye has been underway for several years (e.g., Walleye Farms, Knoxville, Iowa).

Major constraints that impede the development of commercial walleye aquaculture include: (1) the lack of procedures for manipulating reproduction and inducing spawning in walleye broodstock; (2) the lack of captive, domesticated broodstock; and (3) the unreliability of pond management strategies for fingerling production. Research on these topics by the Walleye Work Group of the North Central Regional Aquaculture Center (NCRAC) is presently underway and is described in the Work Group's initial project outline (included in "Program Plan for Grant 2: North Central Regional Aquaculture Center," dated 7 April 1989). The present document constitutes a

proposed addendum to the Walleye Work Group's ongoing regional project and focuses on genetic analyses of walleye populations for potential use as broodstock. The lack of a captive domesticated broodstock is a constraint identified by NCRAC as priority areas for research (NCRAC FY90 Program Development Meeting, 20-23 May, Des Moines, Iowa).

The economic viability of walleye aquaculture lies in the development of selected strains of domesticated broodstock that are adapted to the environment and feeding regimes found in commercial aquaculture. Genetic research is a central factor in the world-wide development of aquaculture (Wilkins and Gosling 1983; Gall and Busack 1986). Genetic research aimed at improving culture technology for walleye was ranked as a high priority at the joint meetings of the Industry Advisory Council and the Technical Committee of NCRAC, held in May 1988, in East Lansing, Michigan and May 1989, in Des Moines, Iowa. Initiation of genetic studies at an early stage of technological development is very important because the generation time of three to four years for cultured walleye to reach sexual maturity is an inherent lag in the improvement of production traits.

The success of the Norwegian Atlantic salmon (*Salmo salar*) farming industry provides an excellent example of the value of initiating genetic analyses at the early stages of a developing aquaculture industry. Selective breeding programs, focused primarily on increasing growth rates, begun in the early phase of the industry (Gjerde 1984; Gjerde and Gjedrem 1984; Standal and Gjerde 1987; Refstie 1987) were a major contributing factor to the Norwegian domination of the international salmon market (Rhodes 1987, 1988). Realized responses to selection for growth rate, a major production trait, have been higher in fish species than those reported for farm animals (Gjerde 1986). Responses in Atlantic salmon breeding, for example, have ranged from a 14% to 30% gain per generation (Gjerde 1986; Kinghorn 1983). These findings suggest that selective breeding of walleye will be of great economic benefit to private aquaculturists.

An efficient regional selective breeding program is needed to obtain broodstock of walleye that will produce progeny adapted to environments that will be used in commercial aquaculture. A regional approach to genetic analyses of walleye populations is needed to ensure that candidate populations from many locations are evaluated in a similar manner. It is imperative, therefore, that the development of a regional selective breeding program begin with a profile of the genetic variation within populations in different geographical areas of the region. Another benefit of a regional approach is that it will bring together the required expertise, including: walleye fry and fingerling culture for strain evaluation (Robert Summerfelt and Bruce Tetzlaff) and genetic analyses (Anne Kapuscinski, Jim Seeb and Lisa Seeb).

Population genetic data (Objective 1) from allozymes and mitochondrial DNA will provide the baseline data needed to recognize and maintain pure strains for aquaculture; they will also provide genetic diversity estimates useful for identifying suitable strains for selective breeding programs. Techniques of quantitative genetics (Objective 2) will be used to evaluate strains for culture and to improve their performance via selective breeding.

**Population Genetic Data on Walleye**

Population genetic data is important to aquacultural development because it permits identification of unique strains or the protection of native populations. These natural gene banks are the future source of brood stock for selection and other aquacultural and genetic manipulations. Information on the relationships among candidate populations is crucial to determine whether or not particular populations from different geographical areas are truly genetically distinct strains. Little data is currently available on the genetics of walleye populations; such data collection is now critical because of the expectation of increased stock transfers with the growing interest in walleye culture.

Additionally, within populations genetic variation provides the basis for the positive gains possible from selective breeding. Conversely, populations with little or no genetic variation have little or no potential for selective gain. Therefore, when starting a selective breeding program, it is important to choose a stock with high genetic diversity. A number of semi-domesticated walleye strains are candidates for broodstock improvement programs in the North Central Region, and our initial genetic analysis will maximize the opportunity for selective gain by providing gene diversity estimates prior to strain selection.

We will use allozyme data derived by protein electrophoresis and restriction fragment analysis of mitochondrial DNA (mtDNA) to profile walleye populations in the North Central Region. The use of two independent lines of evidence will provide an especially powerful approach to discerning relationships between populations and genetic variation within them. As objective 2 develops, and if the walleye (-sauger) hybrid is

proven to be important in the North Central Region, then a similar analyses on sauger populations would be warranted.

Allozyme data are widely acknowledged to be a powerful tool in population studies and are playing an increasingly important role in strain identification, preservation, and selection for aquaculture (e.g., Campton and Utter 1987). Allozyme data can clarify the relationships of discrete stocks, and they can also quantify suspected introgression from stock transfers or hatchery releases. They can provide a basis for genetically marking stocks (Murphy et al. 1983; Seeb et al., in press; Utter and Seeb, in press). Occasionally allozyme marks have also been associated with production characters such as early growth (Allendorf et al. 1983; Liebowitz et al. 1987; Seeb and Miller, in press). Ultimately, as previously stated, within population gene diversity calculated from allozyme frequency data may provide a powerful basis for selection of candidate populations for aquaculture (Allendorf and Ryman 1987).

MtDNA has several unique features that are particularly valuable in the analysis of genetic processes. In higher animals it is maternally inherited (Giles et al. 1980; Brown 1980), and it evolves 5-10 times more rapidly than single copy nuclear DNA, primarily through base substitutions and very small additions and deletions (Brown 1980). Results from restriction fragment analyses of mtDNA indicate that it is an especially sensitive technique for constructing genetic relationships among closely related populations in many different organisms (Avise et al. 1979; Ferris et al. 1981; Lansman et al. l983; Avise and Lansman 1983; Ferris and Berg 1987; Billington and Hebert 1988).

**Quantitative Genetics of Walleye Populations**

The implementation of rationally designed and long-term selective breeding programs into aquaculture operations is an essential means of improving the performance of cultured organisms (Gjedrem 1983; Kinghorn 1983; Tave 1987). The major benefits of selective breeding to commercial aquaculture operations are improvements in product quality and in cost-effectiveness, and increases in harvestable yields and profits. Recently in the North Central Region, the importance of walleye as a food-fish has risen tremendously and a number of private producers and researchers have initiated the development of culture methods for a food-fish product (NCA-23 1987). Yet, virtually no systematic breeding of walleye has been attempted in North America. A major long-term goal of this objective is to establish a North Central Region selective breeding program for walleye.

Initial screening of candidate populations will be done via strain evaluations, i.e., between-strain comparisons of phenotypes for important production traits. Systematic and statistically valid evaluation of walleye strains on a regional basis and for aquaculture purposes is lacking in the North Central Region. We will greatly increase the practical value of initial population screening by including determinations of genotype-environment interactions (Kinghorn 1983). This will make it possible to evaluate the relative suitability of a particular strain varying conditions in different intensive culture facilities (indoor fish culture systems in Iowa versus Illinois) and to different juvenile culture systems (exclusively intensive culture versus tandem extensive fry culture and intensive fingerling culture).

The design of a successful breeding program for any species is dependent on choosing the most suitable populations or "strains" for initiation of the breeding program because it is significantly easier and less expensive to improve a population with an initially good genetic makeup than one with a poor genetic makeup (Kinghorn 1983; Shultz 1986; Tave 1987). Virtually none of the relevant information is currently available to allow reliable choices of the most suitable walleye populations.

After performance evaluations are conducted and one or more suitable populations are identified, the next appropriate step is to estimate genetic parameters (e.g., heritabilities) for important production traits. These parameters are specific to a particular population and are needed to design an efficient selection program. Efficient and precise estimates of genetic parameters are obtainable from half-sib family analysis (Falconer 1981; Tave 1987). Support will be needed beyond the two years of this project proposal to maintain selected strains to sexual maturity and to create half-sib families within one or two populations that are identified to be the most suitable for a regional selective breeding program. By maintaining the separate identity of each family, it will be possible to implement efficient selection schemes (e.g., family selection and index selection), thus leading to the development of selected, pedigreed lines in future installments.

**ATTACHMENT A**

## RELATED CURRENT AND PREVIOUS WORK

### Population Genetic Data on Walleye

Allozyme data may be a powerful tool for discriminating walleye populations. Clayton et al. (1974) examined the population structure of walleye inhabiting lakes within and near Prince Albert National Park, Saskatchewan, Canada. They identified relatively large genetic differences among a number of walleye populations using only one allozyme system, supernatant malate dehydrogenase (MDH, E.C. 1.1.1.37). Some of these populations were only separated by 35 km. MDH allozymes were subsequently used by a number of workers as genetic marks to identify the success of transplanted walleye stocks. Ward and Clayton (1975) and Schweigert et al. (1977) relied on fixed phenotypic differences to successfully evaluate stocking effectiveness in West Blue Lake, Manitoba. Similarly, Murphy et al. (1983) determined that supplemental stocking of juvenile walleyes contributed an average of 67% to the year-class strength in Claytor Lake, Virginia. Paragamian (1988) used both MDH and isocitrate dehydrogenase (IDH, E.C. 1.1.1.42) allozyme data to calculate that stockings of offspring from a lake strain contributed about 19% to the Cedar River, Iowa, walleye population, which was much less than expected; based on this finding he recommended refining the hatchery program.

Genetic marks may have important applications in the development of walleye culture. Fish possessing genetic marks may be selected during the domestication process. The marks are passed from generation to generation, and their use may facilitate the perpetual marking of aquaculturally important strains (Seeb et al., in press; Gharrett and Seeb, in press, reviewed in Utter and Seeb, in press). Additionally, work with other species suggests that the occasional linkages observed between allozyme markers and genes controlling quantitative traits may provide information important for selective improvement (Liebowitz et al. 1987; Seeb 1987).

However, multilocus allozyme studies are necessary to differentiate most populations and provide the diversity estimates that will be of value to fish culturists. Seeb et al. (1981), in a pilot study, resolved 40 allozyme loci in 52 walleye from Lake Erie. Ten loci were polymorphic; the average heterozygosity observed was 0.057, showing that walleye possess an abundance of genetic variation suitable for population structure and gene diversity analyses. Murphy and Lee (1986) only resolved four of ten polymorphic loci they detected in Minnesota populations; yet they observed substantial population differentiation between major watersheds. Billington and Herbert (1988) used 22 endonucleases to study mtDNA variation in ten populations of walleye from the Great Lakes basin. Their data clearly differentiated eastern and western Great Lakes populations and demonstrated the utility of mtDNA analysis in walleye population studies.

Substantial additional study of walleye population structure and genetic diversity has been proposed (David Phillip, Illinois Natural History survey and Anne Kapuscinski, University of Minnesota, personal communication); we have concordant objectives and have initiated communication to eliminate redundancy.

### Quantitative Genetics of Walleye Populations

To the best of our knowledge, no quantitative genetic analyses of walleye populations have been reported in the literature (Ebbers et al. 1988). We are also unaware of any such current investigations by walleye researchers in the U.S. The goal of the first two years of this proposal is to identify strains of walleye that exhibit superior performance (adaptability to extensive and intensive cultural environments and which provide better growth and feed conversion than typical strains) for use in commercial aquaculture. This can reduce the time to reach marketable size and reduced production costs.

To date, quantitative genetic research on aquacultural fish species has focused primarily on rainbow trout (*Salmo gairdneri*), Atlantic salmon (*Salmo salar*), coho salmon (*Oncorhynchus kisutch*), tilapias (*Oreochromis* spp.), channel catfish (*Ictalurus punctatus*), and carp (*Cyprinus carpio*). Studies have been conducted on strain evaluations, genetic parameter estimation, selection, crossbreeding, and inbreeding. Most of the discussion below refers to reviews by Tave (1987), Kapuscinski and Jacobson (1987), and review papers in the proceedings of a World Symposium on Selection, Hybridization and Genetic Engineering (Tiews 1987).

Although not much information is available, performance trials of various strains in a few species have shown that genetic-environmental interactions can influence productivity (e.g., Ayles and Baker 1983; Kinghorn 1983). If similar interactions were to be found in walleye strains, it would become important for walleye farmers to culture the best strain for their particular environments. Numerous studies with other species have shown strain differences in growth, food consumption, disease resistance and heat tolerance.

Previous research has shown that there is great potential for selective breeding in fish. Two major reasons for this are: considerable genetic variance for growth rate and age at maturation has been found in various species; and high selection intensities can be practiced because of the high fecundities in most species (Gjedrem 1983). The opportunity for selective breeding of walleye also should be high due to high fecundities and the great likelihood that levels of genetic variation for production traits are similar to those that have been found in other species. Additionally, realized responses to selection have been higher in fish than those reported in farm animals (Gjerde 1986). Gains in farm animals have been approximately 1% per year, whereas, in salmonids for example, 3-7% gains per year, or 14-30% per generation, from individual and family selection schemes have been reported (e.g., Gjerde 1986; Kinghorn 1983).

Heritability values for a trait reflect the level of additive genetic variance,which is the component of genetic variance most readily exploited in selection programs (Falconer 1981). Although heritabilities greater than 0.1-0.2 are needed for cost-effective individual selection, family selection can be implemented when heritabilities are low (Falconer 1981; Tave 1987). Examples of traits that had relatively high heritabilities in other species (see reviews cited above) and that may be important for selective breeding in walleye include growth rate, age at sexual maturity, and belly wall thickness (e.g., Kinghorn 1983; Gjerde and Gjedrem 1984). Traits that had lower heritabilities, necessitating family selection, and that would be important in walleye selection, include survival and perhaps food conversion. Heritabilities for dressing percentage tend to be low (e.g., Gjerde and Gjedrem 1984). Very little is known about genetic parameters for disease resistance in fish, particularly because it is a very complex trait and meaningful measurement is difficult in genetic experiments (Tiews 1987). Few determinations have been made of genetic correlations between important production traits in fish, but the existing reports suggest the importance of computing correlations in order to ensure the design of successful breeding programs.

## OBJECTIVES

1. Develop baseline information on genetic composition of walleye population for potential use as broodstock.

2. Conduct comparisons of phenotypic characteristics of progeny from selected walleye broodstock.

## PROCEDURES

### Objective 1

Eight populations will be sampled in each of the first two years of study in coordination with private, state, and federal hatcheries in the North Central Region. We will collect 100 individuals from each population; all 100 will be analyzed for allozyme variation, and 20 will be subsampled for mtDNA analysis. Samples will also include the strains evaluated in Objective 1b, i.e., the Iowa semi-domesticated strain (IA), the Ohio domesticated strain (OH) and the Mississippi River (MR) strain collected near Genoa, Wisconsin.

Results from year one will determine the sampling scheme for year two. Samples will be collected from additional populations to increase our understanding of genetic variation in walleye of the North Central Region in areas where the greatest genetic divergence occurs.

Horizontal starch gel electrophoresis will follow Harris and Hopkinson (1976) and Allendorf et al. (1977). Genetic data will be collected from a maximum number of loci, including all those previously detected in walleye (e.g., Seeb et al. 1981; Murphy and Lee 1986; Haas et al. 1988). We will initially perform a thorough screening of buffers and tissues to maximize the number of resolvable loci. Our recent studies have shown that a sequential approach in which additional buffers are used until a locus appears to be adequately resolved is not sufficient. Even small changes in buffer conditions can result in considerable improvement in the resolution of a locus. Thus every system should be screened on a large number of buffer systems. In addition, a large number of tissues should also be screened. Screening multiple tissues will not only result in the largest number of loci, but also in an understanding of the tissue expression patterns. We, therefore, will initially screen six tissues (muscle, liver, eye, heart, kidney, brain) on eight buffers for approximately 60 enzyme systems from one population of walleye. All loci adequately resolved will be analyzed in subsequent populations. Tissue samples will be collected and transported to Carbondale, Illinois on dry ice. Laboratory storage will be at -80ºC.

We will use restriction fragment analysis of mtDNA to estimate both sequence divergence and population relationships. Fresh liver and heart tissue will be collected and stored in a sucrose buffered solution on ice and

transported to the laboratory in Carbondale, Illinois. MtDNA will be isolated and purified in closed-circular form using a cesium chloride/ethidium bromide gradient centrifugation (Lansman et al. 1981). MtDNA will be isolated from the fresh tissues within one week and stored in a 1.1 g CsCl/mL STE solution for subsequent ultracentrifugation. After ultracentrifugation, the cesium chloride and ethidium bromide will be removed by dialysis. Purified mtDNA will be digested with approximately 22 restriction endonucleases following the recommendations of the vendor. Fragments will be radioactively end-labeled with $^{32}$PdXTP using the large fragment of DNA polymerase I (Brown 1980) and separated on agarose gels. Digestion profiles will be visualized by autoradiography (Maniatis et al. 1982).

These end-labeling procedures require the collection of fresh tissue and considerable sample preparation time, but yield highly pure mtDNA as compared to Southern (1975) techniques. These constraints should pose little problem in this study as the specimens will be obtained from midwestern U.S. locations and can be shipped on ice to the laboratory.

Data analysis for both the electrophoretic and mtDNA data sets will be conducted on either an IBM PC/XT or the Southern Illinois University (SIU) mainframe computer. Analysis programs will include BIOSYS-1 (Swofford and Selander 1981), PAUP (D. L. Swofford, Illinois Natural History Survey, Champaign) and PHYLIP (J. Felsenstein, University of Washington, Seattle). We have used all these programs in previous studies (e.g., Wishard et al. 1984).

Each year, sub-samples of fish used in the strain evaluation trials will be electrophoretically genotyped using the same procedures as in Objective 1. Fish will be weighed, measured, and then genotyped for allozyme polymorphisms observed in the parents to test for any potential allozyme genotype-environmental interaction or any heterozygosity-growth interaction.

**Objective 2**

The performances of selected strains (i.e., samples of selected stocks) of walleye with different origins and breeding histories will be evaluated in intensive and extensive culture environments representative of state-of-the-art culture systems. The null hypothesis to be tested is that performance of different populations will be the same when raised in similar cultural environments. The alternate hypothesis is that performance differences will occur among the different evaluated populations. A secondary hypothesis is that the populations will exhibit significant genotype-environment interactions for performance traits, leading to different relative rankings for strain suitability for intensive and extensive cultural environments.

In the first installment of this project performance tests will be done on three populations with different genetic histories. They include: (1) an Iowa strain (IA), a Ohio strain (OH), and a Mississippi River strain (MR). The IA strain is derived from a population native to Spirit Lake, which has been iteratively cultured in a state hatchery from gametes of adults captured in Spirit Lake and then stocked back into the lake as fry or fingerlings. The OH strain is the most domesticated strain available, it consists of 1,200 five-year-old captive broodstock maintained in ponds at the London Fish Hatchery, Madison County, Ohio by Tim O. Nagel of the Ohio Division of Wildlife. These fish are second generation ($F_2$) fish which have been reared their entire life in ponds at the London Fish Hatchery. They are derived from seven year-old parents ($F_1$) that were produced by crossing parents from Lake Erie and the Mosquito Reservoir, Trumbill County in northeast Ohio. The $F_2$ OH strain are well-adapted to the hatchery environment as evident by observations of Nagel that the pond-reared fingerlings from these stocks are more easily converted to formulated feed than offspring of wild stocks. The MR strain will be obtained from the Genoa National Fish Hatchery, Genoa, Wisconsin, near Lock and Dam Number 8 on the Upper Mississippi River. The MR strain is included for comparison to the other strains because it should be the least domesticated and genetically the most heterozygous of the three strains.

In the second year, performance evaluations will be conducted on one or more of the same strains and additional selected populations identified in the genetic analysis of walleye populations in the region. Subsamples of the strains will be maintained by SIU for use in future breeding experiments. After these fish reach maturation, and in collaboration with guidance from University of Minnesta (U MINN), SIU will breed one or more of the strains, selected on the basis of performance characteristics, in a nested mating design (Becker 1984). This mating design will allow us to make statistically reliable estimates of heritabilities and genetic correlations for juvenile traits of the progeny (Falconer 1981).

Strain evaluations will include both intensive and extensive (pond) culture followed by continued maintenance of the intensive cultured fish in tanks and transfer of pond-reared fish to tank culture for training to

formulated feed (Figure 1). The fry stage is defined here as the interval between hatching and 30 d posthatch. However, we propose running the intensive fry culture experiments for 100 d and the pond culture rearing period for 60 d before transferring the pond-reared fingerlings to tanks for training to formulated feed under intensive culture environment.

Fish of all three strains will be reared at both SIU and Iowa State University (ISU): (1) one site for intensive fry culture (ISU); (2) one site for pond culture of fry to fingerlings (SIU) and (3) two indoor sites for training pond-reared fingerlings to formulated feed (ISU and SIU). Pond reared fingerlings from Southern Illinois University will be trained to formulated feeds in indoor troughs at the SIU (Figure 1). The practice of tandem extensive fry rearing and intensive fingerling rearing, as proposed for the latter two sites, is currently the most reliable methodology for juvenile walleye culture (reviewed by Colesante et al. 1986; Nickum 1986).

Procedures for strain evaluations are outlined in Figure 1. Each full-sib group of fertilized eggs will be divided and equally distributed into two hatching jars so that the parentage of the pooled embryos will be similar between jars. Upon hatching of the fry, each of the two hatching jars will be assigned to a different culture site (i.e., SIU, ISU). This means that each site will receive fry coming from three jars, one for each strain. At each site, fry from each jar will be evenly divided among the replicate rearing units assigned to the strain.

The purely intensive culture portions of strain evaluations will be done at ISU. ISU will use three replicate tanks per strain at each site (i.e., a total of nine tanks). The extensive fry culture will be done at SIU using two replicate ponds per strain (a total of six ponds). The training of pond-reared fish to formulated feed will be done at SIU, using three replicate indoor tanks per strain (a total of nine tanks per site) at each of two temperatures (20 and 25$^{o}$C).

At all sites, fertilized eggs will be incubated in hatchery jars according to standard procedures (Nickum 1978, 1986). Intensive culture of fry on formulated feed will be standardized at the intensive culture sites using protocols developed by Dr. Summerfelt in his current research at ISU (Summerfelt 1988, 1989). At ISU, fry are reared in tanks of similar volumes at a density of 20 to 50 fry per liter; larvae are counted by volumetric displacement. Water quality and current is maintained with a complete exchange of ounce/h. Water temperature will be 15$^{o}$C for the first 5 d posthatch, then gradually incremented to 20$^{o}$C by 15 d and 20-23$^{o}$C from 30-100 d. Water flow rates to the rearing tanks will be kept the same and will be adjusted to maintain calculated un-ionized ammonia below 0.001 mg/L. Gas pressures (total, $\Delta$P, and partial pressures of oxygen and nitrogen), dissolved oxygen, pH, alkalinity and total ammonia-nitrogen will be measured daily using a saturometer (Common Sensing Inc., Bainbridge Island, Washington), dissolved-oxygen probe and standard analytical procedures for pH, alkalinity and ammonia-nitrogen as given by APHA et al. (1985).

A distinctive feature of the intensive rearing will be the exclusive use of the "Fry Feed Kyowa," Series B diets (Kyowa Hakko Kogyo Company, Ltd., Japan). Three sizes are used: B-250, B-400 and B-700. The particle size of this feed, as given by the manufacturer, is as follows: B-250, less than 250 µm; B-250, 250-400 µm, and B-700, 400-700 µm. Feed is presented every 3.0 min on a 24 h basis (480 feedings/d ) for the first 30 d, then at 5 min intervals to 60 d and then at 10 min intervals until 120 d. Vibrator type feeders (Model AF6, Sweeney Enterprises, Inc., Boerne, TX) are used to dispense the feed by electronic time-clock. Feeding begins when the fry are 3-d posthatch. The total feed for each day is apportioned by the number of feedings per day.

Experience indicates the importance of frequent upgrading of feed size to accommodate energetics of food capture and processing; failure to increment feed size results in cannibalism. The initial daily amount of food of 10 mg/fish is incremented by 2 mg/fish/d on a regular schedule. These amounts are in excess of the bioenergetic needs of the fish but they are required because the fish consume the feed only while it remains in suspension, and not after it sinks. Thus, only a fraction of the feed is consumed and excessive feeding is necessary to maintain feed density. An upwelling current strong enough to maintain the feed in suspension is too turbulent for the fry, therefore, the feeding needs to be frequent to make feed available continuously.

During the fry rearing interval, waste feed and dead fish are siphoned from the bottom of the tanks daily and counts of dead fish are made to relate daily mortality to fish age and environmental conditions. Waste feed is separated from the dead fish using a number 30 standard sieve with 600 µm (0.0234 inch) openings; the sieve retains the fish but passes the waste feed. Survival over the fry rearing phase (first 30 d) is determined from a final count of fish in the tank at the end of 30 d. At the end of the experiment, a sample of 25 fish from each tank

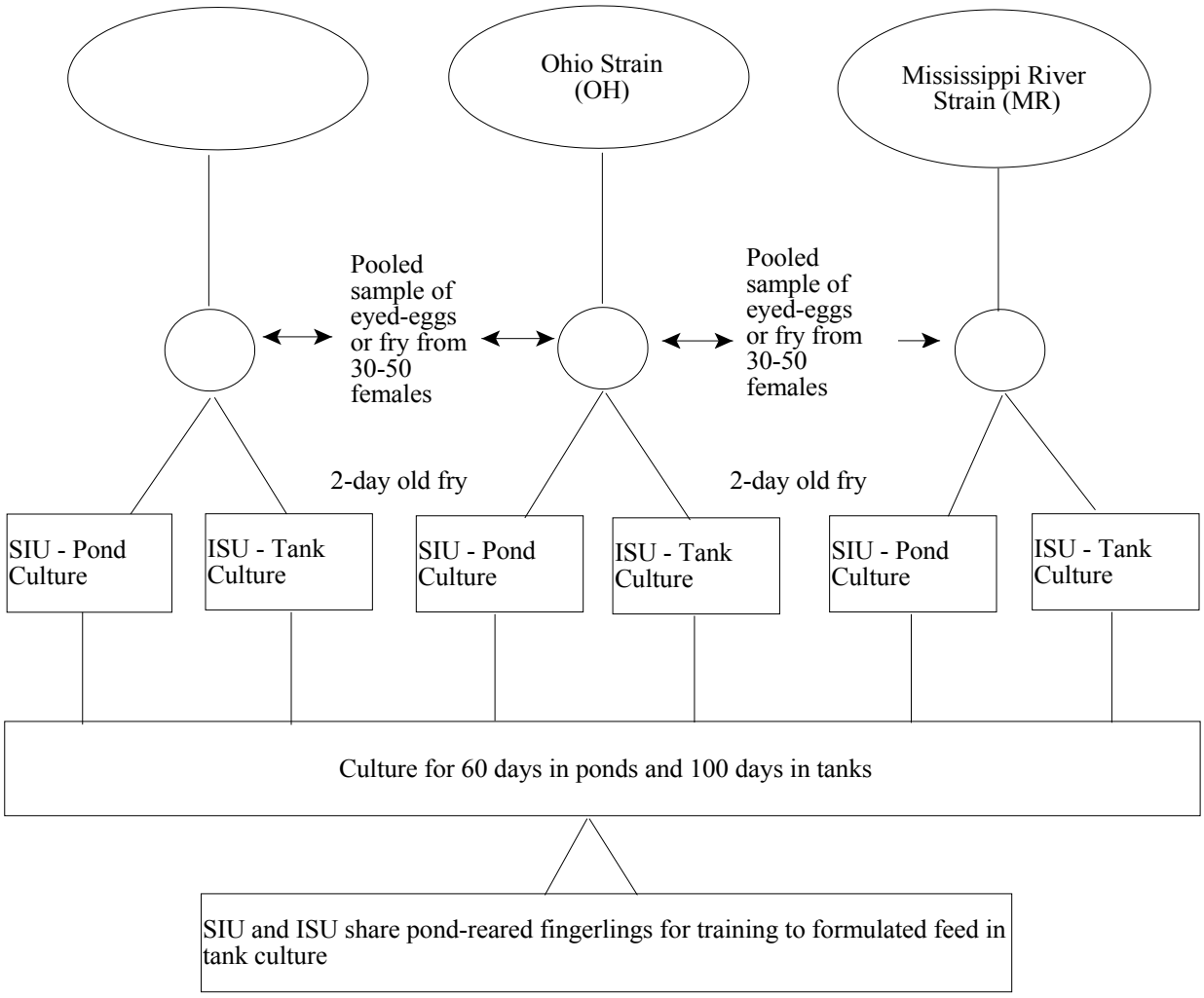


Figure 1. Figure 1.Experimental design for Objective 2 showing collaboration between Southern Illinois University (SIU) and Iowa State University (ISU) for conducting performance evaluation of three strains of walleye fry in extensive (SIU) and intensive (ISU) systems.

is obtained to measure total length and occurrence of an inflated gas bladder. Gas bladder inflation is observed under a dissecting microscope with transmitted light.

Fingerling culture practices for the tank reared fingerlings begin at 30 d. The fingerlings (about 15- to 20-mm) will be transferred from the fry rearing facility to a fingerling facility to free the fry culture facilities for additional groups of fish and also to accommodate the different environmental requirements (light cycle and intensity, stock density, feeding rates) of the fry and fingerlings. Initial stock density for rearing of the fingerlings is one fish per liter. Maximum room light levels for fingerling culture are 25 lux but the tanks are screened which reduces light about 50% over the water (10-15 lux maximum over the water surface). The diurnal light regime (room lighting) is 18 h light and 6 h dark for the last 70 d of fingerling rearing. Densities for intensive culture of fingerlings will start at 1 fish per liter in indoor tanks through 100 d or a length of about 100 mm. Feeding schedules for fingerlings are based on biomass of fish, beginning with 14%/d, adjusted down by 2% every 10 d, to 6%/d by 100 d. Feed is presented at 5 min intervals to 60 d and then at 10 min intervals until 100 d.

Procedures for extensive (i.e., pond culture) fry culture will be standardized. Fry stocking density will be 247,000 fry per hectare (100,000/acre) and the ponds fertilized with alfalfa pellets. Fish will be reared in the ponds until 60 d posthatch when they are expected to be a size of 35 to 40 mm total length. At harvest, fish will be counted and weighed to determine survival, number/kg (no/lb), average length, weight and condition factor. After grading, subsamples will be transferred to indoor tanks (at both ISU and SIU) where performance trials will be conducted to compare the ease at which the fish can be trained to feed on a formulated feed -- Biotrainer diet will be used for training then the fingerlings shifted to Biodry feed (both Biotrainer and Biodry are registered trademarks of Bioproducts Incorporated, Warrenton, Oregon). Pond-reared fingerlings will be cultured intensively for 100 d. Performance differences between strains should become readily apparent during this interval of intensive rearing.

Samples of fingerlings from the selected strains, from both intensive and extensive-intensive culture experiments, will be reared by Southern Illinois University for use in future selective breeding studies.

A number of performance traits will be measured, including but not limited to: percent hatch, percent gas bladder inflation, percent survival to 30 d and to 100 d, length at age, weight at age, condition factor at age, and growth rate. Individual samples for trait evaluation will be taken at 30 d, and 100 d posthatch. Measurements of appropriate traits (e.g., growth and food conversion) will be extended beyond 100 d. This will allow genetic parameter estimation for more advanced life stages. Dr. Kapuscinski (U MINN) will provide advice on collection of data on performance traits, to ensure their suitability for quantitative genetic analyses.

For each evaluated trait, i.e., estimates of the variance between strains, the variance between specific culture environments, and the variance due to interaction of strains with environments will be obtained by analysis of variance in a two-way classification of strains x environments (Falconer 1981; Snedecor and Cochran 1967). Dr. Kapuscinski (U MINN) will provide consultation for these analyses.

## FACILITIES

### Objective 1

The Genetics Lab in the Cooperative Fisheries Research Laboratory, Southern Illinois University, Carbondale, is equipped to perform allozyme analyses, mitochondrial DNA analyses, induced polyploidy experiments, and chromosomal studies. Major equipment in the SIU Genetics Lab includes a Beckman L7-65 ultracentrifuge with vertical rotor; Beckman high speed J-21 centrifuge and rotor; Beckman refrigerated table-top centrifuge; microfuge; 11 power supplies with constant current and constant voltage capabilities; two 0.55 cubic meter -80$^{o}$C freezers; automatic gel cooling systems with capacity for 4 gels at -20$^{o}$C; Gene-Vac chemical vacuum pump; gel dryer; computer interface to analytical balance, digitizer pad, and pit tag reader; and three IBM compatible personal computers with installed BIOSYS-1, PAUP, and PHYLIP genetic analysis software. Miscellaneous rearing systems will be used as necessary. The Genetics Lab has a controlled temperature environmental chamber with two recirculating water systems driving 20 aquaria. Other raceways and tanks are available in the Cooperative Fisheries Research Laboratory's 650.3 square meter wet lab.

**Objective 2**

The on-campus aquaculture facilities at Iowa State University are located in the Sciences 2 Building. Two rooms are used for walleye culture and a third room is used as an analytical laboratory. The latter is equipped with a hood, chemical sink, BOD incubator, and bench space for water chemistry, and microscopic analysis. One of the wet labs has 4,480 L tanks, each with 4-120 L rearing spaces. The tanks can be supplied with either compressed air or pure oxygen and the rearing system operated single-pass or recycle. The other, larger wet lab, has 6-154 L square tanks, 6-185 L cylindrical tanks, and 1,480 L tank with 4-120 L rearing spaces. The tanks may be operated in single-pass or recycle modes. This room also has bench space and a hood for analytical work. Both wet labs are supplied with dechlorinated (activated charcoal) tap water, which is also treated with sodium sulfite from a chemical pump to eliminate residual free chlorine and chloramines. Excess nitrogen is reduced to safe levels by passing all incoming water through a column containing glitch rings for degassing. Refrigerators/freezers are available in each wet lab for storage of feed and reagents.

Southern Illinois University (SIU) has 18-0.058 ha drainable earthen ponds for aquaculture research. Three of these ponds will be used in this study. In addition, SIU aquaculture faculty have access to 17 ha of additional drainable ponds at the LaSalle Aquaculture facility. Three 0.73 ha ponds at this facility will be used in this study. The LaSalle facility also has a wet laboratory containing numerous fish rearing tanks, including a 12,000-L, 6-tank recirculating system and a 9,000-L raceway system designed specifically for training fingerling fish to accept commercial feeds.

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**ATTACHMENT A**

**PROJECT LEADERS**

| State | Name/Institution | Area of Specialization |
|---|---|---|
| **Illinois** | James E. Seeb<br>Southern Illinois University | Molecular Genetics |
| | Lisa W. Seeb<br>Southern Illinois University | Molecular Genetics |
| | Bruce L Tetzlaff<br>Southern Illinois University | Finfish Aquaculture |
| **Iowa** | Robert C. Summerfelt<br>Iowa State University | Finfish Aquaculture/<br>Fish Larval Fish Culture |
| **Minnesota** | Anne R. Kapuscinski<br>University of Minnesota | Quantitative Genetics/<br>Breeding |

**ATTACHMENT A**

**INDIVIDUAL BUDGETS FOR PARTICIPATING INSTITUTIONS**

**Illinois**

Southern Illinois University
James E. Seeb
Lisa W. Seeb
Bruce L. Tetzlaff

**Iowa**

Iowa State University
Robert C. Summerfelt

**Minnesota**

University of Minnesota
Anne R. Kapuscinski

**ATTACHMENT A**

**PROPOSED WALLEYE BUDGET FOR SOUTHERN ILLINOIS UNIVERSITY**

**(J. Seeb and L. Seeb)**

**Objective 1**

| | Year 1 | | Year 2 | | Year 1 | Year 2 |
|---|---|---|---|---|---|---|
| A. Salaries and Wages | No. | FTEs | No. | FTEs | | |
| 1. No. of Senior Personnel & FTEs[1] | | | | | | |
| a. (Co)-PI(s) | 2 | 0.43 | 2 | 0.43 | $6,351 | $6,954 |
| b. Senior Associates | | | | | | |
| 2. No. of Other Personnel (Non-Faculty) & FTEs | | | | | | |
| a. Research Assoc./Postdoc | | | | | | |
| b. Other Professionals | | | | | | |
| c. Graduate Students | 1 | 0.50 | 1 | 0.50 | $9,778 | $10,266 |
| d. Prebaccalaureate Students | | | | | | |
| e. Secretarial-Clerical | | | | | | |
| f. Technical, Shop, and Other | | | | | | |
| **Total Salaries and Wages** | | | | | $16,129 | $17,220 |
| B. Fringe Benefits | | | | | $0 | $0 |
| C. **Total Salaries, Wages and Fringe Benefits** | | | | | $16,129 | $17,220 |
| D. Nonexpendable Equipment | | | | | $3,900 | $0 |
| E. Materials and Supplies | | | | | $5,000 | $5,000 |
| F. Travel - Domestic (*Including Canada*) | | | | | $1,800 | $1,800 |
| G. Other Direct Costs | | | | | $1,200 | $1,200 |
| **TOTAL PROJECT COSTS PER YEAR** (*C through G*) | | | | | $28,029 | $25,220 |
| **TOTAL PROJECT COSTS** | | | | | $53,249 | |

[1]FTEs = Full Time Equivalents based on 12 months.

**ATTACHMENT A**

**PROPOSED WALLEYE BUDGET FOR SOUTHERN ILLINOIS UNIVERSITY**

**(Tetzlaff)**

**Objective 2**

| | Year 1 | | Year 2 | | Year 1 | Year 2 |
|---|---|---|---|---|---|---|
| A. Salaries and Wages | No. | FTEs | No. | FTEs | | |
| 1. No. of Senior Personnel & FTEs[1] | | | | | | |
| a. (Co)-PI(s) | 1 | 0.05 | 1 | 0.05 | $0 | $0 |
| b. Senior Associates | | | | | | |
| 2. No. of Other Personnel (Non-Faculty) & FTEs | | | | | | |
| a. Research Assoc./Postdoc | | | | | | |
| b. Other Professionals | | | | | | |
| c. Graduate Students | 1 | 0.25 | 1 | 0.25 | $5,815 | $6,280 |
| d. Prebaccalaureate Students | | | | | | |
| e. Secretarial-Clerical | | | | | | |
| f. Technical, Shop, and Other | | | | | | |
| **Total Salaries and Wages** | | | | | $5,815 | $6,280 |
| B. Fringe Benefits | | | | | $0 | $0 |
| C. **Total Salaries, Wages and Fringe Benefits** | | | | | $5,815 | $6,280 |
| D. Nonexpendable Equipment | | | | | $0 | $0 |
| E. Materials and Supplies | | | | | $1,200 | $900 |
| F. Travel - Domestic (*Including Canada*) | | | | | $500 | $500 |
| G. Other Direct Costs | | | | | $650 | $750 |
| **TOTAL PROJECT COSTS PER YEAR** (*C through G*) | | | | | $8,165 | $8,430 |
| **TOTAL PROJECT COSTS** | | | | | $16,595 | |

[1]FTEs = Full Time Equivalents based on 12 months.

**ATTACHMENT A**

**PROPOSED WALLEYE BUDGET FOR IOWA STATE UNIVERSITY**

**(Summerfelt)**

**Objective 2**

| | Year 1 No. | Year 1 FTEs | Year 2 No. | Year 2 FTEs | Year 1 | Year 2 |
|---|---|---|---|---|---|---|
| A. Salaries and Wages | | | | | | |
| 1. No. of Senior Personnel & FTEs[1] | | | | | | |
| a. (Co)-PI(s) | 1 | 0.05 | 1 | 0.05 | $0 | $0 |
| b. Senior Associates | | | | | | |
| 2. No. of Other Personnel (Non-Faculty) & FTEs | | | | | | |
| a. Research Assoc./Postdoc | | | | | | |
| b. Other Professionals | | | | | | |
| c. Graduate Students | 1 | 0.50 | 1 | 0.50 | $11,970 | $12,568 |
| d. Prebaccalaureate Students | | 0.10 | | 0.10 | $1,000 | $1,100 |
| e. Secretarial-Clerical | | | | | | |
| f. Technical, Shop, and Other | | | | | | |
| **Total Salaries and Wages** | | | | | $12,970 | $13,668 |
| B. Fringe Benefits | | | | | $300 | $300 |
| C. **Total Salaries, Wages and Fringe Benefits** | | | | | $13,270 | $13,968 |
| D. Nonexpendable Equipment | | | | | $5,800 | $1,800 |
| E. Materials and Supplies | | | | | $800 | $900 |
| F. Travel - Domestic (*Including Canada*) | | | | | $550 | $625 |
| G. Other Direct Costs | | | | | $300 | $400 |
| **TOTAL PROJECT COSTS PER YEAR** (*C through G*) | | | | | $20,720 | $17,693 |
| **TOTAL PROJECT COSTS** | | | | | $38,413 | |

[1]FTEs = Full Time Equivalents based on 12 months.

**ATTACHMENT A**

**PROPOSED WALLEYE BUDGET FOR UNIVERSITY OF MINNESOTA**

**(Kapuscinski)**

**Objective 2**

| | Year 1 | | Year 2 | | Year 1 | Year 2 |
|---|---|---|---|---|---|---|
| A. Salaries and Wages | No. | FTEs | No. | FTEs | | |
| 1. No. of Senior Personnel & FTEs[1] | | | | | | |
| a. (Co)-PI(s) | 1 | 0.02 | 1 | 0.02 | $0 | $0 |
| b. Senior Associates | | | | | | |
| 2. No. of Other Personnel (Non-Faculty) & FTEs | | | | | | |
| a. Research Assoc./Postdoc | | | | | | |
| b. Other Professionals | | | | | | |
| c. Graduate Students | | | | | | |
| d. Prebaccalaureate Students | | | | | | |
| e. Secretarial-Clerical | | | | | | |
| f. Technical, Shop, and Other | | | | | | |
| **Total Salaries and Wages** | | | | | $0 | $0 |
| B. Fringe Benefits | | | | | $0 | $0 |
| C. **Total Salaries, Wages and Fringe Benefits** | | | | | $0 | $0 |
| D. Nonexpendable Equipment | | | | | $0 | $0 |
| E. Materials and Supplies | | | | | $0 | $0 |
| F. Travel - Domestic (*Including Canada*) | | | | | $1,500 | $1,500 |
| G. Other Direct Costs | | | | | $200 | $200 |
| **TOTAL PROJECT COSTS PER YEAR** (*C through G*) | | | | | $1,700 | $1,700 |
| **TOTAL PROJECT COSTS** | | | | | $3,400 | |

[1]FTEs = Full Time Equivalents based on 12 months.

**ATTACHMENT A**

**CULTURE TECHNOLOGY OF WALLEYE**

Budget Summary for Each Participating Institution at 58.6K for the First Year

| | SIU | ISU | U MINN | TOTALS |
|---|---|---|---|---|
| Salaries and Wages | $22,109 | $12,970 | $0 | $35,079 |
| Fringe Benefits | $1,257 | $300 | $0 | $1,557 |
| **Total Salaries, Wages and Benefits** | **$23,366** | **$13,270** | **$0** | **$36,636** |
| Nonexpendable Equipment | $3,900 | $5,800 | $0 | $9,700 |
| Materials and Supplies | $5,178 | $800 | $0 | $5,978 |
| Travel | $2,300 | $550 | $1,500 | $4,350 |
| Other Direct Costs | $1,450 | $300 | $200 | $1,950 |
| **TOTAL PROJECT COSTS** | **$36,194** | **$20,720** | **$1,700** | **$58,614** |

Budget Summary for Each Participating Institution at 53.0K for the Second Year

| | SIU | ISU | U MINN | TOTALS |
|---|---|---|---|---|
| Salaries and Wages | $23,418 | $13,668 | $0 | $37,086 |
| Fringe Benefits | $1,294 | $300 | $0 | $1,594 |
| **Total Salaries, Wages and Benefits** | **$24,712** | **$13,968** | **$0** | **$38,680** |
| Nonexpendable Equipment | $0 | $1,800 | $0 | $1,800 |
| Materials and Supplies | $5,048 | $900 | $0 | $5,948 |
| Travel | $2,300 | $625 | $1,500 | $4,425 |
| Other Direct Costs | $1,590 | $400 | $200 | $2,190 |
| **TOTAL PROJECT COSTS** | **$33,650** | **$17,693** | **$1,700** | **$53,043** |

**ATTACHMENT A**

**RESOURCE COMMITMENT FROM INSTITUTIONS[1]**

(Salaries, Supplies, Expenses and Equipment)

| Institution/Item | | Year 1 | Year 2 |
|---|---|---|---|
| **Southern Illinois University** | | | |
| Salaries and Benefits: SY @ 0.05 FTE | | $1,162 | $1,710 |
| TY @ 0.10 FTE | | $2,500 | $2,650 |
| | | $8,850 | $8,850 |
| | TOTAL PER YEAR | **$12,962** | **$13,260** |
| **Iowa State University** | | | |
| Salaries and Benefits: SY @ 0.05 FTE | | $3,928 | $4,203 |
| Supplies, Expenses and Equipment: | | $6,068 | $6,457 |
| | TOTAL PER YEAR | **$,9996** | **$10,660** |
| **University of Minnesota** | | | |
| Salaries and Benefits: SY @ 0.02 FTE | | $1,192 | $1,192 |
| Supplies, Expenses and Equipment: | | $748 | $748 |
| | TOTAL PER YEAR | **$1,940** | **$2,011** |
| | **GRAND TOTAL** | **$24,898** | **$25,931** |

[1]Since cost sharing is not a legal requirement and due to the difficulty in accounting for small items, documentation will not be maintained.

**ATTACHMENT A**

## SCHEDULE FOR COMPLETION OF OBJECTIVES

Objective 1: Initiated in year 1 and continued in year 2.[1]

Objective 2: Initiated in year 1 and continued in year 2.[1]

[1] Significant progress will be made on both objectives during Years 1 and 2. However, a third year of effort will probably be needed to fully complete the research proposed and thus yield maximum benefits.

**ATTACHMENT A**

**LIST OF PRINCIPAL INVESTIGATORS**

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**James E. Seeb,** Southern Illinois University

**Lisa W. Seeb,** Southern Illinois University

**Robert C. Summerfelt,** Iowa State University

**Bruce L. Tetzlaff,** Southern Illinois University

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**EDUCATION**

B.A. Swarthmore College 1976
M.S. Oregon State University 1980
Ph.D. Oregon State University 1984

**POSITIONS**

Associate Professor/Extension Specialist (Aquaculture), University of Minnesota (1989-Present)
Assistant Professor/Extension Specialist (Aquaculture), University of Minnesota (1984-1989)
Instructor/Project Leader/Research Assistant, Oregon State University (1980-1984)
Research Assistant, Oregon State University (1977-1980)
Aquaculture Research Technician, Weyerhaeuser Company (1976-1977)

**SCIENTIFIC AND PROFESSIONAL ORGANIZATIONS**

American Fisheries Society: Fish Culture Section, Genetics Section, NCD Fish Genetics Technical Committee
Genetics Society of America
International Association of Genetics in Aquaculture (Charter Member)
Society for the Study of Evolution
World Aquaculture Society
Sigma Xi, Phi Kappa Phi, Phi Sigma, Gamma Sigma Delta

**SELECTED PUBLICATIONS**

Yoon, S.J., E.M. Hallerman, M.L. Gross, Z. Liu, J.F. Schneider, A.J. Faras., P.B. Hackett, A.R. Kapuscinski, and K.S. Guise. In press. Transfer of the gene for neomycin resistance into goldfish (*Carrassius auratus*). Aquaculture

Kapuscinski, A.R. In Press. Integration of Transgenic Fish into Aquaculture. Food Reviews International (Invited Paper)

Hallerman, E.M., J.F. Schneider, M.L. Gross, A.J. Faras, P.B. Hackett, K.S. Guise, and A.R. Kapuscinski. 1989. Enzymatic dechorionation of goldfish, walleye and northern pike eggs. Transactions of the American Fisheries Society 117:456-460.

Phillips, R.B., and A.R. Kapuscinski. 1988. High frequency of translocation heterozygotes in odd year populations of pink salmon (*Oncorhynchus gorbuscha*). Cytogenetics and Cell Genetics 48:178-182.

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**EDUCATION**

B.S. University of Puget Sound 1974
M.S. University of Washington 1982
Ph.D. University of Washington 1987

**POSITIONS**

Assistant Professor, Southern Illinois University-Carbondale (1988-present)
Research Assistant Professor, University of Idaho, Moscow (1987-1988)
Graduate Assistant, University of Washington, Seattle (1982-1986)
Fish Biologist, Washington Department of Fisheries, Olympia (1978-1980)
Fish Biologist, Pacific Fisheries Research, Seattle (1976-1978, 1980-1982)

**SCIENTIFIC AND PROFESSIONAL ORGANIZATIONS**

American Fisheries Society
American Genetics Association
American Society of Ichthyologists and Herpetologists
Genetics Society of America
International Association for Genetics in Aquaculture
Sigma Xi

**SELECTED PUBLICATIONS**

Seeb, J.E., and G.D. Miller. In press. The integration of alloenzyme analyses and genomic manipulations for fish culture and management. *In* D.H. Whitmore, editor. Application of electrophoresis and isoelectric focusing techniques in fisheries management. CRC Press, Boca Raton, Florida.

Seeb, J.E., F.M. Utter, and G.H. Thorgaard. 1988. Survival and alloenzyme expression in diploid and triploid hybrids between chum, chinook, and coho salmon. Aquaculture 72:31-48.

Seeb, J.E., L.W. Seeb, D.W. Oates, and F.M. Utter. 1987. Genetic variation and postglacial dispersal of populations of northern pike (*Esox lucius*) in North America. Canadian Journal of Fisheries and Aquatic Sciences 44:556-561.

Scheerer, P.D., G.H. Thorgaard, and J.E. Seeb. 1987. Performance and developmental stability of triploid tiger trout (brown trout x brook trout male). Transactions of the American Fisheries Society 116:92-97.

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**EDUCATION**

A.B. University of California-Berkeley 1973
M.A. University of Montana 1977
Ph.D. University of Washington 1986

**POSITIONS**

Research Assistant Professor, Southern Illinois University-Carbondale (1988-present)
Research Assistant Professor, University of Idaho, Moscow (1984-1988)
Graduate Assistant, University of Washington, Seattle (1982-1988)
Fish Biologist, Pacific Fisheries Research, Olympia, WA (1978-1981)
Fish Geneticist, National Marine Fisheries Service, Seattle (1977-1979)

**SCIENTIFIC AND PROFESSIONAL ORGANIZATIONS**

American Fisheries Society
American Society of Ichthyologists and Herpetologists
Genetics Society of America
Society for the Study of Evolution
Sigma Xi.

**SELECTED PUBLICATIONS**

Seeb, L.W., J.E. Seeb, R.L. Allen, and W.K. Hershberger. 1990. Evaluation of adult returns of genetically marked chum salmon with suggested future applications. American Fisheries Society Symposium 7:418-425.

Seeb, L.W., and D.R. Gunderson. 1988. Genetic variation and population structure of Pacific ocean perch (*Sebastes alutus*). Canadian Journal of Fisheries and Aquatic Sciences 45:78-88. Copeia 1984(1):120-132.

Seeb, J.E., and L.W. Seeb. 1986. Gene mapping of isozyme loci in chum salmon (*Oncorhynchus keta*). Journal of Heredity 77:399-402.

Seeb, J.E., L.W. Seeb, and F.M. Utter. 1986. Use of genetic marks to assess stock dynamics and management programs for chum salmon. Transactions of the American Fisheries Society 115:448-454.

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**EDUCATION**

B.S. University of Wisconsin-Stevens Point 1957
M.S. Southern Illinois University 1959
M.S. Duke University Marine Laboratory 1962
Ph.D. Southern Illinois University 1964

**POSITIONS**

Professor, Department of Animal Ecology, and Associate Director of the North Central Regional Aquaculture Center, Iowa State University (1988-Present)
Professor, Department of Animal Ecology, Iowa State University (1985-1988)
Professor and Chairman, Department of Animal Ecology, Iowa State University (1976-1985)
Leader (Fishery Research Biologist, U.S. Fish and Wildlife Service, GS-13), Oklahoma Cooperative Fishery Research Unit, Oklahoma State University (1966-1976)
Assistant Professor, Department of Zoology, Kansas State University (1964-1966)
Lecturer, Department of Zoology, Southern Illinois University, Carbondale (1962-1964)
Visiting Professor: Utah State Universuty (1983), Oregon Institute of Marine Biology (1975), and Southern Illinois University (1965)

**SCIENTIFIC AND PROFESSIONAL ORGANIZATIONS**

American Fisheries Society: Fish Culture, Fish Health (Charter member), Education (Charter member), Bioengineering, Computer User, and Fisheries Management Sections; Iowa Chapter
American Institute of Fishery Research Biologists (Fellow)
Fisheries Society of the British Isles
Iowa Academy of Sciences
North American Lake Management Society
Societas Internationalis Limnologiae
Honorary: Sigma Xi, Phi Kappa Phi, Gamma Sigma Delta, Sigma Zeta

**SELECTED PUBLICATIONS**

Summerfelt, R.C., and L.S. Smith. In Press. Anesthesia and surgery. Chapter 8 *in* C.B. Schreck and P. Moyle, editors. Methods for fish biology. American Fisheries Society, Bethesda, MD.

Summerfelt, R.C. 1988. Effect of oxygen saturation levels on swim bladder inflation of larval walleye. Iowa Department of Natural Resources, Federal Aid in Fish Restoration, F-121-R, Job Number 1, Des Moines.

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**EDUCATION**

B.S. Southern Illinois University at Carbondale 1974
M.A. Southern Illinois University at Carbondale 1979

**POSITIONS**

Research Project Director, Fisheries Research Laboratory, Southern Illinois University-Carbondale, Illinois (1980-present)
Researcher, Fisheries Research Laboratory, Southern Illinois University (1976-1980)

**SCIENTIFIC AND PROFESSIONAL ORGANIZATIONS**

American Fisheries Society: Fish Culture Section, Bioengineering Section, Fish Management Section

**SELECTED PUBLICATIONS**

Heidinger, R.C., J.H. Waddell, and B.L. Tetzlaff. 1985. Relative survival of walleye fry versus fingerlings in two Illinois reservoirs. Proceedings of the Annual Conference, Southeast Association of Fisheries and Wildlife Agencies 39:306-311.

Miller, S.J., and B.L. Tetzlaff. 1985. Daily growth increments in otoliths of larval walleye (*Stizostedion vitreum*). Transactions of the Illinois Academy of Science 78:115-120.

Heidinger, R.C., B.L. Tetzlaff, and J. Stoeckel. 1985. Evidence of two feeding subpopulations of white crappie (*Pomoxis annularis*) in Rend Lake, Illinois. Journal of Freshwater Ecology 3:133-143.

Heidinger, R.C., L.J. Wawronwicz, and B.L. Tetzlaff. 1983. Applications of water re-use technology for overwintering threadfin shad (*Dorosoma petenense*) at northern latitudes. Aquaculture Engineering 2:153-162.

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