

Title: Evaluation of the Reproductive Success of Wild and Hatchery Steelhead in Natural and Hatchery Environments

Principal Investigators: Dr Tom Quinn* tquinn@u.washington.edu
Dr Lorenz Hauser lhauser@u.washington.edu
Dr Kerry Naish knaish@u.washington.edu

** principal correspondent*

Organization: University of Washington, School of Aquatic & Fishery Sciences

Address: Box 355020, 1122 NE Boat St, Seattle, WA, 98195-5020

Date: April 07 2003

Response to the Request for Studies issued by the Bonneville Power Administration
March 14, 2003

Response to FCRPS Biological Opinion **Action 182** to “determine the reproductive success of hatchery fish relative to wild fish”

Narrative

Title: Evaluate the Reproductive Success of Wild and Hatchery Steelhead in Hatchery and Natural and Hatchery Environments

Project description

ABSTRACT

Complex stochastic and deterministic processes related to breeding dynamics and survival of progeny result in differential reproductive success of adult salmonids with different phenotypic traits. These processes are essential to the long-term health of populations but are markedly different from patterns of mating and subsequent reproductive success in hatcheries. Hatchery populations are on evolutionary trajectories that may reduce their fitness, and their interactions with wild populations are a serious conservation concern. However, it is unclear to what extent hatchery fish can contribute to the stability or recovery of populations. To conserve wild salmonids and wisely manage hatchery populations, we propose to extend a unique study of reproductive success including wild steelhead, hatchery origin fish spawning naturally, and hatchery fish propagated in the hatchery. We have been sampling adults and smolts of the winter steelhead population in Forks Creek, a Willapa River tributary, since winter 1995-96. We are in a rare position – we are able to extend these data to the returning adult F_2 within a year and to the F_3 within the next four years. Our experiment will allow us to compare the genetic diversity from one generation to the next in natural and hatchery environments for males and females, estimate the reproductive success of the offspring of wild-hatchery matings in the wild, and determine the extent to which a wild population “resists” or “amalgamates” the genetic material from hatchery fish after cessation of hatchery releases over several generations. Specifically, we will document the phenotypic traits of fish used for breeding in the hatchery or migrating to spawn in the river, and will then use parentage analysis from DNA microsatellites to determine the reproductive success of individual fish, link these results to various fitness traits in spawning individuals, and examine the changes in gene frequency over three complete generations. Preliminary results from the returning adult F_1 indicate markedly lower survival of hatchery compared to wild fish, with the differences largely in the freshwater rather than marine phases, and hybridization between wild and hatchery fish (despite significant differences in average spawning timing). We have also found great variation in realized reproductive success of hatchery-spawned adults, probably resulting from variation in fertilization success and low but variable marine survival among families. These results leave open the question of whether the population’s long-term health will be affected by the hatchery influence. Our study is poised to address this question within the next few years.

INTRODUCTION

Reproductive Success

Salmon species exist as more-or-less discrete and isolated breeding populations, differing in selectively neutral traits and a variety of adaptive, quantitative traits (Ricker 1972, Taylor 1991; Quinn 1999). This “stock concept” is the cornerstone for the conservation and management of Pacific salmon. It is widely recognized that the health of salmon species depends on the maintenance of the complex population structure (Scudder 1989; Riddell 1993; Waples 1995), characterized by great diversity and adaptation to local conditions among populations.

The differential reproductive success (RS) of individuals that also differ in heritable phenotypic traits is a fundamental part of natural selection and the evolution of populations. RS is a function of the success of individuals in obtaining mates and producing viable offspring.

Mature adults are subjected to sexual selection (e.g., intra-sexual competition and mate choice), resulting in variation in RS. Mortality agents such as nest disturbance, predation, disease, and abiotic factors like gravel scour or ice may take different proportions of the fish from different families, resulting in further variation in RS among the adults of the previous generation. These processes are not unique to salmon but are common to animals (Emlen and Oring 1977; Wade and Arnold 1980; Clutton-Brock and Parker 1992; Andersson 1994), and are a natural component of evolution.

A number of traits have been documented or hypothesized to affect RS in salmon. At the breeding stage, males actively compete for access to ripe females, and large males tend to dominate such competition (Gross 1985; Foote 1988; Quinn and Foote 1994). However, small males may adopt alternative reproductive tactics (sneaking rather than fighting) and successfully fertilize some eggs (e.g., Schroder 1981; Chebanov et al. 1983; Maekawa and Onozato 1986; Foote et al. 1997). The success of males using these tactics changes rapidly over the course of the breeding season as the operational sex ratio (OSR: ratio of sexually active males to sexually active females) increases (Quinn et al. 1996). Behavioral research has tended to focus on variation in RS among males but RS may vary considerably among females as well. Larger females have both more numerous and larger eggs (producing larger fry, with higher survival rates) than smaller females (Beacham and Murray 1993), and they dig deeper (hence less vulnerable) redds (Steen and Quinn 1999), so there could be size-biased variation in female RS. Consistent with this hypothesis, Helle (1989) reported that a chum salmon population was much more productive in years when the females were large than when they were small, and that most of this effect was in addition to the projected increase in fecundity. However, Holtby and Healey (1986) questioned the hypothesis that large females are more productive than small females. Much of the high mortality rate for embryos is related to gravel scour, and this may vary greatly among families but be unrelated to female size. In addition to the factors affecting RS in males and females, there is evidence that salmon tend to mate assortatively by size (Hanson and Smith 1967; Foote 1988; Foote and Larkin 1988). That is, large males tend to breed with large females, and smaller males with smaller females, as a result of both competition and choice. Finally, the date of spawning is heritable, hence the progeny of early spawners will be more likely to mature at the same time as their siblings than non-siblings (Siitonen and Gall 1989), leading to genetic isolation by time within populations (Leary et al. 1989; Gharrett and Smoker 1993; Bentzen et al. 2001). This may tend to make them vulnerable to mortality agents affecting certain segments of the population (e.g., flooding during incubation: Thorne and Ames 1987).

Thus the breeding biology of salmonids is characterized by uneven contribution of adults to the next generation (i.e., selection), and this tends to reduce the effective (as opposed to absolute) population size. In addition, a series of connections between juvenile life history events may result in differential survival of sibling groups, magnifying the variance in RS of the parental generation. Juveniles of most species establish feeding territories in streams, and the ability to acquire and hold territories is largely a function of size and prior residence (e.g., coho salmon, *O. kisutch*: Mason and Chapman 1965; Rhodes and Quinn 1998). Egg size controls the initial size of juveniles, and juveniles that emerge early (because they were spawned early in the fall) will have a size advantage when later-emerging juveniles try to compete with them for space. Territoriality is also strongly related to prior residence, hence there may be great variation in territorial possession among families. Studies with Atlantic salmon and brown trout indicate the importance of these factors for survival (Brännäs 1995; Einum and Fleming 1999, 2000).

In addition to factors related to size and date of emergence, growth rate varies among families (e.g., Beacham 1989). Juveniles are also attracted to siblings (Quinn and Busack 1985) and sibship can affect patterns of foraging and aggression (Brown and Brown 1993), and distribution and growth (Quinn et al. 1994). The combination of family-specific variation in fry size, date of emergence, growth, social behavior, and distribution may lead to considerable variation among families in size at the end of their first summer in the stream. Size is positively correlated with survival through the winter in freshwater (e.g., in coho salmon, Quinn and

Peterson 1996) and at sea (coho salmon: Holtby et al. 1990; cutthroat trout, *O. clarki*: Tipping and Blankenship 1993; and steelhead trout, *O. mykiss*: Ward et al. 1989). Finally, there is evidence for family-specific variation in survival at sea independent of smolt size in pink salmon (Geiger et al. 1997) and chinook salmon (Unwin et al 2003).

Artificial Propagation

Many factors have reduced wild salmon abundance, including impassable or injurious dams, overfishing, land use practices, and variation in marine and freshwater conditions driven by climate (Stouder et al. 1996; National Research Council 1996). To offset the effects of these factors, salmonids have been produced in hatcheries in the Pacific Northwest for over a century. Considerable attention has been focused recently on the benefits and costs of such artificial propagation (e.g., Hilborn 1992). Some of these concerns pertain to the elevated fishing rates on mixed wild and hatchery populations (Wright 1993; Hilborn and Eggers 2000), and possible competitive interactions between populations (Nickelson et al. 1986; Nielsen 1994). These kinds of concerns can be difficult to resolve but are relatively simple to understand. However, the genetic concerns regarding hatcheries are much more intricate and perplexing. Reviews of the subject (e.g., Hindar et al. 1991; Waples 1991; Ryman et al. 1995) identify three major issues: fitness of hatchery populations for post-release survival, interactions between wild and hatchery populations, and reduction in effective population size due to supportive breeding.

Most kinds of animals bred for human consumption (e.g., chickens and pigs) are given no opportunity to interact with wild members of their species (if they even exist). Salmonids are unusual in that we breed them in highly artificial environments but then expect them to range freely on the ocean to feed, grow and return (excepting completely controlled aquaculture operations). A large and growing literature demonstrates that captive rearing induces a number of phenotypic changes in salmonids. Some changes such as color (Maynard et al. 1995) are largely if not exclusively environmentally induced but others may result from both genetic and environmental changes such as aggression (Berejikian et al. 1996; Rhodes and Quinn 1998), growth (Reisenbichler and McIntyre 1977), and anti-predation responses (Berejikian 1995). Changes in the timing of migration and breeding are largely under genetic control (Flagg et al. 1995; Quinn et al. in press).

Many studies indicate that the survival rates of hatchery fish are lower than those of wild conspecifics, but these differences are obviously attributable to many factors. Nevertheless, the genetic changes in hatchery populations are cause for concern, not just for the fitness of the hatchery population but also for wild populations with which they might interbreed. The most convincing studies on this subject, conducted on Kalama River steelhead (Chilcote et al. 1986; Leider et al. 1990), showed that hatchery adults produced markedly fewer offspring, when spawning naturally, than did wild adults. In this case the hatchery stock was derived from another basin so the generality of the finding is not clear. In addition, the hatchery had been operating for many years, so the extent of introgression prior to the study was difficult to ascertain. Finally, the study relied on protein electrophoresis and could assign the fish to population (hatchery or wild) on a probabilistic basis but could not assign them to specific parents. Thus the traits associated with reproductive success (and the poor performance of the hatchery fish) could not be determined. Notwithstanding these drawbacks, the Kalama River work was extremely important and influential, and there is evidence elsewhere of poor performance of hatchery fish in natural environments (e.g., Atlantic salmon: McGinnity et al. 1997; Fleming et al. 2000).

Hatcheries greatly increase egg-smolt survival but often have poor post-release survival. This is largely a result of rearing practices, and there are efforts to improve them (Maynard et al. 1995, 1996a, b), but many phenotypic traits affecting survival have a genetic basis as well. In some cases there has been deliberate selection for a trait that will facilitate operations in the hatchery (e.g., spawning fish that mature early in the season and discarding

those that ripen later: Ayerst 1977; Crawford 1979). More often, though, the selection is inadvertent and less obvious. Since most fish survive in the hatchery, genetic changes probably result from one of two processes. First, hatcheries fail to cull fish of “poor quality” that would be selectively taken by predator or pathogens in nature, and natural processes cull them after release. After eliminating the obviously abnormal fish, most hatchery managers are unwilling to cull large numbers of their fish without a very clear set of criteria. There is very little opportunity in a hatchery to test the vigor of juvenile salmon at a production scale, so this process may be unavoidable.

The second obvious form of selection in hatcheries is the mating system, and this is the main “pressure point” by which we may practically affect salmon evolution in hatcheries. There seems to be little documentary evidence on the specific practices in hatcheries but it is widely acknowledged that staff often tended to select a small number of males for breeding based on size or more subjective criteria related to “quality”. The use of a few males was both a matter of selection and convenience; only a few males are needed to fertilize the eggs of many females. Females tended to be spawned until the capacity of the hatchery was reached. Thus, except for attributes related to timing, there may have been little variation in probability of breeding and reproductive success (other than fecundity) among females but great variation among males.

RATIONALE AND SIGNIFICANCE TO REGIONAL PROGRAMS

There are two major classes of concern with respect to genetics of hatchery and wild populations: the health of the hatchery population and the consequences of interactions between wild and hatchery populations. In the first case, geneticists have expressed concerns about the loss of genetic diversity that might be associated with breeding in hatcheries (e.g., Utter 1998), especially as supportive breeding in itself may cause a reduction in genetic diversity (Ryman *et al.* 1995). In the establishment of breeding protocols, the dominant philosophy has been to prevent loss of genetic diversity. For example, the Alaska Department of Fish and Game Genetic Policy (Davis *et al.* 1985) stated (on p. 14) that “Fitness is enhanced by heterozygosity” and (on p. 15) that “Any loss of genetic variation will be accompanied by a concomitant reduction in fitness.” Highly competent geneticists wrote this report but the statements linking genetic diversity to fitness were made without reference to specific evidence. The report was also very vague with respect to specific protocols (i.e., how the goal of diversity was to be achieved) but it expressed the need to keep the effective population size high ($N_e > 400$).

This is not the universal goal. In National Marine Fisheries Service hatcheries in southern California (e.g., Scott Creek and San Lorenzo River), the policy is designed to more closely mimic natural patterns. The protocol is for each female’s eggs to be fertilized with milt from four males (randomly picked) and each male is given a chance to fertilize eggs from four females. In the Columbia River basin, the Integrated Hatchery Operations Team (IHOT 1994) produced a report on policies and procedures for salmonid hatcheries. The evaluation of “Performance Standards” (p. 35) was all related to aspects of production such as number and size of smolts, date of release, water temperature, etc. However, the report contained a “Genetics Policy” on pp. 65-72. The policy objective was to “... maintain adequate genetic variation and fitness in populations” (p. 66). This policy was to be achieved by “... broodstock collection guidelines designed to minimize selective pressures from hatchery practices.” A series of protocols were recommended, including random matings with respect to fish body size, use of jacks in proportion to their abundance, and absence of selection against fish whose appearance suggested “poor quality”.

Two questions might be posed with respect to this policy. First, is it wise, and second, can it be accomplished? The effort to breed fish at random and ensure equal participation and RS among all adults contrasts starkly with the natural patterns of breeding observed by behavioral ecologists (e.g., Hanson and Smith 1967; Schroder 1981; Gross 1985; Keenleyside and Dupuis 1988; Quinn and Foote 1994; Quinn *et al.* 1996). Aldo Leopold wrote in “Round River” (1953), “If the biota, in the course of eons, has built something we like but do not understand, then who

but a fool would discard a seemingly useless part? To keep every cog and wheel is the first precaution of intelligent tinkering.” To some extent this is the present philosophy with respect to genetic resources: keep all the material and be sure nothing is lost. This may be wise when it comes to biodiversity at the species level but is it necessarily wise at the level of gene pools? Can you repair a machine by putting all the “cogs and wheels” into the case and shaking them up, as may result from such breeding protocols? In addition to uncertainty regarding the wisdom of random matings, there is also the question of whether the goal can be accomplished. Hatchery populations are subject to mortality and selection but most of it takes place after release.

PROBLEM STATEMENT AND JUSTIFICATION

Studies of the natural history and behavioral ecology of salmon reveal complex stochastic and deterministic processes related to survival and breeding dynamics that result in differential reproductive success of individuals with different phenotypic traits. These processes are widely recognized as essential to the long-term health of populations and indeed the species themselves. For better or worse, artificial propagation is now an integral part of the overall evolutionary dynamics of salmon. A large fraction of the salmon in the Pacific Northwest and especially the Columbia River basin are produced in hatcheries. Salmon populations in hatcheries are on their own evolutionary trajectories, and they interact with nearby wild populations, affecting both groups. These interactions are consistently identified as priority concerns in salmon conservation (e.g., Waples 1991; Utter 1998).

There are many questions related to the genetics of hatchery and wild salmon, and some are unique to particular species or settings (i.e., the hatchery and local spawning grounds). However, it is impractical to study each hatchery in detail, so if we are to make progress we must accept the scientific principle that results generated at one site are relevant elsewhere. There are three subjects that are at the core of the problem. First, what is the comparative RS of wild and naturally spawning fish of hatchery origin? If the performance is similar, there may be little grounds for concern. Second, how does the performance of “hybrids” between forms compare to that of pure wild and hatchery fish? This is critical because if hatchery fish are less fit (i.e., lower lifetime RS) than wild fish but still produce viable offspring, we will need to estimate and model the “resistance” of the wild population to depression in RS through interactions with the hatchery population in subsequent generations. Third, we need to know how hatchery populations evolve. Specifically, how strong is the genetic control over important life history traits affecting fitness like age at a maturity, growth, egg size and fecundity, and how much does RS vary among fish spawned in a hatchery?

The Columbia Basin Fish and Wildlife Authority recently produced the “Mainstem and Systemwide Province Draft Artificial Production Program Summary” for the Northwest Power Planning Council (CBFWA 2002). This document makes reference, in numerous places, to the importance of genetic interactions between wild and hatchery salmonids, and includes the three core questions that we have identified. The report stated:

Studies need to be conducted to measure the relative fitness of hatchery fish spawning in the wild. Likewise, experiments need to be conducted to estimate the rate of domestication in a hatchery setting, the rate of readaptation to a natural setting, and to provide information into what traits are under selection in different environments.

PROPOSAL OBJECTIVES, TASKS AND METHODS

Objectives

We propose to take advantage of a unique opportunity to directly determine the reproductive success of steelhead in a production hatchery, fish from the same hatchery population breeding in the river nearby, and the wild fish over two complete generations. This effort, combining DNA parentage analysis with life history data collected from the fish and records of which fish were

bred together, will give unprecedented insights into the genetic outcome of hatchery breeding, differential survival between wild and hatchery fish, and the prospects for recovery of a wild population after hatchery influence ends. We will produce a report documenting the phenotypic traits associated with breeding in the hatchery and the realized RS of these fish, and the RS of naturally spawning steelhead at smolt and adult stages from the first year when hatchery steelhead returned to spawn in this system (winter of 1995-96) onward.

Tasks and Methods

Our overall objective is to determine the factors influencing RS in wild and hatchery-origin salmonids in natural and hatchery environments, using steelhead trout as the test species. Our specific objectives are as follows: (1) quantify the breeding pattern (i.e., selection of fish for spawning) in a production hatchery, (2) directly determine the realized RS of the individual hatchery fish by DNA parentage analysis of the adults returning over 2-3 complete generations (3) directly measure the effective population size (N_e) of the hatchery population over multiple generations (4) determine whether there was any loss of genetic diversity in the hatchery population over two generations, (5) determine the realized heritability of key life history traits for hatchery fish released to the sea, (6) determine the realized reproductive success of wild and hatchery-wild hybrid parents spawning naturally, and compare these levels to those of the first generation of hatchery steelhead spawning naturally. To achieve these objectives, we will combine basic biological data (size, age, fecundity, egg size, date of reproduction of adults, and size and survival rate of progeny) with genetic analyses to link offspring with their parents. Hatchery fish will be sampled as adults; wild fish will be sampled as adults and smolts.

Steelhead are an appropriate species because (1) they are broadly distributed (Burgner et al. 1992), (2) they have been the focus of intense recreational fisheries and are also commercially fished by Native American tribes with treaty rights; (3) many populations are in jeopardy (Stouder et al. 1996) but a status review by the National Marine Fisheries Service indicated that large knowledge gaps will hinder conservation efforts (Busby et al. 1996); (4) previous research with protein electrophoresis indicated that hatchery-produced fish spawning in the wild may have lower reproductive success than wild fish (Chilcote et al. 1986; Leider et al. 1990); (5) there is very extensive hatchery propagation of this species. Light (1987) estimated that about half the adult steelhead in North America, and 70% of those in Oregon, Idaho and Washington, are produced in hatcheries.

Background and Site Description

Our study site, Forks Creek, provides rare opportunities for studying hatchery genetics and management, and our history of research at this site will allow it to make exceptional contributions to pressing management problems in the very first years of the study, rather than having to wait many years for results as would be the case at new sites. The creek, a tributary of the Willapa River in southwest Washington, has a small wild population of steelhead that generally enters from March through May and spawns from April through June (Mackey et al. 2001). A hatchery, situated just above the creek's confluence with the Willapa River, has been operated by the Washington Department of Fisheries to produce coho and chinook salmon since 1895. There had been occasional releases of steelhead in the Willapa River but apparently not in Forks Creek, and there had been no continuous propagation of steelhead in the system. When WDF merged with the Washington Department of Wildlife, a decision was made to produce steelhead as well. Beginning in the winter of 1995-96, the creek received the first adult returns from hatchery-produced steelhead released as smolts in spring of 1994. These fish originated from the Chambers Creek hatchery population but had been propagated at the Bogachiel Hatchery, mixed with unknown proportions of local wild steelhead. The Chambers Creek population is widely released in Washington by the WDFW, and has been artificially selected for early return and spawning timing (generally December through February) to minimize fishery conflicts and interbreeding with wild fish, and to facilitate culture operations. This creation of a

strain of the species for human use that differs from the wild populations has been the state's approach to conserving the abundance and genetic integrity of steelhead. There were additional plants from the Bogachiel Hatchery in 1995 and 1996 but since then the hatchery has produced its own fish without outside influence. We initiated sampling with the first brood year, so this site provides an excellent (and probably unique) opportunity to examine the genetics of hatchery steelhead.

Our past and proposed sampling is designed to take advantage of a special feature of the Forks Creek hatchery operation. Not only do we sample wild adult steelhead and their progeny as smolts and adults, and sample the adults spawned (or killed as surplus) at the hatchery. Most importantly, in the first two years when hatchery adults returned (1996 and 1997), the decision was made to allow surplus adults to spawn in the river (Mackey et al. 2001). Accordingly, 362 hatchery adults were allowed upriver, along with 59 wild fish, and 117 hatchery fish were spawned at the hatchery in those two years. However, the decision was then reversed, to deny hatchery fish access to the spawning grounds. Thus the wild population was exposed to a very strong "pulse" of hatchery influence for two years, followed by an open-ended "recovery" period. This unusual situation allows us to gather critical data on the extent to which a wild population resists hatchery influence once the influence is largely terminated. A few hatchery fish may bypass the weir at high water, and they might be considered to be the equivalent of strays. In addition, a few hatchery fish were used at the hatchery but we were able to sample these fish (Table 1).

Table 1. Numbers of steelhead spawned or sacrificed (i.e., killed but not used for spawning) at the Forks Creek Hatchery. Final data collection from BY 2003 is being completed at this time.

Brood year	Origin of adults in the hatchery	
	hatchery	wild
1996	61	0
1997	56	0
1998	45	0
1999	144	0
2000	131	12
2001	126	1
2002	375	2
2003	> 550	0

Sampling Methods

The hatchery has spawned steelhead since winter 1995-96 (Brood Year 1996 by our designation; Table 1) and we have obtained samples (length, weight, scale for age determination and fin clip for DNA, and egg size and fecundity from females) from almost all of them. There is a weir across the creek that guides salmon and steelhead into a deep concrete channel filled with water from the hatchery. This weir functions well except under very high water conditions. Hatchery staff remove steelhead from the channel and they are examined each week and allowed to remain and mature, spawned, sacrificed, or allowed upriver to spawn in the river, depending on whether they are wild or hatchery, fully mature or not, and whether the hatchery's capacity has been reached. We waited until the hatchery staff selected and spawned the fish according to their standard practice or decision of the moment, and we then sampled the fish. We estimated the fecundity of females by weighing the entire mass of eggs, and weighing and counting a subsample of eggs. We can thus determine how much of the variation in female RS can be attributed to fecundity (the body-size fecundity relationship), as opposed to other factors related to body size or spawning date.

The hatchery rears steelhead for one year before releasing them to sea and most of the steelhead spend two full years at sea before returning (inferred from scale examination, size frequency analysis, and DNA parentage). Thus in winter 1999 most of the hatchery-produced adults returning to Forks Creek were the progeny of adults that we sampled in 1996 (i.e., the first generation), and in winter 2001-2002 we sampled the F_2 (grandchildren) of the first generation. Likewise, we are now completing sampling of the F_2 of the 1997 brood in the winter of 2003. The survival of hatchery progeny to the smolt stage is very high so we evaluate the RS of hatchery parents on an adult-to-adult basis.

In addition to our sampling of hatchery fish and the patterns of breeding there, we have also systematically sampled the naturally spawning fish. Any fish allowed upriver were identified to origin (all hatchery fish have the adipose fin removed) and sex, sampled for length, and scales were removed for age determination and fin tissue removed for DNA analysis. We have also operated a smolt trap from mid-late April to early June. This fan trap catches a very large fraction of the smolts (essentially all except on high water events when a panel must be removed to prevent damage). All steelhead are measured and weighted and a fin clip removed, and these data are recorded with the date of capture. In addition, the trap catches a number of steelhead kelts and this augments our sampling of upstream migrants. We do not know precisely what fraction of the wild population we sample but all indications are that it is high.

Genetic Analysis

We have determined the parentage (and other forms of kinship) for steelhead by genotyping microsatellite loci. The attributes of microsatellites as genetic markers have been reviewed extensively (e.g., Wright and Bentzen 1994; O'Reilly and Wright 1995; McConnell and Wright 1997). Briefly, they consist of 1-5 base pair (bp) repeats that form tandem arrays < 300 bp in length, and exhibit high levels of allelic variation in repeat number. Polymorphism exhibited by specific microsatellites is readily detected by amplification of the microsatellite through the use of oligonucleotide primers specific to the non-repetitive regions that flank the repeat array, in combination with the polymerase chain reaction (PCR). Allelic variation is scored by gel electrophoresis of the PCR products, most commonly on automated systems facilitating running and scoring of genotypes.

Microsatellites have recently come into widespread use in kinship analyses (reviewed in Hughes 1998; Marshall et al. 1998) because the large numbers of alleles and high heterozygosities provide the power for discriminating parent-offspring combinations from unrelated individuals. Power increases dramatically with increasing expected heterozygosity (H_E) (Blouin et al. 1996). For loci with $H_E \geq 80\%$ the average exclusion probability is > 0.999 for nine unlinked loci. We have sampled almost all of the Forks Creek hatchery spawners and so anticipate a high success rate in identifying offspring.

Genomic DNA extractions and PCRs were performed as outlined in McLean et al. (2003a, 2003b). A 96-well capillary system Molecular Dynamics MegaBACE 1000 (Amersham Scientific) was used to size fractionate the eight microsatellite loci examined. Electropherograms were analyzed using Genetic Profiler software version 1.1 (Molecular Dynamics). Observed and expected heterozygosity, probability tests of Hardy-Weinberg equilibrium, tests of linkage disequilibrium, and genetic differentiation estimates between the hatchery and wild populations were calculated using the GENEPOP (version 3.0) software package (Raymond and Rousset 1995), as in McLean et al. (2003a, 2003b). Parentage was assigned through exclusion using the software package WhichParents (version 1.0 alpha; Bodega Marine Lab, UC Davis). Putative parent-offspring groups were further analyzed by direct genotype comparisons. One parent was assigned if an allele match with the offspring occurred at each locus; two parents were assigned if their genotypes together could have produced the offspring genotype. Subsequent statistical analyses did not include offspring that were not assigned at least one parent.

Our detailed analysis of variance in RS among individuals amounts to a direct measurement of the current (variance) effective population size (N_e) of the population. Our

microsatellite assays will also permit indirect estimates of current N_e based on temporal variance in allele frequencies (Waples 1989; Waples 1990a, b; Waples and Teel 1990) and linkage disequilibrium (Bartley et al. 1992), and we will be able to compare our direct observations of N_e with these commonly used (but rarely validated) indirect approaches. N_e is a parameter of fundamental importance in population and conservation biology; hence these comparisons will be of significant interest.

Progress to Date and Preliminary Results

We have been continuing to collect genetic samples from adults and smolts in the river and adults in the hatchery but analysis has been conducted on only a fraction of these samples, and constitutes the Ph.D. dissertation of Jennifer McLean, to be completed in April 2003. Her analysis has considered the production of adult progeny by the first three years of hatchery adults spawning in the hatchery, and the production of smolt and adult progeny from the first two years of natural spawning (including the many hatchery fish and the wild fish). No analyses have been conducted on the next generation of hatchery steelhead, not on the next generation of natural spawners. These are critical because they will allow us to determine the production of smolts and adults from the hybrid wild and hatchery fish, and from hatchery origin fish spawning in the river for a second generation.

Preliminary analysis of the patterns of fish spawned and killed unspawned, and of the fish spawned in the hatchery together through 2001 has revealed fascinating patterns. First, and least surprisingly, the average date when fish were spawned was earlier than the date when fish were killed, indicating directional selection for earlier maturation (3 January vs. 17 January, $t = 12.98$, $P < 0.001$). More surprisingly, given the apparently random choice of fish, is the fact that the fish used for spawning have been significantly larger than those sacrificed at the hatchery (females: 684 vs. 630 mm, males: 687 vs. 656 mm, $P < 0.001$ in both cases). This difference might arise from a correlation between date of maturation and size but the tendency for larger fish to mature earlier was negligible ($r^2 = 0.04$ for females and 0.01 for males). Thus there seems to be some unconscious tendency for the staff to spawn larger fish, though they do not plan to do so and the fish are not lined up prior to spawning in any manner that would facilitate comparisons.

Most surprising of all, the fish that were spawned together (i.e., gametes placed in the same bucket) were more similar in size than would occur by chance. Specifically, the average size of males and females in each spawning group were positively correlated ($P < 0.05$). Finally, although there was an attempt to use males and females equally, the actual ratio of females to males spawned in each group ranged from 6:2 to 7:16. Overall, 212 of the 278 females were used for spawning and only 66 (23.7%) were killed unspawned but 39.4% of the 289 males were killed.

The realized RS of the hatchery fish is often assumed to be quite uniform because of the high survival rates in the hatchery. However, great variation in RS was actually observed, probably resulting from variable fertilization success among males and high and variable post-release mortality. Total marine smolt to adult survival (escapement plus estimated catch) was only 0.88% over the first three years, with only 0.36% return to the hatchery. Figure 1 shows the variation in number of progeny produced among adults. Many parents had no offspring but some had 10 or more, with more variation among males than females.

Figure 1. Number of hatchery parents producing 0 – 24 adult offspring that returned to the hatchery in the first three years of operation. Note that most parents produced 0 or 1 offspring, and that reproductive success varied more among males than females.

The preliminary analysis of the naturally spawning fish was done by assigning the fish to population of origin based on maximum likelihood techniques. As shown in Table 2, the wild females were very much more successful in producing offspring than the hatchery fish but the marine survival rates were generally similar (and very high).

Table 2. Reproductive success of brood year (BY) 1996 and 1997 hatchery and wild steelhead spawning in the wild, based on the number of females spawning in the creek, the number of smolts produced per female, the estimated survival from smolt to adult stages, and RS (number of adults produced on average per female parent; from McLean et al. 2003a, 2003b). Replacement requires RS values of 2 or greater.

BY	Population	Females	Smolts per female	Marine Survival	RS
1996	Hatchery	90	1.07	38%	0.41
	Wild	11	24.45	15%	3.73
1997	Hatchery	73	1.33	12%	0.16
	Wild	10	18.80	36%	6.70

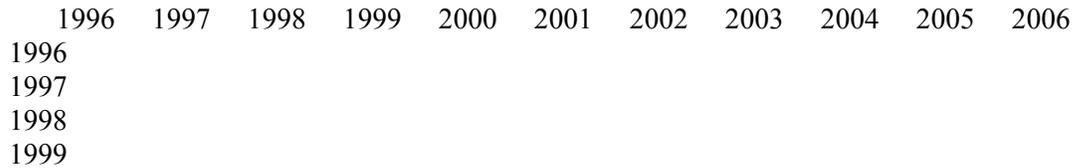
These calculations (in Table 2) assigned all fish to the hatchery or wild populations, and include fish whose parents we may not have sampled. Subsequent analysis determined the specific parentage of many of these fish, and these data confirmed the inferior performance of the hatchery fish in the freshwater stages, the greater variation in RS among males than females, and also indicated the presence of hybrids between wild and hatchery fish (Table 3). It will be extremely important to determine if any of these hybrids reproduced, and what phenotypic traits their progeny show, especially in spawning date.

Table 3. Mean (range), and coefficient of variation (CV) in reproductive success for naturally spawning parents from BY 1996 and BY 1997, as indicated by the number of progeny detected as smolts and adults are shown separately. Because of incomplete sampling, these are relative but not absolute measures of RS.

Population	Smolt production		Adult production	
	Mean	CV	Mean	CV
Hatchery males	0.95 (0 - 17)	1.73	0.39 (0 - 10)	2.56
Wild males	2.24 (0 - 9)	1.12	0.61 (0 - 3)	1.48
Hatchery females	0.73 (0 - 8)	1.38	0.33 (0 - 3)	1.94
Wild females	1.48 (0-5)	0.99	0.42 (0 - 3)	1.72

TIMELINE

Wild spawning fish (some of which are of hatchery ancestry)



Hatchery spawning fish

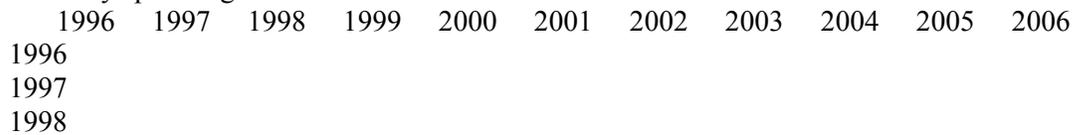


Fig 2: Timeline representing the generations already surveyed, and to be investigated in the current project. Full bars represent freshwater stages, checkered bars ocean life, ending with reproduction.

JUSTIFICATION FOR FINANCIAL SUPPORT

The Forks Creek project was started as part of a suite of studies on the habitat and interactions between wild and hatchery salmonids in the river system, supported by the Weyerhaeuser Foundation, and including projects by Long Live the Kings, Washington Department of Fish and Wildlife, National Marine Fisheries Service, and the Weyerhaeuser Company. This support was augmented by funds from the Hatchery Science Reform Group and the National Science Foundation. However, all of these sources of support have been exhausted and we cannot continue sampling after the 2003 smolt and wild adult trapping, and cannot process some of the samples in our possession. Thus the project is at the brink of being terminated, just at the point when the most important results are becoming available with the return of the second and third generations. We believe that the data are of critical importance for the management of salmonids throughout the region, and especially in the Columbia River basin, where artificial propagation is so widespread and controversial.

We are cognizant of the fact that Forks Creek is not in the Columbia River basin. However, to the extent that the scientific method has any validity, results must be broadly representative of processes not confined to the specific system under study. If this were not the case, scientists would be doomed to conduct identical studies on every lake, island, species, individual, or other unit of organization of interest to us. Indeed, if the results of studies on one hatchery are not applicable to another, we would be forced to conduct detailed studies on all hatcheries of concern. Fortunately, there are basic processes of behavior, ecology, and genetics at work and we can learn lessons and successfully transfer them. Clearly there are local features at individual hatcheries and associated natural spawning areas that may cause results to differ among sites, as well as

some differences in life history among species. Therefore, some diversity of studies will be needed to fully understand the genetic interactions between wild and hatchery salmonids that will permit wise management of these fishes. However, we do not believe that there are fundamental differences between the Forks Creek Hatchery and Columbia River basin hatcheries that make our results inapplicable to the Columbia River basin. To the extent that there are any such considerations, we believe that the years (and generations) that we have already sampled will more than compensate for any concern about the location of the facility. Moreover, the fact that we were able to sample wild fish before hatchery influence, and the fact that the hatchery influence on the wild population is not ongoing make this study system particularly informative. Without further support, the project will end and any subsequent effort to restart it will suffer for the lack of samples in the intervening years. We request support to extend this study for two additional years but we are confident that the insights that it will provide will justify long-term support.

The majority of our budget request for supplies is directed towards the genotyping of the individuals we will sample. Our Center (Marine Molecular Biotechnology Lab) operates as a shared facility, and we operate as a cost center, with a monthly bench fee. The bench fee of USD 450 per month covers plastic consumables, all chemicals, gloves, computers and equipment maintenance. We envisage a large amount of genotyping for this project, and intend to take advantage of the automated methods available in our laboratory. The request for supplies cover DNA extraction (Qiagen system described below), fluorescent primers for microsatellite loci (for visualization on the FMBIO IIe and the Megabace automated sequencer), and *Taq* polymerase for PCR amplification of loci. Finally, we intend using multiplexed groups of genetic loci for kinship analyses and will perform this analysis on our MegaBace 96 well capillary automated sequencer (Molecular Dynamics, Pharmacia). The cost per 96 well plate run on the sequencer is \$150 – and includes the standard ladder, the matrix and the cost of operating the machine.

FACILITIES AND EQUIPMENT

Molecular analyses.

Laboratory:

Our research center comprises a shared facility between four faculty members (two of which are Drs Hauser and Naish) within the College of Ocean and Fisheries Science. We have sole use of a large number of laboratories that include four PI core labs, an “ancient DNA lab”, a sequencing lab, an equipment room, an autoclave room, a dark room and an electrophoresis lab. Half of the space is newly renovated. All work carried out previously on this project has been within our laboratories.

Equipment available and relevant to this project:

One Megabace (Molecular Dynamics, Pharmacia) 96-well capillary sequence analysis system with associated computers and software. This system allows us to process 288 genotypes in forty-five minutes. We also have 4 tabletop centrifuges, one of which is a Sigma centrifuge capable of handling Qiagen’s 96-well DNA-isolation plates. Hence, we are able to process the DNA extractions in a short period of time. The lab houses a large number of sequencing gel rigs, including the “Jumbogel” (Miraibio), adapted to take advantage of the entire scanning area of the FMBIO IIe fluorescent gel scanner, numerous agarose gel rigs, Millipore ultra-pure water filtration, four MJ Research “basestation” thermocyclers for DNA amplification (with interchangeable 96-well and 384-well blocks), a Spectramax plate reader for DNA quantification, numerous multichannel pipettes and electronic repeating pipettes. Finally, we have financial support to purchase an automatic liquid handler, capable of processing a large number of reactions.

Offices:

Each student and faculty member has a computer – we also have a number of shared computers (for analysis of sequencing data and visitors) and surplus computers available for this project. All computers are linked via an Ethernet to a Center LAN and the internet.

Forks Creek:

The Forks Creek facility is described under “sample collection” above. The facility is fully staffed, and our research has received logistic support from this hatchery since 1996. We request support for a smolt trap for our work – we have used equipment “on loan” in the past and wish to install reliable equipment for the duration of this project.

KEY PERSONNEL/QUALIFICATIONS OF PARTICIPANTS

The Forks Creek project was conceived and initiated by Thomas Quinn in collaboration with Paul Bentzen, then on the UW faculty. Dr. Quinn will have overall responsibility for the project, with special emphasis on the meta-analyses. He has over 20 years of experience in salmon behavior, ecology and evolution, including work on reproduction, and studies on hatcheries. Dr. Hauser, who replaced Dr. Bentzen as the molecular ecologist at the UW, is a broadly trained geneticist with experience in the population genetics of many fish and invertebrate species, and will have primary responsibility for the DNA extraction and parentage analysis. Dr. Kerry Naish is a quantitative geneticist with expertise in the molecular basis of quantitative traits, and she will be primarily responsible for conducting analysis of the heritability of fitness traits among wild and hatchery fish, and their hybrids. We request 1.5 months salary for the three faculty members in each year of the project. We request salary support (at 50% FTE) and tuition for a graduate student, and salary support (at 100% FTE) for a staff biologist who will have day-to-day responsibility for supervising the field work, conducting the lab work, and maintaining the increasingly complex database associated with this long-term project. We also request hourly support for staff to sample adults at the hatchery and operate the smolt trap. The trap requires a continuous, on-site staff person though only a few hours a day may be needed. We have hired local staff because it is impractical to commute from Seattle for these tasks. .

Budget September 16, 2003 - September 15, 2006

			Year 1	Year 2	Year 3	Total
Personnel	Mons/hrs	Rate				
Salaries						
T. Quinn, PI	1.5	8,008	12,012	12,492	12,992	37,497
L. Hauser, Co-PI	1.5	6,300	9,450	9,828	10,221	29,499
K. Naish, Co-PI	1.5	5,800	8,700	9,048	9,410	27,158
TBN, Biologist	12	4,000	48,000	49,920	51,917	149,837
TBN, Grad Student	12	1,455	17,460	18,158	18,885	54,503
TBN, Lab Helper	200	10	2,000	2,000	2,080	6,080
TBN, Trap Operator	400	15	6,000	6,400	6,656	19,056
	Total		103,622	107,847	112,161	323,630
Benefits						
Quinn		22.3%	2,679	2,786	2,897	8,362
Hauser		22.3%	2,107	2,192	2,279	6,578

Naish	22.3%	1,940	2,018	2,098	6,056
Biologist	24.5%	11,760	12,230	12,720	36,710
Grad Res Asst (PhD)	11.7%	2,043	2,125	2,210	6,377
Student Asst	9.7%	194	202	210	606
Student Asst	9.7%	582	605	629	1,817
Total		21,305	22,157	23,043	66,506
Services					
DNA extraction	14	175	2,450	2,450	7,350
PCR runs	140	15	2,100	2,100	6,300
MegaBACE	70	150	10,500	10,500	31,500
Bench fees	12	450	5,400	5,400	16,200
Publications costs, mailing, etc			1,000	1,000	3,000
Total		21,450	21,450	21,450	64,350
Travel					
Travel to research site, mileage			1,260	1,260	3,780
Conference travel			2,000	2,000	6,000
Total			3,260	3,260	9,780
Supplies					
Field supplies, (nets, vials, boots, etc.)			800	800	2,400
Lab supplies			2,000	2,000	6,000
Total			2,800	2,800	8,400
Equipment					
PCs	3	2000	6,000	0	6,000
Smolt trap			15,000	0	15,000
Total			21,000	0	21,000
Graduate Student Operating Fee					
\$2200/quarter in academic year			6,600	6,600	19,800
\$600 summer quarter			600	600	1,800
Total			7,200	7,200	21,600
Total Direct Costs			180,637	164,714	515,265
Facilities & Administrative Costs			78,967	81,587	244,824
51.6% MTDC (less equip & grad student fee)					
TOTAL COSTS			259,604	246,301	760,089
F & A Base			153,037	158,114	474,465

REFERENCES

- Andersson, M. 1994. Sexual Selection. Princeton Univ. Press, Princeton.
- Ayerst, J. D. 1977. The role of hatcheries in rebuilding steelhead runs of the Columbia River system. Pages 84-88 in E. Schwiebert, editor. Columbia River salmon and steelhead. American Fisheries Society, Special Publication 10. Bethesda, Maryland.
- Bartley, D.M., M. Bagley, G. Gall and B. Bentley. 1992. Use of linkage disequilibrium data to estimate effective size of hatchery and natural fish populations. Conservation Biology 6: 365-375.
- Beacham, T. D. 1989. Effect of siblings on growth of juvenile coho salmon (*Oncorhynchus kisutch*). Canadian Journal of Zoology. 67: 601-605.
- Beacham, T. D., and C. B. Murray. 1993. Fecundity and egg size variation in North American Pacific salmon (*Oncorhynchus*). Journal of Fish Biology. 42:485-508.
- Bentzen, P., J. B Olsen, J. McLean, T. R. Seamons, and T. P Quinn. 2001. Kinship analysis of Pacific salmon: insights into mating, homing and timing of reproduction. Journal of Heredity 92: 127-136.

- Berejikian, B. A., E. P. Tezak, and A. L. LaRae. 2000. Female mate choice and spawning behaviour of chinook salmon under experimental conditions. *Journal of Fish Biology*. 57:647-661.
- Berejikian, B. A., E. P. Tezak, L. Park, E. LaHood, S. L. Schroder, and E. Beall. 2001. Male competition and breeding success in captive reared and wild coho salmon (*Oncorhynchus kisutch*). *Canadian Journal of Fisheries and Aquatic Sciences*. 58:804-810.
- Berejikian, B. A. 1995. The effects of hatchery and wild ancestry and experience on the relative ability of steelhead trout fry (*Oncorhynchus mykiss*) to avoid a benthic predator. *Canadian Journal of Fisheries and Aquatic Sciences* 52: 2476-2482.
- Berejikian, B. A., S. B. Mathews, and T. P. Quinn. 1996. Effects of hatchery and wild ancestry and rearing environments on the development of agonistic behavior in steelhead trout (*Oncorhynchus mykiss*) fry. *Canadian Journal of Fisheries and Aquatic Sciences* 53: 2004-2014.
- Berglund, A., Magnhagen, C., Bisazza, A., Konig, B. and Huntingford, F. 1993: Female-female competition over reproduction. *Behavioral Ecology* 4: 184-186.
- Bilton, H. T., D.F. Alderdice, and J.T. Schnute. 1982. Influence of time and size at release of juvenile coho salmon (*Oncorhynchus kisutch*) on returns at maturity. *Canadian Journal of Fisheries and Aquatic Sciences* 39: 426-447.
- Blouin, M.S., M. Parsons, V. Lacaille, and S. Lotz. 1996. Use of microsatellite loci to classify individuals by relatedness. *Molecular Ecology* 5:393-401.
- Brännäs, E. 1995. First access to territorial space and exposure to strong predation pressure: a conflict in early emerging Atlantic salmon (*Salmo salar* L.) fry. *Evolutionary Ecology* 9: 411-420.
- Brannon, E. L. 1987. Mechanisms stabilizing salmonid fry emergence timing. *Canadian Special Publications of Fisheries and Aquatic Sciences* 96: 121-124.
- Brown, G.E. and J.A. Brown. 1993. Social dynamics in salmonid fishes: do kin make better neighbours? *Animal Behaviour* 45: 863-871.
- Burgner, R.L., J.T. Light, L. Margolis, T. Okazaki, A. Tautz and S. Ito. 1992. Distribution and origins of steelhead trout (*Oncorhynchus mykiss*) in offshore waters of the North Pacific Ocean. *International North Pacific Fisheries Commission Bulletin* 51: 92 p.
- Busby, P.J., T.C. Wainwright, G.J. Bryant, L.J. Lierheimer, R.S. Waples, F.W. Waknitz, I.V. Lagomarsino. 1996. Status review of west coast steelhead from Washington, Idaho, Oregon and California. *National Marine Fisheries Service Technical Memorandum NMFS-NWFSC-27*.
- Chebanov, N.A., N.V. Varnavskaya, and V.S. Varnavskiy. 1983. Effectiveness of spawning male sockeye salmon, *Oncorhynchus nerka* (Salmonidae), of differing hierarchical rank by means of genetic-biochemical markers. *Journal of Ichthyology* 23: 51-55.
- Chebanov, N. A. 1989. Behavior, assortative mating and spawning success of the pink salmon, *Oncorhynchus gorbuscha*, at different ratios of spawner size groups on spawning grounds. *Journal of Ichthyology* 29:42-50.
- Chebanov, N. A. 1990. Spawning behavior, assortative mating, and spawning success of coho salmon, *Oncorhynchus kisutch*, under natural and experimental conditions. *Journal of Ichthyology* 30:1-12.
- Chebanov, N. A., and B. E. Riddell. 1998. The spawning behavior, selection of mates, and reproductive success of chinook salmon (*Oncorhynchus tshawytscha*) spawners of natural and hatchery origins under conditions of joint spawning. *Journal of Ichthyology* 38:517-526.
- Chilcote, M.W., S.A. Leider and J.J. Loch. 1986. Differential reproductive success of hatchery and wild summer-run steelhead under natural conditions. *Transactions of the American Fisheries Society* 115: 726-735.

- Clutton-Brock, T. H. and Parker G. A. 1992: Potential reproductive rates and the operation of sexual selection. *Quarterly Review of Biology* 67: 437-456.
- Cornuet J.M., Piry S., Luikart G., Estoup A., Solignac M. 1999. New methods employing multilocus genotypes to select or exclude populations as origins of individuals. *Genetics* 153:1989-2000
- Crawford, B. A. 1979. The origin and history of the trout brood stocks of the Washington Department of Game. Washington State Game Department, Fishery Research Report, Olympia.
- Courtenay, S.C., Quinn, T.P., Dupuis, H.M.C., Groot, C. and Larkin, P.A. 1997. Factors affecting the recognition of population-specific odours by juvenile coho salmon (*Oncorhynchus kisutch*). *Journal of Fish Biology* 50: 1042-1060.
- Davis, B., B. Allee, D. Amend, B. Bachen, B. Davidson, T. Gharrett, S. Marshall and A. Wertheimer. 1985. Alaska Department of Fish and Game Genetic policy. Alaska Department of Fish and Game, Anchorage, AK.
- Dickerson, B. R., M. F. Willson and T. P. Quinn. 2002. Body size, arrival date, and reproductive success of pink salmon, *Oncorhynchus gorbuscha*. *Ethology, Ecology, and Evolution* (in press).
- Doyle, R.W., C. Herbinger, C.T. Taggart, and S. Lochmann. 1995. Use of DNA microsatellite polymorphism to analyze genetic correlations between hatchery and natural fitness. *American Fisheries Society Symposium* 15:205-211.
- Einum, S., and I. A. Fleming. 1999. Maternal effects of egg size in brown trout (*Salmo trutta*): norms of reaction to environmental quality. *Proceedings of the Royal Society of London B*. 266:2095-2100.
- Einum, S., and I. A. Fleming. 2000. Selection against late emergence and small offspring in Atlantic salmon (*Salmo salar*). *Evolution*. 54:628-639.
- Emlen, S. T. and Oring, L. 1977. Ecology, sexual selection, and the evolution of mating systems. *Science* 197: 215-223.
- Flagg, T. A., F. W. Waknitz, D. J. Maynard, G. B. Milner, and C. V. W. Mahnken. 1995. The effects of hatcheries on native coho salmon populations in the lower Columbia River. *American Fisheries Society Symposium* 15: 366-375.
- Fleming, I.A. and M.R. Gross. 1994. Breeding competition in a Pacific salmon (Coho: *Oncorhynchus kisutch*): measures of natural and sexual selection. *Evolution* 48: 637-657.
- Fleming, I. A., K. Hindar, I. B. Mjølnerød, B. Jonsson, T. Balstad, and A. Lamberg. 2000. Lifetime success and interactions of farm salmon invading a native population. *Proceedings of the Royal Society of London B*. 267:1517-1523.
- Foote, C.J. 1988. Male mate choice dependent on male size in salmon. *Behaviour* 106: 63-80.
- Foote, C.J. and P.A. Larkin. 1988. The role of male choice in assortative mating of anadromous and non-anadromous sockeye salmon (*Oncorhynchus nerka*). *Behaviour* 106: 43-62.
- Foote, C. J., G. S. Brown, and C. C. Wood. 1997. Spawning success of males using alternative mating tactics in sockeye salmon, *Oncorhynchus nerka*. *Canadian Journal of Fisheries and Aquatic Sciences* 54:1785-1795.
- Gharrett, A. J., and S. M. Shirley. 1985. A genetic examination of spawning methodology in a salmon hatchery. *Aquaculture* 47: 245-256.
- Gharrett, A.J. and W.W. Smoker. 1993. Genetic components in life history traits contribute to population structure. p. 197-202. In J. G. Cloud, and G. H. Thorgaard [eds.], *Genetic conservation of salmonid fishes*. Plenum Press, N.Y.
- Gile, S. R., and M. M. Ferguson. 1995. Factors affecting male potency in pooled gamete crosses of rainbow trout, *Oncorhynchus mykiss*. *Environmental Biology of Fishes*. 42:267-275.
- Geiger, H. J., W. W. Smoker, L. A. Zhivotovsky, and A. J. Gharrett. 1997. Variability of family size and marine survival in pink salmon (*Oncorhynchus gorbuscha*) has implications for conservation biology and human use. *Canadian Journal of Fisheries and Aquatic Sciences* 54: 2684-2690.

- Gross, M.R. 1985. Disruptive selection for alternative life histories in salmon. *Nature* 313: 47-48.
- Hanson, A.J. and H.D. Smith. 1967. Mate selection in a population of sockeye salmon (*Oncorhynchus nerka*) of mixed age-groups. *Journal of the Fisheries Research Board of Canada* 24: 1955-1977.
- Helle, J. H. 1989. Relation between size-at-maturity and survival of progeny in chum salmon, *Oncorhynchus keta* (Walbaum). *Journal of Fish Biology*. 35(Supplement A): 99-107.
- Hendry, A.P., O.K. Berg and T.P. Quinn. 1999. Breeding date, life history, and energy allocation in a population of sockeye salmon (*Oncorhynchus nerka*). *Oikos* 85: 499-514.
- Herbinger, C.M., R.W. Doyle, E.R. Pitman, D. Paquet, K.A. Mesa, D.B. Morris, J.M. Wright, and D. Cook. 1995. DNA fingerprint based analysis of paternal and maternal effects on offspring growth and survival in communally reared rainbow trout. *Aquaculture* 137:245-256.
- Hilborn, R. 1992. Hatcheries and the future of salmon in the northwest. *Fisheries* 17: 5-8.
- Hilborn, R., and D. Eggers. 2000. A review of the hatchery programs for pink salmon in Prince William Sound and Kodiak Island, Alaska. *Transactions of the American Fisheries Society*. **129**: 333-350.
- Hindar, K., N. Ryman and F. Utter. 1991. Genetic effects of cultured fish on natural fish populations. *Canadian Journal of Fisheries and Aquatic Sciences* 48: 945-957.
- Holtby, L. B., B.C. Andersen, and R.K. Kadowaki. 1990. Importance of smolt size and early ocean growth to interannual variability in marine survival of coho salmon (*Oncorhynchus kisutch*). *Canadian Journal of Fisheries and Aquatic Sciences* 47: 2181-2194.
- Holtby, L.B. and M.C. Healey. 1986. Selection for adult size in female coho salmon (*Oncorhynchus kisutch*). *Canadian Journal of Fisheries and Aquatic Sciences* 43: 1946-1959.
- Hughes, C. 1998. Integrating molecular techniques with field methods in studies of social behavior: a revolution in results. *Ecology* 79:383-399.
- Hutchings, J. A., and R. A. Myers. 1988. Mating success of alternative maturation phenotypes in male Atlantic salmon, *Salmo salar*. *Oecologia*. 75:169-174.
- Integrated Hatchery Operations Team (IHOT). 1994. Policies and procedures for Columbia Basin anadromous salmonid hatcheries - Draft. Bonneville Power Administration. 115 pages.
- Keenleyside, M. H. A., and H. M. C. Dupuis. 1988. Courting and spawning competition in pink salmon (*Oncorhynchus gorbuscha*). *Canadian Journal of Zoology*. **66**: 262-265.
- Leary, R.F., F.W. Allendorf and K.L. Knudsen. 1989. Genetic differences among rainbow trout spawned on different days within a single season. *Progressive Fish-Culturist* 51: 10-19.
- Leider, S.A. 1985. Precise timing of upstream migrations by repeat steelhead spawners. *Transactions of the American Fisheries Society* 114: 906-908.
- Leider, S.A., P.L. Hulett, J.J. Loch, and M.W. Chilcote. 1990. Electrophoretic comparison of the reproductive success of naturally spawning transplanted and wild steelhead through the returning adult stage. *Aquaculture* 88: 239-252.
- Light, J.T. 1987. Coastwide abundance of North American steelhead trout. Univ. of Washington, Fisheries Research Institute, report to the International North Pacific Fisheries Commission by the U.S. National Section.
- Mackey, G., J. E. McLean and T. P. Quinn. 2001. Comparisons of run timing, spatial distribution, and length of wild and newly-established hatchery populations of steelhead in Forks Creek, Washington. *North American Journal of Fisheries Management* 21: 717-724.
- Maekawa, K. and H. Onozato. 1986. Reproductive tactics and fertilization success of mature male Miyabe charr, *Salvelinus malma miyabei*. *Environmental Biology of Fishes* 15: 119-129.
- Marshall, T.C., J. Slate, L.E.B. Kruuk, and J.M. Pemberton. 1998. Statistical confidence for likelihood-based paternity influence in natural populations. *Molecular Ecology* 7:639-656.

- Mason, J.C., and D.W. Chapman. 1965. Significance of early emergence, environmental rearing capacity, and behavioral ecology of juvenile coho salmon in stream channels. *Journal of the Fisheries Research Board of Canada* 22: 173-190.
- Maynard, D. J., T. A. Flagg, and C. V. W. Mahnken. 1995. A review of seminatural culture strategies for enhancing the postrelease survival of anadromous salmonids. *American Fisheries Society Symposium* 15: 307-314.
- Maynard, D. J., T. A. Flagg, C. V. W. Mahnken, and S. L. Schroder. 1996. Natural rearing technologies for increasing postrelease survival of hatchery-reared salmon. *Bulletin of the National Research Institute of Aquaculture, Supplement* 2: 71-77.
- Maynard, D. J., G. C. McDowell, E. P. Tezak, and T. A. Flagg. 1996. Effect of diets supplemented with live food on the foraging behavior of cultured fall chinook salmon. *The Progressive Fish-Culturist* 58:187-191.
- McLean, J.E., Bentzen, P., and Quinn, T.P. 2003a. Differential reproductive success of sympatric, naturally spawning hatchery and wild steelhead (*Oncorhynchus mykiss*). *Environmental Biology of Fishes* (in press).
- McLean, J.E., Bentzen, P., and Quinn, T.P. 2003b. Differential reproductive success of sympatric, naturally-spawning hatchery and wild steelhead through the adult stage. *Canadian Journal of Fisheries and Aquatic Sciences* (in press).
- McGinnity, P., and eight co-authors. 1997. Genetic impact of escaped farmed Atlantic salmon (*Salmo salar* L.) on native populations: use of DNA profiling to assess a freshwater performance of wild, farmed, and hybrid progeny in a natural river environment. *ICES Journal of Marine Science* 54:998-1008.
- Meagher, T.R. 1986. Analysis of paternity within a natural population of *Chamaelirium luteum*. I. Identification of most-likely male parents. *American Naturalist* 128:199-215.
- Meffe, G.K. 1987. Conserving fish genomes: philosophies and practices. *Environmental Biology of Fishes* 18: 3-9.
- Morán, P., A. M. Pendás, E. Beall, and E. García-Vázquez. 1996. Genetic assessment of the reproductive success of Atlantic salmon precocious parr by means of VNTR loci. *Heredity* 77:655-660.
- National Research Council. 1996. *Upstream: Salmon and Society in the Pacific Northwest*. National Academy Press, Washington, D.C.
- Nickelson, T. E., M. F. Solazzi, and S. L. Johnson. 1986. Use of hatchery coho salmon (*Oncorhynchus kisutch*) presmolts to rebuild wild populations in Oregon coastal streams. *Canadian Journal of Fisheries and Aquatic Sciences* 43: 2443-2449.
- Nielsen, J. L. 1994. Invasive cohorts: impacts of hatchery-reared coho salmon on the trophic, developmental, and genetic ecology of wild stocks, p. 361-385. *In: Theory and application in fish feeding ecology*. K. L. Fresh, D. J. Stouder, and R. J. Feller (eds.). University of South Carolina Press, Columbia, S.C.
- O'Connell, M., and J.M. Wright. 1997. Microsatellite DNA in fishes. *Reviews in Fish Biology and Fisheries* 7:331-353.
- O'Reilly, P.T., C. Herbinger, and J.M. Wright. 1998. Analysis of parentage determination in Atlantic salmon (*Salmo salar*) using microsatellites. *Animal Genetics* 29:363-370
- O'Reilly, P.T., and J.M. Wright. 1995. The evolving technology of DNA fingerprinting and its application to fisheries and aquaculture. *Journal of Fish Biology* 47 (Suppl. A):29-55.
- Perrin, C. J., and J. R. Irvine. 1990. A review of survey life estimates as they apply to the area-under-the-curve method for estimating the spawning escapement of Pacific salmon. *Canadian Technical Report of Fisheries and Aquatic Sciences*. 1733:1-49.
- Queller, D.C., J.E. Strassmann and C.R. Hughes. 1993. Microsatellites and kinship. *TREE* 8: 285-288.
- Quinn, T.P. 1993. A review of homing and straying of wild and hatchery-produced salmon. *Fisheries Research* 18: 29-44.

- Quinn, T.P. 1999. Variation in Pacific salmon reproductive behaviour associated with species, sex and levels of competition. *Behaviour* 136: 179-204.
- Quinn, T.P. 1999. Revisiting the stock concept in Pacific salmon: insights from Alaska and New Zealand. *Northwest Science* 73: 312-324.
- Quinn, T.P. and C.A. Busack. 1985. Chemosensory recognition of siblings in juvenile coho salmon (*Oncorhynchus kisutch*). *Animal Behaviour* 33:51-56.
- Quinn, T.P. and C.J. Foote. 1994. The effects of body size and sexual dimorphism on the reproductive behaviour of sockeye salmon, *Oncorhynchus nerka*. *Animal Behaviour* 48: 751-761.
- Quinn, T.P., A.H. Dittman, N.P. Peterson and E.C. Volk. 1994. Distribution, survival and growth of sibling groups of juvenile coho salmon, *Oncorhynchus kisutch*, in an experimental stream channel. *Canadian Journal of Zoology* 72: 2119-2123.
- Quinn, T.P., M.D. Adkison and M.B. Ward. 1996. Behavioral tactics of male sockeye salmon (*Oncorhynchus nerka*) under varying operational sex ratios. *Ethology* 102: 304-322.
- Quinn, T.P. and N.P. Peterson. 1996. The influence of habitat complexity and fish size on over-winter survival and growth of individually-marked juvenile coho salmon (*Oncorhynchus kisutch*) in Big Beef Creek, Washington. *Canadian Journal of Fisheries and Aquatic Sciences* 53: 1555-1564.
- Quinn, T. P., M. J. Unwin, and M. T. Kinnison. 2000. Evolution of temporal isolation in the wild: genetic divergence in timing of migration and breeding by introduced chinook salmon populations. *Evolution* 54: 1372-1385.
- Quinn, T. P., J. A. Peterson, V. Gallucci, W. K. Hershberger, and E. L. Brannon. 2002. Artificial selection and environmental change: countervailing factors affecting the timing of spawning by coho and chinook salmon. *Transactions of the American Fisheries Society* 131: 591-598.
- Raymond M, Rousset F (1995). GENETPOP: a population genetic software for exact tests and ecumenicism. *J. Heredity* 86: 248-249.
- Reisenbichler, R. R., and J. D. McIntyre. 1977. Genetic differences in growth and survival of juvenile hatchery and wild steelhead trout, *Salmo gairdneri*. *Journal of the Fisheries Research Board of Canada* 34:123-128.
- Rhodes, J.S. and T.P. Quinn. 1998. Factors affecting the outcome of territorial contests between hatchery and naturally reared coho salmon parr in the laboratory. *Journal of Fish Biology* 53: 1220-1230.
- Ricker, W. E. 1972. Hereditary and environmental factors affecting certain salmonid populations. pp. 19-160. In: *The Stock Concept in Pacific Salmon*, R.C. Simon and P.A. Larkin (eds.). H.R. MacMillan Lectures in Fisheries, Univ. of British Columbia, Vancouver, B.C.
- Riddell, B. E. 1993. Spatial organization of Pacific salmon: what to conserve? p. 23-41. In J. G. Cloud, and G. H. Thorgaard [eds.], *Genetic conservation of salmonid fishes*. Plenum Press, N.Y.
- Ryman N., P.E. Jorde and L. Laikre. 1995. Supportive breeding and variance effective population size. *Conservation Biology* 9. 1619-1628.
- Schroder, S.L. 1981. The role of sexual selection in determining the overall mating patterns and mate choice in chum salmon. PhD dissertation, Univ. of Washington, Seattle, Washington.
- Scudder, G.G.E. 1989. The adaptive significance of marginal populations: a general perspective. *Canadian Special Publication of Fisheries and Aquatic Sciences* 105: 180-185.
- Siitonen, L., and G.A.E. Gall. 1989. Response to selection for early spawn date in rainbow trout, *Salmo gairdneri*. *Aquaculture* 78: 153-161.
- Slate J., T. C. Marshall and J. M. Pemberton. 2000. A retrospective assessment of the accuracy of the paternity inference program CERVUS. *Molecular Ecology* 9: 801-808.

- Smoker, W. W., A. J. Gharrett, and M. S. Stekoll. 1998. Genetic variation of return date in a population of pink salmon: a consequence of fluctuating environment and dispersive selection? *Alaska Fishery Research Bulletin* 5:46-54.
- Steen, R.P. and T.P. Quinn. 1999. Egg burial depth by sockeye salmon (*Oncorhynchus nerka*): implications for survival of embryos and natural selection on female body size. *Canadian Journal of Zoology* 77: 836-841.
- Stouder, D.J., P.A. Bisson and R.J. Naiman (eds.). 1996. Pacific salmon and their ecosystems: status and future options. Chapman and Hall, New York.
- Taylor, E. B. 1991. A review of local adaptation in Salmonidae, with particular reference to Pacific and Atlantic salmon. *Aquaculture* 98: 185-207.
- Thompson, E.A. 1975. The estimation of pairwise relationships. *Annals of Human Genetics* 39:173-188.
- Thompson, E.A. 1976. Inference of genealogical structure. *Social Science Information* 15:477-526.
- Thorne, R. E., and J. J. Ames. 1987. A note on variability of marine survival of sockeye salmon (*Oncorhynchus nerka*) and effects of flooding on spawning success. *Canadian Journal of Fisheries and Aquatic Sciences* 44:1791-1795.
- Tipping, J.M., and H.L. Blankenship. 1993. Effect of condition factor at release on smolt-to-adult survival of hatchery sea-run cutthroat trout. *Progressive Fish-Culturist* 55: 184-186.
- Unwin, M. J., M. T. Kinnison, N. C. Boustead and T. P. Quinn. 2003. Genetic control over survival in Pacific salmon (*Oncorhynchus* spp.): experimental evidence between and within populations of New Zealand chinook salmon (*O. tshawytscha*). *Canadian Journal of Fisheries and Aquatic Sciences* 60: 1-11.
- Utter, F. M. 1998. Genetic problems of hatchery-reared progeny released into the wild, and how to deal with them. *Bulletin of Marine Science*. 62:623-640.
- Wade, M. J. and Arnold, S. J. 1980. The intensity of sexual selection in relation to male sexual behaviour, female choice and sperm precedence. *Animal Behaviour* 28: 446-461.
- Waples, R.S. 1989. A generalized approach for estimating effective population size from temporal changes in allele frequency. *Genetics* 121:379-391.
- Waples, R.S. 1990a. Conservation genetics of Pacific salmon. II. Effective population size and the rate of loss of genetic variability. *Journal of Heredity* 81: 267-276.
- Waples, R.S. 1990b. Conservation genetics of Pacific salmon. III. Estimating effective population size. *Journal of Heredity* 81: 277-289.
- Waples, R.S. 1991. Genetic interactions between hatchery and wild salmonids: lessons from the Pacific Northwest. *Canadian Journal of Fisheries and Aquatic Sciences* 48 (Suppl. 1): 124-133.
- Waples, R.S. 1995. Evolutionarily Significant Units and the conservation of biological diversity under the Endangered Species Act. *American Fisheries Society Symposium* 17: 8-27.
- Waples, R.S. and D.J. Teel. 1990. Conservation genetics of Pacific salmon. I. Temporal changes in allele frequency. *Conservation Biology* 4: 144-156.
- Ward, B.R., P.A. Slaney, A.R. Facchin, and R.W. Land. 1989. Size-biased survival in steelhead trout (*Oncorhynchus mykiss*): back-calculated lengths from adults' scales compared to migrating smolts at the Keogh River, British Columbia. *Canadian Journal of Fisheries and Aquatic Sciences* 46: 1853-1858.
- Withler, R. E., and T. D. Beacham. 1994. Genetic consequences of the simultaneous or sequential addition of semen from multiple males during hatchery spawning of chinook salmon (*Oncorhynchus tshawytscha*). *Aquaculture* 126:11-23.
- Wright, J.M. and P. Bentzen. 1994. Microsatellites: genetic markers for the future. *Rev. in Fish Biol. and Fisheries* 4:384-388.
- Wright, S. 1951. The genetical structure of populations. *Annals of Eugenetics* 15. 15:323-354.
- Wright, S. 1965. The interpretation of population structure by F-statistics with special regard to systems of mating. *Evolution* 19:395-420.

- Wright, S. 1993. Fishery management of wild Pacific salmon stocks to prevent extinctions. *Fisheries* **18**(5): 3-4.
- Ziegle, J.S., Y. Su, K.P. Corcoran, L. Nie, P.E. Mayrand, L.B. Hoff, L.J. McBride, M.N. Kronick and S.R. Diehl. 1992. Application of automated DNA sizing technology for genotyping microsatellite loci. *Genomics* 14:1026-1031.

BIOGRAPHICAL SKETCHES

Name: Thomas P. Quinn
Address: School of Aquatic and Fishery Sciences, Box 355020
Univ. of Washington
Seattle, WA 98195
Phone: (206) 543-9042
e-mail: tqinn@fish.washington.edu

Education: B.A. with Distinction in Biology, Swarthmore College, 1976
M.S. in Fisheries, University of Washington, 1978
Ph.D. in Fisheries, University of Washington, 1981

Employment:

2000-present	Professor, School of Aquatic and Fishery Sciences
1990-2000	Associate Professor, School of Fisheries,
1986-1990	Assistant Professor, School of Fisheries, Univ. of Washington
1984-1985	Research Associate, Department of Oceanography, University of British Columbia, Vancouver, B.C.
1981-1984	Post-doctoral Fellow, University of British Columbia, and Pacific Biological Station, Nanaimo, B. C., Canada

Research Interests:

Behavior, ecology and evolution of salmonid fishes, with emphasis on migrations, life history patterns, reproduction, habitat requirements and conservation.

Selected Relevant Publications (total = 167)

- McLean, J.E., P. Bentzen, and **T.P. Quinn**. 2003. Differential reproductive success of sympatric, naturally-spawning hatchery and wild steelhead through the adult stage. *Canadian Journal of Fisheries and Aquatic Sciences* (in press).
- Seamons, T.R., P. Bentzen and **T.P. Quinn**. The mating system of steelhead (*Oncorhynchus mykiss*), inferred by molecular analysis of parents and progeny. *Environmental Biology of Fishes* (in press).
- Quinn, T.P.**, M.T. Kinnison and M.J. Unwin. 2001. Evolution of chinook salmon (*Oncorhynchus tshawytscha*) populations in New Zealand: pattern, rate, and process. *Genetica* 112/113: 493-513.
- Hendry, A. P., J. K. Wenburg, P. Bentzen E. C. Volk and **T. P. Quinn**. 2000. Rapid evolution of reproductive isolation in the wild: evidence from introduced salmon. *Science* 290: 516-518.
- Quinn, T.P.** 1993. A review of homing and straying of wild and hatchery-produced salmon. *Fisheries Research* 18: 29-44.

Name: Lorenz Hauser
Address: School of Aquatic and Fishery Sciences, Box 355020
Univ. of Washington
Seattle, WA 98195
Phone: (206) 543-4270

Education

1992-1996 **Ph.D.**; University of Wales, Swansea. Thesis "Genetic and morphological differentiation of native and introduced populations of the Lake Tanganyika sardine *Limnothrissa miodon*"
1989-1990 **M.Sc. Fisheries Biology & Management** Course; University College of North Wales, Bangor. Thesis: "Effects of sea trout stocking on the population genetics of landlocked brown trout (*Salmo trutta*)"
1983-1989 **M.Sc. Zoology**; University of Vienna, Austria. Thesis: "Comparative investigations on the food selectivity of 0+ cyprinids"

Employment:

2002- Assistant Professor, University of Washington, Seattle, USA
1996 - 2002 University Research Fellow in Molecular Ecology (Univ. of Hull)
1994 - 1995 Research Assistant (University of Wales, Swansea)
Funded by: The Leverhulme Trust, The Royal Society
Sept-Nov 1994 Visiting Scientist at the Fisheries Centre, University of British Columbia, Vancouver, Canada
1991 - 1994 Research Assistant (University of Wales, Swansea)
Funded by: Overseas Development Administration
1988 - 1989 Part-time Research Assistant (University of Vienna)

Research Interests: Molecular ecology of fishes, with emphasis on the patterns of evolution and population structure of introduced populations.

Selected Relevant Publications (total = 16)

- Hauser L.**, Adcock GJ, Smith PJ, Bernal Ramirez JH, Carvalho GR (2002) Loss of microsatellite diversity and low effective population size in an overexploited population of New Zealand snapper (*Pagrus auratus*). *Proceedings of the National Academy of Sciences* **99**, 11742-11747.
- Hauser L.**, Carvalho G.R., Pitcher, T.J. & Ogutu-Ohwayo R. (1998) Genetic affinities of an introduced predator: Nile perch in Lake Victoria, East Africa. *Molecular Ecology* **7**, 849-857.
- Carvalho G.R., Shaw P.W., **Hauser L.**, Seghers B.H. & Magurran A.E. (1996) Artificial introductions, evolutionary change and population differentiation in Trinidadian guppies (*Poecilia reticulata*: Poeciliidae). *Biological Journal of the Linnean Society* **57**, 219-234.
- Hauser L.**, Carvalho G.R. & Pitcher T.J. (1995) Morphological and genetic differentiation of a clupeid (*Limnothrissa miodon*) 34 years after introduction into Lake Kivu, East Africa. *Journal of Fish Biology* **47**, 127-144.
- Carvalho G.R. & **Hauser L.** (1994) Molecular genetics and the stock concept in fisheries. *Reviews in Fisheries and Fish Biology*, **4**, 326-350.
- Hauser L.**, Beaumont A.R., Marshall G.T.H. & Wyatt R.J. (1991) Effects of sea trout stocking on the population genetics of landlocked brown trout, *Salmo trutta* L., in the Conwy River system, North Wales, U.K. *Journal of Fish Biology*, **39** (A), 109-116.

Name: Kerry Naish
Title: Assistant Professor
Department: School of Aquatic and Fishery Sciences
Institution: University of Washington
Address: 1122 NE Boat Street, Seattle, WA 98105
Tel: (206) 221 6375
Email: knaish@u.washington.edu

Education:

PhD, Biological Sciences, University of Wales, Swansea, United Kingdom, 1993
MSc, Ichthyology and Fisheries Science, Rhodes University, Grahamstown, South Africa, 1990
BSc (Hons), *cum laude*, Ichthyology and Fisheries Science, Rhodes University, Grahamstown, South Africa, 1989
BSc, Zoology, University of Cape Town, Cape Town, South Africa, 1988

Professional experience/ Positions held:

Assistant Professor, School of Aquatic and Fishery Sciences, University of Washington, 2001 – present.
Senior Research Associate, National Research Council, 1999 -2000
Postdoctoral Fellow, University of Guelph, Guelph, Ontario, Canada, 1997 -1998
Aquaculture Geneticist, Overseas Development Administration, Swansea, UK, 1994 – 1996

Research interests: Genetic basis of fitness traits, with emphasis on conservation genetics and the genetic effects of enhancement & restoration

Selected Publications:

- Woram RA, Gharbi K, Sakamoto T, Hoyheim B, Holm LE, **Naish K**, McGowan C, Ferguson MM, Phillips RB, Stein J, Guyomard R, Cairney M, Taggart JB, Powell R, Davidson W, Danzmann RG 2003 Comparative genome analysis of the primary sex determining locus in five species of salmonid fish. *Genome Research* **13** 272-280.
- Naish KA**, Park LK 2002. Linkage relationships for thirty-five new microsatellite loci for genome mapping in the chinook salmon, *Oncorhynchus tshawytscha*. *Animal Genetics*.
- Naish KA** and Boulding EG 2001. Trinucleotide microsatellites for the zebra mussel *Dreissena polymorpha*, an invasive species in North America and Europe. *Molecular Ecology Notes* **1**, 286-288.
- Wilson AB, **Naish KA** and Boulding EG 1999. Multiple dispersal strategies of the invasive quagga mussel (*Dreissena bugensis*) as revealed by microsatellite analysis. *Canadian Journal of Fisheries and Aquatic Sciences* **56**, 2248-2261.
- Naish KA**, Skibinski DOF (1997) Review of techniques applied to the characterization of tilapia species and populations. *Int. Workshop on the Characterization of Ghanaian Tilapia Genetic Resources For Use in Fisheries and Aquaculture, Accra (Ghana), 4-7Jun 1996* Pullin, RSV; Casal, CMV; Abban, EK; Falk, TM (eds), pp. 3-5, ICLARM Conf. Proc., no. 52, ICLARM, Makati City (Philippines).
- Naish KA**, Warren M, Bardakci F, Skibinski DOF, Carvalho GR and Mair GR 1995. Multilocus DNA fingerprinting and RAPD reveal similar genetic relationships between strains of *Oreochromis niloticus* (Pisces: Cichlidae). *Molecular Ecology* **4**, 271-274