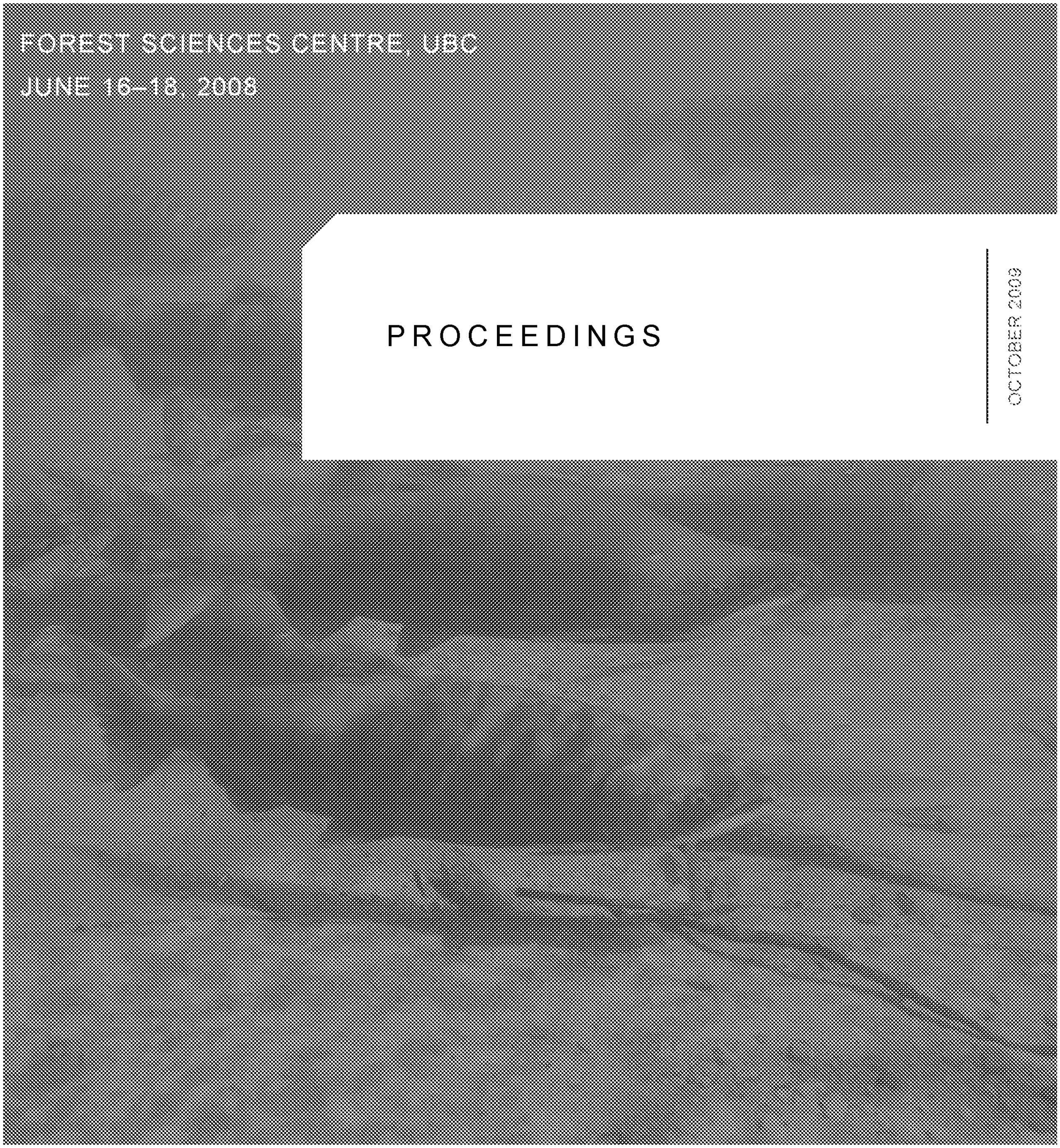
CONFERENCE ON EARLY MIGRATION AND PREMATURE MORTALITY IN FRASER RIVER LATE-RUN SOCKEYE SALMON



FOREST SCIENCES CENTRE, UBC

JUNE 16–18, 2008

PROCEEDINGS

OCTOBER 2009

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PREPARED BY

Scott G. Hinch and Julie Gardner, Editors

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COMING TOGETHER ON THE ROAD TO SOLVING THE EARLY LATE-RUN FRASER SOCKEYE MYSTERY

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This is a report on the proceedings of a conference held at the University of British Columbia (UBC) in June of 2008. Close to 70 participants attended, from universities, private consulting companies, fisheries management organizations, non-governmental environmental organizations and First Nations.[1] Several of the participants were graduate students who learned from the conference rather than contributing to discussions. The conference had an atmosphere of intensity, all aware that forward movement on the question of early migrating Late-run sockeye salmon on the Fraser River is critical.

CONFERENCE RATIONALE

Scott Hinch, Chair of the conference organizing committee (which included Mike Lapointe[2] and David Patterson), explained that, since 1995, large segments of Late-run sockeye salmon populations have forgone their normal estuarine holding behaviour and have migrated up-river several weeks early, resulting in extremely high levels of migration mortality. This phenomenon has presented a huge challenge to the management of the salmon resource. The conference aim was thus to explore the current knowledge of potential causes of the change in migration behaviour and of the high mortality. Investigators from academic, government and consulting groups were invited to present research results from the past five years on these issues to an audience comprised of fisheries scientists and managers.

CONFERENCE GOALS

There were three main goals for the conference:

1. **Fisheries Science:** Obtain feedback and comments on the research, suggest areas for research needs, and suggest how to proceed from current descriptive models of fish behaviour and survival to predictive models.

2. **Fisheries Management:** Involve all participants in discussions on how to convert scientific results into science advice or tools that would be useful for managers given current or future constraints on the fishery.

3. **Extension and Outreach:** Communicate research results to fisheries managers and scientists who have an interest in Late-run Fraser River sockeye salmon. The presentations and discussions form the basis for this document.[3]

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[1] See Appendix 1 for a list of participants. Julie Gardner facilitated the conference and Dawn Steele took notes.
[2] Mike Lapointe, Chief Biologist of the Pacific Salmon Commission, also assisted in editing this document.
[3] Currently, there are links to peer-reviewed publications and project research reports on the Late-run issue at: http://www.psc.org/info_laterunsockeye.htm

FINANCIAL AND OTHER SUPPORT FOR THE CONFERENCE

The conference was made possible by financial support through: a Natural Sciences and Engineering Research Council of Canada (NSERC) Strategic Grant to Scott Hinch, Tony Farrell, Glen Van Der Kraak and Mike Healey; the Pacific Salmon Commission; the Committee for Scientific Cooperation; and the Pacific Fisheries Resource Conservation Council. Conference facilities were provided by the Faculty of Forestry at UBC. The PFRCC funding included note-taker, Dawn Steele, who also produced the first draft of these proceedings.

BACKGROUND TO THE RESEARCH DISCUSSED AT THE CONFERENCE

In February 2001, the Pacific Salmon Commission (PSC) convened a workshop of some 20 experts with backgrounds in oceanography, salmon migration biology and physiology, disease/contaminants, and marine predators, which created a priority ranked list of approximately16 issues that could be explored as possible hypotheses explaining the early migration phenomenon (Table 1). Because the high in-river mortality was believed to be the result of the changes in migration behavior, researchers were asked to focus on hypotheses that might explain why Late-run sockeye were now migrating upstream early. There was little attention paid to the high in-river mortality issue at that time.

The experts brainstormed potential budgets and time frames to conduct research, and investigators were encouraged to form groups and seek research funds. Following the workshop, the Pacific Salmon Commission allocated $100,000 to fund seven pilot studies that investigated topics ranging from contaminants to broad scale oceanographic factors and physiology. In summer 2001, DFO also provided some funds for a couple of conventional tagging studies directed at Weaver and Portage creek sockeye to document the temporal pattern of en-route mortality and some research on the parasite, *Parvicapsula*, that had been implicated as a causal agent in the mortality. PSC and DFO allocated over $1M to fund two large scale marine tagging programs coupled with radiotelemetry in 2002 and 2003 that documented the pattern of river entry and mortality (English *et al.,* 2005)[4]. In 2002, a group of academics lead by S. Hinch, with financial and logistic support of DFO (FREWP) and the PSC, secured a large six-year NSERC Strategic Grant to explore several hypotheses involved with the early migration and high mortality phenomena. As part of that research endeavour, a summary paper was published on these phenomena (Cooke *et al.,* 2004)[5]. This paper included a description of the historical context and a re-worked list of research issues ranked in terms of the likelihood that specific hypotheses were involved with the early migration and high mortality phenomena (Table 1). The research partnerships grew in subsequent years, encouraged by considerable support through the Southern Endowment Fund of the PSC, which culminated in a large, interdisciplinary program in 2006 involving dozens of researchers from government, academic and private organizations.

[4] English, K.K., Koski, W.R., Sliwinski, C., Blakely, A., Cass, A., and J.C. Woodey. 2005. Migration timing and river survival of Late-run Fraser River sockeye salmon estimated using radiotelemetry techniques. Transactions of the American Fisheries Society 134:1342–1365, 2005

[5] Cooke, S.J., Hinch, S.G., Farrell, A.P., Lapointe, M.F., Jones, S.R.M., Macdonald, J.S., Patterson, D.A., Healey, M.C., and G. Van Der Kraak. 2004. Abnormal migration timing and high en route mortality of sockeye salmon in the Fraser River, British Columbia. Fisheries. 29:22-33

TABLE 1. Ranked list of 16 issues and/or hypotheses involved with potential research on the early migration phenomenon for Late-run sockeye salmon (based on February 2001 PSC 'expert' workshop). *Ranking procedure score in parentheses—smaller number indicates an issue or hypothesis perceived as needing more urgent investigation.*

H number and score	Description of hypothesis
1 (4.0)	Exploratory data analysis of ocean parameters related to Late-run upstream timing.
2 (4.4)	One or more Biological Clocks has/have been advanced leading to early upstream migration.
3 (5.3)	Sockeye salmon acquire infection prior to Fraser River entry leading to a behavioral change.
4 (6.0)	Adults approaching Fraser R. are exposed to chemicals that trigger early entry into Fraser.
5 (6.5)	The fish behaviour has changed (monitoring, or EMG tagging).
6 (6.9)	There is a relationship between parasite infection and host mortality.
7 (7.7)	Smolts are infected near mouth of the Fraser River.
8 (7.8)	Exchange processes in Georgia Strait have altered delay behavior.
9 (8.9)	Changes in vertical distribution of Late-run sockeye causes them to experience different physical conditions altering delay behavior.
10 (9.4)	Early upriver migration triggered by low stored energy reserve.
11 (9.7)	Interaction of diversion rate with physical ocean conditions affects Late-run upriver timing.
12 (9.8)	Choice of river entry timing is related to conditions in year of ocean entry (smolt effect).
13 (10.0)	Sockeye are fleeing into the Fraser River to avoid predators.
14 (10.1)	Diet may be responsible for changes to Late-run sockeye behaviour.
15 (10.8)	Predation by harbour seals and killer whales has increased.
16 (10.9)	Outmigrant juveniles are exposed to chemicals that reduce their ability to hold as adults.

Table 2 provides a list of hypotheses for explaining the early migration and high mortality phenomena published by Cooke *et al.* (2004). The hypotheses are categorized into physiology, abiotic environment and biotic environment with the understanding that any given hypothesis may involve interactions with others (e.g., possible synergisms). Hypotheses are not ranked but are identified based on expert opinion as to the relative likelihood that they are responsible for the phenomena. The 'higher priority' issues from Table 1 were deemed 'more likely' by this list. The asterisk indicates that Hypothesis #10 was not in the original list by Cooke *et al.* (2004) but was added to this list because of recent research findings (see presentation by English and Robichaud, p.49 in these proceedings). Much of the research discussed at this conference focused primarily on the priority or 'more likely' hypotheses.

TABLE 2. Hypotheses involved with the early migration and high mortality phenomena (Cooke *et al.*, 2004).

		Relative likelihood	
Hypotheses	**Possible synergisms**	**River entry timing**	**River mortality**
Physiology			
1) Energetics	5,6,8	More Likely	More Likely
2) Osmoregulation	4,5,8	More Likely	More Likely
3) Photoreception	2	Less Likely	N.A.
4) Reproductive Hormones and Sexual Maturation	2,7	More Likely	Less Likely
Abiotic Environment			
5) Oceanic Environment	1,2	More Likely	Less Likely
6) In-River	1,8	Less Likely	More Likely
7) Contaminants	4	Less Likely	Unknown
Biotic Environment			
8) Parasites and Disease	1,2,5,6	More Likely	More Likely
9) Marine Mammals	1	Less Likely	Less Likely
*10) Con-specific Abundance	1,2,4,5	More Likely	N.A.

CONFERENCE AGENDA

On Day 1 the theme was 'Ocean Effects and Migration Timing'. Presentations in the morning focused on historical context, and introductory / observational studies. In the afternoon, the focus was largely on hypothesis testing and experimentation. On Day 2 the theme was 'Consequences of Early Migration'. The first set of presentations dealt with survey observations; the second set explored issues with experimental approaches; and presentations concluded in the early afternoon with examples of management models based on these science results. The second half of the third afternoon involved panel and open discussions. On Day 3, the final day, a discussion was held with the presenters and other interested parties on report writing and future research directions. The agenda is detailed in Table 3.

TABLE 3. Conference agenda (times and details of presentation titles changed during the conference).

Monday June 16 Ocean Effects and Migration Timing		
8:45	*Breakfast*	
9:15	S.Hinch, J.Gardner	Introductions and objectives; hypotheses and research programs
9:30	M. Lapointe	Migration timing and mortality issues—historical to present context, management concerns
10:00	T. Farrell	Introduction to migration physiology and stress—thresholds and relevance to timing and fate
10:30	*Break*	
11:00	D. Patterson	Biological trends in destructive sampling of ocean caught salmon
11:30	K. Miller	Introduction to migration genomics—ocean survey results
12:00	S. Cooke	Ocean biopsy telemetry in 2003—physiological correlations with migration timing, marine fate
12:30	*Lunch*	
1:15	G. Crossin	Ocean biopsy telemetry in 2006—physiological correlations with migration timing, marine fate
1:30	I. Olsson	Coastal water properties and migration timing relationships
2:00	R. Thomson	Predicting early migration timing—ocean water properties
2:30	K. English	Predicting early migration timing—'Stay with the School'
3:00	*Break*	
3:30	M. Cooperman	Experimental evaluation of early migration timing—'maturation and osmoregulation' hypothesis
4:00	H. Herunter	Fraser River water chemistry trends—are there correlations with river entry timing?
4:30	S. Hinch	Revisiting hypothesis list, open discussion of alternative hypotheses, unresolved questions
5:00	J. Gardner	Concluding comments

Tuesday June 17 Consequences of Early Migrations		
8:00	*Breakfast*	
8:30	S. Hinch J.Gardner	Introduction to session and objectives; original hypotheses
8:45	D. Patterson	Historical relationships between flows, temperature and Late-run migrations
9:15	K. English	Migration survival and timing—results from several years of radio telemetry
9:45	S. Cooke	Biopsy telemetry and freshwater fate of migrants in 2003 and 2006
10:15	*Break*	
10:45	S. Hinch	Acute and chronic thermal issues—simulated migration experiments
11:15	T. Farrell	Aerobic scope and temperature: lab and field experiments
11:45	K. Miller	Gene array telemetry experiments and river migration mortality.
12:15	*Lunch*	
1:00	D. Patterson	Pre-spawn mortality, disease and intergenerational consequences
1:45	M. Hague	Thermal-based management models (current and proposed)
2:15	S. Hinch	Revisiting hypothesis list
2:30	J. Gardner	Management 'panel' discussion followed by open discussion
3:30	*Break*	
4:00	J. Gardner	Science 'panel' discussion followed by open discussion
5:00	J. Gardner	Concluding comments

Wednesday June 18 Discussion on Report Writing and Future Directions (presenters should attend, others welcome)	
9:00–12:00	
Gardner	i) Reporting—Discuss next steps with regards to the proceedings/brochure
	ii) Conference impressions—General discussion on feedback from Tuesday afternoon
	iii) Future Research—What research, data or analyses are needed?
	Can current research programs address these gaps?
	Should new collaborations emerge?
	What role might some conference participants have?
	Where would funds come from?

FORMAT OF THE PROCEEDINGS

The general format of the workshop was presentations of 15 to 30 minutes, followed by discussion periods that ranged from 5 to 20 minutes. The oral presentations, subsequent questions and answers, and discussions are summarized in this document. Also included are abstracts of forthcoming research publications provided by the authors. The abstracts precede the presentations in this write-up, and include a list of all participants in the research project to which the presentations relate. Virtually all the presentations included PowerPoint slides with illustrations, which were referred to in drafting the text in this document on the oral presentations. The slides are not included in this document, with the exception of the presentation by M. Lapointe, as they remain to be refined and finalized for publication elsewhere.

Questions were posed by the organizing committee to help initiate discussions reflecting on the proceedings of the workshop and the broader research and management context to which they relate, in the second afternoon of the workshop. Two panels of invited observers provided initial comments on these questions prior to involving all workshop participants in the discussion. The questions put to the panelists were also provided on a questionnaire to participants (Appendix 2). Written comments from the few questionnaires returned are included in Appendix 3.

Two participants, R.F. Addison and Jim Woodey, also submitted written comments. These are included in Appendices 4 and 5.

Discussions from the morning of the third day, involving a smaller group of participants looking ahead to next steps, are also summarized in this document.

This record of the proceedings is organized as follows:

- An overview and synthesis of the presentations and discussion at the conference
- Day 1: Presentations and discussions on ocean effects and migration timing
- Day 2: Presentations and discussions on consequences of early migration
- Day 2: Management and science panel discussions
- Day 3: Discussions of next steps

OVERVIEW AND SYNTHESIS: EARLY MIGRATION AND PREMATURE MORTALITY IN FRASER RIVER LATE-RUN SOCKEYE SALMON

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INTRODUCTION

A fascinating biological situation, a vexing management issue, and a conservation crisis are all apt descriptors for a phenomenon that has been on-going for at least the past 14 years involving adult Late-run Fraser River sockeye salmon (*Oncorhynchus nerka*). These stocks make up the dominant run of Fraser River sockeye in some years, and are one of the most valuable and important groups of salmon in Canada. They are unique among sockeye populations anywhere, and perhaps among Pacific salmon in general, in that following their directed migration from the ocean they 'hold' or 'delay' in the estuary (i.e., the Strait of Georgia) for several weeks (i.e., 3–6) prior to migrating into the mainstem of the Fraser River and onward to spawning areas. Beginning in 1995 and continuing through 2008 this delay period shortened substantially—in several recent years it was 5 days or less (Lapointe, p.15 these proceedings)[6]. The pattern of early migration appears to be occurring in all Late-run populations, including Cultus sockeye for which fence data suggest that the normal migration pattern was observed consistently back into the 1940s. Other salmon species for which delay behaviour in the estuary is not well understood are also migrating into the Fraser River earlier than usual (Lapointe, p.15 these proceedings). Early river entry is not by itself necessarily a large concern; however it is associated with 'en route' mortality during upstream migration and 'pre-spawning' mortality after arrival at spawning areas in sockeye (Lapointe, p.15 these proceedings). In years of the most extremely early entry (e.g., 2000, 2001), total freshwater mortality has exceeded 90% (Lapointe, p.15 these proceedings) meaning less than 10% of some populations have reached natal areas and successfully reproduced.

The early migration / high mortality phenomena pose risks to the perpetuation of these fisheries resources. Prior to 2003, there were few tested hypotheses about potential mechanisms that may underlie these phenomena and no predictive tools to identify when or how they may occur. Such uncertainty challenged fisheries managers and has led to a precautionary approach to harvest decisions. This proceedings document contains a considerable level of new and emerging results and several recommendations are made by individual presenters on what the next research steps should be. The following summary overviews the biological studies conducted to date, the current thinking on causes of early migration and high mortality, potential management uses of this information, and future research needs.

[6] Page numbers referring to articles in these proceedings indicate the first page of the article.

OVERVIEW OF SCIENTIFIC RESULTS

Why Does Early Migration Occur?

This abrupt shift in behaviour has resulted in almost all Late-run fish since 1995 entering the river prior to the historical median upstream date. As a consequence, research examining causal factors for the change in behaviour is hampered by the lack of an experimental control. Thus, the causes of the shortening or complete elimination of the estuarine delay period are still not completely understood but a picture is emerging which illustrates complex links between physiology, environment and behaviour. Reproductive advancement is a key feature in coastal migration speed and in reduced estuarine holding (Cooke *et al.,* p.37 these proceedings; Crossin *et al.,* p.41 these proceedings), and because the physiological changes that initiate reproductive maturation occur prior to fish reaching the coast during their homeward migration (Patterson and Hills, p.31 these proceedings; Miller *et al.,* p.33 these proceedings) the estuarine behavioural change may have its roots in the open ocean. Early entering fish are also not healthy. Their gene array profiles reveal disease, viral, pathogen, and stress responses (Miller *et al.,* unpublished analyses conducted since conference). The fact that 50% of the fish sampled at the Queen Charlotte Islands carried the same disease signatures identified later in the migration at coastal sites and in the lower Fraser River suggests that segments of fish populations may become ill and/or susceptible to diseases while in the high seas (Miller *et al.,* p.77 these proceedings; Miller *et al.,* unpublished analyses conducted since conference). The disease state appears to alter the osmoregulatory physiology of migrants making them osmotically similar to freshwater fish. The physiological processes regulating reproductive maturation and osmoregulation are tightly linked in migrating salmon (Cooperman *et al.,* p.52 these proceedings) so it is possible that the disease state is also responsible for the advanced maturation observed in early-migrating Late-runs. The cause of the disease and/or early maturation and whether these characteristics are driving fish to migrate upstream earlier than normal are unknown. Interannual variation in open-ocean wind speed correlates over the past two decades with interannual variation in river entry timing—suggesting an environmental link (Thomson and Hourston, p.46 these proceedings). How this variable, or others it affects or which affect it, alters the maturation process or disease state of a segment of all Late-run populations is not clear. Further, because genomic and plasma assay analyses have only been conducted in a few recent years on high seas departing migrants (e.g., those sampled at the Queen Charlotte Islands), there is no historic baseline for comparisons and hence only a limited means to argue that some physiological patterns are 'abnormal' (conference comments submitted by J. Woodey, p.111 these proceedings).

Although the genesis of the early migration phenomenon seems to, in part, be in the open ocean, coastal processes appear to further influence migration timing. Specifically, there are several lines of evidence that coastal salinity and osmoregulatory preparedness play important roles: 1) historical correlations show that in years with lower coastal salinity some stocks of Late-run fish entered the river earlier (Thomson and Hourston, p.46 these proceedings); 2) physiological systems of some coastally sampled fish are clearly 'freshwater prepared' and these fish enter the river early (Crossin *et al.,* p.41 these proceedings); 3) estuarine captured Late-runs which were experimentally exposed to freshwater, tagged and released migrated in-river earlier than saltwater exposed fish (Cooperman *et al.,* p.52 these proceedings); and, 4) in that same year, fish which migrated the fastest from estuarine capture sites into the river were associated with the lowest salinity concentration at the capture and tagging site (Olsson *et al.,* p. 44 these proceedings). These more recent findings are consistent with earlier work by Phil Gilhousen decades ago with the Pacific Salmon Commission who found that even in years prior to the beginning of the early upstream migration phenomenon, shorter durations of upstream migration in Adams River sockeye were related to higher Fraser River flows. The other coastal issue that may be important in terms of migration timing of Late-runs is the correlation observed between river entry timing and the proportion of Summer to Late-runs in the Strait of Georgia (English and Robichaud, p.49 these proceedings), which may represent a contributing process responsible for early entry. However, adopting a radically different behaviour that is seemingly non-adaptive (e.g., entering the Fraser River early) would likely not occur without some physiological basis. Early migrating Late-run fish are not in physiological homeostasis during their coastal migration (Farrell, p.28 these proceedings). Early migrants are relatively more reproductively prepared, less osmotically prepared for marine holding, and stressed. These factors could be enough to motivate these potentially 'unhealthy' fish to migrate in-river early. However, additional motivation might come from sensing high abundances of Summer-runs also migrating into the river. It is impossible to determine if the Summer-run mechanism is merely correlative with, or complementary to, the physiological ones because it was not possible to examine the relative physiological states of fish in terms of their local experiences with differing abundances of Summer-runs. There is little evidence that water chemistry or known environmental chemicals (Herunter, p.56 these proceedings; Addison, p.107 these proceedings appendix) are likely triggers or enticements for early migrations. Time-series data are lacking for some therapeutic drugs that possibly could play a role.

Causes of High Enroute Mortality—'Dead Fish Swimming'

Multi-year telemetry and conventional disk tagging studies have confirmed that early river-entering Late-run sockeye perish at relatively high rates during their river migration compared to fish which enter during more 'normal' times (English and Robichaud, p.65 these proceedings; Cooke *et al.,* p.68 these proceedings). Early-timed fish encounter much warmer river conditions and accumulate more thermal units than normal timed fish (Patterson *et al.,* p.61 these proceedings). During their estuarine holding, Late-run sockeye are undergoing reproductive development so they can be prepared to reproduce shortly after arrival at spawning sites. Because adult salmon stocks in the Fraser River appear to be adapted to their historical migration temperatures, early migrants are physiologically stressed during migration (Farrell *et al.,* p.74 these proceedings). Because additional maturation is still required when they get near spawning areas, survivors hold in nearby lakes, instead of holding in the estuary. Spawning times in sockeye salmon are known to be adapted to the thermal regimes in their natal streams that maximize survival of their offspring (i.e., to ensure that eggs incubate and fry emerge at a time when plankton food items are available in nursery lakes), and spawning times have changed very little during this period of early upstream migration (Patterson *et al.,* p.61 these proceedings). Early-timed fish are therefore exposed to freshwater diseases and parasites (i.e., *Parvicapsula minibicornis,* and others) for longer periods of time with disease development being accelerated by higher than normal river temperatures due to early river entry and climate warming, and greater degree days due to earlier summer migrations (Hinch *et al.,* p.70 these proceedings). The evidence suggests that a combination of factors including premature senescence, elevated stress, ionoregulatory dysfunction and disease (Farrell *et al.,* p.74 Hinch *et al.,* p.70 Cooke *et al.,* p.68 Miller *et al.,* p.77; these proceedings) kills many of these fish in freshwater before they reach spawning grounds. These results provide some insights into the selective forces at play that may have originally contributed to the evolution of their estuarine holding, a behaviour which minimizes their freshwater residency times prior to spawning and reduces contact with freshwater diseases and parasites and high migratory temperatures in-river, yet ensures that spawning occurs at a time that maximizes survival of offspring. In some recent years, Late-run fish have experienced temperatures that are acutely lethal so some Late-run fish are likely dying due to cardiorespiratory collapse (Farrell *et al.,* p.74 these proceedings). Lastly, genomics results point towards a further issue—early-timed migrants appear to be 'unhealthy' when they depart the ocean, possibly the result of a viral disease (Miller *et al.,* p.77 these proceedings). Thus, early migrants are physiologically compromised prior to freshwater entry. This would likely increase the lethality of freshwater diseases and exacerbate thermal stressors affecting these fish. All studies emphasized the pivotal role of water temperature, both as an acute and chronic stressor, on the behaviour and survival of Late-run sockeye salmon.

Spawning Ground Fish and Their Offspring—Legacy Effects of Early Migrations?

Spawning ground arrival dates and peak spawning periods of Late-run sockeye have varied within the normal historical range since the onset of the early migration phenomenon (Patterson *et al.,* p.61 these proceedings). Historically, pre-spawn mortality (termed PSM; e.g., dying unspawned) in most stocks was correlated with upstream migration timing and temperature, with higher levels in the earliest arriving fish. Though PSM is still observed in Late-run fish, it does occur throughout their entire spawning period and in some recent years has been as high in the both the earliest and latest arrivals at spawning areas (Patterson *et al.,* p.61 & 81 these proceedings). On average early river migrants are more likely to be the first to arrive on the spawning grounds, however diminishing proportions still continue to arrive throughout the entire spawning period and may contribute to elevated pre-spawn mortality in later time periods. Early entering Late-runs encounter relatively high temperatures and flows in the Fraser River compared to normal-timed Late-runs, and they are on average more reproductively advanced at river entry than fish delaying; therefore, faced with a more difficult migration condition it was reasonable for investigators initially to assume that Late-runs would arrive on spawning grounds with less body energy and thus be more at risk to PSM. However, first arrivals on spawning grounds actually have the highest energy reserves (Patterson *et al.,* p.61 & 81 these proceedings), suggesting that energy exhaustion on spawning grounds is not a factor affecting spawning success in early migrating Late-runs. Results from experiments which have altered energy reserves and physiological stress levels towards the end of river migration in Fraser sockeye support this (K. Hruska, UBC, PhD thesis unpublished. data). The physiology and histopathology of PSM fish indicates gill and kidney diseases play a significant role in mortality, in particular those caused by *Parvicapsula minibicornis,* though also *Loma, Columnaris* and *Saprolegnia* (Patterson *et al.,* p.82 these proceedings). The search for a single cause or suite of casual agents of PSM has been difficult—PSM appears to be complex and can likely result from a multitude of factors whose efficacy can change both annually and seasonally. Most, if not all, of the causal factors are accentuated by increases in temperature and freshwater residency times. Thus once fish enter freshwater it is a race against time for early migrants to be able to spawn prior to succumbing to various disease agents. Considering that the primary determinant of spawning success is longevity on spawning grounds (K. Hruska, UBC, PhD thesis unpublished data), early migrants that are already compromised by their thermal history which reach spawning grounds are at a clear disadvantage to spawn successfully, so they probably have reduced spawning success though there are few data available to test this hypothesis.

There is no evidence that egg quality varies with time of arrival onto spawning grounds, and eggs from PSM females produce highly viable offspring in experiments (Patterson *et al.,* p.84 these proceedings). These findings indicate that gamete quality is highly preserved even at the expense of parental health and survival. Individual variability among spawning ground adults in some physiological characteristics had intergenerational consequences in that it affects egg and offspring survival but it is not clear how much of that variability was caused by an individual's migration experience and/or its genotype (or both) (P. Nadeau, UBC, MSc thesis; K. Hruska, UBC, PhD thesis in progress).

MANAGEMENT USE OF LATE-RUN SOCKEYE SCIENCE

Overview

A primary goal of the presentations at this conference was to provide fisheries managers with information that could be used to change harvest regulations and policy, and, if possible, consider mitigation or adaptation strategies to help prevent the collapse of fisheries and extinction of some stocks. It was hoped that a better understanding of the immediate and intergenerational fitness consequences will provide valuable information for developing pre-season prediction tools, aid in-season decisions, and improve future season prediction models for harvest and stock sizes. Indeed the high rates of en-route and pre-spawn mortality documented during some years have caused managers to take a precautionary approach and prohibit fisheries targeting Late-run sockeye, and limit harvesting to by-catch in fisheries directed at other healthy sockeye stocks. In years of larger Late-run abundances, exploitation rates have been adjusted based on river entry timing predictions that are based on the fraction of fish migration upstream at any given point in time. However, fisheries managers at the conference expressed mixed opinions on whether the research results could convert directly into management actions. Some advocated concentrating more fishing with the Fraser River in early to mid-August when the prospects for survival of Late-run sockeye were particularly poor. Such a tactic could increase harvests of healthier stocks while minimizing short-term impacts on Late-run stocks. However, others suggested that if some of the earliest upstream migrants were able to successfully reach the natal areas and spawn, they would be amongst the most valuable to protect in a long term evolutionary sense (see Management Panel Discussion, p.89 these proceedings).

Early River Entry Phenomenon—Can It Be Predicted?

The inability of the management agencies to predict river entry timing of Late-run sockeye in the past has resulted in hundreds of millions of dollars in foregone harvest. Identifying the causes of early migration would hopefully provide some level of mechanistically based predictability of the behaviour, based on physiological and/or environmental conditions along adult sockeye coastal migratory routes. It should be first noted that the marine approach timing into the Strait of Georgia has not changed—early entry into the Fraser River means early departure from the Strait. There were over a dozen hypotheses proposed when the early entry phenomenon was first realized. Conference presenters have reduced that list substantially and have a much better understanding of the factors affecting early migration. A conceptual model that involves both open ocean and coastal ocean processes is proposed (see above; and Science Panel Discussion, p.93 these proceedings). However, there is still no ‘silver bullet’ factor that managers desire for pre-season prediction as it appears this is a complex, multi-factor phenomenon. Regardless, there are several indices that managers can presently turn to for aid in predicting river entry timing both pre-season and in-season. Specifically, open-ocean wind stress strongly correlated with river entry date for the Adams stock and coastal salinity strongly correlated for the Weaver stock. These relationships could give managers insights into river entry timing several weeks in advance of fish reaching the Fraser River. Summer-run abundance can be estimated pre-season which may give managers several months’ advance insights into Late-run entry timing. During the season, managers have developed models that predict the median date of upstream migration from the fraction of the total run that has migrated upstream to date. While the inputs to these models are subject to assessment error and they do not provide much advanced warning of early entry, the models have allowed managers to react to changes in-season, particularly in years of large Late-run abundances.

As investigators have not discovered any single process that appears to govern entry timing, prudence dictates that managers use multiple sets of information to develop the broadest picture possible for pre- and in-season forecasting. Because behavioural changes are largely driven by physiological processes, physiological indicators that can be readily collected and rapidly processed in real-time during coastal migration could guide future management decisions. Genomic biomarkers (small sets of genes that when expressed may be predictive of a behaviour), which are currently being researched and developed for Late-run entry timing (and river migration fate), may provide such a system, particularly if it turns out that the early migration Late-run phenomenon is being driven by an ocean disease state as some preliminary results suggest.

High River Migration Mortality Phenomenon—It Can Be Predicted

Managers need to know how many fish are likely to perish during river migration in order to adjust harvest levels in the ocean and lower Fraser River to ensure spawning ground escapement targets are achieved. Presently, managers use models based on associations among historic differences between lower and upper-river escapement estimates and environmental conditions and/or entry timing for this purpose. These models are then used to increase escapement targets to compensate for anticipated mortality or differences resulting from adverse river conditions (flows or temperatures) or early river entry. Current adjustment models do not explicitly use much of the mechanistic knowledge presented at the conference, though future adjustment models could benefit from it (Hague and Patterson, p.85 these proceedings). Investigators made considerable progress in understanding the mechanisms, and predicting the levels, of river migration mortality experienced by Late-run sockeye. As with the early migration issue, high mortality is complex and caused by several factors. Nonetheless, the research conclusively demonstrated that physiological factors and higher than normal temperatures kill early migrating fish before reaching spawning areas in expected ways. Managers could use specific water temperature metrics to predict migration mortality (e.g., critical thermal limits, or degree day accumulation). For example, encountering migration temperatures > 20°C resulted in 90–100% enroute mortality of Weaver Creek sockeye because of collapse of metabolic scope. At less extreme temperatures (i.e., 18–19°C) where the processes of disease and stress were paramount, levels of migration mortality were correlated to specific levels of degree day accumulation during migration. Hague and Patterson (p.85 these proceedings) overviewed how environmental variables, which may be surrogates for physiological processes, could be integrated into management adjustment models in order to increase their biological realism. For example in addition to river entry timing, average or extreme river temperatures or flows, weighted temperatures, or degree day accumulation were all suggested as candidate variables. The largest challenge for incorporating environmental variables into adjustment models is the need for spatially and temporally explicit migratory information, which is difficult to obtain and integrate into model predictions on the time scales needed for in-season forecasting. Post-season estimates of en route mortality, independent of differences between estimates, can now be estimated based on these biological relationships explored at this conference to generate better total return size estimates for recruitment curves. These environmental variables can also be used in future-scenario modeling in order to examine how climate change predictions will influence migration survival and escapement.

Do Managers Need to Worry About Legacy or Intergenerational Consequences?

If early entering Late-runs which arrive on spawning grounds suffer high levels of pre-spawn mortality, or if their eggs and offspring are of inferior quality, then this would mean that even higher levels of spawning ground escapement (e.g., lowered levels of harvest) may be needed to ensure stock sustainability. The implications for management of the recent research are that there is likely no need for incremental adjustments to coastal or river migration harvest strategies on early vs. normal timed migrants in terms of their 'on-spawning ground' fitness; compensation for en-route and pre-spawn mortality would appear to be sufficient to ensure sustainability. However, pre-spawn mortality is complex and likely results from a multitude of factors whose efficacy can change both annually and seasonally. Most, if not all, of these factors are accentuated by increases in temperature and freshwater residency times. As experiments that examine the legacy effects of high temperature exposure on adult migrants to their eggs and offspring are still in progress, management should continue to be risk averse and consider the risk of pre-spawn mortality and intergenerational effects in relation to predictable correlates such as migration temperatures and freshwater residence (e.g., accumulated degree days).

FUTURE RESEARCH—WHERE DO WE GO FROM HERE?

The integration of large-scale ocean telemetry with behavioural ecology, physiological biopsy, genomics, and experimental biology, as pioneered by several of the investigators working on the Late-run issue, has proven to be a powerful research approach. Researchers have come a long way in a short time towards identifying a short list of potential causes of the early migration phenomenon. Yet, if indeed the process is mediated through environmental factors in both high seas and coastal areas, then a single predictive explanation will be difficult to uncover without continued large-scale field research. Despite all that has been done, a better understanding is needed of how oceanographic conditions (e.g., salinity, temperature) trigger or control physiological, and hence behavioural, changes in individual migrating and maturing sockeye, and how other environmental factors (both endogenous such as disease states and exogenous such as local abundance) mediate these behavioural changes. Though a small number of manipulation experiments have been undertaken, clearly more are needed as only in this way can cause and effect truly be established. For example, holding Late-run fish in marine pens at different locales relative to the Fraser River mouth, releasing at key times, and tracking using large-scale fixed telemetry systems (e.g., POST) could help elucidate how specific local oceanographic or abundance conditions affects river entry timing by Late-run populations. Physiological interventions and individual-based tracking with mobile telemetry systems at these or other locales are needed to explore specific mechanisms which are thought to be responsible for early river entry. It will remain a challenge to assess the role of prior life history experience in any of these experiments. Some at the conference suggested that other environmental factors including biologically active contaminants and other pollutants also deserve research attention. As some other species of salmon also seem to be eliciting early migrations, the search for answers in early migrating Late-run sockeye needs to include, where possible, analogous studies in other groups of fish.

The causes of en route mortality in early migrants are now reasonably well understood. Early migrants are 'out of synch' with their river migration environment and are clearly not well adapted to deal with the riverine conditions they experience. Only a few Late-run stocks have been examined in detail but there is good evidence that stock-specific adaptations exist. Future work must examine the capacity for adaptation in key Fraser sockeye stocks, not just Late-runs, because as the Fraser River continues to warm from year to year there is significantly increased risk of high mortality even in normally timed Late-runs as well as in other run timing groups. The study of diseases will increase in importance, as diseases will play much larger and obvious roles as agents of en route and pre-spawning mortality in a warming Fraser River. Understanding which stocks will be able to thrive, which ones can barely survive and which ones will be extirpated in the near future is not only a purely scientific query, but also a fisheries management and policy concern.

DAY 1: OCEAN EFFECTS AND MIGRATION TIMING

Presentations in the morning focused on historical context, and introductory and/or observational studies. In the afternoon, the focus was largely on hypothesis testing and experimentation.

MIGRATION TIMING AND MORTALITY: CONTEXT AND MANAGEMENT CONCERNS

Mike Lapointe
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ABSTRACT

Late-run stocks are unique among Fraser River sockeye salmon populations in that they historically held in Georgia Strait for a period of three to six weeks prior to migrating upstream to spawn. Beginning in 1995 and continuing through 2007 this delay period has shortened substantially. The average delay has been five days or less in all years since 2000, with the exception of 2002 when the average delay was 22 days. The decrease in delay has resulted in much earlier than normal upstream migration. In extreme years, like 2000 and 2001, fish entered the Fraser River four to six weeks earlier than normal. In a few years of larger Late-run abundance (e.g., 2002, 2006), fish have tended to delay for longer periods, but even in these years upstream migration has been earlier than historically observed. The pattern of early migration appears to be occurring in all Late-run populations, including Cultus sockeye for which fence data suggest that the normal migration pattern was observed consistently back into the 1940s. This abrupt shift in behavior has resulted in almost all Late-run fish since 1995 entering the river prior to the historical median upstream date. As a consequence, research examining causal factors for the change in behavior is hampered by the lack of an experimental control. Similar patterns of early migration have been observed in Fraser River pink, chum and white-fleshed Chinook salmon. For Fraser River pink salmon, the change to earlier migration occurred coincident with the change in sockeye, and has been associated with decreased delay.

Each year, the early river entry has been associated with variable rates of mortality prior to spawning which increase with earlier upstream timing. Two types of mortality have been observed: 1) en-route mortality—fish that die during their upstream migration prior to reaching spawning areas, and 2) pre-spawn mortality—females that die on the spawning grounds with their eggs intact. The two sources of mortality are incremental, and both increased substantially relative to historical patterns coincident with the change in behavior in 1995. Temporal patterns in intra-annual mortality have been documented through tagging programs in a number of years. These studies have demonstrated that the earlier migrants each year suffer the highest en-route and pre-spawn mortality such that very few fish that migrate upstream prior to mid-August reach the spawning grounds and a high fraction of the few females that do reach the spawning areas from this time period dies unspawned. Thus, in years of the most extreme early entry, the combined annual en-route and pre-spawn mortality has exceeded 90% (e.g., 2000, 2001), whereas in other years of more normal migration behavior annual mortality rates as low as 10% (e.g., 2002) have been observed.

Though in most cases current spawning abundances are not at dire low levels relative to historical values, a declining trend has been observed in most populations since the abnormal migration behavior began. Declines have been very severe in Cultus Lake sockeye where the behavior and subsequent mortality are major factors threatening that stock's long term viability, prompting assessment as endangered by the Committee on the Status of Endangered Wildlife in Canada and recommendations for listing under Canada's Species at Risk Act. Conversely, Harrison River sockeye which migrate upstream the earliest have experienced record high

escapement levels in each of the last three years, exceeding historical averages by as much as 80 fold. The causes of these anomalously high spawning populations are unclear. Harrison sockeye are distinct in that unlike most Fraser sockeye populations whose juvenile rear for one year in a lake, juveniles rear in the Fraser River estuary and Georgia Strait. This estuarine rearing may expose and impart resistance in juveniles to the parasite that has been implicated as one causal agent in the en-route mortality of returning adults. Alternately the increased productivity of Harrison may be caused by other factors un-related to the Late-run problem.

The behavior and its consequences have created a major conservation issue for the Late-run populations. Managers have taken an exploitation rate approach, with the allowable rates on Late-run sockeye based on an attempt to balance conservation risks with the available harvests of co-migrating stocks. On years of relatively low Late-run abundance, exploitation rates have been limited to 15%. Beginning in 2003 models were developed to predict the median upstream migration dates based on in-season estimates of the proportion of the Late-run that had migrated upstream at any given point in time. These models, coupled with models that predict losses based on the median upstream date, have permitted more flexibility in years of larger Late-run abundance and resulted in larger exploitation rate limits in a few years. Lower exploitation rate limits have generally been applied to Cultus sockeye due to its low status.

The clear results from tagging studies that document extremely high mortality in the early migrants each year have led Pacific Salmon Commission staff to recommend potential use of these data in developing in-season harvest tactics. In particular, it would appear that harvesting fish in the Fraser River that are migrating upstream prior to mid-August would have limited negative impact on the number of successful spawners. The intent is not to suggest that all fish migrating during this period should be subject to very intensive harvest, but rather to point out that the tagging data can be used to accurately predict the impacts of harvesting fish in the Fraser River in August on spawning escapement. In contrast, the spawning outcomes of the current harvest practices which harvest the bulk of the fish in marine areas are much less predictable because in any given year a portion of the Late-run fish in marine areas may delay and experience low mortality, while another portion may not delay and experience high mortality. In addition to improved predictability of management impacts, a strategy of focusing fishing in the river in August would diminish fishing pressure on earlier timed stocks in marine areas, decrease the likelihood of excess escapements in co-migrating Summer-run populations and provide more predictable opportunities for Canadian commercial, recreational and First Nations harvesters. The associated shift toward towards fishing later in the migration when in-season assessments are more certain could also reduce risks of over-fishing.

However, implementation of such a scheme faces considerable impediments. First, more studies would be needed to document the intra-annual temporal pattern of mortality in Cultus sockeye which are being afforded extra protection. Second, implementation would be at odds with current domestic allocation policies in Canada that allocate more than 80% of the commercial harvest to areas outside the Fraser River. Third, it is already difficult to schedule in-river fisheries in a manner that provides opportunity to meet allocation targets for commercial and recreational fishers as well as priority obligations to First Nations for food, social and ceremonial purposes. Focusing more harvest in the river during the peak abundance periods would not alleviate and could exacerbate these scheduling and allocation issues. Fourth, some have expressed concern about impact of harvesting the small fraction of successful early migrants on long-term genetic biodiversity and resilience.

ORAL PRESENTATION—MIKE LAPOINTE

The slides referenced in this presentation follow the presentation text.

Lapointe pointed out the Late-run sockeye spawning areas in the Lower Fraser watershed (Slide 1), noting these fish return on four-year abundance cycles, with different stocks dominating each cycle (Slide 2). Historically, normal migration timing included a marine area peak in Johnstone Strait and Juan de Fuca Strait on August 17, with peak arrival about a week later at the mouth of the Fraser River (Slide 3). The fish would normally hold there for three to six weeks before migrating up-river.

Historical data for Weaver Creek sockeye migration past the Mission hydroacoustic station from 1977 to 1994 shows no migration in August and very little in the first week of September (Slide 4). Since 1995, however, every cycle line has dramatically reduced the length of delay at the mouth of the Fraser (average delay is down from 15–25 days to less than 5 days) (Slide 5). Migration timing of Weaver sockeye past Mission each year from 2004 to 2007 has occurred in August and early September (Slide 6). This new pattern shows very little overlap with the historically normal pattern of migration extending from mid-September through October (Slide 7).

Similar patterns of significantly earlier upstream migration since the mid-1990s have been observed for Cultus, Harrison and Adams sockeye (Slide 8). As with Weaver sockeye, the recent migration timing of Cultus and Adams sockeye in August and early September shows very little overlap with the historically normal migration timing in late September and October. The Cultus data are unique in that the migration timing comes from a spawning enumeration fence that has been operated at Sweltzer Creek just downstream from the outlet to Cultus Lake (Slide 9). Historical data from this fence suggest that the normal pattern of upstream migration occurred from the 1940's through 1994 (Slide 7). This early migration pattern has now held true over successive generations of parents and their offspring (Slide 10). Similar patterns of earlier migration are also being seen in other salmon species (definitely for Fraser pinks, possibly for chum and Chinook) (Slide 11, 12).

Consequences of early migration include mortality prior to spawning. En-route mortality (fish that die prior to reaching spawning grounds) is inferred most years by comparing lower and upper river estimates. These estimates are confirmed by tagging in some years. Pre-spawning mortality (fish that reach spawning grounds but die prior to spawning) is measured annually on spawning grounds, though the quality of estimates derived from carcass surveys varies depending on the timing and intensity of surveys.

En-route mortality for Weaver Creek sockeye since 1995 was inferred most years (though there was also some tagging) (Slide 13). The percentage of the run that suffered en-route mortality increased with earlier upstream migration timing. En-route mortality of zero to just over 20% was associated with the historical migration pattern before 1995. In subsequent years, the earlier the migration, the higher the mortality, with en-route losses of over 90% in 2000 when the median date of upstream migration occurred in mid-August. Since 1995, the pattern of en-route mortality in Adams has been consistent with Weaver Creek sockeye, with higher en-route mortality associated with earlier entry (Slide 14). However in some years of larger returns the fish have migrated upstream later and the en-route mortality has been relatively low.

Higher pre-spawning mortality has also been observed for most Late-run sockeye stocks since 1995 (Slide 15), coinciding with this earlier migration pattern (Harrison sockeye is an exception, with continuing negligible levels of pre-spawning mortality).

2002 Ashcroft tagging study: This showed that Adams/Shuswap females that entered earliest suffered 88% en-route mortality, compared to 0.5% for those that entered after September 18 (Slide 16). The same pattern was observed for pre-spawning mortality (Slide 17). So even though overall mortality was low (7%) for 2002 for the run as a whole, most of the earliest migrants (97%) either did not make it to the spawning grounds or were unable to spawn successfully (Slide 18).

There are some exceptions to this temporal pattern in mortality. For example, some early migrating Cultus sockeye were held for brood stock and spawned successfully. In 2006, pre-spawn mortality observed through periods when carcasses were samples on the spawning grounds, but data were not collected to link carcass recovery periods to upstream migration timing.

Population impacts: early predictions, assuming that these trends continued, were dismal. However, actual returns for 2002 and 2006 for Late Shuswap sockeye were significantly higher than predicted (5.5 and 3 million vs. less than 500,000 predicted) (Slide 19). For the Lower Adams, spawning populations on two cycle lines have increased in abundance while two have declined (Slide 20).

Late-run Harrison sockeye spawning escapements have increased dramatically on three cycle lines (Slide 21). Overall, spawning escapements to most of the non-Harrison stocks are declining on three of the four cycles (except 2006 when all are increasing) (Slide 22), while Harrison, which has much earlier marine timing and upstream migration than other Late-runs, has been producing record returns. Unlike the other Late-run stocks, Harrison has an ocean-type life history (juveniles rear in the estuary rather than in a lake). Resistance to the parasite responsible for mortality may be a factor and/or recent productivity may relate to other factors.

Management consequences: This early-entry behavior has created a major conservation issue for Late-run stocks and resulted in severe constraints on harvest. This has led to increased pressure on early-Summer-run sockeye, to escapements greatly exceeding targets for Summer-run stocks in some years and to very low harvests, especially for the Canadian commercial sector. Exploitation rates and marine area catches of all Fraser sockeye stocks have dropped dramatically from historical levels (Slide 23).

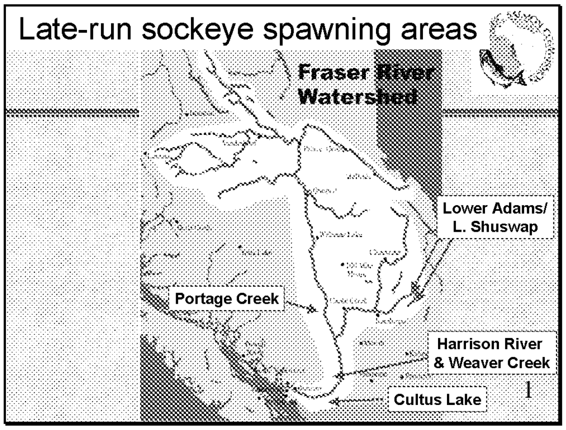
Since 2001, management approaches have sought to balance conservation risks to Late-run stocks against available harvests of co-migrating stocks, but no long-term stock viability or economic viability studies have yet been done (except for Cultus). On Weaver-dominant years, a 15% target exploitation rate has been used. Higher rates have been used on some Adams years, and a lower exploitation rate limit has been used for Cultus sockeye. New models are being developed to predict upstream migration timing in-season as the basis for adjusting exploitation rates (Slide 24, 25). Actual exploitation rates have been close to the targets in most years since 2001 (Slide 26).

Management opportunities: Harvesting Late-run sockeye in-river before mid-August would minimally impact the number of effective spawners. This would provide more stable and predictable fisheries for all sectors. But potential impacts on Cultus sockeye, catch allocations to outside fisheries and the ability to meet First Nations Food Social and Ceremonial fishing obligations would have to be evaluated.

Research is ongoing, but the key question remains unanswered: Why are Late-run sockeye migrating upstream early?

Slides

Slide 1



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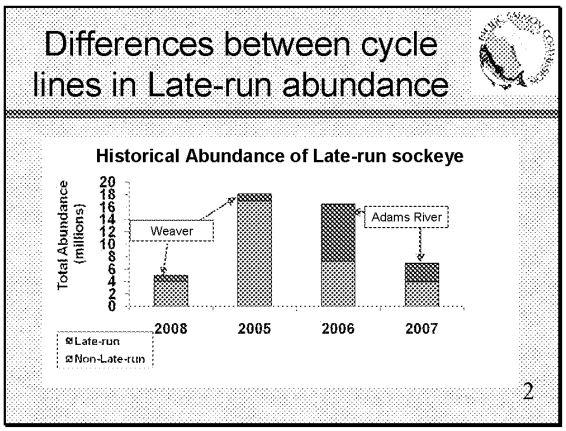

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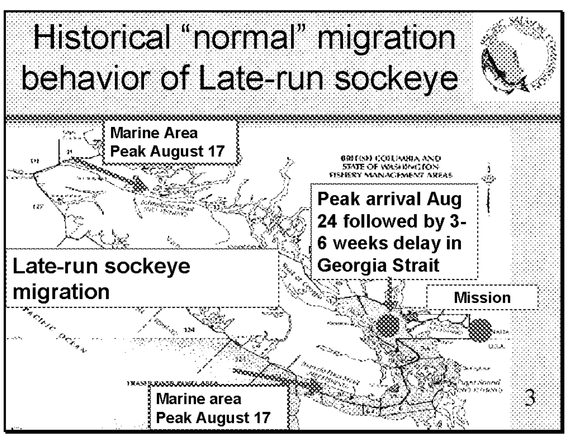

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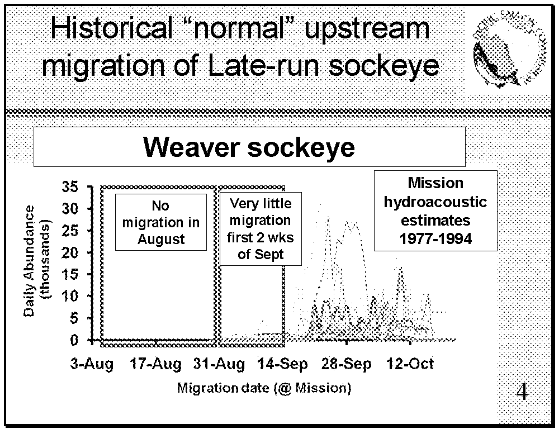

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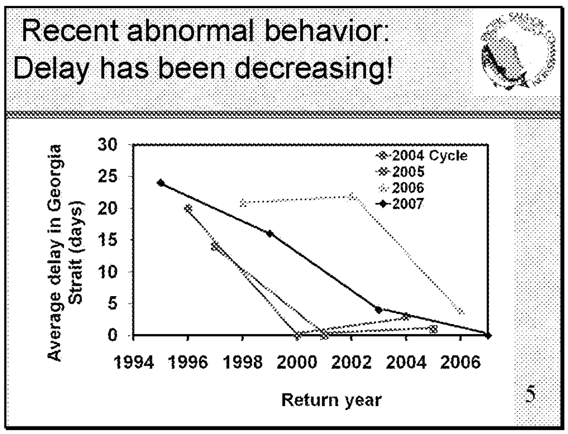

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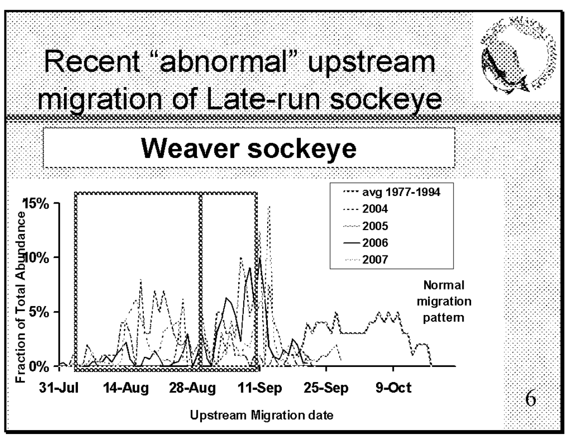

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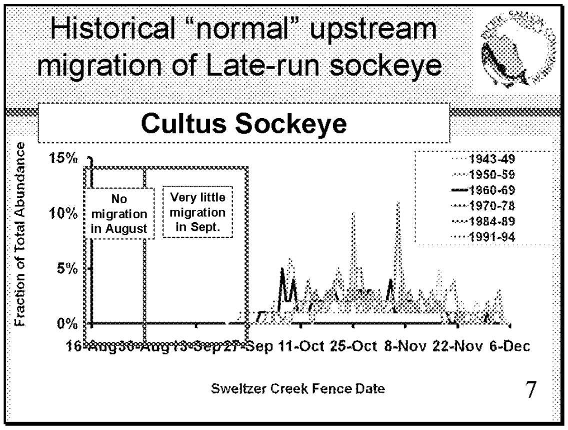

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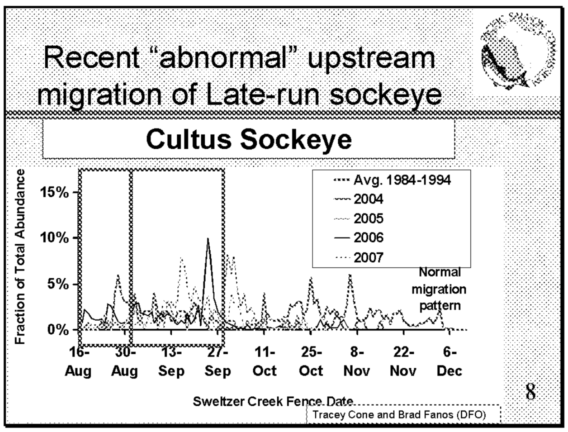

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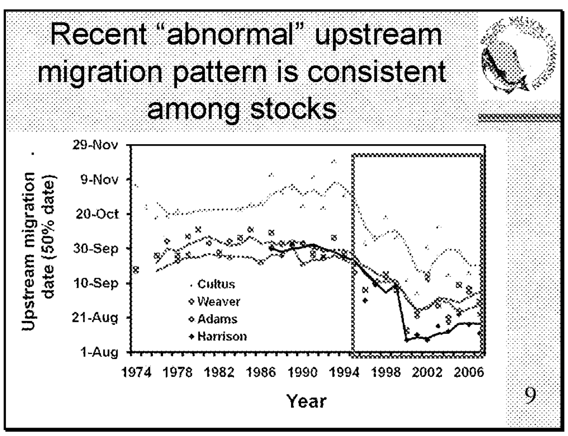

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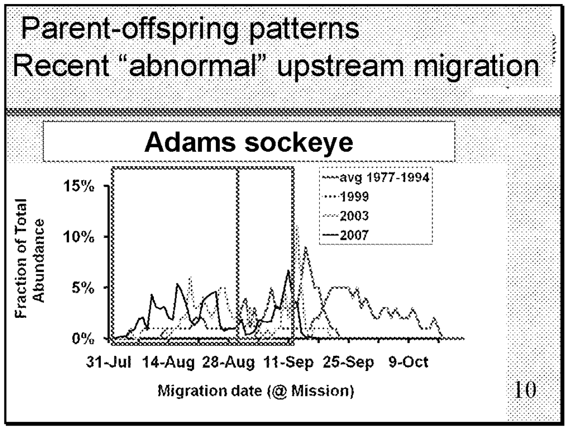

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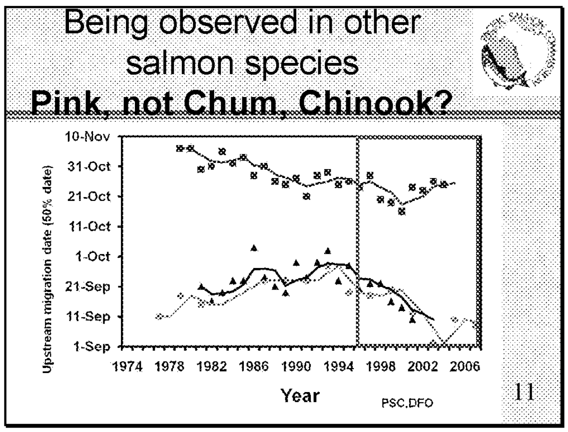

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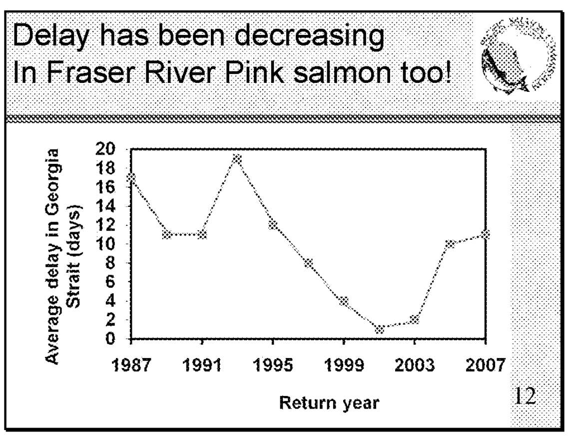

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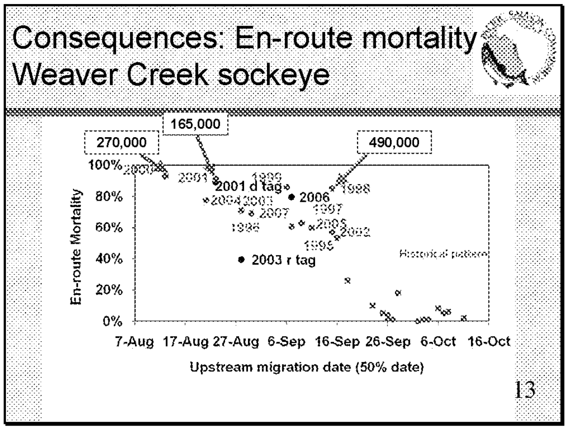

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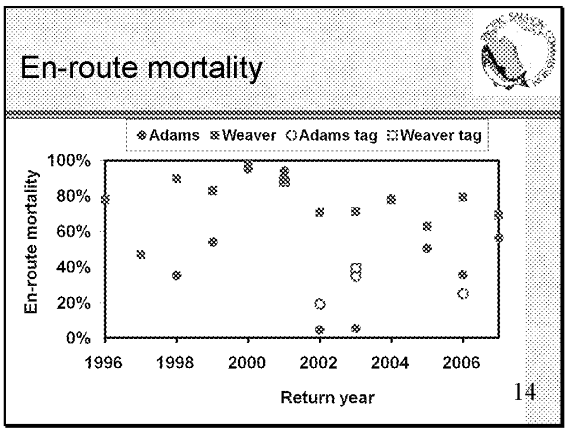

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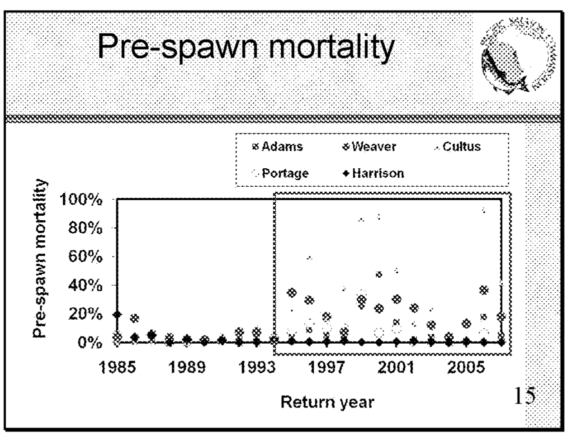

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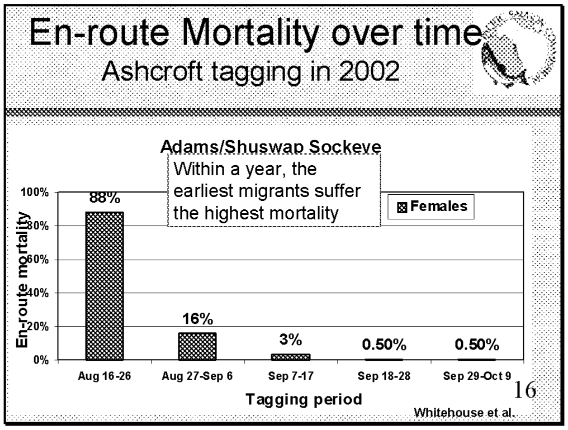

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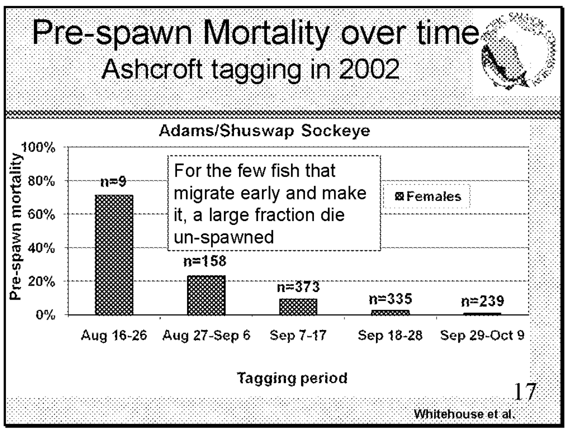

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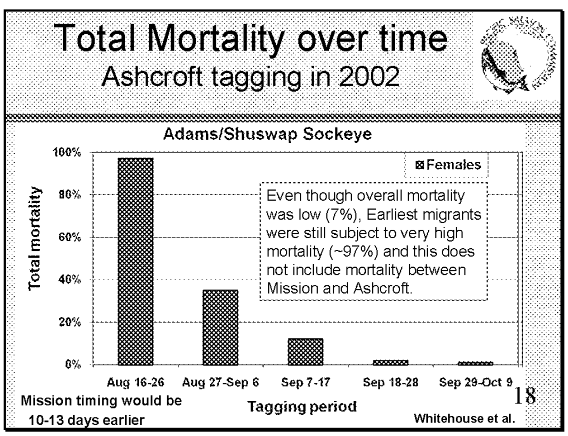

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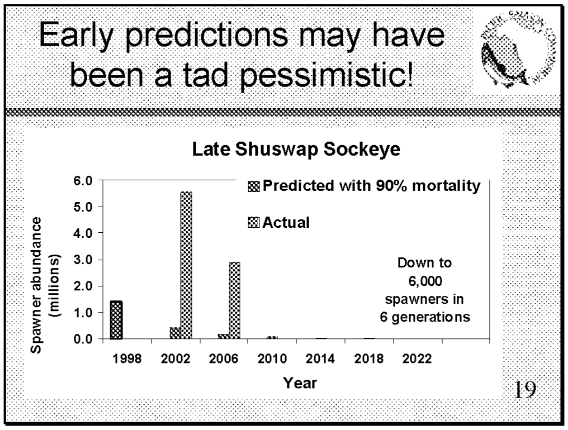

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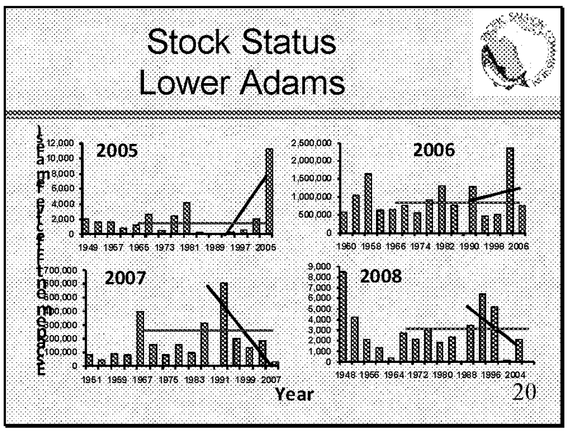

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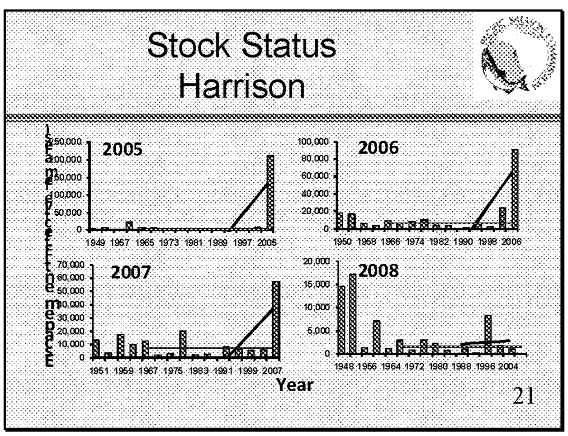

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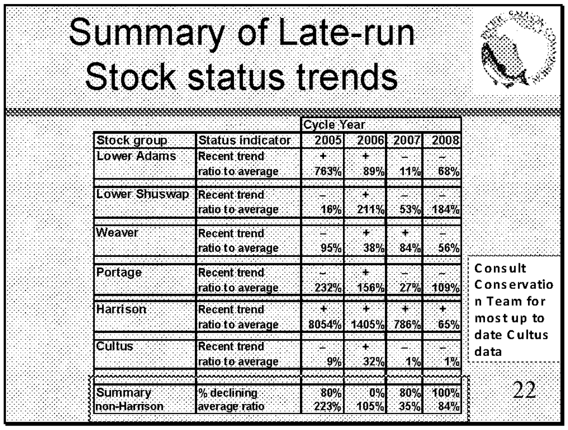

Summary of Late-run Stock status trends

		Cycle Year			
Stock group	Status indicator	2005	2006	2007	2008
Lower Adams	Recent trend	+	+	–	–
	ratio to average	763%	89%	11%	68%
Lower Shuswap	Recent trend	–	+	–	–
	ratio to average	16%	211%	53%	184%
Weaver	Recent trend	–	+	+	–
	ratio to average	95%	38%	84%	56%
Portage	Recent trend	–	+	–	–
	ratio to average	232%	156%	27%	109%
Harrison	Recent trend	+	+	+	+
	ratio to average	8054%	1405%	786%	65%
Cultus	Recent trend	–	+	–	–
	ratio to average	9%	32%	1%	1%
Summary	% declining	80%	0%	80%	100%
non-Harrison	average ratio	223%	105%	35%	84%

Consult Conservation Team for most up to date Cultus data

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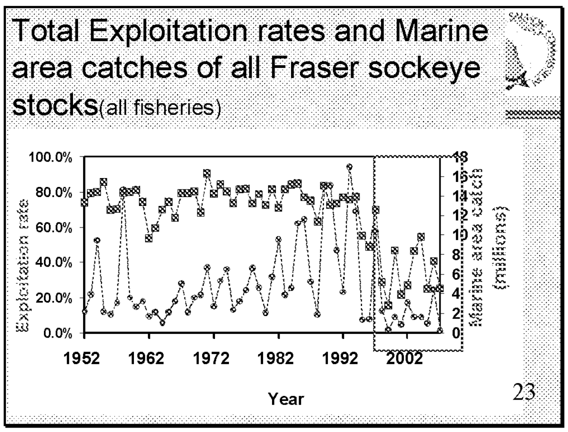
Slide 23



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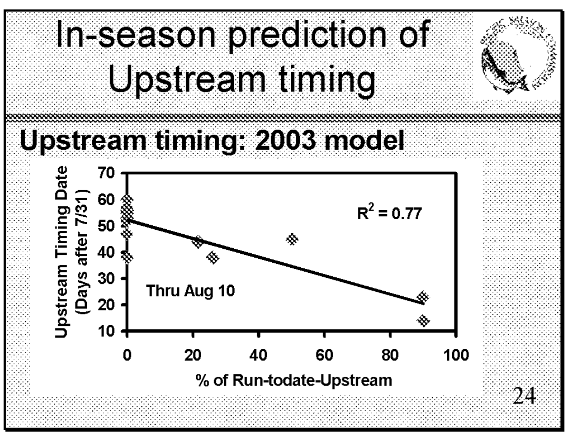

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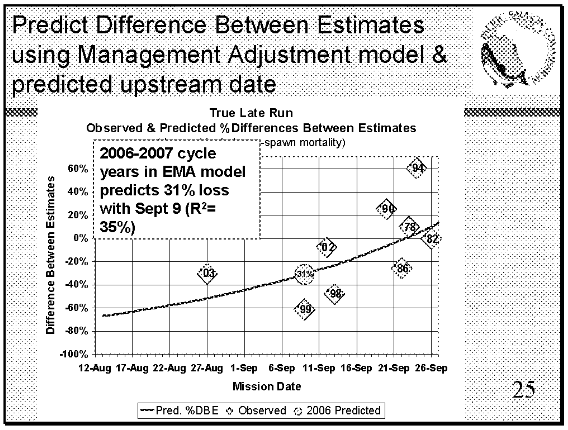

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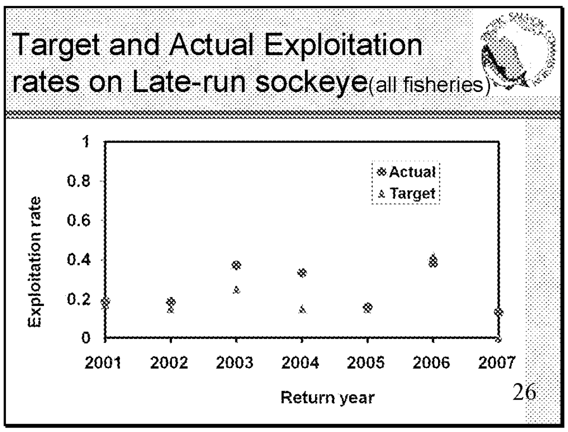

DISCUSSION

- Q/A (Lapointe): Further study is needed, but the little data available for Cultus sockeye supports a pattern of higher mortality associated with early migration.
- Q: How does the ability to distinguish Late-run fish affect the ability to detect early entry? / A (Lapointe): Historical migration data prior to 2000 was based on stock discrimination from scale patterns and since Early and Late Shuswap sockeye rear in the same lake, it is possible that some early entry of Late Shuswap could have occurred and gone undetected. However the data for Cultus sockeye come from an enumeration fence where only Cultus sockeye are migrating, so stock discrimination is not a issue. Thus the Cultus data provide very compelling evidence that the normal migration pattern was present going back to the 1940s.
- Q: What is the entry timing into Georgia Strait of Harrison juveniles? /A (Woodey): Generally, studies from the 1970s or 80s show a build-up until early June, then a decline after that in the area of Deas Slough. It looks like they rear for four to five months in the freshwater estuary and then move out into the Strait but there is no recent data to know whether there has been any change in out-migration timing.

- Q: How confident are we that arrival time in the Georgia Strait hasn't varied? / A (Lapointe): It has varied, but not enough. The recent early migration pattern bears no relation to marine arrival timing—it is linked to the delay at the mouth of the river and to less time spent holding there.
- Q: Is there data on pre-spawning mortality in pinks or other species? / A (Lapointe): There is a time series to 2001 for pinks, but not for chum.

PHYSIOLOGY 100: ADULT MIGRATING SALMON

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ABSTRACT

Central to an understanding of why biopsies are taken from wild, migrating adult salmon and what can be gleaned from these biopsies is the need to appreciate the basic physiology underlying homeostasis, regulatory systems and stress. Homeostasis can be simply viewed as a seesaw in perfect balance. Along with any disturbance to the balance (this can be an environmental change or a stressor) comes a physiological response to return the seesaw to perfect balance. Thus, both the change in variable and the response of the regulators can be used as an indirect indicator of an environmental change or stress. Deviations from the norm for body temperature, blood pH and blood glucose levels in humans (all good examples of homeostatically controlled variables) are routinely used to as indicators of health, which can of course be disturbed too much leading to death.

The challenge for migratory salmon is identifying: What regulated variables can we measure in adult salmon? What are the values? What are the expected changes, unusual changes and thresholds? How do regulators change and are they predictive? and What is the influence of sampling stress? The research performed so far has gone a considerable way to answering each of these questions. Biopsy techniques in conjunction with individual fish biotelemetry have sampled gill and blood tissues, as well as taking biometric measurements including a fat meter reading. As a result information was gained on certain regulated variables (blood glucose, plasma ions (Na^+, Cl^-, K^+), energy stores) and regulators (plasma hormones (cortisol, sex hormones), gill Na^+/K^+-ATPase, gene expression).

Terminal tissue samples, while precluding the possibility of following the fate of individual fish, do allow a broader range of tissue samples (gill, brain, liver, blood, muscles, gonads) and analyses. Some examples of the results were provided. For example, energy use is largely determined by routine metabolism (temperature dependent); swimming, defense and spawning activities; and reproductive development.

We have discovered that initial energy stores vary among stocks (related to migration distance and difficulty), reproductive state at a given spatial location affects energy stores, and a threshold energy level is need for successful spawning. The expected levels for gill Na^+/K^+-ATPase have been established and it was discovered that gill Na^+/K^+-ATPase is lowered in preparation for freshwater entry early in the sea migration. Temporal patterns for various sex hormones have been established and the hormones levels have been used to indirectly assign sex and to assess reproductive preparedness. Measures of stress indicate that we cannot biopsy fish without stressing them to some degree, but the level of stress is tolerated by sockeye salmon as many reach their spawning grounds. New approaches, such as heart rate biotelemetry, are also being developed.

ORAL PRESENTATION—TONY FARRELL

This presentation provided an introduction to relevant concepts in the physiology of adult migrating salmon, including homoeostasis, regulators and thresholds. It also reviewed research challenges and developments.

Homoeostasis is the process whereby key variables such as blood temperature, pH, glucose levels, plasma ions and energy stores are regulated to maintain balance. Hormones are examples of regulators that work to counteract the effects of environmental and stress factors and restore balance. If the imbalance in one or more variables passes a threshold of no return, the result is mortality.

Challenges in understanding these processes include discovering which regulated variables can be measured in adults (through biopsies or destructive sampling) and understanding what are their baseline values, expected changes and key threshold levels. Other questions include how do regulators change and how can the potential effects of sampling stress on measurements be adequately dealt with.

There are two approaches to studying regulated variables and regulators: 1) non-lethal tissue biopsy combined with biotelemetry for subsequent tracking to determine fate; and 2) spatial-temporal sampling freshly killed individuals. Key differences between these two approaches relate to knowing the fate of the individual samples, their relative costs and the extent of tissue analysis possible.

Why Measure These Variables?

Energy stores: These can be measured and graphed relative to sample date (location). Returning adults stop eating before they migrate upstream, so sufficient energy stores are required to last the journey and spawn successfully. Energy needs include maintaining routine metabolism (metabolic rate rises with the transition to warmer freshwater), swimming (need depends on distance and difficulty), defence (e.g., avoiding nets and seals), and spawning and reproductive (e.g., egg, sperm and gonad) development.

Research has discovered that initial energy stores vary among stocks (those that migrate further need bigger stores) and that reproductive state affects energy stores. Studies have also established the minimal energy threshold below which returning adults will not be able to complete the spawning cycle.

Gill enzymes and plasma ions: In the ocean, salmon absorb salt from their environment, so the gills pump out sodium ions to regulate internal levels. In freshwater, they lose salt instead, so the main gill enzyme responsible for maintain ion balance (ATPase) is down-regulated. Studies have established the expected ATPase levels during various stages of the migration, and in doing so found that the Fraser sockeye start to down-regulate gill ATPase levels long before they reach the river, starting near the Queen Charlotte Islands. This knowledge might be useful for making predictions.

Reproductive hormones: Studies have led to development of a series of profiles of expected levels of key reproductive hormones over the course of the migration (ocean, in-river and during spawning). These patterns can be used for sex determination or to assess the reproductive status of an individual. They may also have predictive value (e.g., progesterone levels have been linked with pre-spawning mortality).

Stress values: Studies of key stress hormones such as cortisol and lactate have been useful in identifying patterns associated with capture and handling, vigorous swimming and chronic stress. This information is useful to assess and correct for the confounding effect of sampling stress in further studies. A caveat is that stress levels can only be recorded for a single point in time because only one blood sample is taken. However, these stress hormones have a time-dependent trajectory post-stress and this needs to be considered with only one sample point on this trajectory.

A sockeye sampling study in Johnstone Strait showed lactate levels over time between capture and biopsy. The fish were tagged and released and a comparison of those that went on to enter the Fraser vs. those that were not detected entering showed that virtually all those with lactate levels over a certain threshold were not subsequently detected as reaching and entering the Fraser River. This suggests a maximum plasma lactate threshold beyond which mortality may occur, and this threshold level may be higher for sockeye salmon than the one proposed for rainbow trout in the literature.

New technology also brings new possibilities. For example, a heart rate and temperature logger can be implanted on fish released back into the wild, allowing tracking of oxygen uptake and energy expenditure under different conditions.

In summary, a suite of variables are now available to assess the physiological “state” of adults with respect to their preparedness for migration and spawning and stress levels. Scientists can monitor fish on their migration trajectory and also intervene with experiments that alter the trajectory and measure the consequences. The ideas developed are starting to produce results and suggest new possibilities, with ongoing development of novel approaches.

DISCUSSION

- Q: Were differences detected between males and females with respect to levels and thresholds linked to mortality? / A (Farrell): We are not there yet with respect to stress response. The only things consistently different are reproductive hormones and energy levels.
- Q: Would it be useful to look at other measures of energy stores besides lipids? / A (Farrell): Glycogen is another factor. However, migrating salmon do not deplete these until the last part of the migration (on the spawning grounds) except during stress when it is temporarily depleted and replenished as soon as possible (within hours). The glycogen stores are one of the last things they run down during migration.
- Q: Was there any evidence that fish were being mishandled enough during sampling to cause a lot of mortality? / A (Farrell): There did not appear to be evidence of this. Sockeye appear to be tougher than rainbow trout, which have been extensively studied in this regard.
- Q: How many of the fish that disappeared are we responsible for? / A (Farrell): Last year, the number of surviving fish post-handling was lower than in previous years, but our measures of stress were no different.
- Q: How do you distinguish routine from imposed stress? / A (Farrell): Studies have been done that hold the fish in the lab for a few days until they calm right down and then those levels can be used as a baseline for routine against which to assess an imposed stress.
- Q: Could the biopsies have contributed to mortality? / A (Farrell): All handling is potentially additive. Initially, we had various issues with tagging and biopsy on board a vessel, but fish handling and processing were then improved to get great survival rates. We now take the biopsy as soon as the fish is in the boat to minimize handling. Also, we do controlled experiments to maximize stress (e.g., swimming to exhaustion repetitively, near lethal temperature increases) and sockeye recover remarkably quickly. We have rarely seen plasma lactate higher than 25 mmol/L so we are comfortable that we are not maxing that. Once sockeye burn glycogen, they rebuild it.

LATE-RUN SPAWNING MIGRATION BIOLOGY: MARINE APPROACH AND RIVER ENTRY

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ABSTRACT

Prior to the investigations into the early entry behaviour of Late-run sockeye salmon that began in 2001 there was only limited information on the spatial trends on independent aspects of osmoregulation, reproduction and energetics. However, there was almost no information on the importance of temporal sampling or the interaction of reproduction and energetics in the interpretation of the biological condition of migrating sockeye. This short summary addresses this information gap and provides baseline spatial-temporal physiological information from which hypotheses regarding early entry behaviour can be evaluated against. In addition to being consistent with the biological information, all hypotheses regarding entry timing should not contradict these three basic statements regarding spawning migration timing: 1) spawning times have not changed; 2) marine approach times have not changed; 3) river entry time is earlier.

The general pattern of biological condition of sockeye salmon sampled through time at a fixed marine approach location and at river entry is as follows: there is a slow decrease in energy level with a concomitant increase in maturation status but no change in osmoregulatory capacity. This pattern is based on several thousand Late-run and Summer-run fish sampled at marine approach locations (Port Renfrew, Port Hardy, Port McNeill, Browns Bay, Georgia St.) and at river entry (Cottonwood and Whonnock) from 2002 to 2007. An organsimal physiological approach was taken whereby a variety of tissue samples were taken from the same fish. Osmoregulatory ability was assessed by examining plasma ions levels (e.g., Na^+ and Cl^-) as well as Na/K Gill ATPase activity. Energetic Status was determined by either whole body proximate analysis or fat probe values. Reproductive status was assessed by examining both reproductive hormone levels in blood plasma and by examining gonadal tissue mass relative to body size.

It has been well established that sample location, stock, fish size, and gender are important factors in analyzing body energy data and maturation data. More recently we have discovered the importance of sampling date relative to spawning date and reproductive status relative to final maturation size in interpreting the body energy status of an individual. Similarly, sampling date relative to spawning date and energetic status relative to date and location are also critical to interpreting maturation data. Therefore, energy and reproduction must be analysed in the context of space, time, and with respect to each other. The pattern that emerges from doing this is that for a given date fish are more reproductively advanced and have less body energy the closer they are to the spawning grounds. However, there is still large individual variation in energy and maturation status, as well as osmoregulatory preparedness, within a given sample date and location. This provides the potential basis for different entry behaviours and supports the research into physiological cues for predicting entry timing. In addition, this variance is likely the physiological basis for the different spawning times that are observed for co-river migrants. For example, early river entry can spawn late in the season and late entry fish can spawn early in the season. This information is supported by the telemetry data that shows potential mixing of river entry groups at the spawning grounds.

The information helps management by providing the biological framework against which can be tested competing theories on early entry behaviour. In fact, this information has already helped to eliminate some of the early hypotheses regarding biological condition of the fish and entry timing. We now have a legacy of

biological information on interannual spatial and temporal physiological information on osmoregulation, stress, energetics, and reproduction for Summer and Late-run stocks that has greatly expanded our knowledge of salmon migratory biology.

ORAL PRESENTATION—DAVE PATTERSON

This presentation sets an important foundation for the subsequent ones. Key points include the fact that spawning times have not changed and neither has marine approach timing. What has changed is river entry timing—Late-run sockeye are not holding as long outside the mouth of the Fraser River.

Extensive sampling of biological condition over time in the marine approach areas from early to late August shows a similar pattern as that described in the previous presentation: a slow decrease in energy levels, slow increase in reproductive maturation and no change in osmoregulation (plasma ions and Na/K ATPase). In marine areas Summer-run fish show a similar pattern to Late-run fish in temporal changes to physiological variables. At river entry, the biological condition of Late-run sockeye shows the same pattern.

Importance of energy level in relation to reproductive status and date caught: Destructive and non-destructive sampling was done between 2001 and 2007 at various locations from the Queen Charlottes to spawning grounds. Different tissue types from the same fish were tested for a number of variables. Key findings included the following:

- Marine approach osmoregulation: No temporal trend in plasma ion values of Late-run sockeye with sample date (same pattern was found in Summer-run sockeye and pinks).
- River entry osmoregulation: Large individual variability, but values typical of freshwater entry and no temporal trend.
- Marine approach energetics: Location relative to spawning ground, stock, fish size and gender are all well-established as key factors that relate to energy levels. Date of sampling relative to spawning date and reproductive status relative to final maturation date are emerging as novel key factors. Analysis of stomach contents shows that by the time the fish are approaching Vancouver Island they have stopped feeding and the energy budget is fixed. While there is large individual variability and there are differences relating to factors like stock and gender, the pattern of declining energy stores is consistent: the later you sample, the lower the body energy levels.
- River entry body energy: The pattern is similar to that in the marine approach (later sample date = lower energy), with typically large individual variability and a similar pattern among Summer-run and Late-run fish.
- Marine approach reproduction: Location relative to spawning ground, stock, fish size and gender are all well-established as key factors that relate to reproductive maturation. Date of sampling relative to spawning date is emerging as a key factor, and so is energetic status relative to date and location. Sampling of females shows that female ovary mass and testosterone levels increase by date, though there are large individual differences.
- River entry maturation: Similar patterns of increasing gonad size over time were seen for females, males, for other Late-run stocks and for Summer-run fish. Testosterone levels at river entry showed both males and females that entered the river early had lower testosterone levels than those that entered later. This suggests a “box car model” which applies to the fish seemingly moving together as a unit during river passage but not for maturation (i.e., different states of maturation for fish sampled on a particular date).

When the river entry fish are compared to those potentially holding in the marine environment at the same date, those in the river tended to have slightly lower energy values. As a general rule, fish sampled further upstream on the same date will likely have less energy. Fish in-river also tend to be more reproductively advanced than those further downstream or those potentially holding in the Strait of Georgia, though there is much variability.

These findings suggest that energy and reproductive status in the marine approach and river entry need to be analyzed in the context of place and time and each other. For a given date, fish closer to the spawning ground tend to be more reproductively advanced, though there is large individual variation and early entrants can spawn later and vice versa.

In summary, these findings provide a biological framework to test hypotheses against, and have already allowed the rejection of some hypotheses. Subsequent discussion of entry timing and in-river mortality should be consistent with this data set, which has extended the knowledge of salmon migratory biology. It has not answered all the questions but we are in a better position to say what is or is not normal in future.

DISCUSSION

- Q: Is there historical data on the stomach contents of fish holding in the gulf? / A (Patterson): No, but the sense is that they were not holding to feed.
- Q: The Georgia Strait troll data showed slightly higher energy levels for those fish compared to those caught in the river, but most of those would be early-entry fish anyway. / A (Patterson): It is an issue.

GENOMICS AND FATE OF LATE-RUN FRASER SOCKEYE: FUNCTIONAL GENOMICS 101 AND LARGE SCALE PATTERNS

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ABSTRACT

We used a functional genomics approach to identify the physiological shifts associated with migration. The salmon gene array experiments on gill, whole brain, white muscle, liver, and hypothalamus tissues have uncovered a large amount of new information on the physiological changes associated with adult salmon migration, covering a geographic scale from the Queen Charlotte Islands to the spawning grounds. White muscle is the main fuel source for migrating salmon once they stop feeding, and array data identified the Strait of Georgia as a major shift point in energy utilization consistent with the move towards a catabolic state, with a down-regulation in glycolysis and muscle protein transcription and up-regulation in proteolysis and aerobic respiration. Experiments on gill tissue, important in osmoregulation, disease and stress response validated the findings of direct physiological studies suggesting that osmotic preparation for FW occurs well in advance of arrival to the Fraser River. However, the array study also found that fish migrating in 2006 were more prepared for FW than those in 2005. There were also indications of disease in some fish migrating in SW in both years. Array studies in brain tissue identified strong diversion associated profiles, indicating that fish migrating through Johnstone Strait and Juan de Fuca Strait were coming from different water masses in the ocean. Importantly, these diversion-associated profiles remained with the fish all the way to the spawning grounds, and contained strong signals associated with differences in maturation and navigation. These profiles and their associations with diversion have been validated over two years (2003 and 2005). Interestingly, a refined focus on hypothalamus tissue did not reveal diversion-associated profiles, nor did it reveal strong patterns associated

with the maturation process. However, the hypothalamus did respond strongly to temperature, as noted from the differential response in Savona, a high water temperature region of the river. These are the largest scale "wild ecological genomics" studies carried out in any species to date, and illustrate the immense capacity for gaining physiological insight that this technology has to offer.

ORAL PRESENTATION—KRISTI MILLER

This presentation explained the use of functional genomics technology in several studies to better understand the physiological processes that shift during the course of migration, the relative impact of genetic variation (i.e., patterns linked to stocks or run times) and environmental triggers. This work was part of the larger collaborative study and based on the work done at DFO's Pacific Biological Station molecular genetics laboratory, which has included development of genetic baselines and markers for management purposes like stock identification.

Functional genomics focuses on RNA-level differences instead of DNA, highlighting which genes are expressed or "turned on" to indicate proteins and pathways that may be active. Such analysis can advance understanding of how organisms control their physiology at the molecular level, whether in their development, behavior or response to the environment. It can also help to predict adaptive capacity, fitness and behaviour. The technique used for gene expression profiling uses cDNA microarrays. Individual samples are compared to reference controls to provide a ratio of the level of gene expression in each sample, relative to the control. Further analysis is then done to highlight which genes are differentially expressed, what their functions are and thus what physiological pathways and responses are active.

The three main goals of this work are: 1) to identify environmental and/or physiological cues associated with altered freshwater entry timing; 2) to determine why fish are dying prematurely in-river and whether some fish arrive at the river with compromised fitness; and 3) to reduce the potential genetic variables to a few key genes that can serve as predictive biomarkers for entry timing and spawning success.

The sampling in these studies targets multiple tissues (liver and white muscle, brain and hypothalamus and gill) for different signals associated with energy utilization and reserves, reproductive maturation and freshwater preparedness. Three studies were done involving over 750 slides, using samples from a variety of field locations since 2003. The third and broadest study looked at spatial variation, run time, year, fate, entry time and sex.

Microarrays can provide information at three key levels: 1) Significance and pattern: relationships between physiological distinctions and experimental variables; what happened as the salmon migrated and where the major switch points are; 2) Molecular control of physiology: which genes and pathways are linked to the experimental variables; and 3) Biomarkers: identification of highly differentiated genes that can be used as tools to predict a certain state.

Findings: Looking at genetic variation for different stocks, run-timing groups, tissue type, sex and spatial variation, the key message was that spatial variation was a major factor. Migrating fish undergo profound physiological changes over their final 1,300 km journey, whereas genetic and sex-based differences were minor. Dramatic shifts in gene expression were seen with spatial variation in all tissue types surveyed, with the greatest shifts occurring in brain and white muscle tissue. As expected, major gene expression shifts were associated with freshwater entry and at the spawning grounds. An unexpected finding, however, was the highly differentiated profiles seen within the marine environment.

Hierarchical clustering of genes that showed significant differences in expression in the 2005 study helped to identify different patterns in gill, brain and liver tissue that were associated with different groups of fish. Gill tissue provided three distinctive profiles for JS (Johnstone Strait), JDFS (Juan de Fuca Strait) and W (Whonnock/freshwater) fish. Brain tissue provided a highly distinct profile differentiating JS fish from W/JDFS fish, while liver tissue provided gene expression patterns that differentiated marine from freshwater fish.

Sampling in 2006 again showed a different expression pattern in gill tissue for samples from the different locations, plus differences between those in the marine environment and those in fresh water. Patterns for white muscle tissue showed a physiological shift occurring at the Strait of Georgia estuary and less distinction between JS/JDFS fish, while hypothalamus expression patterns showed no change after a physiological shift just south of the Queen Charlotte Islands (QCI).

Energy reserves: Migrating adults use the lipids and proteins stored in white muscle as fuel. The study shed light on some of the key changes that occur along the migration journey, in terms of the processes that affect energy reserves, and also identified a number of genes that correlate with energy reserves for development as potential biomarkers.

Osmoregulation: Gill was the only tissue for which there were two years of samples for comparison, and there was a big difference between the expression patterns for the two years. In both years, however, only moderate shifts occurred upon freshwater entry, supporting the hypothesis that osmotic changes for freshwater occur out in the ocean, well in advance of arrival at the mouth of the Fraser.

Gene expression relating to osmoregulation showed high variability, with no strong spatial effects. There was a relatively strong signal associated with osmoregulatory preparation between years, and although the most significant genes were not generally the ones that would have been expected based on smoltification research, they showed that fish were more osmotically prepared for freshwater in 2006. Key genes involved in high salinity osmoregulation were down-regulated for fish from all sampling locations, including the QCI, and most notably in Johnstone Strait, for both Summer- and Late-run fish in 2006 compared to 2005. This suggested greater freshwater influence in 2006, which was confirmed by oceanographic data.

Immune response: Comparing the differences in immune response from the 2005 and 2006 gill tissue samples, there was a differential marine immune signal between years, and in 2005, between FW sites, suggesting that some migrating fish may have been responding to a pathogen, possibly viral. The immune signals in 2006 correlated with enhanced osmotic signals.

Diversion-associated profiles: Expression patterns observed in brain tissue were especially vivid, and showed powerful marine diversion-associated profiles linked to JS (the inner coastal route of passage around Vancouver Island) vs. JDFS (the outer route) fish. These patterns were consistent for 2003 and 2005 samples. Importantly, whereas the patterns were strongly linked to diversion, an admixture of the two distinctive expression profiles was observed in QCI, suggesting that the physiological distinctions were already present before fish diverted. Furthermore, an admixture of these two distinct profiles was also observed all the way to the spawning grounds.

Reproduction: Very large differences in signals linked to reproductive maturation were observed within these two patterns. Fish migrating through the inner passage (JS) had higher levels of expression linked to estradiol all along the migration route; for fish taking the outer passage, those levels increased only when the fish reached the spawning grounds.

Navigation: This represented another major difference between these two groups. Fish taking the inner passage were extensively using visual and olfactory cues to navigate through Johnstone Strait and all the way to the spawning grounds. Long-term memory genes were down-regulated, apart from one gene that is involved in both memory and learning. Another memory gene was not turned off until this group reached Johnstone Strait. Fish taking the outer passage used memory and circadian rhythm more extensively while migrating offshore and through Juan de Fuca Strait.

Supporting these findings were gene expressing patterns suggesting differences in the metabolic processes and energy sources utilized by these two groups of fish. The study also found that as few as six key genes could be used as biomarkers to predict whether a sample belonged to the inner or outer passage profile and thus which of the two migration routes they would likely have taken. The biomarkers were accurate until fish reached the spawning grounds, where some further refinement is required to address the significant shifts that occur there.

In summary, migrating adult salmon are undergoing profound physiological change. The environmental cues triggering physiological processes vary. Fish are prepared for freshwater well in advance of arrival at the river, but the degree of osmotic preparation varies by year. Biomarkers to gauge osmotic preparation were identified. Salmon were not energy limited in 2006 but underwent profound shifts in muscle metabolism, initially triggered by arrival in the Strait of Georgia estuary, and then again on the spawning ground. Biomarkers for gross somatic energy were identified, with 40 highly correlated genes involved in glycolysis, muscle contraction, aerobic respiration and proteolysis. Strong diversion-related profiles were revealed in brain expression patterns, largely influenced by differences in maturation processes and navigational strategies (sensory cues vs. memory). Biomarkers for diversion were identified, with as few as six genes required.

DISCUSSION

- Q: Whether looking at muscle or other tissue, given the profound differences between the rates of transcription, doesn't that confound what you are using as a control to allow comparison across sites? / A (Miller): These are not absolute values of expression; they are ratios. The control is a combination of all samples used in the experiment, and is merely used to create a ratio. The control itself has no biological relevance, other than as a constant divisor that allows standardization across slides. Many of the genes are not changing and significant results have validated via quantitative RT-PCR and based on other physiological measures.
- Q: Much effort has been devoted to looking at the possible role of factors like salinity levels in the Strait of Georgia but this suggests the fish have already made up their minds and that environmental factors in the Strait don't play a role. / A (Miller): There may also be environmental factors involved. We think gene expression is affected by the environment but long before they reach the river.
- Q: We are not seeing differences in mortality between fish that migrate through Juan de Fuca vs. Johnstone Strait. / A (Miller): In some years, that may be the case, while in others, perhaps there are differences, and through use of the "diversion" related biomarkers, we could classify successful and unsuccessful spawners based on the ocean routes taken in years where we do not have radio-tagging data. Importantly, we have observed differences in immune stimulation in fish in the marine environment in both 2005 and 2006 stemming from expression patterns in the gill, and in some years, these patterns could be associated with ocean migratory routes.
- Q: Regarding the difference between reliance on visual vs. memory cues, those fish that travel through Johnstone Strait face a very different environment. / A (Miller): Yes, and are likely stimulated by the greater influence of FW salinity and olfactory cues. But the point is that fish were already physiologically predisposed to take one route or the other, suggesting that the differences derive from salmon migrating through different ocean water masses before they reached QCI. We hypothesize that fish taking the inner passage may have used a more coastal route of return, while those taking the outer passage may have migrated a longer distance offshore.

- Q: In the 2005/06 comparison, were the results repeated, given the timing of the return was anomalously late in 2005? / A (Miller): We ran analysis for both Summers and Lates for 2006 and compared it to 2005 Summers, and it didn't matter. Osmotically, the Summer-run fish are a bit more prepared. They came back late and more mature but not as osmotically ready in 2005.
- Q: These studies just look at gene expression; we don't know what the cost of that behavior was. / A (Miller): True.
- Q: These findings relate to the regulatory system and show what is going on when the fish has switched off its machinery for survival in saltwater but still has to survive there. So this identifies things that are critical to their ability to hold. / A (Miller): Yes.

EARLY MIGRATION OF SOCKEYE SALMON: PHYSIOLOGICAL AND ENERGETIC CORRELATES OF BEHAVIOUR AND FATE IN COASTAL ENVIRONMENTS

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ABSTRACT

Physiology and energetics are integral for long distance fish migrations, particularly those involving a transition between marine and freshwater systems and reproductive maturation. By coupling assessments of animal position, movement, behavior, and survival with non-invasive physiological and energetic sampling, it becomes possible to test hypotheses associated with animal condition, behavior, and fate. The ability to link individual behavior and fate with energetics and physiology has eluded researchers for until recently.

Although the concept of releasing transmitter-implanted animals that have been biopsied for physiological variables is simple and the potential insight invaluable, there are only a few published examples where this approach had been employed to study fish migration, all within the last few years. Indeed, it is possible to rapidly and non-lethally collect a number of tissue samples including blood (usually via caudal puncture), gill tissue (using surgical tools), muscle (using muscle biopsy punch), fin tissue, and scales. Cooke *et al.* (2005) demonstrated that with practice it is possible to rapidly sample multiple tissues from un-anesthetized, transmitter-implanted (gastrically) fish without deleteriously affecting animal condition or post-release behavior. These tissue samples can be analyzed for a variety of parameters using a number of techniques including blood and muscle biochemistry (e.g., metabolites, ions, acid/base status), hematology (e.g., hemoglobin, hematocrit),

endocrinology (e.g., hormone titres associated with reproduction or stress), genetics (e.g., stock identification; see above), genomics (e.g., gene expression; see below), and stable isotopes (see below). In addition, a number of new tools enable the non-lethal measurement of energy density using microwave signals.

As part of the Late-run studies, we have used this biopsy technique to sample fish in the Pacific Ocean and then to evaluate the physiological and energetic correlates of fate and behavior. One of our hypotheses explaining why Late-run sockeye salmon were entering the river early was that they had a different physiological status. Specifically, we predicted that abnormally early migrants would have advanced maturation status (as assessed by reproductive hormones), be prematurely prepared for entry to freshwater, and/or have low energetic status. Any of these physiological differences could motivate the fish to enter the Fraser River and reach spawning grounds as quickly as possible. Indeed, our first prediction was correct in that abnormally early migrants tended to have elevated levels of reproductive hormones, including testosterone, 11-ketotestosterone, and estradiol. However, since gill $Na^{+}K^{+}$ ATPase was not significantly different in early migrants, although more variable from normal timed migrants, we found limited evidence to support our second prediction regarding premature preparation for freshwater entry. Using biopsies from fish tagged and released in the ocean, we found evidence that fish which failed to enter freshwater were characterized by elevated levels of stress indicators (e.g., elevated plasma lactate, glucose, and cortisol), whereas fish that were tagged during freshwater migrations but failed to reach the spawning grounds had lower gross somatic energy and higher levels of plasma reproductive hormones than those that reached spawning grounds.

ORAL PRESENTATION—STEVEN COOKE

This presentation discussed data from a 2003 telemetry study that focused on the marine near-shore coastal environment from Johnstone Strait to Mission. The study sought to link individual variation in behavior and survival of migrating adults with physiological and energetic status, recognizing that results could only show correlation, not causation.

Telemetry provided a proven tool to study fish behavior. Sampling included blood, gill biopsy and energy density measurement (using a "fat meter"). Fish were captured by purse seine, individually netted and held in tanks. Blood samples were taken within 30 seconds and pliers were used for the gill biopsy. The sampling procedure was very quick, while the fish was held in a foam-padded trough with water pumped through constantly. Finally, a transmitter was inserted into the stomach (gastric insertion).

Testing was done to demonstrate the ability to perform non-destructive biological sampling on migrating adult salmon. Preseason assessments were conducted using sockeye from an early Summer-run, with one group tagged and the other tagged and biopsied. The fish were then transported to holding pens in Sooke Harbour and monitored. The assessment found no mortality or tag expulsion at 24 hours and no evidence of abnormal behavior during retention.

In season, further field assessment was done using sockeye captured by commercial purse seine over three test periods. Half the fish were biopsied and tagged and the other half just tagged, and the fish were then released in Juan de Fuca or Johnstone Strait. Migration speed and survival between ocean release and river entry was identical for the fish that were and weren't biopsied.

The sampling procedure was thereby validated and has since been modified to make it better and faster. Sampling now also includes scale removal for stable isotope analysis and white muscle tissue samples. Data from studies that used this approach have been summarized in three published papers.

2003 radio tagging study: Fish were tagged in Johnstone Strait and data collected at a series of receiving stations along the migration route. Findings included:

- Migrating radio-tagged fish suffered almost 40% losses prior to reaching Mission, which was associated with higher stress measures (higher osmolality, lactate, glucose, cortisol).
- Fish that did not hold in the Strait of Georgia and which had low energy and high levels of reproductive hormones had higher mortality.
- Late-run fish that hold for less than 7 days were found to have less energy than those that hold for more than 20 days. The difference was more pronounced for Weaver than for Adams sockeye.
- Females that do not hold are more reproductively advanced (estradiol, 11-KT and testosterone levels).
- Early entrants had more variable levels of osmoregulatory preparedness compared to those that entered at the normal time, though there was no difference in the mean (worthy of further investigation).

2003 acoustic telemetry study: Different fish were tagged from the same platform at similar times to the radio-tagged fish. Findings included:

- Reproductive hormones (T) were the strongest physiological correlate with travel time. Fish that come in early move quickly and have higher levels of reproductive hormones.
- Longer holding in Georgia Strait most highly correlates with low reproductive hormone levels in females. Long holding also correlated with high osmolality, Cl^-, glucose and lactate.

The study found differences in survival between radio- and acoustic-tagged fish. Losses between release and Mission was estimated to be 37% for radio-tagged fish compared to 15% for acoustic tagged fish.[7] The two most likely explanations for this difference in survival are the length of holding period prior to release and differential exposure to fisheries. The radio-tagged fish were held for about an hour before release, unlike the acoustic-tagged fish which were released immediately. Radio-tagged fish were subject to higher fisheries influence due to timing of the tagging (August 11–28) and the greater fishing pressure on those fish radio-tagged from August 11–15, prior to the release of any acoustic tagged fish. All the acoustic-tagged fish were released between August 20 and 27.

In summary, fish that die before reaching the river tend to have evidence of stress (some of which may be due to handling). Fish that do not hold tend to have higher reproductive hormone levels, more variable osmoregulatory status and lower energy levels. These were all just correlations, but information from the 2003 study set the stage for refining/developing testable hypotheses.

DISCUSSION

- Q: Was there any comparison of the 37% losses to the river to unhandled fish? / A (Cooke): It is very hard to determine what the level would have been for unhandled fish. The 37% mortality rate was not adjusted for fishing, so we know some were caught. Natural mortality would have to be less than that. / A (Farrell): We know we are causing some of the problem, but we are not sure how much. We were surprised at how well the fish survived, given the parameters we measured.

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[7] K. English notes the reported survival rates from release to Mission numbers were 56% (i.e., 44% mortality) for radio-tagged fish and 81% (19% mortality for acoustic tagged fish (English *et al.*, 2004).

- A (Cooke): There was a companion study two weeks later, with less handling and when the fisheries had stopped briefly, where we saw a 15% loss. So that could be an upper level estimate of what we are doing in the best of circumstances. / A (Cooke): Only QCI fish in the 2006 study had radio tags surgically implanted (because they were still feeding). / A (English): The actual numbers for 2003 were different for the different timing groups. The first group had the highest mortality (54%) compared to 32–34% for the later two release groups.
- Q: How do you estimate survival—do you assume 100% detection at Mission? / A (Cooke): Yes.
- Q: Dead fish found on the spawning grounds showed no evidence of advanced reproductive development, whereas your data show early entrants have more reproductive development. / A (Cooke): Later presentations will show that these fish don't tend to reach the spawning grounds. Early entrants can spawn at any time, but the first to arrive aren't necessarily first to spawn.
- A (Cooke): In earlier years, the fish were still silver bright. The data suggest inter-annual variation in timing relative to normal. They are still dying but perhaps in some years at more advanced physiological states.
- A (Cooke): The evidence is now showing very clearly that fish entering early are more advanced through a series of measures, but you have to get around the problem of not being able to kill the fish. The spawning date is not changing, so they either need to slow down the trajectory or get to the spawning grounds and wait. Studies to date have narrowed the variables and more will be presented on the in-river and spawning ground information tomorrow.

GENERAL DISCUSSION

- Cooke: Considering the cost of energy expended when fish are holding vs. the energy costs of migrating upriver, how long can they afford to hold?
- Patterson: It doesn't matter where they spend their energy because they either need to hold upriver or at the spawning grounds. So they're deciding where to wait.
- Farrell: The big difference is temperature; they can stay in the lower, cooler water or warmer water.
- LeBlond: All this conveys the rather simple idea that many sockeye turn up at the river with sexual maturity but not enough energy and feel they have to move up but cannot make it because they are too weak.
- Cooke: It is not just a matter of being too weak, when you pile on the effects of higher temperatures, fishing and other stressors.
- Cooke: So this suggests that something is causing the fish to show up in a different state.
- Farrell: If the fish come in from the ocean and know when they need to spawn but are running out of gas, they need to make a decision.
- Cooke: It is not clear that they're coming in with less energy. Maybe they just burn off more energy because they enter when it is warmer.
- Cooke: It is possible to use energy levels from mid-Summer-run fish as controls. I don't think you would see that difference across all years.
- Cooke: The Late-run sockeye appear to have taken a different strategy. Their arrival and spawning times have not changed—it is the behaviour in between that is different. The Lates evolved to delay and thus spend less energy when they do migrate. Now there is a significant difference from the previous patterns because even when the fish have entered later in recent years and had better survival, it was still earlier than the previous pattern.

2006 OCEAN BIOPSY TELEMETRY: PHYSIOLOGICAL CORRELATIONS WITH MIGRATION TIMING AND MARINE FATE

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ABSTRACT

Despite recent advances in the bio-telemetry of adult salmon, very few studies have examined the physiology of salmon at sea, and fewer still have sought to examine the physiological bases of ocean behaviours. Here we experimentally test the hypothesis that energetic and reproductive processes in the ocean are the key drivers of migration timing into freshwater and ultimately successful upriver migration to spawning areas. The recent and radical change in migration timing exhibited by some segments of Late-run populations of Fraser River sockeye salmon (*Oncorhynchus nerka*) provides a context in which to address this hypothesis. We use the acoustic telemetry to measure the migratory fate of individual animals, and couple this information with non-lethal biopsies to examine the mechanistic bases of observed homing behaviours.

Homing Adams River sockeye salmon were intercepted 1) near Rennell Sound, off the west coast of the Queen Charlotte Islands, and 2) in the marine approaches to the Fraser River at the northern and southern ends of Vancouver Island. Both locales were well before their arrival at the natal river mouth during reproductive migration. Over 500 salmon were non-lethally biopsied for blood plasma, gill filament tips and gross somatic energy, and then were implanted with acoustic transmitters before release to continue migration. Migratory behaviour was monitored as salmon navigated over 800 km of coastal shelf en route to the Fraser in the former study, and >200 kms in the latter study.

The physiology of salmon at time of capture at QCI is new information about the baseline physiology of ocean salmon, and population-specific variation in somatic energy, reproductive investment were similar to patterns observed 800 kms later as fish begin upriver migrations. Adams sockeye sampled at the northern (Johnstone Strait) and southern (Juan de Fuca Strait) of Vancouver Island show distinct physiological differences between sampling locales, particularly indicators of stress (increases in plasma glucose, lactate, and ion concentrations in the former). In Johnstone Strait where the majority of Adams sockeye were intercepted, gross somatic energy, plasma testosterone, and gill Na^{+},K^{+}-ATPase activities were all found to vary significantly with location and Julian day of sampling, suggesting a seasonality driven by biological rhythms.

Both river entry timing and subsequent ability to reach spawning are strongly related to (a) relative energetic condition and (b) degree of sexual maturation and osmoregulatory preparedness. Salmon that delayed river entry and reached spawning areas had high somatic energy, low testosterone levels, and low gill Na^{+},K^{+}-ATPase

activities. Salmon that entered the river directly and subsequently failed to reach spawning areas had lower energy, higher testosterone, and higher gill Na^+,K^+-ATPase. Once in river, sockeye from the latter group swam at significantly faster rates than the former (~15.5 km·day^{-1} vs. ~20.0 km·day^{-1} respectively). We conclude that hormonally mediated processes tied to biological clocks are important regulators of migration timing and success. This has been observed in other salmon studies, but surprisingly, few studies of this phenomenon exist for other animals.

Physiologically, fish that failed to enter the river where characterized by high stress (plasma lactate and glucose) and ionoregulatory measures (plasma Na^+, Cl^-, osmolality).

High mortality was observed in the QCI study (~95% overall) and in Johnstone/Juan de Fuca Straits (~55% to Mission, BC). At QCI, mortality was likely attributed to a combination of natural, tagging and handling, and fisheries-related mortality, though we cannot determine the degree to which each of these played a role. Assuming that handling-related mortality would occur after 1–2 days from the time of release, the mortality of 17–22% in JS and JDF is generally consistent with previous studies. The remaining >30% could thus be attributed to fisheries, which were active in the Lower Fraser River and in JDF during our study, and natural mortality.

ORAL PRESENTATION—GLENN CROSSIN

Previous studies found that migration timing was positively related to reproductive hormone levels and that success was negatively related to stress measures. Crossin and colleagues tried to build on these findings in two 2006 bio-telemetry studies. The aim was to test the hypothesis that somatic energy, reproductive hormone levels and freshwater preparedness all influence survival to the Fraser River and entry timing, by examining patterns of survival to the Fraser River and physiological correlates of river-entry timing.

Study 1: Fish were captured, sampled and fitted with tags in Johnstone Strait and Juan de Fuca Strait. Acoustic and radio receivers were used to track migration patterns and survival to the river mouth and then right up to the spawning grounds. The second study focused on the role of reproductive hormones as potential drivers of ocean migration rates and river-entry timing. Fish were captured near the Queen Charlottes, injected with hormones and fitted with transmitters.

Results for the first study included the following:

- Mortality from Johnstone Strait for 2006 was higher than in 2003. For 2006, there was 22% mortality to northern Georgia Strait and an additional 14% loss to the river mouth, with a total 48% mortality to Mission. In 2003, a similar study found 15% total mortality, which might have been a best-case scenario. It is thought that most of the early deaths were linked to handling. Harvest could also account for a portion of remaining mortality.
- For fish tagged in Juan de Fuca Strait, they found 17% mortality to the first receiving line and a further 30% to the river mouth (more fisheries were happening on this route), with 65% mortality overall to Mission.
- Survival per day was very similar for both routes (92% for JS vs. 91% for JDF fish).
- Adams sockeye tagged in Johnstone Strait that failed to enter the river had had higher levels of stress and osmolality than those that did successfully enter the river.
- Timing of river entry by Adams sockeye was related to reproductive hormones and freshwater preparedness. Females that did not hold had higher levels of testosterone and fish that did not hold had higher levels of Gill ATPase compared to those that did hold before entering the river.

Conclusions: Survival of Adams sockeye during marine migration to the river was related to stress (higher lactate, glucose, Na^{+} and Cl^{-}) and entry timing was strongly correlated with testosterone and gill ATPase (early entry associated with high testosterone and high gill ATPase). This suggests that early-migrating fish were motivated but not physiologically prepared for freshwater entry.

Study 2: 196 salmon were captured by purse seine between July 31 and August 6 (mix of Adams, early-Summer-run and Summer-run stocks). They were sampled and surgically fitted with radio tags and injected with hormones. Controls were injected with saline solution instead, and then the fish were released and tracked. Results included:

- Survival rates between the Charlottes and the river mouth was very low—only 12 fish made it to Vancouver Island and only four were detected in the Fraser River. However, survival per day was lower than in the previous study and there were certainly fishery impacts.
- High testosterone level before treatment was highly correlated with faster travel time. Low chloride level was also correlated with faster travel times.

Conclusions from the two studies: Testosterone is an important driver of migratory behavior (timing). High female testosterone equated to fast river entry and high testosterone equated with fast ocean travel times. Preparedness for freshwater was also important. High gill ATPase equated to fast river entry but poor survival. Low chloride levels equated to faster ocean travel times.

DISCUSSION

- Q: In the first graphs, is there more detail regarding the spread? / A (Crossin): There was a lot of variation but there was still a difference.
- Q: Were survival rates corrected for tag failure or removal rates? / A (Crossin): The results were adjusted for fish from harvest. We don't think there were tag issues.
- Q: Was any comparison done between fish that had radio vs. acoustic tags? / A (Crossin): There were no differences among them at time of capture or subsequently.
- Q: Regarding stress indicators relative to marine survival and hormone levels relative to river survival, was there any relationship between stress and hormone levels? / A (Crossin): No, but it raises interesting questions.
- Q: It is interesting that you can detect differences between fish that were stressed and entry timing, given the number taken by fisheries. / A (Crossin): Fisheries have different influences in different years. There was not much impact in 2003, but more in 2006. You need to look at each year.

RELATIONSHIPS BETWEEN SITE-SPECIFIC WATER PROPERTIES AND TRAVEL TIMES OF SOCKEYE SALMON FROM MARINE AREAS INTO FRESHWATER WITHIN A SINGLE YEAR

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ABSTRACT

The osmoregulation hypothesis of early-river entry behaviour of sockeye salmon postulates that if individuals experience a strong freshwater signal (i.e., low salinity areas) during their movement through the Strait of Georgia (SOG) then their physiological preparations for entry to the freshwater of Fraser River will be accelerated resulting in earlier dates of river entry compared to con-specifics which did not encounter low-salinity conditions in SOG. The hypothesis is based on empirical correlations that have been detected, using interannual data, between the proportions of Late-run sockeye which migrate in-river early and indices of coastal salinity and wind stress (see Thompson and Hourston presentation). There has been no evaluation of whether daily variability in oceanographic conditions, within a given year, may affect migration rates and river entry times of adult Fraser sockeye.

In this study, migrating sockeye salmon (two populations, Adams and Chilko) were captured, tagged and released with implanted acoustic or radio transmitters at locales in Juan de Fuca Strait (n = 30 Adams, 13 Chilco) and Johnstone Strait (n = 138 Adams, 14 Chilco), approximately 300 km from the Fraser River mouth. We used a CTD device to collect water temperature, salinity and oxygen data from two depth zones: shallow (3, 5 and 7 m sample depths) and deep (20, 35 and 50 m sample depths). We sampled oceanographic conditions (i.e., CTD casts) at sites where fish were collected (n=76). The CTD casts were always executed prior to deployment of fishing gear. Each cast collected measures (twice each meter along the depth profile) of water temperature, salinity and oxygen and within each depth zone, mean values were calculated (n=3) and then used for subsequent analyses to relate in situ oceanographic conditions to sockeye travel times between the tagging locales and the lower Fraser River. Sampling started in Juan de Fuca Strait and continued in Johnstone Strait during three successive time periods in August when sockeye were abundant in the SOG. During that time window, we found that salinity and oxygen levels increased with sampling date probably due to decreasing freshwater discharge from the Fraser River. Water column properties at Juan de Fuca Strait revealed that temperature and oxygen decreased and salinity increased with depth, whereas temperature, oxygen and salinity were more evenly distributed with depth (i.e., mixed water column) in Johnstone Strait.

Because sampling occurred over a three-week period during which oceanographic conditions were changing seasonally, the effect of Julian day of sampling on migration timing needed to be examined and its influence accounted for in order to understand how any additional variation in oceanography may affect migration timing. We conducted a series of ANCOVAs, with Julian day as the covariate, and temperature, salinity and oxygen as

separate explanatory variables. Release areas (JDF, DP) and sampling depth (shallow and deep) were all considered in separate analyses. The class variable in each analysis was based on travel time for fish to migrate from a given release site to Mission. Specifically, we categorized fish into 'fast', 'intermediate' and 'slow' migration groups using the 25th and 75th percentiles. For Adams fish, individuals within the 25th percentile represent fish that reached Mission in < 10 days (the fast group), and individuals within the 75th percentile represent fish that reached Mission in > 15 days (the slow group) after being released in the SOG. Fish taking 10–15 days to reach Mission were classified as the intermediate group. For Chilko fish, individuals within the 25th percentile represent fish that reached Mission in < 7 days (the fast group), and individuals within the 75th percentile represent fish that reached Mission in > 9 days (the slow group) after being released in the SOG. Fish taking 7–9 days to reach Mission were classified as the intermediate group. We interpreted the Type III sums of squares from each analysis thus were able to examine the effects of 'behaviour' (e.g., travel speed class) independent of the effects of Julian date of sampling.

We found that Julian date of sampling was a very important predictor of the variation in oceanographic measures at both sample sites. In particular at Johnstone Strait, Julian date was significant ($P < 0.05$) for all oceanographic measures at both depth classes with the exception of the deep oxygen class. After accounting for variation explained by Julian date (e.g., using Type III sums of squares), there were few significant relationships (all $P > 0.05$) between travel speed class and oceanographic variables measured at either depth zone or release site. Small sample sizes of both populations at JDF and for Chilko fish at Johnstone Strait severely reduced our ability to detect effects. We found a nearly significant relationship ($P = 0.067$) between travel speed class and salinity at Johnstone Strait for Adams fish. Fast Adams migrants experienced lower mean salinity (29.7 ppt) than either intermediate or slow migrants (29.8 ppt). This is a relatively small difference and whether this has biological meaning is uncertain; however other marine organisms can resolve salinity differences smaller than this. Thus the hypothesis that within-season variation in salinity may either trigger sockeye to migrate into Fraser River faster, or that sockeye which may have been exposed to areas of lower salinity earlier during migration are pre-disposed to seek lower salinity along the coastal migration route, cannot be fully rejected. However, there is no clear 'smoking gun' in terms of strong correlations between within-season oceanographic properties and sockeye travel rates into freshwater.

ORAL PRESENTATION—IVAN OLSSON

Background: Multiple year studies suggest sockeye migration timing for entering the Fraser River is negatively correlated with salinity in the ocean (years with lower salinity = early migration). This study looked at whether salinity differences in the ocean in 2006 affected travel speed and migration timing on an individual level. Water properties were collected at the tagging locales and migration was monitored to Mission in the lower Fraser River.

Sampling was done during three periods in August at Juan de Fuca Strait and Johnstone Strait. In total, 195 fish were tagged (mostly Adams and some Chilko; the majority were at Johnstone Strait). Water property sampling was done before each fishing set, using a CTD sampler that repeatedly measures temperature, oxygen and salinity at different depths in the water column. Water properties correlated with sampling date, with salinity increasing over the month of August (presumably due to decreased Fraser River discharges during this period). Water property variables were also analyzed and related to travel time data, with sockeye divided into three groups based on travel times (< 10 days, 10–15 days and > 15 days).

Conclusions: Temperature, oxygen and salinity were more homogenous in Johnstone Strait than in Juan de Fuca in terms of depth distribution. Salinity was strongly correlated with sampling date. After correcting for the influence of Julian day we found that fast migrants (Adams fish only) were associated with lower salinity in

Johnstone Strait; however this relationship was only marginally significant (P = 0.067) and the absolute difference in mean salinity measures were very small (~ 0.1 ppt difference). If this relationship is not spurious, these findings suggest several "chicken or egg" questions. Are fish triggered by certain levels of salinity exposures in Johnstone Strait (the differences were very small—are they even biologically meaningful?)? Or are fish that are exposed to lower salinity earlier during migration predisposed to seek for lower salinity in Johnstone Strait? The fact that we uncovered no strong relationships between oceanographic features and travel times suggests that other factors are probably playing a larger role in within-year variation in travel rates from our capture sites (e.g., individual physiology or behaviour).

DISCUSSION

- Q: With which travel time was intermediate salinity associated? / A (Olsson): There was no 'intermediate' salinity. Fish were grouped by travel times (fast, intermediate, slow) and we compared these groups to the salinity levels at the capture sites.
- Q: The freshwater signal comes from the surface, so any attempt to group it dilutes that signal. If you make the groups smaller, are the differences more obvious? / A (Olsson): We don't think there is a difference. Fraser River water goes down to 70–100 m. deep.
- Q: Has anyone looked at depth profiles for tagged fish? / A (Olsson): Some depth tag data will be discussed tomorrow.

A MATTER OF TIMING: OCEANIC CONDITIONS AND MIGRATION INITIATION OF LATE-RUN FRASER SOCKEYE

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ABSTRACT

The initiation of in-river spawning migration by Late-run Fraser River sockeye salmon (*Oncorhynchus nerka*) changed abruptly in 1995. To account for this change in timing, we have examined correlative links between observed fish arrival dates at Mission (peak and 50%) and ocean conditions encountered by the migrating stocks up to several months earlier. For each stock, we have used the nearly constant marine arrival date as a common reference time for the lagged regressional analysis. The regressional analysis examines correlations over a broad range of lag times and data averaging intervals for both spatially gridded and site-specific data series.

Preliminary findings reveal that the observed times of peak and 50% river entry at Mission in September are correlated with the principal component of wind stress in the northeast Pacific in July. There is also significant correlation between river entry timing and surface salinity in Juan de Fuca Strait in late summer, indicating that the river entry timing of Late-run stocks is strongly determined by oceanic conditions encountered by the fish during both the offshore and inshore stages of inbound migration. Fish migrating in offshore waters at times when the winds are blowing counter to the prevailing surface currents enter the river earlier than those migrating when winds are in the direction of the surface currents. Once the stocks enter Johnstone and Juan de Fuca straits, salinity becomes the dominant environmental factor affecting in-river entry timing such that fish encountering lower than normal salinity enter the river earlier than fish encountering higher than normal salinity.

We speculate that endogenous energy reserves gained by fish swimming in the ocean during times of weaker than normal or reversed prevailing currents are used to advance maturation and osmoregulatory adjustment.

This effect can be further modified by salinity conditions as fish transit the brackish inner coastal waters, such that lower than normal surface salinity advances osmoregulatory adjustment of the fish. It may be possible to use wind stress data from the northeast Pacific in July and August to predict in-river migration timing of Adams stocks at Mission in late August and early September. For Weaver stocks, timely salinity series for Race Rocks will also be needed for in-season predications.

ORAL PRESENTATION—RICK THOMSON

As explained by previous speakers, there was a big shift in return timing in 1995, with earlier peak migration timing up-river past Mission for all Late-run stocks compared to typical migration timing going back to the 1940s. There has been no trend or major shift in the marine arrival date for these same stocks (Cultus Lake, Adams River, Weaver Creek).

This project sought to establish possible causal relationships between migration timing of Late-run Fraser sockeye and oceanic properties observed along the inbound migration routes and to address the question of whether environmental factors have affected the observed shift in migration timing since 1995. The approach used statistical methods to examine correlation between migration initiation timing and environmental time series along inbound migration routes, offshore and in coastal waters.

For migration timing, we used both peak and 50% migration data interchangeably—it made little difference to the results which of the two were used. Environmental data included daily records of ocean wind stress, currents, surface and subsurface salinity and temperature, coastal sea level and Fraser discharge, plus derived time series such as temperature and salinity variance, surface mixed layer depth, coastal upwelling index, etc.

The fundamental idea tested is that returning stocks are affected directly or indirectly by oceanic conditions along their migration path and that succeeding environmental effects along the migration path can enhance but not diminish the impact of each preceding step (i.e., once the physiology starts to change, there is no stopping it). Furthermore, ocean properties must be averaged over migration periods of days to weeks, with periods defined relative to the marine arrival date (50% entrance at Juan de Fuca Strait) for the stock. If ocean properties have a negative lag relative to the Marine Arrival date, then the ocean leads the fish; if ocean properties have a positive lag relative to the Marine Arrival date, then the ocean lags the fish.

Gulf of Alaska: This analysis found a strong correlation between winds in the Gulf of Alaska and arrival timing at Mission up to 40 days later. The stronger the oceanic winds in mid-July blow counter to the prevailing surface currents in the NE Pacific, the earlier the fish arrive at Mission and vice versa. No significant correlation was found between sea surface temperature in the Gulf of Alaska and arrival time at Mission.

Outer coastal region: Arrival timing was linked to northwest (upwelling favourable) winds occurring 20 to 28 days before the arrival date. The stronger the winds in late July blow counter to the prevailing surface currents off the BC coast, the earlier the fish arrive at Mission. Again, no correlation was found between arrival timing and sea surface temperature in the outer coastal region, except for a very small area off the Alaskan panhandle.

Inner coastal region: Arrival timing is related to minimum salinity (maximum temperature) at Race Rocks from 0 to 8 days after the Marine Arrival date. The lower the salinity (higher the temperature) in Juan de Fuca in early August, the earlier the arrival date at Mission.

Strait of Georgia: There was no strong correlation with temperature, salinity or Fraser discharge, but there was marginal correlation with winds such that wind blowing towards the river correlated with early migration.

In summary, arrival time at Mission appears to be significantly correlated with winds in the NE Pacific in mid/late July. Since these winds serve as a proxy for surface currents, it is concluded that fish migrating in offshore waters when winds are blowing counter to the prevailing surface currents will enter the river earlier than normal. Adams fish are affected by both offshore and nearshore coastal currents; Weaver stocks are only affected by nearshore coastal currents. The year 1979 was exceptional, with very strong northward coastal winds during the fish migration period leading to the latest recorded peak fish return timing at Mission.

Once the fish enter Juan de Fuca Strait, salinity becomes the dominant environmental factor affecting entry timing. Fish encountering lower than normal salinity enter the river earlier. Adams River stocks appear to be more affected by ocean currents than by salinity, while with Weaver Creek stocks the opposite occurs. The stronger the winds in the Strait of Georgia are directed towards the river delta in August, the earlier the river entry.

Conclusions: If the fish are swimming against weaker than normal surface currents during migration, they save energy, which may be used to advance maturation and osmoregulatory adjustment. Salinity encountered within inner coastal waters also affects timing, such that lower than normal surface salinity leads to accelerated osmoregulatory adjustment and earlier than normal river entry. Weaker than normal prevailing ocean currents, combined with lower than normal salinities in inner coastal waters since 1995 have advanced fish maturation and adjustment to freshwater, causing stocks to enter the river earlier than normal. This change in behavior has not benefitted spawning success and management of Late-run stocks.

DISCUSSION

- Audience member comment: In a recent publication on Pacific salmon, a Russian researcher concludes, based on 20 years of research, that migration isn't related to sea surface temperature.
- Q: Stronger ocean winds and currents could also mean more mixing and productivity—did the study look at chlorophyll levels? / A (Thomson): We haven't looked at that but it is a potential factor.
- Q: It is not just wind speed but the question of variability vs. steady winds which might affect productivity. / A (Thomson): We haven't looked at that. We looked at different time windows to see which gave the maximum correlation.
- Q: How are the currents helping them? / A (Thomson): Timing is everything. They may be swimming more slowly and still getting there at the same time.
- Q: How many environmental variables were examined? / A (Thomson): Air and water temperature, wind speed and direction, wave variables, SST, blended buoy data, coastal data, current meter data, current velocities and lighthouse data.
- Q: Do you have correction factors? / A (Thomson): We weren't doing a principal component analysis. / A: (Woodey): The data show nothing like this in the earlier period. We can provide peak timing at Mission back to the 1930s for Adams. It is not the same quality of data, but we can give the peak date of arrival for Adams on the dominant line years for a long period. We have gone through regime shifts but whether this is related to a regime shift, we don't know. But we are looking at just one period vs. multiple periods—more data would give us a better sense.

TEST OF 'STAY WITH THE SCHOOL' HYPOTHESIS: 2002, 2003 AND 2006 RADIO-TELEMETRY DATA

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ABSTRACT

Several alternative mechanisms have been proposed to explain the recently observed 'early river-entry' behaviour of Late-run sockeye. One, the "Stay with the School" (SWTS) hypothesis, was tested using river-entry timing data derived from three years of ocean tagging in Juan de Fuca and Johnstone straits, where 912 radio-tagged sockeye were tracked from release to Mission, BC. The SWTS hypothesis states that the early entry behaviour of Late-run (LR) sockeye may result from these fish migrating with large schools of Early summer and Summer-run (ES/SR) sockeye, which enter the Fraser River with little or no delay (English *et al.,* 2005). If the SWTS hypothesis is correct, one would expect a larger portion of the LR sockeye to enter the Fraser River without delay when ES/SR stocks are numerically dominant in the marine tagging areas. Note that this theory does not require individual or groups of sockeye to remain in the same school throughout their return migration. It is expected that schools of migrating sockeye will form and fragment numerous times due to interactions with other schools, fishing gear, predators and other factors. We simply suggest that each LR sockeye that survives to the mouth of the Fraser River will likely be associated with a school of sockeye when its upstream migration is initiated and its behaviour at that critical decision point will be influenced by the behavior of the majority of the sockeye in the school.

The SWTS hypothesis was tested using a logistic regression model constructed to predict the proportion of LR sockeye with 'early river-entry' behaviour from the relative abundance of LR and ES/SR sockeye in the strait. The logistic regression results supported the SWTS hypothesis. There was a highly statistically significant trend ($P <$ 0.0001) showing a decreased probability of LR early entry with increased proportions of LR in the strait (χ_1^2 = 187.8). Both parameter estimates in the logistic regression model were highly significant (Slope = -0.07; SE = 0.01; $P < 0.0001$. Intercept = 0.89; SE=0.15; $P < 0.0001$). This "Stay with the School" hypothesis, combined with the much lower in-river survival rates for Late-run sockeye that enter the Fraser River early, has significant implications for fisheries management and in-season assessments for Fraser sockeye stocks. Estimates of the L:S ratio can be derived from pre-season forecasts and the in-season analysis of DNA samples. These ratios combined with the relationship defined above can be used to forecast the proportion of Late-run sockeye expected to enter without delay, and hence the expected pre-spawn in-river mortality rates. Similar predictions were derived for 2005, 2006 and 2007 using the SWTS Model developed in 2005 (English *et al.,* 2005). The main difference here is that the relationship between the Late-run early entry behaviour and the relative abundance of Late and Summer-run sockeye has been estimated directly from the accurate measurements of travel times from marine release sites to river entry (measured at a consistent location) for 912 Fraser sockeye tracked in the 2002–2006 marine tagging studies.

Reference

English, K.K., Lapointe M.F., Forrest K., and J. Cave. 2005. Influence of Summer-run sockeye on the river entry timing of Late-run Fraser sockeye: "Stay with the School" Hypothesis. Prepared by LGL Limited for Pacific Salmon Commission, Vancouver, BC. 46 p.

ORAL PRESENTATION—KARL ENGLISH AND DAVE ROBICHAUD

This presentation briefly reviewed migration components and the rationale for this hypothesis, and then described how it was tested.

The three major components to migration timing of Late-run sockeye are: 1) migration timing through approach areas of Johnstone Strait and Juan de Fuca Strait, 2) migration delay off the mouth of the Fraser (only the Late-runs do this), and 3) river entry timing.

Arrival at the approach areas is likely determined by large-scale oceanographic conditions in the Alaskan gyre. Small changes in delay behavior could be caused by subtle changes in a variety of factors, such as environmental conditions, arrival timing and fish behavior. However, the large changes observed in recent years are likely tied to a major change in key factors that influence sockeye behavior. River entry timing is likely affected by a whole suite of factors, including biological and environmental factors.

Sockeye have a strong affinity to stay in a school. This hypothesis suggests that when summer stocks are much more abundant and run timings overlap, a large portion of the Late-run will school with the Summers and enter the Fraser in August together with them. DNA analysis in 2000 and 2001 revealed that small Late-runs had migrated through coastal waters with more abundant Summers and all the Late-run fish entered the Fraser in August with the Summer-run stocks. Abundance of Summer-run sockeye increased substantially for three of the four cycle years from 1978 to 2005. For Shuswap (dominant cycle year), Summer-run abundance was greater than that of the Late-run for the first time in 1994 and 1998. Different results were seen in 2002 and 2006 when the Late-run was relatively more abundant again. For the sub-dominant cycle years, summers were more abundant than Late-runs after 1991.

To test the hypothesis that early migration is affected by the relative abundance of co-migrating Summer stocks, travel time data were used from 912 radio-tagged sockeye tracked in 2002, 2003 and 2006. Relative abundance estimates for Late- and Summer-run sockeye in Lower Georgia Strait were reconstructed using escapement, catch and test fishery data. The proportion of Late-run fish that entered the Fraser with the Summers was estimated for each release group. A model was then constructed to predict the proportion of Late-run sockeye with "early entry" behavior based on the L:S ratio for Lower Georgia Strait. Adjustments were also made to account for typically slower migration speed among Late-run sockeye.

The results strongly suggest that "stay with the school" behavior was at play. The higher the proportion of Late-run fish in Lower Georgia Strait, the lower the probability of early entry among Late-run fish. Both parameters in the model were highly significant, regardless of the threshold used to define early-entry behavior.

A model using daily estimates of abundance for Summer-run and Late-run sockeye for 11 years between 1990 and 2003 was developed in 2004 to provide a prediction of the portion of the Late-run return that will enter the Fraser River in August with the Summer-run stocks. This model was applied to 2005, 2006 and 2007 in-season abundance data to estimate how many would enter early. The model predictions of Late-run entry timing were consistent with the estimates derived from post-season analyses but could not explain all the variation in run timing observed in these years. For example, the return timing for Summer-run stocks in 2005 was a full three weeks later than the average run timing for these stocks. The model correctly predicted that all Late-run sockeye would enter with the Summer-run stocks in 2005 but most of the sockeye run entered in September in that year. Clearly, no single factor can explain all the variance in river entry timing for Summer-run or Late-run stocks.

Conclusions: Inter- and intra-annual variation in the abundance of Summer-run sockeye relative to Late-run sockeye appears to account for some of the observed variability in early-entry behavior for Late-run sockeye. The 'Stay with the School' model provides a tool for managers to use pre-season and in-season to forecast the portion of Late-run sockeye that may enter the Fraser in August with Summer-run stocks. Further work is required to identify the key determinates of the overlap in return timing for Summer and Late-run stocks and factors that determine river-entry timing for Late-run stocks in September.

DISCUSSION

- Q: The PFRCC has emphasized the importance of an ecosystem approach, so it is good to see this focus on interactions between stocks and species. / A (English): Something else must have happened around 1995 to explain why this has become more of a factor.
- Q: The relative abundance of Summers relates to things like hatchery production and studies in Alaska showed similar effects. You need to be able to eliminate the effect of date to show the effect of Summer-runs. / A (English): We have run similar analyses after extracting the effect of date. The results were not as clear but there was still a significant relationship between the L:S ratio and the portion of the Late-run sockeye that enter the Fraser River early.
- Q: There must be something else happening. Maybe fish were misidentified but historically there may be years where the data can reject the hypothesis. The question is what else has changed to promote this behavior? / A (English): There is also a changing ocean environment. There are two separate issues—abundance and overlap. Historically, Cultus is the latest run timing, with the least overlap with Summer-run stocks. Also, different stocks may have migrated at different depths and thus had less influence on each other's behaviour due to separation within the water column. Recent environmental changes could be concentrating fish in a portion of the water column.
- Q: As attractive as this is, it has evolutionary consequences. There are few examples in the natural world where you follow someone to their death. We know there are physiological correlates. It has to be behavioural, not genetic. Those that are doing this are not spawning, so they are being eliminated from the gene pool. / A (English): Yes, the SWTS hypothesis is a behavioural hypothesis based on the premise that the schooling behavior combined with the increasing abundance of Summer-run sockeye has influenced the behaviour of Late-run sockeye.
- Q: What might be the role of mammal predators? / A (English): We have eliminated whales as predators, but seal and human populations are both up.

EXPERIMENTAL TEST OF THE OSMOREGULATION HYPOTHESIS FOR THE ABNORMAL MIGRATION TIMING OF FRASER RIVER LATE-RUN SOCKEYE

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ABSTRACT

Prior research into the shift to earlier than historic river entry timing by Late-run sockeye has demonstrated a significant relationship between salinity in the Fraser River estuary (a.k.a., the Strait of Georgia) and timing of river entry, in that lower salinity correspond with earlier dates of river entry. Concurrently, physiological telemetry studies have demonstrated that early migrants had higher concentrations of sex steroids (i.e., testosterone, 11-ketotestosterone, 17β-estradiol) and more variable osmoregulatory capabilities (i.e., gill Na^+, K^+ ATPase activity), relative to "normal" timed migrants. The osmoregulation hypothesis stems from these early findings, and it postulates that the shift in migration behaviour is a response to acceleration of the physiological preparations for freshwater entry caused by exposure to low salinity waters while still in the marine realm. The hypothesis explicitly incorporates state of sexual maturation as an important consideration, as it proposes that more sexually advanced individuals are more susceptible to low salinity cues and therefore more prone to early river entry, owing to interactions among the maturation and osmoregulatory physiological pathways.

Maturation is under the control of the Hypothalmus-Pituitary-Gonadal (HPG) axis, and activation of the HPG axis occurs well before FW entry. In brief, the maturation process involves the release of gonadotrophic releasing hormone (GnRH) from the hypothalamus, which ultimately causes increases in circulating levels of testosterone and its derivatives. Concomitant with the onset of sexual maturation, anadromous Pacific salmon also experience changes in their osmoregulatory system in preparation for the return to FW. Stimulation of the Hypothalmus-Pituitary-Interrenal (HPI) axis yields an increase in plasma cortisol concentration along with multiple other compounds associated with FW adaptability. Amongst it multiple functions, cortisol promotes FW adaptability and it participates in the maturation sequence, perhaps via cortisol sensitizing gonads to the presence of the compounds released from the pituitary. In turn, maturation causes a shift in osmoregulation ability via causing a shift in the number, distribution and function of gill chloride cells and the down-regulation of gill Na^+K^+ ATPase activity. Provided individuals are receptive to treatment, exogenous supply of GnRH can trigger HPG activation and GnRH has been used to accelerate maturation, FW entry timing and upstream movement in many species of salmonids. Effects of exogenous GnRH on circulating hormone concentrations can become manifest within hours of treatment.

During the 2006 Late-run migration, we conducted an intervention experiment as a test of the osmoregulation hypothesis. The experiment was designed to identify the underlying forces responsible for the altered migration timing, an important piece of information necessary for refining projections of Late-run river entry timing and the magnitude of en-route mortality in subsequent years. The experiment involved collecting approximately 250 sockeye of the Adams River and Little River - Little Shuswap populations from the Strait of Georgia (DFO Fisheries management areas 29-4 and 29-9; date of collection Aug 29–Sept. 1), transporting the fish to the DFO West Vancouver Laboratory and dividing sockeye into one of five treatments. Treatments were: 1) Saltwater + GnRH (SW+H), 2) Saltwater (SW), 3) Iso-osmotic water (ISO), 4) Freshwater (FW), and 5) Freshwater + GnRH (FW+H). Salinity in salt water treatments was 28 ppt, iso-osmotic salinity was 13 ppt, and freshwater was 0 ppt. Sockeye were held for one week, followed by biopsy, release to the SOG and telemetry tracking.

Predictions based on the osmoregulation hypothesis and experimental conditions were: i) if osmoregulatory condition is the principle driver of river entry timing and sexual maturation is not an important influence, then sockeye held in low salinity treatments should exhibit the shortest time from post-treatment release to river entry and there will be no evidence of a hormone treatment affect on river entry timing; ii) if osmoregulatory capability is the driver of river entry timing but is modified by state of sexual maturation, then low salinity exposure plus hormone treatment should result in the shortest time between release and river entry than will low salinity exposure with no hormone treatment, and iii) if sexual maturation acts in isolation from osmoregulatory state in determining timing of river entry, then hormone treatment should cause the shortest time between release and river entry regardless of salinity exposure history and the three salinity treatment groups will have similar travel times.

There were large among-treatment differences in mortality during the weeklong treatment period. The ISO and FW treatments experienced the lowest total mortality (15.0% and 15.4%, respectively), FW+GnRH experienced 35% mortality, SW had 40% mortality, and SW+GnRH yielded the highest total mortality (80.0%). These results are consistent with the existing understanding that for Pacific salmon maturation is incompatible with salt water tolerance and that it is more energetically demanding to be in saltwater than in freshwater.

Treatments were able to produce sockeye of distinctly different end of treatment physiology. GnRH treated sockeye had higher testosterone and estradiol concentrations than did either baseline sockeye (i.e., those sacrificed immediately upon collection in the SOG) or salinity-only treatments. Gill ATPase activity was inversely proportional to salinity (i.e., SW>ISO>FW), although FW+H sockeye had higher ATPase values than FW only, consistent with knowledge that sockeye experience a slight upswing in ATPase activity when they near arrival to spawning grounds. Cortisol concentrations also responded to treatments, with values in line with expectations given the role of cortisol in both FW adaptability and maturation (i.e., SW+H = SW < ISO < FW < FW+H). In contrast, other measured physiological variables including hematocric, lactate, glucose, chloride, sodium and osmolality did not differ among treatments, suggesting sockeye rigorously maintain internal homeostasis despite the unique challenges posed by different environments. These results suggest that hormone, ATPase, and cortisol values can be used as viable indicators of maturation and migratory status, while the other variables are less likely to be useful in this capacity.

Physiology of the nine dead or moribund sockeye from which usable biopsy samples were available was greatly different than those of the 122 sockeye that survived to the end of the treatment period. Despite incorporating a few individuals from each of the five treatments, as a group the mortalities had higher mean lactate, cortisol, chloride, sodium, and plasma osmolality and lower glucose concentrations than survivors. These results suggest that death is associated with extensive “system failure,” meaning the loss of ability to maintain homeostasis.

At the end of the treatment period (September 6 and 7), we released 122 telemetry tagged sockeye to the Strait of Georgia. Of these, 104 swam away from the release point. Ultimately, 27 sockeye were subsequently detected within the lower Fraser River. Within each treatment group, the mean (± 1 SD) time elapsed from release to 1st in-river detection was 49.8 (± 17.5) hr in FW (n=7), 84.5 (± 45.6) in ISO (n=10) and 86.0 (± 19.6) hr in SW (n=5). The low number of GnRH treated sockeye entering the river (2 SW+H and 1 FW+H) makes a full assessment of the predications of the osmoregulation hypothesis impossible. However, the salinity component was supported, as FW sockeye entered the Fraser faster than did ISO or SW sockeye (ANOVA for hours from release to river entry for 3 salinity treatments, $p = 0.0266$; multiple range test: FW<ISO=SW).

River entry typically occurred within three days of release, and only 3 of the 27 sockeye that entered the river did so after September 11; one FW+H sockeye entered on September 13, one ISO sockeye entered on September 15th, and one ISO sockeye entered on September 20th (interestingly, this sockeye was detected on the Northern Strait of Georgia acoustic receiver array between WVL release and river entry). It is important to note that the peak of "wild" Late-run sockeye river entry occurred September 7–10, the earliest date for the peak of river entry on record. Given that experimental sockeye were released at the time of peak wild sockeye migration into freshwater, the two day among-treatment difference should be considered ecologically relevant as it is unlikely treatments would generate a larger divergence in entry timing given the river entry timing of the population at large. It is interesting to note that a companion study, also conducted during the 2006 Late-run migration, found that acoustic tagged Adams sockeye released from Area 13 averaged 5.5 days to travel from the NSOG telemetry line to the SSOG line (approximately 150 km) whereas Summer-run stocks made the same journey in 3.5 to 4 days. If we assume this ~ 48 hour difference in travel times between Adams sockeye and Summer-run stocks equates to the period of estuarine 'holding' that Late-run sockeye exhibited during their 2006 migration, then our salinity treatments appear to have succeeded in creating a 'realistic' divergence in river entry timing.

In total, results of our experiment are consistent with the predictions of the osmoregulation hypothesis, and our results suggest that monitoring salinity concentrations in the Strait of Georgia at the time of Late-run sockeye arrival may be a useful tool in predicting the magnitude of early river entry within any given year. However, limited sample sizes and the absence of "true" Late-run behaviours in the wild population make it difficult to offer stronger conclusions.

ORAL PRESENTATION—MIKE COOPERMAN

Background: Early research showed that historically, there was a fairly good relationship between upper layer salinity in Georgia Strait and early river-entry timing (lower salinity = early entry). Further, fish that enter early tend to have higher levels of sex hormones when sampled in Georgia Strait. The hypothesis, therefore, is that more reproductively-advanced sockeye are more susceptible to freshwater cues, which promotes early river entry.

This study involved collecting migrating Late-run sockeye and intervening with salinity and maturation manipulations, followed by biopsy sampling, release back into the Strait of Georgia and tracking the fish to their ultimate fate.

Maturation: Environmental factors trigger the hypothalamus, which produces GnRH to stimulate the pituitary, thus initiating further changes: the gonads respond by producing sex steroids and the gills/kidneys, etc. respond by adjusting in preparation for freshwater entry (e.g., ATPase is down-regulated). Sexual maturation and osmoregulatory processes also interact and affect each other.

In this experiment, fish were collected just outside the Fraser delta and taken to the West Vancouver lab where they were given a shot of GnRH to kick-start the maturation process artificially. Some were then placed in saltwater, some in freshwater and some in intermediate water, held for a week and then released back into the marine environment and tracked.

The fish collected were all Late-run sockeye in Georgia Strait that were displaying "holding" behavior between August 29 and September 1, 2006. About 50 fish were assigned to each of the five different treatment groups after DNA was collected for stock identification and tagging with a visible ID number. The five treatments were saltwater with hormones and without, freshwater with hormones and without, and iso-osmotic water (intermediate salinity) with no hormones.

Fish that died in the tanks were not replaced. Those given hormones and placed in saltwater had by far the highest mortality. These findings were consistent with a growing number of studies that show maturation is inconsistent with an ability to withstand saltwater. Before release, the fish were given a post-treatment biopsy and fitted with transmitters.

Treatment effects: Fish treated with GnRH had higher hormone levels. Fish also responded to salinity treatments, but hormones had an effect on their response to salinity.

Post-release survival: Fish held in freshwater and treated with hormones had the lowest survival rate after they were returned to the Strait of Georgia.

Travel time: Only one of the fish held in freshwater and treated with hormones made it to the natal area and the travel time was slow. It is thought that these fish were unable to do the reversion back to saltwater upon release. Fish held in saltwater and treated with hormones had the fastest travel time. It is thought that maturation plays a role in this. Only two fish (both ISO) entered freshwater on September 15 or later (i.e., normal time).

One caveat is that the early 2006 migration date confounds the ability to evaluate treatment effects, as fish were released just at the peak of the wild run, whereas it had been hoped to have at least a week of potential holding time post-release.

Conclusions: The treatment had the desired effects. The salinity component of the osmoregulation hypothesis was supported. The maturation component was unclear due to small sample sizes but mortality was consistent with predictions that maturity was associated with a loss of saltwater tolerance.

DISCUSSION

- Q: Cannot the fish select salinity gradients in the Fraser plume? / A (Cooperman): Fish outside the river can select where they want to be, but if they are freshwater-adapted, it seems they would choose to enter the river to be in freshwater. They were released from West Vancouver into saltwater and had to find the Fraser plume on their own.
- Q: Is there anything unique to sockeye to suggest that perhaps they could not tolerate the hormone treatments as well as other species, such as Coho? / A (Cooperman): We think sockeye don't tolerate confinement very well. Mortality for those held in freshwater was lower than those in saltwater but it kept going up over time.
- Q: What was the release timing? / A (Cooperman): There were two releases on September 7 and 8, at the same time of day, etc. There was some mixing between the treatment groups and some separation.
- Q: Why only Adams? / A (Cooperman): Adams was the only stock represented in large enough numbers. GnRH only works if the body is prepped for it. The study didn't evaluate other stocks.

FRASER WATER CHEMISTRY TRENDS: CORRELATIONS WITH RIVER ENTRY TIMING?

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ABSTRACT

Since 1995 the Fraser River Sockeye Late-run timing group has exhibited a 4 to 6 week shift to early river entry during their spawning migration (Cooke *et al.,* 2004). High levels of en-route and pre-spawning mortality have been associated with this behaviour. One potential explanation for this behavioural shift is changed in-river conditions. River water quality (WQ) databases were examined to explore the available information and to look for changes in 1995 that may have elicited the early entry behaviour.

Change to river WQ has two mechanisms that may invoke a behavioural shift: 1) late September (normal entry time) water chemistry conditions are being mimicked in August (new entry time), and/or 2) a chemical signal is attracting fish into the river (rather than them being driven from Georgia Strait).

Water quality is divided into two main groups: contaminants and standard parameters. Contaminant concerns in the Fraser were summarized by Johannessen and Ross (2002). They identified several contaminants of concern and rated them as high, medium, low, and unknown risks. The contaminants are of anthropogenic origin and enter the system by both point and non-point means.

The federal-provincial (Environment Canada—Ministry of the Environment) WQ monitoring program measures standard WQ parameters such as acid/base chemistry, carbon, carbon-nitrogen compounds, major ions, metals, non-metals, nutrients, organic contaminants, oxygen, pathogens, and physical aspects. This is a long-term database (initiated in the late 1970s) consisting of bi-weekly samples with on line access (www.waterquality.ec.gc.ca). The concentrations of several parameters were explored for changes at the Hope monitoring station. Both average annual and pre and post 1995 average monthly analyses were performed on six parameters.

Turbidity, a surrogate of gross WQ change and a potential indicator of other parameters showed a marked increase (21 to 37 NTU) and increased variability from 1997 and on.

Turbidity values from this station were quite sensitive to in river conditions, for example the fall 2004 Chilcotin River slide and the very low turbidity conditions of fall 2006 (causing enumeration and capture issues) are clearly seen in the data.

Chloride, a pollutant that was regulated in the late 1980s with the Pulp Mill Effluent Regulations, showed marked decreases post 1991 in annual average (2.6 to 1.3 mg/L) and variability.

Copper, cobalt, pH, and hardness are all known to affect anadromous fish migration and showed little change over the collection period in pre-1995 and post-1995 averages. Copper remained steady at 4.2 ug/L, average Cobalt changed from 1.1 to 1.4 ug/L, pH showed a slight increase from 7.73 to 7.89, and water hardness changed little from 58.5 to 60.5 mg/L. These are fairly modest changes and are likely not statistically significant; no large step-like changes were associated with the 1995 sockeye behaviour shift. Seasonal concentrations did not show marked change.

In summary there may have been some changes to turbidity (increase) and chloride (decrease) concentrations in 1997 and 1991, respectively, however the timing does not coincide with the 1995 sockeye behaviour shift. The other parameters analysed (Cu, Co, pH, and hardness) did not exhibit temporal changes. The database holds several other parameters which could be analysed. WQ stations nearer to the natal spawning and rearing grounds may also be worth analyzing. Environment Canada is currently performing an in-depth time series analysis on WQ trends; the document is due out this fall. Contaminant use and in-river concentrations could be further explored.

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ORAL PRESENTATION—HERB HERUNTER

This presentation discussed efforts to address two key questions relating to water quality: Has some change occurred that is mimicking September conditions or is some chemical signal attracting fish into the river early (vs. something driving them from the Strait of Georgia)?

Available water quality data relates to contaminants and standard parameters. Contaminant concerns were summarized in a 2002 report, so this presentation focuses on data on standard parameters that federal and provincial agencies have been collecting over the years.

Contaminants that correlate with mortality include triclopyr. The application rate for this forestry pesticide correlates well with high pre-spawning mortality among Late-run sockeye. The 2002 study identified contaminants of concern as high, medium, low or unknown risk in terms of impacts on Late-run sockeye. Pesticides, wood preservatives and “new” persistent organic pollutants were identified in the high-risk category. The study concluded that there were considerable gaps, but that there were concerns about new contaminants introduced in the last decade or so. The Forest Practices code was invoked in 1995 and that may account for some of the changes in pesticide use identified by Johannessen and Ross.

Environment Canada and BC’s Ministry of Environment have a long-term data set on standard water quality parameters from the 1970s, based on sampling twice a week at a series of stations all along the river. The data, which are available online, includes parameters such as pH, metals, nutrients, dissolved oxygen, pathogens and physical characteristics such as temperature and turbidity.

Herunter presented data from the Hope station on several parameters. Annual chloride levels have remained low since pulp and paper mill regulations were tightened in the 1980s. Mean annual turbidity levels have been more variable and generally higher since the mid-1990s. Cobalt and copper have known repulsion effects on salmon, but neither has increased since 1995. Hardness and pH have not changed either.

Turning to monthly trends, Herunter presented data comparing average monthly levels since 1995 to the average of previous years. For August/September, chloride has decreased since the mid-1990s, while turbidity is up. Cobalt, copper, hardness and pH remain unchanged.

In summary, if there have been water chemistry changes in the Fraser, they are subtle and the timing of those changes doesn’t precisely match the 1995 early entry changes. More work can be done in this area, including further exploration of the huge federal/provincial database and taking a closer look at data from other stations. Environment Canada is about to release a major long-term trend analysis. Other data to explore include regional district data and industry contaminant data.

DISCUSSION

[Note that Richard Addison as well as Herb Herunter answered the questions below.]

- Q: Previous talks suggest predisposition to early entry happening earlier in the migration, so to what extent are these chemicals being distributed further out to sea? / A: The Fraser plume goes a long way out, though increasingly diluted. / A: Past discussion of contaminants has considered two contexts. Persistent organic pollutants like PCBs, which are fat soluble, have impacts at low concentrations and are also endocrine disruptors. The existing literature focuses on impacts at spawning stages, not river entry timing, but we can probably discount the major organic contaminants. They have also been well regulated in recent years and levels in the environment are declining, apart from PDBEs. So if we are looking at a chemical cause of the disruption of entry timing, we will have to look at water soluble contaminants that fish are exposed to as they enter the river; for example, things found in the outflow from sewage treatment plants. Those are the ones you should focus on, especially two particular groups: 1) steroid hormones: we need to look at something that changed in the mid-1990s. I found there was a dramatic increase, with levels of estradiol that were within 50% of levels known to affect fish. 2) Pharmaceuticals are the other group that has increased most since the mid-1990s. For example, Statins use has increased dramatically. Statins survive excretion and sewage treatment and are known to affect other salmonids, so these are worth considering.
- Q: What about nutrients coming out of the Fraser? / A: Emissions from the Fraser are negligible compared to those coming up in coastal currents.
- Q: The Statins hypothesis might work if there was a change in the time of spawning but they are spawning at the same time. You would also expect testosterone levels to be lower, but in fact they are higher. The concentrations measured in sewage are below the thresholds that have been shown to affect other salmonids. So how would you explain the tremendous success of Harrison if there were some negative factor at play? / A: If there was an attractant, all stocks should be responding and changing their patterns.
- Q: One reason to "stay with the school" is that you are lost. Miller's work showed that they're switching to olfactory cues. There was a study on the effect of herbicides on olfaction; agricultural residues could be coming down the river at that time at low levels that confound olfactory cues, and there are new types that are being used in recent years. / A: We looked at the database and found no obvious smoking gun. The question now is whether we have something that we could go back and look at or do we try some new experiments?

PROGRESS IN STUDYING THE HYPOTHESES

OVERVIEW—SCOTT HINCH

The list of research issues from the 2001 PSC workshop has been presented along with an indication of whether a particular issue has in some way been studied in subsequent years, in Table 1.

The research to date has focused on the hypotheses identified as most likely. A revised list ranks these hypotheses by likelihood and whether they have been explored (Table 4).

TABLE 4. Ranked list of 16 issues and/or hypotheses involved with potential research on the early migration phenomenon for Late-run sockeye salmon (based on February 2001 PSC 'expert' workshop).
X indicates that the issue was studied to some degree since the 2001 workshop. N indicates the issue was not studied. Ranking procedure score in parentheses—smaller number indicates an issue or hypothesis perceived as needing more urgent investigation.

H number and score	Description of hypothesis	Studied? (X=yes)
1 (4.0)	Exploratory data analysis of ocean parameters related to Late-run upstream timing.	X
2 (4.4)	One or more Biological Clocks has/have been advanced leading to early upstream migration.	X
3 (5.3)	Sockeye salmon acquire infection prior to Fraser River entry leading to a behavioral change.	X
4 (6.0)	Adults approaching Fraser R. are exposed to chemicals that trigger early entry into Fraser.	X
5 (6.5)	The fish behaviour has changed (monitoring, or EMG tagging).	X
6 (6.9)	There is a relationship between parasite infection and host mortality.	X
7 (7.7)	Smolts are infected near mouth of the Fraser River.	X
8 (7.8)	Exchange processes in Georgia Strait have altered delay behavior.	X
9 (8.9)	Changes in vertical distribution of Late-run sockeye causes them to experience different physical conditions altering delay behavior.	X
10 (9.4)	Early upriver migration triggered by low stored energy reserve.	X
11 (9.7)	Interaction of diversion rate with physical ocean conditions affects Late-run upriver timing.	X
12 (9.8)	Choice of river entry timing is related to conditions in year of ocean entry (smolt effect).	N
13 (10.0)	Sockeye are fleeing into the Fraser River to avoid predators.	N
14 (10.1)	Diet may be responsible for changes to Late-run sockeye behaviour.	N
15 (10.8)	Predation by harbour seals and killer whales has increased?	N
16 (10.9)	Outmigrant juveniles are exposed to chemicals that reduce their ability to hold as adults.	N

Two hypotheses that can now be rejected are that low energy or *Parvicapsula* might be driving fish into the river. It is now clear that low energy is caused by more advanced reproduction and *Parvicapsula* is not obtained in the Strait of Georgia. Regarding photoreception, studies of UV cone density relative to freshwater preparedness found no differences.

Hinch listed the hypotheses that remain:

- More advanced reproductive state correlated with early entry.
- More 'prepared osmotically' correlated with early entry.
- Oceanographic water properties correlated with early entry.
- High seas and coastal correlations. Additive effects?
- High proportion of Summer-runs in Strait of Georgia correlated with early entry.
- Predator issue not fully explored. No clear patterns linked with Late-run change; should this be explored in terms of the 'stay with the school'?
- Contaminants? Perhaps further study is required.

Several hypotheses seem to be leading and some seem to be inter-related. Are they hierarchical? For example, does high wind stress in the open ocean cause fish to mature earlier? Are more mature fish more likely to stay with the school in Georgia Strait? Are they synergistic? Are fish that encounter low coastal salinity patches coastally more likely to stay with the school?

DISCUSSION

- Audience member comment: Some of these bother me. Proximate cues may be responsible for small-scale changes but don't explain the big shift. There is a sense that something major is still missing, and that we need to be careful in terms of how much these things actually explain.
- Audience member comment: Can any of these be eliminated through discussion in the next two days? The ideas of looking at synergisms and hierarchy are worth exploration (some of the original hypotheses were quite naïve). Another question is whether we need to do more experiments that manipulate fish or more hind-casting. How would we test/reject the hypothesis? There are other ways to test these things without killing fish.
- Q: Has more work been done on smolt theories? / A (Hinch): No.
- Audience member comment: What is killing the fish when they move into freshwater? Could it be a marine viral infection that they've picked up outside that we have missed?
- Q: If fish make landfall around the Queen Charlottes, could they get confused and go north?
  / A (Hinch): There is no array to test that. Initially we worried about the level of mortalities, assuming it was out fault. The control fish also had many problems in that experiment.
- Audience member comment: In 2005 and 2007, they identified a lot more Fraser fish up in Alaska.
- Audience member comment: You could combine the first three items in the list of hypotheses that stay on the table. I'm not convinced that factors that cause early entry and those that cause mortality are the same and that this has been explored enough.

Gardner thanked participants and presenters, noting that Day 2 would focus on the consequences of early migration.

DAY 2: CONSEQUENCES OF EARLY MIGRATION

Reviewing the day's agenda, Gardner explained that the first set of presentations would deal with survey observations and the second would explore issues with experimental approaches. Presentations concluded in the early afternoon with an example of a management model based on these science results.

Hinch noted that discussion would focus on high in-river mortality for adult Late-run sockeye that are migrating earlier. Initially, hypotheses relating to energetics and osmoregulation were seen as most likely to hold explanatory power. Reproductive readiness was initially seen as less likely to relate to what is occuring in the ocean environment. Parasites were seen as important but marine mammals were not.

FRASER RIVER ENVIRONMENTAL CONDITIONS FOR LATE-RUN SOCKEYE

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ABSTRACT

There is a growing awareness that interannual variation and trends in climate patterns can have an impact on fish condition and survival throughout their life history, and thus impact stock productivity. Proxies for Fraser River environmental exposure are already being used to provide management advice for all sockeye salmon run timing groups, including the Late-run. Research conducted in part through the Late-run project has increased our understanding of the subtleties of freshwater environmental exposures experienced by adult salmon migrants and aided the development of more biologically relevant environmental metrics.

The DFO Fraser River Environmental Watch (EWatch) Program monitors water temperatures at multiple locations throughout the Fraser Basin. Although there has been no pronounced change in summer river flows over the past 50 years (data courtesy of Environment Canada), summer river temperatures have been increasing. In addition, the change in river entry behaviour of Late-run fish has resulted in exposure to higher temperatures and flows, and increased freshwater residency, compared to historic levels. Expansion of the temperature monitoring network into Late-run tributaries and lake systems has provided additional information regarding the varying exposures experienced by fish throughout the duration of their run. In addition, internal thermal loggers (ibuttons) implanted in radio-tagged fish provide detailed thermal migration profiles highlighting the strong individual variability with respect to river-entry, thermal refuge and lake residency behaviours. Comparison of re-constructed thermal profiles using EWatch stationary loggers to observed ibutton profiles confirmed that the measured river temperatures are representative of the daily temperatures experienced by Late-run fish, at least in the mainstem of lower Fraser River.

The expansion of the temperature monitoring network, in addition to individual thermal profiles and migration rates from the Late-run tagging studies, has led to the development of several alternative temperature metrics each describing freshwater temperature exposures with varying degrees of detail. These environmental metrics have already proven useful for describing and/or predicting en-route losses of Fraser sockeye with direct application to management adjustment (MA) models used to increase the probability of achieving spawning escapement targets.

1. **Average lower river environmental conditions:** Currently, run timing (Late-run) and symmetric 31-day average lower river temperatures and flows centred on the date 50% of a run passes Hells Gate (Early Stuart, Early Summer and Summer-runs) are used as proxies for en-route environmental exposures in the MA models.

2. **Extreme river environmental conditions:** Although experienced flows will always be highest in the lower river, temperature exposure in different river systems lagged by migration time showed that Late-run fish may actually experience warmer temperatures in the Thompson and South Thompson Rivers than in the lower Fraser.

3. **Weighted average environmental conditions:** The high degree of inter-annual variability in the length and shape of an incoming run means a symmetric 31-day period will capture a varying percentage of the run from year to year. In addition, extreme temperatures/flows experienced at the beginning or end of that 31-day period may skew the estimate of average exposure. Weighting lower river temperature and flow by the proportion of the run passing Hells Gate on a given day may provide an average that is more representative of the conditions experienced by the fish.

4. **Accumulated thermal units (ATUs):** Combining the concepts from items (2) and (3) above, we can integrate information on migration rate and multi-site water temperatures and approximate the weighted average of total thermal units (i.e., degree days) accumulated by Late-run fish from the time of river entry to arrival on the spawning grounds. Comparison of modelled versus observed ATUs (from ibutton data) shows a relatively high degree of correlation in most cases. This information has direct application to disease-progression models.

5. **Individual thermal profiles:** The probability of freshwater survival is influenced by not only by the total number of ATUs, but also the manner in which these thermal units were accumulated (e.g., 100 ATUs resulting from 10 days at 10°C will have a different affect than 100 ATUs resulting from 5 days at 20°C). We are currently investigating whether more detailed information from individual thermal profiles can further enhance our ability to make predictions of successful versus unsuccessful migrants.

The combined shift in river entry timing (~3°C) and increases to average summertime Fraser River water temperatures as a result of climate change (~2°C) have produced a potentially unprecedented 5°C increase in average temperatures experienced by Late-run sockeye over the past 60 years. The projected warming of the Fraser River and uncertainty with respect to Late-run migration behaviour emphasises the need for continued monitoring and modelling of river environmental conditions and their potential effect on sockeye salmon populations, particularly the Late-run. Time series of historic temperature and flow are directly applicable to Fraser sockeye management through use in long range forecasting models used to predict pre-season management adjustments. Detailed information regarding thermal behaviour (e.g., weighted averages, ATUs, thermal profiles) serves as groundwork for the development of explicit biological models which can be used to provide direct predictions of en-route mortality. These models will also assist in the understanding of future climate change effects on the probability of spawning success for different Fraser salmon populations.

ORAL PRESENTATION—DAVE PATTERSON

This presentation focused on river discharge and temperature, including different ways of looking at temperature data. Less is known about encounter velocities than temperature (discharge rates don't necessarily correlate to encounter velocities).

Historical discharge trends show no shift over recent years for the late summer period (not related to early-entry behavior). However, earlier entry would equate with higher discharge levels, which means reduced visibility, but not necessarily higher encounter velocities, as the fish can swim closer to the banks. So it really means a reduced area for optimal swimming performance.

One issue with historical temperature trends is uncertainty about whether the temperatures being monitored were the same as those that the fish were actually experiencing, given behavior linked to thermal stratification and thermal refuges. In order to improve understanding of thermal behavior, temperature data was combined with conventional tagging, plus radio and acoustic tagging and ibuttons (internal archival temperature loggers) to evaluate entry timing and mortality hypotheses.

Patterson provided several examples of the individual thermal behavior profiles that can now be generated by combining these various data sources. The profiles show the major temperature shift that fish encounter on entering the river and fish seeking out cool water in lakes where possible on their migration path. Ibutton data also showed that not all fish spend their time holding time in the Marine environment en-route to Mission but that some fish actually went straight into the river and spent three extra days below Mission in 19° C water instead of in 11° water outside.

Data from the main-stem river showed that internal temperature matched monitored river temperature at different locations, confirming that river temperature was mimicking in-river temperature exposure. Matching temperature profiles with positional information also revealed how fish were seeking out cooler, deeper water as they migrated through lakes.

This new information can be used to advance understanding of early-entry behavior and en-route mortality. Examples include examining the behavior of fish in the marine approach in the absence of positional information or testing predictions regarding entry behavior in the lower estuary. Different exposure models for the main-stem Fraser can now be applied with confidence and generalizations can be made about thermal behavior in lakes.

Researchers can also now develop different temperature metrics and construct more sophisticated models than those which until recently relied on a simple average temperature at one location. More biologically-relevant metrics for temperature exposure include weighted averages (weighting temperature experience by daily abundance) and average environmental exposure for multiple locations. It is now known that fish experience different temperatures along the river migration. For the summers, the warmest temperatures are encountered in the lower river, but Late-run Shuswap experience their highest temperatures further up-river.

Accumulated thermal units: This is a key concept in thermal profiles, derived by combining migration rate data with daily abundance and temperature by location. Such analysis showed that Late-run Adams fish accumulated more thermal units than Chilko and Scotch. Although temperatures are higher in summer, those fish migrate faster and don't spend as long on the spawning grounds. The only stock that accumulated more thermal units during river migration was Early Stuart sockeye.

Work is in progress to determine if individual en-route profiles can be developed that will explain migration success based on daily information—can unsuccessful spawners be identified by their thermal profile?

Applications to biological models include looking back at historical data to match historical temperature experience with stock-specific adaptations in aerobic performance, or doing thermal disease modeling. Interaction between river discharge and temperature can also be modeled (e.g., in some cases the warmer it is, the more able the fish are to deal with higher flows). Historical data on temperature experience can also be used to compare climate change and migration timing behavior.

If temperatures are increasing with climate change and temperature exposure is further increasing due to the behavior shift to earlier migration, the combined effects are unprecedented in any salmonid population, with significant implications for rates of adaptation and selection as well as mortality.

DISCUSSION

- Q: Lab studies of rainbow trout showed that if they were held for three weeks at higher temperatures, they lose sex steroids and eggs. Is that consistent with what has been observed with higher accumulation of thermal units? / A (Patterson): Similar experiments where sockeye are exposed to higher temperatures at the end of the migration have not yet been done.
- Q: What created the higher overall thermal conditions for Early Stuarts? / A (Patterson): As they move up-river, the water gets warmer, plus the overall length of their migration is longer.
- Q: Are Late-run fish that are still spending longer in the ocean still surviving better? / A (Patterson): Yes.
- Q: Early Stuarts are fast-moving fish, despite facing higher discharges. What rate was used for migration time? / A (Patterson): We used actual migration rate profiles from 2006 and 2007. It would be better to include more inter-annual variability.
- Q: The difference between ibutton and environmental temperature data is very small but it seems consistently lower. Why was this—is it a calibration issue? / A (Patterson): They may be seeking out cooler water, but it is an insignificant difference as it is within measurement error.
- Q: What is the response time of fish to temperature changes? / A (Patterson): Technical studies show response time of around 30 minutes. Behaviour doesn't matter in the river in terms of going up and down.
- Q: What are the implications of these data for the recovery of Early Stuarts? / A (Patterson): There is no reason to think the environmental conditions and trends are going to change.
- Q: Over what period was the 10-degree change shown in one graph when the fish entered the Fraser? / A (Patterson): The one shown occurred over four hours, but it can occur in minutes. This relates to the question of the sensitivity of temperature sensors. It is not unusual that the fish are exposed to this thermal shock.
- Q: How many of the fish that delayed went into the river and back out? / A (Patterson): Acoustic data is going to be more reliable than relying on ibuttons for that. It should be able to detect individual movements in fine scale and real time. We also need to be cautious about general conclusions based on individual behaviour.
- A (Patterson): Putting the lack of thermal refuges in the lower river in context, fish in the Columbia swim into the lakes and hang out for days to avoid high temperatures. But there are limited opportunities in the lower Fraser to do so, relative to the upper river reaches.
- A (Patterson): In trying to interpret whether the ibutton data means the fish are entering the river and going back out, they could also be staying in the same spot and it is the river temperature that is changing.
- A (Patterson): At the two test fishing sites, it is quite common to catch fish in the first one and then they don't show up at the next one.

FRASER RIVER SOCKEYE RADIO TELEMETRY STUDIES 2002, 2003 AND 2006—SURVIVAL FROM MISSION TO SPAWNING AREAS FOR LATE-RUN STOCKS

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ABSTRACT

In 2002, 2003 and 2006, the radio-tagged Late-run sockeye that passed Mission in August had a much lower probability of survival to their spawning areas than those that passed Mission in September (English *et al.,* 2005; Robichaud and English 2007). These results are consistent with the 2000 and 2001 observations of higher pre-spawn mortality when a large portion of the Late-run sockeye entered the Fraser River in August. English *et al.* (2005) suggested that changes in environmental factors could affect spawning timing and/or the development of the mortality agent, thus shifting the survival-vs.-entry-date curve to the left or right. It was hypothesized that higher water temperatures could increase the rate of development of the mortality agent (such as a parasite) thus decreasing survival (i.e., shifting the survival curve to the right). Fraser River water temperature data were obtained for the period when Late-run sockeye were migrating upstream in 2002, 2003 and 2006 (i.e., 10 August through 30 September). These data showed that average water temperatures measured in 2003 on the Fraser River near Hope and on the Thompson River near Chase were only slightly higher than those measured in 2002 for the same Late-run migration period. Daily temperature data from a monitoring site near Ashcroft in 2006 combined with individual sockeye temperature log data strongly indicated that accumulated temperature units (ATUs) played a significant role in determining survival to spawning areas and spawning success in 2006. Other research has shown that water temperature is a key determinant of survival for Late-run sockeye. In 2006, DFO increased the number of temperature monitoring stations within the Fraser Watershed. This combined with the ibutton and tracking data for recovered radio-tagged fish provided a unique opportunity to assess the effect of river and lake temperatures on migration rates and spawning success.

While further investigations may be desirable to explain observed variability in survival between Late-run stocks (e.g., Shuswap versus Weaver), the available observations should be considered when formulating fishery management plans. The survival curves derived from the 2002 and 2003 studies were used in 2006 to provide in-season predictions of en-route mortality for Late-run sockeye. Our best estimate of survival for Late-run sockeye from Mission to spawning areas in 2006, excluding fishery removals, is 72.0%. This estimate is consistent with a 28% pre-spawn mortality estimate and very close to the in-season predictions made on 26 August and 1 September 2006, prior to the PSC decreasing the project run size for Late-run stocks.

References

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ORAL PRESENTATION—KARL ENGLISH

These large collaborative studies involved many participants to tag, release and track hundreds of sockeye from ocean tagging sites to spawning areas throughout the Fraser watershed in each of three years (2002, 2003 and 2006), plus smaller releases in the lower Fraser in 2004, 2005 and 2007. In addition to objectives relating to entry timing, the studies sought to estimate in-river mortality and spawning success of Late-run sockeye by Mission timing component. Tracking and monitoring sites extended from the Mission to the spawning areas for all major sockeye stocks.

2002 study: The capture, tagging, release and tracking processes were briefly described. Tracking results showed individual migration patterns and timing to key migration points for the ocean release groups. Some Late-run sockeye in each release group entered the Fraser River with little or no delay in lower Georgia Strait while others from the same release period delayed at the mouth of the Fraser River for one to three weeks before entering and migrating upstream at roughly the same rate as the early entry Late-run sockeye. Comparison of Mission timing of the ocean release of Late-run sockeye in 2002 to PSC estimates of daily abundance showed the sample of radio-tagged fish provided a representative distribution over the actual run.

In-river survival: In the comparison of survivors/mortalities to Mission passage the date of the radio-tagged sockeye showed that most of the survivors passed Mission in September; very few of those that passed Mission in August survived. Plotting the in-river survival rate against Mission passage date for the 2002 data provided a good fit for both the Sigmoid and Michaelis-Menten curves.

2003 study: Additional study objectives included bio-sampling of radio-tagged fish. The same capture and tagging process was used, with the addition of physiological samples (fat probe measurements, blood and gill tissue samples) from roughly half of the tagged fish. Tracking showed similar migration patterns, although the 2003 release groups in mid to late August missed the first part of the Late-run migration. The abundance of Late-run stocks was substantially lower in 2003 (Mission escapement was 10% of the 2002 level), and there was less holding in Georgia Strait. Once again, the majority of survivors were those that passed Mission in September, with most mortalities being among those that passed in August.

Regarding Late-run survival curves, there was more uncertainty earlier in the run. The curve shifted to the right for 2003 vs. 2002, with more early-entry fish dying (river temperatures were warmer).

Results of these two studies were published in 2005 (English *et al.,* 2005).

2006 study: Additional objectives were to determine the location, timing and nature of en-route losses. The tagging and sampling procedures were similar to those in 2003, except that muscle tissue was also sampled, more scale samples were taken and physiological samples were obtained from every radio-tagged and acoustic tagged fish. The marine tagging missed the early part of the run, but this was covered by tagging in the lower river.

Comparison of survival to Shuswap against passage time at Spence's Bridge showed that most of the mortalities were among the fish that passed earlier, when temperatures at Spence's Bridge were 1–2° C higher than normal. Many fish were last detected by the receiver located at Spence's Bridge (near the mouth of the Nicola River). The travel times for these fish were similar to those for fish that reached the spawning grounds successfully.

Most fish that reached the spawning areas passed Mission in late August and early September. Most fish that passed Mission in early and mid-August did not survive.

In conclusion, three years of intensive research on Late-run sockeye have consistently shown a relationship between Mission passage timing and survival to spawning areas. A very large portion of Late-run sockeye that enter the Fraser River in the first half of August die before spawning. Survival rates for Late-run sockeye improve rapidly after mid-August.

DISCUSSION

- Q: 2003 was a warmer year and could have caused higher mortality among earlier fish. Did the studies look at temperature impacts in a different way? / A (English): We looked more at a specific location (e.g., Spence's Bridge). But after three years of study, we still only have three data points. Collaborative work is also being done, looking at temperature in terms of degree days and the relationship between mortality and accumulated temperature units.
- Q: How were fish captured in the lower river? / A (English): We used a 3-inch tangle net. There was very short transport time and the fish recovered from the tangle net capture quickly—there was good post-release survival.
- Q: Can you reconstruct the accumulated thermal units for Harrison in comparison to stocks that didn't do as well? / A (English): The 2004 study focused on the Harrison stock. The clearest information from that study relates to Weaver Creek sockeye. The challenge with Harrison stock is that they spawn in the Harrison River near their pre-spawning holding areas. So you can detect when they enter the river and/or the lake but you don't know whether the fish that entered and were detected ended up successfully spawning. For Weaver, they go up to the lake and then come back down to the spawning channels when they are ready to spawn, so more information is available on Weaver. Harrison is a pretty cool lake and is definitely used as a refuge. It is one of few such opportunities in the lower Fraser.

CONSEQUENCES OF EARLY ENTRY ON LATE-RUN SOCKEYE SALMON: FOCUS ON RIVERINE MIGRATION

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ABSTRACT

Using biopsy and telemetry techniques outlined in the previous Cooke *et al.,* abstract in this document, we studied the consequences of early migration on Late-run sockeye. Perhaps the most meaningful data arose from an in-river study conducted in the Thompson River where fish were captured by dip net with the assistance of First Nations technicians. This sampling technique was sufficiently rapid that the physiological data obtained from blood and other tissues was reflective of the condition of fish in the river and not influenced by capture gear. After fish were biopsied and released we revealed that fish that died en-route tended to have higher levels of plasma lactate than fish that survived to reach spawning grounds. We also attempted to conduct an electromyogram (EMG) telemetry study to evaluate fine-scale patterns of energy use and behaviour in a complex reach of the Thompson River. Interestingly, none of the Late-run fish tagged with EMG transmitters successfully negotiated the study reach. In fact, many fish died in holding pens or on the surgery table. Fish bled excessively during the surgery despite the fact that we have previously conducted these surgeries on other stocks of sockeye. An apparent clotting problem with early migrants may have contributed to this problem although there may be other explanations related to blood pressure and kidney blockages arising from myxosporean parasite infection.

ORAL PRESENTATION—STEVEN COOKE

This presentation focused on consequences of early entry, as decoupled from the phenomenon of early-entry behavior, and specifically at what are the physiological and behavioural correlates of fate in freshwater. It is based on collaborative tagging studies done in 2003 and 2006, results of which were published in three papers in 2006.

2003 results: Although a large number of fish were tagged in this study, when they were broken down into different groups for comparison, it resulted in relatively few in each subcategory. Comparing Adams fish that survived vs. mortalities for each of the three release groups, gross somatic energy levels were higher in each group for those that subsequently survived. It was also found that up-river mortalities (male and female) have advanced reproductive development. This reflects a similar trend to what was seen regarding early entry timing (these fish were radio tagged in saltwater). Findings regarding gill ATPase showed no difference in terms of mean levels, but fish that died in-river have more variable levels of gill ATPase. It is not clear what this means.

2006 results: Comparing travel time for Adams fish from Mission to the Thompson River confluence to holding behavior and fate, it was found that none of the fish that held died in-river. Travel times were significantly different among those that held and succeeded, those that did not hold and succeeded, and those that did not hold and failed. Those that held were more likely to reach the spawning grounds and travelled more slowly. Those that did not hold and failed to reach the spawning grounds had the fastest travel time to the Thompson River confluence. Females that held and reached the spawning grounds had higher levels of somatic energy and lower testosterone. Females that did not hold and perished had low somatic energy and high testosterone.

Summarizing the results of the 2003 and 2006 radio/acoustic data, it seems that energetics and reproductive development are important determinants of migration timing and success. This is based on signatures at the time of capture in the marine environment.

Fish sampled and tagged in freshwater: An in-river telemetry study was done in 2003 in the Thompson canyon to evaluate in-river physiological and energetic correlates of behavior and fate. Fish were caught in dip nets, which provides a very quick method and thus allows reading of actual physiological levels before time of capture.

The results showed a biologically meaningful signal for lactate levels, which was related to fate but not timing. Sampling included Summers, abnormal Lates and normal Lates and in all three groups, those that failed to reach spawning grounds had higher levels of lactate at the time of capture. Those that failed to reach the spawning grounds also had lower energy levels across all three groups. The Late-run sockeye that entered early also had higher energy than those that held and migrated at the normal time. This pattern was consistent with that of Thompson fish in the ocean radio tagging study.

An EMG telemetry study was a failure, with most of the fish bleeding out and dying on the surgery table. This was unusual and may have been related to clotting or blood pressure. Clotting took almost twice as long among early-entry fish, which raises questions about whether this is a mortality agent.

These results provide a lead-in to the upcoming talks on temperature and the parasite *Parvicapsula*. Further work is underway on the role of lakes and thermal refugia and a detailed in-lake analysis of the Shuswap Lake/Adams system.

DISCUSSION

- Q: What are normal lactate levels—is there a baseline? / A (Cooke): They can be non-detectable. It is thought that levels for fish sampled immediately after capture in dip nets are close to actual.
- Q: But those levels were still high. This was a difficult part of the river but can they maintain those levels? / A (Cooke): They were not maxed out but were clearly elevated. Those levels can be cleared if they subsequently find a refuge.
- Q: Do lactate levels interact with water temperature and oxygen available in the water? / A (Cooke): Lactate also clears more quickly with warmer water.
- A (Cooke): When sampled in the marine environment, fish that did not hold had lower somatic energy. But in river, the fish that held were found to have lower energy levels. It may be a question of where you choose to spend your energy.

- Q: If the primary areas of mortality are occurring well up-river, how relevant is an ocean sample? / A (Cooke): We have tried to link lower and upper-river mortality to the ocean data. The higher up-river you go, the weaker the ocean signal. It is still useful, but it is less robust than if you collect in-river samples. Ideally, one should focus tagging and sampling just downstream of those high mortality points. But this raises challenge in terms of being able to keep the fish alive, so destructive sampling is also being done.
- Q: What could cause the blood-clotting problem and was this seen in previous years? / A (Cooke): It wasn't seen in previous years. We had never tried to this in those areas before, but that unusual bleeding was not seen previously. It could be that *Parvicapsula* is blocking urination and the fish is swollen, with high blood pressure, etc. / A (Miller): When we looked at gene arrays for liver tissue, we got a huge signal associated with blood clotting, and also a large signal relating to amino suppression. But it is doubtful that was related to *Parvicapsula* because it would not have been present in fish sampled in Whonnock. There are many opportunities en route, whether they encounter fishing gear or bump into rocks, where they could be injured.

ADULT SALMON IN HOT WATER: MIGRATION THERMAL EXPERIMENTS

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ABSTRACT

Late-run sockeye that enter the Fraser River early encounter warmer water than they have been adapted to dealing with (16–18° C vs. 13–16° C); in recent years they have encountered extremely high temperatures (e.g., 20° C as in 2004); and early migrants mill in lakes near spawning streams instead of holding in the Strait of Georgia, thereby accumulating many more freshwater degree days. We conducted a series of experiments to examine how these temperature issues affect the survivorship of Late-run sockeye. In 2003, Late-run sockeye were captured in the Strait of Georgia, then transported to a facility on Annacis Island where they were exposed to Fraser water (20° or 16° C) for ~1 week, then moved to the Cultus Laboratory where they were held for up to several weeks at high (18°) or low (12°) temperatures. For the fish initially held in 20° degree water (simulating early-timed migrations) half died within 10 days. The surviving fish were then moved to the Cultus lab and all died within the following week in 18° C, but ones held in 12° had slightly better survival (20% survival). Fish initially held in 16° C (simulating normal-timed migrations) had no mortality for the first week. Fish were moved

into cooler or warmer temperatures and held for 3 weeks. Those held in 18° water had some mortality over the next 10 days (~20% mortality), while those held in 12° water continued to have no mortality during the same period. Therefore, this experiment supports the hypothesis that during a typical three-week migratory period, most early migrants can perish because of high temperature exposure. Early migrants that can locate thermal refuge in lakes may have somewhat better survival.

In 2003 Adams sockeye were radio tracked from river entry to spawning grounds. We linked these data with river/lake temperature data to assess the thermal experience of 50 migrating fish. The early-migrating fish spend more time in freshwater than normal-time fish because they do their 'holding behaviour' in lakes adjacent to spawning areas, rather than the estuary as normal-timed fish do. Early-timed fish accumulate ~ 400 degree days by the time they enter Shuswap Lake, and any fish that leave the lake to enter spawning areas have accumulated ~ 700 degree days. That so many fish die in the lake is probably related to high degree day levels. Normal-timed fish enter spawning grounds at ~ 400 degree days. The severity of *Parvacapsula minibicornis* infection, a myxosporean parasite contracted in the estuary which affects kidney function, has been found to increase rapidly when individuals accumulate about 370 degree days. As kidney function declines from disease, high temperatures and exercise could lead to cardiovascular collapse. Lab tests with normal-timed sockeye captured in the Harrison River and transported to the lab and held for 24 days at 18° C showed the same pattern of spiking mortality after about 370 degree day exposure.

In 2004, Weaver Creek sockeye were telemetry tracked from the confluence of the Harrison and Fraser Rivers, across the complete range of arrival times in the Harrison River. We estimated degree day accumulation for 83 fish based on their arrival dates and subsequent potential migration paths to reach spawning grounds on October 10. Early migrants that stayed in Harrison River would have accumulated almost 800 degree days by the time they had reached spawning grounds—in reality, none of these fish survived. Early migrants that used the lake accumulated almost 500 degree days and had 84% mortality. Those that arrived at the normal time and stayed in the river had about 400 degree days, while normal-timed fish that used the lake had the least exposure of all—less than 300 degree days—and also the best survival. To test the hypothesis that migrants exposed to realistic, chronically-high temperatures survive more poorly than those exposed to chronically-low temperatures, in the same year we held Weaver sockeye under two different thermal regimes (8° C vs. 18° C) in the lab for three weeks, then released them back into the Fraser River tracked them towards spawning grounds. Migration mortality was twice as high among the fish held in warm water (60 % vs. 30 % mortality). Mortality rates of these two groups reflect what is often seen in early vs. normal-timed migrants.

In summary, early migrating Late-run fish get thermally 'hit' in three ways which involves higher encountered temperatures and for longer periods of time:1) higher than average temperatures linked to migration timing, 2) climate warming, and 3) more freshwater degree days. A threshold of 400–500 degree days seems to be a critical range for Late-run fish. *P. minibicornis* probably plays a role in mortality, as do other diseases, but most others have not been fully examined.

ORAL PRESENTATION—SCOTT HINCH

This presentation covered three Late-run thermal issues, some thermal mortality concepts, migration thermal simulation experiments, telemetry studies and a field experiment testing the effects of chronic thermal exposure on mortality.

Thermal issues: Late-run sockeye that enter the Fraser River early encounter warmer than normal water (16–18° C vs. 13–16° C). In recent years they have encountered extremely high temperatures, for example the very high temperatures seen in 2004. Trends show that the river is warming and that this is likely to continue. Early migrants are milling in lakes near spawning streams instead of holding in the Strait of Georgia, thereby accumulating many more freshwater degree days.

Thermal mortality: There are several possibilities in terms of how fish could be killed by high temperature exposure. This discussion focuses on chronic processes which involve a slower loss of homeostasis relative to acute processes (examples of these acute ones will be presented by Farrell). In this chronic context, 'accumulated degree days' (AKA 'accumulated thermal units') is a relevant metric. Adult migrating salmon have some special characteristics to consider in this context, including the fact that they are maturing, senescing, and losing their immune functions. Additional unique issues for the early-migrating Late-runs is that they are more physiologically stressed, even more mature, and have irregular osmoregulatory function relative to normal-timed migrants thus we should expect poorer survival in thermal performance experiments from early-timed fish compared to normal-timed fish.

Lab examination of thermal experience and survival: In 2003, Late-run sockeye were captured in the Strait of Georgia, then transported to a facility on Annacis Island where they were exposed to Fraser water (20° or 16°C) for ~1 week, then moved to the Cultus Laboratory where they were held for up to several weeks at high (18°) or low (12°) temperatures.

For the fish initially held in 20° degree water (simulating early-timed migrations) half died within 10 days. The surviving fish were then moved to the Cultus lab and all died within the following week in 18°C, but ones held in 12° had slightly better survival (20% survival).

Fish initially held in 16°C (simulating normal-timed migrations) had no mortality for the first week. Fish were moved into cooler or warmer temperatures and held for 3 weeks. Those held in 18° water had some mortality over the next 10 days (~20% mortality), while those held in 12° water continued to have no mortality during the same period. Therefore, this experiment supports the hypothesis that during a typical three-week migratory period, most early migrants are likely to perish because of high temperature exposure. Early migrants that can locate thermal refuge in lakes may have somewhat better survival.

Role of disease: The parasite *Parvicapsula minibicornis* may play a role in migration mortality. It is contracted by all migrating salmonids passing through the Fraser estuary. Prevalence and severity of infection increase during the up-river migration. The parasite is found in the kidneys and could affect osmoregulation and blood pressure. This is still the leading suspect, but other bacterial and fungal diseases could also be involved, including Columnaris disease and the fungus *Saprolegnia* sp.

The severity of *P. minibicornis* infection has been found to increase rapidly when individuals accumulate about 370 degree days. As kidney function declines from disease, high temperatures and exercise could lead to cardiovascular collapse. Lab tests with normal-timed sockeye captured in the Harrison River and transported to the lab and held for 24 days at 18°C showed the same pattern of spiking mortality after about 370 degree day exposure.

Modeling with telemetry data: The 2003 LGL telemetry data for Adams sockeye was combined with river/lake temperature data to assess the thermal experience of 50 migrating fish from the Fraser River mouth to spawning grounds. The early-migrating fish spend more time in freshwater than normal-time fish because they do their 'holding behaviour' in lakes adjacent to spawning areas, rather than the estuary as normal-timed fish do. Early-timed fish accumulate ~ 400 degree days by the time they enter Shuswap Lake, and of the fish that leave the lake to enter spawning areas, they have accumulated ~ 700 degree days. That so many fish die in the lake is probably related to high degree day levels. Normal-timed fish enter spawning grounds at ~ 400 degree days.

The 2004 Weaver Creek sockeye telemetry study tracked fish from the confluence of the Harrison and Fraser Rivers, across the complete range of arrival times in the Harrison River. We estimated degree day accumulation for 83 fish based on their arrival dates and subsequent potential migration paths to reach spawning grounds on October 10. Early migrants that stayed in Harrison River would have accumulated almost 800 degree days by the time they had reached spawning grounds—in reality, none of these fish survived. Early migrants that used the lake accumulated almost 500 degree days and had 84% mortality. Those that arrived at the normal time and stayed in the river had about 400 degree days, while normal-timed fish that used the lake had the least exposure of all—less than 300 degree days—and also the best survival.

Temperature manipulation field experiment: To test the hypothesis that migrants exposed to realistic, chronically-high temperatures survive more poorly than those exposed to chronically-low temperatures, we captured Weaver Creek sockeye in the Harrison River in 2004 and held under two different thermal regimes (8ºC vs. 18ºC) in the lab for three weeks, then released them back into the Fraser River tracked them towards spawning grounds. Migration mortality was twice as high among the fish held in warm water (60 % vs. 30 % mortality). Mortality rates of these two groups reflect what you often see in early vs. normal-timed migrants. It is interesting that some fish do survive the higher temperatures.

In summary, early migrating Late-run fish get thermally 'hit' in three ways that are additive (higher than average temperatures linked to migration timing, plus climate change, plus more freshwater degree days). A threshold of 400–500 degree days is a critical range for Late-run fish (there could be a different critical range for other timing-groups or stocks of sockeye). *P. minibicornis* probably plays an important role; other diseases are possibly important but have not been examined. Degree days vs. mortality relationships can provide predictions of en-route mortality, although this approach won't work when fish encounter extreme temperatures which causes mortality because of 'acute' processes (i.e., the T_{crit} issues discussed in the next talk).

DISCUSSION

- Q: How did you define 'normal' timed fish in terms of actual dates in 2004? / A (Hinch): The cut-off date for "normal" fish was September 14, but a buffer was used to separate them into those within the first 10 days ('early') vs. the last 10 days ('normal') of the month.
- Q: Can you tease out climate change effects in your data? / A (Hinch): We are calling the general increase seen in the Fraser climate change. The year 2004 could be the surrogate for what is predicted by climate models in the next few decades.
- Q: That could also be due to a large El Nino. / A (Hinch): This is the upward trend based on data going back to the 1920s.
- Q: Do you assume a threshold when you start counting degree days? / A (Hinch): We are starting to do that now.
- Q: The difference between 370 and 4–500 degree days is a big one. Does the temperature range at which degree days are accumulated matter (e.g., whether it is at 10 or 20 degrees)? / A (Hinch): In the lab you can hold temperatures constant, but how do you deal with real life in the real river where temperatures are going up and down?
- Q: What about disease sources and work to determine cause of death? / A (Hinch): We are not working as closely as we need to be on that issue. We are now involved with another group from UPEI which will be helping us with disease assessments.

ADULT SALMON IN HOT WATER: ADAPTATION, PHYSIOLOGY AND BEHAVIOUR

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ABSTRACT

This talk summarizes and illustrates some of our recent discoveries regarding the swimming ability of adult sockeye salmon when faced with high river temperatures. As a background it is essential to acknowledge that the ability of any animal to exercise rests, in the first instance, with its ability to increase its metabolic rate above its routine oxygen requirements. This is true for both aerobic and burst (anaerobic) swimming since recovery from the later incurs an elevated metabolic rate. Aerobic scope is defined as the oxygen available for activities other than routine and can be estimated in fish using swim tunnel respirometers to permit measurements of routine and active metabolic rates as a function of water temperature. Aerobic scope has a temperature optimum (T_{opt}) where is maximal. At higher temperatures aerobic scope ultimately falls to zero at T_{crit} such that fish are unable to swim at this temperature and are merely surviving.

Aerobic scope, T_{crit} and T_{opt} have been estimated for three stocks of Pacific salmon: Gates Creek sockeye, Weaver Creek sockeye and Chehalis coho. These data reveal that absolute aerobic scope, T_{opt} and T_{crit} vary even among sockeye stocks. Furthermore, T_{opt} is coincident with the historic river temperature profile during the migration period for a given stock, raising the possibility that adult salmon are adapted to the river temperatures that they routinely experience in the Fraser River during their migration. There is only 6–7ºC between T_{opt} and T_{crit}, which is a relatively small temperature window between maximal and no aerobic capacity for swimming.

Fraser River temperatures were exceptionally high in 2004, and these high temperatures were associated with massive in-river losses of Weaver Creek sockeye. River temperature during salmon migration in 2004 exceeded the T_{crit} for Weaver Creek sockeye, but not for Gates Creek sockeye. In fact, biotelemetry studies of migrating Weaver sockeye at different dates in 2004 revealed that fish attempting the migration to the spawning area at temperatures > T_{crit} had zero percent success, whereas there was a 70% success rate when river temperature had seasonally declined to around T_{opt}.

We have definitive evidence that adult sockeye actively avoid warm surface water. This evidence comes from a reconstruction of temperature experiences for Weaver sockeye using acoustic biotelemetry of water depth and from temperature I-button data recovered from sockeye salmon migrating through Anderson and Seton Lakes and other portions of the Fraser River system.

Five possibilities were advanced and tested in individual salmon as to why they cannot get more oxygen when swimming above T_{opt}: not enough oxygen in the water (environmental limitation), gills not ventilated with enough water (ventilation limitation), gills cannot extract enough oxygen (gill diffusion limitation), heart cannot pump enough blood (perfusion limitation), or muscles cannot extract oxygen (tissue diffusion limitation). The first

three possibilities were eliminated because arterial blood sampled post-gill in swimming and resting fish remained fully saturated when temperature was increased to a level (19–24°C), despite the fact that water contains less oxygen at high temperature when swimming activity ceased. Instead, heart rate and cardiac output reached maxima above T_{opt} and resting heart rate reached a maximum at T_{crit}—results that clearly point to a cardiac insufficiency fish swimming at > T_{opt}. The additional observation that blood oxygen tension remained unchanged in salmon swimming above T_{opt} even though muscles were releasing lactate as a signal of oxygen lack. In view of these finding, the following death spiral is proposed for swimming at a temperature above T_{opt}: cardiac pacemaker rate reaches a maximum; heart cannot pump/divert any more blood to muscles; muscles cannot extract any more oxygen from blood; muscles become glycolytic and release H^+; and acidosis impairs heart leading to cardiac arrhythmia.

In summary, different salmon stocks can have different absolute aerobic scopes, as well as different T_{opt} and T_{crit}; however, only 6°C exists between T_{opt} and T_{crit}. Salmon routinely avoid high temperature while migrating. Swimming adult salmon continue to extract sufficient oxygen from water above T_{opt}. The heart reaches its maximum pumping capacity above T_{opt}, and the tissue oxygen extraction becomes diffusion-limited.

ORAL PRESENTATION—TONY FARRELL

This presentation was about aerobic scope and temperature changes (only those related to in-river temperature experiences), possible differences between stocks in adaptations to seasonal temperatures, behavioural avoidance of high temperatures and physiological failure at high temperature.

For humans and fish, as temperature increases, metabolic rates go up, and so does oxygen consumption. Swim tunnel experiments have been done with salmon to measure their maximum ability to get oxygen into their bodies. This maximum uptake rate curves up with rising temperature and then starts to decline. Aerobic scope refers to how much oxygen is available for activities other than routine metabolism—i.e., the difference between the maximum ability to consume oxygen and the minimum oxygen consumption required for routine metabolic activities. Expressing this graphically shows an optimal temperature (T_{opt}) where aerobic scope is maximized and a critical temperature (T_{crit}) where the fish cannot do anything but meet the needs of basic metabolic function, and above T_{crit} death will occur.

Aerobic scope temperature profiles developed for three Fraser salmon stocks (Gates and Weaver Creek sockeye, Chehalis coho) showed that absolute aerobic scope and estimated T_{crit} appear to vary among sockeye stocks. For sockeye, there is only about 6–7° C difference between the stock's T_{opt} and T_{crit}. When river temperatures reach T_{crit} it certainly means the fish cannot move upstream.

Fifty years of historical river temperatures during the migration period for each stock (10 years only for coho) represent a perfect fit to the aerobic scope temperature profile for each stock, indicating that T_{opt} for each stock is potentially adapted to what they normally encounter. So in a particularly warm year like 2004, when river temperatures during migration reached or exceed T_{crit}, it means the fish cannot swim upriver and/or they die. They certainly will not spawn. As aerobic scope declines as temperature move towards T_{crit}, the fish must move to the margins of the river to be able to swim upstream by taking advantage of the boundary layer of water.

A biotelemetry study of returning Weaver sockeye in 2004 at different dates and temperatures showed increased survival to spawning areas during seasonal river cooling. Fish tagged and released on dates where temperature was at T_{crit} had zero survival. Those released at T_{opt} had the highest survival rate. Seasonal cooling clearly had an effect on mortality and this has management implications. Several biotelemetry studies have also shown that adult sockeye actively avoid warm surface water.

Experiments have examined why salmon cannot get more oxygen when swimming above T_{opt} and several hypotheses have been eliminated. Remaining possibilities include that the heart cannot pump enough blood. Tests have shown that the heart can only beat so fast, rising with temperature, even for resting fish. Heart rate even in resting reaches its maximum at T_{crit}. Other possibilities are that the heart cannot pump enough blood (perfusion limitation) or that the muscles cannot extract oxygen (tissue diffusion limitation). A death spiral is proposed to occur for salmon swimming above T_{opt}. As the cardiac pacemaker reaches its maximum, the heart cannot pump any more blood to the muscles and the muscles cannot extract any more oxygen. The muscles become glycolytic and release H^+, then acidosis impairs the heart, leading to cardiac arrhythmia.

In summary, different salmon stocks may have different absolute aerobic scopes, as well as different T_{opt} and T_{crit}. There can be a difference of just 6ºC between T_{opt} and T_{crit}. Salmon can routinely avoid high temperatures when migrating, but if they are unable to do so they have to extract oxygen from water above $T_{opt,}$ the heart reaches its maximum pumping capacity and tissue oxygen extraction becomes diffusion limited.

DISCUSSION

- Q: Given the limiting factor is the heart's ability to pump, how much variability is there within stocks? Can selection act on this? / A (Farrell): Absolutely, yes. Genetics will likely play a role because the difference is probably genetically-based. This also has a tremendous bearing on what we do in hatcheries.
- Q: Which of the factors accounts for the difference between T_{opt} and T_{crit} in a population? / A (Farrell): Two potential mechanisms: How well the heart is oxygenated and how many capillaries there are in skeletal muscle to ensure it is well oxygenated.
- Q: What is the relationship between this and *Parvicapsula* and this odd bleeding observed? / A (Farrell): Experiments were done where all these things were measured. Sockeye were divided into two groups: one that never encountered the parasite in the Fraser River and the other that did. However, the results were inconclusive because we ran out of time and didn't get full development of *Parvicapsula*. This experiment is worth repeating. / A (Hinch): Glen did repeat swims, with some fish exposed to the parasite and others not, and then tested how well they swam the second time, relative to the first. The fish infected with *Parvicapsula* didn't swim as well the second time.
- Q: How does *Parvicapsula* relate to Todd's tagging studies in Weaver Creek? / A (Farrell): The fish were exposed in the Fraser River, so it was developing. It is a co-variable. I said the results were all temperature-related, but the question remains unanswered as to how much might be due to *Parvicapsula*.
- Q: Do I understand correctly that the ones that were tagged later entered the river later, when it was colder? / A (Farrell): Yes. Also, it is fairly clear that a salmon sitting off the Fraser is at 7 or 8ºC, unlike the higher temperatures in the river. / A (Cooke): For fish tagged at Chehalis, I believe we were sampling fish that were newer to the system. / A: Hinch: Both acute and chronic issues are happening. Both were reflected in Todd's data. They were exposed to hot temperatures, but there was also chronic exposure, so it was a layering of critical acute over persistent chronic issues. But not all fish chose to go into the lake and take advantage of deep cooler water, and for those that did not do this it didn't seem to matter whether they came early or late as they tended to die regardless.
- Q: What about thermal load and acute changes? / A (Farrell): There seems to be an ability to acclimate because they make a huge jump when they enter the river. But as they go along, they run into the issue of accumulated temperature units.
- Q: If a stock is coming in and every year the temperature is consistent, the fish don't need a big gap between T_{opt} and T_{crit}. But other stocks that have historically encountered more variability might have a

broader range. / A (Farrell): Yes. In addition, there may also be two different stocks that have the same thermal range.

- Q: … or is it potentially linked to warming and future problems? / A (Farrell): Peak temperature has been tracking up for over 50 years, plus salmon been coming in earlier, and so it is a combination of the two real effects.
- Q: Are there other things that are different about the fish going in earlier—predictive things? / A (Farrell): Miller's data suggests there are predictive things, i.e., there were things that characterize a sockeye as an early entrant. The offspring of these fish are coming back this year so we will see how well they did. It may be that Harrison fish are more predisposed to dive deep in the lake compared to Weaver—maybe that explains the difference.

GENOMICS AND FATE OF LATE-RUN FRASER SOCKEYE SALMON: DISEASE, TEMPERATURE, FATE AND ENTRY TIMING

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ABSTRACT

A combination of radio-tagging and gene arrays was used to identify physiological mechanisms associated with entry timing and fate in the river. Two independent studies in SW and FW designed to identify patterns contrasting fish that survived to the spawning grounds versus those that went missing in the upper river both revealed strong fate-associated profiles in gill, while no signal was observed in white muscle, suggesting that energy was not limiting in migrating fish in 2006. While the signal associated with fate was stronger in FW, the actual statistical associations with fate were stronger in SW. In SW, over 400 genes differentiated the two fate categories, whereas over 1700 genes were differentially regulated in SW. While the biological mechanism was not clear from the SW gene list, the strong signals associated immune response, cell death, inflammation and stress emanating from the FW study indicated that the unhealthy profile associated with upper river mortality was, in fact, a disease profile. We hypothesize that the disease agent is intracellular, possibly a virus or intracellular parasite. Moreover, statistical analyses revealed that unhealthy fish, in addition to a greater probability of dying en route, were also more likely to enter the river early. A closer examination of the gene list revealed that unhealthy fish were osmotically more FW, indicating the possibility of osmotic dysfunction in SW that could have pushed the fish to enter the river early. The gene array experiments have revealed a number of less well-studied genes involved in osmoregulation that may be more sensitive indicators of osmotic preparation close to the river.

ORAL PRESENTATION—KRISTI MILLER

This presentation focused on the use of functional genomics as part of a large multi-disciplinary study to better understand the physiological processes that shift during adult salmon return migrations, with key issues including the relative impact of genetic variation (specific patterns relating to stocks or run-times) and environmental triggers. This research has sought to understand the physiological and environmental basis of abnormal shifts in migration timing.

Biomarkers are genes that can classify between two or more states with high fidelity. The goals of gene array work include trying to find signatures and predictive biomarkers that can help distinguish between, say, groups that survive vs. those that don't, thus allowing managers to predict things like survivability or entry timing. It was originally envisioned that biomarkers of salmon "condition" would be used with test fishery samples to predict the expected proportion that would achieve spawning success, thus helping managers decide whether to open or close fisheries.

This discussion focuses on two studies that used radio tagging and biopsies to provide information on entry timing and fate in-river. The studies involved 2005 Summer-run and 2006 Late-run (Adams) sockeye, with a focus on Adams and up-river mortality in the difficult migration areas. Gill and muscle were the only non-destructive tissue samples available to test for physiological signals, which limited the scope somewhat.

Entry timing: In order to maximize the ability to detect divergence between normal and early entry fish, the two groups were constituted from the front and tail ends of the run, leaving out the middle. So the early group consisted of those fish that passed Mission in August, less than 10 days after being sampled at Johnstone Strait (or less than 12 days after Juan de Fuca), while normal-entry fish were those that passed Mission in September, more than 15 days after release at Johnstone Strait (18 after Juan de Fuca). Note that in 2006, most migrating Late-run stocks entered earlier than their "historic normal" peak entrance date.

A primary hypothesis was that osmoregulatory readiness cues salmon to enter the river early. Telemetry was used to determine exact entry timing and microarrays to look for a signal relating to osmoregulatory readiness in gill tissue that might distinguish between the early and normal timing groups. In direct comparisons between entry-time groups, no signal was seen for initially JS fish, but a slight signal was seen for JDF fish.

A second hypothesis is that somatic energy levels were cueing salmon to enter the river. The study showed that many genes were on a spatial trajectory, changing as the fish migrated, but again, there was only a weak signal associated with entry timing in white muscle tissue.

A third hypothesis was that reproductive maturation cues salmon to enter the river. There is no direct way to test this with microarrays, as reproductive tissue cannot be sampled non-destructively. Direct measurements of reproductive hormones in blood samples were taken instead. In 2006, they did test the hypothalamus in migrating summer and Late-run sockeye and saw other differences between early and normal-timed fish (higher levels of several genes relating to stress response among the early migrants), but no signals for reproductive maturation. The tentative conclusion was that there was no strong support for a physiology signal in gill or white muscle tissue associated with entry timing.

Fate: For this portion of the study, five "fate" categories were created: survivors (including pre-spawning mortalities), upper river mortalities, lower river mortalities, tagging mortalities and fishing mortalities. It is to be expected that factors associated with fate will vary from year to year, along with changing river conditions, so any findings for a given year would be preliminary.

The hypothesis was that there is a physiological predisposition to succumb to poor in-river conditions that is measurable in advance of river entry. Initial analysis showed a strong signal in gill tissue (>400 differentially regulated genes) from fish sampled in Johnstone Strait that was associated with in-river fate. In a bigger experiment with more samples, it was found that the identified signature could be used to correctly classify 88% of Johnstone Strait upper river mortalities and 66% of Johnstone Strait survivors. The significant genes associated with this signature were from many different pathways, many not well understood. Only three genes were linked to osmoregulation, but there was a strong signal linked to ion transport and others related to defence, stress and other pathways. While we had a strong association between physiology and fate, the biological mechanism was not clear from these data.

A second independent study conducted on fish tagged in FW (Whonnock/Glenlyon) in 2006 found over 1,700 genes differentiating two profiles derived through outlier analysis comparing survivors and upper river mortalities. This expansive gene list was applied to all locations sampled in 2006, and again two distinct clusters of fish were resolved at each site, suggesting that the two conditional profiles relating to fate were present at all sites in 2006 (QCI to the spawning grounds). Importantly, relationships between fish sampled in JS and JDFS were the same as resolved in the earlier marine fate study, indicating a linkage in the marine and FW profiles. In fact, 77 genes overlapped between the two studies, 95% of which were differentially regulated in the same direction (up or down in "healthy" versus "unhealthy" fish), indicating that both studies identified the same physiological mechanism, but the signal was considerably stronger in FW. The FW fate-associated gene list was also applied to 2005 Summer-run samples for all locations, again resolving two groups, but fewer fish associated with the "unhealthy" (upper river mortality) group (36–56% of fish in 2006 versus 15–30% in 2005 among all sites sampled). In 2007, approximately 36% of fish were unhealthy in FW. Hence, we have shown that these profiles are consistent over multiple years, but the proportion of unhealthy fish varies. The proportion of unhealthy fish is similar among co-migrating stocks within years.

Healthy/unhealthy profiles: The survivor profile was labeled "healthy" while the profile associated with upper river mortality was labeled "unhealthy." The profile of the "unhealthy" fish included evidence of a significant defence response, cell death, inflammation and very clear stress signals indicting the "unhealthy" fish were, in fact, diseased fish. The type of immune signal emanating from these fish indicated an intracellular infection, likely of viral or parasitic origin. Heatmaps were used to delve deeper into the spatial patterns associated with condition and fate, and showed a very clear signature associated with the two patterns all the way to spawning grounds.

Unhealthy fish carried a more advanced FW osmoregulatory signature, possibly indicating osmoregulatory dysfunction in SW. Importantly, statistical analyses revealed that unhealthy fish were not only less likely to make it to the spawning grounds, they also entered the river faster than healthy fish, possibly due to osmotic disruption in SW. It will be important in future studies to examine whether the infective agent is directly or indirectly responsible for shifts in osmotic capacity. This work led to development of 40 biomarkers, which are to be validated on a broader range of samples. Other next steps include examining the significance of interaction between fate and entry timing, examining whether fate in the marine environment is associated with these same profiles (we only incorporated fish that had made it to the river into these studies), and accumulating additional years of data on associations of the relationship between unhealthy profile and fate among stocks and in different environmental conditions.

These findings led to the hypothesis that the unhealthy profile characterizes a response to *Loma salmonae* infection.[8]

Temperature response: The experiments over the spatial trajectory from QCI to the spawning grounds revealed genes associated with temperature response in muscle, hypothalamus and gill tissue in Savona. Savona is the in-river sample point at the entry to Kamloops Lake that saw some of the highest migration temperatures (18–20° C). Fish sampled at Savona would have been exposed to high water temperatures for at least five days

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[8] Subsequent analysis (after the conference) has confirmed that it is not *Loma* or other myxozoa or microsporidian parasites, and the focus has shifted to viral pathogens.

during their migration, so the response observed at this site was chronic, not acute. The strongest temperature-related signal was in white muscle tissue, and the weakest in gill tissue.

Based on white muscle profiles, there appeared to be two divergent types temperature responses of fish (50:50 ratio), one high responder group that contained enhanced defence, inflammation, wounding stress and ion transport/homeostasis compared to the low responders. Only 12% of fish consistent with the high responder profile were present at the spawning grounds, potentially suggesting only 25% survival of this group.

In summary, fish that are heavily infected and stressed are more likely to enter the river early and to succumb to stressful conditions (high temperatures) in-river. Biomarkers have been developed and are being employed on more fish. The survival of Summer-run Chilko fish is potentially less affected by infection status than Stuart or Lower Adams, but larger sample sizes need to be studied through biomarkers. About half the Lower Adams fish respond vigorously to high water temperature stress, as revealed through profiling of white muscle tissue. This may be associated with higher levels of mortality. Importantly, the low responder group may offer some evidence of adaptive capacity within the stock.

A new Genome BC project will support further work linking physiology and fate, focusing on out-migrating smolts and returning adult salmon.

DISCUSSION

- Q: The early results suggested no osmoregulatory link, but then it was seen later. / A (Miller): This relates to the sample size issue and also the statistical approach. The problem is that not every single fish carries those characteristics, so it is not a perfect distinction, but rather one of probabilities.
- Q: Is osmoregulation a primary or secondary issue? Is disease causing it or is it causing disease? / A (Miller): We don't know. Maybe disease is causing osmotic dysfunction in both environments. We know that genes involved in salt pumping go down in freshwater, but maybe they are going too far down. We need to look at that and at validating and also to see whether *Loma* is the disease.
- Q: Could some of those genes be responding to contaminants? / A (Miller): We think this is a disease profile, not a contaminant profile. It is very typical of an intracellular pathogenic response. In the marine environment, we saw early stage responses, with the down-regulation of protein biosynthesis and some minor disease signals, but in FW, fish carried a profile associated with a full blown intracellular infection, with immune-related apoptotic (cell death) pathways being triggered and associated inflammatory response being initiated. This profile marks the beginning of one that could lead to pathogenesis.

PRE-SPAWN MORTALITY: PATTERNS, PHYSIOLOGY AND TIMING

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ABSTRACT

Pre-spawn mortality (PSM) is most commonly defined for Fraser River sockeye as incomplete spawning, or more specifically the proportion of the eggs retained in a salmon carcass after spawning. There has been a long history of information on PSM for Late-run Fraser River sockeye salmon that has been well documented. Historically, PSM has been correlated with en-route river temperatures, and these correlations improve with proximity of river temperatures to the spawning grounds. The normal pattern of PSM within a year is to see a higher proportion of the initial carcasses examined to have a higher probability of high egg retention. This decline in PSM with sample recovery date has been inconsistent in recent years. In 2006, PSM was high throughout the Weaver and Adams River recovery periods. The limited information on PSM for different groups of Late-run that enter the river at different dates, based on in-river tagging in 2001 and 2002, shows that PSM is higher for early river entry groups. This pattern is consistent for Portage, Weaver, and Late Shuswap fish, but it is only part of the story. Although early entry fish may have a higher probability egg retention these fish are still recovered throughout the spawning period. This suggests that fish from different river entry timing groups do mix, distribute, and spawn throughout the spawning period.

Controlled experiments at Weaver creek channel have confirmed that pre-spawn morts can be recovered at any time during the spawning period. There is no clear pattern between longevity and spawning success, with the exception of there being a minimum amount of time on the spawning grounds for which females need to completely extrude all of their eggs. This pattern is important when considering modelling PSM and temperature exposure. Predicting PSM based on temperature will require knowing both when the fish entered the river and when the fish would likely spawn. This has implications for both the impact that fisheries will have on spawning distribution patterns and in the matching of different diseases with different temperature relationships (i.e., threshold, non-linear).

The limited information on the physiology of PSM has been based on sample comparisons of moribund (fish that are destined to be a pre-spawn mort) and healthy vigorous fish. These results indicate that in recent years body energy status is not related to PSM. However, moribund fish are having difficulty in regulating plasma ions and glucose and are producing high levels of lactate associated with increased anaerobic activity. These physiological perturbations are suggestive that a specific gill or kidney disease may be related to the recent episodes of PSM in Late-run fish.

ORAL PRESENTATION—DAVE PATTERSON

There is a long history of information on Late-run pre-spawning mortality (PSM), which has been shown to correlate with temperature and proximity to spawning ground. Recent PSM spikes for Adams sockeye have exceeded anything seen historically, in data going back to the 1950s. The normal pattern of PSM includes higher egg retention among the earliest carcasses recovered in a year. However, in 2006 a pattern of high PSM was seen throughout the run for Weaver and Adams.

Several large tagging studies have shown that early-entry fish have a pattern of higher overall mortality, PSM and en-route mortality. These include the 2001 Weaver Creek tagging study, and the 2002 Late Shuswap study. Studies have also shown that the fish need at least four days on the spawning ground to spawn successfully. Location and timing of fisheries will likely impact spawning distribution patterns. Predicting time of arrival is difficult, and this confounds the ability to predict PSM based on temperature exposure. Prediction is also complicated by different diseases having different relationships with temperature.

Unhealthy fish are having problems regulating ions, with chloride values below critical levels, suggesting the possibility of kidney or gill disease. PSM fish also have critically-high lactate levels, indicative of metabolism problems, and some problems regulating glucose. This could suggest gill problems and/or disease.

DISCUSSION

- Q: You cannot say that fish that enter early will spawn early so the fact that they're re-shuffling when they spawn may affect heritability. / A (Patterson): The earliest fish do have a somewhat higher probability of spawning early.

DISEASE AND PRE-SPAWN MORTALITY FOR LATE-RUN SOCKEYE

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ABSTRACT

Temperature dependent diseases associated with bacterial and fungal gill infections as well as kidney parasites have been identified in the literature as being causal agents of pre-spawn mortality PSM (defined as the proportion of the eggs retained in a salmon carcass after spawning.) in Late-run sockeye. These field survey studies have often suffered from trying to reconcile normal senescence patterns with histopathology results in linking a specific disease to the 'cause' of pre-mature death. Extensive field surveys in 2006 and 2007 for both Summer and Late-run stocks examined the physiological condition and histopathology from individual healthy and moribund fish. The preliminary results from this work confirm that *Parvicapsula minibicornis* infections within the kidney are ubiquitous amongst all Fraser stocks. However, the severity and incidence kidney *P. minibicornis* infections are not different among healthy and moribund individuals, suggesting that at least in 2006 and 2007 it was unlikely to have been the decisive factor in determine PSM.

The pathology results from 2007 clearly showed a high degree of gill pathology amongst all of the Late-run stocks. This is consistent with 2006 results and with the observation of gill blanching in 2006 and 2007 among moribund fish. More interesting was the novel observation and confirmation that the majority of the gill pathology was likely associated with *P. minibicornis* infection within the gill tissue. In addition to confirming the

presence of *P. minibicornis* in the gills there were also *Loma* infections present in gills, heart, and spleen that were present in all Weaver, Late Shuswap, and Cultus fish.

Future work will concentrate on matching the histopathology results with both behavioural observations and the physiological variables collected from the same individual. The search for single cause or suite of casual agents may be difficult and prove too elusive. PSM is complex and can likely result from a multitude of factors whose efficacy can change both annually and seasonally. Most, if not all, of these factors are accentuated by increases in temperature and freshwater residency times. Therefore, the alterative or short-term advice to management would be to consider the risk of PSM in relation to predictable correlates such as temperature and freshwater residence.

ORAL PRESENTATION—DAVE PATTERSON

There is lots of historical information on Late-run PSM and disease, including recent studies on kidney *P. minibicornis*. A key challenge is teasing apart premature from normal mortality.

P. minibicornis is a kidney parasite with well-established connection to disease in Late-run sockeye. All Fraser sockeye get it. Development is linked to temperature and it is correlated with high PSM in the 1990s. It is thought to have been around a long time. Infection in the kidneys causes very little host response or inflammation (although typical symptoms would correlate with the bleeding disorder noted in an earlier presentation).

Recent studies found evidence of the parasite in gill tissue as well. This was unexpected and the presence of *P. minibicornis* was associated with dramatic pathology including a strong inflammatory response and lesions.

Analysis of kidney pathology at the population level showed no difference between healthy and unhealthy fish. Analysis of gill pathology at the population level for different Fraser stocks showed most serious pathology was associated with gill *P. minibicornis* infection. Unhealthy Weaver Creek samples had higher gill pathology and gill *P. minibicornis* infection rates compared to healthy Weaver Creek samples. For Cultus sockeye, spawning ground recoveries had higher gill pathology and gill *P. minibicornis* infection rates than broodstock samples. *Loma salmonae* parasite was also detected in a number of samples.

In summary, a number of problems were found, which is not surprising for senescent fish. These included the same kidney problems and severe gill problems. *Parvicapsula* infection in the gills is a new finding, as is the finding of *L. salmonae* in gill, heart and spleen tissue. Adams, Weaver and Cultus mortalities were heavily infected with gill and kidney *P. minibicornis*. Cultus sockeye held in cool water had lower pathology scores than wild recoveries, which suggests a possible relationship to temperature threshold or location.

There are several challenges in attempting to link disease to behavior. All fish have kidney *P. minibicornis*. Separating cause and effect is difficult. It is also difficult to distinguish between what is and is not healthy. Some initial work has been done to try to link disease pathology and physiology at the individual level. It has shown tight association among plasma ions and that individuals with high gill infections also have high kidney infections.

This work is ongoing. Future work includes engaging disease experts, linking with work underway on gene expression and a number of other projects.

Meanwhile, in terms of management implications, the PSM/temperature link is like Russian roulette—the higher the temperature, the more bullets in the barrel, with each bullet representing a different disease or physiological threshold. While work continues on trying to identify links, management may need to deal with the higher uncertainty by considering risks relating to predictable correlates such as temperature and freshwater residence.

DISCUSSION

- Q: The highest negative correlation was seen with blood cell counts. Could this tie in with the bleeding issue? / A (Patterson): When we started discussing *Parvicapsula*, it was conceptualized as a ticking time bomb that would blow in 28 days. We are still using that. It is not a simple linear relationship between *Parvicapsula* development and time. There is also a relationship with temperature exposure and it may also depend on the maturation stage at the time the infection was acquired.
- Q: There is a question of whether some of these were secondary issues. / A (Patterson): Very few secondary pathogens were seen in the samples

INTERGENERATIONAL EFFECTS: LATE-RUN SOCKEYE

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ABSTRACT

There is strong evidence that both en-route mortality and pre-spawn mortality are associated with early entry behaviour and the resulting exposure to more adverse environmental conditions for Late-run sockeye. However, what is still unknown is the potential legacy effect of parental migratory stress on offspring viability and fitness. A series of experiments and field surveys have been conducted to assess the potential intergenerational impact that early entry behaviour may have on individual offspring fitness. Thus far there is no strong evidence to suggest that quality of eggs deposited by Late-run sockeye will adversely affect future recruitment.

More specifically, there is no evidence that egg quality declines with spawning date based on sequential gamete fertilization tests of Adams and Weaver sockeye throughout the spawning season. Reproductive investment by females is comparable between healthy and pre-spawn morts in both ovary mass and individual egg size and variance. There is also no detectable change in reproductive investment with sample date. Although there is a strong parental influence on gamete viability there is no clear pattern to suggest that unhealthy females produce inferior eggs. Unspawned moribund and fresh dead females still produced viable offspring in similar proportion to healthy spawners. Controlled experiments have also shown that males exposed to higher currents have lower energy reserves and reduce spawning behaviours. However, there were no differences in the quality of sperm and in the swimming ability of the offspring from behaviourally inferior male parents.

Collectively, these results suggest that gamete quality is highly preserved even at the expense of parental health. Also for interventions conservation concerns could occur at the time of spawning allowing for sexual selection or even partial natural spawning to occur prior to human interference. The implications for management are positive in that there is likely no need for adjustment to future recruitment based on the quality of those eggs that are successfully deposited.

ORAL PRESENTATION—DAVE PATTERSON

Is there a legacy effect from being exposed to warm temperatures? Studies to date show no evidence that egg quality declines with spawning date. Egg-to-fry survival data also show no evidence of a legacy effect. At the individual level, reproductive investment is comparable between healthy and PSM fish and does not change with date. Egg quality does not appear to be compromised in unhealthy females. There is some individual variability, but females don't compromise egg quality, with the possible exception of really extreme temperatures which we have not tested. In controlled experiments, males with reduced body energy had a big reduction in reproductive behavior but no reduction in gamete viability.

In terms of advice to management, there is no evidence that the quality of eggs deposited will adversely affect future recruitment. PSM is complex and likely the result of multiple factors. Most of these factors are accentuated by temperature. Work will continue towards building descriptive and hopefully predictive models. However, in the face of uncertainty, it will be necessary to assess the risk of not knowing what the PSM will be.

DISCUSSION

- Q: Is there evidence of impacts by global contaminants such as PCBs and dioxins? Studies have shown that certain contaminants all end up in the eggs and gonads. / A (Patterson): We are not seeing evidence that embryo survival is dropping, but further study could be done.
- Q: Why don't parasites have good years and bad years? / A (Patterson): We need to look at that and also at whether there are predictors, for example, low water years.
- Q: Spawning and migration time are not very well related so is there any relevance of work on intergenerational effects and entry time...? / A (Patterson): We don't know when a fish entered the river unless it was tagged. The challenge will be to determine out how long an individual fish has been in fresh water through physiological or biochemical methods.

LOST IN TRANSLATION: CONVERTING LATE-RUN SOCKEYE SCIENCE INTO MANAGEMENT ADVICE

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ABSTRACT

Several recent studies associated with the Late-Run Project have validated and provided additional insight into the relationship between Fraser River environmental conditions and en-route mortality experienced by adult sockeye salmon migrants. We now have a better understanding of the pathways through which extreme temperatures and flows can lead to chronic conditions such as increased susceptibility to disease, inefficient energetics or sub-optimal aerobic performance. Fraser sockeye salmon "Management Adjustment" (MA) models were used as a case study to highlight the key questions that must be considered when converting these new scientific findings into advice for salmon managers and to outline some examples of standardized model selection procedures that can be used in this process.

Currently, MA models relating freshwater environmental exposure to discrepancies between upriver and lower river escapement estimates are used in the management of Fraser River sockeye salmon to reduce the uncertainty in estimates of migration success for populations that have survived marine and lower river fisheries.

This difference between escapement estimates ("difference between estimates" or DBEs) represents discrepancies arising due to a combination of factors including: in-river losses resulting from natural causes, unreported catch, errors in catch estimates, and errors in escapement estimation. Forecasted DBEs are used to predict the necessary harvest adjustments to account for the anticipated in-river "losses" and thereby increase the probability of achieving spawning escapement targets.

The abrupt shift in river-entry timing for Late-run sockeye salmon and the associated changes in average freshwater temperature and flow conditions experienced by late entry fish have consistently resulted in a good fit between run timing (represented by the date 50% of the run has passed Hells Gate) and historic escapement discrepancies. However, recent advancements in understanding of the complex relationships between the river environment and sockeye biology introduces a suite of potentially new MA predictors. A standardized approach is required to determine whether this new information enhances our current ability to provide accurate Late-run MA forecasts to fisheries managers for use in pre-season and in-season fishing plans. Although numerous MA models have been identified, in the context of Late-run sockeye salmon research we chose to evaluate and discuss a suite of five predictor variables ranging along a continuum of biological realism and data complexity: 1) run timing, 2) average lower river environmental conditions, 3) most extreme average environmental conditions, 4) weighted average environmental conditions, and 5) accumulated thermal units (ATUs).

The first step towards identifying the appropriate MA model is determining whether a "descriptive" or "predictive" model is required, as different questions and model selection criteria are associated with each. Traditional model selection tools (e.g., AIC) are useful for narrowing large variable sets and identifying models that provide a parsimonious fit to the data (i.e., "descriptive models"). For example, small sample-size AIC (AIC_c) selection procedures identified the *weighted average model* as providing the best fit to historic Late-run DBEs from 1995–2007 (of the 5 models listed above). Model selection was sensitive to the years of data used in the analysis. However, fisheries managers typically require forecasts of future events for use in decision-making. The use of AIC as a performance measure for these types of "predictive models" is limited, as AIC does not take into account uncertainty in predictor variables or extraneous management constraints. Selection of a robust predictive model should consider the trade-offs between model fit and predictor variable uncertainty, in addition to modified variable selection procedures, which take into account current management and forecasting limitations. Although future research is required to quantify the precise nature of these trade-offs for MA models, a qualitative examination of AIC_c values and data requirements for the previously listed variables suggests a shift in top model rank to a *run-timing* based model when these limitations are incorporated into the selection procedure.

The difference in the rank order of model selection when forecast uncertainty and management constraints are evaluated in addition to model fit highlights the need to differentiate between MA models used for simulation or post-season evaluations from those used for pre-season and in-season forecasting. In the face of changing climatic conditions and the known importance of temperature (and flow) on survival of aquatic species, it will become increasingly critical for managers to account for the effects of river environmental conditions on species survival. Future research will continue to quantify the trade-offs among the selection criteria discussed here and work towards developing a robust model-selection framework for future Fraser River sockeye management adjustment models. In addition, the lessons learned from the MA model evaluation may provide useful groundwork for the interpretation of other novel biological information concerning Late-run sockeye into sound management advice.

ORAL PRESENTATION—MERRAN HAGUE

This presentation discussed potential management adjustment models to translate some of the recent Late-run research into practice.

Consequences of early entry include increased en-route mortality and changes to environmental exposure. Numerous studies have shown direct or indirect links between high river temperatures and flows and mortality. Management adjustments, based on forecasted differences between lower and upper-river escapement estimates (DBEs), are used to adjust catches to increase the probability of achieving spawning escapement targets. DBE forecasts are based on river environmental conditions and river entry timing for Late-run sockeye. Management adjustments are quantified through simple regressions that fit historic DBEs to river entry timing. This simplified approach ignores much of the knowledge that has been accumulated through recent studies.

A number of other environmental surrogates can be used to estimate en-route loss. Some potential approaches, in order of increasing biological realism and complexity, are based on 1) river entry timing, 2) average environmental conditions, 3) most extreme environmental conditions, 4) weighted average conditions, and 5) accumulated thermal units. Considerations in deciding which approach is best for translating science into management advice include distinguishing between descriptive vs. predictive models. Model selection criteria for predictive models include biological realism, a good fit between observed and predicted outcomes, model predictive power, model sensitivity, forecasting limitations and management constraints.

Assessing potential models based on model fit alone would favour a model based on weighted average temperature and flow. However, making forecasts with this model would require knowing in advance the relative daily abundance of the run, as well as daily temperatures for the length of the run, both of which are highly uncertain. More work is needed to quantify tradeoffs between improved model fit and increased predictor variable uncertainty. For model sensitivity, different models are a better fit for data from different years, though weighted average conditions would provide the best fit for recent years. Predictive capacity is assessed through retrospective evaluations, making annual forecasts using only the years of data that would have been historically available, to assess predictive ability and to quantify model precision and bias. Management constraints include where and when fisheries are occurring and how this affects forecasting uncertainty and model fit.

So while the weighted average conditions model provides the best fit to the data, the information needed to provide such a forecast is far more data intensive. The inputs needed would be highly uncertain if these forecasts had to be produced in time to inform fishing decisions in-season. This explains why current management is still based on the simplest approach based on the 50% migration date at Mission.

When translating science advice for management, considerations for managers include risk tolerance and management constraints. Considerations for scientists include the best fit to historical relationships, uncertainties in forecasted variables, information available for making the forecast, and key trade-offs, such as between biological reality vs. model complexity or between timing of information and uncertainty in the forecasts.

DISCUSSION

- Q: In using weather forecasts, there is a way to quantify whether you're better off assuming average conditions or not. It may work in one year better than others. / A (Hague): It may be better than using nothing at all. A current study is looking at this uncertainty and whether it affects the choice of which model to use.
- Q: From what has been heard, how close are any of these hypotheses to translation into management. Where should we focus? / A (Hague): Model sensitivity is important. You want a model that will be robust in different circumstances.

INITIAL SYNTHESIS AND PANEL DISCUSSIONS

BEGINNING TO SYNTHESIZE PROGRESS TO DATE

OVERVIEW—SCOTT HINCH

Returning to the list of hypotheses, Hinch noted there are now some correlations between some of the hypotheses, as well as causation in some cases. There appear to be links between energetic, osmoregulation and reproductive readiness. In the ocean environment, there seems to be a group of fish that is unhealthy and are coming in early. In the river, there are both acute and chronic high temperature processes that lead to higher mortality. The potential role of contaminants is unknown, but the correlations with parasites and disease are now evident.

In starting to synthesize research results, we know that the early-entry fish are unique physiologically and less healthy, with higher stress, osmoregulatory dysfunction and disease. These early migrants are thus compromised prior to freshwater entry. Once in the river, they encounter higher temperatures than their run-timing groups are adapted to. Temperatures are often extremely high, plus high river temperatures are occurring more frequently in recent years, and this can contribute to thermal-based mortality due to reduction/elimination of aerobic scope. Early migrants also accumulate higher degree days, leading to sub-lethal, chronic temperature-related mortality arising from stress and disease. Early migrating fish that are physiologically compromised are likely especially impacted by these thermal-based mortality issues.

Participants were invited to comment on this synthesis.

DISCUSSION

- Audience member comment: Given the huge number of fish that disappear in the ocean, it would be interesting to look at those that disappear in terms of healthy vs. unhealthy profiles. Those caught in fisheries should be non-selective vis a vis those that disappeared. The puzzling part is that mortality rates are so high that backing up you would have had very high populations, since they could not have persisted a long time with those profiles.
- Q: In 1994–96, the fish underwent a “regime shift” in terms of the change in migration pattern. In recent years, almost no fish have been coming in at the normal time. It is no longer early and normal but early and extra early. I don’t get a sense in all this that there is research that explains that, so we are missing a key component in terms of what the fish are encountering. / A (Hinch): Agreed. Certainly something has changed in Georgia Strait and perhaps further afield. It may be linked to what Miller is finding—not just out in the open ocean but transitionally. It may be this additive change that is causing the fish to do what they’re doing. They’re already different at the Queen Charlottes but even more different as they get closer. / A (Thomson): The correlation that struck me was the one with the open ocean winds.
- Q: One of the things I heard that is not reflected above is the concept that some of these fish are unique physiologically and more reproductively advanced. Also, “stress’ means so many different things to different people. What does stress mean in this context? Is it a separate factor from disease? / A (Hinch): Yes, stress was meant as something different from disease. And reproductive readiness should have been included. Stress here relates to oxidative stress, DNA damage, etc., not defence.
- Q: You may need to refine that term a bit, as others may have different definitions? / A (Hinch): Agreed.

- Q: Are we talking about a 'regime shift' in fish migration patterns (and if so, that has to be tied to something)? / A (Woodey): There was some variation prior to that but if you look back at Adams, which is documented back to the 1930s, you had that later pattern consistently until the mid-1990s and now there is hardly a sockeye in the river at those later times.
- Q: There was a lot of talk about thresholds. We should start categorizing those. / A (Hinch): Agreed.

MANAGEMENT PANEL

Mike Lapointe, Chief Biologist, Pacific Salmon Commission, Vancouver, BC (Representative on Fraser River Panel, Pacific Salmon Commission)

Jeffrey Young, Aquatic Biologist, David Suzuki Foundation, Vancouver, BC (Representative on South Coast Integrated Harvest Planning Committee)

Tim Tynan, Sustainable Fisheries Division, National Marine Fisheries Service, Lacey, Washington (Representative on Fraser River Technical Panel, Pacific Salmon Commission)

The following questions were put to the panelists who were invited to comment on the workshop proceedings from a management perspective:[9]

1. Do you think any of the research results or observations presented could convert directly into management actions? If so, provide some examples.
2. What scientific findings do you feel are missing and are needed to significantly help in management?

PANELIST RESPONSES TO DISCUSSION QUESTIONS

Tim Tynan: On question one, I would say no, we are still in hypothesis testing and culling. The work underway is all good but we are not there yet. The Fraser Panel is doing the right thing, applying a hefty management adjustment and buffering to deliver fish to the spawning grounds. Some of our stocks are not doing well and we are in risk-averse mode. Canada and the US are both cutting back fisheries.

On the second question, I want to find out more about whether something is happening in the Gulf that is driving fish out of the Strait of Georgia earlier. It seems to be something happening in broad, stepped changes. Where did the fish go historically, where are they going now and what is their preferred habitat? In Puget Sound, rockfish and lingcod mortalities are being seen in August. So there should be water quality sampling in the Gulf, where the fish are holding, and vertical sampling. That should be tied in to the biotic and abiotic aspects. We also should look at harmful algae blooms, which are a burgeoning problem in inland waters worldwide, so I'm not sure why we discarded that here. If you are getting nutrients coming from the city of Vancouver and stratification, perhaps it may be causing algae blooms.

Jeffrey Young: On Question One, I think there are some applications like the management adjustment. There are some applications from the information we now have, but also caveats in terms of how much weight we can put on them. Examples include critical thresholds for salmon. We are also starting to learn about biomarkers and their use in coastal environments to look at mortality. But there are some caveats about being able to use them

[9] Workshop participants also received these questions on a form and were invited to submit written comments as well as joining in the discussions after the panel presentations.

to do more than give general warning signs, given the multiple factors affecting salmon. There are also more issues regarding management of aggregates, given findings about how unique the stocks are. We need to be careful and ensure we are capturing diversity and unique differences that might have implications for management. I think climate change is a serious factor for management, and we are already seeing the types of things that we might expect going forward. The Lates and Early Stuarts are hitting these thresholds first, but when is it going to start affecting the Summers? It would be good to understand more about cool water refugia and how to protect them.

Regarding future research in the coastal environment, it is important to find the trigger for this behaviour, though it may not help in management, and it will be useful to understand biomarkers. Some of these results may relate to salmon more broadly than Lates, so it would be useful to translate the findings for other run-timing groups. It would also be useful, when looking at predictive models and risk tolerance, to have more explicit quantification of uncertainty and to start being more explicit about management objectives and confidence in meeting targets and also about how to relate back to see if you are actually doing that. Some existing management processes are already quite effective, but we need a tighter link between management and science.

Mike Lapointe: We need to talk about what we are already doing now to put this in context. We settled on an exploitation rate of 15%, which relates to having an acceptable level of by-catch while still allowing harvest on stocks that are doing well. This is saying that fish caught in the marine area in early August are the same as fish caught in-river in early August, but the data is showing otherwise. This approach also calls for fisheries to go flat out and then try to stop on a dime. It is difficult to do harvesting in the marine area like this when the information is very uncertain. The consequence of this approach has also been increased pressure on early summers.

So what have we learned and what can we do? When you look at data for different stocks on total mortality over time, it is clear that fish that enter the river early almost all die. This shows that harvesting Late-run fish in mid-August is not going to affect the number of fish that reach the spawning grounds. This is not to suggest that you harvest every fish. If the goal is to get a certain amount on the spawning ground, instead of harvesting 15% in marine areas in early August, you harvest 15% in-river in early August. It can be suggested that these will be the fish of the future, but I don't see how this will help select for survival, if climate change is only going to get worse. Understandably, there will be political obstacles to such an approach, but biologically it makes sense.

Until we have an understanding of what is causing this, we need to continue working on predictive models and we also need to work on other species like Harrison and pinks.

DISCUSSION

- Tynan: I think there is value in delivering those fish upriver. They may be our future. We are not heading in the right direction; we are heading in the wrong direction, so the best strategy is to be risk averse.
- Young: If it were the late-migrating fish instead of the early ones dying we could do that, but the few fish that make it may be more important.
- Tynan: I'm coming from the perspective where our stocks are crashing and governments are being sued. There are many issues, including habitat loss and degradation, but people will go after harvest as the easy fix.
- Riddell: Unfortunately, when the environment shifts, species will pay a very high price in mortality to adapt, but it is important even if only three percent survive. Carl Walters' advice is to just fish at a low stable

harvest rate, which is more in line with the animal being able to adapt to environmental change. There are also stocks in the Late-runs that we are trying to conserve, so that limits the flexibility. This needs a genetics approach, not a harvest approach. Also, if it is correct that run timing in the 1950s was more like it is now, it would be very important to know that.

- Patterson: There were periods of earlier migration, but not as extreme as this.
- Woodey: I think that the information is faulty. While there has been variation, there has been no similar regime shift from early to late or from late to early. The timing of Late-runs migrating in late September and October was consistent for many decades until 1996.
- Thomson: We have been involved in looking at harmful algae blooms and I agree that needs to be looked at further.
- Ennis: Given that Harrison falls within the Late-run group, there would be concern about fishing out that productive Harrison stock. In terms of management actions, we tend to focus on fishing management as opposed to broader options. We need to be cautious about techno-arrogance, but what is the feasibility of helping the fish in freshwater? Can you tap into cooler groundwater to cool critical spots? I'm not sure if this is feasible but we should broaden our understanding of what can be done.
- Tynan: We need to be cautious about relying on techno-fixes and hatcheries. We need to restore and preserve wild stocks. Whatever is happening may be on a broad scale.
- Young: We need to protect the groundwater we have and address the water industry when it is considering things like pulling water out of streams in places that affect salmon.
- Lapointe: The strategy I suggested would actually better protect Harrison because we are currently fishing right on the peak. I'm not proposing that we fish harder; this is just about fishing at a different time. You would still have earlier fish getting up to spawn.
- English: Looking just at 2008, it is easy to say we are not planning any fisheries, so we are taking a conservative approach. But if you go back to 2002, there were questions about what to do when there was a big run of Summers and Lates, in terms of impacts on Cultus. If you have a large run, you may have over escapement or negative interactions between two stocks. How can you say no to fishermen, when that is exactly what you did in 2006? So it is an issue of thinking not just about management today, but in the future where there may be different scenarios.
- Young: It is a trade off—one that we are trying to make every year. It should not just be about fishermen and we should be very explicit if we are making that trade-off. I don't really disagree with Lapointe but we need more research on the evolutionary issue. Is 15% okay? Can we go to harvesting at a better place? The concern is that Lapointe's proposal will go forward and turn into something other than what was intended.
- Farrell: No one mentioned temperature and we stressed that temperature is a major factor. I'm pleased that we haven't eliminated looking at Georgia Strait, but when Miller can predict with 80% confidence at the Queen Charlottes that fish are not going to make it, this has clear implications for management, so it is disappointing that people are not taking up the opportunities.
- Tynan: We are trying to be very sensitive to temperature with the management adjustment models. For the Fraser Panel to start applying some of these results we will need more years, more data and more certainty
- Lapointe: We have a ways to go to validate Miller's work.

- Young: Farrell's work supports the idea of how unique individual stocks are and raises new cautions regarding aggregate management and the implications it has for other stocks. Temperature is implicit in a lot of what we focus on, such as climate change.
- Beamish: To respond to climate change impacts, we are going to need 10, 20 and 30-year strategies, because everything suggests it is going to continue. But we are still not hearing that. The focus is still just on what we are going to do this year or next year.
- Steve: As the only population geneticist on the PSC, I would say we need to learn more about the heritability of these things and we haven't presented anything on that. Regarding which fish we want to get to the spawning grounds, the Cultus program is not targeting the earliest fish for breeding. The trait may evolve spontaneously with these fish but that is not the approach we are applying in most cases. Regarding Farrell's comments, it is important to think about the evolutionary sense. Is what the fish are doing adaptive? We don't know whether or not it is. What would be the cost to those fish of staying in the marine areas? So what is going on in marine areas is also very important.
- Welch: If Miller's analysis is right, would breeding early fish imply breeding diseased fish? In the West Vancouver study, the surprising result was that the fish that survived best were the ones not in saltwater. Looking at what is happening with pinks may also shed light on the sockeye issue.
- Lapointe: Assessment of pinks is not currently being done. Aging studies are needed to assess pre-spawning mortalities.
- Q: How is global warming being factored into long-term planning? / A (Tynan): Our agency has been criticized for not factoring it in to our recovery plans. The US perspective is to hope for the best and plan for the worst. / A Lapointe: The Fraser Panel is reacting year by year. There is no bilateral long-range planning going on.
- Hankin: The Fraser discussions would be improved if they were being done at a stock-specific level instead of aggregates. Are you talking about Adams most of the time when you talk about Lates? Down south, we are going to a finer and finer scale and we now have genetic tools that support that. Looking at the distribution over time, the graph appears goofy, and that is likely because it is an aggregate.
- Riddell: The Province just put out a new water strategy for discussion. If implemented, it would be much more appropriate than the current water strategy and would include things like protection of groundwater.
- Orr: There is no mention of the Wild Salmon Policy as a way to manage. We have this policy and we need to move it forward and put more resources into implementing it. Especially if trying to preserve diversity is seen as important.

SCIENCE PANEL

Dick Beamish, Senior Scientist, Pacific Biological Station, Fisheries and Oceans Canada, Nanaimo, BC

Mike Healey, Emeritus Professor, Institute for Resources, Environment and Sustainability, University of British Columbia, Vancouver, BC

Mike Bradford, Senior Scientist, Fisheries and Oceans Canada, Cooperative Resource Management Institute, Simon Fraser University, Burnaby, BC

The following questions were put to the panelists who were invited to comment on the workshop proceedings from a science perspective:[10]

1. What are the two most novel research findings?
2. What type of research could be conducted to address gaps, inconsistencies and satisfy the needs of management?

PANELIST RESPONSES TO DISCUSSION QUESTIONS

Mike Healey: I didn't see an answer emerging yesterday, though I saw more clarity today. It is useful to have at least a straw man, so I drafted a conceptual model to pull it together: An undefined event may occur in the ocean that triggers a whole series of events (it might be the winds that Thomson discussed or something that happens in early life). This causes a premature shutdown of osmoregulation (there may also be a disease component) and sets up a situation in which the fish are in great stress in the ocean (as per the West Vancouver study where fish in saltwater didn't do well). So they are leaving the ocean because they cannot survive there and entering an environment that is also hostile (high temperatures and parasites). Then they experience thermal exhaustion and death. Such a model could help structure further research and frame key questions, such as whether there is capacity in this early migrating group to develop new genetic capabilities. And what is the relevance or link to global warming?

Mike Bradford: Regarding the most novel research findings, the early disease findings at Cultus are exciting. There are diseases affecting Yukon Chinook, Klamath River salmon, and *Parvicapsula* issues with Fraser sockeye. We should be thinking about disease as a consequence of climate change. Regarding genetics, crickets have two body forms with long and short wings. This is a threshold trait expressed as a result of the expression of growth hormones and affected by a complicated chain of factors. So often we are looking for a smoking gun, but when you look at the complex issues involved in reproductive maturation, it is not hard to imagine a chain of little things leading to new phenotype. I was impressed by the differences showing up all the way out in the Queen Charlottes. Regarding the changes in oceanography, we are also seeing changes in salmon abundance in the ocean, so we should look at whether these are factors.

Dick Beamish: We started this project five years ago when it was predicted that the Adams River stock was becoming extinct. When you look at all the work done to date and then you look at the larger picture of salmon in BC, a key lesson learned from these last two days is that if climate change is a factor, considerable and urgent work lies ahead. Secondly, there is a bigger picture, whether it is Georgia Strait or the Gulf of Alaska. Pinks are now migrating about nine days earlier, and at the same time as the Late-runs began migrating earlier we also saw a change in Coho behavior, where they were leaving Georgia Strait and not returning. So we need to spend

[10] Workshop participants also received these questions on a form and were invited to submit written comments as well as joining in the discussions after the panel presentations.

some time looking at that bigger picture. Thirdly, it is possible that we are seeing an overall change in the migration timing of Late-run stocks. The Late-run stocks on the Fraser are unique in the Pacific. If we are seeing an end to that unique behavior, we may need to manage them the same as all other sockeye.

DISCUSSION

- Healey: I agree that it is a bigger picture and that the issue of Late-run Fraser sockeye needs to fit into that in a way that includes other species and a longer time frame that includes climate change.
- Comment: Regarding Bradford's point, with smolt transformation the decision to move into sea water occurs six months earlier. The point is that these things happen much earlier. Thomson's and Miller's presentations both suggest that things are on a trajectory and are happening much earlier than we had imagined. If so, how do we assess that?
- Healey: If something set that in motion, Miller's work is very exciting. If they are still arriving and spawning at the same times, these set up bookends. The information about oceanographic events is interesting but there is still no indication of a mechanism. It may be a series of events but what are they?
- Beamish: Changes are also occurring in terms of smolts that enter the ocean earlier. Those that do enter earlier (pinks and chums) are doing relatively well because they are finding earlier availability of food.
- Bradford: The high mortality of fish tagged in the Queen Charlottes group may relate to a period when many changes are happening to the fish, and where handling contributed to existing stress.
- English: Conditions for tagging out there were much different. Plus there is the question of whether the fish have to leave the marine environment because they are prepared for freshwater versus whether they have the option and it may be other cues encouraging them to make the choice to leave. Miller's work is looking at the extremes of distribution and small samples. We haven't yet done the work; it needs to be done on all available samples.
- Miller: One difference is that smolts can revert but there is no evidence that adults can do the same.
- Thomson: In trying to understand the return migration to the Gulf, what determines where they make landfall? If it is at the Queen Charlottes, where do they get the signal that they are approaching fresh water?
- Healey: They don't have to make landfall in the Queen Charlottes. The zone is fairly broad and can shift year to year. Regarding when they start developing the shift in osmoregulation, we don't know, apart from the data gathered in this project. Other populations may be quite different but we don't know.
- Beamish: In terms of where the fish are before they hit land, we have repeatedly proposed doing tagging in the Gulf of Alaska. Recent Japanese studies suggest their chum all overwinter in the Gulf of Alaska. Plus there is the issue of Russia increasing their hatchery chum production significantly, so there are good reasons to do more studies out there.
- Welch: In warm years, they seem to make landfall further north because more are caught in Alaska fisheries.
- Healey: Previous modeling work certainly indicated that circulation patterns in the ocean had a big impact on where the fish came ashore. They are still subject to being redistributed by ocean currents. But that doesn't tell us much about physiological state.
- Riddell: English and I tried to tag fish in the 1980s and many fish were dying so we stopped. It seems that it might have been because they were in this physiological state. If things are turned on in the Queen Charlottes it suggests trying to go back a few months further out, and seeing if they are in the same state when they are in the Gulf of Alaska.

- Orr: Is there a way to figure out disease effects by manipulating the health of these fish?
- Hinch: We tried to inject them with *Parvicapsula*.
- Cooke: Engaging more fish health professionals would be useful.
- Bradford: In the Klamath, they are also picking them up as juveniles, so there is a double effect.
- Van der Kraak: Are there technical developments in fish tags that would help solve the problem? Are there probes to detect salinity, as with those used for migratory birds?
- Cooke: A conductivity sensor is simple to do, but getting the appropriate resolution is a challenge, at least for the immediate future. There is interesting work with the POST (Pacific Ocean Shelf Tracking) project and tags that turn back on when the fish are returning, though it is a challenge because you have to tag a lot of fish. Other work includes sampling of salmon sharks. They consume tagged salmon and then transmit data to a satellite when they come to the surface.
- Cooke: The interdisciplinary approach is essential because the problem is so complex. This has been an interesting and unique model, so I advocate a continued team approach, incorporating more of the fish health angle.
- Woodey: We need to give thought to several issues. Miller's results are being interpreted as showing something anomalous, but we don't have a baseline. Is it possible to do similar work on another stock that doesn't seem to be under similar stress, because we cannot go back in time to get a baseline?
- Beamish: That is a good point, though there is no obvious choice for a "control" stock—maybe something on the Skeena or in Alaska.
- Miller: We took the summer stock as a control but everything showed that co-migrating Lates and Summers are very similar. We are not sure if they were similar historically and we are not sure if different species or different system like the Skeena might be helpful.
- Healey: I support more between-stock comparisons.
- Beamish: A North Pacific research proposal should be submitted next year.
- Audience member comment: Were there any "Aha!" moments?
- Woodey: Miller's work raises interesting potential to answer many questions from a different way. The biggest overall finding was that we collected useful information during this period. We had very little information about disease and Farrell's information about physiology also adds to our information base.
- Lapointe: Farrell's temperature and aerobic scope findings provide a very powerful tool in the context of climate change. There is also the issue of the bigger picture and Beamish's points. This may help us to do some triage regarding which are the most vulnerable populations and about their capacity to adapt.
- English: We should keep using the ibutton to add temperature logging to our tagging programs. The work on temperature explains a lot about how fish are behaving so it would be foolish not to do it. The question about whether or not the fish are able to stay in saltwater should be fairly easy to test. We can do DNA tests and put them in a net pen and hold them in the marine environment to see how they make out.
- Healey: Work should be done on stock-specific aerobic scope and temperature thresholds. These fish must have a resource allocation program; they still seem to produce viable eggs, etc., so they are still putting all possible resources into producing viable gametes.

WRAP UP

Gardner briefly reviewed the Day 3 agenda. Hinch welcomed all participants to participate in the discussion about future research and next steps, noting that additional comments were also welcome via the questionnaires. He thanked participants and presenters and all those who had contributed to the success of the event.

DAY 3: NEXT STEPS: FUTURE RESEARCH AND SHARING RESULTS

On the morning of the third day a smaller group of participants discussed next steps. A conversation on reporting out the results of the workshop guided the production of this document. Directions for future research were explored, with a commitment to follow-up discussions. Participants also shared their general impressions of the material covered at the conference and generated some ideas for extension and outreach.

CONFERENCE IMPRESSIONS: BRINGING RESEARCH RESULTS TO MANAGEMENT

Gardner invited participants to comment on whether the conference goals related to fisheries science, fisheries management and extension and outreach[11] had been met. Most of the ensuing discussion focused on concerns that the presentations and discussion failed to convince managers present that research findings to date have immediate and practical relevance to management.

Comments about the lack of success in bringing research results to management included the following:

- We have made huge progress with a problem that initially seemed intractable, but we didn't have sufficiently strong data or presentation to convince managers of this.
- Too much is still on the table—we haven't whittled off enough possible hypotheses.
- We didn't distill it down enough or focus the presentations on management—perhaps it was premature in terms of inviting managers.
- At this meeting we couldn't convince managers with synthesized knowledge backed by scientific consensus.
- People are looking for a silver bullet so you have to show where there is an uncoupling of the evolutionary landscape. Woodey tried to put it in context by describing a "regime shift" in behavior. The record of years of evolved behaviour doesn't change based on a two-year event.

A few specific points on key themes and research results that are clear and should be communicated included:

- There is a sense of being on the cusp—i.e., that with a little more research we can start making more definitive statements and solve problems.
- We have answered some of the questions laid out in 2002 and we need to say that. We also need to address other questions, including new questions such as whether this behaviour is adaptive. Set out clearly what has been done and what still needs to be done.
- What didn't come across was that it is something out in the ocean, not in the strait; and that it is not that the fish have changed but that conditions have changed.
- It is not just ocean departure but what happens in-river, and the presentations made it clear that we can now predict what will happen at specific temperatures.
- It was made clear that if you want to manage 100 stocks, you need to go out and get the data for each one.

[11] See page 1.

Participants responded to the question, "What needs to be done to more effectively reach managers?" with the following suggestions or priorities:

- The recent Skeena science panel report was very effective. Perhaps publish a similar report that includes a clear synthesis of what is known, with clear recommendations.
- Clarity and simplicity is critical, even if the issue is not simple, because buy-in from stakeholder groups is critical. Explain how to put it into the current management operational framework. Could this be done as part of the proceedings? It is a process.
- You need to make predictions and show it works before managers will accept it.
- Much progress has been made, with key questions identified, but it hasn't been coherent enough to convince managers. The focus should be on future research; if we are successful, they will come along.
- You must define who the managers are and what question you are trying to answer. For example, in the context of big changes and uncertainty, a key question to focus on is, should managers set prescriptions that take a chunk of the early stock and does that matter?
- It can be made clear that there is consensus on the in-river questions.
- This project started off very tightly focused on a high-profile stock and expanded into something really broad, with big questions about the whole coast and how to manage in a changing climate. You need to communicate in a way that is dramatic enough to get attention and to be very clear about what question you are answering. For example, is this early migrating group an evolutionary significant unit?

Some issues or challenges raised by participants related to getting research to affect management included:

- The continuing uncertainty about why fish are entering early colours everything else.
- A big body of evidence is needed to be convincing because there will always be naysayers.
- The management focus is short term. How do you actually use that information on an annual basis or is it only useful if you are looking at setting benchmarks and long-term goals? Management systems aren't currently set up to use that information.
- Current management systems are set up to deal with 18 degrees, which sits between T_{crit} and $T_{opt,}$ plus there is the time-dependency factor and the idea of different tolerance levels for different stocks. So the current approach takes a very simple best guess at a complex situation. Do managers want it any finer?
- There are potentially better models but we don't have the data series yet. Managers are gun shy because they've seen indicators that worked well for a while until something unforeseen changed.
- There is still uncertainty because we have no controls. Maybe the salmon have always been predisposed to turn on the osmoregulatory clock out in the ocean except that before they were able to hold and now for some reason they cannot.
- It is premature to determine what managers think. The Fraser Panel is already reacting to temperature. It is a huge driver but we can and should be more sophisticated.

EXTENSION AND OUTREACH

Participants were invited to offer comments on how best to communicate the research results. It was noted that those participating represented a diverse group, but that in reporting out to a broad audience, the information must be "clean," and without so many qualifiers.

Brochure: Participants agreed that it would be useful to produce and disseminate a popular piece to translate and communicate research findings in lay terms to a broad audience. It was agreed that the focus would be on the Late-run issue and that draft text would be thoroughly vetted with appropriate scientists before publication.

A broad synthesis: It was suggested that some other reporting format might be needed to communicate effectively with managers and provide practical advice. This would be a broad synthesis that goes beyond brief "take home messages," but shorter than the Skeena panel report. Comments on this proposal for reporting included:

- This is the type of tool that makes sense and matters to managers. Incorporate the questions they have to answer in dealing with their constituents and in managing the stocks. Explain how to use the information we have to this point and how we envision improvements to the science in future. Explain how to operationalize what we have done—for example, if the prediction is for warm water and strong offshore winds, this is how you would incorporate it.
- To convince managers, you have to synthesize and present something backed by scientific consensus. We could do it over the next year. We have to bring it all together and say, "This is the best assessment."
- It will take time to review this work and time is of the essence.
- There is a year to work on research plans, get buy-in and reach managers, with 2010 as the next opportunity to do significant work on Late-run sockeye.
- This wouldn't replace doing something like the Skeena review.
- The transparent, peer-reviewed underpinning is important, instead of asking people to take our word for it. PSARC can provide a peer-review process; there may also be a role for the Nanaimo staff who focus on translating science into management.
- Hinch is doing an overall report to NSERC by fall 2008.

Summarizing, it was agreed that a more significant synthesis than can be achieved through these conference proceedings, that includes advice to managers, will be explored. Possibilities include using the PSARC process and perhaps support from the PFRCC. The NSERC report due in fall 2008 will include material similar to these proceedings, but in a different format.

FUTURE RESEARCH

Hinch recapped various summaries of what has been answered and what questions remain to be tackled, including his updated list, Healy's conceptual model and Kristi Miller's summary of suggestions from the previous day's discussion. Participant comments were invited, including on further opportunities to mine existing data:

- Work is being done to determine whether accumulated temperature units is simply a matter of being exposed to high temperature longer or whether exposures at different high temperatures matters.

- A temperature logger is needed in the Thompson.
- Various efforts are underway to further analyze past/retrospective data.
- Follow up the suggestion about extending the data for 30 years on peak migration date at Mission.
- Question: Is early entry linked to premature freshwater readiness or to added stress? Without controls, we don't know it is premature.
    - There was evidence of discomfort among the fish held in saltwater in the West Vancouver experiment, but the sample size was small and there were confounding factors; the idea of holding them in pens is good.
    - In swim tunnel experiments, those in saltwater had twice the routine metabolic rate of those in freshwater. They swam at the same level, but after recovery there was a difference in terms of the extra oxygen cost. Those are published data.
    - It suggests they may be able to tolerate saltwater but not stress and saltwater.
- Add reproduction to the list.
- Miller's work involved processing and comparison of all samples that were at least a week apart in entry timing. A tool that is able to differentiate between fish that enter just two or three days apart (i.e., trying to find a continual variable) is not realistic.
- New questions relate to the point that it is not just about T_{crit} and T_{opt}, for a stock, but about how much variability there is among the individuals in that stock. If you can show that the ones that make it are more/less reactive and that there is enough variation for adaptive capacity, you can show that by allowing them to reproduce, you could be creating more adaptive capacity in the stock. There are ways to do that. The hard part is the long-term inter-generational studies of how much selection occurred over time.

WORK PLANNED/UNDERWAY

Participants each listed further work planned or still underway with existing data. This included a number of collaborative projects to integrate and/or expand on existing efforts. The importance of having these findings published and refereed was stressed.

POTENTIAL SHORT TERM PROJECTS

- Pink studies: This could be a significant fishery for the future and little is known about pink mortalities.
    - There are some baseline data for pinks.
    - PSC funding for a large telemetry study would not be available before 2010, but there may be some opportunity for studies of pre-spawning mortality.
    - Fishing boats could provide the marine platform, if tagging costs can be covered.
    - The Ocean Telemetry Network is looking for Pacific projects and has money available for fulltime Canadian academics. This might be a good opportunity—i.e., work on pink in 2009 and then do more sockeye studies in 2010.
- Study of capacity for adaptation to a new thermal regime.
- Feasibility of artificially cooled systems: Test the idea that you can artificially cool systems. Engineer a cool flow and use ibuttons to see if the fish follow it. This would link a number of concepts.

PRIORITY STUDIES FOR 2008 AND/OR 2009

- English will be working on fish wheel platforms this summer, with conventional tagging, but not for Late-run sockeye.
- Hinch and Farrell will be collecting 700 Weaver fish, with thermal stressing at Cultus, doing gene array work, taking eggs to look at inter-generational aspects, and looking at acute handling stress to mimic angling.
- Thomson has funding to develop a model for predicting where Fraser sockeye will make landfall and will need some tagging in the Gulf to help validate that model. (Miller would like samples of those fish, if available. There will be the usual test fisheries, so destructive samples will be available.)

FUNDING SOURCES

Possible funding sources to be explored included the following:

- Fraser Basin Council's Fraser Salmon and Watershed Program and Living Rivers Fund can fund in-river projects. This also provides an effective vehicle for communicating results to the public.
- NR-CAN has funding available through its climate change and adaptations program.
- INAC has new small grants available for work by First Nations.
- The Southern Fund might fund a small pilot study to test feasibility of holding returning adults in marine net pens, but a large-scale pink telemetry study is probably not realistic. Fish farms may also provide access to fallow net pens for such a study.

Some participants stated that they would follow up by looking into specific funding possibilities:

- Farrell and Hinch committed to work with Welch to develop a proposal for funding from the Ocean Telemetry Network. This would be a multi-year study looking at different species and bringing in the whole team. That might be the seed money, leading to Southern Fund funding for 2010.
- Miller committed to follow up with Beamish regarding a DFO proposal for the Gulf of Alaska, for submission to the Valdez fund.
- Cooke committed to follow up on availability of NSERC's new small supplemental strategic network grant.

APPENDIX 1: LIST OF PARTICIPANTS

Name	Affiliation
Brian Riddell	Fisheries and Oceans Canada (DFO)
Al Cass	DFO
Anne-Marie Huang	DFO
Timber Whitehouse	DFO
Keri Benner	DFO
Neil Schubert	DFO
Laura Richards	DFO, and the Committee for Scientific Cooperation (CSC)
Dick Beamish	DFO and CSC
Steve Pennoyer	CSC
David G. Hankin	CSC
Steve Latham	Pacific Salmon Commission (PSC)
Paul Ryall	PSC - Fraser River Technical Panel - Canada
Les Jantz	PSC - Fraser River Technical Panel - Canada
Kim Charlie	Chehalis First Nations
Jeff Young	David Suzuki Foundation
Gordon Ennis	Pacific Fisheries Resource Conservation Council
Craig Orr	Watershed Watch Salmon Society
Mike Lapointe	PSC
Dave Patterson	Fisheries and Oceans Canada
Karl English	LGL Environmental Ltd.
Scott Hinch	University of British Columbia (UBC)
Mike Cooperman	UBC
Ivan Olsson	UBC
Glenn Crossin	UBC
Kim Hruska	UBC
Merron Hague	DFO
Steve Cooke	Carleton University
Kristi Miller	DFO
Tony Farrell	UBC
Rick Thomson	DFO
Dave Welch	Kintama Ltd.
Herb Herunter	DFO
Mike Healey	UBC
Carrie Holt	DFO
Glen Van Der Kraak	Guelph University
Mark Shrimpton	University of Northern BC
Mike Bradford	DFO
Dave Roscoe	UBC
Jim Woodey	PSC (retired)
Steve Macdonald	DFO
Paul Leblond	Pacific Fisheries Resource Conservation Council
Jan Lovy	University of Prince Edward Island

Michael Iconomou	DFO
Tim Tynan	US National Oceanic and Atmospheric Administration
Richard Addison	Consultant
Doug Braun	Simon Fraser University
Eduardo Martins	UBC
Dave Robichaud	LGL Environmental Ltd.
Lisa Thompson	UBC
Karia Kaukinen	DFO
Jennifer Bert	UBC
Jayme Hills	DFO
Aswea Porter	Kintama Ltd.
Melinda Jacobs	Kintama Ltd.
Amy Seiders	PSC - Fraser River Technical Panel - US
Andrew Lotto	UBC
Angela Schulze	DFO
Erica Eliason	UBC
Ken Jeffries	UBC
Tim Clark	UBC
Lucas Pon	UBC
Marika Gale	UBC
Mike Donaldson	Carleton University
Norma Ginther	DFO
Shaorong Li	DFO
Todd Mathes	UBC
Sebastian Pieperhoff	UBC
Julia Gardner	Dovetail Consulting Inc.
Dawn Steele	Note taker - Pacific Fisheries Resource Conservation Council

APPENDIX 2: QUESTIONNAIRE

CONFERENCE ON THE EARLY MIGRATION AND PREMATURE MORTALITY IN FRASER RIVER LATE-RUN SOCKEYE SALMON

Forest Sciences Centre, University of British Columbia, June 16–18, 2008

Please hand this in to one of the organizers at the end of the conference. A summary of written comments will be included in the published proceedings and you will remain anonymous. We would however appreciate if you could identify yourself and your contact information so we could follow up with individuals if needed. Any responses to any of the questions are welcome—this isn't a test! Use extra paper if you like.

Part A

(i) Please provide comments you have on any of the presentations you have seen at the conference (results, interpretations, approaches, etc.) that you were unable to raise in the Q&A periods. (Space for writing was provided under each question)

Part B

The following are questions for discussion on Tuesday afternoon. If you have comments on these questions that you were unable to raise at that time, please provide this information below.

Questions for Management Discussion

(i) Do you think any of the research results or observations presented could convert directly into management actions? If so, please provide some examples.

(ii) What scientific findings do you feel are missing and are needed to significantly help in management?

(iii) Can managers or those providing 'science advice' to managers envision how current or potential information could be beneficial given current management constraints?

Questions for Science Discussion

(iv) What are one or two of the most novel research findings?

(v) What type of research could be conducted to address gaps and inconsistencies, and satisfy the needs of management?

(vi) Please comment on the leading hypotheses, and/or on alternative ones, and suggest research approaches.

APPENDIX 3: COMMENTS SUBMITTED BY PARTICIPANTS VIA QUESTIONNAIRES

Participants were invited to submit their views in writing, on the questions posed to the panelists on Day 2 of the conference, or other themes. Three participants did so. Their comments are as follows.

RESPONSE FROM ANDREW LOTTO—UBC

Part A

(i) Simon Jones (DFO) was able to infect juvenile salmon with *Parvicapsula* in the lower Fraser River during summer holding experiments. If juvenile Harrison sockeye are residing in the lower Fraser during their first year of life, shouldn't they becoming infected with *Parvicapsula* in the summer which would result in some levels of mortality? Yet the Harrison stock is one of the few Fraser sockeye groups with healthy (and growing) populations.

Part B

(ii) There are areas of thermal refuge in the Fraser River Canyon from Hope to Thompson that are well documented by First Nations fishers—fish may be hiding/holding in these to their advantage (recovering from temperature stress, hiding from nets).

(iii) Enhancement on certain Summer-run stocks should be curtailed or re-evaluated—before lake fertilization and enhancement facilities (e.g., spawning channels for Horsefly and Chilko sockeye). What were historical spawning escapements?

(iv) Genomic study results of Kristi Miller are very exciting.

(v) Move beyond sockeye to the other species and life histories. Pink salmon should be priority as they may be the fish of the future with fast growth rates and high thermal tolerance.

(vi) Need more temperature manipulation holding studies not only with sockeye but other species. More ocean studies particularly in the high seas.

RESPONSE FROM MARK SHRIMPTON—UNBC

Part B

(v) Could ocean warming and other environmental changes which are currently happening in the ocean be affecting photoperiod perception and thus affecting cues which affect timing of migration initiation?

RESPONSE FROM MICHAEL IKONOMOU—DFO

Part B

(v) and (vi) Most of the panelists recognized the following problems:

A) The coastal environment plays a major role in salmon survivability.

B) The scope of the research should capture all salmon species and not focused only on Late-run sockeye.

C) The early life history of salmon should also be examined.

I am in agreement with these conclusions and I feel that we should examine all factors that impact salmon at the population level. One major factor that impacts the water quality of the coastal waters is contaminants. The continuous population growth in British Columbia links directly to the increases of wastewater related compounds. These come from industrial sources, agriculture, and sewage treatment plants. Numerous studies including some of our own have shown that contaminants that are present in the Fraser River and the BC coastal waters can cause major adverse effects in fish including salmon. It is unfortunate that we were not given the opportunity to present such findings at the workshop. The water quality presentation given at the workshop did not provide a global picture on the issue of concern and the most recent findings.

Our work and that of others have clearly demonstrated that contaminants can have a major impact on salmon populations. They play a significant role throughout their entire life cycle. Contaminants research will complement well with existing studies and will provide the opportunity for a more holistic perspective on the stressors that could impact this very valuable resource.

APPENDIX 4: COMMENTS SUBMITTED BY R.F. ADDISON

The following unedited comments were submitted via Mike LaPointe June 26, 2008.

COULD ENVIRONMENTAL CHEMICAL CONTAMINANTS PLAY A ROLE IN THE "LATE-RUN SOCKEYE" ISSUE?

R.F. Addison
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The "Late-run sockeye" issue involves two main questions:

1. Starting in 1995, why have "Late-run" sockeye begun up-river spawning migrations significantly earlier (early to mid-August) than the "normal" entry date (averaged over the previous 20 years) of early September?
2. Why has "en-route mortality" increased, coincident with the early onset of upstream migration? Various theories attempt to explain both these observations. In this essay, I consider whether environmental chemicals could play a role. I will assume that any agent (including, but not restricted to, environmental chemicals) proposed as a cause of these effects should meet two criteria:
    a. There should be some synchrony between the agent and the changes in migration and mortality observed starting in the mid-1990's;
    b. A mechanistic cause-effect relationship should be conceivable between the agent and the changes in migration and mortality observed starting in the mid-1990's (in other words, correlation is not enough).

IS THERE A ROLE FOR ENVIRONMENTAL CHEMICALS IN EN-ROUTE MORTALITY?

There seems to be a consensus that the unusually high en-route mortality associated with early entry is due to cumulative stress caused mainly by energy depletion and temperature (extremes, or high averages expressed as degree-days of exposure) (Cooke *et al.,* 2006; Patterson, presentation at this workshop). Premature maturation, osmoregulatory disruption and possibly parasitism by *Parvicapsula* may also be associated with this stress (Shrimpton *et al.,* 2005; Cooke *et al.,* 2006; Young *et al.,* 2006), although it is not quite clear whether these indices reflect *causes* or *effects* of stress leading to en-route mortality.

If any contaminants contribute to en-route mortality, the most likely candidates are the bio-accumulative persistent organic pollutants ("POPs") such as PCBs, PCDD/F etc. which have been accumulated by the fish at earlier stages of their life history. These usually do not meet the first criterion listed above: both uses and environmental concentrations of most POPs have been declining since before the 1990's. However, if other potential causes of en-route mortality changed during the 1990's, POPs may have an exacerbating effect. Some of the POPs are toxic in various ways, and so would meet the second criterion. En-route exposure to other water-borne chemicals in the Fraser can be dismissed: Herunter (presentation at this workshop) showed that concentration of most chemicals routinely measured have not changed over the period when sockeye behaviour changed, i.e., they do not meet the first criterion suggested above. (Nor would most of them meet the second criterion.) Furthermore, the fish do not feed while migrating, and food, rather than uptake across gills, is the main route by which POPs are accumulated by fish. As the fish migrate upstream and their energy reserves are depleted, and as lipid is mobilised into the roe, tissue concentrations of previously-accumulated POPs increase since lipid is metabolised much faster than POPs (Debruyn *et al.,* 2004; Kelly *et al.,* 2007). Concentrations of some POPs in roe may reach levels that are associated with egg mortality in rainbow trout (Debruyn *et al.,* 2004). In summary, the increasing tissue concentrations of POPs resulting from lipid mobilisation and catabolism during migration could have two potentially deleterious effects:

- Exacerbation of stresses caused by other factors, especially late in the upstream migration (though this would be very difficult to demonstrate);
- Increased egg mortality.

Clearly, both these potential effects occur towards the *end* of migration (coincident with depletion of lipid reserves).

COULD ENVIRONMENTAL CHEMICALS AFFECT THE TIMING OF RIVER ENTRY BY LATE-RUN SOCKEYE?

Most candidate pollutants do not meet the "synchrony" criterion outlined above. Wastewater discharges from the main GVRD treatment plants have undergone some compositional changes throughout the 1990's (usually reflecting improvement in discharge quality, as more advanced treatments are introduced: Addison 2002). The only chemicals that *might* meet both of the criteria listed above (for synchrony and mechanism) may be a small group of therapeutic drugs: these could include (but may not be limited to) the statins (which interfere with cholesterol synthesis and the cyclo-oxygenase inhibitors ("coxibs", which interfere with prostaglandin synthesis). A third, remoter, possibility might be the fibrates (lipid regulators), which do not quite meet the "synchrony" criterion, as they were first introduced in the mid-1960's, though their use may have increased later. Statins have occasionally been reported in wastewaters and in "receiving" waters (Miao and Metcalfe, 2003a,b) but concentrations were low (in the low $ng \cdot l^{-1}$ range). Some statins may be mildly bioaccumalative, with octanol-water partition coefficients (expressed as log $K_{o/w}$) in the range 4.0–4.5, corresponding to a fish/water bioaccumualtion factors in the range 2–2,000 (Hernando *et al.,* 2007) but equilibrium concentrations in fish exposed over periods of several days would be unlikely to exceed the $ng \cdot g^{-1}$ range. This seems too low to be likely to have significant physiological effects, even though higher doses of statins inhibit 3-hydroxy-3-methylglutaryl coenzyme A reductase (a key step in cholesterol synthesis) (Estey *et al.,* 2008). There are no reports of the occurrence of coxibs in wastewaters or "receiving" waters. The final group of "candidates", the fibrates, are known to be present in both wastewaters and receiving waters, and at environmentally relevant concentrations (the $\mu g \cdot l^{-1}$ range) reduce testosterone synthesis in goldfish (Mimeault *et al.,* 2005). But as noted above, these do not really meet the "synchrony" criterion.

No data have been published describing concentrations of any of these therapeutic drugs in the Fraser River, although GVRD may have undertaken some preliminary analyses in its wastewaters. Thus, any further discussion of the possible role of pharmaceuticals in the Late-run sockeye issue would be purely speculative.

RECOMMENDATIONS: STRATEGIES FOR FUTURE WORK IN THIS AREA

If we ignore the "conventional" pollutants (discussed above) and focus on the potential for pharmaceuticals to be somehow involved in this issue, there are two obvious (and cheap) steps to take.

1. Has GVRD data describing candidate drug residues in its wastewaters? From such data, and knowing the dilution patterns of wastewater from the main GVRD treatment plants, it would be reasonably simple to calculate potential pharmaceutical concentrations in the lower Fraser River.
2. What has been the trend in prescription of the candidate drugs in the "catchment areas" of the GVRD wastewater treatment plants? This information should be available (at a small cost, I believe) from BC PharmaNet.

If the answers to questions (1) and (2) above suggest that pharmaceutical drugs may be a real concern in this issue, than the next step would be to undertake a "scoping" study of candidate drug concentrations in the lower Fraser system. This is obviously going to be fairly expensive ($10K–$20K I would guess, depending on the scale of sampling and analysis). At least two Canadian laboratories are actively interested in the general question of pharmaceuticals in waste and receiving waters (Trent University and Ontario Ministry of Environment) and since they have the appropriate analytical expertise, it would probably be desirable to involve either of them in any further plans.

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APPENDIX 5: COMMENTS SUBMITTED BY JIM WOODEY

The following unedited comments were submitted via Mike LaPointe June 25, 2008.

A PERSPECTIVE OF THE CONFERENCE ON THE EARLY MIGRATION AND PREMATURE MORTALITY IN FRASER RIVER LATE-RUN SOCKEYE SALMON

The conference organizers and scientists who presented their findings obtained in studies on early migration and premature mortality of Late-run Fraser River sockeye are to be congratulated on a job well done! Convening a focused review of the state of science related to the ongoing abnormal behavior and mortality of affected stocks is admirable and a necessary step in the process of finding answers to the two distinct, but intertwined problems. Below I provide a perspective from "afar" in order to try to keep the scientists' "feet to the fire" and to suggest alternate avenues of inquiry into the issue of early river entry. I feel this is necessary in order that potentially productive areas of research are not overlooked in the rush to focus on a few hypotheses.

BACKGROUND

Late-run sockeye salmon normally enter the Strait of Georgia (SOG) in August and early September and delay there for three to six weeks before entering the Fraser River on their upstream migration to spawning grounds. Beginning in 1995, a variable, but often sizable fraction, of these fish has entered the Fraser River with little or no delay and thus, substantially earlier than observed in the prior years of record. Data at hand indicate little, if any, change in migratory arrival dates in marine areas of Juan de Fuca and Johnstone Straits and Strait of Georgia and in spawning timing. However, the allocation of time between SOG and freshwater areas has changed considerably. Mortality during upstream migration or on the spawning grounds of these fish has led to drastic curtailment of fisheries in order to ensure adequate numbers of adults arrive and spawn successfully. Conservation of the stocks via restrictions to fisheries by Canada and the Fraser River Panel has resulted in severe economic and social repercussions in Canada and the United States. In 2002 alone, there was an estimated $62 million loss of revenue to fishers. Currently, the fate of the commercial fishery is problematic due to the widespread use of fishery closures to mitigate the losses. Near total closure of the commercial and recreational fisheries have occurred in some years and constraints on the harvest by marine and lower Fraser First Nations have adversely impacted communities in the affected areas.

PSC staff organized the first science workshop on the phenomenon in February, 2001, and participated in the first research efforts in the fall of that year. Since 2001, documentation of the behavior and mortality of early entry fish by DFO and the PSC has provided a much richer understanding of the more visible aspects of the phenomenon. Over the past seven years, a number of research programs that have been initiated and conducted by DFO, university and private entities have refined our understanding of the behavior and physiology of the fish and have related migration trends to physical and biochemical variables. Research to date has provided a very good insight into the causes of en-route and pre-spawning mortality of Late-run sockeye salmon that enter the river abnormally early.

While the research to date has provided data on the effects of early river entry, the cause of the phenomenon remains elusive. Statistical relationships inform our understanding but do not provide unambiguous information as to the causes of the early migration.

THE PROBLEM

Early river entry of Late-run sockeye is characterized by three observations that must be reconciled with research findings in order to insure that we have the narrowed in on the cause of the abnormal behavior pattern.

1. Approximately five to 25 sockeye stocks (depending on one's definition of a "stock") appear to have switched behavior beginning in 1995 and have maintained early river entry each year since. The stocks encompass small and large populations (Lower Adams River and Cultus Lake); fish from the upper Fraser watershed (Lower Adams River, Lower Shuswap River and several associated populations and Portage Creek) and the lower Fraser (Cultus, Weaver Creek and Harrison River). Different genetic origins for upper and lower Fraser stocks argues that these stocks evolved a migratory behavior that minimizes the risk of mortality prior to spawning. Therefore, early migration does not appear to be merely the result of the expression of genes that evolved in past history but which now are operating in apparently unintended ways that place the fish in jeopardy of mortality.

2. The consistent annual early entry behavior since 1995 suggests that the phenomenon is not the expression of a random, variable behavior. Lack of observation of early river entry behavior between the 1930s (and possibly earlier) and 1994 and the consistent appearance of the behavior since 1995 argues for a change in the environment that has occurred quickly and has been persistent. This also argues against the effects of random environmental phenomena such as El Nino, PDO, etc.

3. Coincidental early river entry of Fraser pink salmon and potentially early river entry in Fraser chum and Harrison River white Chinook suggests that the phenomenon is focused or expressed by conditions existing in the terminal area, i.e., the Strait of Georgia or Fraser River or the approach routes, Juan de Fuca Strait and Johnstone Strait.

CONFERENCE PRESENTATIONS—EN-ROUTE AND PRE-SPAWNING MORTALITY

Research over the past seven years into the causes of premature mortality in freshwater has gone well and, I believe has provided reasonably conclusive evidence on this particular problem. I think that we can safely conclude that fish are physiologically compromised from early river entry and thence face surviving in freshwater for 3–6 weeks longer than normal before spawning. Infection by an endemic parasite, *Parvicapsula minibicornis*, in the lower Fraser most likely leads to a cascade of stressors that ultimately results in the observed en-route and pre-spawning mortality. The stress of encountering higher river discharge levels and river water temperature in August and early September compared with those found in mid September to early October under normal river entry timing undoubtedly piles stress on the fish making them more vulnerable to parasites and diseases. While other parasites and microorganisms (bacterial and fungal infections) undoubtedly act in collusion with *Parvicapsula*, universal infection by this parasite early in river entry must be cited as the most likely primary source of premature mortality. However, I think that we must consciously watch that we separate the conclusion about the fate of fish in freshwater from the primary question of **why the fish enter the river prematurely**?

CONFERENCE PRESENTATIONS—MARINE PHASE STUDIES

I do want to first recognize the excellent work of Tony Farrell, Scott Hinch, Steve Cooke, Dave Patterson and other scientists who worked on the studies presented regarding the physiological status of the migrating sockeye and of Kristi Miller on her team's gene activation research, all looking for a "smoking gun" in the understanding what induces early river entry of Late-run sockeye. As well, I think that we have a much expanded suite of possible causes to consider through the work of Rick Thomson and Karl English on physical oceanographic factors and "stay with the school" hypotheses. The results from the physiological studies, in particular, are tantalizing and may lead us to conclude that some factor(s) are inducing a proportion of the maturing fish of the affected stocks to switch from adaptation to seawater to adaptation to freshwater in seemingly distant areas and times. The idea that these physiological changes observed in a subset of fish causes them to enter the river early and to subsequently die from early infection by parasites and diseases suggests that we have answers within our grasp.

However, I would caution against jumping to conclusions even after such remarkable scientific results, surprising as they are. First, I must wonder if we are simply observing for the first time a natural cycle of physiological change that prepares fish to adapt to freshwater. It is possible that some fish arriving in the leading edge of the coastal migration would enter the river early in the historical migration period because of adaptation to FW while others were destined for later river entry when their physiological state approached full FW adaptation. The statement that those fish showing physiological transition to FW cannot survive in seawater seems incongruous given that they were captured on their normal migration route in the seawater. That the physiological parameters suggest transition to freshwater, they were still active and kicking in when captured in seawater. We may simply be observing for the first time the natural distribution of physiologically transitioning fish that are nearing the estuary of the Fraser River. Those fish showing advanced transition to FW may not be able to survive the deep water areas of the SOG, given their compromised seawater tolerance physiology, but that, in itself, does not necessarily mean that they are destined to enter the river abnormally early. Fishermen in earlier years knew that a portion of the Late-run sockeye delaying in the SOG could always be found in low saline water along Roberts Bank and in the several mouths of the Fraser. We may simply be seeing in the physiological sampling results the expression of natural variation in the sequence of gene activation, and hence the consequences thereof in biochemical measurements.

As I stated at the conference, we could say that nearly every Late-run sockeye arriving at the mouth of the Fraser between 1996 and 2007 has entered the river earlier than "normal". While an oversimplification, even those Adams sockeye that migrated upstream in early-mid September on the dominant line years of 1998, 2002 and 2006 had shifted from a historical average migration pattern to patterns in which river entry was essentially complete by the time that the average peak abundance occurred (September 21–22) in years between about 1938 and 1994. However, the physiological and gene activation samples collected in JS and JDFS in 2006 did not show **every** fish to have transitioned to freshwater. We need to be cautious in concluding that we are seeing anomalous behavior in these physiological sampling results. Undoubtedly, those fish which are physiologically further along in their transition in samples from the JS and JDFS migration areas are very likely to have earlier river entry among the cohort. However, we are not convinced that we are seeing abnormality. I think that we simply lack the baseline studies that would provide convincing proof that the distribution of observations is evidence of abnormal behavior.

When I stand back and examine the results presented at the Conference regarding early river entry of Late-run sockeye salmon, I do not see a mechanism(s) that adequately explains the phenomenon. While this may be my inability to digest and reconcile the many dramatic results of well designed and very informative studies, I argue that we may be simply documenting normal variation that seems striking simply because we "have gone where no man has gone before" rather than answering the central question.

MY PERSPECTIVE

Much of the research to date has documented the early migration behavior. Marine tagging studies have been instrumental in identifying the components of the annual runs that have entered the river earliest and have associated this behavior with en-route and pre-spawning mortality. Individual tagged fish that have entered the river very early have also shown early physiological changes in preparation to enter freshwater. Preliminary gene activation work substantiates that genes associated with freshwater acclimatization have turned on in these early migrating fish in Johnstone Straits and even seaward to the Queen Charlotte Islands. We need to do much more work on these fish to determine if the observations on timing and location of physiological transition from seawater to freshwater adaptation indicates abnormality. We may simply be seeing the normal distribution of fish approaching the Fraser River estuary. Therefore, obtaining data on the physiological state of normally migrating sockeye salmon may be the only avenue of attack. Whether the study of a Summer-run Fraser River sockeye stock would provide an acceptable control group is problematic. If these fish are affected by the same conditions as are Late-run fish, their physiological responses may also be affected and thus, not provide an independent view of normal physiology and gene activation. Selection of a northern stock (e.g., Babine system or Alaskan fish) may be the only way to determine what constitutes normality.

Also, since pink salmon appear to have also modified their migratory behavior, we should discuss how to approach study of this species to identify analogous changes in their gene activation and physiological transition from seawater adaptation to freshwater adaptation. Other species might be examined to determine if the physiological transition signals are consistent between species and areas.

Secondly, as stated above, my sense is that the factor(s) initiating the early river entry of Late-run sockeye are most likely local to the Strait of Georgia or the Fraser River. Richard Addison provided insight into potential candidate chemicals that research has shown to be biologically active in other organisms and have been in much greater use since the mid 1990's, e.g., the family of statins, which are used to control cholesterol in humans and which are not broken down in water treatment plants. Another potential candidate compound, a fire retardant, was identified in an earlier study by Johannsen.

Development of a rapid bioassay protocols, possibly using electroencephalographic techniques on adult sockeye (technique used by D. Bodznick) wherein fish of known origin (via DNA) are exposed to water sources that elicit a natal response and then to the same water contaminated by test chemicals at concentrations analogous to those currently found in water sampling. Other currently used bioassay techniques may provide superior results, but I am not familiar with current state of research on biologically active substances.

CONCLUSION

While I tend to accept at face value sound scientific arguments, I remain skeptical that the observations presented showing gene activation and physiological signals for transition from seawater to freshwater tolerance and the associated organ system changes (kidney, gill, etc.) do, in fact, signal a "smoking gun" in the search for answers to the question: why are Late-run sockeye entering the Fraser River prematurely? Additional research is required to answer questions relating to the interannual variation in the timing of river entry (stay with the school hypothesis?) and to whether local conditions/contaminants may be overriding natural systems wherein a component of a timing cohort of Late-run sockeye may have less delay in the SOG compared to comigrating fish in the migratory areas studied.

Until unambiguous research findings consistently point in one direction, as I believe we see in the apparent cause of en-route and pre-spawning mortality, I urge that a coordinated approach continue to be used to elucidate basic biological features in the genetic, chemical and behavioral profiles of Late-run Fraser River sockeye salmon.

Jim Woodey
June 23, 2008

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