OHRC Research Plan

November 2014

Introduction

The use of hatchery fish to provide fish for fisheries has been practiced in Oregon since the late 1800's. Hatchery salmon and steelhead programs in place today are vital to many Oregon fisheries and have helped restore and maintain natural production in some areas. Research over the last three decades has increasingly indicated that hatchery fish can also be detrimental to wild fish productivity. The challenge for today's fishery managers is how to appropriately manage hatchery salmon and steelhead programs to provide the benefits they provide while also ensuring they have minimal negative impact to the wild populations they interact with.

The Oregon Hatchery Research Center (OHRC) opened in October 2005 with the express purpose of helping fishery managers understand and manage the interactions between hatchery and wild fish. The current OHRC Mission is "to be an internationally-recognized leader in fisheries science, specializing in defining the mechanisms that may create differences between hatchery and wild salmonids, recommending management strategies to manage those differences while meeting fishery and conservation objectives, and educating Oregonians on the role and performance of hatcheries in supporting and protecting Oregon's native fish and fisheries." To achieve this mission, three goals guide the center's work:

1. Understand mechanisms that may create differences between hatchery and wild fish
2. Develop approaches to manage hatchery fish that conserve and protect native fish
3. Educate and train students, fishery biologists, managers and the public on the relationship between hatchery and wild fish, the connection between fish and watershed, estuarine and ocean systems, and the implications for fish management and stewardship.

The OHRC is an Oregon Department of Fish and Wildlife (ODFW) facility operated under a Memorandum of Understanding (MOU) between ODFW and the Fisheries and Wildlife Department of Oregon State University (OSU). The OHRC is intended to serve as a clearing house for all research in Oregon related to the interactions between hatchery and wild fish. The facilities at the OHRC have been, and will continue to be used to conduct such research, but may not be the primary location for other related research activities. The OHRC Advisory Board and the scientists associated with the Center will encourage researchers to use the OHRC as a sounding board for research proposals and will seek to collaborate with researchers addressing issues consistent with the OHRC Mission.

Addressing Goal 3 of the OHRC Mission

Information collected in the past from research conducted through the OHRC has been widely disseminated to the scientific community as well as the public, and has been applied to the management and conservation of hatchery and wild fish where appropriate. Numerous peer-reviewed, scientific papers related to research conducted through the OHRC have been published in journals and symposia, and news releases and displays at the OHRC have shared this information with the public. Several changes to hatchery operations have been implemented, or are being considered for implementation based on the outcome of previous OHRC research. These include:

- Changes in how broodstock are held for spawning
- New techniques for producing sterile trout for stocking
- Changes in how jack salmon should be incorporated into broodstocks
- Changes in how hatchery fish are released to avoid predation
- Use of moist air incubators to mark otoliths of young fish
- Confirmation of ODFW standard protocols for minimizing handling stress
- Water used for incubation and rearing salmon and steelhead

The findings of the research proposed in this plan, as with past research, will be shared with the scientific community and the public, as well as incorporated into fish management as appropriate

Proposed Research Plan

In order to address the Center's goals stated above, the OHRC Advisory Board has adopted a research plan focused on three important areas:

1. If and how the differences in mate selection between hatchery and wild fish influences the reproductive success of hatchery fish in the wild, and how practices could be improved to increase the reproductive success of hatchery fish.
2. If and how hatchery rearing practices alter the selection of traits with resultant fitness consequences, and how hatchery practices could be altered to minimize hatchery versus wild fitness differences related to selection in the hatchery.
3. If and how manipulation of hatchery rearing and water can improve olfactory imprinting by juvenile salmonids and homing of adults to their hatchery of origin.

The proposed research projects in these three areas are described below. The phases of the research are described in a specific order so that the information gathered will build an understanding of what mechanisms influence the fish and which approaches show the most promise to test in a hatchery production setting.

The three research areas defined below are the areas that ODFW, OSU and the OHRC Advisory Board identified as being the most immediately informative and relevant to understanding and managing the interactions between wild and hatchery fish. This is not to suggest that these are the only areas of research surrounding the interactions of hatchery and wild salmon and steelhead that need to be explored. However, with limited funding, ODFW, OSU and the OHRC Advisory Board have identified the research areas listed here as top priorities. The OHRC Advisory Board has identified and will encourage and support research into other important areas as proposals are received. Examples of other important research to be conducted include (in no particular order):

- Effects of hatchery rearing on geomagnetic orientation
- Comparative reproductive success (pedigree) study with Coastal Fall Chinook
- Selection for harvest susceptible adult hatchery steelhead
- Behavior of triploid (sterile) salmon and steelhead
- Imprinting fish to remote locations for supplementation

Focused Research Areas

The three areas are categorized based on which OHRC Mission goal it addresses and described in detail with each phase of research identified.

OHRC Goal 1: Understand mechanisms that may create differences between hatchery and wild fish

Focus Area 1: Differences caused by mate selection

Background

The Calapooya Creek coho pedigree study (Dr. Michael Banks and associates) found that male offspring from wild fish raised in a hatchery were much less successful producing offspring than wild males, even if the fish were released before feeding in the hatchery. Because this loss of reproductive success occurred with very little hatchery intervention (only spawning, egg incubation and fry emergence), the researchers hypothesized that this effect must be due to how spawning occurred in the hatchery versus in the wild. Initial investigations related to this hypothesis suggested that mate selection for disease resistance may play a role in reproductive success (see references 13, 16 below).

Relevance of Strategy

Hatcheries in Oregon randomly mate male and female fish to avoid bias. If offspring survival is tied to successful mate selection in the wild, then being able to understand that selection and mimic it in the hatchery could lead to hatchery fish (with better reproductive success) having a lower risk to wild populations from interbreeding.

This research will help determine if mate selection is important to offspring survival by genomic examination of pairings of wild coho salmon from Calapooya Creek whose success at producing offspring is known. If the analyses demonstrate that most successful matings can be associated with specific genetic combinations observed among most successful mate pairs, then gene markers for relevant traits will be identified and methods to rapidly identify them will be developed. These rapid identification methods will then be applied to mate wild and hatchery fish in a hatchery. Relative reproductive success in the wild of these fish will be compared with reproductive success of normal, random hatchery matings to determine if ‘wild-fish-like’ targeted mating in the hatchery reduces the loss of reproductive success for hatchery fish, and the impact of current hatchery practice on wild populations. Thus results of this research may lead to new procedures for mating fish in a hatchery program that can reduce the risk hatchery fish have on wild populations.

Phase 1: Extensive Genetic Sampling and Analysis: The first step in this research will be re-sampling all of the successful mate pairings (1,387 total mate pairs) from the Calapooya Creek study and conducting an extensive analysis of the genetic composition of all pairings in the study and determining which genomic combinations were most successful in terms of production of young in the next generation (= reproductive fitness).
Timeline: 3 years, **Cost**: $700K

Phase 2: Application of Research Results: The information from the Phase 1 analyses will be used by ODFW and OSU collaborators to develop rapid, cost effective techniques to identify the important markers for mate selection.
Timeline: 1 year; **Cost**: $200K

Phase 3: Field Testing of 'Wild-like' Mating Practices in Hatcheries: The third step in this research program will be conducted at one or more ODFW hatcheries, in collaboration with the OHRC. The research and hatchery personnel will screen hatchery coho using markers to match fish and compare to normal hatchery mating practices. Smolts produced from mating those selected fish will be released following normal stocking procedures. We will use standard genetic pedigree techniques (as in the Banks study) to determine the reproductive success of the fish from the different mating procedures in the wild.
Timeline: 12 years; **Cost**: $3.7M

Focus Area 2: Differences caused by hatchery rearing

Background

Several studies have found that the offspring of wild salmon and steelhead brought into the hatchery were less successful at producing offspring in the wild than wild fish spawning in the wild (see references 1, 2, 3, 4, 5, 6, 10, 13, 15 below). In the Hood River study with steelhead, the reduced reproductive success was found to be inherited by the offspring, the result of strong trait selection occurring in the hatchery. Further research suggested that high rearing densities may be one factor that imposes strong selection, but the traits under selection and the particular rearing conditions that cause strong selection in hatcheries remain unknown.

Relevance of Strategy

Because the hatchery environment appears to select for traits that cause reduced reproductive success in the wild, it may be possible to alter certain rearing practices to reduce or eliminate the selection pressures, thereby producing hatchery fish that have traits that are more similar to wild fish.

Therefore, the goals are to identify which hatchery practices cause strong selection, and to identify what traits of the fish are under selection.

This research will attempt to determine: (1) the level of selection that occurs under different rearing conditions in the hatchery environment (e.g. different densities), and (2) the traits under selection, in order to better understand the mechanisms that may cause lower reproductive success in the wild.

The performance of offspring from the same set of full-sibling families will be compared when grown together under each environmental condition (families are tagged genetically, so individual identities are known). Here family performance is measured as average size and condition at release.

Firstly, environmental conditions that exacerbate differences among families in performance will be considered less desirable (i.e. because they cause stronger selection among families). Secondly, traits that distinguish high performance families ("winners") from low-performance families ("losers") under the high-selection environments will be examined to determine which traits are likely to be under selection. The genomes of "winners" and "losers" will also be compared to determine which genes appear to be under selection.

The various rearing conditions will be examined to find the most appropriate conditions that minimize selection, but maximize hatchery production. The revised rearing conditions would then be tested with wild steelhead in a production hatchery setting, and the reproductive success of the hatchery fish in the wild would be evaluated with a pedigree study. The results of this research may lead to new procedures for rearing fish in hatcheries that reduce the risk hatchery fish pose (due to lower reproductive success) to wild populations.

Phase 1: Investigation of Trait Selection: Multiple families of steelhead will be reared under different environmental conditions at the OHRC with the same goal for size at smolting. All fish will be associated with their parents/family genetically. At the end of the rearing period all fish will be assessed for size and condition factor. Siblings of those same families will be independently assayed for various candidate traits in order to determine which traits are associated with high *versus* low performance in the hatchery (i.e. "winner" *vs* "loser" families).
Timeline: 2 years; **Cost**: $350K

Phase 2: Identify Genes under Selection: The genomes of the "winner" and "loser" families will be analyzed to look for the genes that appear to be selected for under typical hatchery rearing conditions. These will be the genes that likely lead to lower reproductive success in the wild.
Timeline: 2 years; **Cost**: $250K

Phase 3: Field Testing: Once the traits under selection have been determined and the most appropriate rearing density is identified, researchers will release experimental groups of hatchery fish that have been reared under lower and standard densities. Researchers will then follow those fish with a pedigree study to determine if reproductive success in the wild is improved under the revised hatchery practices.

Timeline: 10 years; **Cost**: $3.1M

OHRC Goal 2: Develop approaches to manage hatchery fish that conserve and protect native fish

Focus Area 3: Methods to increase imprinting and homing back to the hatchery

Background

Efforts to reduce the number of hatchery fish that do not return to the hatchery or trap and instead spawn in the wild – defined here as straying – has been identified as a top priority in protecting wild fish (see references 7, 8, 9, 11, 12, 14, 17 below). This issue is a challenge for managing many hatchery programs in Oregon and throughout the Pacific Northwest. Oregon's Coastal Multi-Species Plan (CMP) recently identified a non-viable wild population that is affected by wild spawning hatchery fish, the Elk River Chinook population. The CMP's first priority is to address the non-viability of the wild Chinook population in the Elk River by taking actions to reduce the proportion of spawners that are hatchery fish, as well as addressing habitat improvement. Like most ODFW hatcheries, the Elk River Hatchery utilizes hatchery practices designed to maximize survival of hatchery-reared juveniles, minimize impacts of released juveniles on wild populations and ensure cost-effective operation. In some cases, these well-intentioned practices may contribute to excessive levels of straying, like those observed in the Elk River. Specifically, researchers have identified three hatchery practices at the Elk River Hatchery, commonly utilized at hatcheries throughout the Northwest that may contribute to elevated straying by hatchery fish:

1. Many hatcheries use river water for rearing hatchery Chinook, and it is this same water that is released from the hatchery to attract returning adults into the hatchery trap. This may make it difficult for the returning hatchery fish to differentiate the hatchery from the river itself. The CMP has proposed that the OHRC explore methods to attract more hatchery fish into Elk River Hatchery, including the use of an odorant in the water to give it a distinct scent that would attract more returning hatchery fish to the hatchery trap and keep them from spawning in the wild.
2. Modern hatchery programs are typically designed to attempt to minimize negative ecological interactions between juvenile hatchery and wild salmon. In an effort to minimize ecological overlap between wild and hatchery fall Chinook, the Elk River Hatchery, like several fall Chinook hatcheries in Oregon, typically releases their fish well after wild fish migrate out of the basin. The unintended consequence of these actions may be to increase straying by hatchery fish. The parr-smolt transformation has been identified as a critical period for successful olfactory imprinting and release from the hatchery at inappropriate developmental period before or after smolting can result in elevated stray rates. The third component of this research will

determine whether timing the releases of Elk River fall Chinook to coincide with the normal parr-smolt transformation will improve homing to the Elk River hatchery.

3. In most hatcheries, the need to control embryonic development rate (temperature) and limit exposure to pathogens dictates that salmon are initially reared on ground water. Salmon often return to the vicinity of their natal site, suggesting that in the wild the period of hatching and emergence may be a critical time for olfactory imprinting. Preliminary results from investigations at the OHRC indicated that salmon have an innate preference for stream water over ground water and development of the olfactory system is inhibited in salmon reared in ground water vs. surface water. This research will determine whether exposure to surface water during incubation and early rearing periods will improve homing to the Elk River Hatchery.

Relevance of Strategy

The research necessary to develop methods that may reduce the impact of hatchery fish spawning in the wild, as described in the first two focus areas, will require 15 to 20 years or more to translate into possible hatchery reform actions. In the meantime, short-term strategies need to be developed and tested that can reduce the impact of naturally spawning hatchery fish by removing hatchery fish that are not harvested in fisheries. Improving the ability to remove hatchery fish before spawning in the wild will allow hatchery programs, and the fisheries they support, to continue without jeopardizing the health of wild populations.

This research seeks to increase the homing of hatchery fish back to the hatchery to reduce the negative effects of excessive numbers spawning in the wild, and to better understand the imprinting and homing mechanisms in fish to allow for a broader application to other hatchery programs and situations. Salmon and steelhead use their sense of smell to find their way back to their natal streams which have unique scents. If research can determine how to improve the imprinting of the hatchery scent in juveniles, or create a unique scent for the water coming out of the Elk River Hatchery, and the outcome can significantly increase the number of hatchery fish that swim into the hatchery, the risk to the wild fall Chinook population can be reduced while allowing the same number of hatchery fish to be released if not increased. The results of this research may lead to new procedures for rearing fish in a hatchery program that can reduce the risk hatchery fish have (due to lower reproductive success) on wild populations.

Phase 1: Odorant Selection and Imprinting Effectiveness: Collaborate with NOAA scientist Andrew Dittman (an expert in homing science and fish olfactory systems) to identify the most likely effective scent(s) to incorporate into Elk River Hatchery water.

Researchers will identify and screen a variety of natural compounds released from aquatic plants and organisms and other known fish odorants for their potential use as artificial imprinting/homing cues and identify the most likely effective scent(s) to incorporate into Elk River Hatchery water. Specifically, a panel of potential odorants will be screened using the criteria listed below:

1) safe for release into natural waters
2) inexpensive and readily available
3) stable for storage and after release into natural waters
4) detected by the salmon olfactory epithelium at relatively low concentrations
5) ideally does not elicit innate behavioral (attraction or avoidance)
6) embryo and juvenile salmon are able to learn and respond behaviorally to the compound.

During this phase, an initial analysis will involve:

1. Literature review, permitting procedures and cost analysis to identify safe and cost-effective candidate odorants for subsequent testing.
2. Complex odors will be chemically analyzed to identify candidate compounds.
3. Screening the pool of candidate odorants for neural response (electro-olfactogram) to establish that they are effective and stable olfactory stimulants for salmon.
4. Y-maze analysis for innate behavioral responses.
5. Odor conditioning experiments to establish odor discrimination and learning.

Timeline: 2 years; **Cost**: $300,000

Phase 2: Verification of Imprinting: Researchers will incubate and rear Chinook salmon at the Elk River Hatchery and OHRC under conditions that vary through:

a. Exposure to one or more water-soluble odorants (e.g. those meeting the criteria above) and;
b. The timing of exposure to the above (exposure beginning at incubation versus exposure beginning at ponding)

Researchers will measure the effect of these treatments on juvenile physiology at life history stages relevant to olfactory imprinting (e.g. post emergence, parr-smolt transformation). These experiments will involve two parallel experimental groups.

1. Elk River Fall Chinook will be reared at the OHRC hatchery under controlled conditions and exposed to specific, known imprinting odorants (e.g. arginine) to assess imprint success and timing using a targeted gene approach. Salmon will be exposed to odors at specific developmental periods and sampled for imprinting success using quantitative PCR assessment of odorant receptor expression and smolting-associated physiology (i.e. ATPase, T4). Fish will be reared and sampled through the normal time of release at Elk River hatchery.

This phase will inform managers about the appropriate times for odor exposure and release at Elk River hatchery (and other hatcheries) to ensure successful imprinting.

2. A second group of Fall Chinook will be reared and exposed to added odors at the Elk River hatchery. These experiments will generate fish that have experienced the normal hatchery water and procedures at Elk River but will not readily allow for a targeted gene approach. Salmon will be sampled less frequently and tissues will be collected for transcriptome and epigenetic analysis of imprinting-associated changes in olfactory function. These tools will also allow for assessing imprint in situ at the Elk River hatchery and development for new markers for imprinting success.
Timeline: 3 years; **Cost**: $700,000

Phase 3: Verification of Homing and Field Testing: Researchers will measure the effect of treatments on adult homing behavior to the hatchery. This will be accomplished by releasing smolts with coded-wire tag (CWT) codes that identify their treatment group for three years and subsequent monitoring of adult returns – through creel, surveys and trap collections. If feasible, the effect of juvenile release timing on adult homing behavior will be tested. Feasibility will be determined through statistical power analyses of appropriate release group sizes (in context of typical adult return rates) and hatchery infrastructure. This phase will inform whether the variables tested (see phase 2) can significantly affect adult homing behavior.
Timeline: 10 years; **Cost**: $2.35M

References

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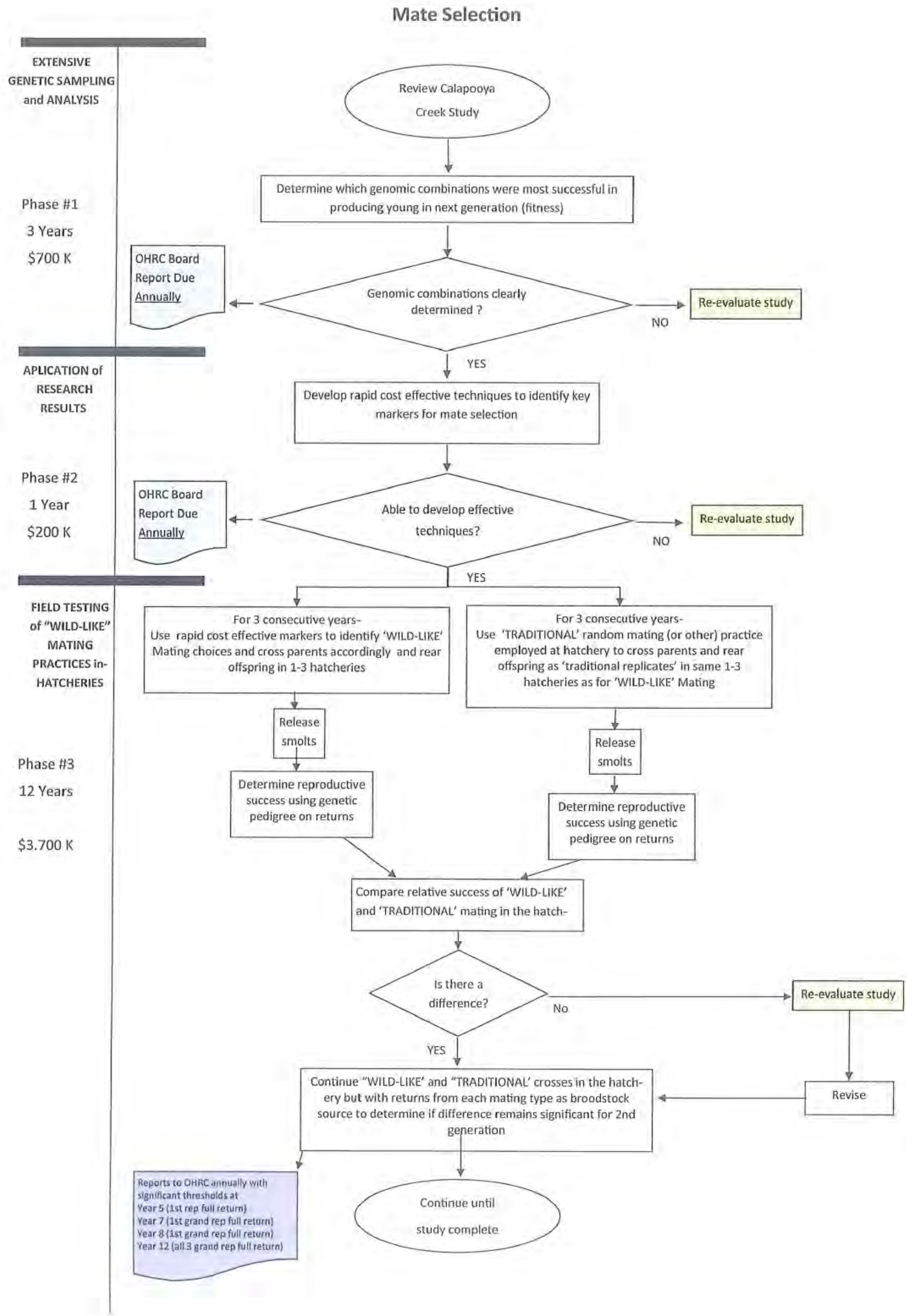
Annual and Biennial Budgets for Oregon Hatchery Research Center Priority Research Projects

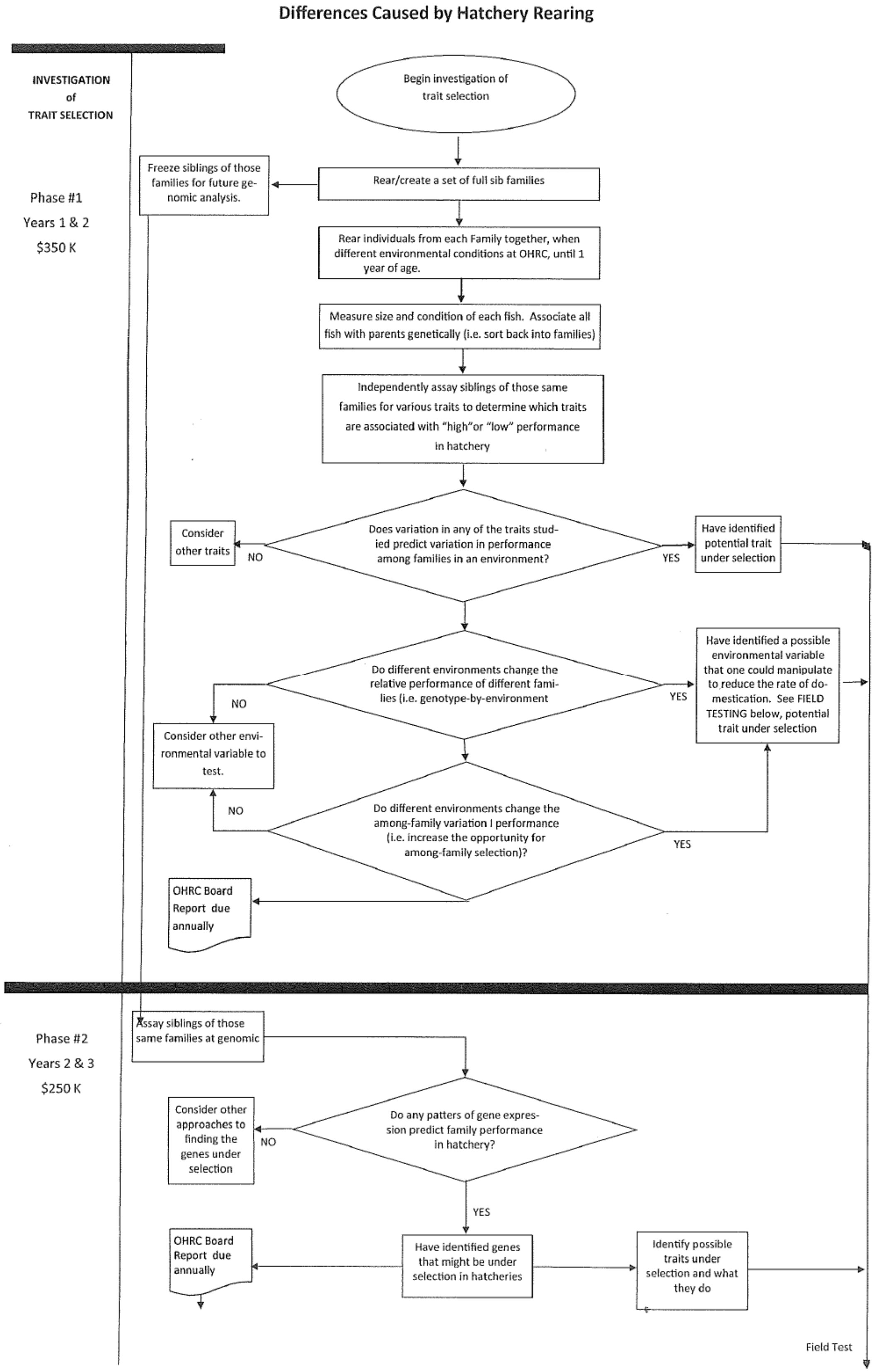
	Year 1	Year 2	Year 3	Year 4	Year 5	Year 6	Year 7	Year 8
Project 1: Mate Selection								
Phase 1: Genetic Sampling and Analysis	$233,333	$233,333	$233,333					
Phase 2: Application of Research Results				$200,000				
Phase 3: Field Testing					$308,000	$308,000	$308,000	$308,000
Project 2: Hatchery Rearing Practices								
Phase 1: Trait Selection	$175,000	$175,000						
Phase 2: Identify Genes under Selection			$125,000	$125,000				
Phase 3: Field Testing					$310,000	$310,000	$310,000	$310,000
Project 3: Imprinting and Homing								
Phase 1: Odorant Selection	$150,000	$150,000						
Phase 2: Verification of Imprinting			$233,333	$233,333	$233,333			
Phase 3: Field Testing						$235,000	$235,000	$235,000
Annual Budget	$558,333	$558,333	$591,666	$558,333	$851,333	$853,000	$853,000	$853,000
Biennial Budget	15-17	$1,116,666	17-19	$1,149,999	19-21	$1,704,333	21-23	$1,706,000

	Year 9	Year 10	Year 11	Year 12	Year 13	Year 14	Year 15	Year 16	Total
Project 1: Mate Selection									
Phase 1: Genetic Sampling and Analysis									
Phase 2: Application of Research Results									
Phase 3: Field Testing	$308,000	$308,000	$308,000	$308,000	$308,000	$308,000	$308,000	$308,000	
Project 2: Hatchery Rearing Practices									
Phase 1: Trait Selection									
Phase 2: Identify Genes under Selection									
Phase 3: Field Testing	$310,000	$310,000	$310,000	$310,000	$310,000	$310,000			
Project 3: Imprinting and Homing									
Phase 1: Odorant Selection									
Phase 2: Verification of Imprinting									
Phase 3: Field Testing	$235,000	$235,000	$235,000	$235,000	$235,000	$235,000	$235,000		
Annual Budget	$853,000	$853,000	$853,000	$853,000	$853,000	$853,000	$543,000	$308,000	$11,645,998
Biennial Budget	23-25	$1,706,000	25-27	$1,706,000	27-29	$1,706,000	29-31	$851,000	

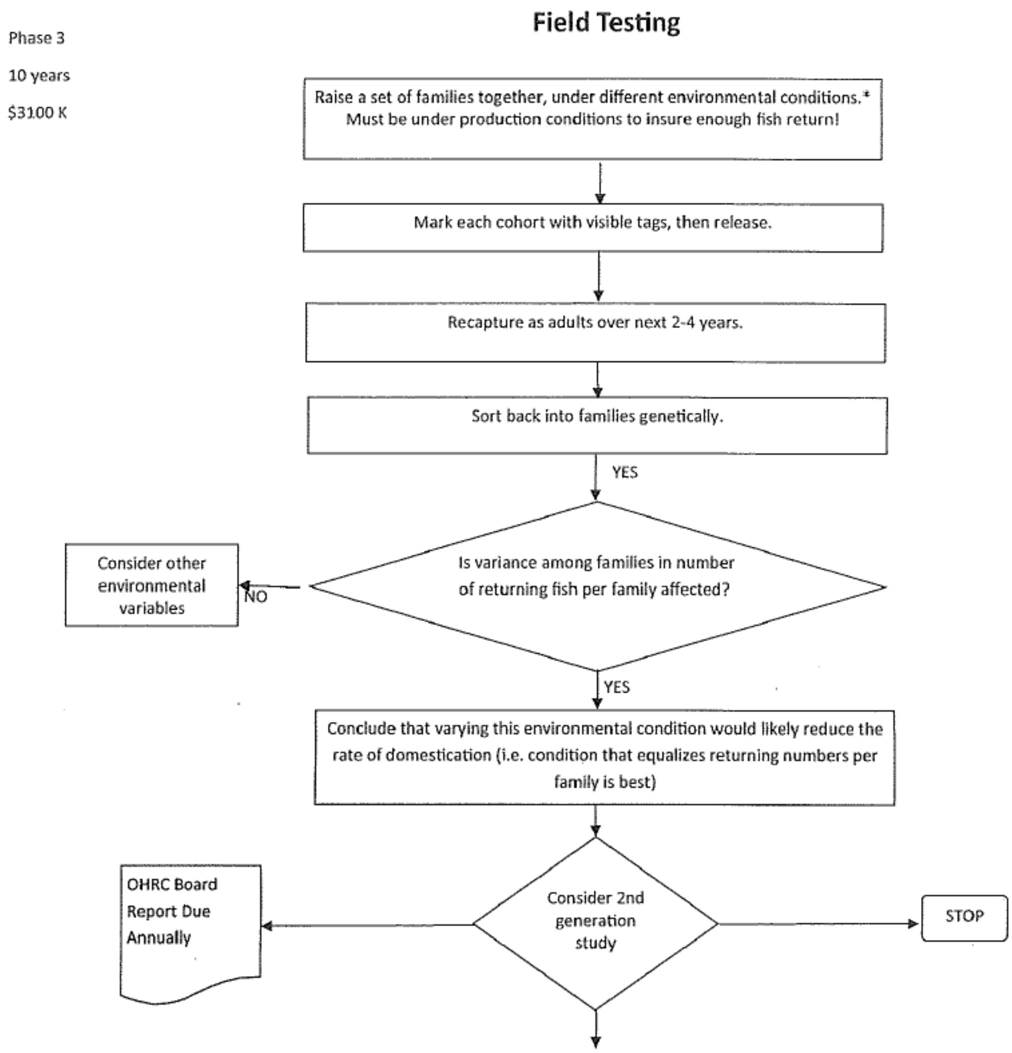
Project 1: Mate Selection project flow chart



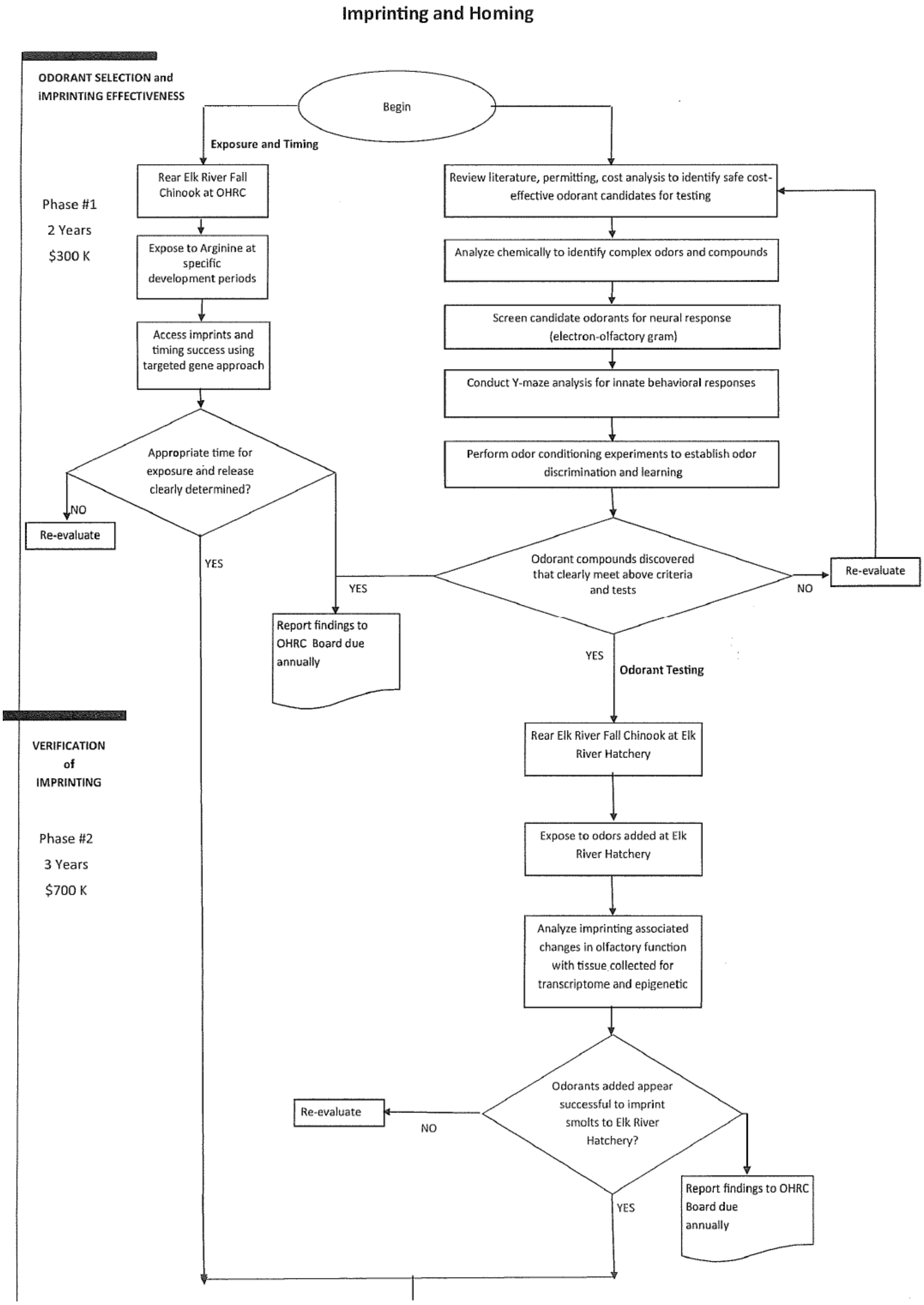
Project 2: Hatchery Rearing Practices project flow chart



Project 2: Hatchery Rearing Practices project flow chart (Page 2)



Project 3: Imprinting and Homing project flow chart



Project 3: Imprinting and Homing project flow chart (Page 2)