

# **Monitoring Sockeye Salmon Health in the Cedar River and Lake Washington**

**July 2010–June 2011**

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## **INTRODUCTION**

This report covers the continued health monitoring of sockeye salmon in the Lake Washington basin between July 2010 and June 2011. The Cedar River sockeye salmon enhancement program began in 1991, with the goals of increasing the number of sockeye salmon returning to the Cedar River and providing biologists with an opportunity to study factors affecting sockeye salmon survival throughout their life history. Washington Department of Fish and Wildlife (WDFW) operates the hatchery with funding provided by Seattle Public Utilities (SPU) through the Cedar River Habitat Conservation Plan. Since the inception of the project WDFW has provided fish health monitoring for the Cedar River Hatchery located at the Landsburg diversion dam. In July of 2005 SPU also began contributing funding to support the Fish Health component.

Since the origin of this project the majority of the pathogen monitoring at the facility has been screening for the presence of the viral pathogen infectious hematopoietic necrosis virus (IHNV). This virus is present in the Lake Washington basin and caused substantial mortalities in sockeye salmon fry during a previous incubation project on the Cedar River. Sockeye salmon are extremely susceptible to this virus and it causes a high rate of mortality, particularly at the higher densities of artificial culture. Stringent disinfection and isolation procedures are in place at the current hatchery to avoid infection of the juveniles and minimize spread of the virus if disease should occur. Each fall adults are tested for IHNV throughout the spawning season to ascertain the prevalence of the virus. Each release of fry is also sampled for IHNV and fish are examined grossly and microscopically to assess the condition of the group at release. Juveniles are monitored closely and examined prior to release if there is a concern. Sockeye salmon are also susceptible to other pathogens such as Bacterial Kidney Disease or Bacterial Coldwater Disease but neither have caused disease in this sockeye population.

During most years the naturally produced sockeye salmon fry are also screened for viral pathogens during the outmigration from the river to monitor for the prevalence of IHNV in the wild. When available, fish collected at other points in their life history such as at smoltification, are also examined for viral pathogens and gross abnormalities. Fish may be examined for other fish pathogens if there is evidence of infection or for periodic monitoring.

## **METHODS**

### **Sample collection**

#### **Adults**

Each year, during spawning at the hatchery, ovarian fluid (OF) samples are collected from 24 mature females each week and a total of 60 kidney and spleen (K/S) samples are also collected. Adult return numbers in 2010 were better than had been seen for a number of years and 3,109 females were spawned. The majority of the broodstock were hauled from the new weir site in Renton, but the SPU passage facility contributed approximately 1/5<sup>th</sup> of the broodstock. All samples taken from the broodstock were tested for IHNV and other regulated viral pathogens, as per requirements of the Salmonid Disease Control Policy of the Fisheries Co-Managers of Washington State. No adults were sampled for Bacterial Kidney Disease this year.

No pathogen samples were collected from prespawning adults returning to the system in 2010. WDFW staff collected otoliths from returning adult sockeye salmon captured in June and July at the Ballard Locks by biologists from the Muckleshoot Indian Tribe but no abnormalities were evident in these fish. No abnormal adult mortalities were observed at the locks or elsewhere in the system.

#### **Juveniles**

At release 60 fry were collected from each rearing unit at the hatchery to test for the presence of IHNV. This monitoring assesses the health of the fish at release and is used to evaluate the effectiveness of the IHNV control strategies. To minimize shipping, most weeks some of the fry were held in the incubators and each week only one shipment was made to the lab. If fish in any of the incubators or rearing containers showed abnormal behavior or excessive mortality, a representative sample was sent to the lab at that time to investigate the cause and test for virus. In addition to virology, a small number of fish from all of the rearing units were also examined. Monitoring consisted of examining gills and skin scrapes microscopically for bacteria, parasites and other abnormalities, and to evaluate gill condition. The fry were dissected and examined grossly for residual yolk material, food in the gut, whether fat was being deposited and any abnormalities

Egg to fry survival at the hatchery was excellent for the 2010 broodyear and 8.36 million fry were released. This year the majority of the fry were fed for at least a few days prior to release and many of them for greater than ten days. There were only a few unfed release groups. This practice of feeding the fry has been shown to improve the survival of the fry after release, but the high level of feeding has the potential to cause gill disease so they are carefully monitored.

This year the WDFW Fish Health Laboratory also tested naturally produced sockeye salmon fry for the presence of IHNV. Each week from the end of February through mid March 100 fry were collected at the floating inclined-plane screen trap that is maintained on the lower river to monitor outmigration. In 2011 hatchery produced fry were released at river mile 13.5 as well as below the fry trap. However, fry were collected at least five days after the most recent hatchery releases so the fry collected in the trap were most likely naturally produced.

In May 2011, outmigrating sockeye salmon smolts were collected on a weekly basis for otolith analysis using a purse seine. Each week 20 of these fish were subsampled and tissues collected for viral analysis. These fish were collected in Lake Union or at Weber Point in Lake Washington.

**Laboratory analysis**

Ovarian fluid is generally a more sensitive specimen to test for IHNV in sockeye salmon so it is emphasized during sampling of the hatchery adults. On a weekly basis OF samples were collected individually from randomly selected females and analyzed to determine the prevalence of IHNV in the population. Once during the spawning season kidney and spleen (K/S) tissues collected from adults at the hatchery were combined in 5 fish pools to screen for a viral pathogen (infectious pancreatic necrosis virus) that is not detected in the OF. All fry samples were sent to the Fish Health Laboratory live, euthanized with MS-222 and processed whole, in pools of up to five fish, to assay for viral pathogens. Kidney and spleen tissues were collected from the smolts and assayed individually for viral pathogens. All viral samples were processed fresh and assayed using standard cell culture procedures. The diluted samples were inoculated onto CHSE 214 and EPC cell lines and observed for a minimum of 14 days at 15°C to monitor for the cytopathic effect from viruses. Confirmation of any viral isolates is done using specific antibody or polymerase chain reaction assays. Methodology is used that will detect IHNV, infectious pancreatic necrosis virus, viral hemorrhagic septicemia virus, or Oncorhynchus masou virus. The CHSE 214 cell line will also detect infectious salmon anemia virus, but not as reliably as some other cell lines.

**RESULTS AND DISCUSSION**

**Adults**

During the summer of 2010 the adult return was estimated to be 156,752 sockeye salmon through the Ballard Locks. The fish examined at the locks had light infestations of copepods and occasional wounds, but no external signs of disease. There were no reports of abnormal mortalities of sockeye salmon adults in the basin.

Mortality levels were normal in the adults that were held at the hatchery for spawning. No signs of disease were evident in the fish spawned at the hatchery and IHNV was the only pathogen detected (Table 1). This virus is commonly found in spawning sockeye salmon and is not necessarily an indicator of a problem. In recent year virus has been detected only at a low prevalence and not until the near the end of spawning. However, this year virus was detected by mid October and the prevalence increased each week until it reached 100% at the last spawn. This was the pattern typically seen prior to the 2006 broodyear.

Table 1. Viral results from adult sockeye salmon

<b>Sample date</b>	<b>Viral results</b>
09/16/10	0/2 K/S virus detected, from prespawning mortality
09/16/10	0/24 OF virus detected

09/20/10	0/24 OF virus detected
09/27/10	0/24 OF virus detected
10/04/10	0/24 OF virus detected
10/12/10	1/24 OF and 2/12 pools K/S (5 fish/pool) <b>IHNV</b> detected
10/18/10	3/24 OF <b>IHNV</b> detected
10/25/10	10/24 OF <b>IHNV</b> detected
11/01/10	15/24 OF <b>IHNV</b> detected
11/09/10	19/24 OF <b>IHNV</b> detected
11/15/10	14/14 OF <b>IHNV</b> detected

No screening for bacterial kidney disease occurred this year, however there were no gross lesions indicative of bacterial kidney disease observed during spawning.

### Juveniles

No major problems were encountered this year. As in prior years, each female's eggs are rinsed and waterhardened in 100 ppm iodophor prior to combining them in the incubator. This practice, along with the spring water supply has been very effective in preventing IHNV. Alevins in one incubator exhibited premature swim up with considerable yolk remaining. No apparent cause was detected and IHNV was not detected in samples taken from this incubator. Loss remained normal and this group of fish was reared and released on schedule. No IHNV or other abnormalities were detected in any of the hatchery releases (Table 2).

In general the fish looked very good, with only very slight swelling of gills in some of the fed groups, which was not surprising considering the intensive feeding. Fry accepted the feed well and those groups that received feed for the longer period showed good growth and fat deposition. Rangen soft moist starter feed was used again this year and it continued to perform well. This year not all fry were fed, or fed for only a few days, and small amounts of yolk remained in these fish when examined at release.

Table 2. Health screening results from hatchery produced sockeye salmon fry

Sample date	Vessel	Rearing	Sample type	Virus results	Visual exam
01/25/11	A-5	C-1 & 2	Routine	0/12 pools VD (60 fish)	appeared normal, but no fish were sent for visual exam
01/27/11	A-1/S-20	R-1	routine	0/12 pools VD (60 fish)	normal
01/27/11	A-2/S-20	R-2	routine	0/12 pools VD (60 fish)	Normal, gills slightly swollen
01/27/11	A-2/S-20	R-2	died in transit	0/1 VD	Cephalic bump, no other exam
01/31/11	A-3	R-3	routine	0/12 pools VD (60 fish)	normal
01/31/11	A-4	R-4	routine	0/12 pools VD (60 fish)	normal
02/04/11	B-5	none	diagnostic	0/6 pools VD (30 fish)	Examined due to early swim up, but fish appeared normal, large yolk still present
02/07/11	S-15	C-2	routine	0/12 pools VD (60 fish)	normal

02/07/11	S-18/19	C-1	routine	0/12 pools VD (60 fish)	normal
02/07/11	A-6	C-3 & 4	routine	0/12 pools VD (60 fish)	normal
02/07/11	A-7	C-5 & 6	routine	0/12 pools VD (60 fish)	Normal, gills slightly swollen
02/07/11	A-8	R-1	routine	0/12 pools VD (60 fish)	Normal, gills slightly swollen, little yolk present
02/10/11	A-14	C-4, unfed	routine	0/13 pools VD (60 fish)	Normal, little yolk present
02/10/11	A-15	R-1, unfed	routine	0/12 pools VD (60 fish)	Normal, little yolk present
02/14/11	A-9/S-17	R-3	routine	0/12 pools VD (60 fish)	Normal
02/14/11	A-10/S-17	R-4	routine	0/12 pools VD (60 fish)	Normal
02/14/11	A-12	C-3 & 4	routine	0/12 pools VD (60 fish)	Normal
02/14/11	A-13	C-5 & 6	routine	0/12 pools VD (60 fish)	Normal
02/14/11	S-14	C-1	routine	0/12 pools VD (60 fish)	Normal
02/17/11	S-16	C-7	routine	0/12 pools VD (60 fish)	Normal, gills slightly swollen
02/17/11	A-11	R-2	routine	0/12 pools VD (60 fish)	Normal, gills slightly swollen
02/17/11	S-12 & 13	C-2	routine	0/12 pools VD (60 fish)	Normal
02/20/11	A-22	C-2, unfed	routine	0/12 pools VD (60 fish)	Normal
02/23/11	A-16	R-1	routine	0/12 pools VD (60 fish)	Normal
02/23/11	A-21	C-7	routine	0/12 pools VD (60 fish)	Normal
02/23/11	A-24	C-1, unfed	routine	0/12 pools VD (60 fish)	Normal, little yolk present
02/23/11	B-1	C-2, unfed	routine	0/12 pools VD (60 fish)	Normal, little yolk present
02/28/11	A-17	R-3	routine	0/12 pools VD (60 fish)	Normal
02/28/11	A-18	R-4	routine	0/12 pools VD (60 fish)	Normal
02/28/11	A-19	C-3 & 4	routine	0/12 pools VD (60 fish)	Normal
02/28/11	A-20	C-5 & 6	routine	0/12 pools VD (60 fish)	Normal
02/28/11	B-3	C-1 & 2, unfed	routine	0/12 pools VD (60 fish)	Normal, little yolk present
03/03/11	B-4	C-1 & 2	routine	0/12 pools VD (60 fish)	Normal, little yolk present
03/03/11	B-5	C-7	routine	0/12 pools VD (60 fish)	Normal, little yolk present
03/08/11	A-23	R-2	routine	0/12 pools VD (60 fish)	Normal
03/08/11	B-2	R-1	routine	0/12 pools VD (60 fish)	Normal
03/08/11	B-10	C-1 & 2	routine	0/12 pools VD (60 fish)	Normal
03/21/11	B-6	R-3	routine	0/12 pools VD (60 fish)	Normal
03/21/11	B-7	R-4	routine	0/12 pools VD (60 fish)	Normal
03/21/11	B-8	C-3 & 4	routine	0/12 pools VD (60 fish)	Normal, some gill clubbing
03/21/11	B-9	C-5 & 6	routine	0/12 pools VD (60 fish)	Normal
04/01/11	B-11	R-2	routine	0/12 pools VD (60 fish)	Normal, gills slightly swollen
04/01/11	B-12	C-1	routine	0/12 pools VD (60 fish)	Normal, gills slightly swollen
04/01/11	S-11	C-2	routine	0/12 pools VD (60 fish)	Normal
04/01/11	S-10	T-1	routine	0/12 pools VD (60 fish)	Normal
04/01/11	S-9	T-2	routine	0/12 pools VD (60 fish)	Normal

\* VD = virus detected

No samples were collected from the naturally produced sockeye salmon fry this year until late February. IHNV was detected in the first sample collected but not in the subsequent samples (Table 3). After three subsequent weeks of sampling with no detection of virus sampling was discontinued. Finding this level of IHNV in the wild fry is fairly typical in this system and there have been years in which virus was observed in the fry much later in the outmigration. These fish were only examined grossly, but all appeared normal. Peak river flows were high during egg deposition and incubation in the river and which resulted in poor survival in the river this year.

Table 3. Health screening results from naturally produced sockeye salmon fry

<b>Sample date</b>	<b>Virus results</b>	<b>Visual exam</b>
02/28/11	2/20 pools <b>IHNV</b> detected (100 fish)	no microscopic exams
03/07/11	0/20 pools virus detected (100 fish)	Same
03/14/11	0/19 pools virus detected (94 fish)	Same
03/22/11	0/20 pools virus detected (100 fish)	Same

All smolts collected appeared healthy with no observable gross signs of disease and no virus was detected (Table 4). Virus testing has been performed on smolts or presmolts many of the years since the beginning of this project and no virus has been detected to date. This is an indicator of good health since the stress of smoltification is known to cause expression of virus, often combined with other bacterial diseases.

Table 4. Health screening results from outmigrating sockeye salmon smolts

<b>Sample date</b>	<b>Virus results</b>	<b>Visual exam</b>
05/10/11	0/20 K/S virus detected	no microscopic exams, fish appeared normal with no gross signs of disease
05/19/11	0/20 K/S virus detected	Same
05/24/11	0/20 K/S virus detected	Same
05/31/11	0/20 K/S virus detected	Same