

Chapter 19

Bacterial Fish Diseases and Treatment



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Abstract Fish food is an inevitable item in human consumption with healthy source of good quality proteins and fat. Aqua industry can help to improve food security, livelihoods for the poorest and to meet the world's food demands. But producing more seafood that is at affordable cost with rich nutrition is challenging for aqua industry. Many factors affect the productivity of aqua industry; one such an important constraint is bacterial diseases. Hence, Aqua industry, a booming business sector, immensely requires continued research with scientific and technical developments, and innovations. Study of bacterial fish disease is one such thrust area which requires intense research to understand the causes and control bacterial diseases in fish. The appearance and development of a fish disease is the result of the interaction among pathogen, host and environment. An insight into bacterial fish diseases, clinical symptoms and treatment may help to manage the bacterial diseases and so can make aqua industry a more profitable field. This chapter deals with different aspects of the most threatening bacterial diseases, occurring in farmed fishes and also in wild fishes, which are results in fish loss and economic loss worldwide. A wide range of gram positive and gram-negative bacteria causing bacterial diseases, clinical symptoms, diagnosis, treatment, vaccines and the nature of water habitat are also discussed in this chapter.

Keywords Bacterial fish diseases · Gram-positive bacteria · Gram-negative bacteria · Clinical signs · Diagnosis · Treatments

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“A disease is the sum of the abnormal phenomena displayed by a group of living organisms in association with a specified common characteristics by which they differ from the norms of their species in such a way as to place them at a biological disadvantage”.

(Campbell et al. 1979)

19.1 Introduction

Water is a habitat for enumerable organisms ranging from microbes to blue whales. Many bacterial species dwell in water as saprophytes in sediments, plants, phytoplankton, and zooplankton. Some of them reside on the skin, gills, and digestive tract of fish, and they live there with mutual beneficial. Fish living in different environments like river, brackish, and marine water and which are reared in farms are prone to various communicable infections caused by a phylogenetically different group of bacterial species (Sudheesh et al. 2012). These microorganisms could also spoil and harm fish health and hence generally considered as pathogenic. In the different growth stages of fish, bacterial diseases have been frequently found in eggs, fry, and fingerlings, thus leading to heavy mortality. These microorganisms are typically harmful pathogens that invade the tissues of a fish host susceptible to infection by various stress factors (Ahmed and Kumar 2005).

Intercommunications between fish and bacteria and the infections caused by microorganisms are the field of research throughout the world, and this plays an important role in fish pathology (Pekala-Safińska 2018). Infectious bacterial diseases are one of the major limiting factors to aqua-industry and for economic and socio-economic development in India and including many other countries of the world (Bagum et al. 2013). Bacterial diseases affecting aqua-industry are mainly due to the intensification of industry without knowledge of the host, pathogen, and environmental factors (Bondad-Reantaso et al. 2005).

The outbreaks of diseases either begin with a sudden increase in an infection leading to high mortality (acute disease) or may develop slowly, with moderate severity for a longer period (chronic disease). A total of 15 different common bacterial diseases have been reported. The main diseases covered are vibriosis, pasteurellosis, enteric red mouth (ERM) disease, furunculosis and marine flexibacteriosis. Other significant diseases, with lower reports, include rainbow trout fry syndrome (RTFS), columnaris disease, motile aeromonas septicaemia, pseudomoniasis, streptococcosis, mycobacteriosis, epitheliocystis, and rainbow trout gastrointestinal syndrome (RTGS) (Toranzo 2004). There has been a constant increase in the number of bacterial species associated with fish diseases, with new pathogens regularly recognized in the scientific literature (Austin and Austin 2012).

19.1.1 Aetiology

Water ecosystems are considered to be a major habitat for aquatic animals. Various changes occurring in water ecosystems seem to be a basic factor in the development of disease, including emerging ones (Johnson and Paull 2011). The evolutionary changes that occur in fish infection mainly depend on the climatic conditions that exist in a given territory, region, or country. In addition to climatic conditions that cause fish infection, external factors such as environmental conditions, water contamination, hypoxia, stress, etc. may also lead to infection (Vijayan and Sanil 2012). Many of these bacterial pathogens are omnipresent in the water ecosystem. Stress can compromise the fish's immune system, and cause them to become more susceptible to the infection with bacteria. Stress is the major causative factor that induces infection in fish, which allows them to succumb to death more easily (Romero et al. 2012). The agents of stress are lowered water quality, overpopulation, redundant treatment and transportation, poor nutrition, and other pathogens. The disease condition depends upon the persistence of the stress varying from mild to severe. Before the onset of any clinical symptoms related to infection, there may be evident damage to the host. The isolation of bacteria from a diseased fish is taken as evidence of infection (Stevenson 1978) (Fig. 19.1).

19.1.2 Factors Affecting Aqua-Industry

Aqua-industry requires good quality of water that includes physical, chemical, and biological factors. The chemical parameters found in pond water, such as increased turbidity, temperature, salinity, pH, water conductivity, and low dissolved oxygen (FAO 2018; Jacobs and Chenia 2007; Nadirah et al. 2012), may induce changes within the water grade which greatly affect fish growth, reproduction, and survival. Acclimation helps us to know the cause clearly why healthy fish get sick under the identical condition at another time. For example, fish can remain healthy when the

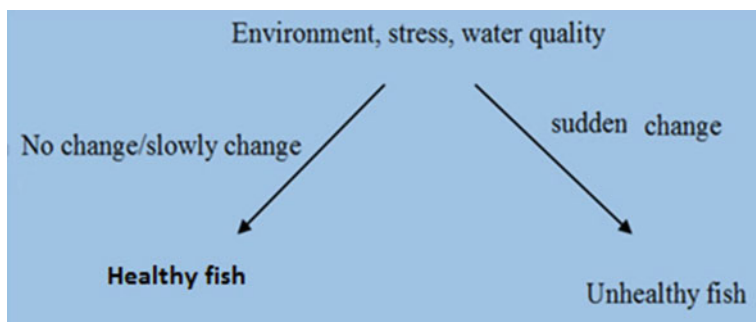
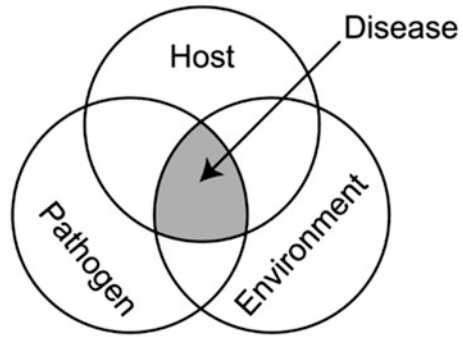


Fig. 19.1 Aetiology of bacterial diseases of fish

Fig. 19.2 Environmental factors and stresses contribute to outbreak of disease in fishes



pH slowly drops from 7.0 to 5.5 over several months, but at the identical time when the pH rapidly gets back to 7.0, the fish may die. The fish can survive in both acidic and basic pH and it is able to tolerate both conditions if they are introduced slowly to the standard environment.

Worldwide there are more than 13 types of bacterial genera that have been reported to cause bacterial diseases in the aqua industry (Pridgeon and Klesius 2012). The genera associated with freshwater bacterial diseases are *Aeromonas*, *Pseudomonas*, *Edwardsiella*, *Flavobacterium*, and *Streptococcus*. Bacteria that cause disease in marine fish are gram-negative. The most commonly associated organisms are genus *Pseudomonas*, *Vibrio*, and *Myxobacteria* (Fig. 19.2).

19.1.3 Types of Diseases

19.1.3.1 Fin Rot

Fin rot is a secondary bacterial infection that is caused by poor water conditions. The major etiological organisms are *Vibrio ichthyodermis* or *Pseudomonas ichthyodermis*. The clinical signs of this infection include frayed or reddened fins and tails. The infection may also lead to total destruction of the fins in extreme cases. In advanced cases it may become a stromal tumour of uncertain malignant potential (STUMPS). Minute haemorrhages and subsequent ulceration of the skin occurs in more serious infections.

19.1.3.2 Vibriosis

It is a type of internal infection caused by gram-negative bacteria *Vibrio*. The highest incidence of infection occurs in larval and juvenile stages of marine species, mainly seabass and seabream varieties (Toranzo et al. 2005). The disease is transmitted through contact with open wounds or stagnant fish, and once a fish is infected, the disease tends to progress quickly. Since vibriosis is an internal infection, fishes

affected by this disease do not show any visible symptoms until the final stages of the disease. The clinical signs with some visible symptoms of this infection include red stripes on the body, red blemishes, dark swollen wounds, and cloudy eyes. Sometimes certain behavioural changes also occur with the symptoms like lethargy, loss of appetite, and difficulty in breathing. The most efficacious therapy for this infection is oral antimicrobial drugs such as kanamycin.

19.1.3.3 Tuberculosis

Mycobacterium species can infect mammals, birds, and reptiles. Some of its species will cause a localized infection, while others cause systemic disease. Mycobacterium that affects fish is called “fish tuberculosis” or wasting disease”. Fish tuberculosis is more prevalent in marine fish species than in freshwater tropical fish. Fin erosion, ulceration on the body, loss of appetite, erythema of the skin, and apathy are the major clinical signs. The disease can be treated with antibacterial medicines which are effective in mild cases. Both vibriosis and fish tuberculosis can be communicated easily to humans through open injuries or sores. Careful handling is necessary while treating infected fish.

19.1.3.4 Photobacteriosis

Photobacteriosis is caused by *Photobacterium damsela* sub sp. *piscicida*, a halophilic member of the Vibrionaceae family. The disease was reported in several Mediterranean countries, the United States and Japan. Transmission of this disease is through the ovarian and seminal fluids from healthy brood stock. The bacterium is able to infect its host through the gills, gastrointestinal tract, and possibly through the skin. Young fish are more susceptible to the pathogenic organism (Toranzo et al. 2005). Seasonal changes that take place in water temperature are responsible for the epidemiology of the disease. At temperatures, lower than 21 °C mortality may decrease but fish become disease carriers (Magarinos et al. 2001). The infected fishes may develop surface darkening, and in several cases, a small red spot appears on the head, gills, operculum, and fin bases due to bleeding into the skin. The infection progresses quickly into acute septicaemia with splenomegaly.

Spleen is the organ of choice for isolation. Characterization and identification of pathogens is carried out using tryptic soy agar (TSA), brain heart infusion agar (BHIA), or blood agar, supplemented with 1–2% NaCl. In advanced cases, typical foci of bacterial microcolonies and severely compromised tissue appear as whitish spots and patches on the spleen surface. Histologically, multifocal necrosis is seen in spleen and liver tissues (Toranzo et al. 1991).

19.1.3.5 Edwardsiellosis

Edwardsiellosis is a systemic disease that affects freshwater and marine water fishes. The causative organism *Edwardsiella tarda* is a gram-negative, motile rod with peritrichous flagella and it belongs to the classification of Enterobacteriaceae. *Edwardsiella tarda* has a worldwide distribution and can be found in the intestines of fish and other marine animals. It is a primary opportunistic enteric bacteria that can survive in phagocytes (Srinivasa Rao et al. 2003). Clinical symptoms of edwardsiellosis differ from species to species of infected fish which may include discolouration of the skin resulting from bleeding underneath, caused by bruising. Ulceration, fin and tail erosion, and, occasionally, exophthalmia and cataracts are observed. The gills appear pale and inflamed. Haemorrhage occurs in internal organs oedematous. The kidney appears enlarged. Whitish granulomatous-like lesions, often visible on their surface, may gradually liquefy into large abscesses and spread into the surrounding musculature.

19.1.3.6 Flexi Bacteriosis

This disease is caused by *Tenacibaculum maritimum* (Flavobacteriaceae) which is an opportunistic gram-negative bacterium commonly found in seawater (Salati and Cubadda 2005) formerly known as *Cytophagamarina* or *Flexibacter marinus* or *maritimus*. The disease is called by various names like “gliding bacterial disease”, “eroded mouth syndrome”, and “black patch necrosis”. Any changes in climatic conditions or fluctuations in water temperature, environmental stressors may trigger the development of the disease (Toranzo et al. 2005). The infected fish develops collapsed and bleeding mouth, fins and tail appear shredded and centres of gills may decay. Several lesions may also occur in the skin. The disease can also infect internal organs of infected fish (Toranzo et al. 2005). Skin and fin abrasions (e.g. following netting or rough handling during grading procedures in nurseries, lesions as a result of feeding activities by parasitic flukes on gills or isopods on the skin) are the major clinical signs that occur quickly.

19.1.4 Classification of Bacterial Fish Diseases

The flowchart (Fig. 19.3) illustrates the classification of fish diseases based on the type of bacteria and nature of water habitat.

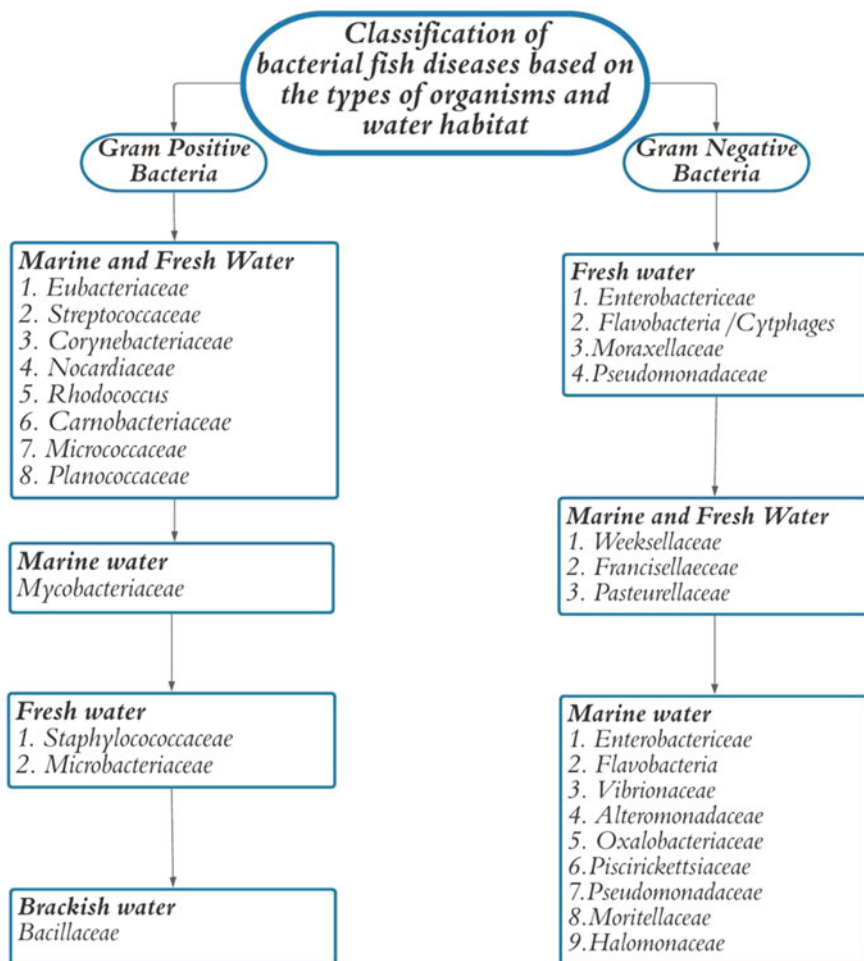


Fig. 19.3 Classification of bacterial diseases based on organism and nature of water habitat

19.2 Gram-Positive Bacteria

19.2.1 Bacterial Diseases of Marine and Freshwater Fishes

19.2.1.1 Eubacteriaceae Representative

Eubacterium tarantellae species has been classified as the anaerobic gram-positive bacteria which have been found to cause a neurological disease known as “Eubacterial meningitis” in marine and river (freshwater) fishes (Austin 2011).

Clinical Signs

Fishes affected by *Eubacterium tarantellae* show twirling in water, darkened pigmentation, uncoordinated swimming movements, and inability to orient themselves which may lead to death of fish. Gram-positive, filamentous anaerobic bacteria are identified by examining the brain tissue smears from the infected fish. Fish deaths due to meningitis caused by gram-positive anaerobic bacteria have been observed in several species of fishes like striped mullet (*Mugil cephalus*), snook (*Centropomus undecimalis*), and gulf flounder (*paralichthys albigutta*) (Austin 2011).

Diagnosis

Gram-positive, filamentous anaerobic bacteria identified by examining the brain tissue smears from the infected fish. To confirm the infection of filamentous and gram-positive anaerobic bacteria, the brain tissue of the fish is examined under a microscope. For diagnostic purposes, isolation and analysis of the causative agent or a direct fluorescent antibody test is carried out. The organism is further characterized by the production of haemolysins and lecithinase and also detecting the presence of lactate acetate in the fermented products.

Treatment

Treatment involves chloramphenicol, erythromycin, novobiocin, penicillin, and tetracycline which is given orally at a dosage of 100 mg/kg per day for 5 days.

19.2.1.2 Streptococcaceae Representative

The gram-positive cocci belonging to the genera *Streptococcus* and *Lactococcus* are considered as the important fish pathogens all over the world. There are several different species of gram-positive cocci, including *Lactococcus garvieae*, *Streptococcus difficilis*, *Streptococcus iniae*, *Streptococcus. parauberis*, *Streptococcus phocae*, *Lactococcus piscium*, *Vagococcus salmoninarum*, and *Carnobacterium piscicola*. The disease caused by *Enterococcus seriolicida*/*Lactococcus garvieae* is called lactococcus or enterococcal infection or enterococcus or streptococcosis (Mata et al. 2004).

Streptococcosis is a “septicaemic disease” that infects both freshwater and sea-water fishes as well as warm- and cold-water fishes. The disease is also known as red boil disease which has been first reported in rainbow trout (*Salmo gairdneri*). Red boil disease mainly affects Golden shiners, Japanese eels, Rainbow trout, Yellow tails, Striped mullet (*Mugil cephalus*) and Hardhead catfish (*Arius felis*). *Lactococcus garvieae* does not affect the internal organs of Golden shiners but produces skin lesions on the body surface. Severe damage is found in the spleen,

intestine, and liver with the accumulation of ascitic fluid in the peritoneal cavity (Mata et al. 2004).

Clinical Signs

The signs of the disease include lethargy, inappetence, swollen abdomen, stomach and intestine filled with yellowish fluid, and in some fish slight haemorrhaging in the eye in *Streptococcus difficilis* infection. Septicaemia along with the brain damage including meningitis occurs in *Streptococcus iniae* infection. Discolouration, loss of orientation, corneal opacity, and internal and surface haemorrhaging lead to death by *Streptococcus iniae* serotype II infection. During summer, weight loss, haemorrhage on the anal, abdomen and pectoral part, pale liver, and congested kidney develop in *Streptococcus parauberis* infection. *Enterococcus* and *Lactococcus* are characterized by haemorrhagic septicaemia. Diseased fish show typical signs of lethargy, anorexia, loss of orientation, and bilateral exophthalmos. Other signs are petechiae on the inside wall of the operculum and congestions of blood vessels in the pectoral and caudal fin (Mishra et al. 2018).

Diagnosis

Enterococcus and *Lactococcus* species are diagnosed by gross appearance which includes haemorrhage of liver, kidney, brain, intestine and spleen. The haematocrit values are significantly lower than that of healthy fish. The above characteristic signs and symptoms indicate the presence of gram-positive cocci in the internal organs and are further confirmed by isolation and determination of the cultural characteristics of *Streptococcus*.

Treatment

The infection can be treated by using antibiotic sodium nifurstyrenate 50 mg/kg/ day which can control streptococcal infection in yellow tails. Oxytetracycline and chloramphenicol at 50 mg/gallon can treat streptococcal infection in golden shiners. Erythromycin at the rate of 25 mg/kg of fish for 4–7 days is used to control the streptococcal infections in yellow tails, which will give better results than daily treatments of Oxytetracycline or ampicillin. Oral administration of peptidoglycan seems to increase the resistance of disease in yellow tails. Recently the use of probiotic bacteria has shown good results in controlling *Enterococcus seriolicida* and *Lactococcus garvieae* infection in rainbow trout.

19.2.1.3 *Corynebacteriaceae* Representative

Corynebacterium is responsible for “bacterial kidney disease (BKD)” which is a systemic infection that causes high mortality among feral and farmed salmonid fish dwelling in river water and marine water. The disease is chronic but sometimes there may be acute outbreaks. The disease was first reported in the United States by Belding and Merrill in 1935. The pathogen is small, non-motile, a sporogenous gram-positive diplobacillus. Based on morphology, the organism was classified under *Corynebacterium* or *Renibacterium salmoninarum*. BKD infects progressively because the pathogen is very slow-growing. It is also called Dee disease, White boil disease, Salmonid kidney disease, or *Corynebacterial* kidney disease (Eissa et al. 2006).

Clinical Signs

The pathogen can be transferred from one fish to another fish and also from adults to their progeny via eggs. Infected fish take a few months to show the signs of disease, so BKD is one of the most difficult diseases to treat. Exophthalmia and small closed blebs or open lateral lesions are typical external signs. The lesions may form large shallow ulcers. Internally, the kidney is the most affected organ by this disease and it also has a swollen abdomen. In some cases, haematopoietic, excretory functions are affected and the kidney is also destroyed.

Diagnosis

The typical signs of the disease and gram-positive Diplobacilli infected tissue which constitutes the presumptive diagnosis. Acute and rapid serological diagnosis is through immunodiffusion-specific antisera and definitive diagnosis is through the lesions in the kidney of the infected fish. In addition, Coagulation test, Indirect and direct fluorescent antibody tests have been developed for rapid diagnosis. Diagnosis can be made within 10 min by direct and within 2 h by indirect fluorescent antibody tests.

Treatment

Erythromycin is given orally at the rate of 9–10 g/kg/day for 3 weeks which will provide partial control over the disease. Sulphonamides fed at 4.2 g/100 kg/day have been successfully used for prophylaxis in salmon. Since BKD is a transmitted disease, avoidance is the most preventive way to treat the fish and prevent transmission by way of the eggs (Hirvela-Koski 2005).

19.2.1.4 Nocardiaceae Representatives

Nocardia Species

Nocardia species are gram-positive, catalase-positive and rod-shaped bacteria that infect river and marine fishes to cause Nocardiosis. It is a long-term and problematic bacterial infection of warm-water fish. The disease finally acts as a major weakening factor in many types of fish. The infected fish often have other secondary infections also. Nocardiosis embarks as a silent infection, which can be unidentifiable for months in fry or juvenile fish. The infection is chronic and the species multiply slowly within fish tissues before any visual symptoms. In some cases lethargy and the death rates also increase. The symptoms of nocardiosis are similar to mycobacteriosis (Wilson 2012).

Clinical Signs

Fishes of all age groups may be infected by this disease with symptoms like lesions, exhibited as small white spots, present in the dermis, muscle, gills, and internal organs. The visual symptoms of this disease are diverse. The visible lesions are found in skin nodules and skin, whereas the opercular lesions are found as fleshy white masses at the base of the gills. As nocardiosis is similar to mycobacteriosis, the internal pathology diagnostic procedures are easily confused especially if mixed infections exist.

Diagnosis

Nocardiosis may be misinterpreted as mycobacteriosis, because of the correlation between the clinical symptoms and pathology linked with the two diseases. Isolation, identification, and histological sections of the infected organs are selected for diagnostic purposes. Nocardioform bacteria can grow on similar kinds of media, with an incubation period ranging from 2 to 14 days.

For the preliminary assessment a thorough visual examination of the fish is needed. The skin and body wall was examined for lumps and ulcers. The presence of dry, grey-yellow, inspissated pus under the skin nodules is one of the basic diagnostic confirmations. Also, pale gills will be seen under the operculum and irregular whitish lumps at the base of the filaments. The colony of nocardia appears as folded, granular, or powdery, pinkish-white to yellow-orange or light brown colonies with aerial mycelium around the edges. For diagnosis Gill, kidney, or spleen imprints or stamps collected (in duplicate), dried, and stained using Grams or an acid-fast stain. 5 mm sections of the infected tissues are used for histological diagnosis and must be preserved in 10% buffered formalin.

Nocardial granulomas without the epithelioid cells in the earliest stages of development are confused easily with piscine mycobacteriosis. It can be confirmed furthermore by the presence of acid-fast organisms in the *nocardia* lesion which show a positive reaction for *nocardia* with Fite-faraco acid-fast stain. There are different types of specific and sensitive methods that have been applied recently for diagnosis, including antibody-based and DNA-based methods (Wilson 2012).

Treatment

Antibiotics such as streptomycin and sulphisoxazole have been used for treating Nocardiosis. Treatment with trimethoprim and sulphamethoxazole at 40 mg/l mixed in feed along with the addition of benzalkonium chloride at 2 mg/l in water decrease the mortality rates of the infected fish. Antibiotic doses need to be high and the duration of treatment must be extended for a prolonged period. The application of protective therapies applied to asymptomatic young fish may be beneficial. Restriction through husbandry and good management methods is the best way to prevent nocardial infections.

19.2.1.5 Rhodococcus Representatives

Rhodococcus Erythropolis

Rhodococcus is a genus of aerobic, non-sporulating, non-motile, gram-positive bacteria which is closely related to Mycobacterium and Corynebacterium. Low level mortalities are recorded in fishes infected by Rhodococcus species. Chinook salmon are a type of anadromous fish (they hatch in freshwater streams and rivers then migrate out to the saltwater environment of the ocean to feed and grow) (Olsen et al. 2006a, 2006b).

Clinical Signs

In Canadian farmed chinook salmon, there is a development of melanosis and ocular oedema, leading to a break in the cornea. There was no other evidence of involvement of any internal organs. With Atlantic salmon, the presence of granulomas in the kidneys is found to be apparent (Olsen et al. 2006a, 2006b).

Diagnosis

Characterization and Identification of Pathogens

Two colonies with denser growth are obtained from diseased tissue following inoculation with blood agar, MacConkey agar, and TSA with incubating at 19 °C for 14 days. Both the colonies are from the organs (Kidney and spleen) of chinook salmon with ocular lesions.

Treatment

Glycopeptide antibiotics (vancomycin, teicoplanin), rifampicin, quinolones, aminoglycosides, carbapenems, and macrolides are effective for this infection.

19.2.1.6 Carnobacteriaceae Representative

Carnobacterium Piscicola

Carnobacterium belongs to gram-positive bacteria. Pseudokidney disease is provoked by Carnobacterium piscicola. This organism can be isolated from rainbow trout, cutthroat trout, coho, and salmon. All river and marine water fishes are susceptible to stress conditions. Lactic acid bacteria alone function as probiotics to fishes. Others are pathogenic to fish (Leisner et al. [2007](#)).

Clinical Signs

Visible signs of infection outside the body are abdominal distension, redness at the base of fins, and subdermal blood sores, and within the body, the liver, spleen, and nephrons are enlarged. Accumulation of ascitic fluid in the peritoneal cavity is common. There are also haemorrhages in the reproductive system of the male, intestinal tract, and muscle. A grey pseudomembrane may be present which resembles BKD infections. Other visible signs of infection outside the body like fin rot were absent. Yet, muscle haemorrhage and hyperaemic air bladder are seen. The heart and gills appeared normal. The disease affects fishes that are greater than a year old and have undergone pressure, during handling and spawning. The general signs and symptoms are diverse which comprise septicaemia, the abdomen distended with ascitic fluid, muscle abscesses, blood blisters below the skin, and internal haemorrhaging.

Diagnosis

Characterization and Identification of Pathogens

Diagnosis is by isolating and characterization of the causative organism. Preliminary isolation is from kidney or lesion using tryptone soya agar (TSA) or Brain heart infusion agar (BHIA) grown aerobically at 15–24 °C for 24–72 h.

Presumptive Diagnosis

Colonies appear pinpoint, opaque, entire, circular, and non-pigmented when grown on TSA. The organism is a non-motile, non-spore-forming, non-acid fast, facultatively anaerobic, gram-positive rods or coccobacillus. The size is 1.1 to 1.4 × 0.5 to 0.6 µm. Other phenotypic analysis showed negative results for biochemical analysis like urease, oxidase, catalase, H₂S, nitrite reduction, and lactose and xylose fermentation. The organism shows positive reactions for arginine dihydrolase and lactic acid production from glucose (no gas), maltose, mannitol, and sucrose.

Confirmatory Diagnosis *Carnobacterium piscicola* can be differentiated from the other *Carnobacterium species* by the fermentation patterns using inulin and mannitol (Leisner et al. 2007).

Treatment

Erythromycin is given as treatment for Lactobacillus-Carnobacterium strains (Michel et al. 1986; Baya et al. 1992).

19.2.1.7 Micrococcaceae Representative

Micrococcus leuteus

Micrococcus luteus is a gram-positive, non-motile, coccus, tetrad in arrangement, pigmented, a saprotrophic bacteria in the family Micrococcaceae. Signs and symptoms are related to Rainbow Trout Fry Syndrome.

Clinical Signs

Rainbow trout (marine fish) and brown trout (freshwater fish) fishes commonly are infected by *Micrococcus leuteus*.

Exophthalmia, enlargement of the abdomen, darkening of the skin, petechiae, and focal lesions are the signs seen in moribund fish.

In many fish inflammation of the intestine, liver obstruction, and bleeding in the tail part of the muscles will be present (Peřkala et al. 2018; Austin and Stobie 1992).

Diagnosis

The organisms were cultured by using scrubbed medium (kidney, liver, spleen) pasteurized milk agar at 25 °C for 48–72 h (Austin and Stobie 1992). In some rainbow trout, a disease called micrococciosis infects the fish but the recognition of the etymological agent of this disease is undetermined (Austin and Stobie 1992).

Treatment

Antimicrobial complexes, i.e. streptomycin, chloramphenicol, and tetracycline, are used for the treatment. This complex is used to suppress the development of infections in the rainbow trout griddle (Austin and Stobie 1992).

19.2.1.8 Planococcaceae Representative

Planococcus Species

Planococcus is a gram-positive or gram-variable, cocci or short rod-shaped bacteria which infects marine and freshwater fishes. Motile gram-positive cocci, planococcus, causes rainbow trout fry syndrome (RTFS) or rainbow trout fry anaemia. The organisms have been connected with a compact off-white 12.4 mm in diameter, disk-like concave patch on the heads of Atlantic salmon (Austin 1985).

Clinical Signs

Large rainbow trout fishes weighing 500 g show symptoms like the accumulation of fluid in the kidney and little quantity of ascitic fluid in the abdominal peritoneal cavity. The fishes also show symptoms of anaemia, enlarged kidney, liver and broadened spleen (Austin and Stobie 1992).

Diagnosis

Characterization and Identification of Pathogens

By using BHIA, pasteurized milk agar and TSA, the bacterium can be isolated from kidney scrub and incubated at 25 °C for 7 days. After 48 h of incubation the organism produces off-white to yellow hoist glassy colonies (1–2 mm in diameter) (Austin et al. 1993).

Treatment

Using antimicrobial compounds: The compounds such as carbenecillin, penicillin G, erythromycin and tetracycline are subtle to the organism which may be virtual for chemotherapy (Austin et al. 1993; Austin and Stobie 1992).

19.2.2 Bacterial Diseases of Marine Water Fishes

19.2.2.1 Mycobacteriaceae Representative

Mycobacteria gram-positive, non-motile, acid-fast rods which are extensively spread in nature. They are copious in the soil, on the skin of vertebrates, on the surface of plants, in foods consumed by humans, and mammals. Mycobacterium have so many species, i.e. *Mycobacterium abscessus*, *Mycobacterium marinum*, *Mycobacterium neoaurum*, *Mycobacterium montefiorensis*, *Mycobacterium shottsii*, *Mycobacterium gordonae*, and *Mycobacterium pseudoshottsii*. Some species of mycobacteria are extremely infectious and cause diseases such as tuberculosis and leprosy in humans. Mycobacteria that cause ailments in fishes differ from the causative organism for animals. Mycobacterial infections in fishes are called “mycobacteriosis of fishes”.

Mycobacteriosis is uncommon in freshwater food and recreation fishes. It is more persistent in wild marine fishes. Mycobacteriosis is a major disease in marine water aquarium fishes. It causes a chronic infection. Mycobacterium genus is a member of the order Actinomycetales and family Mycobacteriaceae; fish mycobacteria can grow in several types of media. Identification can be made using several criteria, including acid-fastness, growth rate, pigment production, colonial morphology.

19.2.2.2 Clinical Signs

The disease shows some physical and extreme changes like non-curing, slight to intense deep skin ulcers, corneal ulcers, discolouration, boniness, white nodules on viscera, loss of appetite, and exophthalmos. The internal signs are small greyish tubercles, or nodules in the liver, kidney, and spleen. Outbreaks occur in fishes of freshwater families like Anabantidae, Characidae, and Zebrafish. Previously it affected only salmonids but recently affects other species of cultured food fish such as tilapia, European sea bass, and striped bass. The initial Mycobacteriosis was noted in carp (*Cyprine carpio*). Increased infection is seen in goldfish (*Carassius auratus*).

19.2.2.3 Diagnosis

Characterization and Identification of Pathogens

Diagnosis of mycobacterium disease is relatively easy by using an acid-fast staining method. Lesions caused by these bacteria are rod-shaped, gram-positive bacilli used for routine diagnosis. Tissues can be smeared and stained, but less preferable. To isolate the organism it takes up to 30 days, for confirmation of the organism it takes up to 90 days. In some cases, mixed infection occurs due to several mycobacterium species being involved.

Treatment

Kanamycin is blended with food to effectively cure the disease. It is absorbed from water by fishes. The dosage of the drug is 0.01%. Other antibiotics such as erythromycin, streptomycin, and rifampicin are effective under experimental conditions.

19.2.3 Bacterial Diseases of Freshwater Fishes

19.2.3.1 Staphylococcaceae Representative

The species in the family of staphylococcaceae are *Staphylococcus aureus*, *Staphylococcus epidermidis*, and *Staphylococcus warneri* (gram-positive).

Clinical Signs

The typical signs are corneal redness caused by the formation of new blood vessels which leads to opaqueness as in eye disease. Because of this disease tissues become degenerated, forming a hollow cup. The optic nerves will be affected and in turn the fishes show lethargicness and become dark. The internal organs are not infected. But the infection causes jaundice in African sharp-catfish (*Clarius gariepinus*). Infectious varieties of *Staphylococcus* sp. were first proclaimed from Japan, in yellow tail (*Seriola quinqueradiata*) and red sea bream (*Chrysophrys major*). The signs and symptoms are exophthalmia, congestion, and ulceration on the tail. The infected fish showed haemorrhage on the fins, gills, and opercula, with distended abdomen with ascitic fluid accumulation, and anaemic liver. The species was also isolated from moribund cultural grass carp and juvenile gilthead sea bream. Mortality is around 12% per day at normal water temperature. The species *Staphylococcus warneri* affects the rainbow trout in Turkey with exophthalmia and ulcerations on the fins. Clinical signs are that the abdomen was swollen due to accumulation of ascitic fluid.

The kidney showed normal architecture while the liver showed change in colouration.

Diagnosis

For diagnosis the eye and brain tissue can be examined. The external change in the fish shows that the fishes are affected by the infection.

Treatment

Treatment with erythromycin at a dosage of 20 mg/kg BW/day for 10 days showed good therapeutic function. Drugs like amoxicillin, erythromycin, and trimethoprim sulphamethoxazole are effective in in vivo experiments.

19.2.3.2 Microbacteriaceae Representative

Microbacterium Paraoxydans

Microbacterium belongs to the family Microbacteriaceae. It contains 96 species. From these 96 species, *Microbacterium paraoxydans* is one of the species which cause disease in fish. This bacteria promotes plant growth. The family microbacteriaceae encompasses a large number of aerobic gram-positive bacteria which have high G + C content. This character can be used to differentiate it from other actinobacteria which have both unusual B group cell wall peptidoglycan and unsaturated respiratory menaquinones.

The bacteria show variation in cell morphology like elongation of coccoid shape, rods are small and irregular and show branched fragmenting hyphae (Park et al. 1993; Stackebrandt et al. 1997). Microbacterium species are yellow pigmented; it is found in various environments like soil and water. The organism can be recovered from Nile Tilapia.

Clinical Signs

Lethargy, erratic swimming, melanosis, red or opaque eyes, scale loss, blindness, and exophthalmia are the symptoms included in this disease.

19.2.4 Bacterial Diseases of Brackish Water Fishes

19.2.4.1 Bacillaceae Representatives

Bacillus Species

The initial outbreak of *Bacillus* species disease led to mortality of farmed populations of *Clarias carpis*, *Clarias gariepinis*, *Clarias nigrodigitatus*, *Heteroclaris*, and *Heterobanchus bidorsalis*.

Clinical Signs

Diseased fish showed symptoms of dizziness, apathy, abnormally thin or weak and necrotizing dermal inflammation, death occurring in a few days. The peritoneal cavity consists of blood tinged fluid. Organs like liver and kidney showed evidence of infection like petechiae and focal necrosis. Other signs include splenomegaly with softness and friability. Myocardium also showed softness and flabbiness. The stomach shows an excess of blood supply in its blood vessels. Bacillary necrosis can be seen in farmed populations of catfish (*Panagasius hypophthalmus*). The signs of the disease are presence of white necrotizing and granulomatous areas (1-3 mm diameter) in the kidney, liver, spleen, and other visceral organs.

Bacillus Cereus

Bacillus cereus is an occasional fish pathogen causing bronchial necrosis in common carp and striped bass.

Bacillus Mycoides

Fishes, which are infected with *Bacillus mycoides*, appear darker in colour, lacked appetite, showed ulcers on the dorsal part of the fish, and focal necrosis is seen in the epaxial muscle.

Bacillus Subtilis

Bacillus subtilis is also a fish pathogen, causing bronchial necrosis in common carp.

19.2.4.2 Diagnosis

Characterization and Identification of Pathogens

Isolation of *Bacillus* species can be carried using nutrient agar and incubation can be at high temperature of 37 °C for an unspecified period. Sample for isolation can be taken from tissues with ulcers, tissues from brain, kidney, liver, and necrotic muscle.

It will be inoculated into a range of different media, like sheep blood in agar base, MH agar, and BHIA with incubation at different temperatures for an unspecified duration. The rhizoidal colonies with filamentous, swirling patterns will be seen.

The colonies are 1 mm in diameter, and are cream in colour on TSA medium after 24 hours of incubation at 28 °C. The organisms are gram-variable, slender and motile.

The biochemical characterization showed oxidase-positives, but non-reactive to sugars, and they favour growth at 15 °C to 37 °C, produce positive for hydrogen sulphide and break down gelatin.

19.2.4.3 Treatment

Bacillus species are sensitive to tetracycline but not penicillin. *Bacillus mycoides* show sensitivity to erythromycin, nalidixic acid, nitrofurazone, novobiocin, and oxytetracycline but not to ormetoprim-sulphadimethoxine.

19.3 Gram-Negative Bacteria

19.3.1 Bacterial Diseases of Freshwater Fishes

19.3.1.1 Enterobacteriaceae Representatives

Edwardsiella ictaluri

Enteric septicaemia is seen among young catfish when the temperature is lower, which is during the month of September and October. *Edwardsiella ictaluri* infection mainly affects river water fish *Pangasius hypophthalmus*. The infected fish may swing enervated at a vertical position at the water surface just right before death (Crumlish et al. 2002).

Clinical Signs

Exterior signs are haemorrhages on the throat and mouth, exophthalmia and open sores on the head, especially on the frontal bone of the skull (Hawke 1979). This

disease is also called as Bacillary necrosis of Pangasius because of the abrasion on the kidney, liver, and spleen (Crumlish et al. 2002).

19.3.1.2 Diagnosis

Characterization and Identification of Pathogens

From the infected fish kidney, liver, spleen, intestine brain, or muscle abrasion are isolated, then inoculated into Brain-Heart Infusion Agar. Then incubated at 26 °C for 48 h. The colonies are circular (2 mm), convex without pigmentation (Hawke 1979). The clear diagnostic approach is attained by the plasmid profiles for *Edwardsiella ictaluri* (Lobb and Rhoades 1987; Speyerer and Boyle 1987).

The serological identification of *Edwardsiella ictaluri* can be achieved by monoclonal antibodies of IIFAT (indirect immunofluorescence antibody test) as an immunoassay enzyme (Rogers 1981; Ainsworth et al. 1986). The use of rapid iFAT (immunofluorescence antibody test) at the same time can recognize two pathogens, i.e. *Flavobacterium columnare* and *Edwardsiella ictaluri* using two fluorochromes which have two different spectra properties (Alexa Fluor 488 discharge green while 594 discharge red fluorescence).

19.3.1.3 Treatment

Vaccination with multivalent *Edwardsiella ictaluri* antigens showed development of sensitive and resistant antibodies and T lymphocytes in channel catfish (Camp et al. 2000).

19.3.1.4 Preparation of Vaccine

Lipopolysaccharide (LPS) extract indicates secured action followed by injection (Saeed and Plump 1987). The defence action is consulted by multiple injections by using FCA with balanced LPC (liver progenitor cell) antigenicity. The systematic vaccine was achieved by engrossment of channel catfish at limited death rates (Plumb and Vinitnantharat 1993). The systematic vaccine results in 96.7% mortality in the control group, while the immerse vaccination decreases mortality to 6.7% and the immerse oral vaccination decreases to 3%.

19.3.1.5 Flavobacteria/Cytophages Representatives

Flavobacterium Species

Flavobacterium is a gram-negative, non-motile rod-shaped bacteria. *Flavobacteria* are found in soil and fresh water. Several species are known to cause disease in freshwater fish.

19.3.1.6 Clinical Signs

Epithelial swelling in gills is noticed and distal tip of adjacent Gill lamellae is fused. In some cases *Flavobacterium* sp. is associated with hyperplasia (Kudo and Kimura 1983a, 1983b).

19.3.1.7 Diagnosis

Characterization and Identification of Pathogens

The organism can be isolated using TSA supplements with Sodium chloride (0.5%-3.0%). Yellow-orange colonies will be formed after 24 h. They are gram-negative, motile, and pleomorphic; arginine dehydrolase and oxidase will be produced. But H₂S, indole and β-galactosidase will not be produced. The Voges Proskauer results are positive (Acuigrup 1980). Several acids are produced by Arabinose, Inositol, Mannitol, and Melibiose.

Diagnosis

Flavobacterium has been diagnosed by the API20E Rapid Identification System (Acuigrup 1980).

Treatment

Oxytetracyclines are considered for the treatment of infected fish (Acuigrup 1980).

19.3.1.8 Flavobacterium Columnare

Columnare was first identified in a wide variety of freshwater fish comprising black bullheads, chub, eel, bass, goldfish, rainbow trout, salmon, sheath fish, white grappie, Chinook salmon and white suckers. Diseases caused by *Flavobacterium*

columnare are seen in various stages. In young fish, there is an inconsequential pathology that occurs in the gills, which causes obstruction of blood vessels decoupled on the epithelium of lamellae. In adult fish, it causes abrasion in the gills or in musculature. On the body, at the end of the dorsal and pelvic fin, small abrasions begin, due to discolouration. The infected areas grow in size up to 3–4 cm in diameter and more than 25% of the total surface area of fish. For the identification of *Flavobacterium columnare* pathogen, Bootsma and Clerx's medium and cytophaga agar are prepared and used. As a result, the pathogen gives rise to yellow-orange pigmented colonies.

Diagnosis

Chemotaxonomic methods consist of whole cell body acids and commercialized design with dominant fatty acids is used for diagnosis.

19.3.1.9 Treatment

Vaccine Development

When heat-destroyed cells are used for vaccination to juvenile coho salmon via food, it leads to the production of antibodies (Fujihara and Nakatani (1971)). In channel catfish, agglutinating antibodies are used as vaccines (Schachte 1978).

Management Techniques

To prevent the fishes from this disease is to keep the water temperature as cool as possible because at high temperature the condition of the columnaris is more severe. There was a high level of expansion and demolition at high virulent culture resulting in the destruction of gill tissue towards the base, which frequently occurs in Carp and Rainbow Trout.

Antimicrobial Compounds

NaCl (sodium chloride) baths are used for treating infections (Farkas et al. 1980). Other than sodium chloride, arsenic, cadmium, selenium mixture, copper, and lead at 1–3 µg/l for 1 day are also used (Macfarlane et al. 1986).

19.3.1.10 *Flavobacterium Johnsoniae* (Cytophaga Johnsoniae)

This disease was first identified among the farmed freshwater barramundi and Lattes Calcarifer in Queensland, Australia. Over a 2-week period 2–5% of mortalities were noted at 27–28 °C. The affected fish expresses the signs of superficial erosion of skin and pectoral fins in the lower jaws (Carson et al. 1993). *Flavobacterium johnsoniae* reacquired from a variety of infected fish like, Skin ulcers, Gill necrosis, and systemic disease (Flemming et al. 2007).

19.3.1.11 Diagnosis

Characterization and Identification of Pathogens

The pathogen can be isolated on Cytophaga agar medium with the incubation period for 7 days at 27 °C (Carson et al. 1993). From phenotypic experiments *Flavobacterium johnsoniae* can be identified (Rintamäki-Kinnunen et al. 1997). These are gram-negative bacteria with gliding filaments with yellow colour colonies at 10–30 °C. These colonies produce β -galactosidase, catalase, and oxidase. Nitrogen is utilized from asparagine, potassium nitrate, and ammonium, and acids are produced from glucose.

19.3.1.12 Treatment

Vaccine Development

From the Streptomycin resistance strain, a vaccine was developed and used for the treatment. Grass carp is protected by this vaccine method. Accordingly, I.P used for 28 days gives the best results in diseased fish (Li et al. 2015).

Antimicrobial Compounds

Effective compounds used for the treatment are acriflavine and oxolinic acid (Carson et al. 1993).

19.3.1.13 Moraxellaceae Representative

Acinetobacter Species

Acinetobacter is a gram-negative bacteria pertaining to Gammaproteobacteria. They give oxidase-negative, show convulsive movement and are present in pairs.

Schizothorax is a cyprinid fish found in south and central Asia. They are primarily found in rivers, streams and lakes. *Acinetobacter lwoffii* infection has been identified among the Atlantic Salmon and cyprinid fish in a group of 60 sexually matured in which the temperature was between 8 and 11 °C through the autumn 1978. Through the event of this disease, the fish which was 5–12 kg in weight were wild reserves from Surma and Norway. After the fifth week of disease, the aggregated transience numbered 92% of the total population.

Clinical Signs

Through, the clinical symptoms were visible only among 40% of the animals consisting of hyperaemia of blood vessels, haemorrhaging with oedema expanded from the base of the fins. Due to this, ulcers were developed, abrasions seen in kidney and liver, and little haemorrhages present in the air bladder (Roald and Hastein 1980).

19.3.1.14 Treatment

Antimicrobial Drugs

Oxytetracycline seems to be implied for chemotherapy (100 mg/fish), when delivered by intramuscular injection.

19.3.1.15 Moraxella Species

Moraxella is a gram-negative bacteria. The transience of this disease resulted in the Potomac River, Maryland, USA, among the juvenile, *Morone saxatilis*, through winter 1987.

Clinical Signs

This disease causes the gills of the fish to be damaged with *Trichodina* and *Ergasilus*. The affected fish shows symptoms like huge haemorrhagic abrasions and misplaced scales on the dorsal surface. The liver became bulge, pale and blotched in emergence. Haemorrhages were found to be present in the swim bladder. The layer substances emerge to attach the body wall with the liver (Baya et al. 1992).

Diagnosis

Characterization and Identification of Pathogens

Growth of round, elevated, luminous and sludge colonies grow at 22 °C for 48 h isolated from kidney, pancreas and liver on TSA medium (Baya et al. 1992). Considering that comparison to *Moraxella* bacteria was distinguished (Juni and Bøvre 2005), the organisms heavily look like the *Acinetobacter*. (Roald and Hastein 1980).

Treatment

Therapeutic Drugs for Microbes

The drugs Chloramphenicol, oxolinic acid, tetracycline, nitrofurantoin, and penicillin were effective against this bacterial species (Baya et al. 1992).

19.3.1.16 Pseudomonadaceae Representatives

Pseudomonas fluorescens

Pseudomonas fluorescens was first identified in the fresh water. This organism is identified in fish spoilages (Shewan et al. 1960). And also this organism primarily causes the damages in fish tissues (Otte 1963). However, the pathogenic capability was inferior. Many wide range of fish species get infected by this organism, including goldfish (*Carassius auratus*) (Bullock 1965), grass carp and black carp (Bauer et al. 1973), rainbow trout (Sakai et al. 1989), tench (Ahne et al. 1982), silver carp and bighead. The infected places were found to be eroded and this organism is mainly associated with tail rot or fin rot (Schäperclaus 1979). It has been reported to 90% of high mortalities recorded in tench fry.

Clinical Signs

Clinical signs have been visualized at the base of the skin, haemorrhagic lesions were noted. In the peritoneal cavity, ascitic fluid was accumulated. In gills, liver, kidney, and lumen and submucosa of the gut, petechial haemorrhages were noticed. Silver carp and bighead have shown the signs of stress and they were triggered by disease (Markovic et al. 1996). Ulcers and haemorrhage were reported in rainbow trout on the gills and fins (Sakai et al. 1989).

19.3.1.17 Diagnosis

Characterization and Identification of Pathogens

These organisms are gram-negative cultures which consist of oxidative rods that are produced by catalases and oxidases. And it exhibits motility by polar flagella. From thiositol, maltose, sorbital, xylose, arabinose, mannitol and sucrose, trehalose acids were produced. Citrate has become utilized. The Voges Proskauer test results were negative. Cultures were grown at 4 °C with the fluorescent pigments.

19.3.1.18 Treatment

On examination, isolates displayed organisms vulnerable to nalidixic acid (quinolone synthetic antibiotics), tetracycline, and kanamycin. These antibiotics prevent the bacterial cells from dividing and repairing, thereby killing them (Sakai et al. 1989). The next examination described extensive vulnerability to neomycin, gentamycin, kanamycin, amikacin, oxytetracycline, complete version of penicillin, erythromycin, chloramphenicol, and sulpha drugs (Markovic et al. 1996).

19.3.2 *Bacterial Diseases of Freshwater and Marine Water Fishes*

19.3.2.1 Weeksellaceae Representatives

19.3.2.2 *Chryseobacterium Scophthalmum* (Flavobacterium Scophthalmum)

These are gram-negative, uniformly rod-shaped organisms which form orange-pigmented colonies. Both freshwater and marine water turbot kind of fish are infected by this disease.

Clinical Signs

The disease causes gill hyperplasia, haemorrhaging in gills and lethargy and extensive internal haemorrhaging and distended abdomen in Turbot (Mudarris and Austin 1989, 1992).

Diagnosis

Characterization and Identification of Pathogens

The pathogenic bacteria was isolated from infected organs using K media (Mudarris and Austin 1989). The bacterial colonies produce oxidase and catalase. From cellobiose and glucose, acids are produced. The Voges Proskauer test shows negative. Casein, tributyrin, aesculin, and gelatin have been deteriorated. However, chitin or starch was not degraded. Thin sections of the colonies exhibit the presence of thick cell envelopes (Mudarris et al. 1994).

Treatment

Antimicrobial Compounds

Mortalities were controlled by using 50 mg/l immersion for 30 min daily for 10 days or furazolidone was used by i.p. injections with an average dosage of 50 mg/kg.

19.3.2.3 Pasteurellaceae Representative

Pasteurella Skyensis

The Pasteurellosis disease is caused by *Pasteurella skyensis*, a gram-negative, non-motile, rod-shaped, and pleomorphic bacteria which were isolated by culture on seawater blood agar. Pasteurellosis in Atlantic salmon can cause symptoms such as boils in skin, at the pectoral fin and internal organs and in some cases so-called “blood eye”. The reproduction of Atlantic salmon occurs in rivers and then on maturation migrate to ocean.

19.3.2.4 Diagnosis

Characterization and Identification of Pathogens

Cultures were isolated from the kidney of infected fishes by using TSA culture medium supplemented with 15% of sea salts and 5% with defibrinated horse blood and incubated for 48 h at 20 ° C. Small grey colonies were formed (Birkbeck et al. 2002). Esterase, indole, lysine, oxidase (weakly), leucine arylamidase, ornithine decarboxylase, and naphthol-ASBI-phosphohydrolase are produced by gram-negative catalase-negative rods. Several acids are produced from carbohydrates but not from sucrose, galactose, raffinose, and rhamnose. It also has a weak haemolytic activity. The analysis of 16S rRNA sequencing of pathogen phenotypically, associated with *Pasteurella phocoenarum* family with 97% of homology. It clearly confirmed that it is closely related as phylogenetically (Birkbeck et al. 2002).

Treatment

Good sanitation and management procedures should be used to avoid overcrowding and other stresses that may predispose fish to disease. Prophylactic chemotherapy with sulphonamides, nitrofurans, or antibiotics has been employed successfully. Sulphonamides at 200–400 mg per kilogram of body weight per day or chloramphenicol at 20–40 mg per kilogram of body weight per day, both fed for a minimum of 6 days, are used to control outbreaks (Matsusato 1975). In vitro studies by Kusuda and Inoue (1976) showed that the antibacterial activity of ampicillin was 8–16 times that of chloramphenicol. They suggested that ampicillin would be useful in treatment of *Pasteurella septicaemia*. Vaccine is under trial and proposed to be completed in the later of year 2020.

19.3.2.5 Francisellaceae Representative

Francisellosis is a disease caused by bacterial species belonging to genus *Francisella* infecting a wide range of animals especially fishes. Francisellosis in fish is characterized by the development of non-specific granulomas in spleen and kidney. This granulomatous infection is mainly caused by *Francisella noatunensis* and *Francisella noatunensis subsp. orientalis*. These are facultative intracellular gram-negative pathogens. TM agar (Soto et al. 2014) and modified cysteine heart agar are used for isolation (Lewisch et al. 2014). During the isolation process the cell culture was grown at 20 °C. Small opaque colonies were formed which then extended in diameter to 2–3 mm (Olsen et al. 2006a, 2006b).

19.3.2.6 Francisella Noatunensis

This is aerobic, non-motile, coccobacilli gram-negative bacteria. *F. noatunensis* bacteria infect fish and results in bacterial francisellosis disease. This bacteria is generally present in freshwater and saltwater ecosystems. The disease was first found in tilapia fry with the homology sequence of pathogens. *Francisella noatunensis* infection was first identified in wild and farmed Atlantic cod in Norway but piscine host range keeps on increasing, so *Francisella* culture should be given priority in routine diagnosis.

Clinical Signs

The infected fish will swim inactive and inappetance and also emaciated. Many fish showed exposed dermal haemorrhagic nodules and corneal opacity. The intestinal mucosa becomes thickened in infected fish. White nodular formations were diagnosed in renal organs, heart, spleen and hepatic tissues along with bloody ascites (Olsen et al. 2006a, 2006b). Chronic inflammatory granulomatous infected fish

consists of a huge number of intracellular gram-negative cocco -bacilli which belong to the Francisella family that is associated with Francisellosis (Zerihun et al. 2011). The white granulomas mainly occurred in marine species of three-line grunts in Japan. By using PCR the small subunits of rRNA were amplified and sequenced. Francisella-like bacteria were found in freshwater reared in hybrid striped bass with the water temperature at 20–29 ° C. Bacterial affected fishes behaved lethargically, skin haemorrhage occurred over the abdomen and darker pigmentation was observed. Enlargement of the kidney and spleen, often exophthalmia and interstitial granulomas was noted (Olsen et al. 2006a, 2006b). 50–60% of farmed tilapia has died in Costa Rica. Also has the signs as loss of appetite, abnormal swimming patterns, anaemia and Exophthalmia (Soto et al. 2009).

Diagnosis

Characterization and Identification of Pathogens

Cultures comprise the non-motile, weakly gram-negative and negatively oxidase. These cultures were incubated at 22 °C on Cystine Glucose Blood agar medium and demonstrated enzyme activities like esterase (C4) and esterase lipase (C8).

Molecular Methods

Cultures depended on 16SrRNA sequencing which was close to *Francisella philomiragia*. So, *Francisella noatunensis subsp. orientalis* and *Francisella noatunensis* (Ottem et al. 2009) were identical to *Francisella philomiragia*.

Treatment

Infected Atlantic cod and three lined grunts were vaccinated by florfenicol, oxolinic acid, erythromycin, streptomycin, rifampin, and flumequine which resulted in control of the disease.

19.3.2.7 *Francisella noatunensis subsp. orientalis*

Clinical Signs

Those infected fry expressed exophthalmia; they become lethargic and also have pale gills. In addition, the spleen and kidney becomes enlarged and it has emptied intestines. Enlargement of gallbladder was also noticed (Jeffery et al. 2010). Nile tilapia and blue tilapia and Mozambique tilapia were recovered from granulomatous disease. Also infection with this organism associated with wild caught Caesar grunt

and French grunt. Spleen and kidney were infected by granulomas. Infected fishes died off (Soto et al. 2012).

Diagnosis

Characterization and Identification of Pathogens

Outbreaks of infections were caused by Rickettsia-like microbes (Chern and Chao 1994). These colonies formed have size more than 1 mm diameter and lateral development occurred by incubation process at 22 °C for 3 days. They are aerobic weak gram-negative bacteria, with shape intermediate between spherical and rod-shape (cocco-bacilli) which liberate hydrogen sulphide when incubated on medium containing cysteine at temperature 10–30 ° C. Replication of organisms from invading head kidney macrophages after infection causes apoptosis and cytotoxicity within 24–36 h. By using the (PQ1104) strain for hybridization of DNA, 60.3% of homology was associated with *Francisella noatunensis* (Mikalsen and Colquhoun 2009). For *Francisella noatunensis subsp. orientalis*, specialized quantitative methods and the real-time PCR has been used for identification.

Treatment

Vaccine Development

Tilapia was protected by immersion vaccination with bacterial suspension of 10⁷CFU/ml for 30 min to 180 min by immersion challenge with wild type (Soto et al. 2011). Antibacterial antibiotics like Florfenicol can be given orally for the treatment of tilapia for 10 days, at an average dosage of 15 mg/kg of fish.

19.3.3 Bacterial Diseases of Marine Water Fishes

19.3.3.1 Enterobacteriaceae Representatives

Edwardsiella tarda

Edwardsiella tarda is a facultative anaerobic, motile, gram-negative rod-shaped flagellated bacteria. This bacteria infects channel catfish, eels, carp, tilapia, chinook salmon and flounder and leads to edwardsiellosis disease (red disease) (Han et al. 2004).

Clinical Signs

The exterior signs are circular abrasion on the postero-lateral part of the body, which then leads to the development of ulcers spreading into hair-filled regions. From the side, these are converse bulge areas and if ruptured, release foul odour. The disease affects fish when the fish is transported to fish processing plants. The scanning of disease affecting tilapia shows the symptoms of loss of pigmentation, blurred of the eyes, abdomen filled with ascitic fluid.

Edwardsiella tarda was identified from the spleen and renal tissue of seawater flounder.

Diagnosis

To determine the presence of *Edwardsiella tarda* in affected fish tissue, fluorescent antibody technique (FAT) has been used for diagnosis (Amandi et al. 1982). Lindstrom et al. (2009) examined the most effective field for diagnosis by using FAT. The monoclonal antibodies of FAT are best suited for diagnosis as being an enzyme for immune assay. ELISA is also used for diagnosis of 44 kd outer membrane proteins.

Treatment

Vaccine Development and Immunostimulants

Vaccination of prophylaxis has been a favourable outcome with oral immune stimulants, namely β -glucan and vitamin C and E for protection. Yano et al. (1989) reported that injection of beta-1, 3 glucans at 2-10 mg/kg of fish inflates resistance for *Edwardsiella tarda*.

Probiotics. *Bacillus subtilis*, *Enterococcus faecium* are those fermented by citrus by-products when nourished to young olive flounder for 70 days gives protection against pathogenic culture as immunostimulation.

19.3.3.2 Flavobacteriaceae Representatives

Tenacibaculum maritimum (= *Flexibacter maritimus* = *Cytophaga marina*)

Tenacibaculum maritimum is a gram-negative, gliding bacteria. *Tenacibaculum maritimum* infection in fishes leads to a disease named as tenacibaculosis, which is one of the most devastating diseases to marine fish community. *Tenacibaculum maritimum* can infect the large population of fish and drastically impact fishing industries all over the world.

Clinical Signs

The infected fish species exhibits clinical signs of fin rot, skin lesions, and paleness of internal organs. This bacteria dwells in host-associated environments, especially in fish renal tissues, and leads to tenacibaculosis, which exhibits clinical signs of lesions and necrosis nearly on all external surfaces of the infected fish (Avendaño-Herrera et al. 2004).

Specifically in juvenile fish, the external changes include mouth erosions, gill erosion and tail rot (Handlering et al. 1997). Initially, on the fins, head and trunk injury develops as grey-white cutaneous in older animals. This condition leads to ulcers from these lesions being depraved (Wakabayashi et al. 1984). Acute and chronic, these are two stages of disease which develop in Salmonids. The acute tenacibaculosis developed 48–72 h after infection with more than $1 \times 10^3 \times 10^3 \times 10^2$ cells/ml epithelial abrasion. In chronic tenacibaculosis tiny blisters appear on the epidermal layer which leads to ulceration and exposing the musculature beneath. Gill necrosis occurred due to lesions formed on the fins and jaw. This bacterial infection results in ulcers in wedge sole (López et al. 2009). Black patch necrosis of Dover sole and bacterial stomatitis of Atlantic salmon (Ostland et al. 1999).

Diagnosis

Characterization and Identification of Pathogens

Tenacibaculum maritimum can be isolated from infected tissues of fish by using Cytophaga agar, mixed with 70% marine water and tryptone-yeast extract-cystine agar medium (Hikida et al. 1979). Cultures containing a very homogeneous group of thin flat layers of colonies which appear pale yellow in colour was identified and is composed of aerobic gram-negative rod-shaped bacteria with size $2\text{--}30 \times 0.5 \mu\text{m}$, which sometimes have filaments up to $100 \mu\text{m}$ long (Bernardet et al. 1990). In older cultures, microcysts do not occur but cells become spherically shaped with a diameter of $\sim 0.5 \mu\text{m}$. The gliding motility is the typical attribute of all isolated bacteria and a non-gliding strain also has been isolated from infected puffer fish (*Takifugu rubripes*) (Tanvir et al. 2014). The growth of pathogens is noticed at a temperature range from 14.6 to 34.3 °C and at pH range from neutral to mild alkaline.

Serology

To detect the pathogen *Tenacibaculum maritimum* whole cell agglutination is effective (Wakabayashi et al. 1984), and also for the identification in tissues of fish FAT can be used (Baxa et al. 1986).

Molecular Methods

16S rRNA sequence amplified with specific PCR primers can be used to identify *Flavobacterium branchiophilum*, *Flavobacterium columnare* and *Tenacibaculum maritimum* (Yeh et al. 2006); however, conventional PCR cannot detect the DNA of the cells because freezing-thawing destroys the cells (Suomalainen et al. 2006a, 2006b) but nested-PCR detected nearly 1–250 cells per PCR cycle in gill, skin and mucus samples of infected fishes. Some asymptomatic fishes show the presence of ten bacterial cells/cycle by nested-PCR (Bader et al. 2003). Mostly, 89% in the tissue blocks from fish detected the DNA from the pathogen, which did not have clear signs of gill disease and in 95% of the tissue blocks from fish with mild-to-severe gill erosion detected the presence of DNA from pathogen. Hence, for the detection of *Flavobacterium psychrophilum* and *Tenacibaculum maritimum*, qPCR technology has been found to be more reliable than culturing techniques (Fringuelli et al. 2012).

Treatment

Vaccine Development

Freund's incomplete adjuvant (FIA) can be injected as immunostimulant to Atlantic salmon (0.1 ml via i.p.) followed by a formulated suspension of *Tenacibaculum maritimum* containing $\sim 1.79 \times 10^1 \times 10^3 \times 10^3 \times 10^3$ cells/ml with relative percentage survival of 79.6% and without relative percentage survival of 27.7% (Van Gelderen et al. 2010). Besides the success of the adjuvanted version of vaccine at reducing mortalities, side effects of the adjuvant are also very likely. Prominent side effects are the development of black/brown pigmentation by melanin formation on the stomach with inflammation in the form of granulomas and cysts (Van Gelderen et al. 2009).

Plant-Based Immunostimulants

1.0–2.0 mg of aqueous or ethanol extracts of an Asian plant *Liriope platyphylla* which is commonly known as big blue lilyturf is used as an immunostimulant for olive flounder (*Paralichthys olivaceus*). Plant-based immunostimulation methods enhance the RBC count with increased haemoglobin concentration, and also enhanced the lymphocyte and monocyte counts and thus complement the phagocytic functions (Harikrishnan et al. 2012a, 2012b, 2012c).

Antimicrobial Compounds

Tenacibaculum maritimum exhibits moderately sensitive to chloramphenicol, doxycycline, oleandomycin, oxytetracycline, sulphamonomethoxine and thiamphenicol, highly sensitive to ampicillin, erythromycin, josamycin, nifurpirinol, penicillin G and sodium nifurstyrenate, and weakly sensitive to nalidixic acid, oxolinic acid, spiramycin and sulphisoxazole (Baxa et al. 1988). Trimethoprim and amoxicillin

dosage have been recommended at 80 mg/kg body for the treatment (Soltani et al. 1996).

19.3.3.3 Vibrionaceae Representatives

Vibrio alginolyticus

Vibrio alginolyticus is gram-negative sea bacteria. Most mortality that occurred in farmed sea bream is by *Vibrio alginolyticus* infection that leads to bacterial septicaemia.

Clinical Signs

This organism causes anaemia. Infected fishes expressed sluggishness and developed ulcer, skin darkening and blockages in air bladder, liver and capillaries in the intestinal wall. Enlargement of intestine and gallbladder are present. Low levels of mortality noted in turbot with gill disease in recirculating aquariums (Austin et al. 1993). This organism causes corneal opaqueness and exophthalmos, their signs correlated with *Vibrio harveyi*. *Vibrio alginolyticus* is also present in ulcerous fish. High mortality occurred in silver sea bream by *Vibrio alginolyticus* (Ye et al. 2008). Also this bacteria is the causative organisms for disease in gilt-head sea bream. Black sea bream fry is associated with *Vibrio alginolyticus* and red spot caused by *Vibrio alginolyticus* in sea mullet fish (Burke and Rodgers 1981). Diseased fish have sluggish swimming and also suffered by exophthalmia which leads to death (Rameshkumar et al. 2017). These organisms are present in damaged tissues.

Diagnosis

Characterization and Identification of Pathogens

The organism can be isolated from blood by using TSA, TCBC with seawater agar. An incubation period is at 15–25 °C for 2–7 days (Colorni et al. 1981). From moribund eels the isolates of *Vibrio alginolyticus* was successfully done by differential medium of *Vibrio alginolyticus* agar which has been combined with high concentration of sodium chloride and bile salts at 37 °C of incubation. Green-yellow colonies will be produced, which comprise motile, gram-negative rod-shaped organism producing hydrogen sulphide, lysine, indole, catalase and ornithine but β -galactosidase or arginine dihydrolase will not be produced. Nitrates are deduced. The Voges Proskauer (VP) test and methyl red (MR) test results are positive. Sensitivity of vibriostatic agent O/129 has been recorded. From maltose, mannose, glycerol, sucrose and salicin the acids will be produced.

Molecular Methods

In diseased marine fish, the diagnosis of the disease is achieved by loop assisted isothermal multiplication with *gyrB* target gene. The limitation of 3.7×10^2 CFU/ml is used for rapid detection.

Treatment

Vaccine Development

Culture of *Vibrio alginolyticus* containing flagellin *flaA* genes given as treatment in red snapper by i.m.(8 µg/fish). The antigen will be accumulated in the liver, spleen and kidney, with the expressions of *flaA* genes corresponding to 88% of relative percentage survival (RPS) after 7–28 days, and 84% of the RPS by recombinant *flaC* genes. Infectious spleen and kidney necrosis virus (ISKNV) evaluation is carried out by combination vaccine with *Vibrio alginolyticus*, with 80% of RPS on orange-spotted grouper. Large-yellow croakers can be protected against the challenge by i.p. injection with 100 µg protein per fish. The booster dose can be given 2 weeks after first vaccination (Qian et al. 2008).

Plant-Based Immunostimulant Agents

In Rockfish, *Aloe vera* has increased the resistance to *Vibrio alginolyticus* for 6 weeks after feeding at 5 g *Aloe vera*/kg of diet. Using chitosan and chitin to kelp grouper has increased the immunostimulation. Also, globin, haemoglobin, albumin, leucocyte, and erythrocyte counts are enhanced. Chitosan gives more protection compared to chitin in challenge (Harikrishnan et al. 2012a, 2012b, 2012c). In Clownfish, white mangrove aqueous leaf extract fed at 1–8%, developed survival after challenge and also enhanced immunostimulation. 70%, 80% and 85% of survival is increased while using extracts at 1, 4, and 8% (Dhayanithi et al. 2015). Clownfish is examined with another *Rhizophora apiculata* (Mangrove) extracts fed at 5% and 10% showed enhanced immunostimulation. Immunostimulation and growth is improved in *Amphiprion sebae* with 5% of mangrove extract and enhancing 85% of survival rates. In another experiment, grouper juveniles fish is tested with *Sauropus androgynus* extracts for 30 days fed at 1.0 and 2.5 g/kg resulted in improved immunostimulation as well as susceptible to the challenge (Samad et al. 2014).

Antibacterial Compounds

Antibacterial compounds such as chloramphenicol have been used for this disease. It was first successfully achieved by Colorni et al. (1981). 50 mg of drug was given per

kg body weight of fish per day. Also nitrofurantoin dosed at 50 mg/l of water/1 h. Both antimicrobial compounds can reduce the fatality rates.

Vibrio Anguillarum (Bacterium anguillarum)

Canestrini 1893 was the first to discover a bacterial fish pathogen of marine species known as *Bacterium anguillarum* (later named as *Vibrio anguillarum*). *Vibrio anguillarum* is rod-shaped polar flagellated gram-negative bacteria. It causes red pest disease in eel fishes. It is damaging to the economy of aqua industries worldwide.

Clinical Signs

The external symptoms of the disease are skin discolouration, red necrotic abrasion in abdominal muscle and erythema surrounding the vent and within the mouth. The gut filled with viscous fluid. This infection was also noticed in Pacific salmon fingerlings. The pathological abnormalities include abnormalities in the blood, connective tissue, gills, renal organ, liver and posterior gastrointestinal track and splenomegaly. Fin rot is another state as cribbed to *Vibrio anguillarum*.

Diagnosis

Characterization and Identification of Pathogens

The pathogen is retrieved from infected tissue, inoculated on BHIA and nutrient agar additive at 0.5–3.5% (w/v) with NaCl, TCBS and seawater at 15–25 °C and incubated for 1 week. The taxonomy of the pathogenic bacteria divided these species into three biotypes on the basis of biochemical reactions.

Type A called as *Vibrio anguillarum* forms typical colonies, giving rise to indole and acid formation from mannitol and saccharose.

Type B called as *Vibrio anguillarum* forms an anguillicide and does not bring about indole or acid formation from saccharose and mannitol.

Type C called as *Vibrio anguillarum* forms an ophthalmia and generates acid from saccharose and mannitol, but does not give rise to indole.

A clarified diagnostic test has been described for *Vibrio anguillarum*, requiring “glucose motility deeps”. Another necessity of GMD is an oxidation fermentation test medium containing dextrose 1.0%(w/v), yeast extract 0.3%(w/v), phenol red broth base (difco) 1.6% w/v, agar 0.3%(w/v).

Treatment

Management Techniques

Infected eels can be moved to well-aerated water and added to reduce complications with vibriosis.

Disease Resistant Fish

The genetically modified disease-resistant fish strain has an opposite confirmation at bestow on resistance to vibriosis. Among various transferrin genes of Coho salmon and Steelhead trout, Winter et al. (1980) stubbornly maintained that there was no difference in resistance to vibriosis. Lately, in the major histocompatibility complex (MHC) 11 α genes in Japanese finders an association has been made between polymorphic and *V. anguillarum*, and in turn three alleles paol DAA*1301, paol DAA*1401, paol DAA*2201 were related notably with resistance (Xu et al. 2011).

Vaccine Development

Johnson and Amend 1983 integrated a vaccine into gelatin and put in oral and anal ingestion to subdue digestion and intestinal symptoms. The results showed the best decrease in mortality.

Vibrio ordalii

Vibrio ordalii is a gram-negative bacillus. In seawater fish vibriosis is the fundamental reason. In Japan, the *Vibrio ordalii* was reported by Muroga et al. (1986a, 1986b). This disorder was recognized as haemorrhagic septicaemia. In gill tissue, skeletal muscle, cardiac muscle, and inside the anterior and posterior of the GI tract, there has been a tendency for the formation of micro colonies among pacific salmon. Contrarily, while in comparison with bacteraemia caused by *Vibrio anguillarum* evolved substantially later in the disease cycle. By the way, the use of seawater and thiosulphate-citrate-bile salts-sucrose agar (TCBS) *Vibrio ordalii* incubated at 25°C for 7 days (Ransom 1978; Ransom et al. 1984). By means of plasmid describing, *Vibrio ordalii* became analogous, ribotyping and serotyping aid lipopolysaccharide (LPS) class and changed into miscellaneous by Biolog-Gen fingerprints. In *Vibrio ordalii*, cynicism plasmid has no longer been determined (Crosa 1980). However, without DNA polymerase I, a 30 kb enigmatic plasmid delegated as PMII01 copied without recreating a single-stranded mean has been set up in all isolation of *Vibrio ordalii* (Bidinost et al. 1999). However, proteases have no longer been entrenched (Kodama et al. 1984).

Diagnosis

Molecular Techniques

The gene for haemolysin, a 112 base pair fragment which is commensurate by using PCR and RT-PCR. The RT-PCR gave 1 h faster copies than the PCR, and it discovered from 5.27×10^2 to 4.13×10^3 CFU/ml or in other words 62–145 versions of voh B gene in spleen and kidney of Atlantic salmon with bacterium.

Treatment

Probiotics

A segregate of *Vibrio alginolyticus* before used as probiotic in Ecuadorian cipher rookery has been practically at commanding disease provoked by *Vibrio ordalii* (Austin et al. 1995).

19.3.3.4 Alteromonadaceae Representatives

Pseudoalteromonas piscicida

Pseudoalteromonas piscicida is a gram-negative gammaproteobacterium present in marine environments. It was reported among the eggs of damselfish, *Amphiprion clarkii* and *Amblyglyphidodon curacao*, characterized by the whitening of the eggs resulting in death within 24 h. (Nelson and Ghiorse 1999). Using with marine agar, the infected eggs were cultivated at 28 °C for 2 days. This culture was experimented among the Damsel fish eggs resulting in balanced mortality which is in contrast to uninfected controls (Nelson and Ghiorse 1999). From *Amblyglyphidodon curacao* eggs were recognized by identifying 16S rDNA sequencing called *Pseudoalteromonas piscicida*.

Shewanella putrefaciens

Shewanella putrefaciens occurs in various distinct forms and is a gram-negative bacterium. It has been extracted from both marine environments and anaerobic sandstone in the Morrison Formation in New Mexico. This disease affects the rabbit fish, *Siganus rivulatus* with high transience occurred mostly in Red sea cages. Gram-negative bacteria was recovered which was effective in affecting the healthy fish mostly during the spring season. This disease has not been reported in any other species. From the API 20 E rapid identification system, a taxon from *Alteromonas putrefaciens* has been rechanneled as *Shewanella putrefaciens*. This disease affects fish that have the external signs such as inertia, exophthalmia, haemorrhaging around the mouth and fin damage. At the same time, the internal signs were not

identified. In Poland, the fish was affected with necrotized gills and necrotized abrasion on the skin among the common carp and rainbow trout particularly during 2007–2012. They have the interior signs such as swollen kidney, enlarged spleen, etc. From the infected spleen, kidney and liver, the bacterium was separated; in turn it was incubated with sodium chloride 3%(w/v) in addition to Brain Heart Infusion agar (BHIA) for an concealed period of time (Saeed et al. 1987).

Treatment

Vaccine Development: The formalin killed vaccine gives clear evidence in limited death rates when administered by i.p. injection (Saeed et al. 1987). When compared to unvaccinated conditions, these two injections reported 40% decreased mortalities. Engrossment vaccination ends in field results.

19.3.3.5 Oxalobacteraceae Representative

Janthinobacterium lividum

The gram-negative rod-shaped bacteria were related with transience which was purple pigmented affecting marine fishes in farms in Scotland and Northern Ireland. The moribund rainbow trout in Scotland was recognized with Rainbow Trout Fry Syndrome. The rainbow trout in Northern Ireland was faced with high death in 2–3 weeks.

Clinical Signs

During January 1992, furthermore purple pigmented bacteria was identified among the large rainbow trout (100-200 g in weight) resulting in the skin abrasions, which were enervating with epiretinal membrane. Small fish exhibit inactive movements, increased skin complexion, skin abrasions, pale gills and abdomen expanded. In the interior, the kidney and spleen become enlarged and serous fluids are filled in the peritoneal cavity. The sockdolager, jumbo, and buster was affected with external scrape, the skin removed off along the loin to tail and showed muscular abnormality (Austin et al. 1992).

Diagnosis

Characterization and Identification of Pathogens

The diversity of media comprises blood agar (5%v/v bovine blood in Gibb blood agar base), cytophaga agar, Histone lysine demethylase 2 (KDM2), Lactose

Fermenters (L-F) medium and Trypticase soy agar or tryptone soya agar (TSA) in which the standardized specimen of whole fish (liver, spleen, kidney and substances from surface abrasions) was grown on the surface of various media at 22 °C for 14 days. After 3 days, the purple pigmented population was identified.

Treatment

Antimicrobial Compounds

The organisms are conscious of oxolinic acid, potentiated sulphonamides and furazolidone (Austin et al. 1992). Accordingly, it is suspected that these chemicals would be convenient for chemotherapy.

19.3.3.6 Piscirickettsiaceae Representatives

Piscirickettsia salmonis

Debased or constrainly parasitic *bacilli* such as Rickettsias and Chlamydias have been habitual bacterium of invertebrates and occasionally referred to attach with fish disease which is termed as Huito disease (Schafer et al. 1990). Salmonid rickettsial septicaemia (Cvitanich et al. 1991) were seen among Quinnat Salmon, Silver Salmon and Rainbow Trout, extended to Atlantic Salmon in Norway (Olsen et al. 1997) and White Bass in California (Arkush et al. 2005). Though the bacterium was at first analogous with Salmon, later it outstretch to other groups integrated to sea dace (McCarthy et al. 2005; Arkush et al. 2005).

Clinical Signs

The infected fish shows inactive and were repletteness. Exterior symptoms include melanosis, gills were paled and anaemia. At first the disease was not observed to take place during fresh water of fish culture. The fish from fresh water was moved into seawater after 6–12 weeks; the death has been recorded (Fryer and Hedrick 2003).

Diagnosis

Characterization and Identification of Pathogens

Isolation of this bacterium was achieved by kidney disease which affects the fish in Salmon egg cells cytoplasm for 5–6 days at 12–21 °C (Fryer et al. 1990). But the growth did not take place in blood agar, BHIA, plasma medium and Loffler medium.

Serology

Isolating of tissue culture is achieved by using iFAT, the spotting in acridine orange stained smears was recommended (Lannan et al. 1991).

Treatment

By Microwave Radiation

For *Piscirickettsia salmonis*, the microwave radiation at 700w was proposed (Larenas et al. 1996a, 1996b).

Vaccine Development

Formalized cells (106.7 TCID₅₀/ml) were delivered showing growth of good preservation in a field experiment with Coho Salmon (Smith et al. 1999). The process of using oral vaccination every 3 days at 6 mg for 30 days called “micromatrix” has been reported and revealed promise.

19.3.3.7 Pseudomonadaceae Representatives

Pseudomonas alcaligenes

These gram-negative rod-shaped organisms of $\sim 2.4 \times 0.8 \mu\text{m}$ mainly infect Moribund fish of fresh and marine water.

Clinical Signs

Infected fishes express signs like pale gills with the swollen and they are affected by anaemic and they also have swollen spleen, haemorrhaging in their kidney and gonad and ascites were also present. Blood spots were developed around the oral cavity. The hybrid sturgeon was taken for further experiments, 33% of mortalities have been recorded with 2×10^6 CFU/Fish after 5 days of infections.

Diagnosis

Characterization and Identification of Pathogens

Using the kidney and liver of Moribund fish, the isolation process was developed with BHIA for an incubation period of 10 days at 28 °C. They are translucent, white in colour and circular; it may be present in individual or in pairs. Colonies are motile and they have unique polar flagella. It is classified as phylogenetic by Biolog -Gn,

which utilize L-alanine, L-arginine, L-malic acid and pyruvate. 16S rRNA sequencing corresponds to 99% of homology with *Pseudomonas alcaligenes* (Xu et al. 2015).

19.3.3.8 Moritellaceae Representatives

Moritella marina (=Vibrio Marinus)

Moritella marina is a halophilic psychrophilic facultative anaerobe gram-negative with curved or straight rods, motile polar flagella that generate PUFAs (polyunsaturated fatty acids) and DHA (docosaehaenoic acid). At low temperature these organisms were correlated with facial skin abrasions of *Salmo salar* orchard in Iceland specifically ~10 °C. In a different part of cold marine environments, *Moritella marina* has been seen from the ocean floor to the intestinal tract of marine organisms. Many of the *Moritella* species are thought to live in collaboration with marine organisms.

Moritella viscosa

In recent times in Scotland, an unspecified cause of ulcer seems on the fringe of *Salmo salar* in seawater (Salte et al. 1994; Lunder et al. 1995; Benediktsdottir et al. 1998). Consequently, *Moritella viscosa* are retrieved from two diseased farmstead Atlantic cod in Norway. In some cold-water fish, *Moritella viscosa* has been found to cause skin ulcers (Urakawa et al. 1998).

Diagnosis

Molecular Methods

The *Moritella viscosa* has been determined conveniently by using PCR which is fixed at the limit of $6.09 \times 10^1 \times 10^{13}$ g of DNA, and is identical to ten bacterial genomes (Grove et al. 2008).

Treatment

Vaccine development: *Salmo salar* which were vaccinated through intraperitoneal injection with an assisting entire cell, authorized interruption carrying *Moritella viscosa* were stored against ensuring summons to attain an RPS of 97% (Greger and Goodrich 1999).

19.3.3.9 Halomonadaceae Representatives

Halomonas cupida

Halomonas are gram-negative, motile, rod-shaped cells. These organisms usually occur in unpigmented or yellow-tinted colour. *Halomonas* bacteria strains require NaCl for growth. They are mostly found in water resources with elevated levels of salinity and usually inhabit deep-sea sediment and deep-sea waters (Okamoto et al. 2004). This bacteria infects *black seabream* and *Acanthopagrus schlegelii* (Kusuda et al. 1986).

Diagnosis

Characterization and Identification of Pathogen

Bacteria obtain through intestinal microflora and in addition to infected tissue. Homogenate cultures were inoculated by BHIA with the incubation period of 24 hours at 25 °C (Kusuda et al. 1986). An oxidative-fermentation test was inactive. Several enzymes and amino acids are produced by catalase, arginine dehydrolase and ornithine dehydrolase and lysine. Hydrogen sulphide and indole are not produced, nitrates became deduced. However, Voges Proskauer reactions and Methyl red test have come to be negative. Haemolysis occurred in eel erythrocytes. Acids are generated by galactose, maltose, adonitol, mannitol, L-rhamnose, D-sorbitol, trehalose, salicin and sucrose (weak). However, acids are futile by inulin and fructose. Above all, these manners clearly resemble the relationship between *Alcaligenes cupidus*. But dependent on 16s rRNA sequencing (Dobson and Franz Mann 1996), this toxon was changed to *Halomonas cupida* (Baumann et al. 1983).

19.4 Summary of Bacterial Fish Diseases

The overview of bacterial fish diseases is given in Table 19.1.

19.4.1 *Bacterial Fish Disease During Hatching of Eggs and Larvae*

In the course of the meticulous hatching of eggs and raising of marine larvae, extraordinary kinds of interactions between microorganisms and organic surfaces can also occur. This ends in the initiation of an autochthonous microflora or be the start of infection. Among habitats and hosts, microorganisms may be effortlessly travelled, inside the aquatic surroundings. Egg surface reflects the bacterial

Table 19.1 Major bacterial fish diseases, causative agents and its host.

Gram-positive bacteria		
Fish diseases	Causative agent	Host range
Eubacteriaceae representative Eubacterial meningitis	<i>Eubacterium tarantellae</i>	Striped mullet, snook, gulf flounder
Streptococcaceae representatives Streptococcosis Lactococcosis (septicaemic disease)	<i>Lactococcus garvieae</i> , <i>Streptococcus difficilis</i> , <i>Str.Iniae</i> , <i>Str.Parauberis</i> , <i>Str. phocae</i>	Rainbow trout, Golden shiners, Japanese eels, yellow tails, striped mullet, hardhead catfish
Corynebacteriaceae representative Bacterial kidney disease	<i>Renibacterium salmoninarum</i>	Salmonid fish
Mycobacteriaceae representatives Mycobacteriosis	<i>Myc.abscessus</i> , <i>Myc.marinum</i> , <i>Myc.neoaurum</i> , <i>Myc.gordonae</i>	Zebra fish, Salmonids, European sea bass, Tilapia, Striped bass, Carp
Staphylococcaceae representatives Eye disease	<i>Sta.aureus</i> , <i>Sta.epidermidis</i> , <i>Sta.warneri</i>	Silver carp, African sharp catfish, Yellow tail, Red sea bream, Gilthead sea bream
Carnobacteriaceae representative Lactobacillosis, Pseudokidney disease	<i>Carnobacterium piscicola</i>	Salmonids
Nocardiaceae representatives Nocardiosis, ocular oedema	<i>Nocardia spp.</i> (<i>Noc. Asteroides</i> , <i>Noc. Salmonicida</i> ; <i>Noc. Seriolae</i>), <i>Rhodococcus sp.</i> , <i>Rhodococcus erythropolis</i> .	Most fish species, <i>Oncorhynchus tshawytscha</i> , <i>Salmo salar</i>
Bacillaceae representatives Septicaemia, bacillary necrosis, branchionecrosis, ulceration	<i>Bacillus spp.</i> , <i>Bacillus cereus</i> , <i>Bacillus mycoides</i> , <i>Bacillus subtilis</i>	Different varieties of freshwater fish species including catfish (<i>Pangasius hypophthalmus</i> , carp (<i>Cyprinus sp.</i>), striped bass (<i>Morone saxatilis</i>), channel catfish (<i>Ictalurus punctatus</i>)
Microbacteriaceae representative	<i>Microbacterium paraoxydans</i>	Nile tilapia
Micrococcaceae representative Micrococcosis	<i>Micrococcus luteus</i>	Rainbow trout
Planococcaceae representative	<i>Planococcus sp.</i>	Salmonids
Gram-negative bacteria		
Enterobacteriaceae representative Enteric septicaemia of catfish	<i>Edw.ictaluri</i>	<i>Amieurus nebulosus</i> , <i>Pangasius hypophthalmus</i> , <i>Danio devario</i> , <i>Pangasius hypophthalmus</i>
Red pest, edwardsiellosis, emphysematous putrefactive disease of catfish	<i>Edw.tarda</i> (<i>Paracolobactrum anguillimortiferum</i> , <i>Edw. anguillimortifera</i>)	Various freshwater species

(continued)

Table 19.1 (continued)

Gram-positive bacteria		
Fish diseases	Causative agent	Host range
Flavobacteria and cytophages Columnaris, saddleback disease	<i>Flavobacterium columnare</i>	Many freshwater fish species
Gill disease, skin disease	<i>Flavobacterium johnsoniae</i> (= <i>Cytophaga johnsonae</i>)	Barramundi (<i>Lates calcarifer</i>), koi carp, rainbow trout, longfin eel (<i>Anguilla mossambica</i>)
Gill disease; generalized septicaemia	<i>Chryseobacterium scophthalmum</i> (= <i>Flavobacterium scophthalmum</i>)	Turbot
Bacterial stomatitis, gill disease, black patch necrosis	<i>Tenacibaculum maritimum</i> (= <i>Flexibacter maritimus</i>)	Various marine fish species
Vibrionaceae representatives Eye disease, septicaemia	<i>Vibrio alginolyticus</i>	Cobia (<i>Rachycentron canadum</i>), gilt-head sea bream, grouper (<i>Epinephelus malabanicus</i>), sea bream (<i>Sparus aurata</i>)
Vibriosis	<i>V. ordali</i>	Various marine fish species
Alteromonadaceae representatives Egg disease	<i>Pseudoalteromonas piscicida</i>	Damsel fish
Septicaemia	<i>Shewanella putrefaciens</i>	Rabbit fish (<i>Siganus rivulatus</i>)
Oxalobacteriaceae representative Anaemia	<i>Janthinobacterium lividum</i>	Rainbow trout
Moraxellaceae representative Acinetobacter disease	<i>Acinetobacter sp.</i>	<i>Salmo salar</i>
	<i>Moraxella sp.</i>	Striped bass
Piscirickettsiaceae representative Coho salmon syndrome, salmonid rickettsial septicaemia	<i>Piscirickettsia salmonis</i>	Salmon, sea bass (<i>Atractoscion nobilis</i>)
Halomonadaceae representative	<i>Halomonas</i> (= <i>Deleya</i>) <i>cupida</i>	Black sea bream (<i>Acanthopagrus schlegeli</i>)
Francisellaceae representative Francisellosis, visceral granulomatosis	<i>Francisella noatunensis</i>	Atlantic cod, <i>Salmo salar</i> , three-line grunt, striped bass, tilapia
Francisellosis	<i>Francisella noatunensis</i> subsp. <i>Orientalis</i>	Tilapia, French grunt, Caesar Grunt
Pasteurellaceae representative	<i>Pasteurella skyensis</i>	<i>Salmo salar</i>

composition of the aquatic surroundings, due to the diverse plant life that in the end develops at the egg floor. Representatives of mucilaginous microflora may damage developing eggs. The microflora of marine invertebrates and plankton can be ruled via probably pathogenic *vibrios* at certain times of the 12 months. End result of including meals at excessive awareness of water is a brilliant medium for the boom of heterotrophic or opportunistic microorganisms. The opportunistic microorganism can cause many fish illnesses. Fish decreased from resistance towards contamination due to pressure in handling or transport of fish. Aquatic invertebrates are natural meal resources for fish larvae and are also co-inhabitants of larval ecosystems. This correlation will suggest that the inception of a larval microflora may also be stimulated by means of the primordial microflora of invertebrates, whether they are food organisms or co-population of larval ecosystems or rearing centers. The fish may also take in invertebrates as meals, the fish may be inflamed through a particular pathogen, which the invertebrates have contained. The pathogen vibrio group comprises microorganisms with a dynamic position in the marine and brackish environments. A particular courting exists among some marine vibrios and their invertebrate hosts. This interaction can be specific, for the reason that bivalves may additionally have lectins that bind vibrios, which include *Vibrio Cholerae*, *Vibrio Vulnificus* which might be pathogenic to fish.

Larval production system have the problems by poor egg quality it leads to heavy mortalities. Eggs are overgrown through incubation techniques. Indigenous microflora may affected by bacteria. And also hatching and hamper egg maturing affected by bacteria. In farmed fish, their short and long period of health has been affected by larval incubators and microflora on eggs. Microflora has been controlled by using antibiotics. Various techniques are used to remove the adherent microflora and during transportation the eggs are have been preventing against the bacterial pathogens in aquaculture. Ozonization, antibiotics, UV-irradiations and membrane filtration methods are used to larviculture. These methods are furtherly interrupt the growth of opportunistic bacteria. Overgrowth of bacteria has led to hypoxia or delayed hatching. Exo proteolytic enzymes were released by adherent bacterial epiflora, it affects the chorion and also zona radiata were destroyed. An intra ovum, infections may transferred by pathogens.

The research reported that in grownup fish the proof encouraged that restrained placenta and excreting immunity is chief in protecting in opposition to bacterial ailment. The regular excretion and discharge of sludge push to goblet cells in the dermis, gills, and membrane of the gastrointestinal vicinity restrict microbial contamination. Excretion of interleukin-like factors recommends that goblet cells may have immunostimulatory functions (Sigel et al. 1986). Relevantly small is familiar about ontogenesis of immunity in fish. It has been deduced that the “resistant ability” in fish larvae is not completely enlarged up to they are many weeks old (Chantanachookhin et al. 1991). Still, this may fluctuate among the fish genus. Ig advantageous cells are confirmed which are observed within a few weeks.

19.4.2 Bacterial Fish Disease During Transportation and Storage

Transportation and storage structures have an effect on the behaviour of fish. The fish becomes vulnerable because of a lack of oxygen supply during transportation and the safety of Betok fish is depleted. Any other aspect, which could affect the prevalence of abnormalities in Betok fish, is advertised within the fish market of Segiri. It is because of the transportation procedure which makes use of pickup and excessive density. The fishes tend to get friction between a fish and another fish. Furthermore, it is also because of the consignment field, so the condition of fish turns weaker.

The fishes had been transported in an easy etching with wet structures that have a tendency to have red spots, reducing, losing scales on the surface of the frame, operculum, and head. The fishes were also discovered to have eminent eyes (exophthalmia) and fingertips. Pathologies can be caused by friction between a fish with any other fish. It is investigated that these fishes have scales and fins which are rigid and sharp, which is able to induce damage to another fish when it is in the contagious container. The length of time also influences the transport of intensity/frequency of fish rubbing. This allows greater harm for the sale procedure. There had been additional triggers for the incidence of accidents to the outside organs of the fish. It results from reconciliation with excessive density without aeration and with a small quantity of water.

It is recognized that there are five kinds of bacteria, specifically: *Aeromonas*, *Pseudomonas*, *Staphylococcus*, *Streptococcus*, and *Bacillus*. The fifth type of bacteria is a bacterium that is generally determined in freshwater fish and numerous human beings reported the infection of fish farming (Austin and Austin 2007). The presence of microorganisms tremendously depends on environmental conditions and the condition of the fish itself. The transportation and storage complex of fish plays a function within the rise of intensity and incidence of bacterial infections in Betok fish.

19.5 Conclusion

The growth of organic matter such as fecal material or uneaten fish food leads to the microbial growth in the water. This greatly affects the fish's health. As well as the dead fish in the pond or a tank for a longer period results in the growth of the microorganisms. Some disease in fish leads to economic losses. Dietary supplements also play a major role in fish health. When the fish are fed with adequate diets like vitamins, natural plant products, and immunostimulants, it leads to better growth and reproduction. Even some bacterial pathogens are capable of creating disease in human beings also when they consume infected fish.

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