

Annual report on health monitoring of wild anadromous salmonids in Norway 2017

– *Health monitoring of returning adult salmon from river
Etne, western Norway*

Abdullah S. Madhun, Øystein Skaala, Rune Nilsen, Rosa Maria Serra-Llinares, Kristine Marit Schrøder Elvik, Pål Arne Bjørn, Ørjan Karlsen, Terje Svåsand and Egil Karlsbakk



Project Report

Report: RAPPORT FRA HAVFORSKNINGEN **No. – Year:** 26-2018 **Date:** 27.06.2018

Title:
Annual report on health monitoring of wild anadromous salmonids in Norway 2017

Authors:
Abdullah S. Madhun, Øystein Skaala, Rune Nilsen, Rosa Maria Serra-Llinares, Kristine Marit Schrøder Elvik, Pål Arne Bjørn, Ørjan Karlsen, Terje Svåsand and Egil Karlsbakk

Distribution:
Open

Project no.:
14986-03

Assignor(s):
Norwegian Food Safety Authority

Program:
Aquaculture

Research group:
Disease and Pathogen
Transmission

Number of pages in total:
13

Photo front page:
Espen Bierud / IMR

Summary (Norwegian):

Svært få funn av virus i tilbakevandrende laks selv om de samme virusene er vanlige i oppdrett

Havforskningsinstituttet har undersøkt forekomsten av flere virus som er prevalent i oppdrett i tilbakevandrende laks fra Etneelva 2015-2016. Laksen stammer fra kultivert smolt som ble snotemerket og satt ut i Etne 2013 og 2014. Forekomsten av SAV-, PRV-, ILAV- og PMCV-infeksjoner ble testet ved å bruke sanntids RT-PCR metoden. Disse virusene kan forårsake sykdommene pankreassykdom (PD) og hjerte- og skjelettmuskelbetennelse (HSMB), Infeksiøs lakseanemi og kardiomyopatisyndrom (CMS, hjertesprekk) hos oppdrettslaks. SAV ble ikke påvist i den tilbakevandrende laksen. Derimot ble PRV1-, ILAV- og PMCV-infeksjoner påvist hhv. i 5 %, 1% og 3% av fisken. Infeksjoner med disse virusene er vanlig i oppdrett. Disse resultatene indikerer at villaksen ikke blir smittet i særlig grad, selv om de kan ha blitt utsatt for smitte fra oppdrett når de vandret ut eller på veien tilbake til elven. Samtidig kan vi ikke utelukke at syk villfisk ikke blir fanget fordi den er svekket og ikke går opp i elv, eller at den dør.

Summary (English):

Very low occurrence of virus infections in returning salmon although the same viruses are common in fish farming.

The Institute of Marine Research has investigated the occurrence of several viruses that are prevalent in salmon farming in returning salmon from River Etne during the period 2015-2016. The salmon is originated from cultivated smolt, which was tagged and released in Etne in 2013 and 2014. The prevalence of SAV, PRV1, ISAV and PMCV infections was tested using real-time RT-PCR method. These viruses may cause pancreas disease (PD), heart and skeletal muscle inflammation (HSMI), infectious salmon anaemia and cardiomyopathy syndrome (CMS) in farmed salmon. SAV was not detected in any of the returning salmon. On the other hand, PRV1, ISAV and PMCV infections were detected in 5%, 1% and 3% of the fish, respectively. Infections with these viruses are common in salmon farming in Norway. These results show low prevalence of virus infections in returning adult salmon, although fish may have been exposed to viruses from fish farming. At the same time, we cannot exclude that diseased wild salmon is difficult to catch because it is either weakened and does not ascend river or it dies.

Geir Lasse Taranger

Research Director

Terje Svåsand

Head of Research Program



Since 2012, the Norwegian Veterinary Institute (NVI) and the Institute of Marine Research (IMR) have been commissioned by the Norwegian Food Safety Authority to carry out an annual health monitoring of wild anadromous salmonids in Norway.

Content

- 1 Introduction5
- 2 Aim6
- 3 Materials and methods.....7
- 4 Results8
- 5 Discussion and Conclusion.....9
- 6 References.....11

1 Introduction

Viral diseases represent a serious problem in Atlantic salmon (*Salmo salar* L.) farming in Norway, often leading to substantial economic losses (Table 1).

Pancreas disease (PD), caused by salmonid alphavirus (SAV), is a major health problem for fish farming in Norway with 99–176 annual cases in the last 5 years [1]. Two subtypes of SAV occur in Norway, SAV3 and the more recently detected SAV2 [2]. Most of the disease outbreaks due to SAV3 occur in the western part of the country, especially in Hordaland county, while SAV2 cases are mostly restricted to an area along Mid-Norway.

Infectious salmon anaemia (ISA) was a major problem in Norwegian aquaculture in the late 1980s. In the last 5 years, the annual number of cases has varied between 10 and 15. ISA virus (ISAV) may be classified based in the sequence of HE gene as HPR0 (avirulent) and HPRΔ (virulent) variants. HPR0 variants are highly prevalent in farmed Atlantic salmon [3, 4], causing non-clinical transient infections (2–4 months). There are increasing evidence that virulent HPRΔ variants originate from different HPR0 types [5]. There is very limited data about the prevalence of ISAV HPR0 in wild salmon in Norway.

Heart and skeletal muscle inflammation (HSMI) is another disease that is caused by a piscine orthoreovirus (PRV-1). High PRV-1 viral loads are found in fish developing HSMI, but may also occur in healthy fish. The disease is an increasing problem in fish farming in Norway with 101–181 annual registered cases of HSMI in the period 2013–2017 [1].

Cardiomyopathy syndrome (CMS) is a growing problem in Norwegian salmon farming which is caused by piscine myocarditis virus. It has been registered between 90 and 107 CMS outbreaks in salmon farming in the last 5 years [1].

Table 1: The number of registered viral disease outbreaks in fish farming in the last 5 years [1].

	2013	2014	2015	2016	2017
PD	99	142	137	138	176
ISA	10	10	15	12	14
HSMB	134	181	135*	101*	93*
IPN	56	48	30*	27*	23*
CMS	100	107	105*	90*	100*

* Underreported.

Disease outbreaks in salmon farms may lead to increased infection and disease risks at neighbouring farms and in wild fish populations [6]. There is increasing public concern of this impacting wild salmonids in Norway. There is limited data on the prevalence of pathogens in wild salmonid populations, but in particular it is difficult to quantify disease incidence and impact in wild fish since sick individuals may be less catchable or may disappear unnoticed (e.g. due to predation). Therefore, it is challenging to evaluate the impact of disease in wild stocks since we normally are only able to collect infected but non-diseased fish such as individuals that has recently acquired or has survived an infection (carriers). There are evidences for pathogen transmission from farmed to wild fish [7–10]. However, the frequency and the consequence of transmission of many viral disease agents are largely unknown.

Wild salmon may be infected by viruses prevalent in salmon farming; in rivers as parr by virus-infected farm escapees and spawning wild salmon or from salmon farms in the fjord when migrating as smolt or returning as adults.

2 Aim

The aim of the current study was to investigate the occurrence (prevalence) of SAV, ISAV, PRV and PMCV infections in returning adult Atlantic salmon, which previously tagged and released as hatchery-reared smolt, recaptured in the river Etneelva, western Norway.

3 Materials and methods

The Institute of Marine Research had a smolt-release project in the river Etneelva to evaluate the effect of salmon lice infestation on the migrating smolt. In 2013 and 2014, approximately 15 000/year of hatchery-reared smolts were adipose fin clipped, tagged and released at the mouth of the river.

Results from 364 adult salmon recaptured at the Etneelva salmon trap in 2015-2016 were used in the current report. The fish weight, length, age, sex and time of capture were determined. The heads of fish were cut posterior to the pericardial cavity and were frozen (-20 °C) in the same day of capture. At autopsy, tissues from the heart were aseptically taken out from the fish while still frozen and transferred to tubes on dry ice. Heart samples were sent on dry ice to accredited commercial laboratories for RNA extraction and virus testing. Analyses for SAV, ISAV, PRV and PMCV viruses (for detection viral RNA) were performed by these laboratories using their in-house real-time PCR assays. Samples with C_t (cycle-threshold) value below 40 are reported as positive.

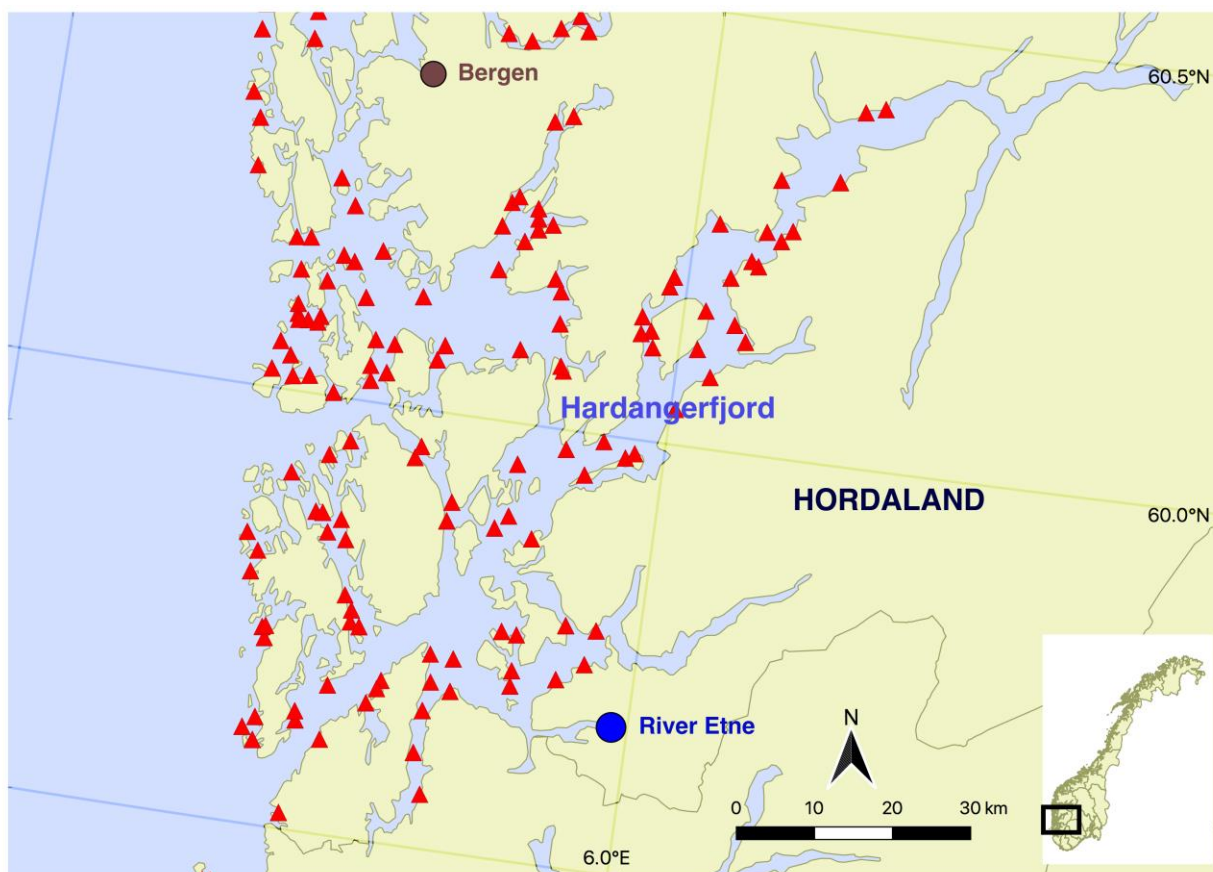


Figure 1: A map showing the location of the river Etneelva and fish farms (red triangle) in the area around.

4 Results

Salmon

We tested 364 returning adult Atlantic salmon recaptured in 2015-2016 at the salmon trap installed in the river Etneelva. Of the fish tested, 228 were released in 2013, 118 in 2014 and 20 had unknown origin.

SAV was not detected.

SAV was not detected in any of the hearts from the tested salmon (Table 2).

The prevalence of ISAV

ISAV was detected in 1% (N=5) of the fish. Most (N=4) of the ISAV-positive fish were released in 2013. Type of virus (ISAV-HPR0 versus ISAV-HPRΔ) could not be determined due to low virus concentration (Ct values; 34.3-36.0) in the positive fish.

The prevalence of PRV

PRV was detected in 5% of the returning adult salmon. PRV prevalence was 7% and 3% in fish from the 2013 and 2014 smolt release respectively. Infected fish had low to moderate viral load (Ct values; 26.9-37.4).

The prevalence of PMCV

PMCV was detected in 3% of the returning salmon. PMCV prevalence was 2% in salmon released in 2013 and 4% in fish released in 2014. Infected salmon had very low viral load (Ct values; 34.5-36.0).

Table 1: The numbers of tested and percentages of virus-positive returning adult salmon captured at salmon trap in River Etne.

Release year	No.	No of virus-positive fish (%)			
		SAV	ISAV	PRV*	PMCV
2013	228	0	4 (2)	6 (7)	5 (2)
2014	118	0	1 (1)	3 (3)	5 (4)
Unknown	18	0	0	0	1 (6)
Total	364	0	5 (1)	9 (5)	11 (3)

* Total number of PRV-tested fish is 184 (85 from 2013 release, 91 from 2014 release and 8 from unknown origin).

5 Discussion and Conclusion

SAV3 is endemic in the Hardangerfjord where fish in the majority of salmon farms become infected during the production cycle [11, 12]. PD outbreaks in the fjord are common in May-June [1]. It is likely that smolt were exposed to virus released from farms with subclinical infections and PD outbreaks during their migration (May-June). However, we could not detect SAV in any of the tested returning adult salmon. The concentration of virus released from farms with clinical PD outbreaks or with subclinically infected salmon is currently unknown. A recent report has shown that a small number of smolts (3 of 24) that were exposed experimentally to low concentration of SAV3 in water was virus-positive in heart samples 1-3 weeks after exposure [13]. The absence of SAV infection in the tested salmon is consistent with previous findings that SAV infections are uncommon in wild salmonids irrespective of farming intensity or the frequency of PD outbreaks at the locations examined [14-18]. These observations could indicate that wild salmon are exposed to a low infection pressure from fish farming. However, the possibility that SAV infection may lead to rapid disappearance (predation) of the infected wild fish cannot at present be completely ruled out.

In contrast to SAV, ISAV (1%), PRV (5%) and PMCV (3%) infections were detected in the tested salmon at low prevalence. The infection intensities measured by were generally low (Ct-values >30) and may indicate a carrier status.

There were no registered ISA outbreaks in the Hardangerfjord in the study period. Therefore, it is unlikely that the tested wild salmon were exposed to virulent ISA-HPR Δ from fish farms. On the other hand, ISAV-HPR0 infection is common in farmed Atlantic salmon [3, 4]. Low concentrations ISAV were detected in 1% (N=5) of the tested fish. We were not able to sequence segment 6 of ISAV strains from positive salmon to determine if the virus is HPR0 or HPR Δ . However, it is likely that the viruses detected in the positive-fish are avirulent strains (ISAV-HPR0). We have previously detected ISAV-HPR0 at low prevalence in wild and escaped salmon captured in northern Norway and in Hardangerfjord [19].

There is no available data about HSMI outbreaks in fish farms located in the area during the 2013-2014 period. However, PRV infection is very abundant in fish farming in Norway [20]. Garseth et al. [8] have suggested that extensive transmission of PRV from fish farms to wild salmon has occurred. However, our recent paper suggests that PRV transmission among salmon may also occur in oceanic feeding area [18]. The impact of PRV infections on the fitness and mortality of wild salmon populations is currently unknown, although the ability of mature salmon to ascent rivers may possibly be affected [21].

CMS is a growing problem in salmon farming in Norway [22]. Very low prevalence (only 2 out of 797 fish) of PMCV was detected in wild salmon collected from 35 different rivers (including river Etneelva) in Norway 2007-2009 [23]. Our current results show a significantly higher prevalence (10 out of 346, $p < 0.001$) of PMCV in returning salmon from Etneelva compared to the previous report. Garseth et al. (2012) have tested 18 salmon from river Etne, 53 from Hardangerfjord and 79 from Hordaland and found to be PMCV-negative. Additionally, we recently found PMCV infections in wild parr in rivers (prevalence 0-32%), brood stock salmon (0-6%) and escaped farmed fish (0-19%) [19]. These observations indicate a higher prevalence of PMCV in wild salmon populations than previously reported and may reflect a recent increase in occurrence of infection.

The results from the current report may suggest low viral transmission from fish farming to hatchery-reared salmon originating from Etneelva. Similar results were also reported in migrating wild smolt from the same area [19]. These observations could be explained by a short exposure time of salmon from river Etneelva, which is located in the outer part of Hardangerfjord, to viruses released from fish farms.

A time series of virus prevalence in parr, migrating smolt and returning adult salmon are necessary to evaluate the effect of infection pressure from salmon farming on the virus prevalence in wild salmon populations and to monitor changes over time.

6 References

1. Hjeltnes, B., et al. (eds), *Fish health report 2017 (in Norwegian)*. 2018, Norwegian Veterinary Institute. p. 108.
2. Hjortaa, M.J., et al., *The first detections of subtype 2-related salmonid alphavirus (SAV2) in Atlantic salmon, *Salmo salar* L., in Norway*. *Journal of Fish Diseases*, 2013. **36**: p. 71-74.
3. Lyngstad, T.M., et al., *Low virulent infectious salmon anaemia virus (ISAV-HPR0) is prevalent and geographically structured in Norwegian salmon farming*. *Diseases of Aquatic Organisms*, 2012. **101**: p. 197-206.
4. Christiansen, D.H., et al., *A low-pathogenic variant of infectious salmon anemia virus (ISAV-HPR0) is highly prevalent and causes a non-clinical transient infection in farmed Atlantic salmon (*Salmo salar* L.) in the Faroe Islands*. *Journal of General Virology*, 2011. **92**: p. 909-918.
5. Christiansen, D.H., et al., *First field evidence of the evolution from a non-virulent HPR0 to a virulent HPR-deleted infectious salmon anaemia virus*. *Journal of General Virology*, 2017. **98**: p. 595-606.
6. Taranger, G.L., et al., *Risk assessment of the environmental impact of Norwegian Atlantic salmon farming*. *ICES Journal of Marine Science*, 2015. **72**: p. 997-1021.
7. Costello, M.J., *How sea lice from salmon farms may cause wild salmonid declines in Europe and North America and be a threat to fishes elsewhere*. *Proceedings of the Royal Society B-Biological Sciences*, 2009. **276**: p. 3385-3394.
8. Garseth, Å.H., T. Ekrem, and E. Biering, *Phylogenetic evidence of long distance dispersal and transmission of piscine reovirus (PRV) between farmed and wild Atlantic salmon*. *PLOS One*, 2013. **8**: p. e82202.
9. Johansen, L.H., et al., *Disease interaction and pathogens exchange between wild and farmed fish populations with special reference to Norway*. *Aquaculture*, 2011. **315**: p. 167-186.
10. Johnsen, B.O., P.I. Møkkelgjerd, and A.J. Jensen, *Furunkulose i norske vassdrag-Statusrapport*, in *NINA Forskningsrapport*. 1993, NINA. p. 1-73.
11. Jansen, M.D., et al., *Pancreas disease (PD) in sea-reared Atlantic salmon, *Salmo salar* L., in Norway; a prospective, longitudinal study of disease development and agreement between diagnostic test results*. *Journal of Fish Diseases*, 2010. **33**: p. 723-736.
12. Jansen, M.D., et al., *Salmonid alphavirus (SAV) and pancreas disease (PD) in Atlantic salmon, *Salmo salar* L., in freshwater and seawater sites in Norway from 2006 to 2008*. *Journal of Fish Diseases*, 2010. **33**: p. 705-705.
13. Jarungsriapisit, J., et al., *Relationship between viral dose and outcome of infection in Atlantic salmon, *Salmo salar* L., post-smolts bath-challenged with salmonid alphavirus subtype 3*. *Veterinary Research*, 2016. **47**.
14. Biering, E., et al., *Annual report on health monitoring of wild anadromous salmonids in Norway 2012*. 2013: Norwegian Veterinary Institute and Institute of Marine Research: www.imr.no www.vetinst.no. p. 13.
15. Plarre, H. and A. Nylund, *Project Report: "The occurrence of SAV 2 in wild salmonids in mid-Norway" (in Norwegian)*. 2014, University of Bergen: University of Bergen. p. 25.
16. Garseth, Å.H., et al., *Annual report on health monitoring of wild anadromous salmonids in Norway 2014*. 2015: Norwegian Veterinary Institute and Institute of Marine Research: www.imr.no www.vetinst.no. p. 14.
17. Madhun, A.S., et al., *Occurrence of salmonid alphavirus (SAV) and piscine orthoreovirus (PRV) infections in wild sea trout *Salmo trutta* in Norway*. *Diseases of Aquatic Organisms*, 2016. **120**: p. 109-13.

-
18. Madhun, A.S., et al., *Prevalence of piscine orthoreovirus and salmonid alphavirus in sea-caught returning adult Atlantic salmon (Salmo salar L.) in northern Norway*. J Fish Dis, 2018. **41**: p. 797-803.
 19. Grefsrud, E.S., et al., *Risk assessment of Norwegian fish farming (in Norwegian)*. 2018: Institute of Marine Research: www.imr.no. p. 181.
 20. Garseth, Å.H., et al., *Piscine reovirus (PRV) in wild Atlantic salmon, Salmo salar L., and sea-trout, Salmo trutta L., in Norway*. Journal of Fish Diseases, 2013. **36**: p. 483-493.
 21. Miller, K.M., et al., *Infectious disease, shifting climates, and opportunistic predators: cumulative factors potentially impacting wild salmon declines*. Evolutionary Applications, 2014. **7**: p. 812-855.
 22. Garseth, A.H., et al., *Cardiomyopathy syndrome in Atlantic salmon Salmo salar L.: A review of the current state of knowledge*. Journal of Fish Diseases, 2018. **41**: p. 11-26.
 23. Garseth, Å.H., E. Biering, and T. Tengs, *Piscine myocarditis virus (PMCV) in wild Atlantic salmon Salmo salar*. Diseases of Aquatic Organisms, 2012. **102**: p. 157-161.

Retur: Havforskningsinstituttet, Postboks 1870 Nordnes, NO-5817 Bergen

HAVFORSKNINGSINSTITUTTET
Institute of Marine Research

Nordnesgaten 50 – Postboks 1870 Nordnes
NO-5817 Bergen
Tlf.: +47 55 23 85 00
E-post: post@hi.no

www.hi.no

