

## XIV

DISEASES AND THEIR  
MANAGEMENT  
IN CAGE CULTURE

S.R.Krupesha Sharma, N. Sadhu and K.K.Philipose

Aquaculture is the farming of aquatic organisms including fish, molluscs, crustaceans and aquatic plants. A cage is a volume enclosed with some type of mesh forming a container for aquatic animals. Farming implies some sort of intervention in the rearing process to enhance production, such as regular stocking, feeding, protection from predators, etc. Aquaculture is a rapidly growing food producing sector in the world, with an average annual growth rate of 8.9% since 1970, compared to only 1.2% for capture fisheries and 2.8% for terrestrial farmed meat production systems over the same period.

The current trend in aquaculture development is towards increased intensification and commercialization of aquatic production. Like other farming sectors, the likelihood of major disease problems occurring increases as aquaculture activities intensify. Disease is now a primary constraint to the culture of many aquatic species, impeding both economic and social

development in many countries. Addressing health questions with both pro-active and reactive programmes has become a primary requirement for sustaining aquaculture production and product trade. It is reported that the principal diseases in cage aquaculture of marine finfish and shellfish in Asia are caused by environmental and management affects, nutritional causes, and viral, bacterial, parasitic, and fungal pathogens.

### Diseases caused by viruses

#### Viral nervous necrosis:

Viral Nervous Necrosis (VNN), also known as viral encephalopathy and retinopathy (VER) causes high mortality with vacuolation of the tissues in the central nervous system of affected fish. The disease occurs mostly in larvae and juveniles but some species such as Asian seabass and grouper are still susceptible at grow-out stages, and the disease with severe mortality has been reported in more than 32 species from 16 families. Electron microscopy revealed that unenveloped round-shaped virus particles, 25–34 nm in diameter, are packed in the cytoplasm of affected retinal and brain cells. The causative agent of VNN was purified from diseased striped jack and identified as a member of the family Nodaviridae, known as striped jack nervous necrosis virus. This disease is included as one of the significant diseases in the OIE Code.

In case of Asian seabass larvae, affected larvae show abnormal swimming behaviour, including spiral swimming, darting or sometimes vertical movement. Affected fish have an inflated swim bladder. Although the pathogenesis of the disease seems to be different among fish species, direct transmission via the epithelium of the skin and the gastrointestinal tract is reported to be important in the infection. The skin of the affected larvae changes from pale to dark. The appetite is affected and fish become lean. The disease occurs as early as at 10 days post-hatching (dph) to 50 dph. In some instances, particularly during stress, fish beyond 50 dph may still be affected. It is also common for the affected fish to have an inflated swim bladder.

The most distinctive lesion in histological sections of the brain and retina from seabass larvae with clinical signs of viral nervous necrosis includes extensive cell vacuolation. In addition, in general, in the central nervous system, the most prominent lesions are observed throughout the spinal cord and medulla oblongata. They include severe massive necrosis of the small nerve cells, spongiform encephalopathy characterized by extensive vacuolar degeneration in the cytoplasm of affected neurons and neuropils. Dark, dense, pyknotic nuclei, neuronal shrinkage and disappearance of Nissle's granule in their cytoplasm is occasionally observed. Focal extensive haemorrhaging in the molecular layer and focal demyelination characterized by degenerated myelin nerve fibres with phagocytic mononuclear foamy cell infiltration were also observed in the area between the Purkinje cell layer and molecular cell layer.

Studies on the development of vaccination methods in groupers are now undertaken.

#### Iridovirus infections:

Iridoviruses, causative agents of serious systemic diseases have been identified from more than 20 fish species in the recent years. Most fish iridoviruses are members either of the genera *Lymphocystis virus* or of the genera *Ranavirus*. Iridoviruses in genera *Lymphocystis virus* cause the development of cluster of extremely hypertrophied fibroblasts or osteoblasts called lymphocystis cells, while viruses in genera *Ranavirus* may lead to systemic disease in infected animals and are associated with high morbidity and mortality. A typical iridovirus has icosahedral symmetry and measures 130–300 nm in diameter. Characterization of iridoviruses has been hindered by the difficulty in isolating and propagating them in tissue cultures.

The most dramatic change in all affected fish is the presence of basophilic, hypertrophied cells, often in large numbers, in various organs. These cells, with a pale foamy or intensely basophilic granular appearance, were often observed in the splenic parenchyma and capsule, in the renal glomerulus and interstitium,



and in the intestinal lamina propria. Hypertrophied cells are also observed in the choroid plexus of the eye, pancreatic interstitium, the connective tissue surrounding ovarian follicles, and in connective tissues throughout the body. They were usually observed in smaller numbers in the liver, heart, gills and brain. Dark staining crescent-shaped cells are frequently observed, forming a "cap" over the hypertrophied cells. Degenerative and necrotic changes are often seen in association with cytomegaly. In some fish, splenic changes consists presence of typical hypertrophied cells accompanied by spongiosis and fibrinoid necrosis of ellipsoids and haemorrhage, with consequent destruction of the entire splenic architecture. Loss of renal glomeruli and interstitium is observed, but the renal tubules are usually unaffected.

#### Lymphocystis:

Lymphocystis disease (LCD), one of the common infectious diseases affecting marine fish cultures, was discovered in 1874. Distribution has been reported worldwide such as Spain, Korea, Japan and China. The causative agent of LCD is lymphocystis disease virus (LCDV) which is a large virus in the genus Lymphocystis virus of the family Iridoviridae. LCDV is an icosahedral symmetry virus, approximately 200-300 nm in diameter, and contains single linear double stranded DNA. LCD is characterized by the external appearance of nodules, either singly or in groups, on skin, fins, or tail of the affected fish. Although, LCD is not a fatal disease, the external appearance might cause a significant economic loss. The principle mode of transmission of LCD is horizontally by direct contact and external trauma. Other factors such as water contamination and stress condition caused by high stocking density, nutrition deficiencies, low dissolved oxygen, suboptimal water quality, or human manipulation may increase the appearance of LCD symptoms. A recent study reported that *Artemia* sp. might act as a reservoir host of this disease (Cano et al., 2009).

The affected fish reveals multifocal to diffuse white, firm, papilloma-like nodules scattered on the skin, fins, eyes and mouth.

The diameters of nodules varies in size, approximately 1-2 mm. Microscopically, many clusters of lymphocystis cells are observed in the connective tissues beneath the epidermis on fins, inner layer of operculum, and gills. Histopathologically lymphocystis disease is characterized by cytomegaly of dermal fibroblasts and only rarely is there any systemic involvement. Numerous hypertrophied cells with basophilic intra-cytoplasmic inclusion bodies are seen in connective tissues of the dermis and between the scales. The lymphocystis cells are also detectable in skeletal muscle, gill lamellae and visceral organs including spleen, head and trunk kidney. The lymphocystis-granulomatous also appear in parenchyma of the spleen and kidney.

Lymphocystis can be diagnosed by several immunological assays like indirect immunofluorescence assay, flow Cytometry and immunodot. There are no vaccines against LCDV which makes necessary the development of tools to control epizootics caused by this virus. It is especially important to detect LCDV before the appearance of tumor-like lesions.

### Diseases caused by bacteria

Fish diseases of bacterial origin have been one of the most important factors of economic loss since the beginning of marine fish culture. Regarding the infectious diseases caused by bacteria in marine fish, although pathogenic species have been described in the majority of the existing taxonomic groups, only relatively small number are responsible for important economic losses in cultured fish. Clinical signs (external and internal) caused by each pathogen are dependent on the host species, age and stage of the disease.

### Vibriosis:

Vibriosis is a disease characterized by haemorrhagic septicaemia and caused by various species of *Vibrio*. It occurs in cultured and wild marine fish in salt or brackish water, particularly in shallow waters during late summer. Within the *Vibrionaceae*, the species causing the most economically serious diseases in



marine culture are *Vibrio anguillarum*, *V. alginolyticus*, *V. ordalii*, *V. salmonicida* and *V. vulnificus*.

Vibriosis has become the economically most important disease in marine fish culture, affecting a large number of species. It is also an important disease of many wild fish populations. Fish affected by vibriosis show typical signs of a generalized septicaemia with haemorrhage on the base of fins, ulcers on body surface (Fig.1), swelling and boils, exophthalmia and corneal opacity. Moribund fish are frequently anorexic with pale gills which reflects a severe anaemia. Oedematous lesions, predominantly centered on the hypodermis, are often observed. On the top of the boils, the epidermis is destroyed and the skin is greyish white. Around the boil the skin is haemorrhaged. Internally there are haemorrhage in liver and intestine, and there is fluid in the heart lumen. Histologically the muscle fibres are widely separated.

*Vibrio* species responsible for vibriosis can be presumptively diagnosed on basis of standard biochemical tests. However, serological confirmation employing serotype-specific polyclonal antisera is necessary. Although commercial diagnostic kits based on slide agglutination or ELISA have been developed for a fast diagnosis of vibriosis, they do not allow the distinction of serotypes and therefore are not useful for epidemiological purposes. Though DNA probe-based detection protocols are available, they are not specific and/or sensitive enough to be used in the diagnosis of vibriosis in the field.



Fig.1. Ulceration in Vibriosis in Asian seabass

### Streptococcal infection:

Streptococcal infection of fish is considered as re-emerging disease affecting a variety of wild and cultured fish throughout the world. Five different species are considered to be of significance as fish pathogens: *Lactococcus garvieae*, *L. piscium*, *Streptococcus iniae*, *S. agalactiae*, *S. parauberis* and *Vagococcus salmoninarum*. Therefore, streptococcosis of fish should be regarded as a complex of similar diseases caused by different genera and species capable of inducing a central nervous damage characterised by suppurative exophthalmia and meningoencephalitis. Warm water streptococcosis typically involves *L. garvieae*, *S. iniae*, *S. agalactiae* and *S. parauberis*. It is important to report that the etiological agents of warm water streptococcosis are considered also as potential zoonotic agents capable to cause disease in humans. Among these fish streptococci, *L. garvieae*, *S. iniae* and *S. parauberis* can be regarded as the main etiological agents causing diseases in marine aquaculture. *S. iniae* was isolated from marine fish including European and Asian seabass in Australia. Streptococcus infection can be diagnosed by biochemical tests.

### Gaffkemia (Red tail disease):

Gaffkemia is one of the most important and well described infectious diseases of lobsters, primarily as a disease of impounded lobsters. The bacterium and resulting disease were first described in a holding facility in Maine. The causative agent, *Aerococcus viridans* (*var.*) *homari* is a free living, gram positive, tetrad-forming coccus (Fig.2), leading to systemic disease in homarid lobsters. The bacterium cannot cross intact lobster integument and does not survive gastric acids; consequently invasion of the host occurs via wounds or punctures of the cuticle. The organism survives well in the benthic environment and in holding facilities, as a free living organism outside the host. Development of the disease is temperature dependent, with death occurring in 180 days at 3 °C and 2 days at 20 °C. Stressors which predispose the lobsters to disease include strain of bacterium, handling, temperature changes, and trauma. Homarid lobsters are most susceptible to infection and disease. Clinically, there are no obvious signs in



infected lobsters, other than weakness or lethargy and a spread-eagle posture which are apparent in later stages of the disease, and which are not pathognomonic for gaffkemia. Lobsters rapidly become anorexic after infection. The bacteria multiply rapidly in the hepatopancreas and then in the heart. The pathogen multiplies in the hemolymph much later in the infection. Pink discoloration of the ventral abdomen and hemolymph will develop. Death results from metabolic incapacity resulting from dysfunction of the hepatopancreas. Additionally, the clotting mechanism is impaired, and it is associated with marked hemocytopenia. Infected lobsters can become exsanguinated, especially in end-stage disease.

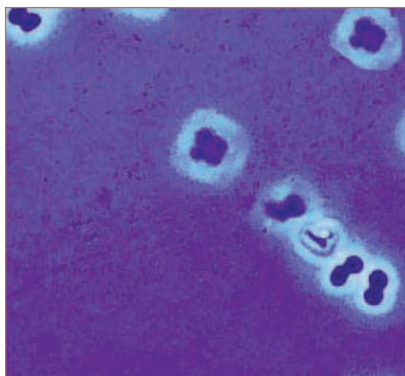


Fig.2. Tetrad forming *Aerococcus viridans*

The disease can be treated with antibiotics after a antibiotic sensitivity test considering other factors. Oxytetracyclin is an antibiotic commonly used in aquaculture. Major challenges associated with use of oxytetracycline in the marine environment include persistence in sediments, bioaccumulation by bivalves or other crustaceans, and development of bacterial resistance. An alternative to chemotherapy is utilization of immunogens derived from virulent strains of *A. viridans*. Low levels of protection were induced with formalin-killed bacteria, and high levels were induced with a vancomycin/live bacteria combination in laboratory experiments.



## Diseases caused by parasites

### Nematodiasis:

Fish are either intermediate or final host for nematode parasites. About 650 species of nematodes parasitize fish as adults and many others use fish as intermediate hosts. While nematodes are common in wild fish, neither adult nor larval nematodes are usually a problem in most cultured fish because of the absence of other hosts in the life cycle. However, pond raised fish or those fed live and wild caught fish/ arthropods (trash fish) can become infested. Marine fish are usually infected by members of the Ascaridoidoidea (*Contracaecum*, *Pseudoterranova*, *Anisakis*), Camallanoidea (*Camallanus*, *Culullanus*), Dracunculoidea (*Philonema*, *Philometra*), and Spiruroidea (*Metabronema*, *Ascarophis*). Most of the camallanoids, dracunculoids, and spiruroids have two host life cycles where fish are the final host. Adult worms are almost always found in the digestive tract, where some (*Capillaria*) can cause chronic wasting if present in high numbers. Some adult nematodes inhabit the peritoneal cavity, gonads or swim bladder.

Faecal examination can be used to identify eggs in the digestive tract. Worms are easily identified as adult or larval nematodes by using wet mounts or tissue sections. The main criteria used to identify species are size, fine structure of the head and tail, position of the excretory pore, and structure of the transitional area between the esophagus and intestine. Anthelmintics can control adult nematodes. Fenbendazole, levamisole and piperazine have been used with some success.

### Disease caused by sea lice:

Sea lice is the term used to describe several species of ectoparasitic copepods of the genera *Lepeophtheirus* and *Caligus* that parasitize cultured fish and may cause diseases with damage to the epidermis and in severe cases death through osmoregulatory failure or secondary infections. *Lepeophtheirus salmonis* and *Caligus elongatus* have economic impact on farmed fishes in many parts of the world.; other caligids pathogenic to cultured or wild



fish are *C. patulus*, *C. orientalis*, *C. epidemicus*, and *Pseudocaligus apodus*. Formaldehyde, malathion and natural compounds show either poor efficacy or unsuitable therapeutic margins. Pyrethroids are at present the most used therapeutic against sea lice in Norway. Diflubenzuron and teflubenzuron added to feed are also used in significant amounts. Carbaryl and diflubenzuron are efficacious but the compounds make them unsuitable due to undesirable environmental toxicological characteristics. Despite these problems, chemotherapy remains an important component of control strategies. As appropriate sea lice control strategies appear to be prevention of cross-contamination by avoiding overlaps of salmon generations in cultures or biological control by stocking locally obtained wrasse (cleaner-fish of the family Labriadae) into cages when salmon first go into the sea cages. Development of vaccines against sea lice is also a perspective area of control of these parasites.

### Cryptocaryon disease

Cryptocaryon disease in the fish, also called the Marine white spot, is caused by the ciliated protozoan parasite. This disease spreads if the quality of water is not good. If the pH level of the water is reduced then, it might lead to the Cryptocaryon outbreak.

One of the signs of Cryptocaryon infection is lethargy in the fish and it might also rub itself against all the objects found in the aquarium. This type of behaviour is obvious if there is Cryptocaryon infection. We can notice white spots (2 mm) on the body and fins. The white spot first appears in the pectoral fins and then spread to the other parts. The gill is one part that has many of these organisms. The white spots spread to the entire body and it might also lead to hemorrhage later. During the advanced stages of this infection the eyes of the fishes would be clouded which might cause blindness and lead to other diseases like fungal infections which adds to the already existing problem.

Since Cryptocaryon is a parasite it will need a host for its development. During the initial stage the Cryptocaryon will be free swimming and it will try to find a host for its development.

Once it finds a host it will penetrate the skin of the host i.e. the fish. The free swimming stage of *Cryptocaryon* is called tomite and the parasitic stage is called trophont. It feeds on the tissues of the fish and grows. The size of the parasite double every single day and it is visible to the naked eye after two days. After about 4 days a cyst is formed which will give rise to another 100 to 300 tomites. It is only about 5 to 10 percent of the tomites succeeds in finding another host for its development. Every week you can find the population of *Cryptocaryon* increases ten times. Due to this rapid increase the effect of the infection will also increase dramatically.

Acting at the earlier stage of detection of this parasite will be helpful in eradicating this infection altogether. Dangerous levels of the infections are reached within 12 days of the infection. So it is better to start the treatment at the earlier stage itself to remove those infections.

In the treatment of *Cryptocaryon*, copper based mediations are useful. The dosage of copper used should be appropriate so that it does not affect the fish. The treatment using copper should be done for several times daily. Even after removal of the parasite the treatment has to be continued for about a week so that you remove all the latent tomons too. This is to ensure that the fish is not affected again. Fresh water bath will be tolerated by the fish for about half an hour only. However we have to maintain the pH of this water too.

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### Source:

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