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Viral Infections of Cultured Fish in Japan

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ABSTRACT

Since infectious pancreatic necrosis virus and infectious hematopoietic necrosis virus were first isolated in the 1970s, more than 20 fish viruses have been isolated and at least 5 viruses have been observed by electron microscopy. Viral diseases are major problems and cause economic losses among cultured fishes in Japan and other countries. This paper reports our current understanding of the extent of viral infection in the cultured fishes of Japan.

Introduction

A virological study of cultured fishes in Japan was initiated when an unknown disease occurred among rainbow trout *Oncorhynchus mykiss* in the 1960s. The causative agent was identified as infectious pancreatic necrosis virus (IPNV) (Sano 1971). Subsequently, infectious hematopoietic necrosis virus (IHNV) was isolated from kokanee salmon *O. nerka* (Kimura and Awakura 1977). Since then, various viral infections of fish have been reported. At present, more than 20 fish viruses have been isolated and at least 5 viruses have been observed by electron microscopy studies (Table 1).

Viral Diseases of Salmonid Fishes

Infectious Pancreatic Necrosis

Infectious pancreatic necrosis is an acute systemic disease affecting the fry and fingerlings of rainbow trout. Its occurrence is widespread in Japan. Susceptibility of fish to IPNV depends on body weight; smaller fry are more susceptible. The signs of this disease are darkening of body coloring, moderate exophthalmia, and abdominal distention. Internally, the spleen, heart, liver, and kidneys are pale and the digestive tract is almost always devoid of food. Rainbow trout is the fish most affected by IPNV, but the virus has also been isolated from amago (*O. rhodurus*) and masu salmon (*O. masou*). Recently, the fry of rainbow trout were observed to be less susceptible to IPNV and, consequently, damage attributed to IPN has decreased (Okamoto et al. 1987).

Infectious Hematopoietic Necrosis

Infectious hematopoietic necrosis is an acute systemic disease which mainly affects the fry of rainbow trout and masu and kokanee salmon but which has also been isolated from moribund ayu *Plecoglossus altivelis* (Yoshimizu et al. 1987c). The characteristic sign of IHNV infection is V-shaped hemorrhages located in muscle tissue. Recently, a large rainbow trout, with a body weight of 50–80 g was found to be infected with IHNV and subsequently died (Mori et al. 1987). In this case, petechiae were observed in the fatty tissues and on the wall of the body cavity. This virus is widespread and especially prevalent in the central part of Honshu, the Japanese mainland (Sano et al. 1977). In several districts, river waters have been contaminated with IHNV and are now unsuitable for rainbow trout culture. Although the vertical transmission of IHNV is doubtful (Yoshimizu et al. 1988, a and b), it can be controlled by disinfecting eggs with iodine during the early eyed stage. Fish at the fry stage are very susceptible to IHNV. They should be reared in either well water or ultra-violet irradiated river water. When fish are past this sensitive stage, they can be transferred to the usual rearing ponds.

Table 1
Viral infection in cultured fishes in Japan.

Isolated virus	Host
DNA virus	
Nerkavirus in Towada Lake, Aomori and Akita Prefecture (NeVTA)	Kokanee salmon
Oncorhynchus masou virus (OMV)	Masu salmon
Yamame tumor virus (YTV)	Yamame (masu salmon)
Icosahedral cytoplasmic deoxyribovirus (ICDV)	Japanese eel
Herpesvirus cyprini	Fancy carp
Herpesvirus	Japanese eel
Unidentified small virus	Tiger puffer
RNA virus	
Infectious pancreatic necrosis virus (IPNV)	Salmonid fish
Infectious hematopoietic necrosis virus (IHNV)	Salmonid fish
Chum salmon virus (CSV)	Masu salmon
Yellowtail ascitic virus (YAV)	Yellowtail
Rhabdovirus olivaceus (HRV)	Various marine fish
Eel virus from European eel (EVE)	European eel
Eel virus of America (EVA)	American eel
Eel virus of Europe X (EVEX)	European eel
Papovavirus	Japanese eel
Birnavirus	Yellowtail
Birnavirus	Japanese flounder
Birnavirus	Red sea bream
Coronavirus cyprini virus (CACV)	Common carp
Picornavirus	Japanese eel
Reovirus	Common carp
Reovirus	Japanese eel
Observed by electron microscopy	
Viral erythrocytic necrosis virus	Various marine fish
Lymphocystis virus	Various marine fish
Paramyxovirus	Black rockfish
Herpesvirus	Japanese flounder
Picornavirus	Ishidai

Herpesvirus Infection

A herpesvirus, nerkavirus in Towada Lake Akita and Aomori Prefecture (NeVTA), was first isolated from diseased kokanee salmon in Towada Lake (Sano 1976). In 1978, another herpesvirus was isolated from the ovarian fluid of apparently normal mature masu salmon (Kimura et al. 1980). This virus was named *Oncorhynchus masou* virus (OMV) from the scientific name of the host fish. *Oncorhynchus masou* virus was found to be pathogenic and significantly more oncogenic in young masu salmon and several other salmonid fish (Kimura et al. 1981, a and b; Yoshimizu et al. 1987a). In 1983, a similar herpesvirus were isolated from tumor tissue of yamame (landlocked *O. masou*) and was named yamame tumor virus (Sano et al. 1983). Subsequent study showed that OMV is enzootic in the northern part of Japan (Yoshimizu et al. 1988b) and that the characteristics of these three herpesviruses are similar except that NeVTA lacks oncogenicity (Hedrick et al. 1987; Sano et al. 1988). In 1983, we recommended the disinfection of fish eggs with iodine at the early eyed stage in Hokkaido. Now OMV cannot be detected in most of the hatcheries in this area (Yoshimizu et al. 1988b). Although the host species of this virus is primarily masu salmon, OMV has also been isolated from the tumor tissues of pen-cultured coho salmon, *O. kisutch.*

Chum Salmon Virus Infection

In 1978, a reovirus was isolated from an apparently normal adult chum salmon, *O. keta* returning to its hatchery in Hokkaido (Winton et al. 1981). After initial isolation and characterization, it was named chum salmon virus (CSV). This virus was not observed again until 1986, during an episode of mass mortalities of masu salmon fry for which it was responsible. Since then, the virus has been detected in stocks of adult masu salmon at new locations in Hokkaido (Yoshimizu 1988). Artificial infection studies of this virus showed no significant mortality in several species of salmonid fishes (Winton et al. 1989).

Viral Erythrocytic Necrosis

Inclusion bodies stained with Giemsa were observed in the erythrocytes of chum and pink salmon, *O. gorbuscha,* collected in Okhotsuku and along the north Pacific coast of Hokkaido. The causative agent of viral erythrocytic necrosis (VEN), an iridovirus, was subsequently observed by electron microscopy (Yoshimizu et al. 1988b).

Viral Infections of Eels

Many viruses have been isolated from cultured eels (*Anguilla anguilla, A. japonicus,* and *A. rostrata*) by Sano (1976) and Sano and Fukuda (1987). They include a birnavirus, eel virus from the European eel (EVE); the rhabdoviruses, eel virus of America (EVA) and eel virus of Europe X (EVEX); papovavirus; herpesvirus; picornavirus; and a reovirus. These viruses are not recognized as pathogenic against eel except for EVE (Nishimura et al. 1981). Sorimachi (1982, 1984) reported having isolated icosahedral cytoplasmic deoxyribovirus (ICDV) from a diseased eel. This

virus was shown to be pathogenic against Japanese eels following artificial infection. Mortality was 40–75% at water temperatures of 14.5–18.5° C, 15% at 22.8° C, and 0% at 24.1° C. Infected fish showed signs of decoloration; congestion of the anal, pectoral, and dorsal fins; and an increase of mucus on the body surface.

Viral Infections of Carp

Herpesvirus cyprini was isolated from papilloma tissue of cultured fancy carp (*Cyprinus carpio,* also called common or asagi carp) and confirmed as the agent of infection by induction of epithelial tumors by artificial infection (Sano et al. 1985). *Coronavirus cyprini,* carp coronavirus (CACV), was isolated from diseased common carp raised in the laboratory. Fish infected with CACV showed acute mortality showing no external signs except erythematous skin on the abdomen. Experimentally, CACV was virulent for carp fry at 20° C. Cumulative mortality for 3-week-old fry was 72.5%. The affected fish manifested swollen and hemorrhagic abdomens filled with ascites and eventually died. Reovirus was also isolated from common carp (Sano and Fukuda 1987).

Viral Infection of Other Marine Fishes

Viral Pancreatic-Hepatic Necrosis of Yellowtail

A yellowtail ascites virus (a birnavirus) was isolated from the fry of yellowtail *Seriola quinqueradiata* (Sorimachi and Hara 1985). This epizootic is an acute viral infection of both naturally grown and hatchery-raised fry. The epizootic period occurs from May to June at water temperatures of 18 to 22° C. The moribund fry typically show anemic gills, hemorrhaging in the liver, and ascites and petechiae in the pyloric caea. The disease name, viral pancreatic-hepatic necrosis, was proposed by Egusa and Sorimachi (1986).

Rhabdovirus Infection of Japanese Flounder

The rhabdovirus, *Rhabdovirus olivaceus*—also referred to as hirame rhabdovirus (HRV)—was isolated from diseased hirame (Japanese flounder), *Paralichthys olivaceus,* and black sea bream, *Milio macrocephalus* (Gorie et al. 1985; Kimura et al. 1986). This virus is pathogenic for marine fish such as hirame, black sea bream, red sea bream *Pagrus major* and black rockfish *Sebastes inermis,* and also for salmonid species, especially rainbow trout and masu salmon (Yoshimizu et al. 1987b). Signs of HRV infection are gonadal congestion, focal hemorrhage of skeletal muscle and fins, and accumulation of ascitic fluid. Hirame rhabdovirus is distributed widely from Hokkaido to Honshu in Japan.

Kuchishiroshou of Tiger Puffer

From cultured tiger puffer, *Fugu rubripes,* an unidentified, small virus was isolated (Inoue et al. 1986). The epizootic period is from May to June during water temperatures of 18–22° C. Moribund fish had necrosis around the mouth and had been observed to be fighting with each other. From the signs of this infection, the disease was named "Kuchishiroshou", from the Japanese words "kuchi", meaning mouth, "shiro", meaning white, and "shou", meaning disease. Viral particles were observed in the brain by electron microscopy. Kuchichiroshou occurs in southwest Japan where tiger puffer are cultured.

Epidermal Hyperplasia of Japanese Flounder.

Outbreaks of a disease resulting in mass mortalities of larval and juvenile Japanese flounder was reported by Iida et al. (1989). Once the disease occurs in a pond, the resident fish populations usually become extinct within one month. Affected fish are characterized by opaque fins. Histopathologically, hyperplasia is observed in the epidermal layer of the fins and skin. In the epidermal tissues of infected fish, hexagonal virus particles were observed by electron microscopy. Japanese flounder larvae experimentally exposed to the filtrate of infected tissue homogenate suffered 18–50% mortalities with 93-100% of the survivors exhibiting epidermal hyperplasia. This virus has not been isolated in any of the 33 fish cell lines in which culture has been attempted, including that of the host species.

Birnaviruses were also isolated from Japanese flounder and red sea bream (Yoshimizu and Kimura, unpubl. data). These viruses were neutralized with antibody against IPNV; the pathogenicities of these birnaviruses have not been clarified.

Lymphocystis Disease

In several species of marine fishes, suzuki, *Lateolabrax japonicus,* yellowtail, red sea bream, Japanese flounder, and others, lymphocystis disease was re-

ported and iridovirus was observed by electron microscopy (Matsusato 1975; Miyazaki and Egusa 1972; Tanaka et al. 1984). Seasonal variation in the prevalence of lymphocystis was noted with increased prevalence in summer. Lymphocystis cells were observed mainly on the fins or body surface. The virus particles were polyhedral, presenting hexagonal or pentagonal profiles in tissue sections. They may be seen in a crystalline array and are always located in the cytoplasm.

Other Diseases

Yoshikoshi and Inoue (1988) reported picornavirus in moribund fry of ishidai, *Oplegnathus fasciatus*, and Miyazaki et al. (1989) reported herpesvirus in the epidermal necrosis of Japanese flounder and paramyxovirus in the epithelial necrosis of black sea bream. To date, these viruses have not been isolated.

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