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Characterization of viral haemorrhagic septicaemia virus (VHSV) to inform pathogen transfer risk assessments in British Columbia

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Foreword

This series documents the scientific basis for the evaluation of aquatic resources and ecosystems in Canada. As such, it addresses the issues of the day in the time frames required and the documents it contains are not intended as definitive statements on the subjects addressed but rather as progress reports on ongoing investigations.

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TABLE OF CONTENTS

LIST OF TABLES

LIST OF FIGURES

ABSTRACT

Viral haemorrhagic septicaemia virus (VHSV) is a rhabdovirus and the causative agent of the disease viral haemorrhagic septicaemia which can occur in a wide range of wild and farmed fish species in both marine and freshwater environments. Phylogenetically, VHSV groups into four major genotypes (I, II, III, IV) and ten subtypes (Ia-If and IVa-IVd). In British Columbia, VHSV-IVa is endemic in the marine environment where it causes significant mortality events in Pacific Herring and Pacific Sardines. Less commonly, the virus has been detected in cultured and wild salmon; however, not all salmon species are susceptible to VHSV. Specifically, Sockeye Salmon appear refractory compared to other species such as Atlantic Salmon. Government surveillance programs of farmed Atlantic Salmon in the Discovery Islands area of British Columbia from 2002-2018 detected VHSV in four of the seventeen years tested while there has been no confirmed detection of VHSV among independent surveys collectively testing over 5000 wild Sockeye Salmon. Furthermore, laboratory studies exposing Pacific Herring, Atlantic Salmon, and Sockeye Salmon to VHSV, corroborate a gradient of susceptibility such that Pacific Herring were the most susceptible experiencing upwards of 100% mortality after exposure to extremely low levels of virus. Atlantic Salmon demonstrated low to moderate susceptibility as they remained free of VHSV infection after being exposed to waterborne virus for a short duration; however, became infected when exposed for a longer duration via cohabitation with VHS diseased Pacific Herring. Conversely, Sockeye Salmon, regardless of whether exposed to VHSV through waterborne or cohabitational exposure, for short or long periods, proved refractory to VHSV infection. Consequently, without evidence of either natural or non-invasive experimental VHSV infections, Sockeye Salmon are not considered susceptible to VHSV based on the World Organization for Animal Health (OIE) criteria for listing species as susceptible to infection with a specific pathogen.

INTRODUCTION

Fisheries and Oceans Canada (DFO) has a regulatory role to ensure the protection of the environment while creating the conditions for the development of an economically, socially and environmentally sustainable aquaculture sector. In recent years, restoring funding to support federal ocean science programs to protect the health of fish stocks, to monitor contaminants and pollution in the oceans, and to support responsible and sustainable aquaculture industries in Canada has been identified as a top priority of the Minister of Fisheries, Oceans and the Canadian Coast Guard.

It is recognized that there are interactions between aquaculture operations and the environment (Grant and Jones, 2010; Foreman et al., 2015). One interaction is the risk to wild salmon populations resulting from the potential spread of infectious diseases from Atlantic Salmon (*Salmo salar*) farms in British Columbia (BC) (Cohen, 2012). While several Atlantic Salmon farms are located within the migratory routes of Pacific salmon species, several individual risk assessments have been conducted to date to specifically determine the risk to wild fish populations associated with pathogens released from Atlantic Salmon farms in the Discovery Islands area.

DFO Aquaculture Management Division (AMD) requested formal science advice on the risks of pathogen transfer from Atlantic Salmon farms to wild fish populations in BC. Given the complexity of interactions between pathogens, hosts and the environment, DFO will deliver the science advice through a series of pathogen-specific risk assessments.

PURPOSE OF THIS DOCUMENT

The information summarized in this document will assist in the environmental assessment of the risk to Fraser River Sockeye Salmon (*Oncorhynchus nerka*) due to the transfer of viral haemorrhagic septicaemia virus (VHSV) from Atlantic Salmon farms located in the Discovery Islands area of BC. This document is not designed to be an exhaustive review of VHSV but rather focuses on the occurrence of VHSV in BC with an emphasis on its transmissibility and ability to infect and cause disease in farmed and wild fish species occurring in the Discovery Islands area.

METHODS

A literature search of peer-reviewed articles was undertaken using PubMed, Google Scholar, Google, and the Federal Science Library (FSL), a partnership of seven federal science libraries. The following search terms were used singularly: “viral haemorrhagic septicaemia virus”, “viral haemorrhagic septicaemia”, “VHSV”, “VHS”, and most in combination with "Atlantic Salmon”, “Sockeye Salmon”, “Pacific salmon”, “outbreak”, “infection”, “disease”, “mortality”, “transmission”, “shedding”, “vaccine”, “British Columbia”, and “susceptible species”.

Peer-reviewed literature was the primarily focus however any relevant references cited in any of these papers were also retrieved for use along with non peer-reviewed literature, or “grey literature”. Fish disease reference books and manuals including: Fish Diseases and Disorders Vol. 3 (Woo and Bruno, 2011) and the World Organization for Animal Health (OIE) Manual of Diagnostic Tests for Aquatic Animals were also searched for relevant information. The World Animal Health Information Database (WAHIS) interface was also accessed to determine the number of countries that have reported VHSV detections to the OIE. In addition, using the search terms “fish health reports”, specific searches of the following organization websites were conducted: Government of British Columbia and Government of Canada.

Laboratory data and interpretation of results were requested from DFO AMD. As necessary, phone calls were conducted to supplement information provided through government records or industry reports. Lastly, unpublished results generated from studies conducted by the authors and other DFO science colleagues were incorporated into sections of the document herein where information from peer-reviewed publications were limited. In these instances, the information provided was cited as “unpublished data”.

CHARACTERIZATION

PATHOGEN AND DISEASE

Viral haemorrhagic septicaemia virus is the causative agent of the disease viral haemorrhagic septicaemia which can occur in a wide range of wild and farmed fish species in both marine and freshwater environments. It is an endemic pathogen in marine waters in BC.

By electron microscopy, the virus is an enveloped, bullet-shaped particle, approximately 70 nm in diameter and 180 nm in length. The viral genome is a linear single-stranded negative sense RNA of approximately 11,000 nucleotides with six genes in the order 3’-N-P-M-G-NV-L-5’, encoding the nucleoprotein (N), phosphoprotein (P), matrix protein (M), glycoprotein (G), non-virion protein (NV), and RNA-dependent RNA polymerase (L). Due to its genetic composition, this virus has been taxonomically placed into the species *Piscine novirhabdovirus*, genus *Novirhabdovirus*, family *Rhabdoviridae,* order *Mononegavirales* (Walker et al., 2018). Given its contagious nature and potential to cause significant disease in fish, VHSV is listed as reportable to the OIE.

Gross signs of disease associated with VHSV infection are non-pathognomonic and may include subcutaneous hemorrhage, lethargy, exophthalmia and darkening of the skin. Histological changes in fish with VHS disease most commonly include focal hepatic necrosis, necrosis and hemorrhage in hematopoietic tissues, and necrosis in the intestinal submucosa (Kocan et al., 1997; Marty et al., 1998; Lovy et al., 2012). In addition to causing host pathological changes, infection with VHSV can also trigger both innate and humoral immune responses (Tafalla et al., 2008; Hansen et al., 2012; Millard and Faisal, 2012; Lovy et al., 2013).

GEOGRAPHIC DISTRIBUTION AND GENETIC TYPES

The disease caused by VHSV was first described in 1931 in European farmed Rainbow Trout (*O. mykiss*) and currently the virus is widespread throughout the temperate Northern Hemisphere occurring in 33 countries (WAHIS, accessed Sept 20, 2019).

Phylogenetic analyses have classified VHSV isolates into four major genotypes (I, II, III and IV) and ten subtypes (Ia-If and IVa-IVd) which to some degree correlate with geographical distributions (Einer-Jensen et al., 2004; Elsayed et al., 2006; Schönherz et al., 2018; Guðmundsdóttir et al., 2019) (Figure 1). In addition, genotypes also appear to have some differentiation concerning pathogenicity and host range associations as discussed in the section “Genotype specific host range, susceptibility, and virulence”.

Geographically, genotypes I, II, and III are predominately limited to Europe while genotype IV is present in North America, Iceland and the Asian countries of China, Japan, and Korea (Figure 1). Within North America, VHSV genotype IV isolates are further delineated into three subtypes (IVa, IVb, and IVc) with IVa detections occurring on both the east and west coasts, while IVb is only present in the Laurentian Great Lakes region, and IVc is limited to the estuarine waters of New Brunswick and Nova Scotia (Figure 1). All VHSV isolations in BC belong to genotype IVa

(Garver et al., 2013b). To date, VHSV genotype IVd has only been identified in Iceland (Guðmundsdóttir et al., 2019).

Figure 1. Geographic distribution of viral haemorrhagic septicaemia virus (VHSV). Legend for the genotypes: genotype IVa (●; magenta), genotype IVb (●; orange), genotype IVc (●; beige), genotype IVd (●; purple), genotype III (●; green), genotype II (●; black), genotype I (●; blue).

GENOTYPE SPECIFIC HOST RANGE, SUSCEPTIBILITY, AND VIRULENCE

Infections with VHSV have been described in over 80 species encompassing the bony fish orders of: Clupeiformes (herring, pilchard, sprat); Gadiformes (cod, hake, burbot, pollock); Pleuronectiformes (flounders, soles, plaice, dab, halibut, turbot); Osmeriformes (smelt); Perciformes (perch, drum, sand lance, sand eels, gobies, temperate basses and sunfish); Salmoniformes (salmon, trout, whitefish, grayling); Esociformes (pike, muskellunge); Scorpaeniformes (rockfishes, sculpins); Cypriniformes (minnows, carp); Gasterosteiformes (sticklebacks, tubesnouts); Cyprinodontiformes (mummichog); and Anguilliformes (eels).

Despite VHSV's capacity to infect a broad range of hosts, not all species are universally susceptible to all genotypes of VHSV. Across the different VHSV genotypes, there appears to be specific host preferences such that one genotype may exhibit a higher virulence to a particular host species while another genotype is avirulent to that same species. For example, genotype Ia is highly virulent in Rainbow Trout causing serious disease and mortality while genotype IV demonstrated low virulence in this same species (Emmenegger et al., 2013).

Sequence comparisons between virulent and avirulent isolates have identified a number of changes across the viral genome that may dictate whether an isolate becomes pathogenic or not (Betts and Stone, 2000; Campbell et al., 2009). The viral G-protein, due to being the major antigenic protein, has been suggested to be an important contributor to the pathogenic mechanism of VHSV (Bearzotti et al., 1995; Stone et al., 1997) yet others have demonstrated that differences in virulence among phylogenetically distinct isolates of VHSV are not explained by variability of the G-protein or the non-virion (Nv) protein (Einer-Jensen et al., 2014). Further, subsequent studies using reverse genetics rejected the hypothesis that the glycoprotein (G), non-virion (NV) and polymerase (L) were major determinants of virulence (Yusuff et al., 2019), but rather the nucleoprotein (N) and phosphoprotein (P) genes are the likely determinants of VHSV virulence in Rainbow Trout (Vakharia et al., 2019).

As the genetic make-up of VHSV is a major determinant of virulence, it is essential that assessments evaluating the risk of VHSV infection be specific to those genotypes which occur

in the location of interest. Given that genotype IVa is the only VHSV genotype found in BC and the surrounding northeastern Pacific Ocean (Hedrick et al., 2003; Garver et al., 2013b), this review focuses on the occurrence, host susceptibility and virulence of this specific genotype. VHSV genotype IVa is an endemic pathogen to the marine waters of BC (Garver et al., 2013b).

VHSV-IVA: HISTORICAL PRESENCE ON THE WEST COAST OF NORTH AMERICA

VHSV was isolated for the first time in North America in 1988 during routine fish health checks on apparently healthy returning adult Coho (*O. kisutch*) and Chinook (*O. tshawytscha*) salmon at two different Washington State hatcheries (Brunson et al., 1989; Hopper, 1989). Due to its novel identification, increased testing was undertaken (particularly of salmonids) for several years resulting in five more isolations, again from apparently healthy adult Coho Salmon returning to hatcheries in Washington State (Meyers and Winton, 1995). By the early 90's, the host and geographic range of the virus in North America was found to extend from Alaska to California with the virus primarily being detected in wild marine fish species experiencing high mortality. In Alaska, VHSV was found responsible for die-off events of Pacific Herring (*Clupea pallasii*), Pacific Hake (*Merluccius productus*), and Walleye Pollock (*Theragra chalcogramma*) (Marty et al., 1998; Meyers et al., 1999). Likewise, in BC, numerous detections of VHSV were made in Pacific Herring and Pacific Sardine (*Sardinops sagax*) displaying external hemorrhagic lesions and often undergoing epizootic losses (Traxler et al., 1999). In Oregon, the virus was detected in Eulachon (*Thaleichthys pacificus*), Surf Smelt (*Hypomesus pretiosus pretiosus*) and Pacific Sardine, while in California, the southernmost range of the virus, VHSV was evident in Pacific Sardine and Pacific Chub Mackerel *(Scomber japonicus*) (Hedrick et al., 2003).

It remains unclear as to the exact origin of VHSV-IVa in the marine water of the northeastern Pacific Ocean. Based on historical reports from BC that describe mortality events of Pacific Herring and Pacific Sardine in 1941 (Foerster, 1941) and 1942 (Tester, 1942), with clinical signs similar to that of present day VHSV die-off events, it is probable that VHSV was present in this watershed prior to its identification in 1988.

Although few studies have estimated evolutionary timescales for VHSV, it is postulated that the primary clade divergence of the European VHSV lineages and that of the North American genotypes is on the order of 300-500 years (Einer-Jensen et al., 2004; He et al., 2014). As the evolutionary history of VHSV appears to involve ancestry endemic to the Pacific Ocean, it is speculated that the divergent split of VHSV likely occurred in the Pacific Ocean rather than the North Atlantic and presumably disseminated from the Pacific westward to East Asia, eastward to the North Atlantic/Baltic Sea, the Great Lakes, Atlantic Canada watersheds, and the Atlantic coast (He et al., 2014).

VHSV-IVA: HOST RANGE AND VIRULENCE

In the coastal waters of the northeastern Pacific Ocean, VHSV-IVa has been detected in 17 fish species (Table 1) that differ in susceptibility to the virus. Pacific Herring, Pacific Sardine, Pacific Sandlance (*Ammodytes hexapterus*), Pacific Hake, and Walleye Pollock are extremely susceptible to VHSV-IVa with infection often resulting in severe disease epizootics of these species (Meyers et al., 1999; Traxler et al., 1999; Kocan et al., 2001). In contrast, Pacific salmonids (*Oncorhynchus* spp.) appear to be of negligible susceptibility with VHSV-IVa infections being uncommon and occurring in apparently healthy adult fish (Meyers and Winton, 1995). For farmed Atlantic Salmon in this region, the species is considered of low to moderate susceptibility due to the occasional detection where in some instances it has resulted in VHS diseased fish (Garver et al., 2013b).

Table 1. Host species from which viral haemorrhagic septicaemia virus genotype IVa (VHSV-IVa) has been detected in the coastal northeastern Pacific Ocean.

Common Name	Scientific Name	Reference
Pacific Herring*	*Clupea pallasii*	Meyers et al. (1999); Traxler et al. (1999)
Pacific Sardine*	*Sardinops sagax*	Meyers et al. (1999); Traxler et al. (1999)
Pacific Sandlance	*Ammodytes hexapterus*	Kocan et al. (2001)
Pacific Hake*	*Merluccius productus*	Meyers et al. (1999)
Walleye Pollock*	*Theragra chalcogramma*	Meyers et al. (1999)
Pacific Cod	*Gadus macrocephalus*	Meyers et al. (1992)
Three Spine Stickleback*	*Gasterosteus aculeatus*	Kent et al. (1998)
Surf Smelt	*Hypomesus pretiosus pretiosus*	Hedrick et al. (2003)
Pacific (Chub) Mackerel	*Scomber japonicus*	Hedrick et al. (2003)
Eulachon	*Thaleichthys pacificus*	Hedrick et al. (2003)
Sablefish*	*Anoplopoma fimbria*	Traxler et al. (1999); Hedrick et al. (2003)
Shiner Perch*	*Cymatogaster aggregata*	Hedrick et al. (2003)
Steelhead Trout	*Oncorhynchus mykiss*	Brunson et al. (1989)
Tubesnout*	*Aulorhynchus flavidus*	Margolis (1995)
Chinook Salmon*	*Oncorhynchus tshawytscha*	Meyers and Winton (1995)
Coho Salmon*	*Oncorhynchus kisutch*	Meyers and Winton (1995)
Atlantic Salmon*	*Salmo salar*	Garver et al. (2013b)

**Species within BC with VHSV detections*

This gradient of host susceptibilities to VHSV-IVa as experienced in the field has been further substantiated through laboratory exposure studies. Pacific Herring, a species associated with recurring VHSV epizootics in the field, were highly susceptible to VHSV in laboratory exposures with upwards of 100% mortality occurring when fish were either injected with or waterborne exposed to extremely low levels of virus as low as 0.07 plaque-forming units (PFU) per mL (Kocan et al., 1997; Hershberger et al., 2007; Hershberger et al., 2010b; Hershberger et al., 2011). Pacific salmonids, with limited occurrences of VHSV infections in the natural environment, have been shown to be equally refractory to VHSV infection under laboratory conditions (Table 2). Among studies that simulated natural VHSV exposure pathways, such as waterborne immersion, Chinook, Pink, and Sockeye salmon remained free of VHSV infection and VHSV-associated mortality despite exposures to VHSV-IVa at doses one hundred to ten thousand times greater than that used in the aforementioned Pacific Herring experiments (Follett et al., 1997; Traxler et al., 1999; Gross et al., 2019). In addition, Sockeye Salmon proved refractory to VHSV-IVa infection when exposed to the virus for long periods of time through cohabitation with VHSV-infected Pacific Herring (Gross et al., 2019).

Table 2. Summary of laboratory studies that exposed Pacific Salmon species to viral haemorrhagic septicaemia virus genotype IVa (VHSV-IVa) under conditions mimicking natural transmission pathways. PFU: plaque-forming units.

Species	Challenge Method (Dose)	Average VHSV associated mortality	Reference
Rainbow Trout Coho Salmon	Immersion (10^5 pfu/mL)	7%[a] 3%[a]	Winton et al. (1991)
Chinook Salmon Coho Salmon Pink Salmon Sockeye Salmon Rainbow Trout	Immersion (10^3 and 10^5 pfu/mL)	0%[b] 0%[b] 0%[b] 0%[b] 12%[b]	Follett et al. (1997)
Chinook Salmon Sockeye Salmon	Immersion (Not reported)	0% 0%	Traxler et al. (1999)
Sockeye Salmon	Immersion ($6.7x10^3$pfu/mL)	0%	Gross et al. (2019)
Sockeye Salmon	Cohab w/ VHSV-infected Herring	0%	

[a] *Testing to confirm presence of VHSV in mortalities was not reported. VHSV-associated mortality is based on differences between VHSV exposed and unexposed treatments.*

[b] *Non-VHSV-associated mortality occurred in both the VHSV exposed and unexposed treatments. All mortalities were screened for VHSV to differentiate mortalities associated with VHSV infection.*

In Atlantic Salmon, laboratory studies have reflected a low to moderate susceptibility across multiple VHSV genotypes (Dale et al., 2009; Lovy et al., 2013; Guðmundsdóttir et al., 2019). Specific to VHSV-IVa, Atlantic Salmon exposed to 6.3 x 10^3 PFU/mL by immersion for one hour remained absent of infection (Gross et al., 2019); however, when virus was administered by prolonged exposure through cohabitation with VHS-diseased Pacific Herring, VHSV infection was evident and a low level of disease (5-8%) occurred in the residing Atlantic Salmon (Lovy et al., 2013; Gross et al., 2019). Higher levels of Atlantic Salmon mortality, 40-90%, were only observed through the invasive procedure of intraperitoneal injection of VHSV at a dose of $7x10^3$ PFU/fish (Traxler et al., 1999) or $5x10^5$ PFU/fish (Lovy et al., 2013). Clinical signs associated with VHS disease including dark colouration, hemorrhaging of the skin and exophthalmia were observed in Atlantic Salmon succumbing to VHSV infection in laboratory challenges (Lovy et al., 2013).

VHSV-IVA IN BRITISH COLUMBIA

In BC, VHSV-IVa was first identified in 1993 from Pacific Herring incurring mortalities in and around the marine waters of Prince Rupert Harbour (Traxler and Kieser, 1994). Since this report, mortality events of Pacific Sardine and Pacific Herring have been reported at various locations in coastal BC. During these epizootics, VHSV is typically isolated from moribund and fresh dead fish. The geographic extent of the losses varies from being quite localized to huge areas covering many square kilometers. Field observations made by experienced Pacific Herring biologists estimate several of the die-offs involving Pacific Sardine to be in excess of 5000 metric tons, with the surface of some inlets being covered with dead and dying fish for

distances of over 15 km (Garver et al., 2013b). Routine surveillance or monitoring programs for VHSV-IVa within Pacific Herring populations of BC have not been undertaken, however, through opportunistic sampling of mortality events, VHSV has been detected 44 times over a 19-year period (Garver et al., 2013b), revealing the endemic nature of this virus in these two pelagic species. Though Pacific Herring and Pacific Sardine appear to be natural hosts for VHSV-IVa, in BC, the virus has also been identified in nine other species as listed in Table 1. Among the salmonid species VHSV-IVa has been detected in both farmed and wild populations.

Farmed Salmonids

The first reported isolation of VHSV in farmed salmonids in North America occurred in Atlantic Salmon in the marine waters of BC in March, 1995 (Margolis, 1995). It has since been sporadically detected in farmed Atlantic and Chinook salmon whereby detections often co-occur with VHSV detections in other pelagic fish species found in and around the net-pens (Garver et al., 2013b). VHSV within farmed salmonid populations has been identified either through the Fish Health Audit and Surveillance Program (FHASP) or through industry reporting of fish health events.

Fish Health Audit and Surveillance Program

The FHASP aims to audit 30 active farms each quarter. Audits were conducted by BC provincial government from 2002-2010 and afterwards by DFO AMD (2011-present). A maximum of 30 fresh fish are selected for histopathology, bacteriology and molecular diagnostics/virology (Wade, 2017). It is worthwhile to note, however, that a positive VHSV test result does not necessarily indicate clinical disease or equate to a farm-level diagnosis. Veterinarians review all relevant records, on-site observations and test results to establish whether or not there is a farm-level diagnosis of VHS.

Atlantic Salmon

A total of 1446 audits have been conducted on BC Atlantic Salmon farms between 2002 and 2018 (Table 3), screening over 8,700 fish, resulting in 17 farm-level diagnoses of VHS disease (Table 4). Within the Discovery Islands area (sub-zone 3.2), 279 audits have been performed, resulting in four molecular detections (Table 5) and no farm-level diagnosis of VHS (Table 4). Three of the four molecular detections in zone 3.2 occurred in the first quarter (Jan-Mar) while the detection in 2012 was from an April 5th, 2012 audit (DFO, 2019b).

Table 3. Number of audits from the Fish Health Audit and Surveillance Program (FHSAP). Values in parentheses are the numbers of farms that were audited. The three farms licensed in fish health surveillance zone 3.3 (Althorpe, Hardwicke and Shaw Point) are grouped in zone 3.2 as per Aquaculture Management Division (AMD) practices. Source: data collected by the BC provincial government, 2010, and DFO Aquaculture Management Division (2011-2018 data were downloaded from the Open Canada website September 6th, 2019). 'N/A' indicates no audits.

Year	Fish Health Surveillance Zone and Sub-Zone									
	2.1	**2.2**	**2.3**	**2.4**	**3.1**	**3.2**	**3.3**	**3.4**	**3.5**	**Σ_{year}**
2002	1 (1)	N/A	5 (5)	3 (3)	1 (1)	3 (3)	4 (4)	4 (3)	N/A	**21**
2003	3 (1)	N/A	13 (10)	13 (6)	3 (2)	10 (4)	19 (10)	11 (5)	N/A	**72**
2004	N/A	N/A	13 (6)	13 (8)	8 (4)	13 (8)	22 (14)	9 (5)	1 (1)	**79**
2005	N/A	N/A	15 (9)	12 (7)	7 (3)	18 (12)	23 (15)	11 (5)	3 (2)	**89**
2006	N/A	N/A	16 (9)	13 (10)	4 (3)	19 (12)	20 (13)	8 (5)	6 (4)	**86**
2007	N/A	N/A	19 (10)	17 (11)	4 (2)	24 (13)	22 (12)	12 (5)	6 (3)	**104**
2008	N/A	N/A	15 (9)	12 (7)	3 (2)	28 (14)	24 (14)	8 (6)	8 (3)	**98**
2009	N/A	N/A	20 (11)	15 (10)	4 (2)	23 (14)	20 (12)	13 (6)	5 (3)	**100**
2010	N/A	N/A	6 (6)	4 (4)	1 (1)	4 (4)	5 (4)	5 (5)	N/A	**25**
2011	N/A	N/A	11 (7)	9 (6)	4 (3)	13 (8)	11 (8)	6 (4)	3 (3)	**57**
2012	N/A	N/A	18 (11)	16 (7)	3 (2)	23 (12)	22 (12)	9 (6)	9 (4)	**100**
2013	N/A	N/A	23 (11)	14 (6)	1 (1)	12 (8)	27(14)	9 (6)	8 (4)	**94**
2014	N/A	N/A	19 (11)	18 (9)	3 (2)	16 (8)	22 (11)	8 (5)	6 (3)	**92**
2015	N/A	N/A	22 (11)	17 (8)	11 (6)	18 (9)	21 (13)	10 (7)	8 (3)	**107**
2016	N/A	N/A	22 (11)	11 (6)	8 (4)	21 (11)	25 (13)	11 (6)	7 (4)	**105**
2017	N/A	N/A	19 (11)	20 (9)	7 (3)	16 (9)	25 (12)	14 (7)	7 (3)	**108**
2018	N/A	N/A	18 (9)	18 (10)	10 (5)	18 (9)	23 (12)	15 (7)	7 (3)	**109**
$\Sigma_{subzone}$	**4**	**N/A**	**274**	**225**	**82**	**279**	**335**	**163**	**84**	**1446**

Table 4. Number of Fish Health Audit and Surveillance Program (FHSAP) farm-level diagnoses of viral haemorrhagic septicaemia (VHS) in farmed Atlantic Salmon. Values in parentheses are the numbers of farms on which farm-level audit-based diagnoses were made. The three farms licensed in fish health surveillance zone 3.3 (Althorpe, Hardwicke and Shaw Point) are grouped in zone 3.2 as per Aquaculture Management Division (AMD) practices. Source: data collected by the BC provincial government, 2010, and DFO Aquaculture Management Division (2011-2018 data were downloaded from the Open Canada website September 6th, 2019). 'N/A' indicates no audits, dashes indicate no VHS farm-level diagnosis.

Year	Fish Health Surveillance Zone and Sub-Zone									
	2.1	**2.2**	**2.3**	**2.4**	**3.1**	**3.2**	**3.3**	**3.4**	**3.5**	**Σ_{year}**
2002	-	N/A	-	-	-	-	-	-	N/A	-
2003	-	N/A	-	-	-	-	-	-	N/A	-
2004	N/A	N/A	3 (3)	1(1)	-	-	-	-	-	4 (4)
2005	N/A	N/A	4 (3)	-	-	-	-	-	-	4 (3)
2006	N/A	N/A	1 (1)	-	-	-	-	-	-	1 (1)
2007	N/A	N/A	1 (1)	-	-	-	1 (1)	-	-	2 (2)
2008	N/A	N/A	-	-	-	-	1 (1)	1 (1)	-	2 (2)
2009	N/A	N/A	2 (2)	-	-	-	1 (1)	1 (1)	-	4 (4)
2010	N/A	N/A	-	-	-	-	-	-	N/A	-
2011	N/A	N/A	-	-	-	-	-	-	-	-
2012	N/A	N/A	-	-	-	-	-	-	-	-
2013	N/A	N/A	-	-	-	-	-	-	-	-
2014	N/A	N/A	-	-	-	-	-	-	-	-
2015	N/A	N/A	-	-	-	-	-	-	-	-
2016	N/A	N/A	-	-	-	-	-	-	-	-
2017	N/A	N/A	-	-	-	-	-	-	-	-
2018	N/A	N/A	-	-	-	-	-	-	-	-
$\Sigma_{subzone}$	**0**	**N/A**	**11 (5)**	**1(1)**	**0**	**0**	**3 (2)**	**2 (2)**	**0**	**17 (10)**

Table 5. Number of Fish Health Audit and Surveillance Program (FHASP) molecular detections of viral haemorrhagic septicaemia virus (VHSV) in farmed Atlantic Salmon. The three farms licensed in fish health surveillance zone 3.3 (Althorpe, Hardwicke and Shaw Point) are grouped in zone 3.2 as per Aquaculture Management Division (AMD) practices. Source: data collected by the BC provincial government, 2010, and DFO Aquaculture Management Division (2011-2018 data were downloaded from the Open Canada website September 6th, 2019). 'N/A' indicates no audits, dashes indicate no VHSV detections via molecular testing.

Year	Fish Health Surveillance Zone and Sub-Zone									
	2.1	**2.2**	**2.3**	**2.4**	**3.1**	**3.2**	**3.3**	**3.4**	**3.5**	**Σ_{year}**
2002	-	N/A	-	-	-	-	-	-	N/A	0
2003	-	N/A	-	-	-	1	1	-	N/A	2
2004	N/A	N/A	2	1	-	-	-	-	-	3
2005	N/A	N/A	4	-	-	1	-	-	-	5
2006	N/A	N/A	4	-	-	-	-	-	-	4
2007	N/A	N/A	3	2	-	-	3	-	-	8
2008	N/A	N/A	-	1	-	1*	3	1	-	6
2009	N/A	N/A	3	-	-	-	3	2	-	8
2010	N/A	N/A	1	-	-	-	-	-	N/A	1
2011	N/A	N/A	-	-	-	-	1	-	-	1
2012	N/A	N/A	-	-	-	1	-	2	-	3
2013	N/A	N/A	-	-	-	-	-	-	-	0
2014	N/A	N/A	-	-	-	-	2	-	-	2
2015	N/A	N/A	-	-	-	-	1	-	-	1
2016	N/A	N/A	-	-	-	-	1	-	-	1
2017	N/A	N/A	-	-	-	-	1	-	-	1
2018	N/A	N/A	-	-	-	-	-	1	-	1
$\Sigma_{subzone}$	**0**	**N/A**	**15**	**4**	**0**	**4**	**16**	**6**	**0**	**47**

**Note this detection occurred on a farm that has not been in production since 2010 and therefore does not meet the criteria for an active farm for the purposes of the VHSV risk assessment.*

Pacific salmon

A total of 291 audits have been conducted on BC Pacific salmon farms between 2002 and 2018, screening over 1600 fish, resulting in three farm-level diagnosis of VHS. All three farm-level diagnoses were made in 2005, twice in zone 2.3 and once in zone 3.2. Over this same time period, FHASP molecular testing recorded six VHSV detections at six different farm sites within zones 2.3 and 3.2; five PCR detections at five sites in zone 2.3 and one in zone 3.2 (DFO, 2019b).

Fish Health Events

A fish health event (FHE) is "a suspected or active disease occurrence within an aquaculture facility that requires the involvement of a veterinarian and any measure that is intended to reduce or mitigate impact and risk that is associated with that occurrence or event" (DFO, 2015). Since 2002, as a condition of licensing (with the exception of 2013-2015), industry must report FHEs (Wade, 2017).

Atlantic Salmon

Between 2002-2019 (excluding 2013-2015), a total of 17 FHEs attributed to VHS have been reported by BC's Atlantic Salmon farming industry. Within the Discovery Islands area, a single FHE was reported in 2003 (Table 6) (DFO, 2019a).

Table 6. Number of Fish Health Events (FHE) (2002-2019) attributed to viral haemorrhagic septicaemia (VHS) in farmed Atlantic Salmon reported by industry to DFO. Values in parentheses are the numbers of farms on which a FHE was reported. The three farms licensed in fish health surveillance zone 3.3 (Althorpe, Hardwicke and Shaw Point) are grouped in zone 3.2 as per Aquaculture Management Division (AMD) practices. The 2016-2019 FHE data were downloaded from the Open Canada website on January 30th, 2020. 'N/A' indicates no requirement to report FHEs, dashes indicate no fish health events reported.

Year	Fish Health Surveillance Zone and Sub-Zone									
	2.1	**2.2**	**2.3**	**2.4**	**3.1**	**3.2**	**3.3**	**3.4**	**3.5**	**Σ_{year}**
2002	-	-	-	-	-	-	-	-	-	0
2003	-	-	1 (1)	-	-	1 (1)	-	-	-	2 (2)
2004	-	-	-	-	-	-	-	-	-	0
2005	-	-	4 (3)	-	-	-	-	-	-	4 (3)
2006	-	-	1 (1)	-	-	-	-	-	-	1 (1)
2007	-	-	-	-	-	-	1 (1)	-	-	1 (1)
2008	-	-	-	-	-	-	-	-	-	0
2009	-	-	3 (3)	-	-	-	-	-	-	3 (3)
2010	-	-	3 (2)	-	-	-	-	-	-	3 (2)
2011	-	-	-	-	-	-	-	-	-	0
2012	-	-	-	-	-	-	1 (1)	-	-	1 (1)
2013	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A
2014	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A
2015	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A
2016	-	-	-	1 (1)	-	-	1 (1)	-	-	2 (2)
2017	-	-	-	-	-	-	-	-	-	0
2018	-	-	-	-	-	-	-	-	-	0
2019	-	-	-	-	-	-	-	-	-	0
$\Sigma_{subzone}$	**0**	**0**	**12 (7)**	**1 (1)**	**0**	**1 (1)**	**3 (2)**	**0**	**0**	**17 (11)**

Pacific salmon

No FHEs have been attributed to VHS in Pacific salmon between 2002 and 2018 (excluding 2013-2015) (DFO, 2019a).

Mortality Events

DFO (2015) defines a mortality event as “a) fish mortalities equivalent to 4000 kg or more, or losses reaching 2% of the current facility inventory, within a 24 hour period; or (b) fish mortalities equivalent to 10,000 kg or more, or losses reaching 5%, within a five day period”. As a condition of license, any mortality event must be reported to DFO no later than 24 hours after discovery with details including facility name, fish cultured, number of dead fish, suspected proportion affected, suspected carcass biomass, probable cause, and action taken (DFO, 2015).

Atlantic Salmon

Between 2011 and 2019, one mortality event attributed to VHS has been reported in BC. The event occurred in March 2012 on an Atlantic Salmon farm located in zone 3.3 (DFO, 2020).

Pacific salmon

Between 2002 and 2019, there have been no mortality events attributed to VHS in Pacific salmon (DFO, 2020).

Wild Salmonids

Although VHSV-IVa is endemic in the BC marine environment, there have been no confirmed detections of VHSV in Sockeye Salmon. The presence of VHSV nucleic acid was suggested in three of 90 wild Sockeye Salmon (Dr. G. Marty, Ministry of Agriculture,1767 Angus Campbell Rd., Abbotsford, BC V3G 2M3, pers. comm., 2020); however, as there were uncertainties as to whether the specimens were indeed Sockeye Salmon and no follow-up testing was conducted to confirm VHSV infection, the results are insufficient to ascertain a natural transmission in Sockeye Salmon. Furthermore, across multiple independent surveys, VHSV was not detected in over 5000 Sockeye Salmon collected in British Columbia (Table 7). Overall, the detection of VHSV in Pacific salmon appears uncommon. Understanding that there are differences in methodologies for the various studies, face value detection prevalence of VHSV in over 10,000 wild Pacific salmon in BC demonstrated detections occurred in less than 1% of the samples tested, with only Coho and Chinook salmon testing positive (Table 7). It is important to note that gill tissue was included in the Fluidigm studies, therefore, it remains unknown as to whether the VHSV detections are representative of a systemic infection or whether the detections reflect an occurrence of VHSV on an exterior tissue as demonstrated in Gross et al. (2019).

Table 7. Viral haemorrhagic septicaemia virus (VHSV) detection prevalence in wild Pacific salmonids surveyed in British Columbia. Number in parentheses is detections per number screened. Results are reported at face value without consideration of differences in sampling or assay design. Test method is identified in parentheses under publication heading.

Species	Kent et al 1998 (Virus Isolation)	S. Johnson, unpublished (RT-qPCR; OIE)	Fluidigm studies* (Microfluidics qPCR using OIE primers/probe)	Overall
Sockeye Salmon	0% (0/436)	0% (0/1555)	0% (0/3474)	0 % (0/5465)
Chinook Salmon	0% (0/96)	-	0.52% (9/1744)	0.49% (9/1840)
Coho Salmon	0% (0/84)	-	0.68% (18/2656)	0.66% (18/2740)
Pink Salmon	0% (0/27)	-	-	0% (0/27)
Chum Salmon	0% (0/347)	-	-	0% (0/347)

**(Jeffries et al., 2014; Miller et al., 2014; Bass et al., 2017; Teffer et al., 2017; Nekouei et al., 2018; Teffer et al., 2018; Thakur et al., 2018; Tucker et al., 2018; Laurin et al., 2019; Nekouei et al., 2019; Thakur et al., 2019)*

FACTORS MODULATING VHSV-IVA DISEASE IN THE NORTHEASTERN PACIFIC OCEAN

In the northeastern Pacific Ocean, VHSV-IVa typically occurs at low prevalence and intensity. For instance, annual surveys of Pacific Herring collected from the marine waters of Prince William Sound Alaska rarely revealed VHSV detections despite using a standard 60 fish sample set (Kocan et al., 2001; Hershberger et al., 2015). Nevertheless, massive epizootic events have been reported to occur in this and other marine species in Alaska and BC. The driving factors of such mortality events are unclear, yet results from controlled laboratory and field surveillance studies particularly with Pacific Herring have provided some insights. Environmental factors, for instance, likely influence the potential for VHS outbreaks. Typically, die-off events of Pacific Sardine and Pacific Herring occur during the winter and early spring months in periods of colder ocean temperatures (Garver et al., 2013b). Furthermore, laboratory studies demonstrate that the susceptibility of Pacific Herring to VHS is inversely related to ambient seawater temperature, as evidenced by higher cumulative mortalities, greater viral shedding, and longer viral persistence in the tissues of survivors at cooler temperatures (Hershberger et al., 2013). Host factors are also likely pivotal in modulating VHS epizootics. In laboratory studies, Pacific Herring fed various commercially available pelleted feeds, differed in susceptibilities suggesting the importance of host nutritional status in subverting VHS disease (Beaulaurier et al., 2012). Another critical determinant of VHS potential is whether a host population has acquired resistance. Although naïve Pacific Herring are highly susceptible to VHS, survivors of the disease can develop a robust, long-lived immunity to the virus making them resistant to subsequent disease (Hershberger et al., 2015). Consequently, the susceptibility of Pacific Herring to VHS (and the potential for VHS epizootics) typically decreases with the host age, as older cohorts are more likely to have survived prior infection and developed acquired resistance. This acquired resistance supersedes all other disease co-factors, and a resistant population will not experience a VHS epizootic even if all other disease co-factors (e.g., exposure to high levels of the virus, cool temperatures, tight aggregations, etc.) are present (Kocan et al., 2001; Hershberger et al., 2013).

In the case of Pacific salmonid species, their inherent resistance to VHSV greatly limits infection and safeguards them against development of disease regardless of whether they are exposed to VHSV in freshwater or saltwater (Follett et al., 1997; Gross et al., 2019). Similarly, with Atlantic Salmon, although shown to be capable of contracting VHS disease experimentally, their reduced susceptibility to VHSV-IVa is evidenced by limited infections in BC marine farm populations despite sharing the environment with highly susceptible endemic species (see section “Shedding and transmission of VHSV-IVa in the northeastern Pacific Ocean”). Nevertheless, the sporadic occurrence of VHSV-IVa in farmed Atlantic Salmon has raised concern in regard to the potential for this endemic virus to evolve higher virulence towards salmon when present in a farm environment (Garver et al., 2013b). For instance, in Europe, VHSV-Ia which is highly virulent in farmed Rainbow Trout, is hypothesized to have evolved from a genotype I marine ancestor driven by the historical practices of feeding unpasteurized raw marine fish to the farmed Rainbow Trout (Meyers and Winton, 1995; Dixon et al., 1997; Dixon, 1999). In British Columbia, there is no empirical evidence to date to suggest VHSV-IVa adaptation or virulence changes are occurring in the salmon aquaculture industry. VHSV-IVa has been shown to remain relatively stable genetically since it was first detected in farmed salmon in the mid-1990’s (Garver et al., 2013b) and there has been no temporal trend towards increasing VHS diagnoses or VHS-associated fish health events with either farmed Atlantic Salmon or Chinook Salmon.

TRANSMISSION OF VHSV

Waterborne transmission is a natural route of VHSV infection with laboratory studies effectively transmitting virus through either bath exposure or via cohabitation with infected fish (Lovy et al., 2013). VHSV, in some instances, has been successfully transmitted via an oral route through intubation of infected feed (Schönherz et al., 2012) or ingestion of infected fish (Getchell et al., 2013), suggesting predation of VHSV-infected prey as a potential transmission route. Vector-mediated transmission has also been speculated given the occurrence of VHSV in some invertebrate species such as leeches (Faisal and Schulz, 2009; Faisal and Winters, 2011); however, given that direct horizontal transmission of VHSV is readily achieved, transmission via a multicellular parasite is unnecessary in spreading VHSV. Currently, there is no evidence to suggest a vector is required for VHSV transmission. Lastly, there are no indications or evidence of vertical transmission of VHSV (Bovo et al., 2005a).

SHEDDING AND TRANSMISSION OF VHSV-IVA IN THE NORTHEASTERN PACIFIC OCEAN

Pacific Herring and other forage species native to the northeastern Pacific Ocean are exceptionally susceptible to VHSV and upon infection shed copious amounts of VHSV into the water (Hershberger et al., 2010a). Laboratory studies with infected herring have demonstrated that shed VHSV can be detected in the water as early as 4–5 days post exposure (PE), prior to the onset of host mortality from the resulting disease. Viral shedding peaks at 6–10 days PE with each diseased herring shedding an average of 500 million plaque forming units (PFU) into the water each day (Hershberger et al., 2010a). Laboratory studies examining VHSV infections at 8ºC,11ºC and 15ºC further demonstrated that the progression of viral shedding is extremely temperature dependent, with lower temperatures generally resulting in higher shedding levels and delayed peaks in viral shedding (Hershberger et al., 2013). The capacity of Pacific Herring as super shedders has also been observed in wild populations where shed VHSV was in sufficient quantities to permit detection in open seawater areas surrounding wild free ranging and impounded spawning Pacific Herring (Hershberger et al., 1999). This inherent susceptibility of Pacific Herring and other forage species to VHSV and their capacity to amplify the virus in the natural environment makes them ideally suited as a natural reservoir and source of VHS virus. In fact, in cases where VHSV has occurred in marine net-pen salmon in BC, there has often been the co-occurrence of Pacific Herring or other forage species suggesting them as a source of VHSV to the farmed salmon. Whether drawn to the net-pen facilities as a refuge from predators (e.g., sea lions and seals) or for food, forage fish with the potential to harbour VHSV have been observed in or around the net-pen area. Consequently, it is likely that if VHSV susceptible forage fish succumb to VHS disease while in the area of a salmon farm, the farmed salmon would be subjected to high concentrations of shed virus. Genetic typing of VHSV isolates obtained from Pacific Herring and Pacific Sardine sampled around net pen salmon farms often identified an exact match to the virus type that was subsequently detected in the net-pen farmed salmon (Garver et al., 2013b). The mode of viral transmission between the forage fish and farmed salmon is likely through waterborne exposure as laboratory studies have successfully infected Atlantic Salmon with VHSV through cohabitation with VHSV infected Pacific Herring (Lovy et al., 2013; Gross et al., 2019). VHSV transmission to farmed salmon could also potentially occur orally via the ingestion of VHSV infected Pacific Herring, however, this route of transmission is unlikely as predation rates of 1.1% or less have been reported in farmed salmon (Johannes and Hay, 2006) (K. Shaw, DFO, 1520 Tamarac St., Campbell River, BC V9W 3M5, pers. comm., 2020). Nevertheless, in free range Pacific salmonid and Pacific Hake populations where Pacific Herring and other forage species are a primary food source, ingestion exposure to virus infected prey could be a likely source of VHSV periodically detected in wild fish populations (Meyers and Winton, 1995).

In instances where farmed Atlantic Salmon become infected with VHSV, the duration and extent of virus shedding from infected individuals is unknown. A laboratory study transmitting VHSV from Atlantic Salmon to Pacific Herring demonstrated that VHSV-infected Atlantic Salmon are capable of shedding virus and infecting a highly susceptible species like Pacific Herring (Lovy et al., 2013). It is worthy to note that successful transmission of VHSV from Atlantic Salmon to Pacific Herring in laboratory trials has been inconsistent and likely reflects the limited susceptibility of Atlantic Salmon to VHSV. For instance, in the Lovy et al. (2013) study, VHSV was not transmitted from Atlantic Salmon to Pacific Herring across all replicate tanks and detectable levels of VHSV remained absent from water samples collected from tanks containing only VHSV-exposed Atlantic Salmon. Similarly, Gross et al. (2019) failed to recover VHSV from Atlantic Salmon subsampled over the course of a waterborne VHSV challenge and Traxler et al. (1999) was only able to isolate VHSV from 1/6 Atlantic Salmon survivors 25 days post VHSV-injection and did not recover VHSV from the bath-exposed Atlantic Salmon survivors.

MINIMUM INFECTIOUS DOSE

The virus exposure level required to establish infection is dependent upon host susceptibility. Pacific Herring are extremely susceptible to VHSV-IVa and can become diseased when exposed to extremely low levels of VHSV. Laboratory studies which immersed Pacific Herring for 24 hours in seawater with 10 PFU/mL of VHSV (a level typically undetected using standard laboratory detection methods), initiated infection among the exposed population and caused upwards of 100% mortality (Hershberger et al., 2010a). In contrast, Sockeye Salmon exposed to 670 times higher VHSV levels, remained free from virus infection revealing their inherent resistance to the virus and improbability of receiving an infectious dose within a natural environment (Gross et al., 2019). VHSV infection of Sockeye Salmon was only achieved through the artificial and invasive procedure of injecting virus (3.9×10^3 plaque-forming units [PFU] per fish) intraperitoneal into fish (Gross et al., 2019). Likewise, Chinook, Coho, and Pink salmon were largely resistant to infection when freshwater exposed to high levels of VHSV ($>10^3$ PFU/mL), revealing their refractory nature to VHSV and likely accounting for the rare occurrence of VHSV in free ranging Pacific salmon populations (Winton et al., 1991; Follett et al., 1997).

VIRUS DECAY AND INACTIVATION METHODS

Laboratory studies have demonstrated that VHSV shed from an infected host can remain infectious in either marine or freshwater environments, yet the duration at which the virus remains infectious is highly dependent upon environmental parameters such as water salinity, temperature, organic load, microbial content, and exposure to ultraviolet light. In a controlled laboratory study whereby decay of VHSV genotype IVa was measured in freshwater and seawater held in the dark at various temperatures, the virus remained infectious longer in freshwater than in seawater with 99.9% of VHSV rendered inactive by day three in seawater at 15°C versus day 11 in freshwater (Hawley and Garver, 2008) . These viral decay times are contingent upon temperature with a duplicate experiment performed 5°C colder prolonged virus inactivation by three and eight days, in seawater and freshwater, respectively, revealing that virus stability is inversely proportional to temperature (Hawley and Garver, 2008). The stability of VHSV in seawater has also been shown to increase significantly with aqueous protein concentrations (Kocan et al., 2001) suggesting an increased probability of virus transmission around fish spawning aggregations. Laboratory studies have also demonstrated that VHSV can be retained on fomites such as plastic pieces, glass, fishing hooks and fishing lines yet the duration by which VHSV remains infectious on the objects was dependent upon storage condition and material type (Pham et al., 2012). When plastic, glass, and aluminum objects stored in the dark under moist conditions were seeded with high concentrations of VHSV (10^9

tissue culture infectious dose per mL ($TCID_{50}$/mL), infectious virus could be retained for at least 10 days. However, when objects were stored under dry conditions, infectious virus was not recovered past the first sampling day (Pham et al., 2012). Additionally, rusted metal also proved to have low virus retention capacity with virus inactivation likely expedited by the iron oxides present from the rusting process (Pham et al., 2012) (Pham et al., 2012). VHSV is also sensitive to UV light (Øye and Rimstad, 2001) and, given its morphological resemblance and genomic similarities to infectious hematopoietic necrosis virus (IHNV), it would be expected that VHSV would display equally rapid inactivation when exposed to sunlight as demonstrated for IHNV (Garver et al., 2013a).

In addition to natural inactivation methods, physical chemicals have been shown to be efficacious at rendering VHSV inactive (Bovo et al., 2005b; Bowker et al., 2016). Some of the common chemicals used in the aquaculture industry as disinfectants include Virkon® Aquatic, Peroxigard™, chlorine compounds and hydrogen peroxide. These compounds effectively inactivate VHSV when it is exposed to the chemical at the prescribed concentration for the required contact time. For example, Virkon® Aquatic, the most commonly used disinfectant in the BC aquaculture industry, inactivates VHSV at a concentration of 0.5-1% with a contact time of 10 minutes (Bowker et al., 2016).

HEALTH MANAGEMENT

VACCINES

In laboratory studies, various vaccine types, including DNA vaccines, have been shown to be effective at protecting fish against VHS disease (Lorenzen et al., 1999; Lorenzen et al., 2000); however, to date there are no commercially available vaccines against VHSV (Kim and Kim, 2019).

DISEASE CONTROL IN ENZOOTIC AREAS

Detection of VHSV in farmed salmon in BC initiates industry fish health management actions aimed at reducing disease as described in Wade (2017) which can include, but is not limited to, changes in the visitation order of farms with VHS positive farms being the last farm visited as well as implementation of low stress protocols such as reducing handling and feeding at a site.

Moreover, the Canadian Food Inspection Agency (CFIA) has a response plan for VHS (Dr. K. Klotins, CFIA, 59 Camelot Dr., Ottawa, ON K1A 0Y9, pers. comm., 2020). The CFIA has zoned Canada for the presence and absence of VHS in susceptible species of finfish. The zones identified as Pacific Ocean, Pacific Ocean Watershed of British Columbia and Pacific Ocean Watershed of Yukon are declared as infected areas for VHS genotype IVa as well as specific zones on the east coast of Canada (accessed October 6, 2020).

The goal of CFIA's disease response policy for responding to VHS IVa notifications in the Pacific Ocean and Pacific Ocean Watershed of BC is to contain the disease to these zones; eradication of the disease will be considered under certain circumstances. The following activities are carried out where possible regardless of the whether notifications originate from private or public premises.

Upon notification of a suspicion of VHS, the CFIA will:

- Confirm the disease and publish the confirmation on the CFIA web site and report to the OIE.

- Identify if the epidemiology of the disease has changed, for example, infection in a new species or a change in the expected clinical picture. Changes in epidemiology are published on the CFIA web site and reported to the OIE.
- Identify if any movements of infected animals, virus contaminated water and/or fomites occurred out of the infected area from the time when disease was likely introduced into the affected facility until the CFIA was notified.
- Identify and control any planned movements of animals or things from the affected facility that are not covered by the domestic movement control program.
- Identify if surveillance for potential spread of VHS-IVa is required outside of the infected area because the notification was from a wild finfish population or because of a high-risk movement of affected cultured finfish.
- Determine if eradication of a cultured population is required, for example, because of a significant impact on wild fisheries.
- Provide advice on disease containment and elimination on any disease response procedures that may be undertaken by industry or government, such as disposal of affected finfish and cleaning and disinfection, if requested.

OIE CRITERIA FOR DEFINING A SUSCEPTIBLE SPECIES: ASSESSMENT OF SOCKEYE SALMON SUSCEPTIBILITY TO VHSV

Based upon criteria established by the OIE, a species of aquatic animal is considered susceptible to infection with a pathogenic agent, when each of the following criteria are met:

1. Transmission has been obtained naturally or by experimental procedures that mimic natural pathways for the infection;
2. The identity of the pathogenic agent has been confirmed in accordance with OIE diagnostic criteria or equivalent; and
3. Evidence indicates that presence of pathogenic agent constitutes an infection (OIE, 2019).

Therefore, in assessing whether Sockeye Salmon are susceptible to VHSV, it is clear that this species does not fulfill these criteria as there is no confirmed detection of VHSV in wild fish to reveal evidence of a natural transmission nor have experimental exposures that simulated natural infection pathways been successful at generating infections in this species.

SUMMARY

Viral haemorrhagic septicaemia virus is endemic in the northeastern Pacific Ocean where it can cause significant disease in numerous marine forage fish species such as Pacific Herring and Pacific Sardine. Yet despite the virus' high virulence in marine forage fish species, it is of low to moderate virulence in Atlantic Salmon and low virulence in Pacific salmon species. Given that the marine forage fish species can be found in and around marine net-pen farmed salmon and have the capacity to shed enormous quantities of VHSV, they have been shown to be a natural source of VHSV to the marine reared farmed salmon. The frequency of VHSV detections in farmed Atlantic Salmon, as reported through industry fish health events or government surveillance programs, varies across regions but overall is relatively irregular with a total of five detections made over an eighteen-year period in the Discovery Islands area (zone 3.2).

In Pacific salmon, the occurrence of VHSV is equally sporadic and is undoubtedly a reflection of their refractory nature to VHSV infection as measured in controlled laboratory studies.

Specifically, in Sockeye Salmon, no confirmed detection of VHSV has occurred in over 5000 fish tested. Furthermore, laboratory studies have demonstrated that Sockeye Salmon remain free from VHSV infection despite exposures to high concentrations of virus that are known to be lethal to Pacific Herring. Consequently, without evidence of either natural or non-invasive experimental infections with VHSV, Sockeye Salmon do not fulfill the criteria of a VHSV susceptible species as defined by the OIE.

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