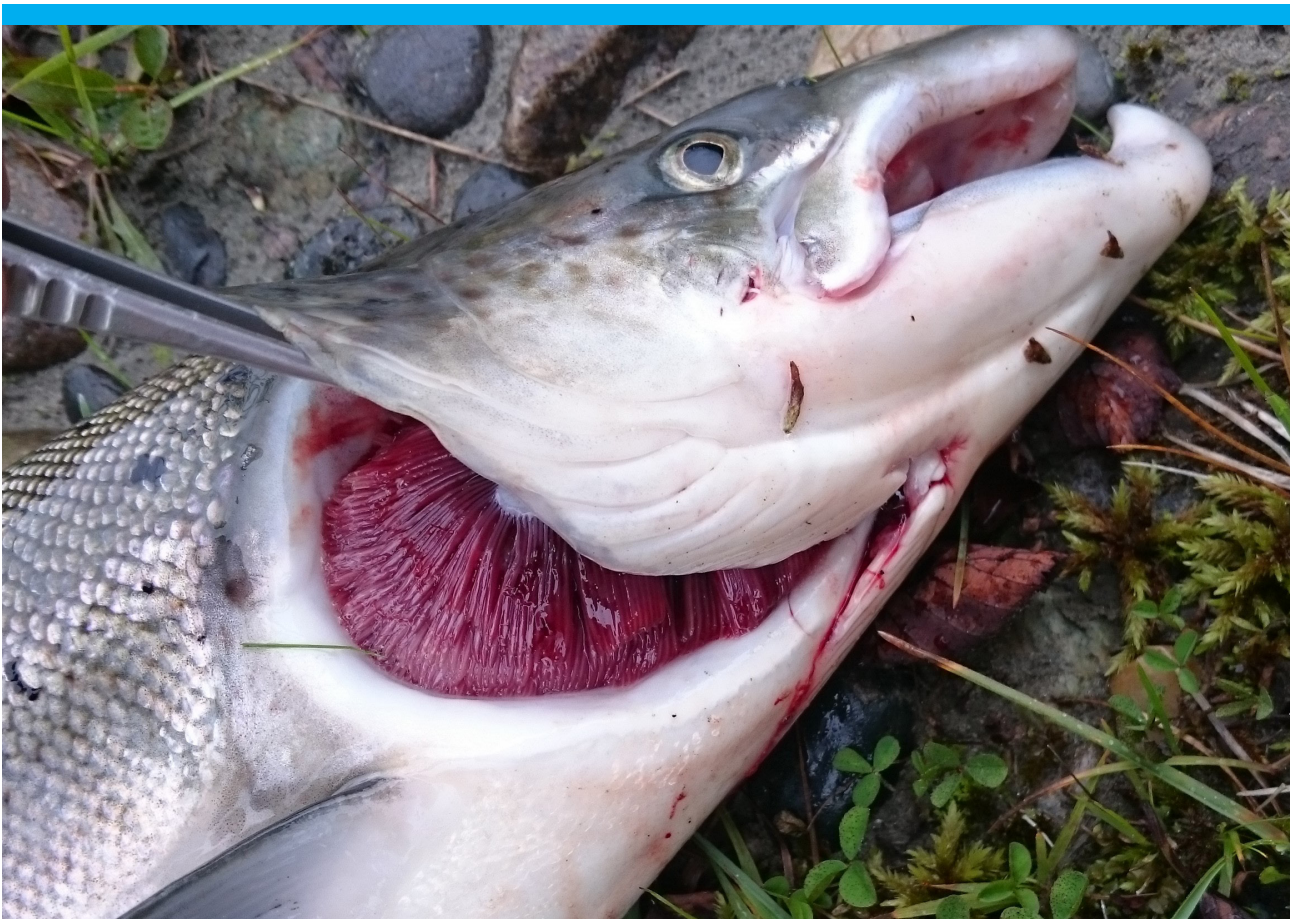


Health monitoring of wild anadromous salmonids in freshwater in Norway 2018



Veterinærinstituttet
Norwegian Veterinary Institute



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Introduction

Gill disease is a significant problem in Atlantic salmon farming in Norway, both in production of juveniles on land and in on-growing sites in the sea [1]. A number of infectious agents are associated with gill disease in farmed salmon and rainbow trout, and our knowledge is continuously increasing.

Serious recurring gill diseases in Atlantic salmon in seawater is often multifactorial, and referred to as proliferative gill inflammation (PGI) [2]. Microscopic cysts in the gill epithelium, also called epitheliocystis, is a common finding by histopathological examination. Candidatus *Branchiomonas cysticola* has been identified as the main contributor to epitheliocystis in the gills in both Norway and Ireland, but is also common in gills from unaffected salmon [3]. Candidatus *Piscichlamydia salmonis* is a bacteria also found in association with epitheliocystis [4].

The microsporidian *Paranucleospora theridion* (also known as *Desmozoon lepeophtherii*) is found in gills of clinically healthy fish, but with higher loads in fish with PGI [5, 6]. When discovered, the microsporidian was first associated with sea lice [7]. *Paranucleospora theridion* is suggested to be the primary agent in cases with PGI in the late autumn and winter in Western Norway [6].

In 2005 Atlantic salmon paramyxovirus (ASPV) was found to be a contributor to proliferative gill inflammation [2], but it could not reproduce disease in an infection trial alone [8].

Salmon gill poxvirus (SGPV) is present at all stages of the production cycle in farmed salmon, and it is associated with disease and mortality in both juvenile salmon in freshwater and in adult fish after seatransfer [9]. Garseth et al. reported in 2017 that SGPV was widely distributed in wild Atlantic salmon returning from marine migration [10]. They also found characteristic gill lesions, including apoptosis.

Amoebic gill disease (AGD) is caused by the protozoan parasite *Paramoeba perurans* in the marine environment, and was first reported in Norway in 2008 [11]. The disease has been reported in several farmed species around the world, and is known to cause significant losses [12].

Aim

In 2018, the objective of the health monitoring program in freshwater was to investigate the occurrence of Atlantic salmon paramyxovirus (ASPV), Candidatus *Branchiomonas cysticola*, *Paranucleospora theridion* (*Desmozoon lepeophtherii*), Candidatus *Piscichlamydia salmonis*, Salmon gill poxvirus (SGPV) and *Paramoeba perurans* in wild salmonids.

Materials and methods

The study sample comprised four species of wild salmonids from different parts of Norway. Wild caught brood fish of Atlantic salmon (*Salmo salar* L.), sea trout (*Salmo trutta* L.) and Atlantic char (*Salvelinus alpinus* L.) and non-anadromous salmon were obtained from stock enhancement hatcheries and the Genebank for wild Atlantic salmon. Fish from the same river were held together in tanks from a few days up to 6-7 weeks before stripping and tissue sampling.

Samples from sea trout and Atlantic salmon at sea were obtained from shore-based coastal fisheries and in three different locations. Additional samples of Atlantic salmon were obtained from six rivers by recreational fishers. Brown trout from three different lakes and one anadromous watercourse, and European whitefish (*Coregonus lavaretus*) from one lake. Additional samples of Atlantic char and sea trout were obtained from one anadromous watercourse and one non-anadromous lake (Atlantic char only).

Atlantic salmon were classified as wild or farmed based on scale reading, although scales from one salmon was not available. Wild caught broodfish were also classified by genetic tests. [13-17].

Gill samples fixed in RNAlater™ were sent to Patogen AS for specific real-time RT-PCR analysis of Atlantic salmon paramyxovirus (ASPV), *Candidatus* Branchiomonas cysticola, *Paranucleospora theridion*, *Candidatus* Piscichlamydia salmonis, Salmon gill poxvirus (SGPV) and *Paramoeba perurans*. Only gill samples from Atlantic salmon and seatrout caught in sea were analysed for *Paramoeba perurans*.

Gill samples from landlocked salmon was tested at the Norwegian Veterinary Institute in 2016 by real-time RT-PCR analysis for Salmon gill poxvirus (SGPV).

Table 1. Overview of study sample including species, morphology, year and number of counties, watercourses and fish.

Species		Counties	Locations	No. Fish	Sampling Years
Anadromous	Atlantic salmon <i>Salmo salar</i> L.	4	9	132	2018, 2017
	Seatrout <i>Salmo trutta</i> L.	3	4	69	2011, 2016
	Brown trout <i>Salmo trutta</i> L.	1	1	13	2017
	Arctic char <i>Salvelinus alpinus</i> L.	2	2	24	2016, 2018
Non-anadromous	European whitefish <i>Coregonus lavaretus</i>	1	1	21	2018
	Arctic char <i>Salvelinus alpinus</i> L.	1	1	20	2018
	Landlocked salmon <i>Salmo salar</i> L.	2	2	71	2016
	Brown trout <i>Salmo trutta</i> L.	3	3	48	2016, 2018

Results

Table 2. Results from gill samples analyzed by RT-PCR. Results reported as number of positive samples and in % (*: , **tested by NVI in 2016 [10], ***).

Fishgroup	# localities	# fish	Atlantic salmon paramyxovirus (ASPV)		<i>Candidatus Branchiomonas cysticola</i>		<i>Paranucleospora theridion</i>		<i>Candidatus Piscichlamydia salmonis</i>		Salmon gill poxvirus (SGPV)		<i>Paramoeba perurans</i>	
			Result	Ct-value	Result	Ct-value	Result	Ct-value	Result	Ct-value	Result	Ct-value	Result	Ct-value
Sea trout - sea	1	2	0		2	31.8-32.2	1	35	0		0		0	
Sea trout - anadromous watercourse	2	42	5 (11.9)	33.1-36	39 (92.9)	18.4-36.5	0		14 (33.3)	22.6-36.5	0		0*	
Sea trout - wild caught brood fish	1	25	0		13 (52.0)	25.6-36.5	6 (24.0)	33-36.7	0		0		-	
Brown trout - anadromous watercourse (Suspected stationary)	1	13	0		7 (53.9)	13.9-36.9	0		2 (15.4)	23-35	0		-	
Brown trout - non-anadromous freshwater	3	48	0		19 (39.6)	21.2-36.8	0		8 (16.7)	21.8-34.4	0		-	
Atlantic salmon - non-anadromous	2	71	0		0		0		0		0**		-	
Atlantic salmon - sea	3	33	0		28 (84.9)	17.7-36.5	3 (9.1)	32-36.6	6 (18.2)	22.5-35.7	0		0	
Atlantic salmon - river	6	70	0		59 (84.3)	17.9-36.1	13 (18.6)	19.9-36.6	24 (34.3)	21.4-36.7	9 (12.9)	23.9-36.9	-	
Atlantic salmon - wild caught brood fish	1	29	0	0	29 (100)	19.6-34.1	28 (96.6)	17-35	13 (44.8)	21.5-36.9	26 (89.7)	20.6-36.8	-	
Arctic char - non-anadromous	1	20	0		0		0		0		0		-	
Arctic char - anadromous watercourse	1	13	0		6 (46.2)	21.5-33.2	0		0		0		0***	
Arctic char in farm - wild caught brood fish	1	11	0		11 (100)	18.9-31.1	0		0		0		-	
European whitefish	1	21	0		5 (23.8)	25.5-34.7	0		0		0		-	

Based on the scale reading, four Atlantic salmon was classified as hatchery-reared smolt for stock enhancement purposes and two as either early escaped farmed salmon or hatchery-reared smolt for stock enhancement purposes. Scales from one salmon was not readable. All other Atlantic salmon was classified as wild.

Discussion and Conclusion

In this study, a limited number of individuals were examined across species and anadromously. Nevertheless, some of the findings deserve attention.

Ca. Branchiomonas cysticola was present in all the groups of salmonids except in non-anadromous Atlantic salmon and Atlantic char above anadromous watercourse. Mitchell et al. reported the bacteria to be highly prevalent in farmed Atlantic salmon, and it appeared to be common in gills of both diseased and apparently healthy farmed Atlantic salmon [3]. This corresponds with the results in this study, where it is the most common gill pathogen in wild salmonids in both freshwater and seawater.

Atlantic salmon paramyxovirus was only detected in sea trout caught in an anadromous watercourse. This implies that the virus is not widely distributed in wild salmonids.

In this study, *Ca. Branchiomonas cysticola* was detected in 23.4% of the gill samples from European whitefish that were included. The results show that it could be relevant to investigate the occurrence of pathogens in European whitefish and other freshwater fish in the freshwater sources of aquaculture facilities.

The occurrence of salmon gill poxvirus in various groups of salmon varied considerably. The virus was neither detected in non-anadromous salmon (Småblank and Byglandsblega) that were examined in 2016, nor in 33 salmon caught at sea in 2018. The occurrence in rivers was 12.9%, while 90% of the wild-caught broodstock collected for stock enhancement purposes were virus positive. This matches the findings from 2016 where SGPV had a high overall prevalence (83.7%). It is assumed that this is due to infection spreading in the tanks. In 2016, the prevalence of SGPV in salmon that were captured in a river and not held in tanks (River Rana) was 46.2% [10]. The virus can thus be quite prevalent in wild Atlantic salmon in rivers.

This study revealed a similar pattern for the microsporidia *Paranucleospora theridion* (*Desmozoon lepeophtherii*). This marine parasite was not detected in non-anadromous Atlantic salmon, while the prevalence in salmon in the sea (9 %) and in rivers (19 %) was considerably lower than in the gills of wild caught broodfish (97%). These results are potentially important with regards to the health of wild caught broodfish. Nevertheless, the limited number of individuals examined across geography, species and anadromously imply that one should be careful not to draw strong conclusions.

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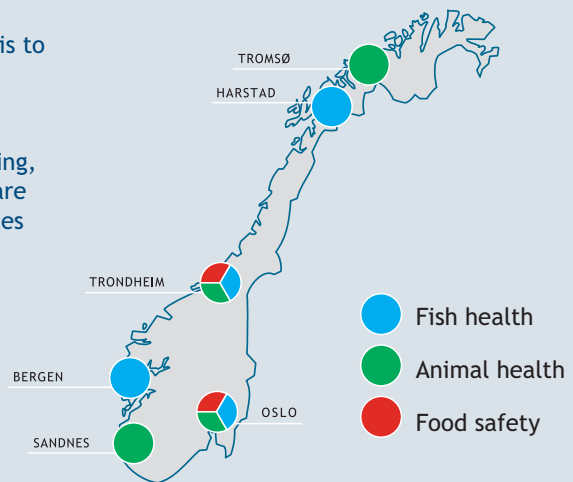
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