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Emerging viruses in aquaculture

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Aquaculture remains the world's fastest-growing sector producing food of animal origin. Unlike in terrestrial animal agriculture, in aquaculture both farmed and wild aquatic animals in the same water column experience the same virus challenges. Additionally, the burgeoning international aquaculture expansion and expanding global trade in live aquatic animals and their products have been accompanied by long distance geographical redistribution of aquatic animal species and their viruses. The outcome is a continuous emergence of viral diseases in aquaculture, which may be driven by virus factors, animal host factors, environmental factors, and/or anthropogenic factors. Examples of emerging viruses in aquaculture include viral haemorrhagic septicaemia virus, infectious haematopoietic necrosis virus, infectious salmon anaemia virus, piscine orthoreovirus, Tilapia lake virus, Covert mortality nodavirus, Shrimp hemocyte iridescent virus, and Abalone herpesvirus.

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Introduction

Aquaculture, the intensive water rearing of fish, mollusks, and crustaceans, remains the world's fastest-growing sector producing food of animal origin. As in the case of terrestrial animal agriculture, bringing together large numbers of animals than occur naturally involves substantial animal stress which facilitates virus multiplication and clinical disease. However, aquaculture presents unique challenges in contrast to all other intensive animal production systems, in that the aquatic farmed and wild animals occur in the same water column, and the aquatic environmental parameters cannot be very closely controlled as for captive livestock agriculture (for example as in the poultry and swine industries). Viruses, carried by wild aquatic animals where they are often not sufficient to sustain the natural transmission cycle density, are readily facilitated by the high density of hosts in aquaculture, which with the associated chronic stress provide opportunities for the emergence of viral diseases [1••]. Additionally, the burgeoning international aquaculture expansion and expanding global trade in live aquatic animals and their products have been accompanied by rapid long-distance geographical redistribution of aquatic animal species and their viruses with emergence in the same or different aquatic animal species. The outcome of these events is a continuous emergence of viral diseases in aquaculture, which may be driven by virus factors, animal host factors, environmental factors, and/or anthropogenic factors [1••]. For example, the wide use of 'cleaner fish' in marine farmed salmonids as a biological control for sea lice *Lepeophtheirus salmonis* in Europe and Canada is now considered a new route of emergence of viruses (such as viral haemorrhagic septicaemia virus (VHSV) and Cyclopterus lumpus virus) in fish aquaculture [2]. This practice is not only akin to mixing of species in fish farms, but has routinely involved use of wild-caught cleaner fish directly in the salmon farms or as broodstock for hatchery-raised cleaner fish [2]. Moreover, progressive farming practices now enable discovery of emerging viruses through surveillance and laboratory diagnosis. Indeed, several new viruses infecting aquatic organisms have been discovered through Next-generation sequencing (NGS) methods [3].

Several emerging, and re-emerging viruses in aquaculture will be highlighted in this overview. Many are listed by the World Organization for Animal Health (OIE), which means that countries free of these viruses can refuse imports of live aquatic animals and their products from areas that have not been declared virus-free, regardless of existing free trade agreements [4].

Carrier status in global movement of live aquatic animals and their products

The potential for dissemination of aquatic viruses because of aquaculture and movements of live cultured aquatic animals or their eggs is extremely high where persistent viral infections occur in the absence of clinical disease (i.e. 'healthy carrier' aquatic animals/subclinical infections in aquatic animals). Although life-long infections are known to occur among herpesvirus infections and retrovirus infections, there are several other virus groups where infection is not cleared by the host and the virus persists in a carrier state including species susceptible to infection without displaying clinical signs, age-related resistance to virus infection (e.g. adult fish), and infection with virus strains of low pathogenicity. There could also be situations of persistent infections where the virus level falls below detectable levels but not completely cleared from the host. All such infected animals are considered 'healthy' and may pass regulatory inspections for movement and/or export. This would be expected not only for new emerging viruses like piscine orthoreovirus (PRV) and tilapia lake virus (TiLV), which have been in existence but unknown until they were discovered [1••], and diagnostic tools developed not only for their detection, but also for re-emerging viruses such as VHSV, infectious haematopoetic necrosis virus (IHNV), infectious pancreatic necrosis virus (IPNV), and infectious salmon anaemia virus (ISAV) that cause persistent viral infections associated with lower virus levels in affected fish that may be difficult to detect through routine surveillance programs [5]. Most recently, 8000 juvenile Atlantic salmon at a commercial hatchery in Washington State-USA had to be destroyed because they tested positive for a strain of PRV found in Iceland. The virus is considered to have originated from fish eggs imported from Iceland. The source company for the eggs reported that they have an optional service of screening against PRV customers may choose as an extra risk measure to avoid vertical transmission (Owen E, 2018. https://salmonbusiness.com/egg-supplier-responds-to-washington-prv-salmon-cull/). In both examples above of new emerging viruses and re-emerging viruses where broodstock would have been persistently infected, the viruses would be disseminated via broodstock, fry or smolt movements, or egg transport into disease free farms, zones or countries. Where apparently 'healthy' aquatic animals are delivered to processing plants, the viruses would be disseminated via global trade in aquaculture products. In areas where these viruses are enzootic, clinical disease may manifest with the introduction of virus in imported aquatic material as for example with IPNV in Ireland where all reported clinical outbreaks were associated with imported IPNV isolates. In case of IHNV, in European countries where the main mode of virus transfer is by trade in infected fish, IHNV may remain undetected once introduced on a farm site [5].

The situation is even more concerning where international regulatory methods of control (e.g. for OIE listed diseases) dictate depopulation of affected farms upon virus detection in a few animals with few or none with clinical disease. In such situations, the affected animals may be allowed for human consumption and through international trade serve to introduce virus to new geographical areas. For example, White spot syndrome virus (WSSV), a highly infectious virus with a very wide crustacean host range has spread to all prawn-producing countries in the world with global movement of live shrimp. Until 2016, the Australian prawn industry was considered free of WSSV. Australia's biosecurity arrangements were breached by WSSV from Asia resulting in an outbreak in commercial *Penaeus monodon* prawn farms in Queensland in November–December 2016. The most likely route of infection appears to be via imported infected retail prawns used for human consumption and as bait by fishers (Loynes K. 2017.https://www.aph.gov.au/About_Parliament/Parliamentary_Departments/Parliamentary_Library/pubs/rp/rp1718/Chronology/WhiteSpotDiseaseAustralia). It is generally accepted that freezing seafood results in reduced infectivity of associated aquatic viruses.

Selected emerging viruses in fish aquaculture

Viral haemorrhagic septicaemia (VHS) virus (VHSV)

VHSV belongs to the species *Oncorhynchus 2 novirhabdovirus*, genus *Novirhabdovirus* within the family *Rhabdoviridae* [6]. Genotyping in accordance with VHSV G-gene and N-gene reveals four major genotypes (I–IV) that correspond with the broad geographical origins and host specificity of isolates. VHS is a notifiable disease to the OIE [7]. VHSV has been isolated from more than 82 marine and freshwater fish species, with at least 44 of these species shown to be susceptible [7] although its economic importance is primarily to the rainbow trout and turbot aquaculture in Europe and Japanese flounder (*Paralichthys olivaceus*) in Japan and olive flounder (*Paralichthys olivaceus*) in Korea.

VHSV is assumed to be endemic among a wide range of marine and anadromous fish species in the northern hemisphere [7], occasionally emerging in aquaculture as shown by transmission events reported for rainbow trout reared in marine and brackish waters in Finland, Norway, and Sweden, and the recent detections of VHSV III in wrasse species (*Labridae*) used as cleaner fish in Atlantic salmon farms in Scotland and VHSV IVd in wild lumpfish (*Cyclopterus lumpus*) brought to a land-based farm in Iceland, to serve as broodfish [8••].

Infectious haematopoietic necrosis virus (IHNV)

IHNV belongs to the species *Oncorhynchus 1 novirhabdovirus*, genus *Novirhabdovirus* within the family *Rhabdoviridae* [6]. In contrast to VHSV in the same genus, IHNV has a relatively narrow host range restricted to salmonids, fish families *Oncorhynchus* and *Salmo*. Genotyping according to the glycoprotein gene reveals five major genogroups. Three of the genotypes, on the basis of a 303-nucleotide variable region ('mid-G'), are designated as U (upper), M (middle), and L (lower), respectively, to correlate with the geographic areas in the Pacific Northwest of North America; the fourth and fifth genogroups based on the full-length glycoprotein gene, are 'E' and 'JRt' or 'J', consisting of European and Japanese rainbow trout isolates, respectively. IHNV is endemic to western North America where it was first described in Sockeye salmon (*Oncorhynchus nerka*) fry hatcheries in the early 1950s, and is considered to have spread to Europe and Japan via shipments of IHNV-contaminated rainbow trout eggs or fry. IHNV appears to travel through Europe without significant restrictions, termed viral 'tourism' as a consequence of frequent fish trade between private farms [9••]. IHN is a notifiable disease to the OIE [10]. Phylogenetic analysis of recent IHNV isolates in China indicate existence of a recently introduced virus via transfer of eggs or fish from North America where endemic virus continues to circulate undetected [11].

Infectious salmon anaemia virus (ISAV)

ISAV belongs to the species *Salmon isavirus*, genus *Isavirus* within the family *Orthomyxoviridae*. Genotyping based on the haemagglutinin-esterase (HE) gene reveals two basic genotypes, North American and European. ISAV strain designation is mostly based on sequence deletions/insertions in a 35-amino acid highly polymorphic region (HPR) of the HE protein [12]. Viruses without any deletion/insertion in HPR are designated ISAV-HPR0 to indicate 'full-length HPR' and are resistant to growth in cell culture, nonpathogenic, replicate only in epithelial cells of Atlantic salmon gills, and cause transient infection [12]. All ISAV isolated in fish cell lines to date from clinical disease have deletions in HPR relative to HPR0 and are referred to as ISAV-HPR-deleted (ISAV-HPRΔ). Virulent ISAV-HPRΔ targets endothelial cells resulting in systemic haemorrhagic disease. ISA is one of the most important salmonid viruses and is notifiable to the OIE [13]. Since 2012, ISAV outbreaks have been reported mostly in Norway, Canada and Chile. ISAV-HPRΔ was detected by RT-PCR but could not be isolated in cell culture, at a Chinese entry-exit port in 1 of 79 batches of eviscerated fresh salmon imported from Norway in 2015; the shipment was disposed of without entering Chinese aquaculture [14]. China currently has one of the world's biggest fully submerged net cage farming Atlantic salmon in the Yellow Sea (Owen E, 2018. https://salmonbusiness.com/chinas-gets-ready-to-harvest-first-batch-of-farmed-salmon-from-huge-deep-sea-fully-submersible-fish-cage/). The level of risk of introducing ISAV into a disease free country via importation of frozen whole salmon or fillets may be lower than with non-frozen salmon products as ISAV is sensitive to freezing and thawing [15].

Another fish orthomyxovirus, rainbow trout orthomyxovirus (RbtOV) isolated from juvenile rainbow and spawning steelhead trout (both *Oncorhynchus mykiss*) has been suggested to belong to a new genus, proposed name *Mykissvirus*, in the family *Orthomyxoviridae* [16]. RbtOV appears to have a relatively low prevalence in trout populations, grows in cell culture but is nonpathogenic in fish [16].

Tilapia lake virus (TiLV)

TiLV is a new orthomyxovirus of fish. It has a genome of 10 segments of linear negative sense single stranded RNA. It belongs to the species *Tilapia tilapinevirus*, genus *Tilapinevirus* within the family *Orthomyxoviridae*. Since its discovery as the etiological cause of massive losses of tilapia in Israel and Ecuador in 2009 [17••], TiLV has emerged as a significant cause of fish disease with mortality rates of 10–90% in farmed tilapia and the wild population in 12 countries across 3 continents (Asia, Africa, South America) [18]. TiLV represents an important risk for the fast-growing worldwide tilapia production sector. Tilapia is the world's second-most-farmed fish after carp [19]. It is possible that international trade may have been circulating TiLV worldwide through movement of live fish for aquaculture in the absence of knowledge of the existence of an associated risk [19,20]. It was recently shown that TiLV is inactivated in tilapia fillets stored at $-20°C$ for 90–120 days [21] demonstrating that frozen seafood (e.g. whole fish or fillets) imports may be associated with lower risk of virus dissemination than non-frozen products. TiLV has not yet been detected in North America tilapia stocks [22].

Salmonid alphavirus (SAV)

SAV belongs to the genus *Alphavirus* within the family *Togaviridae*. SAV is the cause of pancreas disease (PD) and sleeping disease (SD), viral diseases of serious concern for salmon aquaculture in Northern Europe [23,24]. Genomic, antigenic, and histopathological studies have shown that SPDV and SDV isolates are closely related strains of the same virus now referred to as SAV. Six different subtypes of SAV (SAV1-6) have been identified using phylogenetic analysis with partial glycoprotein E2 and nonstructural protein-3 (nsP3)-gene sequence data, providing evidence that some subtypes are dominant in certain geographical regions [25], and each subtype likely represents a single and separate introduction to aquaculture from a wild reservoir in or around the North Sea. SAV has been isolated from wild common dab *Limanda limanda* and plaice *Pleuronectes platessa* in Scotland and Ireland. The disease, which was first recorded in 1976 in Scotland, has continued as a significant threat to sustainable salmon production in Scotland, Ireland, Norway, France, Spain, Germany, Switzerland, and most recently Poland. SAV infections are on the OIE list of notifiable aquatic animal diseases [25]. To date there has been no confirmed reports of SAV in North America [26].

Piscine orthoreovirus (PRV)

PRV belongs to the family *Reoviridae*, subfamily *Spinareovirinae*. The PRV genome comprises of 10 segments of double-stranded RNA and all of them have been sequenced [27,28]. PRV is considered to be ubiquitous in farmed Atlantic salmon. It is an emerging virus of salmon aquaculture that is associated with an ever-increasing list of clinical syndromes including heart and skeletal muscle inflammation (HSMI) in farmed Atlantic salmon in Norway, Chile and BC-Canada [27,29–31]. The PRV genomic segment S1 sequence differentiates PRV isolates into two genotypes, I and II [28,29], and each of them into two major subgenotypes designated Ia and Ib, and IIa and IIb (Kibenge *et al.*, unpublished). Figure 1 shows the PRV genotypes and subtypes, and their geographical locations and associated clinical conditions.

Selected emerging viruses in crustacean aquaculture

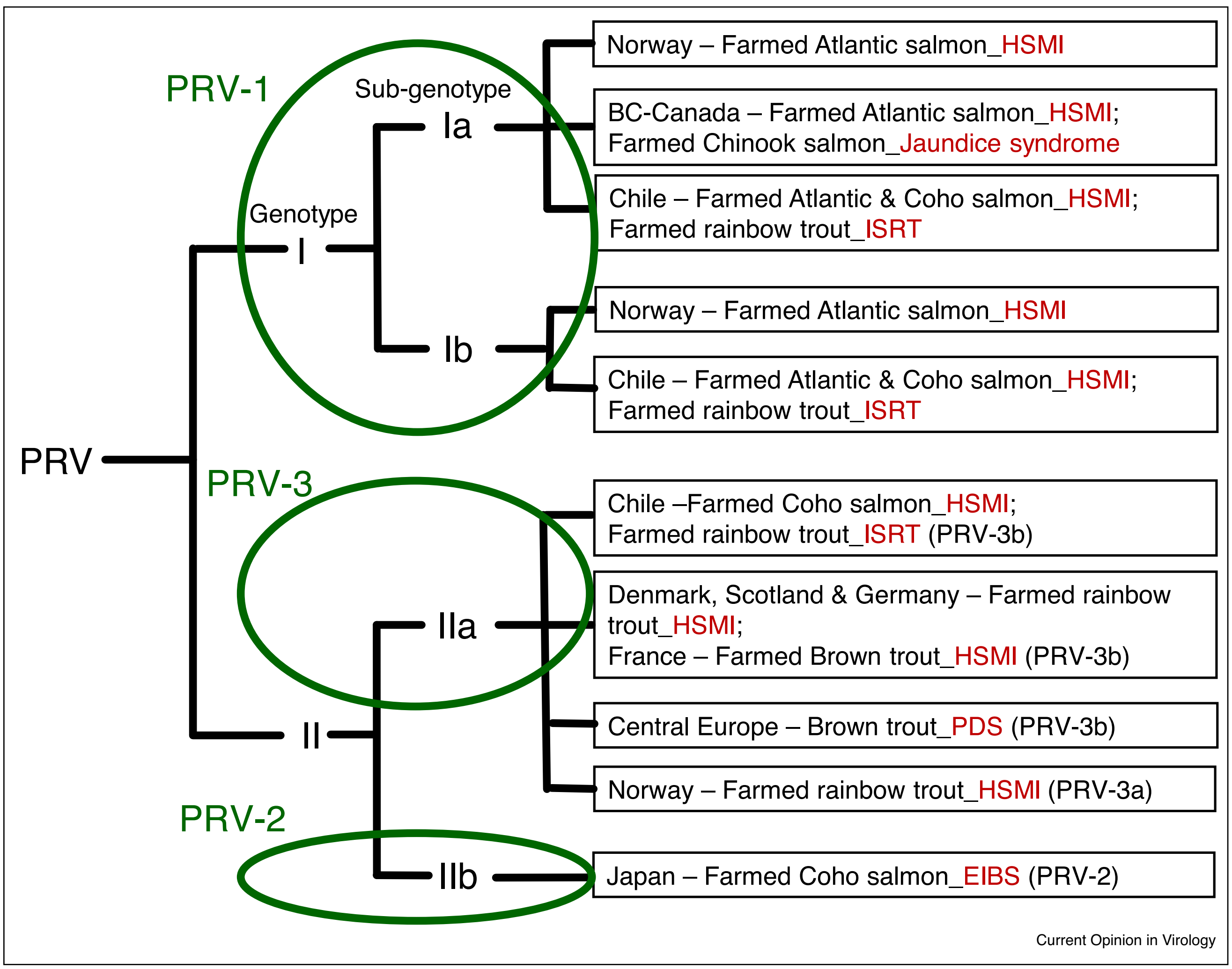
Shrimp hemocyte iridescent virus (SHIV)

A new virus of the family *Iridoviridae* isolated in China, results in a high mortality rate in white leg shrimp (*Litopenaeus vannamei*) [32]. The virus is proposed to be a member of the new genus *Xiairidovirus* [33] in family *Iridoviridae*. SHIV was detected in *L. vannamei*, *Fenneropenaeus chinensis*, and *Macrobrachium rosenbergii* in samples collected during 2014–2016 from 5 provinces in China [32].

Covert mortality nodavirus (CMNV)

CMNV is a new virus of the family *Nodaviridae*, genus *Alphanodavirus*. It is the cause of viral covert mortality disease of shrimp [34] which has caused serious loss in China since its emergence in 2002–2003. Shrimp infected with CMNV are commonly found in deep water on the

Figure 1



Piscine orthoreovirus (PRV) genotypes, subtypes, geographical location and associated fish diseases. Phylogenetic analysis of genome segment S1 groups PRV into two genotypes (I and II) and four subgenotypes (Ia, Ib, IIa and IIb) [28,29, Kibenge *et al.*, unpublished]. Following order of discovery, Genotype I is also referred as PRV-1, subgenotype IIb as PRV-2 [29], and subgenotype IIa as PRV-3 [Kibenge *et al.*, unpublished]. All the PRV-3 isolates can be further subdivided into PRV-3a from rainbow trout from Norway [41,42], and PRV-3b from the rest of Europe [42–44] and Chile [45,46].
HSMI = Heart and skeletal muscle inflammation in farmed Atlantic salmon [29–31]. In other fish species (coho salmon, rainbow trout, brown trout), the disease is referred to as 'HSMI-like' disease [29,41,42,44].
Jaundice and anemia (Jaundice syndrome) in farmed Chinook salmon in BC-Canada [47,48].
ISRT = Idiopathic syndrome of rainbow trout in fared rainbow trout in Chile [45].
EIBS = Erythrocyte inclusion body syndrome in farmed juvenile coho salmon in Japan [29].

bottom of the shrimp pond rather than swimming on the surface or in shallow water like shrimp infected with White spot syndrome virus (WSSV) [35]. The disease causes economic losses in hatcheries and farms due to high mortality rates of up to 80% commonly found within 60–80 days post-stocking. CMNV should not be confused with other nodavirus infections such as infectious myonecrosis virus (IMNV), *Macrobrachium rosenbergii* nodavirus (MrNV) and *Penaeus vannamei* nodavirus (PvNV) [35]. These viruses do not cause hepatopancreatic atrophy and necrosis, unlike CMNV.

CMNV has a wide host range among cultured shrimp species, with a high prevalence and wide distribution in Southeast Asia, and Latin American countries [34]. CMNV was found in eleven species of invertebrates collected from shrimp ponds of cultured shrimp species affected with VCMD, which may be vectors and reservoirs of CMNV [36]. CMNV has also naturally crossed the species barrier (i.e. jumped species) and infected several species of fish such as *Mugilogobius abei*, a common marine fish in shrimp farming ponds and coastal water in China, another marine fish *Chaeturichthys hexanema* found in the Yellow sea [37], and farmed Japanese flounder (*Paralichthys olivaceus*) [38••].

Selected emerging viruses in molluscan aquaculture

Abalone herpesvirus (AbHV)

AbHV is the cause of abalone viral ganglioneuritis (AVG) in farmed and wild abalone primarily in Australia and Chinese Taipei [39] and is listed by the OIE [40]. The virus is a member of the family *Malacoherpesviridae* [39] which includes Ostreid Herpesvirus-1 and is tentatively placed in a new genus *Haliotivirus*. The disease first occurred in Australia in 2005 [40].

Future perspectives

Aquaculture is important now and will continue in the future as a principal source of animal protein for human consumption, as will the global trade in live aquatic animals and their products. Aquatic animal viral diseases are inherent in aquaculture, and they continue to negatively impact aquaculture significantly. Considering that seafood is the most traded commodity globally, it, therefore, virtually impossible to have 'aquatic virus-leakproof' international borders. The implementation of strict biosecurity measures on aquaculture farms on land, in lakes and the sea, and in processing plants or other natural source for aquaculture helps to limit but does not eliminate the risk of dissemination of aquatic viruses. Biosecurity management will remain an on-going effort for the foreseeable future. The best options for keeping abreast of the continuous emergence of viral diseases in aquaculture are ideally at the farm level where better knowledge about the viral diseases and their improved diagnosis, inspection and surveillance programs translate into higher profits for the farmer and, therefore, motivation for a sustainable industry.

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References and recommended reading

Papers of particular interest, published within the period of review, have been highlighted as:

- • of special interest
- •• of outstanding interest

1. Kibenge FSB: **Determinants of emergence of viral diseases in aquaculture**. In *Aquaculture Virology*. Edited by Kibenge FSB, Godoy MG. Elsevier Inc., Academic Press; 2016:95-116.
••

This book chapter is a 'must read' about emerging viruses in aquaculture. It presents a detailed context for understanding the drivers responsible for the emergence of viral diseases in aquaculture and fisheries. The most recent definition of an 'emerging infectious disease' is used to group emerging aquatic animal viruses into eight categories: (1) a virus introduced in a new host and new geographic area; (2) a virus infecting the same host but introduced in a new geographic area; (3) enzootic virus with increased pathogenicity in same host (manifesting as increased incidence or more severe disease); (4) enzootic virus with increased host range; (5) enzootic virus with expanded geographic range; (6) increased awareness; (7) improved diagnosis and surveillance; and (8) newly discovered viral cause of existing disease. An emerging aquatic animal viral disease may fit in several of these categories depending on the sequence of events involved and geographical area concerned. The drivers (attributes or causal factors) of emerging viruses can be categorized into virus drivers, animal host drivers, environment drivers and anthropogenic drivers. The interplay of these drivers manifests into new or previously unrecognized viral diseases or re-emerging viral diseases in aquaculture and fisheries. A separate distinct category is introduced for those viral diseases whose incidence is decreasing as a result of anthropogenic means or through natural means, as this is the ultimate goal of disease control and prevention.

2. Murray AG: **A modelling framework for assessing the risk of emerging diseases associated with the use of cleaner fish to control parasitic sea lice on salmon farms**. *Transbound Emerg Dis* 2016, **63**:e270-e277.

3. Munang'andu HM, Mugimba KK, Byarugaba DK, Mutoloki S, Evensen Ø: **Current advances on virus discovery and diagnostic role of viral metagenomics in aquatic organisms**. *Front Microbiol* 2017, **8**:406.

4. World Organization for Animal Health (OIE): *Aquatic Animal Health Code*. edn 21. 2018 . Available at: *http://www.rr-africa.oie.int/docspdf/en/Codes/en_csaa.pdf*.

5. Dixon P, Paley R, Alegria-Moran R, Oidtmann B: **Epidemiological characteristics of infectious hematopoietic necrosis virus (IHNV)**. *Vet Res* 2016, **47**:63.

6. Afonso CL, Amarasinghe GK, Bányai K, Báo Y, Basler CF, Bavari S, Bejerman N, Blasdell KR, Briand F-X, Briese T *et al.*: **Taxonomy of the order Mononegavirales: update 2016**. *Arch Virol* 2016, **161**:2351-2360.

7. World Organization for Animal Health (OIE): *Manual of Diagnostic Tests for Aquatic Animals; Viral Haemorrhagic Septicaemia*. 2018 . Chapter 2.3.10. Available at: *http://www.oie.int/fileadmin/Home/eng/Health_standards/aahm/current/chapitre_vhs.pdf*.

8. Guðmundsdóttir S, Vendramin N, Cuenca A, Sigurðardóttir H, Kristmundsson A, Iburg TM, Olesen NJ: **Outbreak of viral haemorrhagic septicaemia (VHS) in lumpfish (*Cyclopterus lumpus*) in Iceland caused by VHS virus genotype IV**. *J Fish Dis* 2019, **42**:47-62 http://dx.doi.org/10.1111/jfd.12910.
••

The authors provided a detailed account of characterization of new VHSV isolates originating from wild lumpfish on the south coast of Iceland. These VHSV isolates clearly constitute a new subtype (IVd) never reported before, although the authors remain very cautious in interpretation of their findings.

9. Cieslak M, Wahli T, Diserens N, Haenen OLM, Schuétze H:
•• **Phylogeny of the infectious hematopoietic necrosis virus in European aquaculture**. *PLoS One* 2017, **12**:e0184490.
The authors confirm what has been known for a long time that IHNV is transmitted throughout Europe through active trout trade.

10. World Organization for Animal Health (OIE): *Manual of Diagnostic Tests for Aquatic Animals; Infectious Haematopoietic Necrosis*. 2018 . Chapter 2.3.4. Available at: *http://www.oie.int/fileadmin/Home/eng/Health_standards/aahm/current/chapitre_ihn.pdf*.

11. Jia P, Breyta RB, Li Q, Qian X, Wu B, Zheng W, Wen Z, Liu Y, Kurath G, Hua Q *et al.*: **Insight into infectious hematopoietic necrosis virus (IHNV) in Chinese rainbow trout aquaculture from virus isolated from 7 provinces in 2010–2014**. *Aquaculture* 2018, **496**:239-246.

12. Kibenge FSB, Kibenge MJT: **Orthomyxoviruses of fish**. In *Aquaculture Virology*. Edited by Kibenge FSB, Godoy MG. Elsevier Inc, Academic Press; 2016:299-326.

13. World Organization for Animal Health (OIE): *Manual of Diagnostic Tests for Aquatic Animals; Infectious Salmon Anaemia*. 2018 . Chapter 2.3.5. Available at: *http://www.oie.int/fileadmin/Home/eng/Health_standards/aahm/current/chapitre_isav.pdf*.

14. Xiao L, Lin H, Yang M, Chen S, An W, Wu X, Liu H, Li D, Yan Y, Hu J *et al.*: **The first instance of HPR-deleted ISAV detection in eviscerated, fresh salmon at a Chinese entry-exit port**. *Aquaculture* 2018, **485**:220-224.

15. Ramírez R, Marshall SH: **Identification and isolation of infective filamentous particles in infectious salmon anemia virus (ISAV)**. *Microb Pathog* 2018, **117**:219-224.

16. Batts WN, LaPatra SE, Katona R, Leis E, Ng TFF, Brieuc MSO, Breyta RB, Purcell MK, Conway CM, Waltzek TB *et al.*: **Molecular characterization of a novel orthomyxovirus from rainbow and steelhead trout (*Oncorhynchus mykiss*)**. *Virus Res* 2017, **230**:38-49.

17. Bacharach E, Mishra N, Briese T, Zody MC, Kembou Tsofack JE,
•• Zamostiano R, Berkowitz A, Ng J, Nitido A, Corvelo A *et al.*: **Characterization of a novel Orthomyxo-like virus causing mass die-offs of tilapia**. *MBio* 2016, **7** e00431-00416.
The authors were the first to characterize a novel orthomyxovirus with 10 genome segments as the cause of a new viral disease of tilapia. Using next-generation sequence methodology, the authors found only one of the genomic segments (segment 1) to have weak sequence similarity with Gammainfluenzavirus; all the other nine genome segments were identified based on conserved terminal sequences characteristic of orthomyxoviruses.

18. Jansen MD, Dong HT, Mohan CV: **Tilapia lake virus: a threat to the global tilapia industry?** *Rev Aquacult* 2018:1-15 http://dx.doi.org/10.1111/raq.12254.

19. Food and Agriculture Organization of the United Nations (FAO): *FAO Fisheries and Aquaculture Circular No. 1135/5 2017*. 2017 . Available at: *http://www.fao.org/3/a-i6875e.pdf*.

20. Dong HT, Ataguba P, Khunrae T, Rattanarojpong T, Serapin S: **Evidence of TiLV infection in tilapia hatcheries from 2012 to 2017 reveals probable global spread of the disease**. *Aquaculture* 2017, **479**:579-583.

21. Thammatorn W, Rawiwan P, Surachetpong W: **Minimal risk of tilapia lake virus transmission via frozen tilapia fillets**. *J Fish Dis* 2019, **42**:3-9 http://dx.doi.org/10.1111/jfd.12924.

22. Al-Hussinee L, Subramaniam K, Ahasan MS, Keleher B, Waltzek TB: **Complete genome sequence of a tilapia lake virus isolate obtained from Nile tilapia (*Oreochromis niloticus*)**. *Genome Announc* 2018, **6** e00580-18.

23. Hjortaas MJ, Bang Jensen B, Taksdal T, Olsen AB, Lillehaug A, Trettenes E, Sindre H: **Genetic characterization of salmonid alphavirus in Norway**. *J Fish Dis* 2016, **39**:249-257.

24. Deperansińska I, Schulz P, Simicki AK: **Salmonid alphavirus (SAV)**. *J Vet Res* 2018, **62**:1-6.

25. World Organization for Animal Health (OIE). Manual of diagnostic tests for aquatic animals; Salmonid alphavirus, Chapter 2.3.6. Available at: http://www.oie.int/fileadmin/Home/eng/Health_standards/aahm/current/chapitre_salmonid_alphavirus.pdf.

26. Purcell MK, Powers RL, Evered J, Kerwin J, Meyers TR, Stewart B, Winton JR: **Molecular testing of adult Pacific salmon and trout (*Oncorhynchus* spp.) for several RNA viruses demonstrates widespread distribution of piscine orthoreovirus (PRV) in Alaska and Washington**. *J Fish Dis* 2018, **41**:347-355.

27. Kibenge FSB, Godoy MG: **Reoviruses of aquatic organisms**. In *Aquaculture Virology*. Edited by Kibenge FSB, Godoy MM. Elsevier Inc, Academic Press; 2016:205-236.

28. Takano T, Nawata A, Sakai T, Matsuyama T, Ito T, Kurita J, Terashima S, Yasuike M, Nakamura Y, Fujiwara A *et al.*: **Full-genome sequencing and confirmation of the causative agent of erythrocytic inclusion body syndrome in coho salmon identifies a new type of piscine orthoreovirus**. *PLoS One* 2016, **11**:e0165424.

29. Godoy MG, Kibenge MJT, Wang Y, Suarez R, Leiva C, Vallejos F, Kibenge FSB: **First description of clinical presentation of piscine orthoreovirus (PRV) infections in salmonid aquaculture in Chile and identification of a second genotype (Genotype II) of PRV**. *Virol J* 2016, **13**:98.

30. Alarcon M, Dahle M, Taksdal T: **Heart and skeletal muscle inflammation**. In *The Health Situation in Norwegian Aquaculture 2015.* Edited by Hjeltnes H, Walde C, Bang Jensen B, Haukaas A. Veterinærinstituttet Rapportserie; 2016:28-29. (in Norwegian).

31. Di Cicco E, Ferguson HW, Schulze AD, Kaukinen KH, Li S, Vanderstichel R, Wessel Ø, Rimstad E, Gardner IA, Hammell KL *et al.*: **Heart and skeletal muscle inflammation (HSMI) disease diagnosed on a British Columbia salmon farm through a longitudinal farm study**. *PLoS One* 2017, **12**:e0171471.

32. Qiu L, Chen MM, Wan XY, Li C, Zhang QL, Wang RY, Cheng DY, Dong X, Yang B, Wang XH *et al.*: **Characterization of a new member of Iridoviridae, Shrimp hemocyte iridescent virus (SHIV), found in white leg shrimp (*Litopenaeus vannamei*)**. *Sci Rep* 2017, **7**:11834.

33. Qiu L, Chen MM, Wang R-Y, Wan XY, Li C, Zhang QL, Dong X, Yang B, Xiang JH, Huang J: **Complete genome sequence of shrimp hemocyte iridescent virus (SHIV) isolated from white leg shrimp, *Litopenaeus vannamei***. *Arch Virol* 2018, **163**:781-785.

34. Zhang Q, Xu T, Wan X, Liu S, Wang X, Li X, Dong X, Yang B, Huang J: **Prevalence and distribution of covert mortality nodavirus (CMNV) in cultured crustacean**. *Virus Res* 2017, **233**:113-119.

35. Pooljun C, Direkbusarakom S, Chotipuntu P, Hirono I, Wuthisuthimethaveea S: **Development of a TaqMan real-time RT-PCR assay for detection of covert mortality nodavirus (CMNV) in penaeid shrimp**. *Aquaculture* 2016, **464**:445-450.

36. Liu S, Wang X, Xu TT, Li X, Du L, Zhang Q: **Vectors and reservoir hosts of covert mortality nodavirus (CMNV) in shrimp ponds**. *J Invertebr Pathol* 2018, **154**:29-36.

37. Zhang QL, Liu S, Li J, Xu TT, Wang XH, Fu GM, Li XP, Sang SW, Bian XD, Hao JW: **Evidence for cross-species transmission of covert mortality nodavirus to new host of Mugilogobius abei**. *Front Microbiol* 2018, **9**:1447.

38. Wang C, Liu S, Li X, Hao J, Tang KFJ, Zhang Q: **Infection of covert**
•• **mortality nodavirus in Japanese flounder reveals host jump in the emerging alphanodavirus**. *J Gen Virol* 2018 http://dx.doi.org/10.1099/jgv.0.001177. [Epub ahead of print].
The authors report a unique occurrence of newly discovered virus in shrimp (belonging to a virus group,*Alphanodavirus*, known to infect only insects) having jumped from shrimp to a new host species (fish). It is not clear how long the virus had existed in shrimp before successfully infecting fish.

39. Renault T: **Malacoherpesviruses of mollusks**. In *Aquaculture Virology.* Edited by Kibenge FSB, Godoy MG. Elsevier Inc, Academic Press; 2016:513-524.

40. World Organization for Animal Health (OIE): *Manual of Diagnostic Tests for Aquatic Animals; Abalone Herpesvirus*. 2018 . Chapter 2.4.1. Available at: In: *http://www.oie.int/fileadmin/Home/eng/*

Health_standards/aahm/current/chapitre_abalone_herpesvirus.pdf.

41. Hauge H, Vendramin N, Taksdal T, Olsen AB, Wessel Ø, Mikkelsen SS, Alencar ALF, Olesen NJ, Dahle MK: **Infection experiments with novel Piscine orthoreovirus from rainbow trout (*Oncorhynchus mykiss*) in salmonids**. *PLoS One* 2017, **12**: e0180293.

42. Dhamotharan K, Vendramin N, Markussen T, Wessel Ø, Cuenca A, Nyman IB, Olsen AB, Tengs T, Dahle MK, Rimstad E: **Molecular and antigenic characterization of Piscine orthoreovirus (PRV) from Rainbow Trout (*Oncorhynchus mykiss*)**. *Viruses* 2018, **10**:170.

43. Kuehn R, Stoeckle BC, Young M, Popp L, Taeubert J-E, Pfaffl MW, Geist J: **Identification of a piscine reovirus-related pathogen in proliferative darkening syndrome (PDS) infected brown trout (*Salmo trutta* fario) using a next generation technology detection pipeline**. *PLoS One* 2018, **13**:e0206164.

44. Adamek M, Hellmann J, Flamm A, Teitge F, Vendramin N, Fey D, Riße K, Blakey F, Rimstad E, Steinhagen D: **Detection of piscine orthoreoviruses (PRV-1 and PRV-3) in Atlantic salmon and rainbow trout farmed in German**. *Transbound Emerg Dis* 2019, **66**:14-21 http://dx.doi.org/10.1111/tbed.13018.

45. Cartagena J, Tambley C, Sandino AM, Spencer E, Tello: **Detection of piscine orthoreovirus in farmed rainbow trout from Chile**. *Aquaculture* 2018, **493**:79-84.

46. Bohle H, Bustos P, Leiva L, Grothusen H, Navas E, Sandoval A, Bustamante F, Montecinos K, Gaete A, Mancilla M: **First complete genome sequence of piscine orthoreovirus variant 3 infecting coho salmon (*Oncorhynchus kisutch*) farmed in southern Chile**. *Genome Announc* 2018, **6** e00484-18.

47. Miller KM, Günther OP, Li S, Kaukinen KH, Ming TJ: **Molecular indices of viral disease development in wild migrating salmon**. *Conserv Physiol* 2017, **5** cox036.

48. Di Cicco E, Ferguson HW, Kaukinen KH, Schulze AD, Li S, Tabata A, Günther OP, Mordecai G, Suttle CA, Miller KM: **The same strain of Piscine orthoreovirus (PRV-1) is involved with the development of different, but related, diseases in Atlantic and Pacific Salmon in British Columbia**. *FACETS* 2018, **3**:599-641.