PHYSIOLOGICAL, BEHAVIOURAL AND SURVIVAL EFFECTS OF ASSISTING THE
POST-CAPTURE VENTILATION OF ADULT SOCKEYE SALMON EXPOSED TO
CAPTURE AND RELEASE IN FRESH WATER

by

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Abstract

Fish that are released from fisheries capture exhibit physiological and behavioural changes that can result in mortality. The ability to release fish that do not experience subsequent fitness consequences is fundamental to fisheries conservation and management tools that mandate live release. Thus, researchers have evaluated methods that fishers can use to reduce the potential for negative capture-related effects. Indeed, modifying capture and landing practices can limit the severity of the physiological and behavioural impairments. Moreover, release techniques that enhance the metabolic recovery process essential for mitigating capture-related physiological changes may help to enhance survival. Because this essential recovery process requires oxygen consumption that exceeds basal metabolic needs, I evaluated a ventilation assistance technique that forced a high flow of water over the fish's gills in an attempt to provide additional oxygen. This assisted ventilation technique mimics manual recovery attempts that are recommended by fisheries managers and often employed by recreational anglers.

The physiological, behavioural and survival responses of adult migrating Fraser River sockeye salmon (*Oncorhynchus nerka*) to capture and release, with and without ventilation assistance, were assessed in laboratory and field experiments. A simulation of capture and release consisting of 3 minutes of strenuous exercise and 1 minute of air exposure resulted in significant physiological impairment in the laboratory experiment. In a field experiment, this simulation resulted in an approximate 30% overall reduction in post-release survival to reach natal spawning grounds. Female fish exposed to simulated capture and release exhibited poorer survival relative to control females and males of all treatments in both of these

experiments. The 1-minute assisted ventilation technique did not enhance survival. In fact, further reductions in survival were observed in the laboratory experiment for females subjected to ventilation assistance before release from capture.

Capture and release can result in delayed mortality and it appears that a recovery technique recommended by fisheries managers to recreational fishers does not help to reduce capture-related mortality. Mitigating negative capture-and-release effects by disseminating capture and landing best practices, while incorporating scientifically-defensible post-capture mortality rates into management plans, may be the best approach to meeting conservation and management objectives.

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Preface

This research was carried out as a component of a multi-disciplinary research program looking to quantify and reduce mortality of released fish in multi-sector Pacific salmon fisheries in the coastal rivers of British Columbia. I held primary responsibility for research design and experimental protocols, collection and analysis of data, and preparation and submission of manuscripts. During this process, I received considerable logistical support from my colleagues, and guidance from my supervisor Dr. Scott G. Hinch and my supervisory committee members, Dr. Steven J. Cooke and Dr. John S. Richardson. Collaborators on individual projects who were instrumental in development, experimentation or manuscript preparation are listed as coauthors on manuscripts that will be submitted for publication. All experimental procedures were approved by the University of British Columbia Animal Care Committee (#A08-0388) and conducted in accordance with guidelines set forth by the Canadian Council on Animal Care.

Chapter 2: **Effects of post-capture ventilation assistance and elevated water temperature on sockeye salmon in a simulated capture-and-release experiment**

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Chapter 3: **Influence of assisted ventilation on migration behaviour and survival of adult sockeye salmon after capture and release in fresh water**

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Table of Contents

List of Tables

List of Figures

Acknowledgements

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Chapter 1: Introduction

An understanding of the stress experienced by fish during capture and the effects it has on those that are released can inform the management and conservation of fish populations. The live release of fish is a commonly employed management tactic designed to reduce fisheries pressures on particular populations (Cooke and Schramm 2007); however, the efficacy of this technique relies on the subsequent survivability of fish that are released (Wydoski 1977). Indeed, fish exposed to fisheries capture elicit a stress response and experience physiological changes that require a considerable period of metabolic recovery for their future performance, and ultimately their survival upon release (Milligan 1996). This thesis evaluates a method of aiding this essential recovery process prior to release from fisheries capture, thereby attempting to enhance post-release survival and achieve management objectives.

1.1 Physiological changes associated with fisheries capture and release

An encounter with fisheries gear elicits a stress response (e.g., Davis 2002) which consists of physiological and behavioural adjustments that work to maintain or re-establish homeostasis and thus promote survival (Pickering and Pottinger 1995; Wendelaar Bonga 1997; Barton 2002). The stress response can be conceptualized into three levels: primary, secondary, and tertiary (Wedemeyer and McLeay 1981; Barton 2002). The primary (neuroendocrinological) response consists of the immediate release of catecholamines (i.e., epinephrine and norepinephrine) and corticosteroids (i.e., cortisol; Barton 2002). These hormonal secretions induce secondary responses related to cardiorespiration, carbohydrate metabolism, and osmoregulation (Pickering *et al*. 1982; Pickering and Pottinger 1995). The

tertiary response is a whole body response to the stimuli (e.g., changes in growth, immune function, and reproduction) that may ultimately influence survival (Muoneke and Childress 1994; Barton 2002).

The primary and secondary responses are adapted to overcome threats to homeostasis that are associated with encountering short-term stressors, such as fisheries capture. For example, a capture event results in the increased consumption of oxygen at the tissues (Arlinghaus *et al*. 2007). The stress response alters blood chemistry and increases heart rate and ventilation rate to enhance the transport of oxygen to the tissues and thereby attempt to maintain aerobic processes such as energy production (Pickering and Pottinger 1995). Indeed both aerobic and anaerobic pathways can be used to produce energy; however, it is advantageous to the fish to maintain aerobic production as it is the more efficient method of producing energy and does not result in the accumulation of metabolites (Pickering and Pottinger 1995). Thus, these biochemical and physiological alterations are intended to maintain internal stability when the fish is exposed to fisheries capture.

Fish use burst swimming in attempts to evade capture or escape landing during a fisheries gear encounter (Milligan 1996; Kieffer 2000). Burst swimming consists of powerful, yet short bouts of exhaustive movement and the nature of this activity requires energetic output that exceeds that which is producible by aerobic metabolism alone (Pickering and Pottinger 1995). Therefore, burst swimming events are primarily fueled by the anaerobic breakdown of glycogen within white muscle fibers (Black *et al*. 1966). This glycolysis results in the depletion of glycogen reserves and the production of lactate and proton metabolites that accumulate in the muscle (Wood 1991). The accumulation of lactate in the muscle tissue creates an osmotic gradient that results in ionic and fluid volume

alterations (Turner *et al*. 1983; Wood 1991). As the lactate and protons in the muscle slowly leak into the blood, elevated levels of protons, along with an increase in carbon dioxide levels, result in metabolic and respiratory blood acidoses (Wang *et al*. 1994). Thus, it is crucial for fish to metabolically recover from these capture-related physiological changes, particularly as the resynthesis of glycogen stores and the clearance of anaerobic metabolites are required for future burst swimming events (Milligan 1996). The recovery processes require the uptake of oxygen in excess of basal metabolic rates, termed excess post-exercise oxygen consumption (EPOC; Gaesser and Brooks 1984). Hormones secreted during the stress response increase the diffusion of oxygen across the gills and increase the rate/volume of ventilation in order to meet the increased oxygen demands (Pickering and Pottinger 1995) and thereby promote physiological recovery to pre-capture levels. Research has indicated that evaluating how fish respond to capture at the physiological level is important to assessing the potential for negative consequences of capture and release (Cooke *et al*. in press).

1.2 Mortality and sublethal consequences of fisheries capture and release

An understanding of the stress imposed on natural populations by human-induced capture stressors is essential to predicting their influence on released fish (Pickering *et al*. 1982). Fish that are released from fisheries capture events can suffer delayed mortality or harmful sublethal effects (Chopin and Arimoto 1995); therefore, these released fish may be impaired in a manner that affects their subsequent fitness. By definition, delayed mortality occurs after the fish swims away from release and is the result of injury and/or physiological stress (Arlinghaus *et al*. 2007). For example, the strenuous burst exercise that is associated with capture and landing leaves fish with depleted glycogen stores and accumulated

anaerobic metabolites (Wood 1991). As a result, repeat burst swim performance is limited by the duration required to resynthesize energy stores and clear lactate (Milligan 1996). Early research evaluating recovery durations suggested complete metabolic recovery takes 2 – 24 hours (Milligan 2000); however, subsequent burst events do not require complete recovery (Farrell *et al*. 1998). The resynthesis of glycogen and the clearance of lactate can recover in as little as 40 minutes, depending on the situation (Wood 1991). However, during this recovery period, fish are vulnerable to mortality associated with predation or recapture (Chopin and Arimoto 1995). Even more, fish that do not suffer fatal repercussions may experience growth, immune system, and reproductive impairments (Chopin and Arimoto 1995) and thereby potentially suffer fitness consequences. These sublethal effects of fisheries capture are not as well understood as they are difficult to evaluate.

Fisheries capture events can vary greatly in consideration of the capture circumstances. For example, the gear type, angling duration, air exposure, species of fish, and numerous environmental factors (e.g., water temperature) can influence the potential for negative effects of capture and release (Cooke and Suski 2005; Arlinghaus *et al*. 2007; Gingerich *et al*. 2007). As a result, delayed mortality levels can range from 0 – 89% on a case-by-case basis (Muoneke and Childress 1994; Arlinghaus *et al*. 2007). Indeed, the severity (duration and magnitude) of the stressors can impact both the time trajectory of the stress response and the recovery process (Donaldson 2012). Therefore, the duration required to mitigate metabolic disturbances resulting from a capture event can vary greatly as well. Thus, the mechanism by which fish recover from capture and avoid fatality is highly relevant to fisheries practices, and evaluating methods to reduce capture-related mortality is an integral contribution to conserving and managing populations (Cooke and Schramm 2007).

1.3 Efforts to reduce capture-related mortality and sublethal effects upon release

Understanding and reducing capture-related effects on released fish is important for the effectiveness of capture-and-release management tools. As a result, studies have evaluated recovery devices as methods of reducing the negative effects capture events have on fish. Researchers have looked to facilitate the recovery of fish post-capture using recovery devices built on the premise of increasing oxygen availability during EPOC (Farrell *et al*. 2001a; Donaldson *et al*. 2013; Nguyen *et al*. in press). For example, Farrell *et al*. (2001a) attempted to enhance recovery of coho salmon (*Oncorhynchus kisutch*) bycatch in a marine commercial gillnet fishery off the coast of British Columbia. The fish were placed within a wooden, rectangular box sized to restrain movement and orient the fish towards the inflow of a jet nozzle to assist ventilation. The authors found that a $1 - 2$ hour recovery duration promoted metabolic recovery (i.e., decreased muscle lactate and stabilized glycogen levels) and reduced post-release short-term (24 hour) mortality. These results suggest that the assisted flow of water over the gill surface provides the additional ventilation needed to aid in the metabolic recovery and ultimately reduce mortality of fish after release from capture.

Best practice recommendations describing manual recovery techniques prior to release are included in a number of recreational capture-and-release angler guidelines (Arlinghaus *et al*. 2007; Pelletier *et al*. 2007). Such techniques include physically holding the fish into the flow of a fluvial system or moving fish back and forth, side to side or in an S-shaped pattern. All of these methods attempt to assist the flow of water over the gill surface prior to release. The logic behind such a technique may be seeded in research that shows oxygen is necessary for metabolic recovery from strenuous exercise (Wood 1991). However, the methods mentioned here are built on the underlying assumption that the use of manual

restraint to facilitate an increased flow of water over the gills will outweigh the potential negative consequences of further physical confinement. Thus, validation of current release strategies that aim to facilitate this essential recovery process is necessary to strengthen our knowledge of fisheries best practices.

1.4 Study species and local managerial and environmental challenges

The Fraser River (British Columbia, Canada) produces salmon of cultural, ecological, economical, and political importance to Canadians. Beginning in midsummer and continuing into late fall, this river is home to millions of migrating adult sockeye salmon (*Oncorhynchus nerka*). After spending 2 – 3 years in the ocean, these fish return to their natal spawning grounds for their sole opportunity to reproduce. However, this freshwater migration passage poses numerous bioenergetic and navigational challenges to securing fitness (Hinch *et al*. 2006). Maturing sockeye salmon cease feeding prior to re-entry into fresh water. Thus, they depend only on endogenous energy reserves for fueling aerobic and anaerobic activity to complete their hydraulically challenging migration (Hendry and Berg 1999). These finite energy stores are also being diverted to gonadal growth and the development of secondary sexual characteristics (Brett 1995). Upon reaching their natal spawning grounds, sockeye salmon will find a mate, spawn and die, leaving behind the next generation of sockeye salmon developing within gravel nests (Burgner 1991). The failure to reach spawning grounds and successfully pass on their genes will result in zero lifetime fitness.

In addition to this naturally arduous migration journey, Fraser River sockeye salmon are currently experiencing elevated river temperatures. From 1953 – 2006, the Fraser River mean summer water temperature has increased by ~ 2°C (Patterson *et al*. 2007) and this

warming trend is predicted to continue (Morrison *et al.* 2002; Ferrari *et al.* 2007; Hague *et al.* 2011). Elevated water temperatures during spawning migration have been shown to reduce the survival of adult sockeye salmon (Gilhousen 1990; Macdonald *et al.* 2010; Martins *et al.* 2012) and further reduce the likelihood of survival if fish are captured and released (Martins *et al.* 2011).

Adult migrating sockeye salmon will likely encounter a suite of fisheries gears that further intensify the difficulty of reaching spawning grounds to reproduce. Commercial, First Nation, and recreational fishery sectors target these fish in coastal estuarine and freshwater environments. Fisheries and Oceans Canada (DFO) regulates this multi-sectoral sockeye salmon fishery with advice and recommendations from the Pacific Salmon Commission (PSC). Based on estimates of run size, timing and stock composition of sockeye salmon returning to spawn each year, managers can calculate spawning escapement targets (the number of fish to reach their natal spawning grounds) and harvest rates. Thus, managers will adjust the multi-sectoral harvesting of sockeye salmon to ensure they reach their management objectives. A method of curtailing harvest while maintaining fishing openings uses the mandated live-release of fish after capture in order to reduce fisheries pressures. Fish that are not harvested after encountering fishery gear, whether they have escaped landing or were actively released as in commercial bycatch or capture-and-release angling, can continue their journey to reach natal spawning grounds. This assumes that the fish that are released will survive to reach their natal sites and successfully reproduce.

Sockeye salmon are released from the Fraser River recreational fishery. They can be released voluntarily or in response to mandated harvest restrictions. Non-retention regulations may be applied to reduce mortality and thereby meet escapement targets or

reduce fishing pressures on co-migrating populations of concern (DFO 2012a). Thus, it is important that managers quantify delayed mortality in order to understand the effects of live-release conservation and management tools on expected results. By knowing this mortality level, they can then adjust harvest rates to ensure targets are met. Techniques that can enhance post-release survival will reduce the need to decrease harvest rates. Thus, the ability to manage for sustainable fisheries can benefit from a reduction in capture-induced mortality of fish intended for release (Cooke and Cowx 2006). Validating and implementing techniques that assist ventilation to minimize the lethal consequences of fisheries capture and release may aid in ensuring the stability of Fraser River sockeye salmon populations, particularly in light of climate change.

1.5 Thesis goal and objectives

The goal of this thesis is to evaluate the use of post-capture ventilation assistance as a technique for facilitating the recovery of Fraser River sockeye salmon prior to release. I examined physiological, behavioural and survival responses to this recovery technique in a controlled laboratory setting and in the natural environment of two field-based experiments. Chapter 2 reports the findings from the laboratory experiment that compares the physiological and survival responses of adult sockeye salmon to capture and release with or without ventilation assistance. The influence of elevated water temperature on the efficacy of this facilitated recovery technique was assessed by holding fish at two ecologically relevant temperatures. Chapter 3 provides the results of two field experiments that used biotelemetry to evaluate the migration behaviour and survival of adult sockeye salmon after release with or without ventilation assistance. All experiments evaluated the same assisted ventilation procedure, thus linking the benefits of both laboratory and biotelemetry approaches. In my

concluding chapter (Chapter 4), I provide a synthesis of my findings, discuss the implications

of my research for fisheries management and suggest possible areas for future investigation.

Chapter 2: Effects of post-capture ventilation assistance and elevated water temperature on sockeye salmon in a simulated capture-and-release experiment[1]

2.1 Introduction

The ability of organisms to respond to stressors encountered in the natural environment is essential to their persistence, but the response itself can be energetically costly and requires additional energy and time to recover to homeostasis (Wendelaar Bonga 1997). Fisheries capture and handling results in a physiological stress response that begins aerobically with the release of catecholamines and corticosteroids (Pickering and Pottinger 1995). The secretion of these hormones increases heart rate and ventilation rate, and stimulates changes in hematocrit and hemoglobin to increase the carrying capacity of blood to meet mounting tissue oxygen requirements (Pickering and Pottinger 1995). However, a prolonged or high intensity response to the fisheries capture event will trigger anaerobic pathways as the tissue oxygen demand exceeds supply (Pickering and Pottinger 1995). For example, burst swimming, a short duration of rapid movement typically associated with fisheries gear encounters, is fueled by the anaerobic breakdown of glycogen in white muscle fibers (Black *et al*. 1966). The subsequent glycolysis results in lactate and metabolic proton accumulation in the muscle and blood. These metabolites alter the acid-base status (Milligan

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1996), and the buildup of muscle lactate attracts plasma fluid resulting in subsequent ion-osmoregulatory imbalances (Turner *et al*. 1983; Wood 1991; Kieffer 2000). The metabolic recovery from these changes requires oxygen in excess of basal metabolic needs for processes such as lactate clearance and glycogen resynthesis (Wood 1991). This incurred recovery cost, termed excess post-exercise oxygen consumption (EPOC; Gaesser and Brooks 1984), is partially facilitated by an increase in the capacity for oxygen diffusion across the gills and an increase in the frequency or volume of ventilation (Scarabello *et al*. 1991; Wood 1991). Although homeostasis can be regained following these metabolic disturbances associated with burst activity, fish released from fisheries capture events may experience a considerable period of physiological limitations.

Performing subsequent burst swim events within an ecologically relevant timeframe is important for fish returned to their natural environment after a capture-and-release fishing event. If the physiological disturbance is severe enough and homeostasis cannot be readily achieved, the fish could perish from physiological stress (e.g., blood acidosis; Wood 1983). In less severe instances, fish that are immediately released from capture, prior to the restoration of energy reserves, are incapable of subsequent burst swims (Milligan 1996). Studies suggest that the duration of recovery needed for repeat burst events ranges from 40 minutes to several hours (reviewed in Wood 1991). Indeed, during this period, these fish are vulnerable to predators or recapture, or in fluvial systems could be forced to fall back downstream as a result of physiological and behavioural impairments (Donaldson *et al*. 2012). Even more, the circumstances surrounding the capture event (e.g., handling and air exposure) can prolong the recovery process (Kieffer 2000; Suski *et al*. 2006), For example, fish exposed to air during handling (e.g., removal from net, unhooking, photography) incur

additional physiological alterations that occur as tissues are starved of oxygen (Ferguson and Tufts 1992), much like the effects of hypoxia (reviewed in Cooke and Suski 2005). The ability for repeat performance requires aerobic metabolism to restore anaerobic ability in a timely manner (Farrell *et al*. 1998) and this process is vulnerable to the specific circumstances of the capture-and-release event (Suski *et al*. 2006).

Adult sockeye salmon (*Oncorhynchus nerka*) that return to the Fraser River, British Columbia, Canada during their once-in-a-lifetime spawning migration are highly valued by commercial, First Nation, and recreational fishers. They are captured in both marine and freshwater fisheries, generally by rod and reel, gillnet, and beach seine in the latter environment. In order to protect threatened stocks and to ensure spawning escapement targets are met for all stocks, managers implement catch limits and temporal fishery closures that force the live release of fish. In addition, sockeye salmon are voluntarily released by recreational rod-and-reel anglers. In 2010, the recreational fishery released one-third of sockeye salmon (~ 100,000 fish) captured in the lower Fraser River, and in the previous 4 years released an average of 21,000 fish per year (DFO 2011). However, little is known about the effects this fishery has on the survival of sockeye salmon released from freshwater capture. A recent telemetry experiment estimated that ~ 20 – 35% of Fraser River sockeye salmon released from freshwater angling or beach seine perished as a result of the capture-and-release event prior to reaching spawning grounds (Donaldson *et al*. 2011). In addition to the capture-related stressors, Fraser River sockeye salmon are exposed to elevated water temperatures during their spawning migration. Fraser River mean summer water temperatures have increased by ~ 2°C over the past 60 years with 13 of the warmest summers on record occurring over the last 20 years (Patterson *et al*. 2007). High water

temperatures have been correlated with in-river mortality during spawning migration (Macdonald *et al*. 2010), and more specifically, high temperatures at the time of capture and tagging have also been associated with elevated mortality post-release in the wild (Martins *et al*. 2011). A corroborating laboratory study, evaluating capture-related stressors on sockeye salmon, found that warm water temperatures during simulated capture-and-release events that incorporated air exposure depressed ventilation rates immediately after capture (Gale *et al*. 2011), potentially limiting oxygen availability at the gills, and those that exhibited reduced ventilation rates were less likely to survive the subsequent 24 hours (Gale 2011). These results suggest that releasing physiologically impaired sockeye salmon which have not adequately recovered can leave fish vulnerable to delayed capture-related mortality, particularly when in warm waters.

Researchers have evaluated various devices for promoting physiological recovery to reduce the mortality of Pacific salmon (*Oncorhynchus* sp.) after release from capture. It is known that the delivery of adequate oxygen to the tissues following fisheries capture is important for recovery (Wood 1991). Indeed, approaches to facilitating recovery have attempted to assist ventilation during metabolic recovery by ensuring that water is forced across the gill surface to provide the oxygen required during EPOC (see Farrell *et al*. 2001a; Donaldson *et al*. 2013; Nguyen *et al*. in press). The Fraser Box, a device which forces ventilation by restricting fish movement while orienting it into an inflowing water jet, promoted physiological recovery and reduced short-term (24 hour) mortality for coho salmon (*Oncorhynchus kisutch*), which are caught as bycatch in marine commercial gill net fisheries (Farrell *et al*. 2001a). Manual recovery techniques that do not rely on devices are recommended to recreational anglers by North American natural resource agencies (Pelletier

et al. 2007). These methods facilitate the flow of water over the fish's gills by moving the fish in an S-shape or back and forth in the water, or by orienting the fish upstream in an attempt to enhance recovery prior to release. Currently however, there is a clear lack of conclusive evidence regarding the efficacy of these manual recovery techniques (Arlinghaus *et al*. 2007; Pelletier *et al*. 2007).

The objective of this study was to evaluate ventilation assistance as a means of facilitating the recovery of adult migrating Fraser River sockeye salmon after a simulated fisheries capture-and-release event. Unlike previous studies, we did not use restraining devices and instead manually held fish into a set water flow to more closely simulate simple and inexpensive approaches that could be readily adopted by fishers (e.g., holding a fish facing into river flow). We incorporated a thermal treatment in consideration of research that has reported a reduction in ventilation rate after capture and release at ecologically relevant high water temperatures (Gale *et al*. 2011). This addition enabled us to compare post-treatment survival at a typically experienced water temperature and at a relatively high water temperature that is occasionally experienced now and which is expected to be commonly experienced as the Fraser River continues to warm. We predicted that assisting post-capture ventilation would increase the survival of adult sockeye salmon following a fisheries capture-and-release simulation.

2.2 Methods

2.2.1 Study site and animals

Adult sockeye salmon were collected by beach seine on August 10 – 13, 2010 from the Fraser River at Chilliwack, British Columbia, Canada soon after commencing their

freshwater spawning migration. River temperature during collection was 18 – 20°C. Fish

were transported in aerated ~ 14°C water using truck-mounted transport tanks to the

Fisheries and Oceans Canada (DFO) Cultus Lake Salmon Research Laboratory (~ 26 km;

Figure 2.1). Upon arrival, Passive Integrated Transponder (PIT) tags (~ 8.5 mm x 2 mm size,

134.2 kHz, Biomark Inc., Boise, Idaho) were inserted into the coelomic cavity for individual

fish identification. Fish were then transferred to 10 circular 1400 L aerated tanks (2 m

diameter; 12 – 13 fish tank^{-1}) supplied with filtered and UV sterilized fresh water (LS-

Permabead Filtration System, Integrated Aqua Systems Inc., Escondido, California) from

Cultus Lake. The fish were held at 14°C for $\geq$ 15 h to recover from transport.

2.2.2 Experimental design

One hundred and three fish (55 males and 48 females) were used in the experiment.

On August 14, 2010, the holding temperature was gradually increased (~ 0.5°C h^{-1}) to 16°C

(five tanks) and 21°C (five tanks). These temperatures were chosen to approximate current

average temperatures and current peak temperatures experienced by adult sockeye salmon

migrating through the lower Fraser River at this time of year (Patterson *et al*. 2007). The fish

were held at these temperatures for $\geq$ 36 h before the experiment commenced.

Fish were subjected to one of three simulated fisheries capture-and-release

treatments: (i) control (20 males (10 at 16°C, 10 at 21°C) and 13 females (8 at 16°C, 5 at

21°C)), (ii) simulated capture without assisted ventilation (18 males (10 at 16°C, 8 at 21°C)

and 16 females (8 at 16°C, 8 at 21°C)), and (iii) simulated capture with assisted ventilation

(17 males (10 at 16°C, 7 at 21°C) and 19 females (9 at 16°C, 10 at 21°C)). The control fish

were not subjected to the simulated capture or assisted ventilation. The simulated fisheries

capture consisted of 3 min of manual chasing by four experimenters leaning over a ~ 800 L

doughnut-shaped tank (2 m diameter) corresponding to the appropriate experimental

temperature. This was followed by 1 min of air exposure in a dip net. Similar techniques

have been used extensively in stimulating exhaustive exercise in fish (reviewed in Milligan

1996; Kieffer 2000) and have been refined for fisheries capture simulations with adult Fraser

River sockeye salmon (Donaldson *et al*. 2011; Gale *et al*. 2011). This capture-and-release

simulation does not impose the physical injury that is often associated with fisheries gear

encounters. Injury from fishing gear and capture-release practices can contribute to

immediate and delayed mortality (reviewed in Chopin and Arimoto 1995).

Half of the fish that were exposed to the simulated fisheries capture event were then

subjected to the assisted ventilation treatment in which fish were oriented into a jet of water

flow (~ 0.50 m s^{-1} measured at the mouth) from a submersible pump for a maximum of 1

min, thus forcing ventilation. We chose a water speed of ~ 0.5 m s^{-1} because it is similar to or

greater than speeds regularly found in the Fraser River and its tributaries (Hinch and Rand

2000), and because it is comparable to the water speeds assessed in a recent study evaluating

portable recovery bag outcomes for Fraser River sockeye salmon (~ 0.1 to 0.4 m s^{-1};

Donaldson *et al*. 2013). Opercular beats were observed as the experimenter physically held

the fish with the mouth ~ 20 cm from the jet outlet. If the fish became highly vigorous and

attempted to escape from the experimenter's hands during this time, it was released (similar

to an angler releasing an active fish) and the duration of assisted ventilation was recorded. Of

the 36 fish subjected to assisted ventilation, 12 were released prior to the completion of the

1-min treatment. Due to logistics, two fish were subjected to identical and simultaneous

protocols within the same doughnut-shaped tank at the same time.

After treatment, fish were transferred by dip net to an individual rectangular holding tank (~ 100 L, 1 m x 0.5 m x 0.3 m deep) with fresh flowing water of the appropriate experimental temperature. Thirty minutes after the initiation of the capture-and-release simulation, fish were sampled for blood (see below) in a flow-through, foam-lined trough and then transferred to 7000 L aerated tanks (3 m diameter) at the appropriate experimental temperature for long-term monitoring. Control fish were dip-netted directly from the 1400 L tanks and sampled for blood immediately before transfer to the 7000 L tanks. All fish were maintained in their respective temperatures for up to 33 days to examine survival. By that point, we expected these fish to have arrived at their natal spawning grounds (see below; Gilhousen 1990).

Mortalities were dissected as they occurred; fork length (FL), body mass (M_B) and sex were recorded post-mortem. A piece of opercular tissue (7 mm diameter) was removed for DNA stock identification (Beacham *et al*. 2005). This confirmed that the 103 fish were from Early Summer and Summer-run stock groups, a classification scheme for fisheries managers based on the timing of river entry. Early Summer and Summer-run sockeye salmon enter the river in July and August and experience overlapping river temperatures (Patterson *et al*. 2007) with their arrival on natal spawning grounds typically occurring < 33 days following freshwater entry (Gilhousen 1990). These run-timing groups were pooled for analyses.

2.2.3 Blood sampling and laboratory assays

Blood samples were collected to examine the short-term effects of the treatments on physiological variables. This allowed us to ensure that our fisheries capture simulation

17

resulted in physiological disturbances and then compare these disturbances to other related studies. Blood samples were attained via caudal puncture (~ 3 mL) using a heparinised vacutainer (detailed in Cooke *et al*. 2005) and stored in a water-ice slurry for $\leq$ 1 h (Clark *et al*. 2011). A hand-held hemoglobin analyzer (HemoCue Hb 201^{+}; HemoCue, Ängelholm, Sweden; calibrated for fish blood) was used on whole, well-mixed blood (Clark *et al*. 2008). Percent hematocrit was quantified using hematocrit tubes centrifuged at 10,000 x g for 3 min. Mean corpuscular hemoglobin concentration (MCHC) was calculated as [hemoglobin] / ([hematocrit] / 100) (as in Donaldson *et al*. 2010b). The remaining whole blood was centrifuged at 7,000 x g for 5 min. Plasma was isolated and flash frozen in 1.5 mL cryogenic vials in liquid nitrogen prior to storage at -80°C. Plasma was analyzed for cortisol (Neogen ELISA with Molecular Devices Spectramax 240pc plate reader), lactate and glucose (YSI 2300 Stat Plus analyzer), osmolality (Advanced Instruments 3320 freezing-point osmometer), chloride (Haake Buchler digital chloridometer), sodium and potassium (Cole-Palmer, model 410 single-channel flame photometer), as described in Farrell *et al*. (2001a).

2.2.4 Statistical analyses

Two-way analyses of variance (ANOVA) were used to test for differences in size (FL and M_B) among treatment groups. No significant differences were found in either FL or M_B across groups (ANOVA: $P > 0.05$), and so groups are compared directly herein. The effect of simulated capture-and-release and temperature treatments on physiological variables was examined using two-way ANOVA. One fish in the 21°C and assisted ventilation group died within 20 min of blood sampling, 17 h earlier than all other instances of mortality, and exhibited anomalous blood physiological parameters. This individual was subsequently removed as an outlier in all analyses of physiological variables. Plasma cortisol values were

analysed separately for sex in consideration of the naturally higher plasma cortisol values of migrating female sockeye salmon in comparison to migrating male sockeye salmon, as reported in the literature (Sandblom *et al*. 2009; Hruska *et al*. 2010; Roscoe *et al*. 2011). Data for sodium, potassium, and hematocrit were $\log_{10}$ transformed to meet parametric assumptions. We used a power transformation on lactate values and a rank transformation on osmolality values in order to meet parametric assumptions. Significance levels were set at 0.05. Where significant differences were detected among simulated capture treatments, Bonferroni multiple comparisons tests were used. Percent survival at 10 days or 15 days was compared between simulated capture treatments of males and females at 16°C with Fisher's exact tests using Bonferroni corrections ($P = 0.017$).

2.3 Results

The fisheries capture-and-release simulation significantly increased plasma lactate, cortisol (male and female), glucose, osmolality, and hematocrit relative to controls (Table 2.1). Female cortisol concentrations were roughly two-fold greater than male concentrations of corresponding groups. Plasma potassium and MCHC significantly decreased in response to the capture-and-release simulation relative to controls. Assisted ventilation did not have a significant effect on any blood parameters when compared with the unassisted group, with the exception of plasma lactate at 21°C and hematocrit. In both these instances, the magnitude of change in reference to the controls increased in the assisted ventilation groups. The temperature treatment had a significant effect on plasma potassium, hematocrit, and hemoglobin. Treatment × temperature interactions were detected for plasma lactate and cortisol (males only; see Table 2.1).

Fish held at 21°C exhibited 100% mortality across all groups within 3 days after the simulated capture-and-release treatment (Figure 2.2). At 16°C, mortality began 4 days after the simulated capture treatment and continued until the end point of the experiment (33 days) when seven fish remained. Due to the rapid mortality exhibited at 21°C, we focused on fish held at 16°C for subsequent treatment comparisons. Females subjected to assisted ventilation exhibited poorer survival in comparison with non-assisted and control females, and compared with males of all treatments (Figure 2.3). We focused our survival analyses at 10 and 15 days after the simulated capture-and-release treatment because these durations reflect a range of times it would take Early Summer and Summer-run sockeye salmon to reach their natal tributaries from our capture locale (Gilhousen 1990). At day 10, 11% (1 of 9) of females that received assisted ventilation survived compared to 63% (5 of 8) of females that did not receive assisted ventilation and 75% (6 of 8) of control females (Figure 2.4). By day 15, females from both of the simulated capture groups exhibited poor survival (0% (0 of 9) for the assisted ventilation group, 25% (2 of 8) for the non-assisted group) compared to the female controls (63% (5 of 8); Figure 2.4). At both day 10 and day 15, the percent survival of females that received assisted ventilation was significantly lower than the percent survival of control females. There were no significant differences in survival for males between treatment groups at either day 10 or day 15. By day 15, males across all groups exhibited 70 – 90% survival.

2.4 Discussion

This study is one of the first direct assessments of the long-term survival benefits of manually holding fish into a current prior to release, despite the fact that this tactic is

routinely used by conservation-minded anglers and encouraged by many natural resource agencies (Pelletier *et al*. 2007). Contrary to our original predictions, our method of ventilation assistance did not increase the survival of sockeye salmon after the simulated capture-and-release event. For fish held in the peak water treatment, relatively rapid mortality was observed without significant differences among the simulated fisheries capture-and-release treatments. Fish held in the average water treatment exhibited a sex-specific mortality increase in response to our simulated fisheries capture and subsequent recovery tactic. We will discuss possible explanations for the physiological changes and survival patterns observed, as well as the implications for fisheries management.

The premise of using ventilatory assistance as a recovery tactic depends on the assumption that forcing water over the gill surface aids in oxygen uptake at a time when excess oxygen is needed to regain physiological homeostasis. Thus, in order for this technique to be effective, fish need to reach a state of metabolic impairment that requires subsequent physiological recovery. In the present study, analyses of the plasma variables clearly indicated that the simulated fisheries capture event was sufficient to metabolically impair fish (Table 2.1). Plasma cortisol, the principle corticosteroid measured in response to capture-and-release stressors, was significantly higher (roughly two-fold greater) for male and female fish that were subjected to the capture-and-release simulation in comparison to control fish. Moreover, the significant increase in glucose concentration for these fish is consistent with the mobilization of energy stores triggered by stress hormones (e.g., cortisol; Gamperl *et al*. 1994; Wendelaar Bonga 1997). Fish subjected to the simulated fisheries capture event (with or without assisted ventilation) exhibited lactate concentrations (~ 13 mmol L^{-1}) that are proposed to be characteristic of anaerobic activity during burst swimming

in sockeye salmon (i.e., concentrations greater than 6 mmol L^{-1}; Eliason Parsons 2011) and reached the level proposed as the threshold for negatively affecting repeat swim performance in rainbow trout (Jain and Farrell 2003). Our plasma lactate concentrations for these fish subjected to the capture-and-release simulation were six-fold greater than concentrations measured for fish in our control group (~ 2.1 mmol L^{-1}). These results cumulatively suggest that the fisheries capture simulation caused fish to mount a stress response and use anaerobic pathways that require subsequent excess oxygen uptake. Consequently, it is unlikely that the lack of enhanced survival in fish that received ventilation assistance was due to insufficient metabolic impairment of the fish post-capture.

We incorporated two ecologically relevant water temperatures into our study (16°C and 21°C) to assess if current warming trends observed in the Fraser River may affect the outcomes of recovery attempts after capture. Fish that were held at the current Fraser River peak water temperature (21°C) exhibited relatively rapid mortality, and there were no significant differences in mortality among all simulated fisheries capture-and-release treatments, including controls. Studies have shown that water temperatures greater than 18°C have negative survival consequences on sockeye salmon during their freshwater spawning migration (Naughton *et al*. 2005; Keefer *et al*. 2008; Martins *et al*. 2011). Suggested mechanisms responsible for this mortality include the depletion of energy stores (Rand *et al*. 2006), the collapse of aerobic scope and cardiovascular function (Farrell *et al*. 2008), and the increased progression of pathogens and disease (Crossin *et al*. 2008; Bradford *et al*. 2010). Regardless of the exact cause, the present results emphasize the deleterious effects of warming migration temperatures on Fraser River sockeye salmon and are particularly

relevant as Fraser River water temperatures are expected to continue to rise (Morrison *et al*. 2002).

At the 16°C water temperature, we found that the assisted ventilation tactic was ineffective in enhancing survival after our simulated fisheries capture and release. Elevated mortality rates were observed for female sockeye salmon subjected to the simulated fisheries capture and release in comparison to control females. Moreover, those females that received ventilatory assistance after capture exhibited a higher rate of mortality relative to females that did not receive assistance and control females. Other studies of Fraser River sockeye salmon have found that females suffer significantly (often two-fold) higher mortality than males when migration conditions are stressful (e.g., high water temperatures, high flows, excessive handling; Patterson *et al*. 2004; Crossin *et al*. 2008; Nadeau *et al*. 2010; Jeffries *et al*. 2011; Roscoe *et al*. 2011; Martins *et al*. 2012). Indeed, migrating female sockeye salmon naturally have higher plasma cortisol values in comparison to males, largely due to the role of cortisol in reproductive maturation (Sandblom *et al*. 2009; Hruska *et al*. 2010; Roscoe *et al*. 2011). This elevation of cortisol concentrations in females assists in mobilizing energy stores for gonad development (Mommsen *et al*. 1999; Jeffries *et al*. 2011). However, plasma cortisol acts as an immunosuppressant (Schreck *et al*. 2001) and females may therefore be more susceptible to infection (Martins *et al*. 2012). The additional stress of the handling in our experiment may have further compromised females.

Previous studies evaluating methods of enhancing survival of Pacific salmon using recovery devices suggest that any benefit of using these devices must be balanced with the potential for additional stress resulting from added handling and confinement, particularly for more vigorous fish (Farrell *et al*. 2001a; Donaldson *et al*. 2013; Nguyen *et al*. in press).

23

Thus, our attempt to manually restrain fish for 1 minute may have compounded the physiological stress that our approach was attempting to alleviate, particularly as elevated cortisol will prolong the removal of lactate and the re-synthesis of glycogen after anaerobic burst activity (Pagnotta *et al*. 1994; Eros and Milligan 1996; Milligan *et al*. 2000). However, considering our one sampling point and the dynamic process of the cortisol response trajectory (Barton 2002), we cannot determine if the extra handling required for this method of ventilation assistance resulted in a prolonged elevation of cortisol concentrations.

This study allowed us to evaluate the efficacy of this recovery tactic at two water temperatures within a controlled setting, removing some of the natural variables associated with migration through the Fraser River after release from capture (e.g., secondary capture encounters, hydraulic challenges). However, the restraint within the holding tanks results in confinement stress that these fish would not experience under natural conditions (Roscoe *et al*. 2011). Furthermore, this confinement can be particularly harmful for female sockeye salmon with naturally elevated cortisol levels (Mommsen *et al*. 1999; Jeffries *et al*. 2011). Thus, biotelemetry of wild fish offers an approach for determining the fate of fish released into their natural environment after fisheries capture and subsequent recovery techniques, and indeed this is the focus of ongoing studies within our research groups.

Most North American fisheries regulations recommend manual recovery techniques for fish intended to be released after capture (Pelletier *et al*. 2007). The manual ventilation assistance that we examined here was designed to simulate these attempts at recovery that would be implemented by Fraser River recreational anglers. While the method we assessed did not result in any obvious benefits under these experimental conditions, there is a need to expand this research of ventilation assistance before general recovery technique

recommendations can be made. The lack of clear support for the ventilation assistance described herein provides an example of both the need to test assumptions regarding universal best practices for handling sockeye salmon in moderate temperatures and the primary role of thermal stress driving survival at high temperatures. Managers may need to consider the implications of future climate scenarios that indicate an increase in frequency and duration of water temperatures exceeding 21°C for the Fraser River (Hague *et al*. 2011); particularly because thermal stress may overshadow any attempt at facilitating recovery that is associated with in-river fisheries
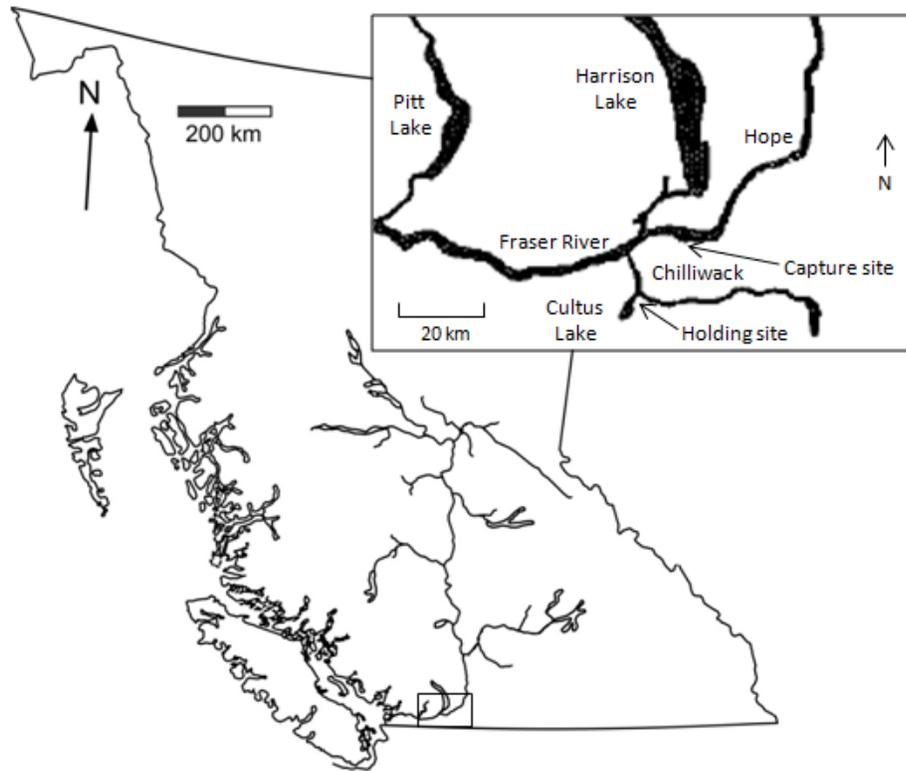
Figure 2.1 – Map of the Fraser River system in British Columbia, Canada. The inset shows the Fraser River capture site at Gill Road near Chilliwack and the holding site at the Fisheries and Oceans Canada (DFO) Cultus Lake Salmon Research Laboratory where experiments occurred.

Table 2.1 – Sample sizes (*n*), means, and standard errors of the means (S.E.M.) for blood physiological variables of sockeye salmon after one of three simulated fisheries capture-and-release treatments – control (C), no assisted ventilation (N), and assisted ventilation (A) – at two temperatures (16 and 21°C). Two-way ANOVA results are presented for each physiological variable. The superscript letters present the results of Bonferroni multiple comparisons tests. Dissimilar lower case letters denote significant differences between values, where A – D denotes differences with respect to significant treatment $\times$ temperature interactions and X – Z denotes significant differences with respect to the treatment main effect across both temperatures.

Physiological Variable	Fisheries Capture Treatment	*n*	16°C Mean ± SEM	*n*	21°C Mean ± SEM	Treatment *F*	*P*	Temperature *F*	*P*	Interaction *F*	*P*
Plasma lactate (mmol L^{-1})	C	18	1.65 ± 0.15 ^{a}	15	2.62 ± 0.38 ^{c}	250.1	<0.0001	0.9	0.3444	4.19	0.018
	N	18	13.38 ± 0.64 ^{b}	16	11.73 ± 0.98 ^{d}						
	A	19	13.06 ± 0.78 ^{bd}	16	14.11 ± 0.98 ^{b}						
Plasma cortisol (ng mL^{-1}) males	C	10	120.0 ± 19.6 ^{a}	10	162.8 ± 29.3 ^{a}	28.25	<0.0001	4.79	0.0335	4.71	0.0136
	N	10	387.5 ± 35.9 ^{b}	8	268.3 ± 15.3 ^{c}						
	A	10	350.5 ± 26.7 ^{b}	6	273.6 ± 34.4 ^{c}						
Plasma cortisol (ng mL^{-1}) females	C	8	301.0 ± 41.3 ^{x}	5	339.8 ± 64.0	13.84	<0.0001	0.07	0.7885	0.64	0.533
	N	8	531.0 ± 59.7 ^{y}	8	526.2 ± 57.1						
	A	9	586.1 ± 32.5 ^{y}	10	521.5 ± 24.1						
Plasma glucose (mmol L^{-1})	C	18	6.10 ± 0.34 ^{x}	15	7.06 ± 0.63	19.42	<0.0001	0.72	0.3988	0.51	0.6023
	N	18	9.33 ± 0.57 ^{y}	16	9.15 ± 0.70						
	A	19	9.70 ± 0.59 ^{y}	16	10.08 ± 0.47						
Plasma Na$^+$ (mmol L^{-1})	C	18	155 ± 3	15	152 ± 5	1.22	0.2986	3.84	0.0531	0.27	0.7673
	N	18	159 ± 3	16	154 ± 3						
	A	19	163 ± 3	16	154 ± 3						
Plasma Cl$^-$ (mmol L^{-1})	C	18	131.3 ± 1.3	15	131.3 ± 1.8	2.07	0.1314	0.07	0.794	0.03	0.97
	N	18	134.1 ± 1.3	16	133.3 ± 2.3						
	A	19	134.9 ± 2.1	16	134.5 ± 1.0						

Physiological Variable	Fisheries Capture Treatment	16°C		21°C		Treatment		Temperature		Interaction	
		n	Mean ± SEM	n	Mean ± SEM	F	P	F	P	F	P
Plasma K$^+$ (mmol L^{-1})	C	18	1.1 ± 0.1 ^{x}	15	1.4 ± 0.2	17.36	<0.0001	11.77	0.0009	1.66	0.1955
	N	18	0.4 ± 0.04 ^{y}	16	0.8 ± 0.1						
	A	19	0.6 ± 0.1 ^{y}	16	0.8 ± 0.1						
Plasma osmolality (mOsm kg^{-1})	C	18	316 ± 2 ^{x}	15	314 ± 3	33.7	<0.0001	0.15	0.6981	0.62	0.5388
	N	18	349 ± 3 ^{y}	16	348 ± 3						
	A	19	343 ± 6 ^{y}	16	350 ± 4						
Hematocrit (%)	C	18	41.1 ± 0.9 ^{x}	15	41.7 ± 1.4	23.05	<0.0001	4.19	0.0434	1.35	0.2652
	N	18	44.6 ± 1.5 ^{y}	16	49.9 ± 2.1						
	A	19	50.0 ± 1.0 ^{z}	16	52.0 ± 1.6						
Hemoglobin (g L^{-1})	C	18	102 ± 2 ^{x}	15	104 ±3	4.56	0.0128	6.41	0.013	2.85	0.0628
	N	18	98 ± 3 ^{xy}	16	112 ± 4						
	A	19	110 ± 2 ^{y}	16	112 ± 3						
Mean corpuscular hemoglobin concentration (MCHC; g L^{-1})	C	18	249 ± 3 ^{x}	15	252 ± 6	34.57	<0.0001	0.09	0.7641	0.74	0.4816
	N	18	221 ± 3 ^{y}	16	226 ± 4						
	A	19	222 ± 4 ^{y}	16	217 ± 4						
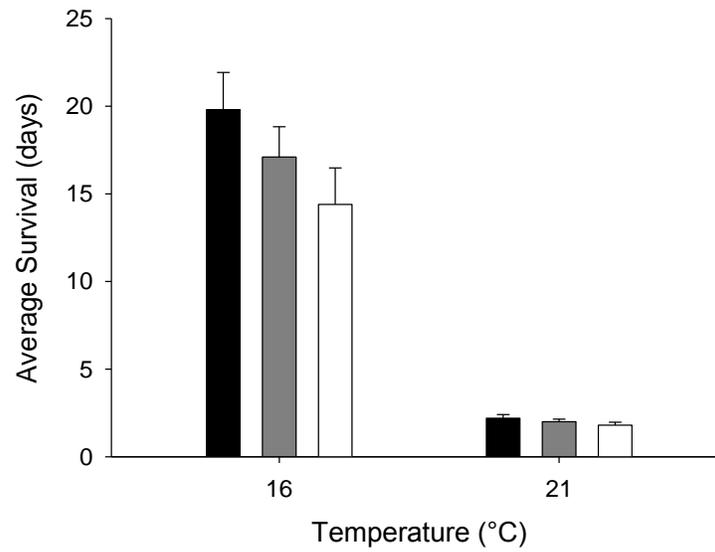
Figure 2.2 – Average days survived after experimental treatment – control (black bars), fisheries capture simulation without assisted ventilation (grey bars), and fisheries capture simulation with assisted ventilation (white bars) – for sockeye salmon held at 16°C and 21°C, with standard error bars ($n = 15 - 19$).
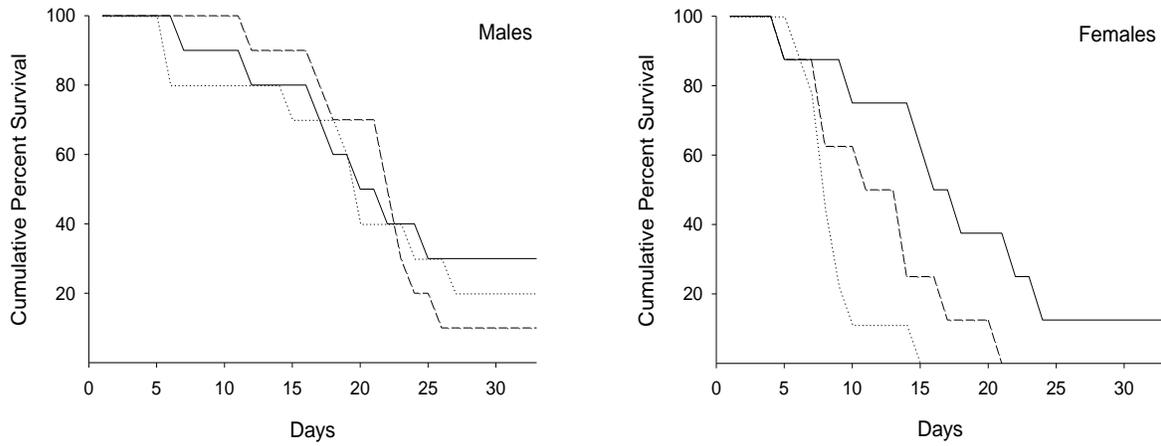
Figure 2.3 – Cumulative percent survival after experimental treatment for male and female sockeye salmon held at 16°C ($n = 8 - 10$). Seven fish (6 males, 1 female) remained at the endpoint of the experiment (33 days). Line type indicates simulated fisheries capture treatment – control (solid line), fisheries capture simulation without assisted ventilation (dashed line), and fisheries capture simulation with assisted ventilation (dotted line).
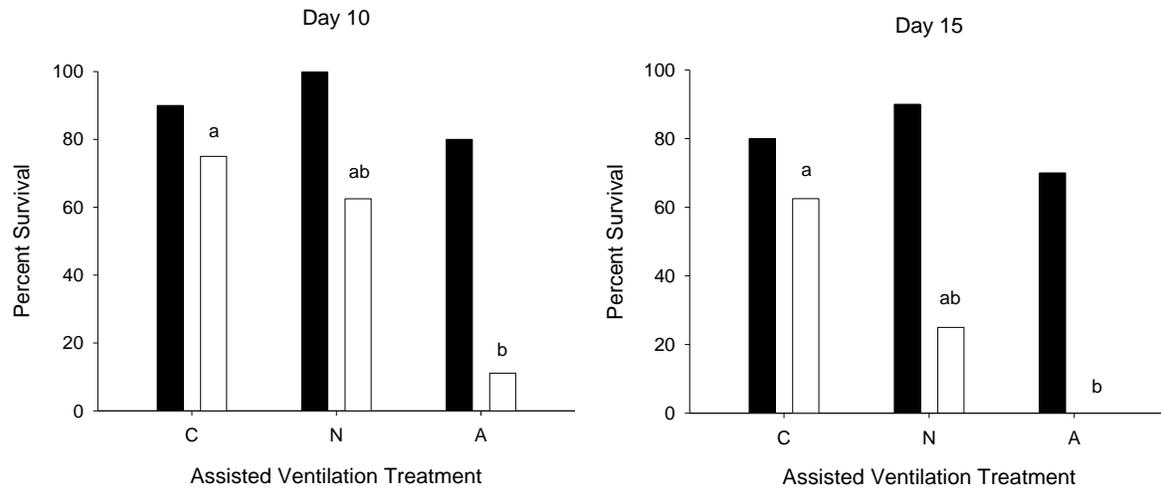
Figure 2.4 – Percent survival 10 and 15 days after experimental treatment (C = control, N = no assisted ventilation, A = assisted ventilation) for males (black bars; n = 10) and females (white bars; n = 8 – 9) held at 16°C. Different lower-case letters indicate significant differences between simulated fisheries capture-and-release treatments after Bonferroni corrections for multiple comparisons. Significant differences were not detected among male treatment groups.

Chapter 3: Influence of assisted ventilation on migration behaviour and survival of adult sockeye salmon after capture and release in fresh water[2]

3.1 Introduction

The live release of captured fish is commonly employed world-wide on many fish taxa (Cooke and Suski 2005; Arlinghaus *et al*. 2007). The motivation for release may be based on ethical and voluntary grounds, or may be a requirement based on mandated harvest restrictions (Cowx 2002; Cooke and Suski 2005; Arlinghaus *et al*. 2007). Mandated live-release is used to minimize fishing pressure on a population of fish (Policansky 2002; Cooke and Schramm 2007). However, the success of this conservation and management tactic is contingent on the assumption that the released fish survive to reproduce or, at minimum, the rates of survival following release are known (Wydoski 1977). Studies on recreational catch-and-release fisheries have demonstrated that capture can result in delayed mortality of released fish and, in some cases, mortality following release can be extremely high (range 0 – 95%; mean 18%; Bartholomew and Bohnsack 2005). Delayed mortality can arise from physical injury and/or the inability of the fish to regain physiological homeostasis (Wood 1983; Chopin and Arimoto 1995). Clearly, approaches that can reduce delayed mortality can benefit live-release management efforts.

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[2] A version of this chapter will be submitted for publication. Robinson, K.A., S.G. Hinch, G.D. Raby, M.R. Donaldson, S. Tyerman, D. Robichaud, D.A. Patterson and S.J. Cooke. Influence of assisted ventilation on migration behaviour and survival of adult sockeye salmon after capture and release in fresh water.

The physiological changes that occur from capture stressors can be best described in the context of the burst swimming events that are provoked during attempts to evade capture or escape landing. Burst events are fueled by the anaerobic metabolism of glycogen resulting in the accumulation of metabolites (e.g., lactate and protons) which will ultimately alter the acid-base status and cause ion-osmoregulatory imbalance (Wood 1991; Kieffer 2000). The depletion of glycogen reserves and the accumulation of lactate during anaerobic breakdown will inhibit repeat burst swimming events (Milligan 1996), which can leave fish vulnerable to predation or recapture and more likely to fall back downstream when released into a fluvial system. Thus, the ability of impaired fish to recover from these physiological alterations within an ecologically relevant timeframe will influence post-release behaviour and survival. The resynthesis of energy stores and the removal of anaerobic metabolites occur during metabolic recovery from fisheries capture (Wood 1991). This recovery process requires oxygen uptake that exceeds basal metabolic rates (termed excess post-exercise oxygen consumption (EPOC); Gaesser and Brooks 1984). Efforts to reduce the physiological impact fisheries have on released fish by promoting metabolic recovery may help to mitigate the capture-related mortality and sublethal impacts.

Considering the negative consequences of releasing physiologically impaired fish, the ability to enhance recovery to minimize capture-related mortality is important for meeting live-release management objectives. Indeed, managers inform recreational fishers of recommended landing and handling techniques that can be used to minimize playing and handling time, avoid unwarranted physically injury, and eliminate air exposure (Cooke and Suski 2005; Pelletier *et al*. 2007). Even more, a number of North American natural resource agencies currently recommend the use of manual recovery techniques prior to release.

Examples of these techniques include orienting the fish into a water flow (e.g., in a fluvial system) or moving the fish back and forth in the water (Pelletier *et al*. 2007), both of which attempt to facilitate the flow of water over gill surfaces. This underlying recovery foundation seems intuitive given that the physiological changes inherent with capture events are rectified using excess oxygen consumption (Wood 1991); however, an adequate evaluation of this fundamental technique is lacking.

Adult migrating sockeye salmon (*Oncorhynchus nerka*) are often released from capture in the Fraser River (British Columbia, Canada). Each fall, the Fraser River and its tributaries are home to millions of adult sockeye salmon returning from the ocean to begin their freshwater migration to natal spawning streams (Hinch *et al*. 2006). The predictable nature of this life history event and the economic, social, and cultural value that these fish have for British Columbians make them targets for fishers during their approach and upon entry into the Fraser River. Within the freshwater environment, sockeye salmon are targeted by a growing number of recreational fishers (Kristianson and Strongitharm 2006). Fisheries and Oceans Canada (DFO) manages this pressure by implementing harvest restrictions (i.e., no retention mandates or daily catch limits) in an effort to achieve sockeye salmon escapement targets or reduce the fishing pressure on a co-migrating population of concern. Sockeye salmon are also voluntarily released by anglers. In 2011 alone, freshwater recreational anglers released approximately 63,000 of the estimated 145,000 sockeye salmon captured (43% released; DFO 2012b).

Recent studies have shed light onto the delayed mortality associated with these fisheries events (Donaldson *et al*. 2011; Martins *et al*. 2012; Raby *et al*. 2012). Research using biotelemetry to evaluate post-release survival of Fraser River sockeye salmon has

indicated that recreational angling can reduce survival to natal tributaries by approximately 35% (Donaldson *et al*. 2011). However, recent field studies have indicated that the post-release survival of sockeye salmon can be enhanced under certain circumstances by facilitating recovery with a flow-through recovery bag (Donaldson *et al*. 2013). This recovery device allows for the flow of water over the gills of the fish while isolating the fish within a safe environment. In Chapter 2, I expanded on the results of this research by evaluating a manual recovery technique designed to assist ventilation by physically orienting the fish into a high flow water source. This recovery technique mimics the methods of recovery that recreational anglers may use upon the recommendation of local managers (Pelletier *et al*. 2007). However, in the laboratory study (Chapter 2), we found no benefit of this technique. In fact, we observed a relative increase in female mortality rates in response to the assisted ventilation, which we attributed to the increased sensitivity of female sockeye salmon to the stress of long-term laboratory confinement. Therefore, as a result, the current study attempts to evaluate this particular recovery technique for fish released into their natural environment.

The purpose of our study was to determine the influence of post-capture ventilation assistance on the migration behaviour and survival of Fraser River sockeye salmon released to resume their freshwater spawning migration. We assessed the effects of this recovery tactic using biotelemetry in two field experiments. The first experiment exposed sockeye salmon to a riverside capture and release that simulates angling and the second experiment released sockeye salmon after capture by rod-and-reel anglers. In both experiments, a group of captured fish received ventilation assistance prior to release. The simulated capture experiment evaluated migration behaviour and successful arrival to natal spawning grounds

of two populations in the Harrison River system (a tributary of the Fraser River), whereas the angling experiment evaluated migration times and survival to reach the upmost receivers station in the Fraser River system. For both experiments, we predicted that the ventilation assistance would promote the metabolic recovery from capture and thus reduce behavioural alterations and delayed mortality upon release.

3.2 Methods

3.2.1 Harrison River experiment

3.2.1.1 Experimental site and fish capture

This experiment was conducted on Weaver Creek and Harrison Rapids sockeye salmon during their migration up the Harrison River, British Columbia, Canada to reach natal spawning areas. Weaver Creek sockeye salmon spawn in Weaver Creek, a tributary of the Harrison River, or in an artificial spawning channel ~ 1 km up the creek. Harrison Rapids sockeye salmon spawn in the near shore gravel that lines the Harrison Rapids channel in the middle reaches of the Harrison River (Schaeffer *et al*. 1951; Donaldson *et al*. 2012). Peak spawning period for these populations is approximately October 20 and November 15, respectively (Gilhousen 1990).

There are regulated fishery openings for Pacific salmon in the Harrison River. During this experiment, the First Nation fishery sector harvested sockeye salmon in the economic opportunity fishery and for food, social and ceremonial (FSC) purposes during openings in August and September (DFO 2012c, 2012d). Recreational retention fishing for sockeye salmon in the lower reaches of the Harrison River was open August 13 to September 18

(DFO 2012b). Two of our study fish were captured in the economic opportunity fishery and reported to researchers.

Fish capture and treatment were conducted on the Harrison River over 5 days (August 23, August 29, September 1, September 21, and September 22) in 2011. Hourly river temperature readings were recorded during treatment using a temperature logger (TidbiT v2, Onset Computer Corporation Inc., Bourne, MA) that was deployed across from the capture site. The river temperature during the hours of capture and experimental treatments was 14.96 ± 0.14°C (mean ± S.E.M.). Adult sockeye salmon were captured by beach seine at the site indicated in Figure 3.1 (see Harrison release site). Seining was accomplished using a mesh seine net with one end anchored on shore and the other pulled to the centre of the river then arced closed with a power boat, forming a circular area of containment. The seine was drawn in from both ends to concentrate fish close enough to shore (while maintaining sufficient water depth to minimize unnecessary stress and crowding) to allow for dip-netting and removal of fish.

Control fish were immediately placed inside a cylindrical fish bag for processing. This black hypolon fish bag (100 cm length x 20 cm diameter) is constructed with mesh ends to allow for a continuous flow of fresh river water (Donaldson *et al*. 2011, 2013). During processing, a blood sample was taken, a scale was removed, a fork length measurement was recorded, and a radio transmitter was gastrically inserted (see below). Control fish were released directly from the bag following these procedures. As controls were processed, treatment fish were dip-netted from the seine and transferred to an in-river holding pen (1.0 m x 2.5 m x 1.0 m deep) with mesh ends providing a constant flow of fresh river water. A maximum of 11 fish were used per seine to minimize the duration spent in the holding pen.

Fish were held in the pen for < 45 min. Treatment fish were dip-netted from the holding pen and placed in a flow-through, foam-lined trough to complete sampling and tagging procedures (as described above for control fish). These fish were then transferred to a riverside tank for treatment.

3.2.1.2 Blood sampling, stock identification and tagging procedures

In consideration of the sex-specific mortality patterns observed in Chapter 2, blood samples were collected to determine the sex of study fish. Samples of ~ 2 mL were attained using caudal venipuncture with a 21-guage needle and a heparinised vacutainer (detailed in Cooke *et al*. 2005). Whole blood was centrifuged (7,000 x g) for 5 min and plasma samples were stored in 1.5 mL cryogenic vials in liquid nitrogen prior to storage at -80°C. Plasma was analyzed for testosterone and 17 B-estradiol (Neogen ELISA with Molecular Devices Spectramax 240pc plate reader).

A scale sample was obtained from all fish in order to determine stock complexes. As a result, the populations were not identified until after treatment and release. The scale analysis determined that 102 Harrison Rapids and 40 Weaver Creek sockeye salmon were used in this experiment.

All fish had an individually-coded radio transmitter (Pisces, Sigma Eight Inc., Newmarket, ON) gastrically inserted by holding the fish supine with its head just out of the water and pushing the transmitter down the esophagus using a smooth plastic plunger (Ramstad and Woody 2003; Cooke *et al*. 2005). The transmitters were 16 mm in diameter, 46 mm long with a 460 mm long antenna. The tags transmitted on the 150 MHz band on six frequencies (320, 360, 440, 460, 600, 800 KHz) with pulse intervals of 5.5 s. These coded

transmitters allowed us to identify individual fish as they were detected at receiver stations or during manual tracking (explained below).

3.2.1.3 Experimental design

Three treatment groups were established: (i) control (25 females (14 Harrison, 11 Weaver) and 17 males (13 Harrison, 4 Weaver)), (ii) simulated capture without assisted ventilation (25 females (20 Harrison, 5 Weaver) and 27 males (20 Harrison, 7 Weaver)), and (iii) simulated capture with assisted ventilation (23 females (14 Harrison, 9 Weaver) and 25 males (21 Harrison, 4 Weaver). This treatment protocol was developed to extend the laboratory-based evaluation of assisted ventilation as a technique for facilitating recovery (Chapter 2) to a field-based telemetry study. To this end, a doughnut-shaped tank (~ 800 L; 2 m diameter) was set up riverside for fisheries capture-and-release simulations consisting of 3 min of strenuous exercise and 1 min of air exposure of two fish at a time. Exercise was stimulated by three experimenters leaning over the edge of the tank and touching the tail or splashing behind the fish to elicit burst swimming. After 3 min, fish were then immediately dip-netted from the tank and exposed to 1 min of air. There was no physical injury component in this simulation, though injury is typical of capture events and can result in direct and indirect (e.g., disease and predation) mortality (Chopin and Arimoto 1995). Following each round of fisheries capture simulation, one fish was released and the other was subjected to ventilation assistance. Comparable to the protocols of Chapter 2, the mouth of the fish was oriented into a jet of river water (held ~ 20 cm from the jet nozzle) from a submersible pump. The fish was supported with one hand around the caudal peduncle and the other on the ventral surface, just posterior to the pectoral fins. The flow speed was ~ 0.50 m s^{-1} (as measured at the mouth), consistent with the speed evaluated in Chapter 2.

Ventilation was assisted for 1 min unless the fish became vigorous prior to cessation, at which time the fish was released early to minimize additional stress. Seven of the 48 fish subjected to the ventilation assistance were released prior to treatment completion. Opercular beats were observed during assisted ventilation, indicating successful forced ventilation.

3.2.1.4 Radio-tracking and determination of fate

The coded radio transmitters were detected using five fixed radio telemetry receiver stations (SRX400, Lotek Wireless Inc. Newmarket, ON) with 3-, 4-, or 5-element Yagi antennas. Four stations were setup on the Harrison River, two downstream of the release site and two upstream of the release site (see Figure 3.1). The downstream receiver nearest the release site was 2.5 km away and the nearest upstream receiver was 1.5 km away (distances in river kilometers). The fifth station was set up on Weaver Creek, ~ 250 m upstream of the mouth. In addition, manual tracking by boat occurred throughout the study to supplement the fixed-station array.

Survival was assessed as arrival to natal spawning grounds. Weaver Creek sockeye salmon were recorded as successful arrivals on spawning grounds if they were detected at the Weaver Creek receiver (station 6 in Figure 3.1). For Harrison Rapids sockeye salmon, the nearest downstream receiver (station 3) and the nearest upstream receiver (station 4), relative to the release site, flank their natal spawning area. Successful arrivals of Harrison fish on spawning grounds were assessed as fish exhibiting movement within the spawning area on or after October 20, 2011 (for reasons outlined in Donaldson *et al*. 2012). Manual tracking allowed us to confirm movement and location, and thus successful arrivals to natal spawning areas.

3.2.1.5 Data analysis and statistics

Significance levels were set at 0.05. Pearson's chi-squared analysis was used to test for differences in overall and population-specific post-release survival to reach spawning grounds among the three treatment groups. Where significant differences were detected among simulated fisheries capture-and-release treatments, Bonferroni multiple comparisons tests were used. Differences in survival to reach spawning grounds between male and female fish of each treatment group were assessed using Fisher's exact tests with Bonferroni multiple comparisons corrections used.

3.2.2 Fraser River experiment

3.2.2.1 Experimental site and fish capture

Both the experimental site and angling method used were identical to those in Donaldson *et al.* (2011). All capture and tagging occurred at Grassy Bar (49°10'0.20" N, 122°1'9.14" W) on the Fraser River mainstem near Chilliwack, British Columbia, Canada from August 15 to September 2, 2011. The daily mean water temperature during the capture and tagging period was 16.7 ± 0.1°C (mean $\pm$ S.E.M.) while the daily mean water discharge was 4413 ± 71 m^3 s^{-1} (mean $\pm$ S.E.M.). The Fraser River discharge in August was ~ 35 – 40% above normal for that time of year (e.g., normal for August is ~ 3000 m^3 s^{-1}; Morrison *et al.* 2002). The water temperatures were relatively low for August, the month during which the Fraser River normally reaches its annual peak temperatures. Temperature and discharge data for the Fraser River were obtained by DFO's Environmental Watch Program using monitoring stations on the mainstem of the Fraser River at Qualark (49° 31'58.13" N, 121° 25'20.67" W) and Hope (see Figure 3.1), respectively.

Adult sockeye salmon ($n = 70$) were caught by volunteer anglers using standard "bottom-bouncing" gear designed to target this species. The angling method uses long (> 3 m) leaders with barbless J-shaped hooks sized 1 to 3/0 and a heavy metal weight. The weight bounces along the riverbed and suspends the hook in the water column. Duration from hooking to landing ranged from $1 - 5$ min, but was ≤ 2 min in 82% of the cases. Fish were landed using a knotless nylon landing net in which the fish were unhooked and then transferred into cylindrical fish bags (see description above in Harrison River study) for processing. This resulted in $15 - 45$ s of air exposure.

3.2.2.2 Post-angling treatment, tagging and release

Once in the fish bag, each fish was randomly assigned to one of three treatment groups: (i) tagging and release, (ii) 1 min additional air exposure, tagging and release, or (iii) 1 min additional air exposure, tagging, 1 min assisted ventilation (see below), and release. Tagging consisted of gastrically inserting radio transmitters as described in the Harrison River experiment (see above). Air exposure (when applicable) occurred immediately after capture by lifting the fish bag completely out of the water and allowing the water in the bag to quickly drain through the mesh ends. After transmitter insertion, a small clip of tissue was removed from the adipose fin using a hole punch and stored in 95% ethanol for stock identification via laboratory analysis (see Beacham *et al*. 2005). Unlike the Harrison River experiment, blood was not sampled from these study fish. Each fish was measured (fork length), and rapidly assessed for reflex impairment (in < 10 s; see Raby *et al*. 2012, data not reported here).

Once tagging and processing were complete, fish were either released (groups 1 and 2), or provided with the assisted ventilation treatment (group 3). This ventilation assistance was carried out as detailed in the Harrison River study (see above). Fish were oriented to face into the river flow (~ 0.5 m s^{-1} water speed) and held in this position for 1 min. If the fish became vigorous and struggled against restraint during treatment, it was released. All 23 fish that received the assisted ventilation treatment were held in the flow for 1 min. Once the time elapsed, the handler simply let go of the animal and in every instance the fish swam away in the upstream direction. All fish exposed to the revival technique were ventilating while being held into the river flow.

3.2.2.3 Radio-tracking and determination of fate

Transmitters were individually-coded and used seven frequencies (320, 360, 440, 460, 500, 600, 800 KHz) on the 150 MHz band to reduce signal collisions as fish passed receiving stations (see below). All fish were tracked using an array of seven receiver stations on the mainstem of the Fraser River and an additional four on the Harrison River (including the station on Weaver Creek; see Figure 3.1). The downstream receiver nearest to the release site was 22 km away at Mission (station 1); the nearest upstream receiver was 9 km away at the junction of the Harrison and Fraser rivers (station 2; distances in river kilometers). Short-term survival was estimated for each fish based on whether the fish stopped migrating upstream within 72 h of release. Fish were assessed as successful migrants (i.e., survivors to upmost receiver station) if they were detected at the most upstream receiver station on the migration pathway towards their DNA-identified natal spawning area (see stock composition in Table 3.2). Based on this information, the receiver at the junction of the Thompson and Fraser rivers (station 10) would be the terminal detection station for 58 of the 70 fish tagged.

An additional 10 fish would have to pass the receiver at the junction of the Nicola and Thompson rivers (station 11) to successfully reach natal spawning areas. One fish was identified as belonging to a Harrison River system population (Birkenhead). DNA analysis was not completed for one fish so it was assumed that its population identification was reflected in its migration pathway (explained in the results). In order to compare migration times among treatment groups, we used time from release to first detection at upstream receivers (in hours). For analyses, we focused on time to reach five upstream receivers for which sufficient sample sizes were attained: Harrison River, Hope, Sawmill Creek, Hell's Gate, and Thompson River (stations 2 and 7 – 10, respectively; Figure 3.1).

In order to facilitate reports of captured and harvested study fish, the tagging study was widely advertised with an associated reward for reporting and returning radio tags. The radio tag return program has been active on the Fraser River for 10 years and there is an awareness of the program in the angling community. Fish that were recaptured and reported by anglers were included in survival estimates, accounting for the location and time of recapture. For example, one fish was recaptured and euthanized by an angler 200 m upstream of the release site on the day of treatment. This fish was not included in the analyses. Other fish were captured and reported near spawning areas in the upper watershed beyond the terminal receivers; these fish were assessed as successful migrants.

3.2.2.4 Data analysis and statistics

We compared short-term (72 h) survival and survival to upmost receiver station among the three treatment groups using Pearson's chi-squared test. For analyses of migration times, we first evaluated whether fish size (fork length) influenced time to reach any of the

five upstream receivers using Spearman rank correlation. There was no significant correlation between fish size and migration times ($P > 0.05$ for all five receivers). Therefore, migration times were subsequently analyzed as the number of hours from release to each of the five upstream receivers. A Kruskal-Wallis test was then used to compare migration times (in hours) among the three treatment groups to each of the five upstream receivers. Finally, we evaluated whether fish that migrated more quickly after release were more likely to complete their migrations. To do so, migration times to the Harrison River and Hope receivers (stations 2 and 7 in Figure 3.1, respectively) were compared using Kruskal-Wallis tests between fish that did and did not ultimately reach the upmost receiver station.

3.3 Results

3.3.1 Harrison River experiment

Two Harrison Rapids sockeye salmon were confirmed captured in the Harrison River First Nation sockeye salmon economic opportunity fishery on August 24, 2011. Five tags were not detected at a fixed station receiver; however, two of these tags were picked up near the release site using manual tracking. They were confirmed to be mortalities. The three tags that were not detected by fixed stations or manual tracking were considered to be tag malfunctions, and were removed from the data set.

3.3.1.1 Survival to reach spawning area

In total, 25 of the 142 (17.6%) sockeye salmon that were released survived to reach their natal spawning area. Treatment had a significant effect on the survival of sockeye salmon to reach spawning areas ($\chi^2 = 26.727$, df = 2, $P < 0.001$). Eighteen of 42 (42.9%) control fish reached the spawning area, whereas only 5 of 52 (9.6%) fish and 2 of 48 (4.2%)

fish in the groups without ventilation assistance and with ventilation assistance were successful, respectively. There was no significant difference in survival to reach spawning areas between the group that received assisted ventilation and the group that did not ($P = 0.439$).

Comparable percentages of total Harrison Rapids and Weaver Creek sockeye salmon reached spawning areas (18.6% and 15.0%, respectively; Table 3.1). Treatment had a significant effect on the survival of Harrison Rapids sockeye salmon to reach spawning areas ($\chi^2 = 21.338$, df = 2, $P < 0.001$). Harrison fish that were immediately released following capture by beach seine (i.e., controls) were more likely to reach spawning areas (13 of 27 fish [48.1%]) relative to those subjected to the simulated capture-and-release event (with or without assisted ventilation; 2 of 35 fish [5.7%] and 4 of 40 fish [10.0%], respectively). A similar significant effect was observed for Weaver Creek sockeye salmon ($\chi^2 = 6.667$, df = 2, $P = 0.036$). Immediately released Weaver fish exhibited a greater likelihood to reach spawning areas (5 of 15 fish [33.3%]) relative to fish that endured the simulated capture-and-release event (with or without assisted ventilation; 0 of 13 fish [0.0%] and 1 of 12 fish [8.3%], respectively). For both the Harrison Rapids and Weaver Creek stocks, there was no significant difference in survival between fish that did not receive ventilation assistance and those that did ($P > 0.05$ in both cases).

A trend in sex-specific differences in survival to reach spawning areas was detected (Figure 3.2). Of the females that were subjected to the simulated capture-and-release event, zero survived to reach spawning areas, whereas 9 of 25 (36.0%) female control fish survived. Overall, the trend of reduced female survival relative to males of corresponding treatment groups was not statistically significant ($P > 0.05$ in all cases).

3.3.1.2 Migration behaviour

Sixty-six Harrison Rapids (21 of 27 [81.5%] controls, 26 of 40 [65.0%] without assisted ventilation, 19 of 35 [54.3%] with assisted ventilation) and all of the Weaver Creek sockeye salmon (40 fish) swam upstream following release and were detected at the most upstream Harrison Lake receiver (station 5). Of these fish, 19 (28.8%) and 6 (15.0%) survived to reach spawning areas, respectively. The remaining fish were first detected at the downstream station nearest the release site (31 Harrison fish) or died close to the release site prior to detection (5 Harrison fish). Nine of the 31 Harrison fish that were first detected downstream of the release station then swam to the most upstream station; however, these fish did not survive to reach spawning areas.

3.3.2 Fraser River experiment

The one fish with "unknown" population origin (DNA analysis was not conducted; Table 3.2) moved through the Harrison River system within 2 days and was last detected at the upmost Harrison River receiver (station 5 in Figure 3.1). That fish was assessed as having ultimately survived with the assumption that it was from a Harrison-Lillooet River system population. One fish in the angling plus additional air exposure group was not tracked (see Table 3.2) because the tag information was not recorded prior to release. Two fish were detected downstream at Mission (station 1) and did not subsequently move upstream. These fish were deemed mortalities.

Across all treatment groups, short-term survival (72 h) was 88.7% while survival to the upmost receiver station was 73.3%. Survival was not significantly different among the three treatment groups for short-term survival ($\chi^2 = 0.10$, df = 2, $P = 0.95$) or survival to

upmost receiver station ($\chi^2 = 0.18$, df = 2, $P = 0.91$; Table 3.2). Migration times (in hours) to reach any of the five upstream receivers were not significantly different among the three treatment groups ($P > 0.35$ in each case). Short-term post-release migration times (hours from release to the Harrison River (station 2) and Hope (station 7) receivers) did not differ significantly between those fish that reached the upmost receiver station and those that did not ($P > 0.05$ in both cases).

3.4 Discussion

By assisting the flow of water over the gills of fish during EPOC, we intended to facilitate the recovery of these fish prior to release from capture. However, this study demonstrated that a simple, yet practical approach to manually assisting the ventilation of migrating sockeye salmon after freshwater capture did not affect subsequent migration behaviour or survival in either of our field experiments. The results of the Fraser River experiment indicated that there was no significant effect of assisted ventilation treatment on either migration times or survival rates. In the Harrison River experiment, we found that the capture simulation resulted in a clear reduction in survival to reach natal spawning grounds; however, we did not detect an effect of our ventilation assistance technique. To our knowledge, this is the first study to use biotelemetry to evaluate this manual recovery technique that assists ventilation using a method that is comparable to recreational angler recovery attempts in which they orient the fish into the flow of a fluvial system.

Overall, in the Harrison River experiment, fish exposed to the simulated fisheries capture and release exhibited an overall reduction of ~ 30% in survival to reach natal spawning grounds. Previous research evaluating the delayed mortality of angling and release

has been conducted using volunteer anglers for capturing migrating sockeye salmon from the same release site as our Fraser River experiment (Donaldson *et al*. 2011). Unlike our experiment, Donaldson et al. (2011) used a more extensive radio receiver array that monitored post-release survival to natal sub-watersheds, thus accounting for even more of each fish's migration. As a result, the authors were able to compare their observed long-term survival to an estimated level of natural survivorship to spawning areas (~ 70%; Martins *et al*. 2011) and thus calculate an approximate 35% mortality attributable to angling and release. This value is similar to the survival reduction detected in the Harrison River experiment.

The present examination of assisted ventilation used the same recovery technique that was evaluated in our earlier laboratory experiment (Chapter 2). The results from the present field experiments are consistent with Chapter 2 in that we did not detect a benefit of attempting to facilitate recovery from capture using ventilation assistance. From the laboratory experiment we suggested that this may be the result of stress associated with the additional handling inherent of manual recovery techniques. Furthermore, we speculated that this additional stressor may be particularly harmful for female sockeye salmon as we found reduced survival rates of females that received assisted ventilation. However, this may be in part the result of the chronic stress that is associated with laboratory confinement (Pickering and Pottinger 1995) and thus we proposed evaluating this recovery technique upon release into a natural environment. Thus, our field-based experiments of this chapter compliment the lab-based experiment of Chapter 2. In our Harrison River experiment, we found a trend of increased female mortality after exposure to simulated fisheries capture and release; however, we did not detect a difference in female mortality for those fish that received the

assisted ventilation treatment relative to those that did not. This result could be explained by the observation that no females that were exposed to simulated capture and release survived to reach natal spawning grounds. In consideration of this, our long-term endpoint assessment may not have been sensitive to detecting survival differences, particularly as the results of Chapter 2 were evaluated at 10 and 15 days post-treatment. Overall, in both chapters, for male and female sockeye salmon that received the assisted ventilation treatment, there was no benefit observed.

The capture-related reduction in survival observed in the Harrison River experiment reaffirms the need for a means of mitigating the mortality associated with fisheries capture events. However, as discussed, the current study provides evidence that a simple means of attempting to facilitate the recovery of sockeye salmon prior to release from capture fails to affect fish migration behaviour or survival. Considering that the technique evaluated here uses a post-capture ventilation assistance method and duration that is comparable to techniques recommended to recreational anglers by local managers, this result raises questions as to the efficacy of this attempt to facilitate recovery. However, irrespective of attempts to recover fish, these results indicated that delayed mortality of sockeye salmon released from this freshwater recreational fishery is likely greater than the mortality rate currently used in management models (i.e., 3%, DFO 2011), and the level of mortality may be higher for released females. This implies that in order for managers to adjust for the effects of the sockeye salmon recreational fishery sector on escapement targets or populations of concern, they may need to limit fishery openings to curtail capture-induced mortality.

Research should continue to evaluate case-specific attempts at reducing the negative effects of capture and release. Previous research has evaluated methods of facilitating recovery using devices such as Fraser Boxes (Farrell *et al*. 2001a; Nguyen *et al*. in press) and recovery bags (Brownscombe *et al*. 2013; Donaldson *et al*. 2013). These recovery devices have shown context-specific benefits in promoting physiological recovery (Farrell *et al*. 2001a) and reducing capture-associated delayed mortality (Farrell *et al*. 2001a; Donaldson *et al*. 2013). However, these studies have suggested that the extent to which fish are exhausted as a result of the fisheries capture event may determine the potential effectiveness of recovery techniques. For example, more vigorous fish may not benefit from recovery, whereas more impaired fish may reap the potential benefits of increasing oxygen available for uptake during EPOC (Farrell *et al*. 2001a; Donaldson *et al*. 2013; Nguyen *et al*. in press). If this is the case, then methods of evaluating the extent to which captured fish are impaired and predicting post-release mortality could be valuable to the success of attempts to facilitate recovery (e.g., using reflex action mortality predictors (RAMP); Raby *et al*. 2012). Future studies may be able to shed more light onto recovery attempts by evaluating levels of capture-related impairment, thereby reducing the unnecessary handling stress on more vigorous fish.
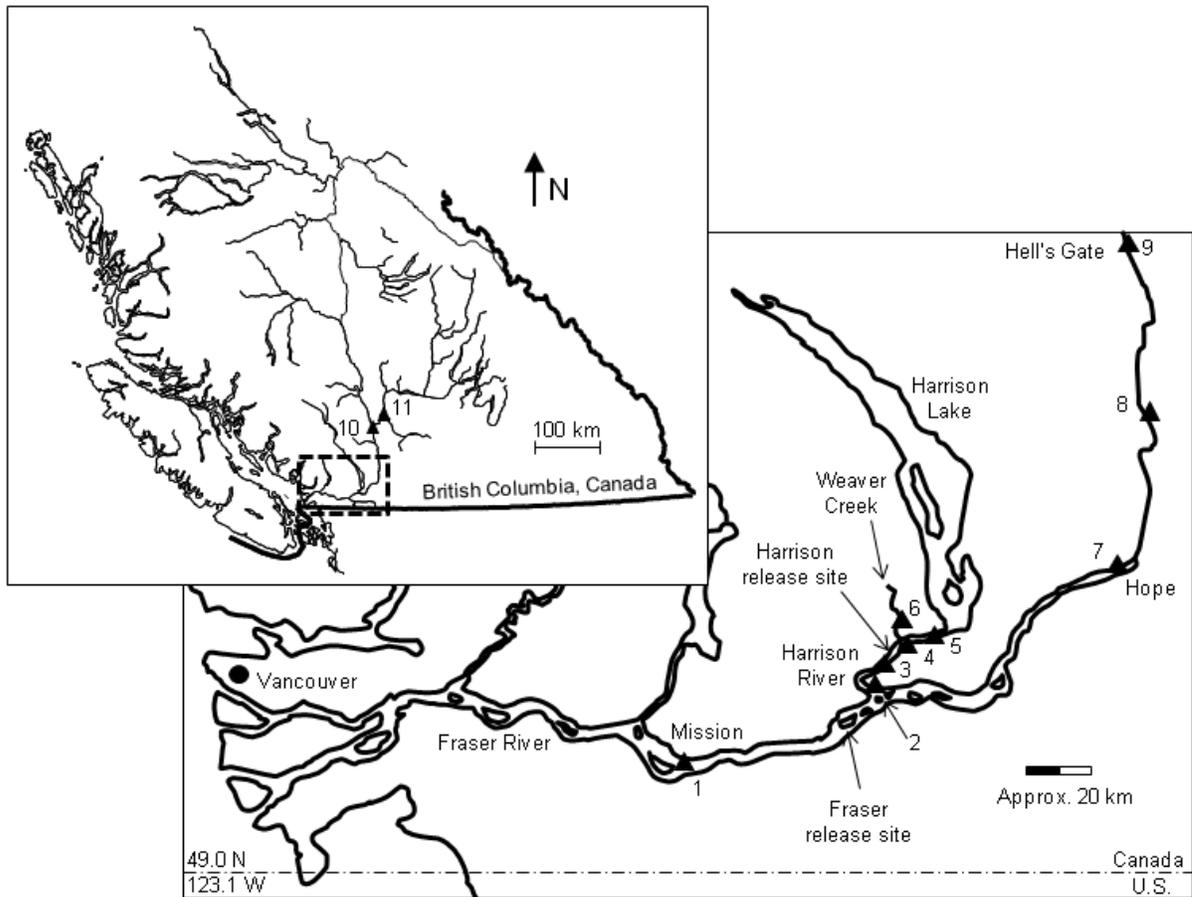
Figure 3.1 – Map of the Fraser River system in British Columbia, Canada. The inset shows the Harrison River and Fraser River (Grassy Bar) release sites where sockeye salmon were captured, treated, and released. The numbered triangles represent the radio receiver stations. The inset shows stations 1 through 9 and the full system map shows stations 10 and 11.

Table 3.1 – Percentage of adult Harrison Rapids and Weaver Creek sockeye salmon that survived to reach their natal spawning areas after capture and release in the Harrison River, British Columbia, Canada. Upon capture, fish were assigned to three treatment groups. The first group (i.e., control group) was released immediately. The second group was subjected to the fisheries capture simulation consisting of 3 min of strenuous exercise and 1 min of air exposure. The third group was subjected to the fisheries capture simulation plus 1 min of assisted ventilation that was accomplished by orienting the fish's mouth into a jet of river water.

		Survived to spawning area
Population, treatment group	*n*	(% [*n*])
Harrison Rapids:		
Control	27	48.1 [13]
Capture simulation	40	10.0 [4]
Capture simulation + assisted ventilation	35	5.7 [2]
Total	102	18.6 [19]
Weaver Creek:		
Control	15	33.3 [5]
Capture simulation	12	8.3 [1]
Capture simulation + assisted ventilation	13	0.0 [0]
Total	40	15.0 [6]
Grand Total	142	17.6 [25]
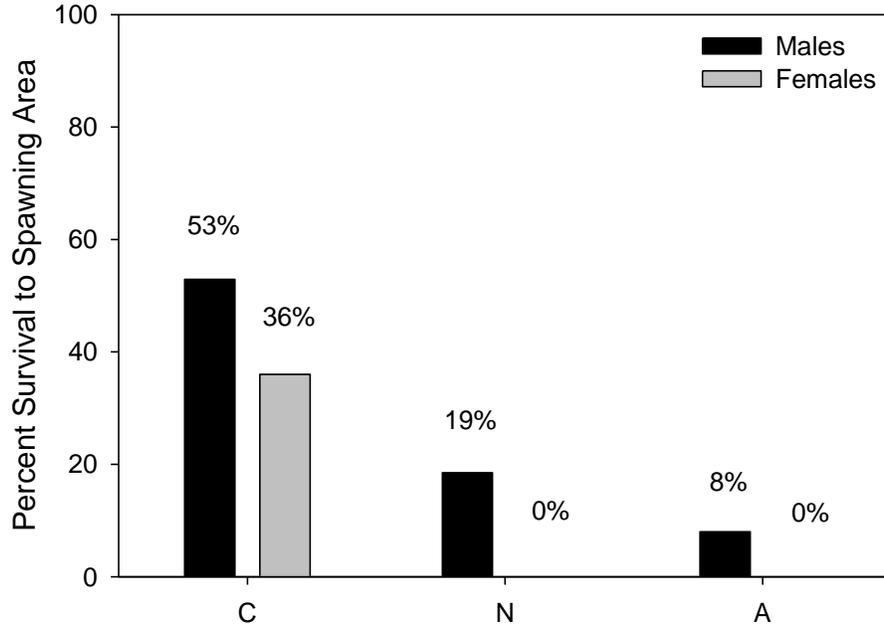
Figure 3.2 – Percent survival to reach natal spawning area after treatment (C = control, N = no assisted ventilation, A = assisted ventilation) for male (black bars, $n = 17 - 25$) and female (grey bar, $n = 23 - 25$) Harrison Rapids and Weaver Creek sockeye salmon. The control fish were released immediately after processing. The 'no assisted ventilation' fish were subjected to the fisheries capture simulation consisting of 3 min of strenuous exercise followed by 1 min of air exposure. These fish did not receive ventilation assistance prior to release. The 'assisted ventilation' fish were subjected to the fisheries capture simulation and then received 1 min of ventilation assistance that was accomplished by orienting the fish's mouth into a jet of river water. The trend of reduced female survival relative to males of the corresponding treatment group was not statistically significant ($P > 0.05$).

Table 3.2 – Sample sizes, study fish characteristics, and post-release survival for the three treatment groups. Angling duration refers to the time from hooking to landing. Data for angling duration and fork length are presented as means ± S.E.M. 'Air' refers to the 1 min of air exposure that was completed immediately after landing. 'Assisted ventilation' was accomplished by holding the fish by hand and orienting its mouth into the river flow (~ 0.5 m s^{-1}). All fish were gastrically tagged with radio transmitters for monitoring post-release survival. Stock composition and thus natal spawning areas were determined using DNA analysis of adipose tissue taken during processing (Beacham *et al*. 2005). The "*n* recaptured" column refers to tagged fish that were recaptured, killed, and reported by recreational fishers. Survival differences among groups were not significant for 72 h survival (Pearson's Chi Square, $\chi^2 = 0.10$, df = 2, $P = 0.95$) or survival to the upmost receiver station ($\chi^2 = 0.18$, df = 2, $P = 0.91$).

Treatment group	*n*	Angling duration (mm:ss)	Fork length (cm)	Stock composition	*n* recaptured	72 h survival (% [*n*])	Survival to upmost receiver station (% [*n*])
Angling	24	1:43 ± 0:13	63 ± 1	18 Chilko 2 Thompson 2 Stellako 1 Quesnel 1 unknown	3 of 24	87.0 [20 of 23]	69.6 [16 of 23]
Angling + air	23 (22 tracked)	1:39 ± 0:09	64 ± 1	11 Chilko 6 Thompson 3 Stellako 3 Quesnel	3 of 22	85.7 [18 of 21]	80.0 [16 of 20]
Angling + air + assisted ventilation	23	1:57 ± 0:15	64 ± 1	18 Chilko 2 Thompson 2 Quesnel 1 Birkenhead	6 of 23	94.4 [17 of 18]	70.6 [12 of 17]

Chapter 4: Conclusion

My thesis looks to evaluate and help mitigate the negative effects fisheries capture can have on released fish. Using a laboratory experiment (Chapter 2) and two field experiments (Chapter 3), I examined the influences of fisheries capture and release on adult Fraser River sockeye salmon that recently transitioned to fresh water during their spawning migration to natal sites. With this information, I have assessed a recovery technique designed to assist the captured fish's ventilation during its metabolic recovery from the physiological changes that are inherent with capture. This method of facilitating recovery is intended to increase the oxygen that is available to these fish during the post-exercise period that requires excess oxygen uptake (termed excess post-exercise oxygen consumption (EPOC); Gaesser and Brooks 1984; Lee *et al.* 2003). The fish's ability to fulfill this oxygen requirement in a timely manner can be vital to survival (Milligan 1996). Therefore, by assisting ventilation prior to release, I attempted to help mitigate the capture-related metabolic changes and thereby reduce the potential for physiological and behavioural impairments, and ultimately mortality. From linking my laboratory and field experiments, I was able to evaluate the physiological, behavioural and survival responses of sockeye salmon to capture and release and ventilation assistance. The assisted ventilation procedure that I developed for this recovery technique is comparable to techniques recommended by fisheries managers and used by recreational anglers attempting to recover fish prior to release (see Figure 4.1). The following synthesis of my results will inform managers regarding the efficacy of assisted ventilation as a recovery technique and encourage the integration of my results into managerial decisions regarding the use of live-release conservation and management tools.

56

4.1 Effects of fisheries capture and release on physiology, behaviour and survival

The physiological changes that resulted from my simulation of a fisheries capture-and-release event were assessed in the laboratory experiment (Chapter 2). The results indicated that the simulation elicited a stress response and provoked the fish to burst swim, using anaerobic metabolism for energy. As expected, fish that were exposed to the capture and release event exhibited significantly elevated plasma cortisol concentrations relative to control fish (Pickering and Pottinger 1995). The burst swimming events resulted in elevated plasma lactate levels and ion-osmoregulatory alterations that are consistent with fisheries gear encounters (e.g., Wilkie *et al*. 1997; Suski *et al*. 2006). Thus, my laboratory examination confirmed that the simulated fisheries capture and release induced metabolic changes that would require a period of recovery to regain homeostasis (Wood 1991).

This fisheries capture-and-release simulation was used in the laboratory experiment (Chapter 2) and the field-based Harrison River experiment (Chapter 3). In the laboratory environment, I observed reduced post-release survival of female sockeye salmon; however, a significant effect of the simulation on male survival was not detected. In the field experiment, both females and males exhibited a capture-related reduction in survival. This experiment also resulted in a trend towards reduced female survival relative to males. These sex-specific results corroborate previous studies suggesting that additional stressors experienced by female migrating sockeye salmon will elevate mortality levels (Patterson *et al*. 2004; Crossin *et al*. 2008; Nadeau *et al*. 2010; Jeffries *et al*. 2011; Roscoe *et al*. 2011; Martins *et al*. 2012). Overall, in this field experiment, fish exposed to capture and release exhibited a reduction of approximately 30% in survival to reach natal spawning grounds. This value is comparable to previous research that assessed an approximate 35% capture-

related reduction in survival to reach natal tributaries after release from angling in the lower

Fraser River (Donaldson *et al*. 2011).

4.2 Attempts to help mitigate the negative effects of fisheries capture and release

Assisting the post-capture ventilation of sockeye salmon for 1 minute prior to release

did not have a beneficial effect in the laboratory (Chapter 2) or field (Chapter 3) experiments.

In Chapter 2, I did not detect any positive influence of ventilation assistance on the

physiological variables of captured fish that received ventilation assistance compared to

those that did not. The results from chapters 2 and 3 indicate that the attempt at facilitating

recovery did not affect post-release migration behaviour or enhance survival. In fact, in the

lab experiment (Chapter 2), the technique appeared to reduce the survival of females when

compared to females that did not receive the assistance after capture. My attempt to evaluate

the influence of elevated water temperature on the assisted ventilation technique (Chapter 2)

resulted in rapid mortality (i.e., 100% mortality in < 3 days) and undistinguishable survival

rates among treatments.

Other attempts at reducing the negative effects of capture and release on fish have

used recovery devices (summarized in Table 4.1). Using devices for facilitating recovery has

the potential to assist ventilation while isolating fish from their natural environment. This

allows fish to recover from capture-related metabolic changes within a safe atmosphere (e.g.,

no risks of predation, recapture, or fall back). It also allows for long treatment durations

without manual restraint. As an example, a recovery device called a Fraser Box has been

used in the marine environment for the recovery of coho bycatch after capture and release in

a commercial gillnet fishery (Farrell *et al*. 2001a). The authors found that $1-2$ hours in the

device promoted physiological recovery and reduced short-term (24 hour) mortality. As a result, this device has been assessed in the freshwater environment on Fraser River sockeye salmon using 15 minutes of recovery after gillnet capture simulation (Nguyen *et al*. in press). In this experiment, the device did not significantly affect survival. Though both studies evaluated the efficacy of the Fraser Box after gillnet capture, the parameters of their use varied considerably (e.g., capture and release environment, species tested, and recovery duration; see Table 4.1). This example indicates the complexity of interpreting the results of these recovery device studies.

I sought to evaluate a manual recovery technique that does not require a device and imitates methods currently used by recreational anglers. I found that the tactic was not beneficial to adult migrating Fraser River sockeye salmon after capture and release in fresh water. And overall, research into facilitating the recovery of fish with and without devices has not indicated clear benefits; however, it has highlighted the need for case-by-case evaluations that assess the various influential parameters that may affect future research attempts to facilitate post-capture recovery.

4.3 Implications for local fisheries management

The live release of fish is a conservation and management tool employed to reduce the impact of a fishery on a population of fish. Fisheries and Oceans Canada (DFO), the organization responsible for the management of the multi-sectoral sockeye salmon fisheries in the Fraser River, can employ mandates that result in live release in order to ensure escapement targets are reached and to reduce fishing pressure on co-migrating populations of concern. In this multi-species, multi-sectoral management system, the ability to release live

fish that survive to spawn allows for continued fisheries openings. However, if the fish that are released due to these regulations are not able to survive and ultimately reproduce, then this can inhibit the efficiency of this conservation and management tactic.

For sockeye salmon released from freshwater recreational angling, DFO uses a post-release mortality rate of 3% (DFO 2012a). However, my thesis supports previous research that indicates post-capture delayed mortality levels of approximately 35% (see Donaldson *et al*. 2011). Thus, live-release management objectives would benefit from methods of reducing this post-release mortality. A method of facilitating recovery to reduce mortality that is currently recommended to recreational anglers by local managers and consequently evaluated in this thesis, did not aid in enhancing post-release survival. Therefore, this attempt to reduce the mortality associated with capture and release does not appear to help managers meet their objectives (i.e., low post-release mortality rates). Furthermore, there appears to be a higher level of post-release mortality than is accounted for in DFO's management of the recreational sockeye fishery. If managers cannot turn to live-release mandates to mitigate fishing pressures to the extent currently assumed, then there may be the requirement to reduce harvest allocations. Complicating managerial decisions further, is the knowledge that this capture-related mortality increases in warm water temperatures, particularly for female sockeye salmon (Martins *et al*. 2012).

4.4 Limitations to recommendations and future research directions

Future studies can build on the results I have presented here, particularly as my research is specific to semelparous migrating sockeye salmon that recently transitioned to fresh water. Research on Pacific salmon has found context/gear-specific, sex-specific, species-specific, and environment-specific differences in the consequences of capture

stressors on released fish (Gale 2011; Martins *et al*. 2011; Donaldson 2012). Even more, it appears that the success of attempts at facilitating the recovery of these fish is also situation-specific, potentially depending on the status of fish captured (e.g., exhaustion level, life history stage) and the environment in which the fish is released (e.g., predator load, fluvial system, water temperature). This complexity inhibits the ability to make broad-scale suggestions for the utility of various recovery techniques with or without devices. Thus, evaluating both the impacts of capture-and-release fisheries and methods of facilitating recovery should be assessed on a case-by-case basis.

Lastly, my research has focused on the lethal effects of capture and release and the subsequent impacts of assisted ventilation on this mortality. However, with this focus, I have not evaluated the implications of this technique on long-term sublethal effects. Even if fish survive release from capture, the sublethal effects (e.g., behavioural, growth and reproductive alterations) can manifest at the population level and thus influence management objectives (Cooke and Schramm 2007). Indeed, a recent study found negative effects of catch-and-release on the reproductive success of Atlantic salmon (Richard *et al*. 2013). Although these effects can be difficult to quantify, they can play a key role in overall population fitness and the achievement of management targets.
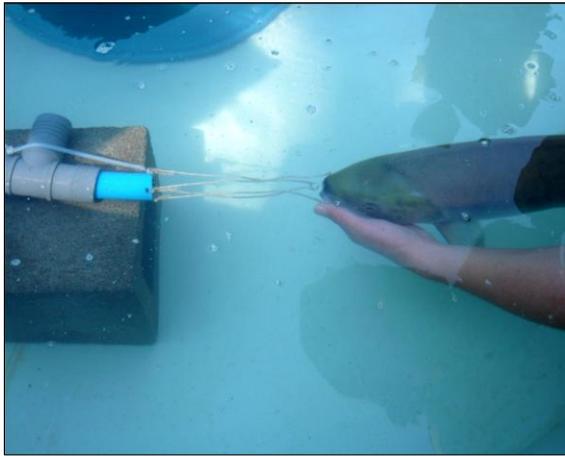
Figure 4.1 – Photos of the assisted ventilation technique performed under laboratory (left) and field (right) conditions. Fish were oriented into the water flow (~ 0.5 m s^{-1} as measured at the mouth) for a maximum of 1 min to aid the movement of water over the gill surface.

Table 4.1 – Research that has evaluated recovery devices intended to reduce the negative effects of capture and release on fish after encounters with various types of fishing gear in freshwater and marine environments.

Reference	Species	Capture gear	Recovery method	Recovery duration (min)
Fresh water				
Bettinger *et al*. 2005	Striped bass (*Morone saxatilis*)	Rod and reel	Live-release tube	120, 240, 360
Donaldson *et al*. 2013	Pink salmon (*Oncorhynchus gorbuscha*) Sockeye salmon (*Oncorhynchus nerka)*	Simulated angling Rod and reel	Recovery bag	15
Nguyen *et al*. in press	Sockeye salmon (*Oncorhynchus nerka*)	Simulated gillnet	Fraser Box	15
Marine				
Farrell *et al*. 2000	Coho salmon (*Oncorhynchus kisutch*)	Seine, troll, gillnet	Recovery box	30 – 60
Farrell *et al*. 2001a	Coho salmon (*Oncorhynchus kisutch*)	Gillnet	Fraser Box	60 – 120
Brownscombe *et al*. 2013	Bonefish (*Albula vulpes*)	Simulated angling	Recovery bag	15

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