INFORMATION TO USERS

This manuscript has been reproduced from the microfilm master. UMI films the text directly from the original or copy submitted. Thus, some thesis and dissertation copies are in typewriter face, while others may be from any type of computer printer.

The quality of this reproduction is dependent upon the quality of the copy submitted. Broken or indistinct print, colored or poor quality illustrations and photographs, print bleedthrough, substandard margins, and improper alignment can adversely affect reproduction.

In the unlikely event that the author did not send UMI a complete manuscript and there are missing pages, these will be noted. Also, if unauthorized copyright material had to be removed, a note will indicate the deletion.

Oversize materials (e.g., maps, drawings, charts) are reproduced by sectioning the original, beginning at the upper left-hand corner and continuing from left to right in equal sections with small overlaps.

ProQuest Information and Learning
300 North Zeeb Road, Ann Arbor, MI 48106-1346 USA
800-521-0600

UMI®
Conservation Value of Alternative Life-History Tactics in Steelhead Trout

*(Oncorhynchus mykiss)*

By

Joshua Clyde Walters

A thesis submitted in conformity with the requirements
for the degree of Master of Science,
Department of Zoology
University of Toronto

2005

© Copyright by Joshua Clyde Walters, 2005
NOTICE:
The author has granted a non-exclusive license allowing Library and Archives Canada to reproduce, publish, archive, preserve, conserve, communicate to the public by telecommunication or on the Internet, loan, distribute and sell theses worldwide, for commercial or non-commercial purposes, in microform, paper, electronic and/or any other formats.

The author retains copyright ownership and moral rights in this thesis. Neither the thesis nor substantial extracts from it may be printed or otherwise reproduced without the author’s permission.

In compliance with the Canadian Privacy Act some supporting forms may have been removed from this thesis.

While these forms may be included in the document page count, their removal does not represent any loss of content from the thesis.

AVIS:
L’auteur a accordé une licence non exclusive permettant à la Bibliothèque et Archives Canada de reproduire, publier, archiver, sauvegarder, conserver, transmettre au public par télécommunication ou par l’Internet, prêter, distribuer et vendre des thèses partout dans le monde, à des fins commerciales ou autres, sur support microforme, papier, électronique et/ou autres formats.

L’auteur conserve la propriété du droit d’auteur et des droits moraux qui protège cette thèse. Ni la thèse ni des extraits substantiels de celle-ci ne doivent être imprimés ou autrement reproduits sans son autorisation.

Conformément à la loi canadienne sur la protection de la vie privée, quelques formulaires secondaires ont été enlevés de cette thèse.

Bien que ces formulaires aient inclus dans la pagination, il n’y aura aucun contenu manquant.
Conservation Value of Alternative Life-History Tactics in Steelhead Trout

(Oncorhynchus mykiss)

Abstract of a thesis submitted in conformity with the requirements for the degree of Master of Science, Department of Zoology

University of Toronto

Joshua Clyde Walters

Abstract

Salmonid conservation programs typically include captive breeding, fishery regulation and habitat enhancement. This research addresses the consequences of these programs for the demography, ecology, life-history, genetics, and phenotypes of steelhead trout (Oncorhynchus mykiss) in the Keogh River, British Columbia.

The conservation program altered life-history by producing resident hatchery fish, which altered ecology by consuming resources and conspecifics. Residents had phenotypes that diverged from the wild type, and potentially altered genetics and demography by participating in both wild and hatchery anadromous matings.

Two possible negative consequences are an increase in density-dependent competition in the freshwater environment, and a decrease in marine survival due to phenotypic change. Positive outcomes are an increase in total males spawning and thus increased genetic diversity, decreased genetic drift, decreased inbreeding due to breeding across age cohorts, and increased gene flow between hatchery and wild fish. The net effect for conservation remains to be determined.
Acknowledgments

I am truly grateful to my supervisor, Mart Gross, for his advice, guidance, and encouragement. His passion for conservation is contagious and he kept me focused on seeing “the big picture”. He provided a wealth of opportunities that have greatly benefited my academic and personal development. Thank you for being my mentor.

I am grateful to my committee member Donald Jackson for his expertise in statistics and experimental design, and for his support throughout the thesis.

I am also thankful for the financial support provided by NSERC (through a grant to Mart Gross) and by the Department of Zoology at the University of Toronto.

I acknowledge Bruce Ward, and the Ministry of the Environment, for access to facilities, historic data, and knowledge gained through three decades of observation at the Keogh River. I thank Don McCubbing, Lloyd Burroughs, and Peter Troffe for excellent support and advice at the field site. Thanks to Ken Fuller, and the North Vancouver Island Salmonid Enhancement Association, for access to facilities and equipment. Thanks to Dan Hayward, and the Vancouver Island Trout Hatchery (VITH), for information on LGB production, as well as access to VITH facilities.

I also thank Bruce Morrison, and the Ontario Ministry of Natural Resources, for providing access to their coded wire tag reading facilities at the Glenora Fisheries Station.

Within the Gross Lab, I greatly benefited from the assistance, discussions, and friendship of my lab mates. Thanks to Aviva Patel, Cory Robertson, Michelle Herzog, Ian Craine, Carli Halpenny, Ho Young Suk, Cristian Correa, Andrea Bravo and Jamon Camisso – your support is very much appreciated.

And lastly, I would like to thank my family and friends. The unwavering support and encouragement of my parents, Clyde and Sheila Walters, helped me to get here and continues to propel me onwards. Thanks to my friends in both Toronto and BC – you motivated and entertained me along the way. I would like to especially thank Ilke van Hazel. Her support has been phenomenal and diverse, ranging from in-depth scientific discussions to maintaining a smile on my face. Thank you.
Table of Contents

Abstract ......................................................................................................................................................... ii

Acknowledgments ........................................................................................................................................ iii

Table of Contents ........................................................................................................................................ iv

List of Tables ................................................................................................................................................ v

List of Figures ............................................................................................................................................... vii

List of Appendices ..................................................................................................................................... viii

Part 1: Introduction .................................................................................................................................. 1

Part 2: Conservation of a wild fish population (steelhead trout): influences of captive breeding, habitat improvement and fishing regulations on alternative life histories ........................................... 3

  Introduction .............................................................................................................................................. 4

  Methods ..................................................................................................................................................... 7

  Results ....................................................................................................................................................... 12

  Discussion ............................................................................................................................................... 17

Part 3: Phenotypic divergence in a conservation hatchery ..................................................................... 46

  Introduction ............................................................................................................................................ 47

  Methods ................................................................................................................................................... 49

  Results ..................................................................................................................................................... 54

  Discussion .............................................................................................................................................. 57

Part 4: Conclusions and Recommendations .............................................................................................. 77

Appendices .................................................................................................................................................. 83

Literature Cited ........................................................................................................................................... 94
List of Tables

Part 2

Table 1. Terms used to differentiate among wild and LGB steelhead life history tactics, ontogenetic stages, and cohorts ................................................................. 26

Table 2. Timing and methods of sample collection, data collected and sample size for developmental stages and life history types in wild and LGB origin fish .......... 27

Table 3. Population estimates, sex ratio, average mass, and estimated biomass of LGB and wild fish in 2004 .................................................................................. 28

Table 4. Population estimates for each river reach from snorkel surveys conducted in 2003 ...... 29

Table 5. Freshwater, ocean and total ages of male and female wild and LGB anadromous adult steelhead from the Keogh River in 2004 ..................................................... 30

Table 6. Age classification of wild and LGB juveniles from the Keogh River ......................... 31

Table 7. Gonad factor scores, by sex, for wild and LGB smolt and parr sampled in 2004 ....... 32

Table 8. Mass and fork lengths of wild smolt, LGB smolt and 2004 LGB parr expressing various gonad factor scores ................................................................. 33

Table 9. Potential egg deposition by wild and LGB anadromous and resident females from 2002-2004 ........................................................................................................... 34

Table 10. LGB parr feeding success and the diversity of invertebrates each fish type consumed at the Keogh River in 2004 ................................................................. 35

Table 11. Allometrically scaled relative food consumption for wild and LGB fish on an individual- and population-specific basis during summer 2004 ......................... 36

Table 12. Issues for wild steelhead conservation of various steelhead-coastal cutthroat trout hybridization rates in LGB progeny ......................................................... 37

Part 3

Table 1. Fish types differentiated by life stage, origin, and sampling period .................. 64

Table 2. Multi-response permutation procedure results for comparisons of shape variables .... 65

Table 3. Linear regression statistics and parameters for 17 non-geometric variables ............ 66

Reproduced with permission of the copyright owner. Further reproduction prohibited without permission.
Table 4. Multi-response permutation procedure results for comparisons of non-geometric morphological variables................................................................. 67

Table 5. Fish-type specific mean residuals from independent regressions of each non-geometric variable on body size ................................................................. 68

Table 6. Average mass, length, and centroid size for the wild and LGB origin life histories..... 69

**Part 4**

Table 1. Summary of potential changes to biological patterns in steelhead trout as a result of conservation actions........................................................................... 82
List of Figures

Part 2
Figure 1. Schematic diagram of life-history patterns exhibited by both LGB and wild fish at the Keogh River................................................................. 38
Figure 2. Keogh River watershed area and location on Vancouver Island........... 40
Figure 3. Schematic diagram of the Keogh River enumeration fence infrastructure ............ 42
Figure 4. Marine survival estimates for wild and LGB smolt cohorts with total smolt output .... 44

Part 3
Figure 1. A) Landmark locations for geometric morphometric analyses from the left lateral view of wild and LGB fish. B) Morphological characters measured for non-geometric analyses ...... 70
Figure 2. A) Scatterplot of individual scores from a relative warps analysis of all wild and LGB specimens, with the uniform component omitted. (B) & (C) Graphical illustrations of the deformation in shape from the consensus configuration implied by positive trends along relative warp1 (RW1) and RW2 ............................................................................................................................ 72
Figure 3. A) Scatterplot of individual scores from the canonical variates analysis of all wild and LGB specimens, with the uniform component included. B) Deformation grids represent shape change from the consensus configuration implied by each combination of positive and negative trends on CV1 and CV2........................................................................................................................................ 74
List of Appendices

Appendix A: Details of Sampling and Euthanasia ................................................................. 83
Appendix B: Population Estimation Methods ................................................................. 87
Appendix C: Snorkel Survey Methods and Population Estimation ............................. 89
Appendix D: Snorkel Survey Efficiency Tests ................................................................. 91
Appendix E: Age Determination Methods ................................................................. 93
Part 1: Introduction

The future of wild salmonids (salmon and trout) is uncertain. Salmonid populations have declined dramatically in many areas of the Pacific coast of North America (e.g. Nehlson et al. 1991; Slaney et al. 1996; Smith and Ward 2000; Ward 2000) and disappeared from almost 40% of their historical range (NRC 1996). The loss of biodiversity is not the only concern – society attributes immense economic and cultural value to salmonids. Although highly valued, much of the salmonid decline can be attributed to human-mediated abuses: primarily through habitat destruction, overexploitation, and the use of fish hatcheries (reviewed in Lackey 2003).

Faced with decreasing fish abundance and intense societal pressure, biologists are challenged to develop effective recovery measures before populations reach extinction. In practice, there are three broad categories of conservation tools: fishery management (primarily harvest and gear restrictions); habitat enhancement; and, supplementation using hatcheries (Molony et al. 2003). Paradoxically, fish hatcheries, one of the major tools for conservation, have been blamed for causing major problems for wild populations (e.g. Hilborn 1992; Waples 1999). Historically, all three tools have been widely applied, although their effectiveness at recovering fish populations remains in question.

Steelhead populations have declined precipitously in many areas of British Columbia (BC) (Welch et al. 2000). Facing the loss of steelhead, the BC government responded in the late 1990s by initiating the watershed-specific "Vancouver Island Steelhead Recovery plan" (Wightman et al. 1998). The Keogh River has been monitored since 1976 (Ward and Slaney 1988) and continues to act as the province’s only steelhead research site (Lill 2002). Ambitiously, all three conservation tools were piloted in this single watershed and their effectiveness for wild population recovery is currently being evaluated. This thesis addresses many demographic, ecological, life-history, genetic, and phenotypic consequences of this multifaceted and experimental conservation program.

A widespread characteristic of salmonid populations is the existence of alternative life-histories (Gross 1984, 1996). Some individuals in many populations follow life-history trajectories that are unlike those commonly described for the species. For instance, some males may mature precociously and adopt a sneaker tactic by which they garner reproductive success through avoiding the fighting of "hooknose" males (Gross 1985). The proportions of individuals that become sneakers varies with both genetic and environmental factors (Gross 1991). An increase in growth rate, for example, can increase the proportion of precocious males (Gross 1991).
Part 2 discusses the influence of three conservation actions on the expression of alternative life-histories, and the implications of this expression on the recovery of the wild population. Captive breeding, fishery regulation, and habitat enhancement were all applied to the single depressed Keogh River steelhead population. For wild- and hatchery-origin fish I delineate life-history tactics and ontogenetic stages, and quantify their respective population sizes, sex ratios, age structures, biomasses, relative food consumption, potential reproductive output (as potential egg deposition), and overall potential to influence wild population recovery. I also discuss the potential for the hybridization of steelhead and coastal cutthroat trout in the captive breeding program and the implication of hybrid production for wild population recovery. Using this broad scope, I discuss the potential for these life-history and demographic shifts to have both positive and negative influences on the abundance, behaviour, ecology, and genetics of a depressed wild population.

Part 3 describes the effects of captive rearing on the phenotypes of individuals used for conservation, and the possible impacts this has on their survival. A sophisticated captive broodstock program was designed and implemented to manage genetic diversity while attempting to increase population size. However, management of phenotypic diversity may allow considerable change from the wild type. Using traditional and geometric morphometric techniques, I describe how captive-bred individuals differ from the wild type. I conclude by discussing the implications for any phenotypic divergence from the wild type for subsequent survival in nature.

Part 4 provides conclusions and recommendations from this research for conservationists and managers of conservation programs. The BC government's use of three conservation tools - captive breeding, fishery regulation and habitat enhancement - has implications for life-history and demographic patterns, reproductive and ecological interactions, and the expression of phenotypes. I synthesize the conservation issues and suggest methods to maximize the benefits and minimize the costs to wild population recovery.
Part 2: Conservation of a wild fish population (steelhead trout): influences of captive breeding, habitat improvement and fishing regulations on alternative life-histories
Introduction

Conservation programs have a variety of recovery tools to prevent extinction of fish populations (Blankenship and Leber 1995; Molony et al. 2003). Three common tools are improving the habitat, regulating the fishery, and supplementing the population using hatcheries.

Habitat improvement usually aims to increase habitat quality or area for at least one life stage (e.g. Ward and Slaney 1979, 1993a; Reeves et al. 1991). For diadromous species such as salmonids, the primary focus has been on improving freshwater habitat (e.g. Hunt 1976; Hartzler 1983; House and Boehne 1985; Annantrout 1991). Many freshwater habitats have been “improved” (see Roni et al. 2002), but the effectiveness of these actions in recovering fish has been evaluated rarely (Everest et al. 1991; Reeves et al. 1991; Frissell and Nawa 1992; Kauffman et al. 1997). Critics believe that most habitat improvements are “…at best a reversal of the degradation towards an anthropomorphic vision of what the river should look like” (Cowx and van Zyll de Jong 2004:247).

Fishery regulation is complicated by social, economic, and political resistance from fishers (e.g. Cook et al. 2001; Lester et al. 2003). Moreover, regulations may not protect all members of a population (e.g. restricting the sex, size, or time of harvest), and thus cause altered selective forces on life-history, behavioural and morphological traits (e.g. Gross 1991; Conover and Munch 2002). In addition, anglers tend to target the larger members of a population (Peterson and Evans 2003), further altering selective forces. Seasonal fishery closures may temporarily limit angler effort, but the effect is lessened substantially if angler effort remains uncontrolled during the balance of the year (Cox et al. 2002).

Many wild populations are supplemented by hatchery programs – worldwide, billions of salmonid juveniles are released annually (Brown and Day 2003). Recently, these programs have increasingly focused on conserving genetic diversity and natural adaptation (Utter 2004). Yet, anthropocentric concerns (such as ensuring the population is dominated by large, harvestable individuals) are frequently incorporated into conservation programs, either implicitly or explicitly. By incorporating exploitative biases the conservation program does not conserve genetic diversity and natural adaptation exclusively, but is also designed to produce a product more desirable to humans. For example, methods to reduce the number of non-migratory steelhead from hatchery releases have been developed, although this reduction may represent a loss in genetic variability from the stock (Viola and Schuck 1995). Also, precocious males successfully breed in the wild (Gross 1984, 1996; Hutchings and Myers 1988; Jordan and Youngson 1992; Thomaz et al. 1997; Seamons et al. 2004), but may be excluded from breeding.
in hatcheries (McLean et al. 2005). Although conservation programs may depend on the maintenance of the genetic characteristics of the wild population (Frankham et al. 2002), recovery efforts do not always treat all life-history types equally.

*Oncorhynchus mykiss* exhibits considerable diversity in life-history. Two major life-histories can co-exist sympatrically and interbreed: an anadromous form, “steelhead trout”, and a stream- or lake-resident form, “rainbow trout” (Shapovalov and Taft 1954). In addition, the anadromous form may also demonstrate summer- and winter-run reproductive ecotypes (Smith 1960; Withler 1966). In short, *O. mykiss* may migrate to the ocean at various ages, may return to freshwater after one or more years, may spawn across multiple years, may spawn before entering the ocean, and may remain in freshwater for their entire lives (Shapovalov and Taft 1954; Withler 1966).

Steelhead populations have declined precipitously in many areas of British Columbia (BC) (Welch et al. 2000). The decline is correlated with reduced adult returns, due to reduced freshwater and oceanic survival (Smith and Ward 2000; Ward 2000; Welch et al. 2000). The principal management agency, the BC Ministry of Water, Land, and Air Protection responded in the late 1990s by initiating the “Vancouver Island Steelhead Recovery plan” with the goal of “restoring healthy steelhead populations and habitats in selected watersheds on the east coast of Vancouver Island” (Wightman et al. 1998:55). They define a healthy steelhead population as one that “will always exceed minimum genetic spawner abundance levels while seeding at least 50% of the estimated fry rearing capacity in a given stream each year” (Wightman et al. 1998:55). As the Keogh River has been monitored since 1976 (Ward and Slaney 1988) and continues to be the province’s only steelhead research site (Lill 2002), it was selected to pilot all three recovery tools. These include: 1) a fishery closure from 1 December to 31 May commencing 1997 (Lill 2002); 2) habitat improvement, including in-stream habitat structure placement and inorganic nutrient addition since 1997 (Ward et al. 2003); and 3) a Living Gene Bank (LGB) hatchery program since 1998 (Wightman et al. 1998).

The LGB program aims to increase population size while retaining natural genetic diversity and adaptation. This sophisticated captive broodstock program is detailed by Amman et al. (2005a, 2005b). The program collected smolt rather than adults as broodstock because this allowed the production of large numbers of progeny without appreciably decreasing the number of returning wild adult spawners. Each year approximately 100 wild outmigrating smolts, identified as pure steelhead by visual inspection by contract biologists, were collected for broodstock as greater changes in allele frequency may occur at lower spawner numbers (Waples
Broodstock were raised to maturity in fish culture, spawned as adults according to a genetic protocol (genotyping, and later one-on-one random mate assignment), and their progeny reared for one year. As a result of the LGB program, five cohorts of hatchery-produced juveniles have been released during the annual wild smolt migration. After release, LGB-produced fish complete their life-cycle in nature. The program was limited to five years, or one life-cycle, to ensure that progeny of LGB-produced fish would not be collected as broodstock in later years (Ammann et al. 2005a), a problem in past hatchery programs (Waples and Do 1994).

Although assumed to be pure steelhead, without molecular analysis, it was possible for the program to include natural hybrids as LGB broodstock. *Oncorhynchus mykiss* and *O. clarki clarki* (coastal cutthroat trout) are naturally sympatric salmonid species that are known to hybridize in nature (Campton and Utter 1985; Johnson et al. 1999; Young et al. 2001; Baker et al. 2002; Docker et al. 2003; Ostberg et al. 2004). Of 30 sympatric cutthroat and steelhead populations that were recently sampled on Vancouver Island, 29 demonstrated evidence of hybridization (Bettles 2004). Furthermore, the probability of correctly classifying a juvenile steelhead can be low in sympatric cutthroat and steelhead populations due to similar appearance and frequent hybridization (Baumsteiger et al. 2005). This thesis does not address the genetic identification of LGB and wild fish; future molecular work will address this issue.

Multiple life-history tactics are being expressed by the LGB progeny at release (Werlen 2003). Our initial observations and those by McCubbing (2002), McCubbing and Ward (2003) and Werlen (2003), suggest these tactics include (Table 1): 1) migrants – individuals that have smoltified and immediately migrate to the ocean at release; 2) delayed-migrants – individuals that remain within the river, to outmigrate as unspawned smolt in subsequent years; 3) resident breeders – individuals that remain in the river to spawn as precocious parr; and 4) delayed-resident-breeders – individuals that remain as immature parr within the river after release, to become precocious parr in subsequent years. In addition, life-history patterns predict that resident breeders should retain the potential to smoltify and migrate to the ocean after one or more years of spawning. Despite three decades of monitoring by the BC government, no precocious parr and only a few rainbow trout have been observed in this wild population (B. Ward, personal communication 20 July 2005). The life history patterns of wild fish are relatively simple in the Keogh River, whereas LGB progeny apparently exhibit alternative life history tactics (Figure 1). Hence, the LGB conservation program has either created new life-history tactics, or amplified extremely rare tactics that were not previously seen in the wild population.
All three conservation tools – fishery regulations, habitat improvement and the LGB program – are influencing the life-histories of *O. mykiss* in the Keogh River. To evaluate BC’s program, I estimate the relative numbers of LGB and wild fish, their age and sex, their potential contribution to reproduction and biomass, and their potential resource competition. I also discuss the implications of three potential hybridization scenarios for wild-population recovery. Through these analyses I demonstrate the potential for alternative life-histories to make both positive and negative contributions to the wild population through reproductive, ecological and behavioural pathways. I conclude with suggestions on how conservation programs may be modified to better reach their goals of increasing population increase and maintaining genetic diversity.

**Methods**

*Study site*

The Keogh River, at the northeastern end of Vancouver Island, is a third-order coastal stream, 31km in length, with a drainage of 130km² and has been detailed elsewhere (Figure 2, Ward and Slaney 1979, 1993b, 1988; Johnston et al. 1986; Irvine and Ward 1989). An enumeration fence (Figure 3), 300m upstream of the estuary, traps migrating salmonids in both upstream and downstream directions – details of fence operations are in Ward and McCubbing (1998) and Appendix A.

*Sampling*

Fish were sampled in the river and from the LGB program in 2003 and 2004. As the wild smolt run was effectively complete by 15 June (McCubbing and Ward 2003, 2004), any LGB progeny encountered in-stream after this date were assumed to be resident (e.g. “LGB parr”; Table 1).

In 2004, collection dates, data types and sample sizes vary by fish origin and life stage (Table 2) – full collection details are provided in Appendix A. Since the 2004 LGB parr were sampled at three times, I refer to the samplings as early, middle, and late (Tables 1 and 2). Any fish that was sampled internally (e.g. sex, gonad factor, and/or gut contents) was euthanized prior to sampling using tricaine methane sulfonate (MS222) or a 1:10 solution of eugenol and 95% ethanol – details of anesthesia are in Appendix A.

From 17 June to 30 July 2003, the 2003 LGB parr were angled, marked (with an individually numbered Floy FD-68B tag; Floy Tag Manufacturing Company, Seattle).
Washington 98105) and released for use in a mark-recapture population estimate. From 7 to 26 July 2003, the 2001-2002 LGB parr were angled, marked and released for population estimation.

**Population Estimation**

In 2004, the anadromous wild and LGB populations were estimated by McCubbing and Ward (2004). LGB and wild anadromous adult numbers were estimated separately. Due to differential mortality of anadromous adult males and females during freshwater residency (Ward and Slaney 1988), male and female numbers were also estimated separately. Population estimations were done via the adjusted Petersen estimate (Ricker 1975). The operculum of adult steelhead were bus punched upon capture, and these operculum-marked kelts acted as recaptures (Ward and Slaney 1988, 1990) – further population estimation details are provided in Appendix B. As anadromous wild males were marked, but not recaptured, they were estimated by estimating the total number of anadromous wild fish and subtracting the number of anadromous wild females.

Wild smolt were enumerated by adding the daily totals of outmigrating individuals encountered at the enumeration fence (details provided in McCubbing and Ward (2004)). To estimate the number of LGB smolt released, the mass of each LGB release was divided by the average individual LGB fish mass (n~100 from each release). Due to sampling methodology, wild and LGB smolt population estimates are without probability regions.

LGB parr that were sampled and released live were marked using Floy tags (as above) to allow the identification of recaptures. As Petersen mark-recapture methods have problems when data are sparse (Cormack 1968; Ricker 1975), I used the Bayesian sequential mark-recapture algorithm (Gazey and Staley 1986) to estimate LGB parr populations in 2003 and 2004 (Appendix B). To verify the similarity between the hypergeometric and binomial Bayesian algorithms, I use both models to independently analyze all data from periods in which sampling was done without replacement.

Because LGB smolt were sampled at release, the sample contains fish that will: 1) immediately migrate to the ocean (LGB migrants); or 2) choose to adopt a resident tactic, as they never underwent smoltification (LGB parr). To determine the sex ratio and number of LGB migrants I: 1) estimated the number of males and females in the LGB smolt and early 2004 LGB parr populations using their respective population estimates and sex ratios (see below for sex ratio calculations); and 2) subtracted the number of male and female early 2004 LGB parr from the male and female LGB smolt.
Survival rates have been calculated for the full 2001 and partial 2002 wild and LGB smolt cohorts (see McCubbing and Ward 2004). To determine if LGB and wild estimated marine survivals differ within each smolt cohort, I compared the numbers of smolt that lived to adulthood and the number that died at sea for LGB and wild fish using a \( \chi^2 \) test.

During spring 2003, LGB parr from the 2001-2002 releases were observed approximately 5.5km upstream of the release site (D. McCubbing personal communication 30 June 2003). During summer 2003 we (J.C. Walters and C. Werlen) used a stratified mark-recapture sampling technique to estimate the population of 2001-2002 LGB parr (Appendices C and D). Briefly, the estimation entailed: prior to 11 July 2003 we angled, marked (using Floy tags, as above) and released LGB parr in four 500m reaches. From 11 July to 17 August 2003, we surveyed each reach four times via snorkeling (as in Thurow 1994). Using the Bayesian mark-recapture algorithm (Gazey and Staley 1986), I estimated the population of LGB parr within each reach. Reach-specific population densities were then extrapolated throughout the remainder of the river.

Age Determination

The individual ages of wild and LGB fish were determined using scale-pattern analysis and by analyzing coded-wire tags. Scales were collected as in Maher and Larkin (1954).

Anadromous adult ages were determined by scale-pattern analysis exclusively. Acetate impressions of scales from anadromous adult fish (LGB and wild) were projected onto a microfiche reader (Narver and Anderson 1974; Bernard and Myers 1996) and then aged by multiple independent readers (wild scales: P.M. Troffe, B.R. Ward, 100% concordance between readers; LGB scales: P.M. Troffe and J.C. Walters, 96.5% concordance between readers). Total ages (freshwater age + ocean age) of anadromous wild and LGB adults were compared by Fisher’s exact test.

I profiled the age structure of wild smolt, wild parr, and the 2001-2003 LGB parr sampled in 2004. LGB smolt and 2004 LGB parr were not included in the scale-pattern analysis as they were of known age (1 year).

Scales from parr and smolt were cleaned using an ultrasonic cleaner and a solution of pancreatin (Whaley 1991), then rinsed and mounted on glass slides. Scales were then digitized using a dissecting microscope and a digital camera. Two independent readers then determined ages (P.M. Troffe and J.C. Walters: 95% concordance between readers). Scale preparation and analysis are detailed in Appendix E.
In 2002, hatchery staff injected coded wire tags into the snout of each LGB fish. Upon
detection via a metal detector, tags were surgically extracted and subsequently read using a
dissecting microscope. Coded wire tag-based age determinations took precedence over a scale-
based age estimate when a discrepancy occurred (11% discrepancy rate).

**Sex Ratio and Gonadal Development**

Gonad factors were determined in LGB parr, LGB smolt and wild smolt (Table 2). The
gonad factor scale rates an individual's sexual maturity from 0 (no development) to 4 (spawning
condition) (Rempel et al. 1984). Category 5 was added to the gonad factor scale, representing
recently spawned fish. Gonad factors were then compared between populations using Fisher's
exact test.

In addition, population sex ratios were compared using a $\chi^2$ test. I also used an analysis
of variance (ANOVA) to test for differences in length and mass among immature and
precociously maturing LGB smolt and parr.

**Potential Egg Deposition**

Anadromous potential egg deposition (PED) for 2004 was calculated by: 1) using the
fecundity and length relationship for Keogh River steelhead to predict average anadromous LGB
and wild fecundity, and 2) multiplying this average by the estimated number of anadromous
LGB and wild females. In addition, LGB parr PED was calculated by multiplying their average
fecundity by the estimated number of mature female LGB parr.

I also estimated average individual fecundity and PED for 2002 and 2003 using estimates
of anadromous adult populations (McCubbing 2002; McCubbing and Ward 2003) and
corresponding unpublished length data (B. Ward unpublished data). These PEDs were then used
to estimate wild parr biomass (see next section).

**Biomass estimation**

The biomass of wild and LGB anadromous adults was estimated by: 1) conducting a
linear regression using length and mass data from 494 wild steelhead (1981-1995, B. Ward
unpublished data), 2) using this relationship to predict the average individual mass of
anadromous wild and LGB adults sampled in 2004, and 3) multiplying the estimated average
mass of anadromous wild and LGB adults by their respective population estimates.
For the years 2002-2004, fry biomass from wild and LGB anadromous adult spawning was estimated by: 1) multiplying PED by average egg-to-fry survival rate for the Keogh River (6.53% from 1976-1985, Ward and Slaney 1993b), and 2) multiplying the estimated fry abundance by 1.98g - the average fry mass during watershed restoration (McCubbing and Ward 2002).

To estimate the number of 2002 and 2003 cohort fry that survived to become 1+ parr, I assumed overwinter survival to the 1+ stage to be 25%, as: 1) this survival rate has been observed (Ward and Slaney 1993b); and 2) virtual population analyses demonstrated that, for many cohorts, less than 25% survival would necessitate unrealistic survival rates (e.g. ≥100%) through successive years of freshwater rearing (Burrows 1997). To estimate the number of 2003 cohort 1+ parr that remained in-stream during summer 2004, I then subtracted the estimated number of 1-year-old smolt that outmigrated in 2004 (McCubbing and Ward 2004) from the estimate of surviving 1+ parr. To estimate the number of 2002 cohort 1+ parr that remained in-stream during summer 2003, I then subtracted the estimated number of 1-year-old smolt that outmigrated in 2003 (McCubbing and Ward 2003) from the estimate of surviving 1+ parr.

To estimate the number of 2002 cohort 1+ parr that survived to become 2+ parr, I assumed 50% overwinter survival, as: 1) a plausible range for annual survival beyond the 1+ parr stage is 50-95% (Burrows 1997), and 2) due to reduced smolt-per-spawner output, overwinter parr survival is currently close to 50% (B. Ward, personal communication 20 July 2005). I then subtracted the estimated number of 2-year-old smolt that outmigrated in 2004 (McCubbing and Ward 2004) from the estimate of surviving 2+ parr.

After 2001, 4+ smolt have not been observed migrating to the ocean (McCubbing and Ward 2004). Therefore, if 3+ parr did exist during summer 2004, their numbers were likely low and did not comprise a significant portion of the in-stream biomass - thus, 2+ parr are the oldest juveniles included in these biomass calculations.

To estimate the biomass of each parr age (1+ and 2+) I multiplied each cohort population estimate by the cohort-specific average individual mass.

Hatchery staff determined the 2004 LGB smolt biomass by summing the biomass from the five releases. The biomass of LGB parr was estimated by multiplying the average mass of an early 2004 LGB parr by their population estimate. The biomass of the 2001-2003 LGB parr was estimated by multiplying their average mass by their population estimate.
Diet and Impacts

In 2004, the diet of LGB parr was determined by examining their stomach contents and identifying the invertebrates (to ordinal level) and other non-food items present. These results were compared to published records from wild Keogh River steelhead (Johnston et al. 1990).

Relative feeding success was determined by classifying stomach contents into three categories: 1) greater than 50% of stomach content volume was non-food items (e.g. conifer needles, stones); 2) greater than 50% of stomach content volume was invertebrate prey; 3) empty. Differences in feeding success among LGB parr groups were then evaluated (via Fisher’s exact tests). As fry tend to emerge in mid-June at the Keogh River (B. Ward personal communication 8 August 2005) I examined the stomachs of any LGB parr sampled after this date for evidence of steelhead fry predation.

As food consumption in salmonids scales to body mass by the exponent of 0.73 (Steingrimsson and Grant 1999), I use $W^\alpha$ (where: $W =$ body mass; and $\alpha=0.73$) from the standard allometric equation ($y = bW^\alpha$, (Huxley and Teissier 1936) as an estimate of relative food consumption among wild and LGB fish types. By multiplying $W^\alpha$ by the estimated population sizes of each wild and LGB fish type, I estimate the relative food consumption by each fish type.

Results

Population estimation

The anadromous LGB population in 2004 was 146 females (95% CI: 60-362) and 130 (95% CI: 62-299) males (Table 3). The anadromous wild population was 53 females (95% CI: 33-192) and 33 males (no CI due to calculation method; Table 3). Thus, the total anadromous population was estimated to be 361 fish, of which 76% were LGB and 24% were wild. (McCubbing and Ward 2004)

Enumeration at the fence in 2004 counted 551 wild smolt (McCubbing and Ward 2004). By totaling LGB release estimates, the 2004 LGB smolt release was 27506 fish (Table 3).

In 2004, the 2001-2003 LGB parr population was estimated using the binomial (585 fish, 95% HPD: 283-983) and hypergeometric (586 fish, 95% HPD: 263-963) – both algorithms demonstrated similar results. The 2004 LGB parr population was estimated using the binomial model exclusively (1634 fish, 95% HPD: 1441-1791; Table 3).

By subtracting the estimated 1634 LGB parr (with a male:female ratio of 1.0:0.45; Table 3 – see below) from the 27506 LGB smolt (with a male:female ratio of 1.0:1.13; Table 3 – see...
below), the number of LGB smolt immediately migrating to the ocean was 25872, with a male:female ratio of 1.0:1.34.

For the 2002 smolt cohort, the provisional marine survivals (i.e. of 2-ocean fish only) of wild and LGB smolts were significantly different (0.43% and 0.24%, respectively; $\chi^2_1 = 15.08$, $p=0.0001$). For the 2001 smolt cohort, the marine survivals of LGB and wild smolts were not significantly different (3.23% and 1.95%, respectively; $\chi^2_1 = 2.40$, $p=0.1213$).

In 2003, the 2003 LGB parr population was estimated to be 513 individuals (95% HPD: 353-713). Also in 2003, there were an estimated 534 2001-2002 LGB parr resident in the river (Table 3, Appendix C), with greater fish densities observed in the lower river than in areas further upstream (Table 4). Given the 2001-2003 LGB resident populations totaled 1047 (513 ÷ 534) in 2003 and 585 in 2004, they experience an annual survival rate of 56%.

**Age Determination**

Most 2004 wild anadromous adults had migrated to the ocean as smolt at age two (Table 5). Repeat spawners were the most common wild anadromous adult tactic, although maiden spawning at ocean age 2 and 3 were also demonstrated by both sexes (Table 5). Only wild males returned after a single year in the ocean (22.2%; Table 5).

All LGB anadromous adults migrated to the ocean after one year (Table 5). Ocean ages differed among the sexes: LGB females primarily spent 3 years at sea, whereas most LGB males spent either 2 or 3 years at sea (Table 5).

Within each sex, anadromous wild and LGB adults were similar in total age (males: Fisher's exact test, $p=0.0788$; females: Fisher's exact test, $p=0.0927$), thus the LGB program does not accelerate population recovery by reducing average total age.

The freshwater age distributions of the wild and LGB smolt populations were different: LGB smolt are all age 1, whereas wild smolt age averaged 2.2 years (age 1= 5.1%, age 2= 69.2%, age 3= 25.6%, n=39: Table 6).

In 2004, the 2001-2003 LGB parr sample was determined to contain individuals from the 2002 and 2003 LGB cohorts only (2002 LGB cohort n=17; 2003 LGB cohort n=49). Thus, by 2004 it appears that resident 2001 LGB parr did not exist in the river.

**Sex Ratio and Gonadal Development**

The sex ratios of LGB and wild fish did not differ significantly for either anadromous adults ($\chi^2_1 = 2.02$, $p=0.1554$, N=362) or smolts ($\chi^2_1=0.12$, $p=0.7286$, N=328; Table 3). In
contrast, the sex ratios of LGB smolt and the early 2004 LGB parr were significantly different ($\chi^2 = 31.19, p<0.001, N=340$); LGB smolt are female biased, whereas the 2004 LGB parr are strongly male biased (Table 3).

Gonad factors (GF) differed between wild and LGB smolt (Fisher’s exact test, $p=0.0017$). No wild smolt demonstrated signs of precocial development, whereas 19.6% of LGB smolt exhibit advanced maturation (GF 2 and 3 combined; Table 7). The early 2004 LGB parr demonstrated significantly higher GF scores than the LGB smolt (Fisher’s exact test, $p<0.0001$; Table 7). The late 2004 LGB parr demonstrated significantly higher GF scores than the early LGB parr (Fisher’s exact test, $p=0.0258$; Table 7).

A gonad factor score of 3 is nearing spawning condition, but does not exhibit free-flowing gametes (Rempel et al. 1984). Upon release, 0.7% of LGB smolts exhibited a GF=3 (Table 7). This suggests that 193 of the 27506 2004 LGB smolts were approaching spawning condition upon release. In comparison, 14.5% of the early 2004 LGB parr exhibited a GF=3 (Table 7). This suggests that 237 of the 1634 2004 LGB parr were approaching spawning condition upon residency. For LGB smolts and 2004 LGB parr, only males demonstrated a GF of 3 or greater (Table 7).

In 2004, the resident 2002 and 2003 LGB parr exhibited a strong male bias: 2002 LGB parr, 71% male ($n=17$); 2003 LGB parr, 69% male ($n=49$). Most 2002-2003 LGB parr had undeveloped or partially developed gonads (GF 1 and 2, respectively; Table 7). The two female 2002-2003 LGB parr with ripe gonads (GF=4) contained 312 and 319 eggs. Mature females (GF 4 and 5) represent 4.2% of the 585 2002-2003 LGB parr – suggesting 25 potential spawning females. Mature males represent 13.9% of the 585 2002-2003 LGB parr – suggesting 81 potential spawning males.

On average, precociously maturing (i.e. GF 2 and 3) LGB smolt and 2004 LGB parr were larger than those that were not maturing (Table 8). Among gonad factors expressed by LGB smolt, there are significant differences in mass ($F_{2,282}=19.81, p=0.0001$) and length ($F_{2,282}=3.53, p=0.0304$). Among early 2004 LGB parr, precocious and immature individuals are similar in mass ($F_{2,52}=1.55, p=0.2220$) and length ($F_{2,52}=1.66, p=0.2010$). In contrast, precocious middle 2004 LGB parr are larger in mass ($F_{2,277}=27.68, p<0.001$) and length ($F_{2,277}=25.38, p<0.001$) than immature individuals. Precocious late 2004 LGB parr were greater in mass ($F_{2,63}=3.78, p=0.0282$), but not length ($F_{2,63}=2.32, p=0.1068$), relative to immature individuals.
Potential Egg Deposition (PED)

The average estimated fecundity was greater for anadromous wild females due to their larger size relative to anadromous LGB fish, but due to their large numbers the LGB female PED was approximately 2.5 times that of wild females in 2004 (Table 9). Mature female LGB parr PED is low relative to anadromous wild and LGB females (Table 9).

In 2003, average estimated fecundity was much greater for wild females than LGB females (Table 9). By 2003, LGB females spent two years growing in the ocean, whereas wild anadromous females were both 2- and 3-ocean age fish. As larger females have higher fecundity (Ward and Slaney 1993b), due to their larger average size, a wild female would likely contain more eggs. In 2003, wild female PED was much greater than that of LGB females (Table 9).

As no anadromous LGB females returned in 2002 (McCubbing 2002), PED was from wild anadromous females only (Table 9).

Biomass estimation

Using archived data (B. Ward unpublished data), the relationship between adult length and mass is described by:

\[
\text{mass}_{kg} = -8.597 + 0.01785 \times \text{Length}_{mm} ; r^2 = 0.77, p<0.001.
\]

Based on this relationship, anadromous wild adults were, on average, heavier than anadromous LGB adults (Table 3). Although lighter on average, due to their relatively large numbers the anadromous LGB adults represented roughly 2.7 times more biomass than wild anadromous adults (Table 3).

Midsummer fry abundance in 2004 was estimated to be 57200 individuals; and based on their relative population sizes and average estimated fecundity, anadromous wild and LGB females contributed 16240 and 40960 fry, respectively (Table 3). There was an estimated 113.25kg of fry in the river during 2004 (Table 3), with anadromous wild and LGB parents contributing 32.15kg (28%) and 81.10kg (72%), respectively.

Midsummer fry abundance in 2003 was estimated to be 14788 individuals. Assuming 25% survival and subtracting the estimated 24 1+ smolt that migrated in 2004 (McCubbing and Ward 2004), there were an estimated 3673 1+ parr in the river during summer 2004. With an average mass of 24.60g (SD=5.70, n=10), total 2003 cohort 1+ parr biomass was estimated to be 90.36kg (Table 3).
Midsummer fry abundance in 2002 was estimated to be 17478 individuals. Assuming 25% survival and subtracting the 963 1+ smolt that migrated in 2003 (McCubbing and Ward 2004), there were an estimated 2036 1+ parr in the Keogh River during summer 2003. Assuming 50% mortality to the 2+ stage and subtracting the 321 2+ smolt that migrated in 2004 (McCubbing and Ward 2004), there were an estimated 1382 2+ parr in the river during summer 2004. With an average mass of 36.81g (SD=8.80, n=19), total 2002 cohort 2+ parr biomass was estimated to be 50.87 kg (Table 3).

The estimated biomass of the early 2004 LGB parr (94.48 kg) and 2001-2003 LGB parr (75.45 kg) were much greater than that of the wild smolts (27.43 kg, Table 3). LGB smolt releases were the greatest source of biomass in 2004 (2170.70 kg), with LGB migrants contributing the vast majority at the time of release (2050.99 kg; Table 3).

**Diet and Impacts**

Most 2001-2003 LGB parr stomachs contained food only (Table 10). Many early 2004 LGB parr contained food, although most stomachs contained non-food items (Table 10). A large proportion of the middle 2004 LGB parr had empty stomachs or contained non-food; few contained food only (Table 10). The late 2004 LGB parr had much greater success in obtaining food; most contained non-food and food only (Table 10).

Feeding success was different among the LGB parr groups, with more experienced parr demonstrating greater feeding success (e.g. 2001-2003 LGB parr > late 2004 LGB parr > early 2004 LGB parr; Fisher's exact test, p<0.0001). Feeding success also increased with age among 2004 LGB parr (late 2004 LGB parr vs. early 2004 LGB parr; Fisher's exact test, p=0.0137).

The middle 2004 LGB parr were excluded from statistical analyses as at the time of sampling they had been retained in a small area below the fish counting fence for a minimum of 6 weeks. Hence, this high-density population likely demonstrated high rates of empty and non-food filled stomachs because of entrapment and intense competition for limited food. This issue may also influence the amount of non-food present in the stomachs of the early 2004 LGB parr, but as <5% of them had empty stomachs, food availability appeared to be adequate. In contrast, as the early 2004 LGB parr contained mostly non-food items, they may have had difficulty discriminating between prey and non-food items.

Although LGB parr stomachs primarily contained insects from the orders Trichoptera, Plecoptera, Diptera, and Ephemoptera, other invertebrate groups were also observed in a number of cases. These included insects (Coleoptera, Orthoptera, Lepidoptera and Odonata), arachnids.
crayfish, clams, and snails. The late 2004 LGB parr contained the widest variety of invertebrate orders among the LGB parr (Table 10).

After fry emergence in 2004, 16.7% and 1.20% of the 2001-2003 and 2004 LGB parr, were observed to prey on steelhead fry, respectively. Thus, an estimated 98 2001-2003 LGB parr (16.7% of the estimated 585 individuals) and 20 late 2004 LGB parr (1.20% of the estimated 1634 individuals) cannibalized steelhead fry. This indicates that fry predation increases with age – likely a result of increased size.

Due to their larger average size, an LGB parr will consume 2 and 21 times more food than wild parr and fry, respectively (Table 11). On a population-basis, LGB parr will potentially increase in-stream food consumption by steelhead by 41% in 2004 (Table 11). Assuming LGB migrants are similar in mass to LGB smolt, an LGB migrant will consume 33% more food than a wild smolt, but due to their greater population size, LGB migrants cause a 67-fold increase in migrating steelhead food consumption (Table 11).

Discussion

The multifaceted conservation program of the BC Ministry of Water, Land, and Air Protection had many effects on the depressed steelhead population. Life-history and demographic shifts occurred within and between the wild and hatchery populations. Below I discuss how the population has changed as a result of the conservation actions and the implications for wild-population recovery.

The marine age distributions of wild and LGB anadromous adults are very different. Some wild steelhead returned to spawn after only one year in the ocean, whereas LGB steelhead spent a minimum of two years at sea. This suggests LGB fish cannot obtain similar rapid rates of growth and development in the ocean. Conversely, the most rapid growing LGB fish may have adopted a resident tactic and matured precociously. Furthermore, the repeat spawner class (1st spawning at ocean age-2, 2nd spawning at ocean age-3) was the most prevalent for both sexes of wild anadromous adults, while uncommon in anadromous LGB adults. The predominance of the repeat spawner tactic in 2004 may be anomalous as it comprised <10% of wild anadromous returns during 2002 and 2003 (McCubbing 2002; McCubbing and Ward 2003).

On a per-spawner basis anadromous wild steelhead likely have a greater impact on population recovery than anadromous LGB fish. The reason for this is four-fold: 1) within a given year, a greater proportion of anadromous wild fish may spawn for a second year, relative to anadromous LGB fish; 2) the average estimated fecundity is greater for wild anadromous
females relative to anadromous LGB females due to the former’s larger size at age; 3) radiotracking results demonstrate that anadromous LGB females apparently spawn in high densities in the lower reaches of the river, whereas wild females tend to spawn further upstream at lower densities, (although spawning areas overlap, incomplete spawning separation); and 4) hatchery steelhead tend to achieve lower reproductive success relative to wild adults. As the lower river reaches have historically supported low juvenile production (B. Ward personal communication 20 July 2005), poor spawning habitat and high spawning density will likely further reduce the average reproductive success of anadromous LGB fish. Hatchery steelhead tend to achieve low reproductive success in comparison with wild anadromous adults: in other rivers hatchery steelhead produced 4.4-7.0% (McLean et al. 2004), 18-37% (Kostow et al. 2003), and 28% (Chilcote et al. 1986) the number of smolts per wild female. On a per-spawner basis, hatchery steelhead females have been shown to produce 2.3-11.1% (McLean et al. 2003) and 4-13% (Kostow et al. 2003) of the adult offspring produced by wild females. Although LGB smolt have survived to adulthood at over half the rate of wild smolt (McCubbing and Ward 2004), due to differences in spawning location, behaviour and fecundity, the average individual reproductive success is likely much greater for wild anadromous females relative to their LGB counterparts.

The reduced reproductive success of hatchery fish may result not only from selection of poor spawning habitat and lower fecundity relative to wild fish – but considerable evidence suggests competitive inferiority relative to wild fish is also responsible. Due to diminished competitive ability, hatchery males tend to be outcompeted by wild males during spawning and hatchery females tend to dig fewer nests than wild females (Berejikian et al. 1997; Fleming et al. 2000). Hatchery males also tend to be less reproductively active relative to wild males (Fleming and Gross 1992). Although they demonstrate lower individual reproductive success relative to wild fish, anadromous LGB steelhead may assist population recovery in the Keogh River. Even with lower individual fecundity, the large numbers of anadromous LGB females potentially deposit about 2.5 times more eggs than wild anadromous females in 2004. Large numbers of anadromous LGB fish may also have dramatic effects on increasing the effective population size in the Keogh River. For example, if 15% of anadromous LGB fish are as successful at spawning as anadromous wild fish, the number of successfully fertilized eggs will increase by almost 50%.

By altering patterns of life-history and demography, conservation measures could influence anadromous adult inbreeding rates. Watershed restoration efforts on the Keogh River have been attributed with increasing smolt recruitment, producing record smolt-per-spawner output, and reducing average smolt age from three to two years (Ward et al. 2003). Although
average wild smolt age was 2-years in 2004, LGB smolt development was even shorter (1 year), potentially increasing anadromous steelhead returns sooner than watershed restoration could alone. If anadromous LGB fish spawn at a younger total age than wild fish, then the relatedness between wild and LGB fish is reduced. Unfortunately, this is not the case; statistically, there is no difference in overall age distribution between wild and LGB anadromous fish. Hence, LGB-mediated changes to marine juvenile life-history patterns likely do not decrease inbreeding rates during subsequent adult spawning.

By dramatically increasing total smolt output, the LGB program may contribute to further declines in wild smolt marine survival. LGB migrants may compete with LGB smolt for marine resources; in 2004, LGB migrants represented almost 51 times the number, almost 75 times the biomass, and over 67 times the relative food consumption of wild smolt. Within the 2002 smolt cohort, wild smolt experienced significantly greater marine survival than LGB smolt (0.43% and 0.24%, respectively; McCubbing and Ward 2004). In contrast, the 2001 smolt cohort fared better: marine survival of wild smolt was greater than for LGB smolt, but this difference was not significant (3.23% and 1.95%, respectively; McCubbing and Ward 2004). In addition, LGB smolt releases were typically 2-3 times the size of the largest wild Keogh River smolt runs on record (see Ward and Slaney 1993b). Although data exist for only two LGB releases, marine survival decreased for both wild and LGB smolts as total smolt output increased (Figure 4).

Density-dependent growth and mortality has been demonstrated among salmonids in the marine environment (e.g. Peterman 1978, 1984; McCarl and Rettig 1983; Peterman and Routledge 1983; Ruggerone et al. 2003), and negative density-dependent effects may be increased by hatchery supplementation (Levin et al. 2001). Also, being winter-run steelhead, the Keogh population is not vulnerable to incidental by marine commercial salmon fisheries (Smith and Ward 2000). Thus, negative density-dependent effects, related to the large annual releases of LGB smolt, may be partially responsible for recent declines in marine survival.

Although it appears as if conservation actions increased the prevalence of alternative life-histories, precocious wild parr likely exist. After nearly 30 years of monitoring, precocious parr have yet to be observed in the Keogh River, but they have not been directly targeted for discovery (B. Ward personal communication 20 July 2005). Given: 1) the female bias in the LGB migrants; 2) the male bias in the 2004 LGB parr; and 3) the similarity in female bias between LGB migrants and wild smolt; wild precocious parr likely exist at low numbers. Recall that 0.7% of the LGB smolt demonstrate precocity (e.g. 237 of 27 506). Assuming a similar rate of precocity exists in the wild population, there would have been about 4 wild precocious parr in
the river during 2004 (e.g. 0.7% of 511 wild smolt). As juvenile numbers were previously much greater (see Ward and Slaney 1993b), the number and influence of precocious wild parr may have been more significant in the past.

Watershed-restoration efforts and the LGB hatchery program have acted in concert to favour a life-history tactic not prevalent in the wild. In-stream fertilization increases the production of benthic invertebrates (Johnston et al. 1990). Prior to watershed restoration, many hatchery fish became resident after release, but very few were enumerated in subsequent years (Ward and Slaney 1990), implying extremely high annual mortality rates and/or extensive up- or downstream migration outside the annual smolt migration. In contrast, LGB parr experienced only moderate annual mortality rates; the estimated 1047 LGB parr present in summer 2003 declined to 585 individuals by summer 2004 (44% annual mortality). Also, only 4.1% of the 2001-2003 LGB parr migrated to the ocean as smolts during 2004. Thus, recent watershed restoration efforts likely increased prey abundances, potentially increasing LGB parr survival.

During river residency, LGB parr mortality is due to many factors acting simultaneously. As natural predators are excluded from the rearing facilities, the hatchery does not mimic the wild environment with regards to encounters with predators. Predation can be a major source of mortality to juvenile salmonids (Ruggerone and Rogers 1984; Fresh and Schroder 1987). As prior experience increases the ability of juvenile salmonids to avoid predators (Ginetz and Larkin 1976; Patten 1977; Olla and Davis 1989), the naive LGB parr may initially succumb to high rates of predation. In addition, angler retention also contributes to LGB parr mortality. Fishing regulations stipulate that from 31 May to 30 November any *O. mykiss* over 30cm in length are eligible for harvest (BC WLP 2005). Prior to the LGB program, the rarity of *O. mykiss* of this size kept angler success low. When interviewed, local anglers boasted of landing upwards of 20 LGB parr per day during the summers of 2002 and 2003. Also, between the summers of 2003 and 2004 the abundance of LGB parr over 30cm in length declined greatly (J.C. Walters and M.R Gross unpublished data) – further supporting my presumption of intense angling pressure.

The estimated numbers of precocially maturing LGB smolt and LGB parr are similar in 2004. This suggests that precociously maturing LGB smolt chose to forego oceanic migration and become LGB parr, likely adopting one of the resident breeder tactics. Alternatively, the precocious LGB smolt may have exited the river and a similar number of LGB parr matured between samplings. This alternative explanation is unlikely as the stress associated with handling, transport, and release to a novel environment may take anywhere from minutes to
weeks to subside (Schreck et al. 1997). Thus, prior to release all precociously maturing LGB progeny have apparently decided to become precocious parr and remain resident in the river.

Precociously maturing LGB fish also achieve the greatest growth in relative condition. At the time of river residency, immature and precocious LGB parr were similar in size. By the late LGB parr sampling, precocious fish were greater in mass and similar in length relative to immature individuals. Sexually maturing resident hatchery steelhead typically have high condition factors (Viola and Schuck 1995). This suggests that relative to immature fish, precocious individuals are more successful at feeding and can allocate more resources to increasing their condition and advancing their reproductive development.

During river residency, LGB parr will exhibit differential reproductive success between the sexes. In 2004, the 2002-2003 LGB parr contributed an estimated 0.89% of the total potential egg deposition. Fish experiencing only the lower reaches of a river as juveniles (e.g. LGB migrants) hesitate to move upstream into the spawning grounds (Jonsson et al. 1994). Furthermore, individuals without previous river experience wander more within the river and are more likely to exit unspawned (Jonsson et al. 1990). Although their PED is low, the individual reproductive success of female LGB parr may be greater than expected due to extensive experience in the river relative to anadromous LGB females.

Mature male LGB parr may achieve high reproductive success and have made considerable contributions to in-stream spawning. Although small in size relative to anadromous adults, mature 2002-2003 LGB parr outnumbered wild anadromous males by almost 2.5 times in 2004. L’Abée-Lund (1989) demonstrated that within a small population of Atlantic salmon, mature male parr increased the effective population size above the recommended minimum (Allendorf and Ryman 1987). Mature male LGB parr likely have similar positive effects on effective population size. Indirect molecular evidence suggests that non-anadromous male steelhead achieve a large number of fertilizations (Seamons et al. 2004). Although not well studied in steelhead, the reproductive success of mature parr is high in other salmonids. In Atlantic salmon (Salmo salar), precocious parr have demonstrated large reproductive contributions to individual redds: 1-29% (Hutchings and Myers 1988), 1-28% (Jordan and Youngson 1992), 26-40% (Thomaz et al. 1997), and 45-86% (Garcia-Vazquez et al. 2001). In situations of sperm competition, sneak spawners experience higher fertilization success than larger guarding males (Fu et al. 2001). Precocious male O. mykiss demonstrate elevated sperm concentrations, potentially compensating for their lower milt volume relative to larger adults (Liley et al. 2002). Since the 2004 LGB parr population was more than three times that from the
previous LGB cohort, the relative reproductive contributions of male LGB parr may increase dramatically in subsequent years.

LGB parr may even experience higher fitness than wild and LGB anadromous fish. The fitness of a life-history tactic is defined by the probability of surviving to maturity and by the reproductive success of that tactic (Gross 1991). In 2004, the average time from release to maturity was 1.36 years for mature LGB parr (7 were 2003 LGB parr, 4 were 2002 LGB parr). Assuming that: 1) all mature LGB parr were ripe and/or spawning for the first time in 2004; 2) constant probability of survival within a year; as annual LGB parr survival is 56%, the probability of surviving 1.36 years to first maturity is 41%. The previous two cohorts of anadromous wild and LGB adults averaged 1.83% and 1.10% survival to maturity, respectively. Thus, LGB parr are about 22 and 37 times more likely to survive to maturity than wild and LGB anadromous adults, respectively. Therefore, to equal the fitness of wild and LGB anadromous adults, mature LGB parr must only achieve 4.5% and 2.7% of their respective reproductive successes. Given the demonstrated reproductive success of precocious salmonids and the potential for LGB parr to spawn in freshwater or migrate to the ocean in multiple years, the resident-breeder LGB parr tactic likely experiences greater fitness than wild and LGB anadromous life-histories.

Spawning by mature parr may also reduce inbreeding rates. Some LGB parr reached spawning condition within one year of release, whereas few wild and LGB anadromous fish did the same. Due to their similar total age, wild and LGB anadromous fish likely experience similar probabilities of producing inbred crosses. In contrast, cross-cohort spawning by precocious parr serves to avoid losses of genetic variability in small populations (Moran and Garcia-Vazquez 1998; Garcia-Vazquez et al. 2001). Thus, as LGB parr apparently mature and spawn at a younger age, they may conserve genetic diversity by increasing cross-cohort spawning and reducing the probability of inbreeding in this small population.

As the LGB program aimed to release hatchery fish that would spawn with wild adults, mature parr may also increase the rates of cross-breeding between LGB and wild fish. As spawning between wild males and hatchery females can be more common than the converse (e.g. Jonsson 1997), LGB parr spawning may increase the relative rate of hatchery male and wild female crossing. By increasing the probability that wild-hatchery crosses occur in both directions, LGB parr provide an alternative way to interbreed wild and hatchery fish.

Mature parr may also influence the relative amount and effect of gene flow among depressed populations of steelhead. As mature parr do not migrate prior to spawning they do not
risk ‘straying’ into adjacent rivers upon completing their marine migration – a phenomenon well-documented within some anadromous salmonids (for a review see Quinn 1993). As the magnitude of gene flow is partially determined by population size (Endler 1977), increases in mature parr numbers may increase the effective population size and thereby decrease the relative reproductive contributions of strays from adjacent rivers. Hence, during periods of population decline, spawning by mature resident hatchery steelhead may minimize the genetic homogenization with adjacent populations.

Although resident hatchery parr may benefit conservation, increasing the number and biomass of steelhead present in the river may also be negative. Keogh River steelhead fry primarily feed on benthic invertebrates (Johnston et al. 1990), including similar invertebrate orders as LGB parr. Due to dietary overlap with wild juveniles, LGB parr may increase food competition within the Keogh River.

LGB parr also cause dramatic increases to food consumption by steelhead. In summer 2004, LGB parr increased total parr biomass by an estimated 120%, or total juvenile biomass (fry and parr) by 76%. Thus, LGB parr potentially increased steelhead food consumption in the river by 41% in 2004. As habitat fertilization increased juvenile survival (Ward et al. 2003), food availability apparently limits juvenile production in the Keogh River. By dramatically increasing food consumption, LGB parr may increase food competition and greatly reduce the positive effects of fertilization on juvenile production.

LGB parr tend to be larger than wild parr and fry. As territory size increases with increasing body size (Abbott et al. 1985; Keeley 2000) and increases in body size tend to increase dominancy (MacPhee 1961), LGB parr have the potential to dominate and displace smaller wild conspecifics. Juvenile salmonid growth may also be limited by population density (Egglishaw and Shackley 1977; Randall 1982; Bergheim and Hesthagen 1990; Keeley 2001). Non-migrant hatchery steelhead increase salmonid density and biomass (Viola and Schuck 1995) resulting in reduced wild salmonid growth (McMichael 1997). Reductions in wild juvenile size could decrease overwinter survival (Oliver and Holeton 1979; Toney and Coble 1979, 1980; Biro et al. 2004). Thus, due to their larger size and large numbers, LGB parr may displace and reduce the growth and survival of wild juveniles.

LGB parr also negatively influence wild population recovery by preying on steelhead fry. Although I estimate that 118 LGB parr preyed on steelhead fry, fry predation rates were likely higher in 2004 for two reasons: 1) immediately after fry emergence, LGB parr, and the sampling efforts for them, were concentrated in lower reaches of the river (J.C. Walters and M.R. Gross

Reproduced with permission of the copyright owner. Further reproduction prohibited without permission.
unpublished data) that are known to have low fry output (B. Ward personal communication 20 July 2005); and 2) as the 2004 LGB parr disperse upstream they encounter higher fry densities and likely increase their fry predation accordingly. If low fry output coincides with large LGB parr numbers, fry predation by LGB parr may have detrimental effects on fry survival and subsequent wild smolt recruitment. For example, if each piscivorous LGB parr consumed only three fry per week, from fry emergence in June until early October (16 weeks), about 10% of all 2004 fry would be consumed. In reality, fry predation may be more or less intense than this scenario, but this demonstrates how a small number of piscivorous LGB parr may significantly reduce fry survival and subsequent wild smolt production.

Conservation actions may not only alter demographic, life-history, and ecological patterns in steelhead – the LGB program may have inadvertently amplified the wrong species’ genes. Despite the early published accounts of steelhead and coastal cutthroat trout hybridization in natural streams (e.g. Campton and Utter 1985), LGB broodstock were not screened for the presence of cutthroat trout genes. Hybridization is widespread and highly variable where steelhead and coastal cutthroat are sympatric on Vancouver Island; found in 29 of 30 sampled streams, at rates of 3-88% (Bettes 2004). In 2002, of 50 individuals visually identified as steelhead from the Keogh River, 6% were determined to be hybrids (Ostberg 2003 in McCubbing and Ward 2003). As the probability of correctly visually classifying a juvenile steelhead can be low in sympatric hybridizing trout populations (Baumsteiger et al. 2005), the LGB program may have inadvertently altered hybridization rates through broodstock collection and subsequent spawning.

Hybridization could be detrimental to the preservation of steelhead genetic diversity: the genetic extinction of parental genotypes may ultimately result from the transfer of genes between parental gene pools (Rhymer and Simberloff 1996). The predicted benefits for conservation of the LGB program hinge upon amplifying the target species’ genes and numbers. As hybrid broodstock may have been incorporated into the LGB program, I discuss the conservation issues related to hybridization in LGB progeny, assuming that the natural wild rate in the Keogh River is about 5% (Table 12).

The benefits for steelhead conservation are greatest when the LGB program does not produce hybrids (Table 12). In contrast, as the hybrid proportion of LGB progeny increases, the ability of the LGB program to conserve steelhead trout genes decreases (Table 12). Although hybrid fitness may be higher, lower, or equivalent to the parental types (Edmands and Timmerman 2003), even when hybrid fitness is very low (e.g. 10% of the parental type)
introgression may spread and eliminate the parent type within a few generations (Epifanio and Philipp 2001). Because all progeny of hybrids are hybrids themselves (Allendorf et al. 2004), introgression can rapidly spread throughout a population, resulting in a 'hybrid swarm'. If the LGB program produces hybrids at a similar frequency as in the wild, there will be nearly a 53-fold increase in hybrid smolt abundance (Table 12). Hybrids may fertilize pure steelhead eggs and increase the spread of introgression or, if hybrid fitness is low, hybrid spawning may increase steelhead gamete wastage. Thus, if the LGB program produced any hybrids, wild steelhead conservation will likely suffer if those hybrids spawn.

Due to the differences in steelhead and coastal cutthroat trout life-histories, hybridization may decrease marine survival and alter life-history frequencies. As steelhead undertake extensive migrations and coastal cutthroat trout rarely stray far from the estuary (Wydoski and Whitney 1979), non-migratory LGB parr may be hybrids expressing the coastal cutthroat life-history, rather than pure steelhead forgoing ocean migration – this hypothesis will be tested by future molecular analyses by the Gross Lab. Also, as the swimming ability of hybrids is intermediate to those of pure steelhead and coastal cutthroat trout (Hawkins and Quinn 1996), the phenotypically intermediate hybrids may be selected against in the ocean environment (Young et al. 2001). Regardless of hybridization rates, introgression is negative for wild steelhead conservation and should be avoided.
Table 1. Terms used to differentiate among wild and LGB steelhead life-history tactics, ontogenetic stages, and cohorts; year designations emphasize attributes pertinent to the focus of this study.

<table>
<thead>
<tr>
<th>Fish type</th>
<th>Definition</th>
</tr>
</thead>
<tbody>
<tr>
<td>LGB anadromous adult</td>
<td>Andadromous fish that have returned to the river to spawn, but originate as smolt from the LGB program. Where necessary, referred to by the year of their return to freshwater (e.g. “2002 anadromous LGB adult”) – emphasizing spawning year. “Kelt” refers to a post-spawned individual.</td>
</tr>
<tr>
<td>LGB smolt</td>
<td>Fish produced by the LGB program. This includes all fish in the net pens prior to release, and all fish released. Depending on life-history tactic, these fish are later identified as LGB parr or LGB migrants. Where necessary, referred to by release year of (e.g. “2004 LGB smolt”) – emphasizing release year.</td>
</tr>
<tr>
<td>LGB parr</td>
<td>The portion of the LGB smolt population that become resident in the river at release – likely having never undergone smoltification. Any LGB smolt residing in the river after 15 June was assigned to this category. Historically, residents have been termed “residuals” or “residual fish” (e.g. McMichael 1997; Viola and Schuck 1995; Seelbach 1987). Where necessary, referred to by the year of release (e.g. “2002 LGB parr”) – emphasizing duration of river residency. The 2004 LGB parr were sampled at three times - early, middle and late (e.g. “late 2004 LGB parr”). LGB parr exhibit three tactics: 1) delayed-migrants – individuals that remain within the river, to outmigrate as unspawned smolt in subsequent years; 2) resident breeders – individuals that remain in the river to spawn as precocious parr; and 3) delayed-resident-breeders – individuals that remain as immature parr within the river after release, to become precocious parr in subsequent years.</td>
</tr>
<tr>
<td>LGB migrants</td>
<td>The portion of the LGB smolt population that chose to immediately migrate to the ocean at release. Where necessary, referred to by year of release (e.g. “2004 LGB migrants”) – emphasizing year of outmigration and origin.</td>
</tr>
<tr>
<td>LGB fry</td>
<td>Fry that were produced by the spawning of anadromous LGB females in the wild. Where necessary, referred to by their year of hatch (e.g. “2004 LGB fry”) – emphasizing year produced and spawning year of their parents.</td>
</tr>
<tr>
<td>Wild anadromous adult</td>
<td>Anadromous fish that have returned to the river to spawn and originated as smolt from the river as opposed to the LGB program. Where necessary, referred to by the year of their return to freshwater (e.g. “2002 anadromous wild adult”) – emphasizing spawning year. “Kelt” refers to a post-spawned individual.</td>
</tr>
<tr>
<td>Wild smolt</td>
<td>Smolt that were produced by wild-origin parents. Where necessary, referred to by their year of migration from freshwater (e.g. “2004 wild smolt”) – emphasizing year of outmigration.</td>
</tr>
<tr>
<td>Wild parr</td>
<td>Parr that were produced by wild-origin parents. Where necessary, referred to by age and year of hatch (e.g. “1+ parr (2003 cohort)”) – emphasizing year produced, age and spawning year of their parents.</td>
</tr>
<tr>
<td>Wild fry</td>
<td>Fry that were produced by anadromous wild female spawning. Where necessary, referred to by their year of hatch (e.g. “2004 wild fry”) – emphasizing year produced and spawning year of their parents.</td>
</tr>
</tbody>
</table>
Table 2. Timing and methods of sample collection, data collected and sample size for developmental stages and life-history types in wild and LGB origin fish. (L=fork length, M=mass, S=sex, F=gonad factor, A=age estimation, G=gut contents)

<table>
<thead>
<tr>
<th>Origin</th>
<th>Fish type</th>
<th>Collection Period</th>
<th>Total</th>
<th>Sample Size</th>
<th>Collection Methods Used</th>
</tr>
</thead>
<tbody>
<tr>
<td>Wild</td>
<td>anadromous adults:</td>
<td>18 December 2003 - 16 July 2004</td>
<td>21</td>
<td>21</td>
<td>Angling, trap, seine</td>
</tr>
<tr>
<td></td>
<td>smolt: 2004</td>
<td>5 April - 24 May 2004</td>
<td>362</td>
<td>362^2</td>
<td>Trap</td>
</tr>
<tr>
<td></td>
<td>parr: 2002-2003</td>
<td>4-17 August 2004</td>
<td>45</td>
<td>45^1</td>
<td>Angling and trap</td>
</tr>
<tr>
<td>LGB</td>
<td>anadromous adults:</td>
<td>18 December 2003 - 16 July 2004</td>
<td>57</td>
<td>57^1</td>
<td>Angling, trap, seine</td>
</tr>
<tr>
<td></td>
<td>smolt: 2004</td>
<td>17-26 May 2004</td>
<td>301</td>
<td>301</td>
<td>Dipnet</td>
</tr>
<tr>
<td></td>
<td>parr: early 2004</td>
<td>15-20 June 2004</td>
<td>446</td>
<td>442^2</td>
<td>Angling and trap</td>
</tr>
<tr>
<td></td>
<td>parr: middle 2004</td>
<td>7-21 July 2004</td>
<td>284</td>
<td>284</td>
<td>Angling and trap</td>
</tr>
<tr>
<td></td>
<td>parr: late 2004</td>
<td>4-13 August 2004</td>
<td>83</td>
<td>79</td>
<td>Angling and trap</td>
</tr>
</tbody>
</table>

1 Released live due to concerns regarding small population size
2 Contains both individuals released live and euthanized
3 Scale samples collected, but not age verified – age known to be 1 year
Table 3. Population estimates, sex ratio, average mass, and estimated biomass of LGB and wild fish in 2004.

<table>
<thead>
<tr>
<th>Origin</th>
<th>Fish Type</th>
<th>Population Estimate</th>
<th>95% Probability Region</th>
<th>Sex (M; F)</th>
<th>Average Mass g ± SE (N)</th>
<th>Estimated Biomass (kg)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Wild</td>
<td>anadromous females</td>
<td>53</td>
<td>33 – 192^1</td>
<td>N/A</td>
<td>5256 ±405.6 (12)</td>
<td>278.57</td>
</tr>
<tr>
<td></td>
<td>anadromous males</td>
<td>33</td>
<td>None^1</td>
<td>N/A</td>
<td>6947 ±470.8 (5)</td>
<td>222.31</td>
</tr>
<tr>
<td></td>
<td>anadromous total</td>
<td>85</td>
<td>35 – 210^1</td>
<td>1.00:1.66</td>
<td>N/A</td>
<td>500.88^10</td>
</tr>
<tr>
<td></td>
<td>smolt</td>
<td>511</td>
<td>N/A^4</td>
<td>19, 24</td>
<td>53.67 ±2.43 (46)</td>
<td>27.43</td>
</tr>
<tr>
<td></td>
<td>parr: age 2+ (2002 cohort)</td>
<td>1382</td>
<td>N/A^4</td>
<td>N/A</td>
<td>36.81 ±2.02 (19)</td>
<td>50.87</td>
</tr>
<tr>
<td></td>
<td>parr: age 1+ (2003 cohort)</td>
<td>3,673</td>
<td>N/A^4</td>
<td>N/A</td>
<td>24.60 ±1.80 (10)</td>
<td>90.36</td>
</tr>
<tr>
<td></td>
<td>fry</td>
<td>16,240</td>
<td>N/A^4</td>
<td>N/A</td>
<td>1.98^7</td>
<td>32.15</td>
</tr>
<tr>
<td>LGB</td>
<td>anadromous females</td>
<td>146</td>
<td>60 – 362^2</td>
<td>134, 151</td>
<td>4880 ±251.6 (17)</td>
<td>712.45</td>
</tr>
<tr>
<td></td>
<td>anadromous males</td>
<td>130</td>
<td>62 – 299^2</td>
<td>1.00:1.12</td>
<td>N/A</td>
<td>636.86</td>
</tr>
<tr>
<td></td>
<td>anadromous total</td>
<td>276</td>
<td>130-705^2</td>
<td>N/A^6</td>
<td>79.29^8</td>
<td>2180.70</td>
</tr>
<tr>
<td></td>
<td>smolt</td>
<td>27,506</td>
<td>N/A^4</td>
<td>1.00:1.34</td>
<td>79.29^9</td>
<td>2050.99</td>
</tr>
<tr>
<td></td>
<td>migrants</td>
<td>25,872^1</td>
<td>N/A^4</td>
<td>53, 24</td>
<td>128.97 ±7.74 (99)</td>
<td>75.45</td>
</tr>
<tr>
<td></td>
<td>parr: 2001-2003</td>
<td>585</td>
<td>283-983^3</td>
<td>1.00:0.45</td>
<td>57.82 ±1.57 (446)</td>
<td>94.48</td>
</tr>
<tr>
<td></td>
<td>parr: early 2004</td>
<td>1,634</td>
<td>1441-1791^4</td>
<td>1.00:0.45</td>
<td>55.87 ±1.88 (284)</td>
<td>N/A</td>
</tr>
<tr>
<td></td>
<td>parr: middle 2004</td>
<td>N/A</td>
<td>N/A</td>
<td>1.00:0.48</td>
<td>84.25 ±4.31 (79)</td>
<td>N/A</td>
</tr>
<tr>
<td></td>
<td>parr: late 2004</td>
<td>N/A</td>
<td>N/A</td>
<td>1.00:0.41</td>
<td>1.98^7</td>
<td>81.10</td>
</tr>
</tbody>
</table>

^1 Calculated by subtracting early 2004 LGB parr from LGB smolt
^2 Confidence interval (Zar 1984)
^3 Probability region not possible as no recaptures occurred
^4 Probability region not possible due to estimation method
^5 Highest probability density (Gazey and Staley 1986)
^6 Calculated from corresponding anadromous male and female population estimates
^7 From McCubbing and Ward (2002)
^8 Calculated by dividing the total biomass by population size
^9 For demonstrative purposes, assumed to be similar to LGB smolt
^10 Calculated by summing the estimated biomass for males and females
Table 4. Population estimates for each river reach from snorkel surveys conducted in 2003.

<table>
<thead>
<tr>
<th>Reach name</th>
<th>Length (m)</th>
<th>Estimated density in reach (fish(m^{-1}))</th>
<th>Population estimate</th>
</tr>
</thead>
<tbody>
<tr>
<td>A</td>
<td>1100</td>
<td>0.182</td>
<td>200</td>
</tr>
<tr>
<td>B</td>
<td>2250</td>
<td>0.140</td>
<td>315</td>
</tr>
<tr>
<td>C</td>
<td>6300</td>
<td>0.003</td>
<td>19</td>
</tr>
<tr>
<td>D</td>
<td>21050</td>
<td>0</td>
<td>0</td>
</tr>
</tbody>
</table>

\(^1\) No probability region due to no recaptures
Table 5. Freshwater, ocean and total ages of male and female wild and LGB anadromous adult steelhead from the Koehl River in 2004 by scale analysis (1s denotes repeat spawner: 1st spawning at ocean age-2, 2nd spawning at ocean age-3; wild age data from McCubbing and Ward (2004)).

<table>
<thead>
<tr>
<th>Origin</th>
<th>Sex</th>
<th>Freshwater Age</th>
<th>Ocean Age</th>
<th>Total Age</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>% (N)</td>
<td>% (N)</td>
<td>% (N)</td>
</tr>
<tr>
<td>Wild</td>
<td>M</td>
<td>0 80 20 22.2</td>
<td>11.1 22.2</td>
<td>44.4 20.0</td>
</tr>
<tr>
<td></td>
<td></td>
<td>(4) (1) (2)</td>
<td>(1) (2)</td>
<td>(4) (1)</td>
</tr>
<tr>
<td>Wild</td>
<td>F</td>
<td>0 100 0 0</td>
<td>8.3 25</td>
<td>66.7 12.5</td>
</tr>
<tr>
<td></td>
<td></td>
<td>(8)</td>
<td>(1) (3)</td>
<td>(8) (1)</td>
</tr>
<tr>
<td>LGB</td>
<td>M</td>
<td>100 0 0 0</td>
<td>34.3 51.4</td>
<td>14.3 34.2</td>
</tr>
<tr>
<td></td>
<td></td>
<td>(35)</td>
<td>(12) (18)</td>
<td>(5) (12)</td>
</tr>
<tr>
<td>LGB</td>
<td>F</td>
<td>100 0 0 0</td>
<td>13.6 68.2</td>
<td>18.2 13.6</td>
</tr>
<tr>
<td></td>
<td></td>
<td>(22)</td>
<td>(3) (15)</td>
<td>(4) (3)</td>
</tr>
</tbody>
</table>

Reproduced with permission of the copyright owner. Further reproduction prohibited without permission.
Table 6. Age classification of wild and LGB juveniles from the Keogh River; wild fish were aged by scale analysis. LGB fish are of known age (regenerated scales had incomplete freshwater histories and could not be aged).

<table>
<thead>
<tr>
<th>Origin</th>
<th>Sex</th>
<th>Freshwater age % (N)</th>
<th>Regenerated</th>
<th>Total N</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>1 (1)</td>
<td>2 (10)</td>
<td>3 (4)</td>
</tr>
<tr>
<td></td>
<td>M</td>
<td>5 (1)</td>
<td>53 (10)</td>
<td>21 (4)</td>
</tr>
<tr>
<td></td>
<td>F</td>
<td>4 (1)</td>
<td>71 (17)</td>
<td>25 (6)</td>
</tr>
<tr>
<td></td>
<td>Both Sexes</td>
<td>5 (2)</td>
<td>63 (27)</td>
<td>23 (10)</td>
</tr>
<tr>
<td>LGB smolt</td>
<td>M</td>
<td>100 (134)</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td></td>
<td>F</td>
<td>100 (151)</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td></td>
<td>Both Sexes</td>
<td>100 (285)</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>Wild parr</td>
<td>Both sexes</td>
<td>30 (14)</td>
<td>46 (21)</td>
<td>7 (3)</td>
</tr>
<tr>
<td>Origin</td>
<td>Fish Type</td>
<td>Male Gonad Factor Scores</td>
<td>Female Gonad Factor Scores</td>
<td></td>
</tr>
<tr>
<td>------------</td>
<td>---------------</td>
<td>--------------------------</td>
<td>---------------------------</td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td>% (N)</td>
<td>% (N)</td>
<td></td>
</tr>
<tr>
<td>Wild</td>
<td>smolt</td>
<td>100 (19)</td>
<td>100 (24)</td>
<td></td>
</tr>
<tr>
<td>LGB</td>
<td>smolt</td>
<td>91.8 (123)</td>
<td>70.2 (106)</td>
<td></td>
</tr>
<tr>
<td></td>
<td>parr: early 2004</td>
<td>50 (19)</td>
<td>88.2 (15)</td>
<td></td>
</tr>
<tr>
<td></td>
<td>parr: middle 2004</td>
<td>63 (119)</td>
<td>77.7 (70)</td>
<td></td>
</tr>
<tr>
<td></td>
<td>parr: late 2004</td>
<td>41.5 (17)</td>
<td>88.2 (15)</td>
<td></td>
</tr>
<tr>
<td></td>
<td>parr: 2001-2003</td>
<td>60 (30)</td>
<td>40.9 (9)</td>
<td></td>
</tr>
</tbody>
</table>
Table 8. Mass and fork lengths of wild smolt, LGB smolt and 2004 LGB parr expressing various gonad factor (GF) scores.

<table>
<thead>
<tr>
<th>Origin</th>
<th>Fish Type</th>
<th>Mass g ±SD (N)</th>
<th>Length cm ±SD (N)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>GF 1</td>
<td>GF 2</td>
</tr>
<tr>
<td>Wild</td>
<td>smolt</td>
<td>53.67 ±16.53 (46)</td>
<td>-</td>
</tr>
<tr>
<td>LGB</td>
<td>smolt</td>
<td>82.09 ±25.80 (241)</td>
<td>100.95 ±29.20 (42)</td>
</tr>
<tr>
<td></td>
<td>parr: early 2004</td>
<td>85.51 ±34.71 (34)</td>
<td>95.59 ±35.27 (13)</td>
</tr>
<tr>
<td></td>
<td>parr: middle 2004</td>
<td>47.44 ±24.82 (189)</td>
<td>70.87 ±33.32 (59)</td>
</tr>
<tr>
<td></td>
<td>parr: late 2004</td>
<td>75.11 ±30.40 (34)</td>
<td>95.68 ±32.33 (16)</td>
</tr>
</tbody>
</table>
Table 9. Potential egg deposition by wild and LGB anadromous and resident females from 2002-2004.

<table>
<thead>
<tr>
<th>Spawning Year</th>
<th>Origin</th>
<th>Life-History</th>
<th>Estimated Fecundity eggs ± SD (N)</th>
<th>Estimated Number of Females</th>
<th>Potential Egg Deposition</th>
</tr>
</thead>
<tbody>
<tr>
<td>2004</td>
<td>Wild</td>
<td>anadromous adult</td>
<td>4 692 ± 351 (12)</td>
<td>53</td>
<td>248 676</td>
</tr>
<tr>
<td></td>
<td>LGB</td>
<td>anadromous adult</td>
<td>4 296 ± 184 (17)</td>
<td>146</td>
<td>627 216</td>
</tr>
<tr>
<td></td>
<td>LGB</td>
<td>resident parr</td>
<td>316 ± 5.0 (2)</td>
<td>25</td>
<td>7 900</td>
</tr>
<tr>
<td>2003</td>
<td>Wild</td>
<td>anadromous adult</td>
<td>4 559 ± 975 (26)</td>
<td>90</td>
<td>410 364</td>
</tr>
<tr>
<td></td>
<td>LGB</td>
<td>anadromous adult</td>
<td>2 287 ± 435 (41)</td>
<td>99</td>
<td>226 443</td>
</tr>
<tr>
<td>2002</td>
<td>Wild</td>
<td>anadromous adult</td>
<td>4 118 ± 1 180 (33)</td>
<td>65</td>
<td>267 642</td>
</tr>
<tr>
<td></td>
<td>LGB</td>
<td>anadromous adult</td>
<td>-</td>
<td>0</td>
<td>0</td>
</tr>
</tbody>
</table>
Table 10. LGB parr feeding success and the diversity of invertebrates each fish type consumed at the Keogh River in 2004.

<table>
<thead>
<tr>
<th>Fish Type</th>
<th>N</th>
<th>Empty % (N)</th>
<th>Food only % (N)</th>
<th>Non-food % (N)</th>
<th>Observed</th>
</tr>
</thead>
<tbody>
<tr>
<td>parr: early 2004</td>
<td>62</td>
<td>4.8 (3)</td>
<td>53.2 (33)</td>
<td>41.9 (26)</td>
<td>4</td>
</tr>
<tr>
<td>parr: middle 2004</td>
<td>93</td>
<td>48.4 (45)</td>
<td>15.0 (14)</td>
<td>36.6 (34)</td>
<td>4</td>
</tr>
<tr>
<td>parr: late 2004</td>
<td>83</td>
<td>8.4 (7)</td>
<td>60.2 (50)</td>
<td>31.3 (26)</td>
<td>12</td>
</tr>
<tr>
<td>parr: 2001-2003</td>
<td>76</td>
<td>23.7 (18)</td>
<td>69.7 (53)</td>
<td>6.6 (5)</td>
<td>4</td>
</tr>
</tbody>
</table>

1 Feeding success categories defined in Methods.
Table 11. Allometrically scaled relative food consumption for wild and LGB fish on an individual- and population-specific basis during summer 2004 ($\alpha=0.73$, Steingrimsson and Grant 1999).

<table>
<thead>
<tr>
<th>Fish Type</th>
<th>Average mass $W$ (in g)</th>
<th>$W^{\alpha}$</th>
<th>$W^{\alpha} \times$ Population Size $^1$</th>
</tr>
</thead>
<tbody>
<tr>
<td>Wild fry</td>
<td>1.98</td>
<td>1.65</td>
<td>94,380</td>
</tr>
<tr>
<td>Wild 1+ parr</td>
<td>34.60</td>
<td>10.36</td>
<td>38,052</td>
</tr>
<tr>
<td>Wild 2+ parr</td>
<td>36.81</td>
<td>13.90</td>
<td>19,210</td>
</tr>
<tr>
<td>Wild smolt</td>
<td>53.67</td>
<td>18.31</td>
<td>9,356</td>
</tr>
<tr>
<td>LGB migrants</td>
<td>79.29</td>
<td>24.35</td>
<td>629,861</td>
</tr>
<tr>
<td>LGB parr. late 2004</td>
<td>84.25</td>
<td>25.45</td>
<td>41,585</td>
</tr>
<tr>
<td>LGB parr. 2001-2003</td>
<td>128.97</td>
<td>34.73</td>
<td>20,317</td>
</tr>
</tbody>
</table>

$^1$ Population estimates in Table 2
Table 12. Issues for wild steelhead conservation of various steelhead-coastal cutthroat trout hybridization rates in LGB progeny.

<table>
<thead>
<tr>
<th>Conservation Issues</th>
<th>No Hybridization (0%)</th>
<th>Equivalent to natural hybridization (~5%)</th>
<th>Elevated Hybridization (10-100%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Use of conservation resources</td>
<td>No waste of conservation resources</td>
<td>Amplified genes of non-target species at natural rates, mixed species gene pools in captivity, ~5% of LGB production is wasted</td>
<td>Amplified genes of non-target species, mixed species gene pools in captivity, 10-100% of LGB production is wasted</td>
</tr>
<tr>
<td>Probability of hybrid swarm</td>
<td>LGB-induced hybrid swarm improbable</td>
<td>Hybrid swarm unlikely -- hybridization rate similar to wild, but still possible due to LGB progeny abundance (e.g. in 2004: 5% of 27,508 LGB progeny = 1375 hybrids, vs. 5% of 511 wild smolt = 26 hybrids)</td>
<td>Probability of hybrid swarm increases with increased rate of hybridization</td>
</tr>
<tr>
<td>Potential explanation of alternative life-history expression in LGB progeny</td>
<td>LGB parr result from alternative life-histories in O. mykiss</td>
<td>LGB parr may result from hybrids expressing coastal cutthroat life-history and/or from alternative life-histories in O. mykiss</td>
<td>LGB parr may result from hybrids expressing coastal cutthroat life-history and/or from alternative life-histories in O. mykiss</td>
</tr>
<tr>
<td>Hybrid fitness</td>
<td>No problems related to hybrid fitness</td>
<td>1) Elevated hybrid fitness may disrupt/dilute natural steelhead genes</td>
<td>1) Elevated hybrid fitness may disrupt/dilute natural steelhead genes</td>
</tr>
<tr>
<td>Implications of LGB parr spawning</td>
<td>Spawning by LGB parr likely benefits conservation</td>
<td>2) Reduced hybrid fitness may increase steelhead gamete wastage</td>
<td>2) Reduced hybrid fitness may increase steelhead gamete wastage</td>
</tr>
<tr>
<td>Marine survival</td>
<td>Hybridization does not influence marine survival</td>
<td>Probability of marine survival likely reduced for hybrids</td>
<td>Probability of marine survival likely decreases with increased hybridization</td>
</tr>
</tbody>
</table>

37
Figure 1. Schematic diagram of life-history patterns exhibited by LGB and wild fish at the Keogh River (based on our initial observations and: McCubbing (2002); McCubbing and Ward (2003, 2004); and Werlen (2003)).
smolt

hatchery

spawn in the hatchery after 2-3 years

LGB Wild

ocean

return to spawn in river after 1-3 years at sea

adult

moved to lake-based net pens at parr stage

fry

parr

Release (1 year old)

migrate to ocean after 1-3 years of rearing in river

adult

smolt

breed

Ocean
Figure 2. Keogh River watershed area and location on Vancouver Island (Courtesy of www.steelheadrecoveryplan.ca).

Reproduced with permission of the copyright owner. Further reproduction prohibited without permission.
Figure 3. Schematic diagram of the Keogh River enumeration fence infrastructure (adapted from Ammann et al. 2005a).
Figure 4. Marine survival estimates for wild and LGB smolt cohorts with total smolt output (wild + LGB). Note that 2001 was the first release of a LGB cohort (with data from Ammann et al. (2005a) and McCubbing and Ward (2004)).
Part 3: Phenotypic divergence in a conservation hatchery
Introduction

Conservation programs increasingly use captive rearing and breeding in an attempt to save wild salmonid populations. These programs use various designs, with the common goal of retaining wild genotypic and phenotypic structure. This is paradoxical: the rearing in artificial environments will alter the phenotype. Thus, the questions are: 1) How much will the conservation program change the phenotype, and what are the ramifications of these changes in terms of survival and reproductive success of the captive-bred as well as the wild individuals? 2) Will the phenotypic change decrease the survival and reproductive success, or otherwise impact the wild population, to such a degree that the conservation program is a failure? 3) What changes can be implemented to mitigate the negatives and enhance the positives?

Thus far, few studies have provided clear and complete answers to these questions. Researchers have investigated the influences of culture on salmonid phenotype, but results rarely suggest how the observed phenotypic change may influence the subsequent survival and reproduction of wild and hatchery-produced populations. Hatchery-origin fish have larger gonads, investing more energy in gamete production than wild conspecifics (Fleming and Gross 1992). Although they may invest more in gametes, evidence suggests that cultured fish experience reduced breeding success (Chilcote et al. 1986; Fleming and Gross 1993, 1994; Berejikian et al. 1997; Kostow et al. 2003; McLean et al. 2004), although phenotypic divergence from the wild type is rarely investigated as a contributing factor. Morphological divergence occurs between cultured and wild juveniles (Taylor 1986; Swain et al. 1991; Fleming et al. 1994; Kostow 2004), although little attention has been paid to the implications of these differences on subsequent survival. Conversely, where phenotypic effects on survival have been considered, studies have neglected body shape and most morphological characters other than length and mass (e.g. Kostow 2004).

In 1998, as a response to the precipitous decline of steelhead populations in many areas of British Columbia (BC) (Welch et al. 2000), the BC government initiated the “Living Gene Bank” (LGB) – a conservation hatchery program for the Keogh River steelhead population (Wightman et al. 1998). The LGB program implemented sophisticated rearing protocols, with the primary goal of maintaining population genetic diversity (Part 2, Ammann et al. 2005a). Phenotype management within the LGB program included segregating progeny into size classes and rearing them in low densities to produce healthy fish (i.e. low rate of disease. Halpenny 2005 - MSc thesis in prep.), that were well-fed and large relative to wild conspecifics (Ammann et al.
There was no specific program to maintain other phenotypic characters, such as shape, body dimensions, and internal organ size.

Salmonid morphology is known to be very plastic. Morphological variation in salmonids has been demonstrated to have environmental origins (Winans 1984; Taylor 1986; Currens et al. 1989; Swain et al. 1991; Hard et al. 2000). Thus, rearing salmonids in an unnatural environment will lead to phenotypic change. Life-history patterns also change in response to culture (in Part 2). Thus, the unnatural rearing environment of conservation hatcheries may greatly influence morphological and life-history patterns expressed by captive-bred salmonids.

In addition to the prevalent anadromous life-history, the LGB program has produced resident parr—fish that likely have not smoltified and do not migrate to the ocean upon release, but remain in the river for one or more years. Three life-histories are demonstrated by the resident LGB parr: 1) delayed-migrants—individuals that remain within the river and outmigrate as smolt in subsequent years; 2) resident breeders—individuals that continue maturing upon release and become precocious parr in the river; and 3) delayed-resident-breeders—individuals that remain as immature parr immediately after release, but become precocious parr in subsequent years.

In this part, I examine the influences of a conservation-focused captive broodstock program on juvenile phenotype and consider the implications on fitness of any deviations of the LGB fish from the wild type. I compare the morphology of various wild and LGB life-history tactics and ontogenetic stages utilizing a combination of geometric and traditional morphometric techniques.

Geometric morphometrics uses the positions of homologous anatomical landmarks to evaluate shape variation, whereas traditional morphometrics includes the multivariate analysis of distances measured on an organism (Rohlf and Marcus 1993). Recently, morphometric methods have focused on the analysis of landmark data as landmarks correspond to physical locations, thereby retaining the geometric relationships among structures (Rohlf 1990; Rohlf and Slice 1990; Rohlf and Marcus 1993). Shape variation is then characterized by analyzing partial-warp scores using the thin-plate spline (Bookstein 1989, 1991). Traditional morphometric analyses allow the investigation into the influences of culture on other important phenotypic characters, including the length of fins, internal organ masses, and the lengths of and linear distances between various structures. Lastly, I contrast the size differences between hatchery and wild juveniles, and discuss the reproductive and ecological implications of any differences.

Consequently, these morphometric analyses demonstrate how captive rearing can inadvertently
influence the phenotype of juvenile salmonids in sophisticated conservation programs. I conclude by discussing the implications of phenotypic changes for population recovery.

Methods

Study site and specimen collection

Specimens were collected from the Keogh River, British Columbia, Canada, between May 12 and August 17, 2004. The Keogh River, at the northeastern end of Vancouver Island, is a third-order coastal stream, 31km in length, with a drainage of 130km² and a mean annual discharge of 5.6m³s⁻¹, and has been detailed elsewhere (Ward and Slaney 1979, 1988, 1993a; Johnston et al. 1986; Irvine and Ward 1989; Johnston et al. 1990).

At the time of sampling, one does not know whether an individual remained in the river to adopt a resident-breeder, delayed-resident-breeder, or delayed-migrant tactic. Thus, as the wild smolt run was effectively complete by 15 June (McCubbing and Ward 2004), any LGB progeny encountered in-stream after this date was assumed to be resident and referred to as “LGB parr” (Table 1).

I sampled six fish types, differentiated by life-history tactic (parr or smolt) and origin (wild or LGB) (Table 1). In addition, as I sampled the LGB parr at three times within a single season, I refer to the three samplings as early, middle, and late (i.e. “early LGB parr”). I staggered the three LGB parr samplings by about one-month intervals to determine if LGB parr phenotype changed within a season in response to the river environment. Depending on fish type, specimen collection utilized a combination of angling, trapping (using large minnow traps, baited with salmon roe), and dipnetting. For example, I used a dipnet to sample LGB smolt from the tanker truck used to deliver them from the rearing facility, whereas I angled and trapped LGB parr because they were already living within the river.

Geometric morphometric analyses

Each specimen was anaesthetized and the left lateral side was digitally photographed, ensuring a millimeter ruler was visible to retain a common scale. To ensure adequate visibility of physical features, fish were photographed against a neutral blue background with fins fully extended. Although the initial positioning of a specimen’s body can alter the shape of landmark configurations, the influence of this type of measurement error is likely low as fish were positioned with a straight lateral line. This position is similar to that used by a juvenile that is holding (or swimming in one place; M.R. Gross and J.C. Walters unpublished data), and likely

Reproduced with permission of the copyright owner. Further reproduction prohibited without permission.
represents a body position common among swimming fish. In addition, all specimens were positioned using the same protocol, thus any deformations in shape implied by human error would likely be distributed consistently across all fish types.

Individual landmarks that correspond to important skeletal features (Figure 1A; as in Winans (1984) and Hard et al. (1999)) were digitized from the photographs with tpsDig 2.0 software (Rohlf 2004). All digitizing was performed by myself; the average standard deviation of 10 repeated measurements from each of 10 images was used as an estimate of measurement error due to the digitization process.

Non-shape variation (i.e. position, orientation, scale, and size) is retained in the raw landmark data of a given specimen. To investigate shape variation among specimens that differ in any of these non-shape components, one must first eliminate the influence of non-shape variation. In geometric morphometrics, this is done by creating shape variables through superimposing the landmark configurations for each specimen. The most commonly used superimposition method is the Generalized Least Squares procedure (GLS) and is a type of ‘Procrustes technique’ (Rohlf and Slice 1990).

GLS iteratively translates, rotates and scales the landmark configurations of each specimen until the least-squares fit of all specimens to the mean shape (the ‘consensus configuration’) can no longer be improved upon, thus reducing the locations of landmark coordinates to shape coordinates (Gower 1975; Bookstein 1991). Effectively, this entire procedure removes non-shape variation and differences in the coordinates of corresponding landmarks represent shape differences only. The generalized least-squares superimposition was calculated using IMP Coordgen6f software (Sheets 2003a).

The deformations in shape between a specimen and the consensus configuration consist of uniform and nonuniform components. Uniform components describe shape changes that show linear, uniform deformations throughout the body, whereas nonuniform components describe non-linear deformations that are localized among different regions of the body (Bookstein 1991, 1996). The nonuniform components were described using the thin-plate spline interpolation function (Bookstein 1989, 1991), with nonuniform shape deformations of a specimen from the consensus configuration expressed as a set of shape variables known as partial warps (Bookstein 1991; Rohlf 1993). Twenty partial warps (2p-6; where p is the number of landmarks) were generated for each specimen as six degrees of freedom are lost in the analysis of two-dimensional data (two for translation, one for scale, one for rotation, and two for...
the uniform components; Bookstein 1991). The uniform components were calculated as Bookstein’s $u_1$ and $u_2$ (Bookstein 1996) and appended to the partial-warp matrix.

General description of shape variation: Relative warps analysis (RWA)

To identify and summarize major trends in shape variation among all specimens, I conducted a principal component analysis (PCA) on the partial warps using IMP PCAGen6n (Sheets 2003b). This procedure is termed a ‘relative warps analysis’ (RWA) and summarizes major trends in nonuniform shape variation (Rohlf 1993). As the two uniform components are in different units than the nonuniform partial warps, they must be excluded from the RWA to avoid generating arbitrary results (Rohlf et al. 1996). The first relative warp, or principal component axis, represents the direction of maximal nonuniform shape variation among all specimens (Bookstein 1991; Rohlf 1993), with successive relative warps describing additional proportions of nonuniform shape variation.

After completing an RWA, one needs to determine the number of components that are interpretable. In this part I use a stopping rule called ‘the broken stick model’ (Frontier 1976), which is demonstrably more accurate and less arbitrary than other commonly applied criteria (Jackson 1993).

Quantifying differences among fish types: Multi-response permutation procedure (MRPP)

As the normality assumption inherent in multivariate parametric tests was not met, the multi-response permutation procedure (MRPP) was used on the combined partial warp/uniform component matrix to test for significant differences in shape among the six fish types.

MRPP can produce test analogous to many parametric tests, and remains more versatile when the assumptions of parametric tests are violated (Mielke and Berry 1994). Using Blossom Statistical Package Version 2003.02 (Cade and Richards 2000), all MRPP statistics were computed using a Euclidean distance function (i.e. $v=1$), group weighting of $C_k = n_k/N$ (where: $n_k$ = number of individuals in group $k$ and $N$ = total number of individuals; as described in Mielke (1984)), with no commensuration (none needed as, by definition, Procrustean superimposition standardizes data), for 100 000 permutations. A Euclidean distance function was selected as it has been shown to be more robust and less influenced by outliers relative to a Euclidean squared option (Mielke and Berry 1994). As of yet, there are no post-hoc tests associated with the MRPP, thus I conducted specific comparisons of interest to evaluate among- and within-group variation in shape.
I opted to not utilize the sequential Bonferroni adjustment (Rice 1989), although this technique has become a prevalent approach to reduce the probability of falsely concluding statistical significance. Recently, strong arguments have been made against the arbitrary nature of this method based on mathematical, logical and practical objections (Moran 2003). Hence, I present uncorrected p-values, but remain wary of drawing strong conclusions from p-values that are close (e.g. p≥0.025) to the conventional α=0.05 when presenting large numbers of comparisons.

**Visualizing differences among fish types: Canonical Variates Analysis (CVA)**

A canonical variates analysis (CVA) was performed in IMP CVAGen6j (Sheets 2003c) to graphically identify the patterns of shape variation among fish types. Specifically, this ordination was conducted by independently regressing partial warp/uniform component matrix onto each canonical axis (Rohlf et al. 1996). The resulting deformation grids reflect how the consensus configuration is deformed to create the most extreme form observed in each combination of canonical variates. As with the RWA, the broken-stick model was used to determine how many CVA axes to interpret.

**Non-geometric Morphological Measurements**

To ensure samples remained viable for pathological screening (part of a related study - see: Halpenny 2005 - MSc thesis in prep.), only a sub-sample of the individuals listed in Table 1 were used in further evaluations of phenotypic variation. The fish types that were sampled include: wild smolt (n=42), LGB smolt (n=62), and LGB parr at 3 times (n=43, 87, and 58 respectively). Unfortunately, wild parr could not be included in this section due to a moratorium on sacrificial sampling instituted by the BC Ministry of Water, Land, and Air Protection.

I measured three suites of morphological characters. First, I measured the masses of three internal organs (spleen, liver, and heart) and then the entire body to the nearest 0.01 g. Second, I measured the maximum length of the dorsal, anal, left pectoral and left pelvic fins, parallel to the fin rays to the nearest 0.1 mm. Third, I measured other trunk-based characters not included in the landmark dataset (Figure 1B). These characters included snout-eye length, postorbital-hypural length, head width at the top of the orbit, head depth through the center of the orbit and perpendicular to the longitudinal axis, maximum distance from the snout to the furthest point of the left operculum, length of maxilla, length of premaxilla, and girths perpendicular to the lateral line at: the anterior insertion of the dorsal fin ('dorsal girth'), the anterior insertion of...
the anal fin ('anal girth'), and the narrowest point of the caudal peduncle ('caudal girth'). During sampling, additional length and mass measurements were collected from all of the fish types except wild parr – sample sizes are included in Results.

Quantifying differences among fish types: Multi-response permutation procedure (MRPP)

Although analyzing these non-geometric morphological variables via a MANCOVA would account for the covariation of these variables with body size, I analyzed these variables using nonparametric methods as these data did not meet parametric multivariate assumptions. To my knowledge, there is no nonparametric version of a MANCOVA for unbalanced designs. Therefore, to account for the effects of size I regressed each untransformed morphological variable against a representative body size variable: organ mass variables were regressed against body mass, whereas trunk measurements and fin lengths were regressed against centroid size. Centroid size is the square root of the summed squared distances of all landmarks (landmarks seen in Figure 1) to the centroid (Bookstein 1986, 1991) and corresponds to a summation of the dimensions of a specimen in many directions simultaneously. Fish types were then compared using the residuals from these regressions as the dependent variables for MRPPs. MRPPs were conducted using parameters similar to those used in the geometric morphometric analyses (v=1; Ck= n_k/N; 100 000 permutations), with Euclidean commensuration used to standardize the dependent variables. Euclidean commensuration is consistent with the parameters v=1 and Ck= n_k/N and prevents any of the response variables from being overwhelmed by variables recorded in different scales and/or units (Mielke and Berry 1999). Again, specific MRPP comparisons of interest are presented to evaluate among- and within-group variation for these suites of morphological variables.

Size Contrasts

In addition to shape contrasts, I also analyzed three types of size data (fork length, mass, and centroid size) to understand the relative size differences among LGB and wild juvenile life-histories. As the data were not normally distributed, I conducted specific non-parametric comparisons of interest to evaluate size differences among the six sampled fish types. When I compared any single size variable for two populations I used a Mann-Whitney U test; when I compared three populations I used a Kruskal-Wallis ANOVA.
Results

Geometric morphometric analyses

Measurement error averaged 0.18mm (range: 0.08-0.34mm) for all of the landmarks. Hence, with the large amount of size and shape variation observed, digitizing error was insignificant.

General description of shape variation: Relative warps analysis (RWA)

A broken-stick model suggested only the first two relative warps were interpretable, explaining 28.67% and 16.95% of the total shape variation, respectively (Figure 2A).

Positive RW1 scores represent a ‘bowing’ along the longitudinal axis, with the head and tail moving downward and the middle region moving upwards (Figure 2B). As the centroid of each fish type is near zero in terms of RW1 (Figure 2A), there is no strong relationship between fish type and the presence of longitudinal bowing. Thus, longitudinal bowing comprises the majority of shape variation among all juvenile steelhead in this study, regardless of life stage (parr or smolt) or origin (wild or LGB).

Positive RW2 scores represent anterior movement of the snout tip, vertical widening of the anterior, upwards and posterior movement of the dorsal surface, and anterior movement of the caudal peduncle (Figure 2C). Therefore, positive values along RW2 can be thought to represent a reduction in streamlining. For RW2 the centroid of LGB smolt is positive indicating LGB smolts tend to be more robust, whereas the centroid for wild parr is negative along RW2, indicating wild parr tend to be more streamlined. Conversely, the centroid for wild smolt and all the three LGB parr groups remain near zero, indicating on average, individuals from these groups are intermediate in terms of RW2-related streamlining.

Quantifying differences among fish types: Multi-response permutation procedure (MRPP)

MRPP demonstrated specific trends in shape variation within and among fish types. Overall, there is a significant shape difference between LGB and wild origin fish, regardless of life-history and/or ontogenetic stage (Table 2). Sexual dimorphism was not observed within any fish type (Table 2). Sexual dimorphism was not examined within wild parr to avoid lethal sampling.

Shape differences were also demonstrated within and among life stages. Within the smolt life stage, I found that LGB and wild fish differ significantly in shape (Table 2). Among the wild fish, significant morphometric differences exist between smolt and parr – thereby
confirming that different body shapes are expressed in river residency versus oceanic migration. Also, I found significant shape differences among the three LGB parr samplings (Table 2), confirming that LGB parr shape changes through time. Wild parr were also significantly different in shape from the late LGB parr (Table 2), demonstrating that even after a few months of river residency, LGB parr do not have a shape similar to wild parr.

Visualizing differences among fish types: Canonical Variates Analysis (CVA)

According to a broken-stick model, the first two canonical variates were interpretable, explaining 50.29% and 28.52% of the shape variation among populations, respectively (Figure 3A).

In terms of discriminating among populations, the first two CVs do not completely separate each of the six populations, although the centroids for each fish type (seen as enlarged symbols) demonstrate clear patterns in response to both axes (Figure 3A). The wild fish (parr and smolt) and late LGB parr tend to respond negatively in terms of CV1, whereas the other three LGB populations tend to respond positively in terms of CV1 (Figure 3A). Also, the smolts (wild and LGB) tend to respond negatively in terms of CV2, whereas the other four populations respond more positively in terms of CV2 (Figure 3A). Therefore, the CVA demonstrates there are two distinct body shape trends among the sample populations. The most significant axis, CV1, separates production history (wild vs. LGB), and CV2 demonstrates a life-history effect (i.e. smolts vs. parr). In addition, the centroids of the three LGB parr groups approached the centroid of the wild parr as the season progressed, indicating that over time the LGB parr are approaching a shape that is closer to that of wild juveniles.

The patterns of ordination among the fish types correspond to complex morphological patterns (Figure 3B). Simultaneous positive responses on both CV1 and CV2 — associated with a vertical compression in the anterior landmarks and a vertical expansion in the posterior landmarks (Figure 3B) — tend to correspond to the early and middle LGB parr (Figure 3A). A positive response on CV1 and simultaneous negative response on CV2 — associated with a vertical compression in the anterior landmarks and vertical expansion in the central landmarks (Figure 3B) — tend to correspond to LGB smolts (Figure 3A). A negative response on CV1 and simultaneous positive response on CV2 — associated with a vertical expansion of the anterior region and vertical compression in the central landmarks (Figure 3B) — tend to correspond to late LGB parr (Figure 3A). Simultaneous negative responses on both CV1 and CV2 — associated
with a vertical expansion of the anterior region and pronounced narrowing of the caudal peduncle (Figure 3B) – tend to correspond to wild smolts and wild parr (Figure 3A).

Non-geometric Morphological Measurements

All regressions of the non-geometric variables against representative size variables were significant (Table 3): centroid size explained a small amount of the variation in each trunk and fin variable, whereas mass explained a moderate proportion of the variance in each organ mass variable (Table 3).

Quantifying differences among fish types: Multi-response permutation procedure (MRPP)

There was a significant difference among non-geometric morphological measurements between LGB and wild origin fish, regardless of LGB life stage (parr or smolt: Table 4). For spleen mass, liver mass, heart mass, and dorsal fin length characters, on average, wild fish demonstrated larger residuals than did LGB fish. For the remaining characters, LGB fish tend to demonstrate larger residuals that the wild fish.

Among smolt, significant differences in non-geometric measurements existed between wild and LGB smolt (Table 4). On average, wild smolt had significantly larger organ mass, pectoral fin length and dorsal fin length residuals than LGB smolt (Table 5). For the remaining characters, LGB smolt tended to have larger residuals than wild smolt (Table 5).

Within LGB smolt, strong sexual dimorphism existed among non-geometric measurements (Table 4). On average, female LGB smolt demonstrated larger dorsal fin length and organ mass residuals than males; for all other characters, males demonstrated larger average residuals (Table 5). In contrast, significant sexual dimorphism was not observed in wild smolt (Table 4). Although average residuals were not identical for all variables between the sexes (Table 5), overall, male and female wild smolt were similar regarding the non-geometric variables measured.

Morphological divergence was also evident among the LGB parr samplings. Strong differences in non-geometric measurements were observed among the three within-season samples of LGB parr (Table 4). Also, significant differences in non-geometric measurements were observed for each possible pairing of the three LGB parr samplings (Table 4). For most characters (all except anal girth, liver mass and heart mass), the late LGB parr tended to exhibit larger residuals than the other two samplings (Table 5).
Sexual dimorphism was only observed in one of the LGB parr samplings for non-geometric measurements (Table 4). To determine if this sexual dimorphism was due to precocial maturity within the males, I reran the MRPP with precociously maturing males excluded (i.e. remove gonad factor 2 and 3 males). With precociously maturing males removed from the dataset, no significant differences remained between male and female middle LGB parr (Table 4). This suggests that within this particular sampling of the LGB parr, precocial maturity appears to influence phenotype more than does sex alone (i.e. undeveloped males and females are more similar than undeveloped and precocious males). These results are interesting as although all three samplings of LGB parr demonstrate similar rates of precocial maturity (see Part 2), only the middle LGB parr group exhibits evidence of sexual dimorphism.

LGB smolt and LGB parr also differed in non-geometric measurements at the time of river-residency (Table 4). Early LGB parr exhibited greater residuals than LGB smolt for 12 non-geometric characters (Table 4).

Size Contrasts

LGB smolt were significantly larger in mass \((M-W Z=10.67, p<0.0001)\), fork length \((M-W Z=16.44, p<0.0001)\), and centroid size \((M-W Z=4.96, p<0.0001)\) than wild smolt (Table 6). I compared wild parr to early LGB parr to determine the size differences that exist between the two groups at the time of LGB parr initial residency. Compared to wild parr, early LGB parr were significantly larger in mass \((M-W Z=3.01, p=0.0026)\), fork length \((M-W Z=3.23, p=0.0013)\), and centroid size \((M-W Z=5.73, p<0.0001; \text{Table 6})\). If one compares the size of LGB parr to each separate wild parr age class \((1+ \text{ and } 2+)\), significant differences in size are still observed – for simplicity I only present the overall wild parr vs. LGB parr size comparisons.

I also compared the three samplings of LGB parr to see how rapidly LGB parr change in size after initial residency. The three samplings of LGB parr were significantly different in mass \((K-W H=41.58, p<0.0001)\), fork length \((K-W H=56.87, p<0.0001)\) and centroid size \((K-W H=35.43, p<0.0001; \text{Table 6})\) – late LGB parr were larger in mass and length than early and middle LGB parr.

Discussion

This study demonstrates substantial phenotypic and life-history divergence between wild juveniles and the juveniles produced by a conservation hatchery. These differences are likely due to the developmental experience in the hatchery and wild, as well as to different life-history
decisions made as a consequence of this experience. I have also found that hatchery-produced juveniles, once released into the wild, become increasingly more like wild juveniles, demonstrating a continuing phenotypic response to changes in environmental conditions.

The canonical variates analysis demonstrated that wild and LGB-produced juveniles demonstrated two distinct trends in shape: 1) canonical variate 1 represents a continuum in shape from wild to hatchery. Along this continuum, LGB smolt and early LGB parr were the furthest from the wild type, with middle and late LGB parr closer to the wild populations; 2) canonical variate 2 demonstrates a separation of life-history stages, with a separation between parr and smolt. The combination of these two trends resulted in distinct phenotypic trends for each wild and LGB fish type sampled.

Smolts produced by the LGB program tend to be more robust and less streamlined when compared to the wild type, whereas the early and middle LGB parr demonstrate greater body depth only in the caudal region. At the time of release, the visceral cavity of LGB smolts contains large fat deposits, whereas the visceral cavity of LGB parr tend to contain considerably less visible fat – decreasing from the early to late LGB parr samplings. This ‘thinning’ of the central region may correspond to a reduction in fat levels during continued and prolonged river residency. Containing large fat deposits at time of release may increase LGB progeny survival while they adjust to foraging in the natural environment and while their body shape responds to the new environmental conditions.

The convergence to the wild parr phenotype is extremely rapid – if current rates of morphological change continue, the centroids of wild and LGB parr could show similar positions on CV1 within an additional two months. Also, the centroid of LGB parr becomes positioned more positively in terms of CV2 over time – indicating that LGB parr become more similar to wild parr than to wild smolt. In addition, the location of the early LGB parr centroid differs from that of LGB smolt, indicating that LGB parr exhibit a shape that is unlike that of ocean-migrating LGB juveniles at the time of release. This observed phenotypic convergence could be due to phenotypic plasticity, selective mortality, or both. If selective mortality is the case, the LGB parr population is expected to change from high variance in shape to lower variance. Moreover, the range of phenotypes expressed by the late LGB parr should be a subset of those expressed by the early sampling. However, Figure 3A demonstrates that this pattern does not exist. Therefore, this within-season change in shape is most likely a consequence of phenotypic plasticity within the LGB parr.
Although graphically the LGB and wild smolt are positioned similarly in terms of CV2, these similarities do not appear to result in similar trends in body shape. These two groups remain obviously different in body shape although they respond similarly in terms of one canonical variate. Wild smolts have a very fusiform body shape; a shape that is commonly associated with fish species that exhibit efficient and rapid long-distance cruising (Webb 1984a, b). The expression of this fusiform shape by wild smolt is logical in life-history terms as salmonid smolts undertake substantial oceanic migrations. The absence of a fusiform shape in LGB smolt is likely to make these fish less efficient in oceanic migrations. By using an acoustic array, LGB smolt were suggested to experience early oceanic survival rates that were considerably lower than wild smolt (Welch et al. 2004). Reduced streamlining relative to the wild type may be partially responsible for the reduced early oceanic survival of hatchery progeny.

Although the LGB program appears successful at accelerating skeletal and muscular growth relative to wild fish, LGB fish remain smaller than wild conspecifics for some morphological characters. For a given size, wild fish tend to have larger than predicted internal organs than LGB fish, as well as have longer dorsal fins. The trend of longer dorsal fins in wild fish is likely a result of damage to the dorsal fin of LGB fish due to fin erosion and agonistic interactions – both frequently observed in hatchery settings (Latremouille 2003). Conversely, larger dorsal fins have been observed in salmonids reared in faster water (Pakkasmaa and Piironen 2001), thus the discrepancy between wild and LGB dorsal fin size may be a result of phenotypic plasticity related to rearing-water velocities. As ontogenetic relative growth follows the law of allometry (Huxley 1932), the smaller than predicted organ size in LGB fish is likely a consequence of: 1) greater LGB fish body size relative to wild fish; and 2) allometric scaling of internal organs to an exponent of <1.

The LGB and wild juveniles also differed in sexual dimorphism. Within the LGB smolt, body shapes are similar between the sexes, but dimorphism is observed for the non-geometric variables. Female LGB smolt had larger internal organ masses and dorsal fin lengths, relative to males. Differential allocation of energy to internal organ development may be due to differences in reproductive roles among the sexes (e.g. Casselman and Schulte-Hostede 2004). Egg production is energetically expensive and larger livers are associated with higher production of yolk proteins and higher energy content (Lambert and Dutil 1997; Dahle et al. 2003; Guijarro et al. 2003). The lack of sexual dimorphism among wild smolt is not surprising, as it is likely that during initial stages of the oceanic migration both male and female smolt experience similar
selective pressures (i.e. foraging and predation). Relative to wild conspecifics, sexual
dimorphism develops prematurely in LGB smolt. This early expression of sexual dimorphism is
likely another phenotypic consequence of the rearing environment.

The previous section demonstrated that a substantial proportion of male LGB parr exhibit
signs of accelerated gonadal development (approximately 42%). It also noted that a high
proportion of ripe precocious male LGB parr from pre-2004 LGB releases had partially empty
gonads, suggesting that they may have already attempted to spawn. Integrating the precocious
maturation and morphometric results suggests that precocious maturation does not contribute to
sexual dimorphism within the LGB parr. Although precociously maturing individuals are
nearing spawning condition they are apparently not developing any secondary sexual
characteristics, such as a kype. Small, precociously maturing male salmonids tend to show little
or no development of secondary sexual characteristics relative to the larger anadromous forms
(Morton 1965). As these precociously maturing LGB parr are of small size and not visibly male,
it is likely that these precocious individuals will adopt a ‘sneaker’ reproductive tactic in breeding
attempts (Gross 1996).

The length and weight advantage of the LGB smolts relative to wild smolts does not
confer an apparent survival advantage in the marine environment. Marine survival has averaged
1.95% and 0.24% for the 2001 and 2002 releases of LGB smolt, respectively, whereas
corresponding cohorts of wild smolt experienced survival rates of 3.23% and 0.43% (McCubbing
and Ward 2004). At the Keogh River, smolt-to-adult survival is positively correlated with smolt
size (Ward and Slaney 1988; Ward et al. 1989): a 40% increase in smolt length increased marine
survival by a factor of 8 (Ward et al. 1989). As marine survival of LGB progeny has been
considerably lower than that of wild smolt, the observed size advantage is obviously not
providing the LGB fish with elevated survival relative to wild conspecifics. Conversely, if wild
and LGB smolt were released at a similar body size, the negative effects of phenotypic change
on survival would likely appear more extreme.

Although LGB parr are apparently converging with wild parr shape through time, LGB
parr are considerably larger in size than wild parr. Although the two groups were sampled
simultaneously, late LGB parr demonstrate a relatively large size advantage over wild parr.
Larger size tends to confer advantages with respect to dominance (MacPhee 1961), thereby
influencing territory acquisition and size, as well as resource competition (Abbott et al. 1985;
Keeley 2000). Thus, as LGB parr move upstream from the release site they are likely displacing

Reproduced with permission of the copyright owner. Further reproduction prohibited without permission.
wild parr from optimal habitats, thereby increasing resource competition. The potential influences of LGB parr on wild parr are discussed in Part 2.

Inadvertent artificial selection, resulting from the unnatural environmental conditions of the hatchery environment, will shift the phenotype off of the adaptive peak of the wild phenotype, thereby reducing the performance of the hatchery-produced fish under conditions experienced in the natural environment (Goodman 2005). The reduced survival of LGB smolt relative to wild smolt may well be due to the many striking morphological differences found between LGB and wild fish. Alternatively, the reduced survival of LGB smolt may be due to other factors, such as the naivety of hatchery progeny when subjected to the wild environment and any potential predators.

Other factors may be contributing to the reduced survival of LGB progeny, but these are not partitioned out in this study. For example, I did not investigate the potential impacts of the non-volitional release strategy used by the LGB program on subsequent hatchery smolt survival. Although volitionally released salmon are thought not to experience greater survival than force-released conspecifics (Wagner 1968; Evenson and Ewing 1992; Appleby et al. 2004), my description of phenotypic variation within LGB fish will likely capture phenotypic divergence between wild and hatchery juveniles that arise due to differences in “readiness” to migrate to the ocean.

Although salmonid morphology is partially under genetic control (Riddell et al. 1981; Taylor and McPhail 1985; Fleming and Einum 1997; Hard et al. 1999; Hard et al. 2000), unless interspecific hybridization is common, I believe that genetic divergence had only a minor role in contributing to the phenotypic change demonstrated in this study. The BC LGB program employs captive broodstock for a single generation to minimize artificial selection and genetic divergence in captivity. It has been demonstrated that environmental influences on salmonid growth can be stronger than genetic effects alone (Nicicza 1995; von Cramon-Taubadel et al. 2005). Second, water-velocity changes in the rearing environment induces rapid phenotypic change in juvenile salmonids (Pakkasmaa and Piironen 2001). My study supports these earlier results: when subjected to natural conditions, the hatchery progeny exhibit a plastic response and their phenotype changes to become more like the wild type. Hence, to increase the post-release survival of hatchery progeny, conservation hatcheries should also be concerned with phenotype conservation – altered rearing conditions will result in phenotypically altered progeny.

To my knowledge, no other study has so comprehensively evaluated phenotypic variation between hatchery and wild juvenile life-history tactics and ontogenetic stages. Furthermore,
earlier studies were not as relevant to a single-generation captive broodstock program as their comparisons involved a hatchery stock that had: 1) originated from a different stream than the wild stock used in their contrasts; 2) spent multiple generations in captivity, and thus, were subjected to multiple generations of relaxed selection (e.g. the hatchery increases fertilization success and fish survival); or 3) a combination of different origin stream and multiple generations in captivity. Hence, my characterization of morphometric variation between wild and hatchery salmonid juveniles is not only the most comprehensive to date, it also has direct implications for understanding how sophisticated conservation culture programs inadvertently influence the phenotype of their product.

Here, I return to the three questions raised in the introduction:

1) How much will the conservation program change the phenotype, and what are the ramifications of these changes? The conservation program caused dramatic phenotypic change— not only in shape, but also for a variety of other internal and external phenotypic characters. The observed phenotypic change may represent a major contributing factor to the reduced marine survival experienced by hatchery fish relative to wild conspecifics.

2) Will the phenotypic change decrease the survival and reproductive success, or impact the wild population, to such a degree that the conservation program is a failure? In addition to the potential for LGB fish to negatively impact wild fish growth and survival through density-dependent effects (Part 2), the reduced marine survival of LGB progeny is likely a consequence of culture-induced phenotypic divergence from the wild type. This phenotypic divergence, and associated reduced survival, may greatly reduce the ability of this conservation program to recover the wild population. At this point, the success or failure of the conservation program is undetermined.

3) What changes can be implemented to mitigate the negatives and enhance the positives? A variety of changes could be made to the conservation program to maximize its benefit to wild population recovery. Although the effectiveness of conservation actions were evaluated in two parts, phenotypic, life-history, and demographic responses to conservation actions do not occur entirely independent of one another. Therefore, in Part 4, I synthesize the demographic, ecological, genetic, life-history and phenotypic issues raised in Parts 2 and 3, and suggest
methods in which to maximize the benefits and minimize the costs to wild population recovery associated with conservation programs.
Table 1. Fish types differentiated by life stage and origin (wild or LGB), and sampling period; including capture methods and dates, and collection sizes used for morphometric contrasts.

<table>
<thead>
<tr>
<th>Sample group</th>
<th>Capture Method</th>
<th>2004 Capture Dates</th>
<th>N</th>
</tr>
</thead>
<tbody>
<tr>
<td>Smolt</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Wild</td>
<td>Trapping at weir</td>
<td>May 12-19</td>
<td>46</td>
</tr>
<tr>
<td>LGB</td>
<td>Dipnet below weir</td>
<td>May 17-26</td>
<td>80</td>
</tr>
<tr>
<td>Parr</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>late Wild</td>
<td>Angling &amp; trapping in river</td>
<td>August 4-17</td>
<td>42</td>
</tr>
<tr>
<td>early LGB</td>
<td>Angling &amp; trapping in river</td>
<td>June 15-20</td>
<td>89</td>
</tr>
<tr>
<td>middle LGB</td>
<td>Angling &amp; trapping in river</td>
<td>July 7-21</td>
<td>52</td>
</tr>
<tr>
<td>late LGB</td>
<td>Angling &amp; trapping in river</td>
<td>August 4-13</td>
<td>77</td>
</tr>
</tbody>
</table>

Reproduced with permission of the copyright owner. Further reproduction prohibited without permission.
Table 2. Multi-response permutation procedure results for comparisons of shape variables (partial warps + uniform components).

<table>
<thead>
<tr>
<th>Comparison of Interest</th>
<th>N</th>
<th>MRPP Δ</th>
<th>P-value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Origin: LGB vs. wild</td>
<td>298.88</td>
<td>0.04575</td>
<td>0.00001</td>
</tr>
<tr>
<td>Smolts: LGB vs. wild</td>
<td>80.46</td>
<td>0.03665</td>
<td>0.00001</td>
</tr>
<tr>
<td>LGB smolt: male vs. female</td>
<td>34.29</td>
<td>0.03569</td>
<td>0.08000</td>
</tr>
<tr>
<td>Wild smolt: male vs. female</td>
<td>20.23</td>
<td>0.03707</td>
<td>0.23000</td>
</tr>
<tr>
<td>Early LGB parr: male vs. female</td>
<td>32.13</td>
<td>0.03337</td>
<td>0.20000</td>
</tr>
<tr>
<td>Middle LGB parr: male vs. female</td>
<td>61.27</td>
<td>0.04623</td>
<td>0.26000</td>
</tr>
<tr>
<td>Late LGB parr: male vs. female</td>
<td>41.18</td>
<td>0.05033</td>
<td>0.55000</td>
</tr>
<tr>
<td>Wild juveniles: smolt vs. parr</td>
<td>46.42</td>
<td>0.04027</td>
<td>0.00001</td>
</tr>
<tr>
<td>LGB juveniles: smolt vs. early parr</td>
<td>80.52</td>
<td>0.03547</td>
<td>0.00001</td>
</tr>
<tr>
<td>LGB juveniles: smolt vs. late parr</td>
<td>80.77</td>
<td>0.04405</td>
<td>0.00001</td>
</tr>
<tr>
<td>Within season LGB parr shape differences: early, middle and late</td>
<td>89.52.77</td>
<td>0.04534</td>
<td>0.00001</td>
</tr>
<tr>
<td>Late LGB parr vs. wild parr</td>
<td>77.42</td>
<td>0.04909</td>
<td>0.00001</td>
</tr>
</tbody>
</table>
Table 3. Linear regression statistics and parameters for 17 non-geometric variables (for each variable N=292: organ mass variables were regressed against body mass, all others were regressed against centroid size).

<table>
<thead>
<tr>
<th>Non-geometric variable</th>
<th>$r^2$</th>
<th>p-value</th>
<th>F</th>
<th>Intercept</th>
<th>Slope</th>
</tr>
</thead>
<tbody>
<tr>
<td>postorb-hypural length</td>
<td>0.1666</td>
<td>&lt;0.0001</td>
<td>57.969</td>
<td>10.945</td>
<td>0.200</td>
</tr>
<tr>
<td>snout-eye length</td>
<td>0.0981</td>
<td>&lt;0.0001</td>
<td>31.547</td>
<td>0.675</td>
<td>0.010</td>
</tr>
<tr>
<td>head width</td>
<td>0.1271</td>
<td>&lt;0.0001</td>
<td>42.240</td>
<td>0.786</td>
<td>0.016</td>
</tr>
<tr>
<td>head depth</td>
<td>0.1140</td>
<td>&lt;0.0001</td>
<td>37.303</td>
<td>1.531</td>
<td>0.020</td>
</tr>
<tr>
<td>left operculum</td>
<td>0.1240</td>
<td>&lt;0.0001</td>
<td>41.042</td>
<td>2.932</td>
<td>0.038</td>
</tr>
<tr>
<td>maxillary length</td>
<td>0.0945</td>
<td>&lt;0.0001</td>
<td>30.265</td>
<td>1.187</td>
<td>0.0144</td>
</tr>
<tr>
<td>premaxilla length</td>
<td>0.0773</td>
<td>&lt;0.0001</td>
<td>24.294</td>
<td>0.408</td>
<td>0.004</td>
</tr>
<tr>
<td>anal fin length</td>
<td>0.0878</td>
<td>&lt;0.0001</td>
<td>27.925</td>
<td>1.451</td>
<td>0.0185</td>
</tr>
<tr>
<td>dorsal fin length</td>
<td>0.0528</td>
<td>&lt;0.0001</td>
<td>16.161</td>
<td>1.305</td>
<td>0.020</td>
</tr>
<tr>
<td>left pelvic fin length</td>
<td>0.0790</td>
<td>&lt;0.0001</td>
<td>24.886</td>
<td>1.399</td>
<td>0.016</td>
</tr>
<tr>
<td>left pectoral fin length</td>
<td>0.0471</td>
<td>0.0002</td>
<td>14.330</td>
<td>1.732</td>
<td>0.017</td>
</tr>
<tr>
<td>dorsal girth</td>
<td>0.1158</td>
<td>&lt;0.0001</td>
<td>37.988</td>
<td>6.114</td>
<td>0.107</td>
</tr>
<tr>
<td>anal girth</td>
<td>0.0976</td>
<td>&lt;0.0001</td>
<td>31.350</td>
<td>5.076</td>
<td>0.081</td>
</tr>
<tr>
<td>caudal girth</td>
<td>0.1159</td>
<td>&lt;0.0001</td>
<td>38.024</td>
<td>3.119</td>
<td>0.046</td>
</tr>
<tr>
<td>spleen mass</td>
<td>0.4224</td>
<td>&lt;0.0001</td>
<td>212.064</td>
<td>-0.038</td>
<td>0.002</td>
</tr>
<tr>
<td>heart mass</td>
<td>0.7064</td>
<td>&lt;0.0001</td>
<td>697.644</td>
<td>0.0139</td>
<td>0.001</td>
</tr>
<tr>
<td>liver mass</td>
<td>0.5490</td>
<td>&lt;0.0001</td>
<td>352.965</td>
<td>0.150</td>
<td>0.007</td>
</tr>
</tbody>
</table>
Table 4. Multi-response permutation procedure results for comparisons of non-geometric morphological variables.

<table>
<thead>
<tr>
<th>Comparison</th>
<th>N</th>
<th>MRPP Δ</th>
<th>P-value</th>
</tr>
</thead>
<tbody>
<tr>
<td>LGB vs. wild</td>
<td>250, 42</td>
<td>4.50289</td>
<td>0.00001</td>
</tr>
<tr>
<td>Smolts: LGB vs. wild</td>
<td>62, 42</td>
<td>4.02088</td>
<td>0.00001</td>
</tr>
<tr>
<td>LGB smolts: Male vs. female</td>
<td>32, 30</td>
<td>4.78391</td>
<td>0.01255</td>
</tr>
<tr>
<td>Wild smolts: male vs. female</td>
<td>18, 24</td>
<td>4.90933</td>
<td>0.32759</td>
</tr>
<tr>
<td>LGB parr: early, middle, late</td>
<td>43, 87, 58</td>
<td>4.09164</td>
<td>0.00001</td>
</tr>
<tr>
<td>LGB parr: early vs. middle</td>
<td>43, 87</td>
<td>4.37048</td>
<td>0.00001</td>
</tr>
<tr>
<td>LGB parr: early vs. late</td>
<td>43, 58</td>
<td>4.63302</td>
<td>0.00001</td>
</tr>
<tr>
<td>LGB parr: middle vs. late</td>
<td>87, 58</td>
<td>4.04295</td>
<td>0.00001</td>
</tr>
<tr>
<td>Early LGB parr: Male vs. female</td>
<td>31, 12</td>
<td>4.79050</td>
<td>0.60257</td>
</tr>
<tr>
<td>Middle LGB parr: Male vs. female</td>
<td>61, 26</td>
<td>4.59047</td>
<td>0.00056</td>
</tr>
<tr>
<td>Middle LGB parr: Male vs. female, with Gonad Factor 3 males removed</td>
<td>50, 26</td>
<td>4.67907</td>
<td>0.00623</td>
</tr>
<tr>
<td>Middle LGB parr: male vs. female with Gonad Factor 2 and 3 males removed</td>
<td>40, 26</td>
<td>4.83201</td>
<td>0.08173</td>
</tr>
<tr>
<td>Late LGB parr: Male vs. female</td>
<td>40, 18</td>
<td>4.88094</td>
<td>0.87137</td>
</tr>
<tr>
<td>Early LGB parr vs. LGB smolt</td>
<td>43, 62</td>
<td>4.79393</td>
<td>0.00001</td>
</tr>
<tr>
<td>Late LGB parr vs. LGB smolt</td>
<td>58, 62</td>
<td>3.78246</td>
<td>0.00001</td>
</tr>
</tbody>
</table>

Reproduced with permission of the copyright owner. Further reproduction prohibited without permission.
Table 5. Fish-type specific mean residuals from independent regressions of each non-geometric variable on body size (regression details in Methods; variable abbreviations as in Figure 1B)

| Fish Type | Sex | N | PH | SE | HW | HD | OP | MA | PM | AF | DF | PV | PC | DG | AG | CG | SP | HE | LV |
|-----------|-----|---|----|----|----|----|----|----|----|----|----|----|----|----|----|----|----|----|----|----|
| Early LGB | Both | 43 | 0.79 | 0.06 | 0.07 | 0.12 | 0.06 | 0.05 | 0.07 | -0.15 | 0.03 | -0.09 | 0.86 | 0.78 | 0.36 | -0.05 | -0.05 | -0.33 |
|           | M   | 31 | 0.83 | 0.08 | 0.08 | 0.15 | 0.08 | 0.11 | -0.15 | 0.05 | -0.09 | 0.92 | 0.79 | 0.40 | -0.05 | -0.05 | -0.33 |
|           | F   | 12 | 0.68 | 0.03 | 0.02 | 0.03 | -0.01 | 0.03 | -0.05 | -0.15 | -0.02 | -0.11 | 0.72 | 0.73 | 0.25 | -0.06 | -0.05 | -0.32 |
| Middle LGB | Both | 87 | -1.33 | -0.07 | -0.12 | -0.15 | -0.08 | -0.03 | -0.07 | -0.05 | -0.06 | -0.08 | -0.92 | -0.76 | -0.42 | -0.01 | -0.02 | -0.09 |
|           | M   | 61 | -0.87 | -0.05 | -0.07 | -0.09 | -0.17 | -0.02 | -0.01 | -0.02 | -0.04 | -0.68 | -0.54 | -0.31 | -0.02 | -0.02 | -0.12 |
|           | F   | 26 | -2.39 | -0.14 | -0.22 | -0.28 | -0.44 | -0.05 | -0.21 | -0.12 | -0.17 | -1.49 | -1.27 | -0.68 | 0.00 | -0.01 | -0.02 |
| Late LGB | Both | 58 | 1.52 | 0.12 | 0.12 | 0.27 | 0.54 | 0.23 | 0.04 | 0.31 | 0.37 | 0.31 | 0.42 | 0.92 | 0.77 | 0.46 | 0.01 | -0.04 |
|           | M   | 40 | 1.57 | 0.12 | 0.13 | 0.29 | 0.57 | 0.25 | 0.04 | 0.32 | 0.42 | 0.34 | 0.46 | 0.95 | 0.79 | 0.48 | 0.00 | -0.04 |
|           | F   | 18 | 1.40 | 0.10 | 0.10 | 0.23 | 0.48 | 0.19 | 0.03 | 0.28 | 0.26 | 0.24 | 0.34 | 0.87 | 0.72 | 0.41 | 0.02 | -0.05 |
| LGB smolt | Both | 62 | 1.29 | 0.04 | 0.11 | 0.05 | 0.04 | 0.03 | 0.01 | -0.05 | -0.26 | -0.10 | -0.18 | 0.92 | 0.60 | 0.31 | -0.19 | -0.04 |
|           | M   | 32 | 1.63 | 0.07 | 0.15 | 0.12 | 0.11 | 0.04 | 0.02 | -0.01 | -0.32 | -0.07 | -0.08 | 1.21 | 1.00 | 0.39 | -0.10 | -0.04 |
|           | F   | 18 | 0.94 | 0.01 | 0.07 | -0.03 | -0.05 | -0.11 | -0.04 | -0.09 | -0.13 | -0.29 | 0.64 | 0.36 | 0.22 | -0.08 | -0.03 | -0.24 |
| Wild smolt | Both | 42 | -2.06 | -0.13 | -0.16 | -0.22 | -0.40 | -0.17 | -0.06 | -0.27 | 0.13 | -0.19 | -0.06 | -1.60 | -1.30 | -0.59 | -0.03 | 0.00 | 0.29 |
|           | M   | 18 | -2.46 | -0.15 | -0.22 | -0.27 | -0.46 | -0.18 | -0.07 | -0.25 | 0.10 | -0.20 | -0.09 | -1.76 | -1.42 | -0.73 | -0.02 | -0.01 | 0.29 |
|           | F   | 24 | -1.76 | -0.11 | -0.12 | -0.18 | -0.36 | -0.15 | -0.06 | -0.29 | 0.16 | -0.18 | -0.03 | -1.48 | -1.21 | -0.49 | -0.03 | 0.00 | 0.30 |
Table 6. Average mass, length, and centroid size for the wild and LGB origin life-histories.

<table>
<thead>
<tr>
<th>Fish Type</th>
<th>Mean Mass (g) (± SE, N)</th>
<th>Mean Length (cm) (± SE, N)</th>
<th>Mean Centroid Size (cm) (± SE, N)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Smolt</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>wild</td>
<td>53.70 (± 3.99, 105)</td>
<td>17.01 (±0.09, 362)</td>
<td>21.46 (±0.31, 46)</td>
</tr>
<tr>
<td>LGB</td>
<td>81.79 (±1.27, 301)</td>
<td>19.94 (±0.11, 301)</td>
<td>24.07 (±0.20, 62)</td>
</tr>
<tr>
<td>Parr</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>wild</td>
<td>43.95 (±5.13, 37)</td>
<td>15.81 (±0.51, 41)</td>
<td>18.24 (±0.71, 42)</td>
</tr>
<tr>
<td>early LGB</td>
<td>57.82 (±1.57, 446)</td>
<td>17.39 (±0.16, 442)</td>
<td>23.61 (±0.46, 52)</td>
</tr>
<tr>
<td>middle LGB</td>
<td>55.86 (±1.88, 284)</td>
<td>18.05 (±0.19, 284)</td>
<td>20.20 (±0.40, 89)</td>
</tr>
<tr>
<td>late LGB</td>
<td>84.25 (±4.31, 79)</td>
<td>20.58 (±0.34, 79)</td>
<td>23.38 (±0.44, 77)</td>
</tr>
</tbody>
</table>

1 Weighted mean and pooled variance used as during each LGB release a different number of LGB smolt were released and sampled.
Figure 1. A) Landmark locations for geometric morphometric analyses from the left lateral view of wild and LGB fish: 1) anterior tip of snout at upper jaw, 2) most posterior aspect of neurocranium, 3) origin of dorsal fin, 4) insertion of dorsal fin, 5) origin of adipose fin, 6) anterior attachment of dorsal membrane of caudal fin, 7) centre of tail fork, 8) anterior attachment of ventral membrane of caudal fin, 9) insertion of anal fin, 10) origin of anal fin, 11) origin of pelvic fin, 12) origin of pectoral fin, and 13) posterior-most point of maxillary. For landmarks 12 and 13, landmarks are placed at their respective positions at the closest point to the body on a line perpendicular to the horizontal axis of the specimen. [Landmark locations and image adapted from: Winans (1984); Hard et al. (1999)].

B) Morphological characters measured for non-geometric analyses: maximum length of the dorsal fin (DF), anal fin (AF), left pectoral fin (PC) and left pelvic fin (PV), parallel to the fin rays; SE= snout-eye length; PH = postorbital-hypural length; HD = head depth through the center of the orbit and perpendicular to the longitudinal axis; OP= maximum distance from the snout to the furthest point of the left operculum; MA= length of maxilla; PM = length of premaxilla; and girths perpendicular to the lateral line at: the anterior insertion of the dorsal fin (DG = dorsal girth), the anterior insertion of the anal fin (AG = anal girth), and the narrowest point of the caudal peduncle (CG = caudal girth). Variables not shown: head width at the top of the orbit, and spleen, liver and heart masses.
Figure 2. A) Scatterplot of individual scores from a relative warps analysis of all wild and LGB specimens, with the uniform component omitted. Symbols represent fish type: ♦ = wild parr, ● = wild smolt, △ = LGB smolt, LGB parr (◇ = early, □ = middle, and ■ = late). Enlarged symbols represent the centroid of each fish type. Relative warps 1 and 2 explained 28.67% and 16.95% of the total variation in shape in the sample, respectively. (B) & (C) Graphical illustrations of the deformation in shape from the consensus configuration implied by positive trends along relative warp 1 (RW1) and RW2, respectively.
Figure 3. A) Scatterplot of individual scores from the canonical variates analysis of all wild and LGB specimens, with the uniform component included. Enlarged symbols represent the centroid of each fish type. CV1 and CV2 explain 50.29% and 28.52% of the among population variation in shape, respectively. B) Deformation grids represent shape change from the consensus configuration implied by each combination of positive and negative trends on CV1 and CV2.
Part 4: Conclusions and Recommendations

The “Vancouver Island Steelhead Recovery Plan” has the potential to greatly influence the recovery of wild Keogh River steelhead. The use of three conservation tools – captive breeding, fishery regulation and habitat enhancement – had demonstrable effects on life-history and demographic patterns, reproductive and ecological interactions, and the phenotypes expressed by hatchery-origin juveniles (Table 1).

The LGB hatchery program influences life-history patterns at each developmental stage. Only wild fish spawned after only one year in the ocean and wild fish were also more likely to be repeat spawners. Conversely, only LGB progeny have been observed to mature and spawn precociously in this river. Due to a greater average fecundity, higher repeat spawning rate, and higher quality spawning locations, an individual anadromous wild steelhead likely has a much greater impact on population recovery than an individual anadromous LGB fish. But due to their great numbers, the population of anadromous LGB fish may still positively influence steelhead recovery. Therefore, due to their apparent lower marine survival and individual reproductive success relative to wild anadromous adults, conservation hatcheries will likely need to produce large numbers of anadromous adults to assist wild population recovery.

Conservation initiatives also greatly influence both wild and LGB fish that migrate to the ocean. Watershed restoration efforts reduced average wild smolt age from three to two years (Ward et al. 2003). Although the average age of LGB migrants was less than that of wild smolt (1 year vs. 2.2 years in 2004), as LGB fish tend to spend longer in the ocean than wild fish, the average total ages of LGB and wild anadromous spawners were similar. Thus, anadromous LGB fish likely did not increase cross-cohort spawning rates. Increases in cross-cohort spawning reduces the probability of inbreeding and may help conserve genetic variability in small populations (Garcia-Vazquez et al. 2001). Although LGB smolt were younger, they demonstrated advanced maturity and precocious development relative to wild conspecifics. Thus, the LGB program is also apparently accelerating maturation rates – likely a result of intensive culture and accessibility to resources (e.g. LGB progeny are well-fed in the hatchery).

During oceanic migrations, LGB fish outnumbered, outweighed, and outconsumed their wild counterparts. LGB migrants represented almost 51 times the number, almost 75 times the biomass, and over 67 times the relative food consumption of wild smolt in 2004. Increases in marine resource consumption due to increased smolt output may result in negative density-dependent growth and survival – a potential explanation of recent further declines in both wild and LGB marine survival. Reduced LGB migrant survival, relative to that of wild anadromous
adults, may be a result of LGB fish tending to spend longer in the ocean than wild conspecifics—a phase where mortality rates are the highest. Alternatively, the vast numbers of LGB progeny may diffuse the pressure of predation of wild smolt—essentially providing protection in numbers.

Besides reducing wild smolt age, habitat restoration efforts may have favoured life-history tactics not prevalent in the wild. Prior to habitat enhancement, stream-resident hatchery fish apparently experienced low survival (Ward and Slaney 1990). As in-stream fertilization increases the production of benthic invertebrates (Johnston et al. 1990), recent watershed restoration efforts likely increased prey abundances, thereby inflating LGB parr survival. Although LGB parr survival may be high, anecdotal and phenotypic evidence suggests that it is likely depressed due to angler harvest—thus, the partial fishery closure protects only the anadromous life-history from exploitation.

By becoming resident in the river, consuming invertebrates and other conspecifics, and maturing precociously, LGB parr represent great changes to in-stream steelhead population dynamics. LGB parr achieve the greatest growth rates among LGB progeny, and hence, may dramatically increase food consumption by steelhead in the river. Due to their larger size relative to wild parr, LGB parr may displace wild fish from optimal habitat and compete with them for limited food resources. In addition to potentially increasing in-river competition for food and space, LGB parr may interfere with wild population recovery by consuming wild steelhead fry. Thus, LGB parr may depress the capacity of the wild population to recover through in-river competition and conspecific predation.

Conversely, both male and female precocious LGB parr may assist population recovery through reproductive functions. In contrast to LGB anadromous adults, LGB parr may help conserve genetic diversity by exhibiting a short time-to-maturity and increasing cross-cohort spawning. Female LGB parr mature in low numbers and with low fecundities, but their extensive experience in the river may give them higher than expected reproductive success. Male LGB parr mature in greater numbers than do females. Male LGB parr likely increase effective population size, reduce gene flow and increase the frequency of LGB and wild crosses. Due to their similarity with other salmonids that demonstrate precocity, LGB parr likely reduce inbreeding rates, achieve high relative reproductive success, and may represent the life-history tactic with the highest relative fitness during current environmental conditions and recovery efforts.

Reproduced with permission of the copyright owner. Further reproduction prohibited without permission.
If marine survival remains low, the importance of LGB parr to conservation will increase. Based on current population sex ratio and size, any decrease in marine survival will result in virtually no surviving wild anadromous males. As mature Atlantic salmon parr have successfully mated with anadromous females in the absence of anadromous males (Myers and Hutchings 1987) and given the similarity between Atlantic salmon and steelhead life-history patterns, mature LGB parr may experience similar spawning scenarios. Hence, if the anadromous male population collapses, mature male parr may prevent complete recruitment failure by ensuring anadromous females can mate. Similarly, if anadromous female spawner numbers continues to decline the relative reproductive contributions of female LGB parr may increase dramatically. Thus, the production of non-migratory mature parr may provide an avenue for conservation hatcheries to conserve genetic diversity and increase effective population size during periods of population decline and reduced marine survival.

Although successful at producing prodigious numbers of juveniles for release, the BC LGB program produced individuals that exhibited phenotypes that differ greatly from that of wild juveniles. LGB smolt tend to be more robust and less streamlined when compared to the wild type, whereas LGB parr (early and middle samplings) are of intermediate body depth – possibly related to the LGB progeny losing visceral fat deposits as they adapt to the river environment. The lack of fusiform shape may be detrimental to LGB migrant survival – with reduced streamlining they are likely inefficient in oceanic migrations. LGB parr also rapidly converge to the wild parr phenotype, likely as a result of phenotypic plasticity. Although LGB parr shape converged with the wild type, LGB parr are larger and may dominate wild parr during in-stream interactions. Furthermore, LGB and wild fish tend to differ greatly for a wide variety of other morphological variables.

Successful conservation programs will likely focus not only on conserving wild genotypes, but also on conserving wild phenotypes. Phenotypically, salmonids are very plastic. Unfortunately, conventional captive environments, such as the BC LGB program, unwittingly exploit this capacity, and may produce hatchery progeny that are better adapted for survival in culture rather than survival in the wild. Although hatchery fish also demonstrate rapid phenotypic convergence with the wild type, evidence suggests that the effects of culture on phenotypes may never fully subside (e.g. Fleming et al. 1994). Currently, the US National Marine Fishery Service, in conjunction with other agencies, is testing a Natural Rearing Enhancement System (NATURES) in which hatchery salmon are reared in raceways that simulate natural environments (Maynard et al. 2001). At present, the NATURES program has

Reproduced with permission of the copyright owner. Further reproduction prohibited without permission.
focused primarily on mitigating behavioural changes associated with culture – as of yet, no detailed phenotypic evaluations have been published.

The suggested positive effects of conservation actions on wild population recovery may be negated if they result in steelhead and coastal cutthroat trout hybridization. The probability of correctly classifying a juvenile steelhead can be low in sympatric hybridizing trout populations (Baumsteiger et al. 2005). As LGB broodstock were selected by visual inspection only, steelhead-coastal cutthroat hybrids, and their backcrosses, may have been incorporated into the captive breeding program. By including only a few hybrids and/or hybrid backcrosses in the LGB broodstock, the LGB program would likely dramatically increase the number of hybrids and hybrid backcrosses in the wild. Any hatchery-mediated addition of hybrids may trigger the formation of a hybrid swarm, even if hybrid fitness is extremely low relative to pure steelhead. In addition, hybrids may be intermediate in phenotype relative to steelhead and coastal cutthroat trout parental types, contributing to decreased marine survival. Thus, when hatchery broodstock contains the genes of non-target species, not only are conservation funds and resources wasted, but also captive breeding could be counterproductive and actually accelerate the loss of genetic diversity in the wild. Thus, when conservation hatchery programs are initiated for any species that has the potential for interspecific hybridization, it is vital that molecular techniques are used to screen for non-target genes in the broodstock.

Hence, my results demonstrate that sophisticated recovery plans have the potential to interfere with population recovery in three ways: 1) by increasing density-dependent effects in both the marine and freshwater environments; 2) by producing hatchery juveniles that experience low marine survival due to their phenotypic divergence from the wild type; and 3) by altering natural rates of interspecific hybridization. With many potential problems arising from captive breeding and partial fishery closures, perhaps the greatest positive influences on wild population recovery would result from combining full fishery closures and habitat enhancement – essentially increasing freshwater survival and only minimally altering wild selective forces and life-history patterns.

Thus, this broad and sophisticated conservation program resulted in dramatic changes to this steelhead population. Patterns of demography, life-history and morphology were altered in predicted, as well as unforeseen ways. In future, conservation programs should be aware that inadvertent changes to these patterns may have serious positive and negative implications on their ability to meet conservation goals. Although such conservation actions have the potential to avert complete population collapse, my results also demonstrate that conservationists should...
be careful and diligent in implementing and monitoring recovery plans – apparently simple conservation plans can have a wide array of unanticipated effects on wild population recovery.
Table 1. Summary of changes to steelhead trout and potential positive and negative outcomes as a result of conservation actions.

<table>
<thead>
<tr>
<th>Change</th>
<th>Positive</th>
<th>Negative</th>
</tr>
</thead>
<tbody>
<tr>
<td>Watershed restoration increased invertebrate production</td>
<td>1) Reduced wild smolt age, likely increasing the proportion of wild juveniles surviving to smolt stage &amp; favours new (resident) life-history tactic – positive if LGB parr spawn</td>
<td>1) Favours new (resident) life-history tactic – negative if LGB parr compete with and consume wild juveniles</td>
</tr>
<tr>
<td>Lower repeat spawning rate in anadromous LGB fish</td>
<td>2) Reduces rearing costs and may reduce intensity of artificial selection in captivity</td>
<td>2) Accelerating development may negatively affect marine survival</td>
</tr>
<tr>
<td>LGB program introduced new life-history tactic</td>
<td>3) None</td>
<td>3) Decreases reproductive success and fitness of an anadromous LGB fish</td>
</tr>
<tr>
<td>Anadromous LGB fish are numerous and increase population size</td>
<td>4) Potential positive genetic effects (see below)</td>
<td>4) Potential negative ecological effects (see below)</td>
</tr>
<tr>
<td>LGB parr prey on conspecific fry</td>
<td>1) Increases the number of breeders</td>
<td>1) As marine juveniles, they outnumber wild conspecifics, possibly creating negative density-dependent marine survival</td>
</tr>
<tr>
<td>LGB parr consume similar food as wild juveniles</td>
<td>1) Unlikely</td>
<td>1) May reduce juvenile recruitment and impede population recovery</td>
</tr>
<tr>
<td>LGB parr distributed throughout river</td>
<td>2) Unlikely</td>
<td>2) May increase density-dependent competition for food</td>
</tr>
<tr>
<td>LGB parr spawn with wild anadromous adults</td>
<td>3) Unlikely</td>
<td>3) May displace wild juveniles from optimal habitat</td>
</tr>
</tbody>
</table>
| LGB program altered rates of steelhead-coastal cutthroat trout hybridization | 1A. Reduced age-at-maturity may increase cross-cohort spawning and reduce inbreeding  
B. Increase effective population size  
C. Reduce gene flow and outbreeding depression  
D. Increase frequency of wild-hatchery crosses  
E. Reservoir of males, ensuring anadromous females can mate | 1) If LGB parr are predominantly hybrids, their spawning will dilute pure steelhead genes |
| Anadromous LGB fish produce fewer eggs than wild conspecifics | 2) Unlikely | 2) Hybrid spawning may dilute pure steelhead genes and trigger hybrid swarm |
| LGB progeny differ in morphology from wild fish | 1) Unlikely | 1) Reproductive success of an anadromous LGB fish likely lower than a wild conspecific |
|  | 2) Unlikely | 2) LGB phenotype is off the adaptive peak of the wild type, reducing LGB survival |
Appendices

Appendix A: Details of Sampling and Euthanasia

The Keogh River has an enumeration fence 300m from the river mouth, that has been in operation since 1975 (Figure 2, Mottram 1977; Johnston et al. 1986; McCubbing and Ward 2004). The fish fence directs migrating salmonids into traps during various times of the year based on the life stage targeted for sampling. In general, each year the fence is used to sample steelhead in three ways: 1) the adult trap is used to collect upstream migrating anadromous adults from January to March, 2) the kelt trap is run from Feb to July to collect downstream migrating kelts, and 3) the smolt trap is operational from April to mid-July. During smolt sampling, the fence configuration uses stop logs to divert the flow of the Keogh into the side channel and the fish are funneled into the smolt trap. In times of extremely high flow secondary smolt traps are used to catch outmigrating smolts (Part 2, Figure 3). The fence is located 300 meters upstream from the ocean.

Capture and Sampling: Winter 2003-Summer 2004

From 18 December 2003 to 28 March 2004, anglers captured anadromous adults, marked the fish by bus punch on the right operculum, and released them at the site of capture. Prior to release, technicians recorded fork length, sex (from external features) and collected scale samples for age determination (as in Maher and Larkin 1954). We can distinguish between LGB- and wild-origin fish easily as the adipose fin has been removed from LGB progeny during culture. From 1 March to 16 July 2004, technicians captured outmigrating kelts in the downstream trap at the fish fence and via seine-netting the pool immediately above the fence structure a minimum of once per week.

Anglers captured, marked and released 81 anadromous adults throughout the river. Later, from 4 April to 16 July 2004, 62 downstream migrants were captured at the fish fence: 15 wild (Recaptures: 3 total, all female) and 47 LGB (Recaptures: 9 total, 5 females).

From 5 April to 24 May 2004, technicians used the enumeration fence to count migrating smolts. As the fence structure directs all outmigrating smolt into a single trap, technicians apparently encountered and tallied the entire wild smolt population. A number of the wild smolt were sampled for fork length (n=362) and mass (n=105).

Wild smolt were collected between 12-19 May 2004, (n= 45). I euthanized these fish with tricaine methane sulfonate (MS222), after which I collected scale samples, measured fork length and mass, and determined sex and sexual maturity (gonad factor as in Rempel et al. 1984).
Near the end of the wild smolt migration, 'LGB smolt' were transported from the rearing facilities to be released into the Keogh River, immediately below the fence structure. I use the term 'LGB smolt' to refer to the LGB-produced juveniles released each year. During each LGB release, LGB smolt were collected from the tanker truck via dipnet. Five releases occurred between 17-26 May 2004. Sampled fish were euthanized (n=301) using MS222 or a solution of clove oil and 95% ethanol, and then recorded fork length, mass, sex and gonad factor.

To sample LGB parr we utilized angling and trapping (using large minnow traps, baited with salmon roe). Depending on data requirements, captured fish were either: a) sacrificed and sampled internally and externally, or b) externally sampled and returned to the river live.

LGB parr from the 2001, 2002 and 2003 LGB releases were sampled throughout the river during two periods. From 19 December 2003 to 11 March 2004, individuals (n=43) were sampled for scales, fork length, and mass. Sex and sexual maturity were also determined using external characteristics where possible (i.e. gamete extrusion). After sampling, individuals were marked with an individually numbered Floy FD-68B tag (Floy Tag Manufacturing Company, Seattle, Washington 98105). Fish were released live for use in a mark-recapture population estimate. From 12 March to 3 June 2004, I captured 2001-2003 LGB parr (n=75), euthanized them using a solution of clove oil mixed with ethanol in a 1:10 ratio, and sampled them for scales, fork length, mass, sex and gonad factor.

As the Keogh River smolt run is typically complete by mid-June (B. Ward, personal communication 20 July 2005), any LGB fish remaining in the river after June 15 was deemed to be a resident and was classified as LGB parr. The 2004 LGB parr were sampled (via angling and trapping; euthanized via MS222 or clove oil) at three times: early, 15-20 June (n=62); middle, 7-21 July (n=93); and late, 4-13 August (n=69). From these individuals I collected scale samples, and recorded sex, gonad factor, fork length and mass.

During mark-recapture efforts, additional fork length and mass measurements were collected from the early 2004 LGB parr (n=366). An additional sample (n=191) of middle 2004 LGB parr were sacrificed to augment my profile of the sex, gonad factor, and size (fork length and mass) of fish that have chosen to adopt one of the LGB parr life-history tactics.

I angled wild parr (n=45) from 4-17 August 2004. Upon capture I lightly sedated an individual (using a clove oil:95% ethanol solution), then collected a scale sample and measured fork length and mass. Fish were then revived in freshwater and released at the site of capture. Sex determination and gut content analysis of wild parr could not be included due to a
moratorium on sacrificial sampling instituted by the BC Ministry of Water, Land, and Air Protection relating to concerns over low wild juvenile numbers.

Capture and Sampling: Summer 2003

During summer 2003 we (C. Werlen and myself) conducted a mark-recapture population estimate of the 2001 and 2002 LGB parr. LGB parr had been encountered approximately 5.5km upstream of the enumeration fence during spring angling in 2003 (D. McCubbing personal communication 30 June 2003). Thus, we chose four 500m reaches within the lower 15km of the river as representative river sections based on accessibility, gradient, habitat variability and encounter data from previous angling efforts (D. McCubbing personal communication 30 June 2003). Reaches consisted of a 300m capture-marking zone, within which we angled, marked (using Floy tags, as above) and released LGB parr. Additional 100m buffer zones were added on either end of the capture-marking zone to allow for short-distance movements of tagged fish without the appearance of migration.

Presented as distance upstream of the estuary, the reaches were: A (Fence) 0.4-0.9km, B (Pumphouse) 1.9-2.4km, C (Cutthroat Creek) 4.9-5.4km, and D (Highway) 14.5-15.0km. These reaches, and their associated capture data, were later used in a mark-recapture population estimate (see subsequent Appendices: B-Population Estimation, C-Snorkel Survey Methods, and D-Snorkel Efficiency Testing).

After the 2003 LGB release an unknown number of LGB juveniles remained below the fish fence. Again, as these individuals did not exit the river system during the annual smolt period, I assumed that these fish were resident, thereby adopting one of the LGB parr life-history tactics. From 17 June to 17 July 2003 the 2003 LGB parr were angled and trapped. Upon capture, fish were Floy tagged and released for use in a mark-recapture population estimate.

Use of Anesthesia/Euthanasia

Sampled fish were anaesthetized with either clove oil or with tricaine methane sulfonate (MS222), depending on the data to be collected from the individual (i.e. samples destined for use in a related epidemiological study were treated with MS222, all others were subdued with clove oil).

Clove oil is an effective anaesthetic for fishes – including *O. mykiss* and other salmonids (Munday and Wilson 1997; Taylor and Roberts 1999; Prince and Powell 2000; Woody et al. 2002). As clove oil does not completely dissolve in water below 15°C, and Keogh River water...
temperatures tend to be below this throughout most of the year (B. Ward personal communication 20 July 2005), clove oil was mixed with ethanol in a 1:10 ratio to facilitate mixing. Ethanol has no known anaesthetic properties on fishes at low doses (Munday and Wilson 1997).
Appendix B: Population Estimation Methods

Based on the collection effort detailed in Appendix A, various mark-recapture techniques were applied to estimate the population size of wild and LGB life-histories. Here I detail specific techniques and how they were used to estimate population sizes.

Both frequentist and Bayesian methods of mark-recapture calculations tend to operate on a similar change-in-ratio concept. Essentially, when one applies a mark to individuals randomly sampled from a population and subsequently returns those marked individuals to the greater population, consecutive samples will allow a comparison of the ratio of marked to unmarked fish. Regardless of whether one uses a frequentist or Bayesian approach to analyze the mark-recapture data, this method provides the following information:

\[ M_t = \text{total number of marked fish at large at the beginning of the } t^\text{th} \text{ sampling interval}; \]
\[ C_t = \text{total number of fish sampled during interval } t; \text{ and} \]
\[ R_t = \text{number of recaptures in sample } C_t. \]

The use of frequentist mark-recapture techniques

Frequentist methods were used to calculate a mark-recapture population estimate of the anadromous steelhead for two reasons: 1) to remain consistent with the historic use of estimation techniques on the Keogh River; and 2) there is only one true recapture period (kelt capture at outmigration) as upstream migrating fish continue to move through and may exit, the sampling area. Thus using recapture data from sampling throughout the river would artificially deflate recapture rates, thereby artificially inflating the population estimates.

As one can visually distinguish between anadromous LGB and wild fish, their population sizes were estimated separately. Due to differential mortality of adult males and females during freshwater residency (Ward and Slaney 1988), the numbers of males and females within each of the LGB and wild populations were estimated separately. This was done using the adjusted Petersen estimate (Ricker 1975). Marking was done by bus punching the operculum of upstream migrant adults and subsequently capturing kelts during their downstream migration (Ward and Slaney 1988, 1990). Captured kelts were examined for operculum marks to enumerate recaptures.

Thus, the following equation was applied separately on the LGB and wild adult mark-recapture data:
\[ \hat{N} = \frac{(M_i + 1)(C_i + 1)}{(R_i + 1)}, \]  
\[ (\text{Ricker 1975}) \]

using the above variable definitions, as well as:

\[ \hat{N} = \text{estimate of steelhead adults, and } t = 1. \]

The use of Bayesian mark-recapture techniques

Traditional frequentist methods of population estimation have problems when data are sparse (i.e. low capture and/or recapture numbers - discussed in Cormack (1968) and Ricker (1975)). As sampling for each LGB parr group lasted for extended periods and typically exhibited low capture and/or recapture rates, I opted to use the Bayesian sequential mark-recapture algorithm (Gazey and Staley 1986) to estimate the LGB parr populations for data collected in 2003 and 2004. Reasoning and the explanation of the algorithm follows:

When sampling is done with replacement (i.e. captured individuals are checked for a mark and then released), the probability that \( C_i \) contains \( R_i \) individuals for a population size \( (N_i) \) is described by the following binomial distribution:

\[
P(R_i \mid N_i) = \binom{C_i}{R_i} \left( \frac{M_i}{N_i} \right)^{R_i} \left( 1 - \frac{M_i}{N_i} \right)^{C_i - R_i} \quad \text{(Gazey and Staley 1986).}
\]

When sampling is done without replacement, the following hypergeometric distribution is more appropriate:

\[
P(R_i \mid N_i) = \binom{M_i}{R_i} \frac{(N_i - M_i)}{C_i - R_i} \left( \frac{N_i}{C_i} \right) \quad \text{(Gazey and Staley 1986).}
\]

As the binomial equation is usually a very good approximation to the hypergeometric distribution (Gazey and Staley 1986), I conducted the population estimates in two ways: 1) applying the binomial equation for all sampling periods, regardless whether I sampled with or without replacement, and 2) using either model where appropriate (i.e. binomial equation for periods of sampling with replacement and the hypergeometric equation for sampling without replacement). Overall, estimates using either equation were similar.
Appendix C: Snorkel Survey Methods and Population Estimation

During summer 2003 we (C. Werlen and myself) conducted a mark-recapture population estimate of the 2001 and 2002 LGB parr. LGB parr had been documented approximately 5.5km upstream of the fish fence during angling in spring 2003 (D. McCubbing personal communication 30 June 2003). Thus, we chose four 500m reaches within the lower 15km of the river as representative river sections based on accessibility, gradient, habitat variability and encounter data from previous angling efforts (D. McCubbing personal communication 30 June 2003). We angled within the four reaches – angling details in Appendix A – marked (via Floy tags as above) and released LGB parr. The entire 500m lengths were used in the snorkel surveys.

Snorkel survey techniques according to Thurow (1994) were employed in the recapture phase of the population estimation. From July 11 to August 17, 2003, 4 snorkel surveys were made on each of the sampling reaches, 16 swims in total.

To extrapolate the data collected from a reach into adjacent areas, we allowed a given reach to represent a length of river half of the distance from its center to the center of adjacent reaches in up- and downstream directions. Presented as distance upstream of the estuary, the reaches were: A (Fence) 0.4-0.9km, B (Pumphouse) 1.9-2.4km, C (Cutthroat Creek) 4.9-5.4km, and D (Highway) 14.5-15.0km. As reach D does not have another reach in the upstream direction, we selected it to represent the balance of the upstream portion of the river. Conversely, reach A can only represent an additional 100m of river in the downstream direction as the fish fence restricts fish movement beyond that point. These reaches, and their associated capture data, were employed in a mark-recapture population estimate (Table 4: calculation methods in Appendix B).

As we did not encounter any marked fish in the 100m buffer zones on the ends of each sample reach, nor did we encounter marked fish in a reach other than the one in which they were initially tagged, we treated each reach as a small, closed population.

The 2001-2002 LGB parr population in reach A was estimated to be 91 individuals (95% HPD: 58-118), or 0.182 fish per meter of river length. Extrapolating this population density throughout the representative section, there were 200 2001-2002 LGB parr in section A.

The 2001-2002 LGB parr population in reach B was estimated to be 70 (95% HPD: 17-218), or 0.140 fish per meter of river length. Extrapolating this population density through the representative section, there were 315 2001-2002 LGB parr in section B.
During marking efforts, no LGB parr were encountered in reach C. One untagged 2001-2002 LGB parr was captured in each of two snorkel surveys. As this may have been a single fish encountered on two occasions or two different individuals, to estimate population densities we treated this as 1.5 LGB parr in the 500m reach – equivalent to 0.003 fish per meter.

Extrapolating this population density throughout the representative section, there were 19 2001-2002 LGB parr in section C.

During both angling and snorkel efforts, no LGB parr were encountered in reach D. Therefore, I assume that reach D, and the river length it represents, contained no 2001-2002 LGB parr.

Thus, by totaling the estimated numbers of 2001-2002 LGB parr in the four river sections I estimate that there were 534 2001-2002 LGB parr present during summer 2003.
Appendix D: Snorkel Survey Efficiency Tests

Methods and Rationale

To ensure that underwater visibility was equal among the three different Floy tag colours we (C. Werlen and myself) used and between tagged and untagged fish, we tested our sampling efficiency on a representative, 77m length of river. To do this we set up nets on both ends of the reach to prevent any fish from moving in or out of the test reach. Next, we used a Smith-Root 12-B electrofisher to remove all unmarked LGB parr. After depletion was suspected (i.e. electrofishing failed to capture additional LGB parr), we introduced 40 LGB parr to be sampled via snorkel surveys: 30 marked with Floy tags (10 marked with each colour) and 10 without tags. A total of eight efficiency swims were conducted on 30-31 July 2003.

After efficiency swims began, it became apparent that not all untagged were initially removed via electrofishing. Thus, I used two independent estimation techniques to determine the number of unmarked fish present during the efficiency snorkel surveys. After all surveys were completed, we conducted extensive fish removal using electrofishing and minnow trapping. I then estimated the number of unmarked fish present in the reach using the depletion data and the Seber-LeCren (1967) formula for depletion curves.

To verify the Seber-LeCren estimate, I also applied the sequential Bayesian mark-recapture algorithm (Gazey and Staley 1986) to the snorkel survey data (see Appendix B for mark-recapture methods).

Once I corrected for unequal initial sample sizes, I analyzed the snorkel data using an analysis of variance (ANOVA) to determine if visibility was different among the three tag colours and unmarked fish. As the data were represented as the proportion of each population observed in a given survey, I arcsine transformed (Zar 1984) the data prior to analysis.

Results

First, I estimated the number of unmarked fish present using the depletion data and the Seber-Le Cren calculation (Seber and Le Cren 1967). As the third pass of electrofishing produced larger catches of unmarked fish than the first two, I treated the third pass as c2 (c2=7) and the fourth pass as c1 (c1=3; i.e. a point where capture rates were beginning to decline). To this estimate I added the catches of unmarked fish from electrofishing attempts 1 and 2, as well as captures from two additional trapping attempts. Thus, the Seber-Le Cren method suggests that there were 22 unmarked fish in the reach prior to the visibility test swims.
Second, the binomial Bayesian sequential mark-recapture algorithm, suggests that there were 22 unmarked fish in the reach prior to the visibility test swims [52 (95% HPD: 46-57) minus the 30 marked fish added initially]. Therefore, as two independent analyses produced identical estimates of the number of unmarked fish present in the swim efficiency reach, I have great confidence in my estimate.

No difference in visual detection was evident among the three mark types and unmarked fish (ANOVA, $F_{3,28} =1.32$, $p=0.28$). Therefore, I am confident that neither tag colour nor the presence or absence of a tag influenced our ability to observe a fish while snorkeling.
Appendix E: Age Determination Methods

Due to a high degree of scale contamination (i.e., mucous, dirt, etc.), LGB parr, LGB smolt, wild parr and wild smolt scales were cleaned prior to mounting using an ultrasonic cleaner (Model UL-009; IKOHE Mahwah, NJ, 07430) in combination with a solution of 0.02g/1mL solution of USP pancreatin (Whaley 1991). Cleaning times ranged from 6-20 minutes, depending on how soiled the scales were initially. Scales were then rinsed with distilled water and mounted on microscope slides in a 60% glycerol medium and sealed. From each individual, up to five scales with a clear freshwater history were digitized using a Leica MZ8 dissecting microscope, connected to a digital camera (CoolSnap by RS Photometrics). Images were then used by two independent readers for age analysis, after which they conferred and mediated on differences.

During the course of the LGB program, the hatchery staff injected coded wire tags into the LGB smolts released in 2002 and 2004, with a different binary code corresponding with each release cohort. Therefore, any individual found to contain one of these tags could be aged conclusively. These tags were surgically extracted and then read using a Leica MZ8 dissecting microscope. The coded wire tag based ages were compared to the age estimates produced by scale reading – with coded wire tag readings taking precedence over a scale-based age estimate if a discrepancy occurred (11% discrepancy rate).

Anadromous adult ages were determined by scale-pattern analysis exclusively. Scales from adults (LGB and wild) were prepared for age determination by making acetate impressions (Narver and Anderson 1974) and projecting the impressions onto a microfiche reader. Three independent readers aged the scales, after which they conferred and resolved any differences.
Literature Cited


Hawkins, D.K. and T.P. Quinn. 1996. Critical swimming velocity and associated morphology of juvenile coastal cutthroat trout (Oncorhynchus clarki clarki), steelhead trout (Oncorhynchus mykiss), and their hybrids. Canadian Journal of Fisheries and Aquatic Sciences 53(1487-1496).


Reproduced with permission of the copyright owner. Further reproduction prohibited without permission.


Kostow, K.E. 2004. Differences in juvenile phenotypes and survival between hatchery stocks and a natural population provide evidence for modified selection due to captive breeding. Canadian Journal of Fisheries and Aquatic Sciences 61(4): 577-589.


Lambert, Y. and J.-D. Dutil. 1997. Can simple condition indices be used to monitor and quantify seasonal changes in the energy reserves of Atlantic cod (Gadus morhua)? Canadian Journal of Fisheries and Aquatic Sciences 54(Suppl. 1): 104-112.


Reproduced with permission of the copyright owner. Further reproduction prohibited without permission.


Shapovalov, L. and A.C. Taft. 1954. The life histories of the steelhead rainbow trout (Salmo gairdneri gairdneri) and silver salmon (Oncorhynchus kisutch) with special reference to Waddell Creek, California, and recommendations regarding their management. California Department of Fish and Game Fish Bulletin 98: 375pp.


Reproduced with permission of the copyright owner. Further reproduction prohibited without permission.


Reproduced with permission of the copyright owner. Further reproduction prohibited without permission.


Reproduced with permission of the copyright owner. Further reproduction prohibited without permission.