# INTRODUCTION

The vast and varied inland water resources of India are potentially one of the richest in the world. Recently, inland fish production of India has gone up through adoption of improved technology and culture method. Fish culture is also important from ecological point of view because fish is the most efficient among farm animals in converting feed into nutritious food. Fishes have always been an important source of protein for human and there is ever increasing demand for more fish protein. However, outbreak of diseases hinders effort to increase fish production. Fishes are susceptible to various types of diseases. Fish disease is a global problem affecting fresh water and marine fish, wild, cultured, sport fish and even ornamental fish with large scale of mortality (Trust, 1986).

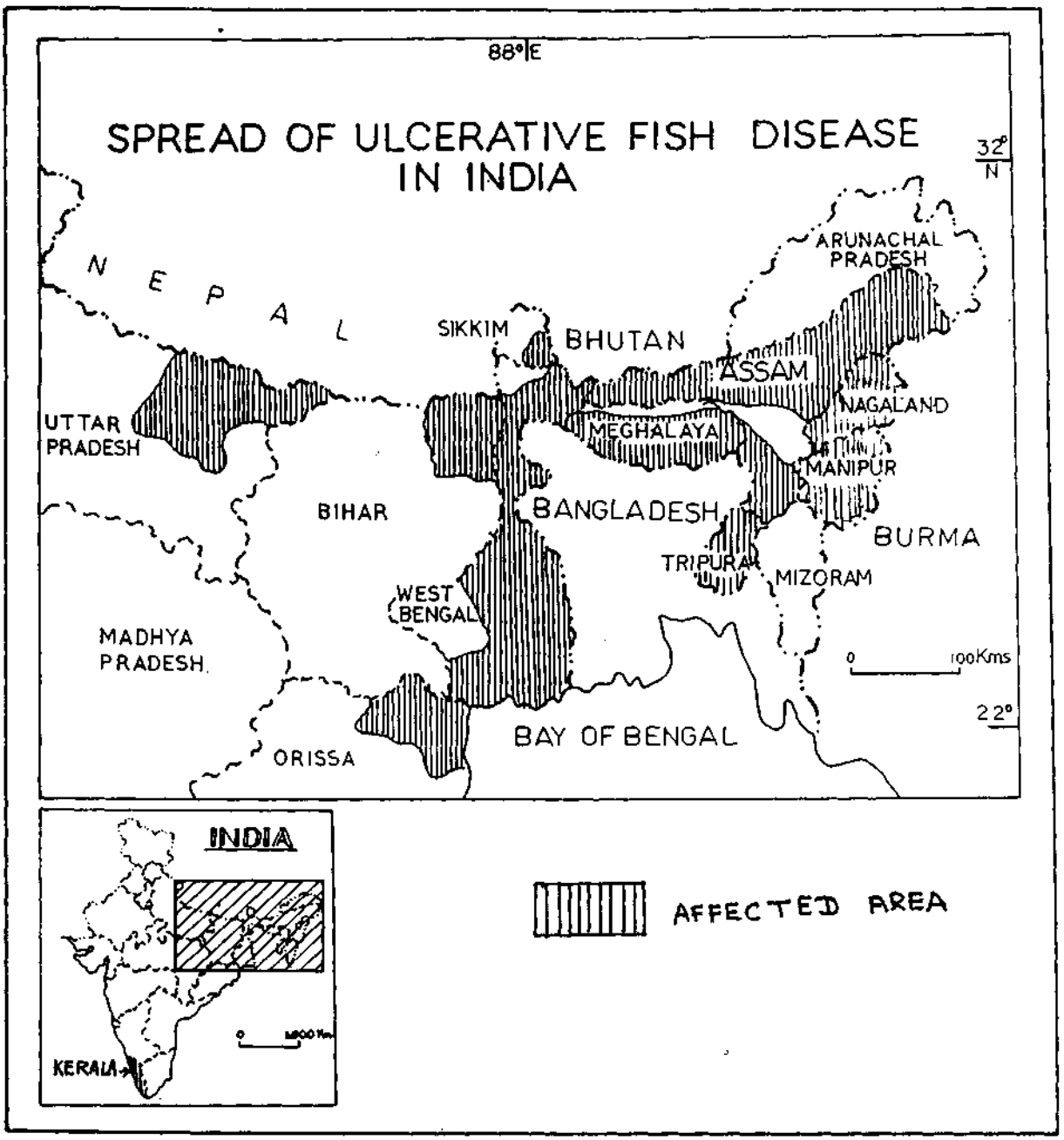
A severe outbreak of ulcerative fish disease in epizootic form affecting various types of fishes, wild and cultured, have occured for the first time in May, 1988 in various states of north-eastern India such as Tripura, Meghalaya and Assam. Subsequently, the disease has spread to some northern districts of West Bengal such as Cooch Behar, Jalpaiguri, West Dinajpur, Malda and plains of Darjeeling district (Das, 1988; Pal and Pradhan, 1990b). The disease has severely affected

almost all the districts of West Bengal except Purulia in 1989. A panic has been created among the people of West Bengal that the rate of consumption of fish declined. A sharp decline in the trade of fish in urban, sub-urban and rural market has been noticed after the outbreak of the ulcerative disease. The fish farmers and fisherman have suffered heavy economic losses. It has become a serious concern to the fishery scientists also. The situation has gone to such an alarming condition that the West Bengal Government and Ministry of Agriculture, Government of India have organised the "National Workshop on Ulcerative Disease Syndrome in Fish" at Calcutta, March 6-7, 1990. Scientists from all over India have participated in the workshop and exchange their views. Scientists have differed in their opinions regarding the etiology of the ulcerative fish disease. Pal and Pradhan (1990a) have opined that it is a mixed bacterial infection; Das et al., (1990) have indicated that the disease is caused by Micrococcus sp.; others have expressed it could be a viral infection (Prasad and Sinha, 1990).

This paper deals with i) Ulcerative disease in air-breathing fishes and isolation of bacteria from ulcer tissues, ii) Experimental induction of ulcer in fishes, Channa punctatus and Anabas testudineus by

bacterial culture, iii) Histopathological observation of liver, kidney and spleen of experimentally infected fishes, Clarias batrachus and Channa punctatus, iv) Effect of bacterial culture on Total erythrocyte count and Haemoglobin content in fish, Channa punctatus, v) Evaluation of the role of bacteria, $R_1$, $R_2$ and $R_3$ in causing ulcer disease, vi) Drug sensitivity testing and preliminary observation on vaccination by formalin-killed bacteria in mixed condition in fish, Anabas testudineus, vii) Ulcerative disease in Indian major carps and isolation of bacteria from ulcer tissues of Cirrihinus mrigala.

Air-breathing fishes are chosen for experimental studies for their easy handling and maintenance in laboratory. The above mentioned experiments were designed depending on the supply of healthy fishes, the supply of which was very irregular. Sometimes we have to wait months after months for getting a particular type of healthy fish. On several occasions even after purchase of healthy fishes without any external visible sign of the disease ulcers developed on the bodies of the fishes in the laboratory within 48 hours.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SPREAD OF ULCERATIVE FISH DISEASE IN INDIA
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BAY OF BENGAL
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AFFECTED AREA

Fig. 1.

Cooch Behar, Jalpaiguri, West Dinajpur, Malda and plains of Darjeeling district. The incidence of the disease has also been reported from some southern districts of West Bengal such as Murshidabad, Nadia, 24-Parganas and Midnapur (Pradhan and Pal, 1990). In 1989, the disease had severely affected also, almost all the districts of West Bengal except Purulia.

In 1990, the disease has spread in some areas of other states of India such as Orissa , Bihar, Uttar Pradesh, Sikkim, Manipur and Nagaland (Prasad and Sinha, 1990; Prusty and Nayak, 1990; Kumar et al. , 1991). At the end of August, 1991 reports of the disease have came from some areas of Kerala, a southern state of India. Though,at present,the declining trend of the disease has been reported we are still getting affected air-breathing fishes from local markets viz., Siliguri, Matigara, Shibmandir and Bagdogra of Darjeeling district, West Bengal.

## Species affected

The disease have affected both wild and culturable fresh water fishes (Table 1). The susceptible wild fishes are Clarias batrachus Heteropneustes fossilis, Anabas testudineus, Channa punctatus,

Table 1. Fish species affected by ulcerative disease.

| CULTURED | WILD |
|---|---|
| Catla catla, | Anabas testudineus, |
| Labeo rohita, | Heteropneustes fossilis, |
| L. calbasu, | Clarias batrachus, |
| Cyprinus carpio, | Channa punctatus, C.striatus, |
| Cirrhinus mrigala, | C. gachua, Chanda chanda, |
| Puntius javanicus, | Mastacembelus sp. |
| P. sarana, | Callichrous pabda, |
| Ctenopharyangodon | Mystus vittatus, Nandus |
| idellus. | nandus, Colisa sp. |
| | Puntius sophore, |
| | Amblypharyngodon mola, |
| | Ambassis ranga |
| | Glossogobius giuris |
| | Gadusia chapra |

C. striatus, C. gachua, Colisa sp., Puntius sophore, P. ticto, Mastacembelus pancalus, M. armatus, Amblypharyngodon mola, Ambassis ranga, Mystus vittatus, Nandus nandus, Glossogobius giuris, Chanda chanda, Gudusia chapra and the affected culturable fishes are Catla catla, Cirrhinus mrigla, Labeo rohita, L. calbasu, Cyprinus carpio, Puntius javanicus, P. sarana (Jhingran and Das, 1990; Pal and Pradhan, 1990b; Kumar et al., 1991).

The severely affected fishes are mainly air-breathing fishes like Anabas, C batrachus, H fossilis and Channa sp., Moderately affected fishes are major carps e.g. C. catla, C. mrigala, L. rohita, C. carpio.

Prior to the recent outbreak of ulcerative disease in epizootic form in 1988, several Indian workers have reported ulcerative skin lesion in fishes (Gopalakrishnan, 1963; Monohar et al., 1976; Pal et al., 1978, Pal, 1984; Karunasagar et al., 1986; Kumar et al., 1987a).

Since 1972 different countries of the Asia-Pacific region have witnessed severe ulcerative condition occuring suddenly and often causing mass mortalities in wild brackish water fishes (Roberts et al., 1986). In Queensland, Australia, the disease charac-

terised by shallow haemorrhagic ulcers is named "red spot disease" which affected marine and estuarine fishes in 1972 with recurrence in subsequent years (Rodgers and Burke, 1977, 1981). Fish disease characterised by dermal ulcer from the rivers of the south Papua, New Guinea in 1975-76 has been reported by Haines in 1983. In Indonesia the disease is known as infectious dropsy or haemorrhagic septicemia has spread to west central and eastern Java in 1980 and recurrence thereafter have been reported from Thailand (Tonguthai, 1985). Rahim et al., (1985) have reported bacterial haemorrhagic ulcerative disease from Bangladesh. Llobrera and Gacutan (1987) have reported ulcer disease in Phillipines. Costa and Wijeyaratne (1989) have recorded the ulcer disease in fishes in Sri Lanka.

Infectious disease of fishes are of common occurence. Causative agents of the infectious diseases are bacteria, virus, fungi and several groups of protozoan and metazoan parasites.

BACTERIAL DISEASES

One of the limiting factors in fish production is mortality due to bacterial infection (Bullock,1971; Wolke, 1975; Kabata, 1985). Bacteria which are ubiquitous in nature can enhance the disease under favourable condition such as poor nutrition, weakened immune system of the fish and poor water quality. Though a large number of pathogenic bacteria have been isolated from diseased fishes, the most and frequently occuring bacterial diseases are described below.

(i) Red Spot Disease

Wakabayashi and Egusa (1972) have reported the outbreaks of a new bacterial disease 'Sekiten byo'(red spot disease) in epizootic form during late spring to early summer of 1971 causing considerable mortalities among pond cultured eel populations in both Shizuoka and Tokushima prefectures, Japan. The characteristics symptom of the disease is a remarkable petechial haemorrhage on the body surface and the pathogen is Pseudomonas anguilliseptica. Muroga et al., (1973) have reported an epizootic in Japanese eel (Anguilla japonica) in Tokushima prefecture, Japan and the

pathogen is P. anguilliseptica. Jo et al.,(1975) have recorded red spot disease in the European eel in 1974. Miyazaki and Egusa (1977) have studied the histopathology of the Red spot disease of the Japanese eel. The histopathological studies have revealed that infected lesions appeared in dermis, subcutaneous adipose tissue, interstitial tissue of body musculature, vascular walls, bulbus arteriosus and heart. Various pathological changes have also been recorded in the visceral organs such as congestive edema and intensive fatty degeneration of the hepatic cells in the liver; serous exudation, tissue liquification and cellular proliferation in the spleen; infectious glomerulitis, activation of the reticulo-endothelial cells lining sinusoids and atrophy of the haematopoietic tissue in the kidney. Kuo and Kou (1978) isolated P. anguilliseptica from red spot disease of pond cultured eel A. japonica in Taiwan. Muroga and Nakajima (1981) have investigated different methods for artificial infection in Japanese eels (A. japonica). Nakai et al., (1981) have studied the serological properties of P. anguilliseptica isolated from eels (A. japonica and A. anguilla) of Japan. Nakajima et al., (1983) have isolated the pathogen from Black Sea bream Acanthopagrus schlegeli. Nakai et al., (1985a) have

studied the multiplication and distribution of P. anguilliseptica in artificially infected Japanese eel. Nakai et al., (1985 b) have recorded P. anguilliseptica infection in pond cultured ayu, Plecoglossus altivelis.

In 1981 red spot disease has occured in European eel (A. anguilla) in Scotland and this seems to be the first record of the disease in Europe (Nakai and Muroga, 1982; Stewart et al., 1983). Red spot disease has also been reported from Danish eel farm (Mallergaard and Dalsgaard, 1986). In Finland, P. anguilliseptica is responsible for severe disease outbreak in several species of farmed salmonid fish (Wiklund and Dalsgaard, 1987; Wiklund and Bylund, 1990).

(ii) Motile aeromonad disease

Aeromonas hydrophila has been regarded as a potential pathogen for various kinds of fishes. The infection of Aeromonas hydrophila was commonly known as 'Motile aeromonas septicemia' (AFS,1975). The disease has been named as red pest for European eels, A. anguilla (Schäperclaus, 1934), red disease for Japanesee eel, A. japonica (Hoshina, 1962), red disease for carp, Cyprinus carpio (Egusa,1978), red

sore for large mouth bass Micropterus salmonides (Huizinga, et al., 1979), Aeromonas disease of cyprinids (Takahashi, 1984b). An extensive work has been done to clarify the causes, infection, mechanism of attack and methods of prevention and treatment of the Aeromonas disease occuring in carp, gold fish, crucian carp and river dodger by Takahashi (1984 b). Infected fishes shows exopthalmus, cutaneous haemorrhage in the tail and anal region; and some times showed severe haemorrhage and ulceration on the body surface. Haemorrhage and necrosis were also seen in liver and kidney with haemosiderosis in liver, spleen and kidney (Miyazaki and Jo, 1985; Miyazaki and Kaige, 1985). Miyazaki and Kaige (1985) experimentally induce the disease in crucian carp by intra-peritoneal injection of the pathogen from naturally infected crucian carp.

Ulcer disease caused by bacteria has been reported in Indian major carps by Gopalakrishnan (1963). The symptom of the disease is the presence of open sores or ulcers on the body of the fish which gradually increase in size exposing the muscle. Karunasagar et al., (1986) have reported similar disease condition in Catla catla . Kumar et al., (1987a) have reported

several outbreaks of such disease. Presence of *Aeromonas hydrophila* in the ulcers is detected in all such cases.

(iii) Ulcer disease

This disease was first described by Calkins (1899) and was common among the various species of trout; the causative factor was gram negative motile rod, *Haemophilus piscium*. An extensive review on the ulcer disease of trout has been done by Mawdesley-Thomas and Jolly (1968). The primary symptom of the disease is the appearance of white tufts on the skin. The tufts often coalesce and ultimately an open sore is formed which may expose underlying musculature or, if over the body wall may erode into the peritoneum. Ulcer disease may also affect the fins, jaws and mouth, occasionally producing almost total destruction of the lower jaw.

Gold fish ulcer disease (GUD) was first described in England in 1969 (Mawdesley-Thomas,1969). The ulcer disease in gold fish appears to be widely distributed in the United States, United Kingdom and Japan (Elliot and Shotts, 1980); Hungary (Csaba, *et al.*,1984), Canada (Munkittrick and Leatherland,1984), Germany (Mirle, *et al.*, 1986); Bohm *et al.*, 1986) and Australia

(Hamilton et al., 1981; Whittington et al., 1987 Carson and Handlinger, 1988). In Gold fish ulcer disease the symptoms appear as skin lesions and haemorrhages with loss of scales. An atypical Aeromonas salmonicida is the causative agent of GUD.

Ohtsuka et al.,(1984) have isolated atypipal A.salmonicida from diseased eel and they tentatively have called it the head ulcer disease because of its unique sign of ulcerative lesions on the head part of the affected eels.

(iv) Vibriosis

Vibriosis is a infectious disease with a world wide occurence mainly in farmed and wild marine and brakish water fishes (Anderson and Conroy, 1970; Ghittino, 1972; Levin et al.,1972; Harrel et al., 1976; Mc Carthy, 1976; Muroga and Tatani, 1982; Ransom et al., 1984; Lewis, 1985). In Japan, Muroga and Egusa (1967) isolated the bacterium Vibrio anguillarum from ayu (Plecoglossus altivelis) in salt water lake of Hamana. The pathogen has also been isolated by other workers from diseased fishes (Anderson and Conroy,1970; Wolke, 1975; Horne, 1982; Lewis, 1985). The disease has

been reported in cultured ayu (Muroga and Egusa,1967), in Japanese eel, A. japonica (Jo and Muroga,1972; Miyazaki, 1980), in Yellow tail, Seriola quinquerata (Jo et al., 1979). Muroga et al., (1984) have detected V. anguillarum in wild ayu fingerlings. In Norway, the first outbreaks of cold water vibriosis (Hitra disease) have occured in the late seventies and the bacterium isolated from diseased fish is named as Vibrio salmonicida (Egidius et al., 1986).

The vibriosis is characterised with haemorrhagic septicemia and is frequently associated with superficial ulcers, haemorrhages at the bases of fins and bloody discharges from the vent (Bullock et al., 1971; Novotony, 1978). From histopathological observations in naturally and experimentally infected fishes (A. japonica), Miyazaki (1980) has mentioned that the characteristics of V. anguillicida are strong invasiveness into the tissue and the cause of septicemia in the terminal case. Lewis (1985) has mentioned that vibriosis appears to be a disease in which the agent is localised in selected tissues. Muroga and Cruz(1987) have concluded that the first colonization site of V. anguillarum in ayu is the skin.

(v) Furunculosis

Furunculosis is an infectious disease of numerous fish species caused by Aeromonas salmonicida (Mc Graw, 1952; Klontz, et al., 1966; Mawdesley - Thomas, 1967 and 1969; Mawdesley-Thomas and Jolly, 1968; Herman, 1968, Ghittino, 1972; Mc Carthy,1975; Miyazaki and Kubota 1975; Furguson and Mc Carthy,1978; Mc Carthy and Roberts, 1980; Ellis et al., 1981). There are two main types of furunculosis (i) acute type in which fish dies without showing any external signs of disease. (ii) sub acute type in which skin or muscle lesion seen in many places. Various types of histopathological changes in the gills, gastro-intestinal tract, kidney and spleen have been recorded by various authors (Mawdesley-Thomas and Jolly, 1968; Ghittino, 1972; Miyazaki and Kubota, 1975; Furguson and Mc Carthy, 1978; Boomker et al., 1984).

(vi) Bacterial Kidney Disease

It is a chronic systematic infectious disease caused by Renibacterium salmoninarum (Sanders and Fryer, 1980; Fryer and Sanders, 1981; Bruno and Munro, 1982). The gross external symptoms of this disease is

variable. In certain cases there is no sign of the disease, but in some cases it shows swelling of abdomen, exopthalmous, petechial haemorrhage and haemorrhage around the base of the fins (Mac Lean and Yoders, 1970; Hendricks and Leek, 1975; Kimura, 1978; Hayakawa et al. 1989). Fishes infected with Bacterial Kidney Disease shows various histopathological changes in kidney, liver and spleen. The kidney may be swollen, granular and purulent. Sometimes ascitic fluid may be present in the peritoneum and within the opaque membrane covering the internal organs like, Kidney, spleen and liver (Wood and Yasutake, 1956; Smith, 1964). Bruno (1986) has experimentally induced the disease in rainbow trout and Atlantic salmon with viable R. salmoninarum.

(vii) Enteric septicemia

Hawke (1979) has described a disease called Enteric septicemia of catfish externally characterised with haemorrhages, at the base of pectoral fins, on the lower edge of operculum. The causative factor has been identified as Edwardsiella ictaluri, an enteric bacterium. (Hawke et al., 1981; Chen and Kulmin, 1989). Miyazaki (1980) has described E. tarta infection in Japanese eel. Miyazaki and Plumb (1985) reported that

the disease can affect brain, liver, spleen and kidney of infected fishes. Initially, it is thought that the E.ictaluri is specific to ictalurids (Waltman et al., 1985). However, isolation of the bacteria has been reported from non ictalurid species (Kent and Lyons, 1982; Blazer et al., 1985; Waltman et al., 1985; Kasornchandra et al., 1987). Plumb and Sanchez(1983) have experimentally shown that tilapia Sarotherodon aureus is susceptible to E ictaluri. Baxa et al., (1985) have reported that the diseased red sea bream exhibited typical symptoms of Edwardsiellosis with haemorrhagic lesions on the caudal and dorsal fins and white spots on the liver and kidney. Two kinds of bacteria E. tarta and Staphylococcus aureus have been isolated from the diseased fish. E. tarta is pathogenic but S. aureus is non-pathogenic. Baxa et al, (1990) have shown that E. ictaluri is a potential pathogen of salmonid fishes also.

(viii) Myxobacteria and other external bacterial infection in fish

Infectious fish diseases like, "Columnar disease", "Peduncle disease, "Gill disease" and "Tail rot disease" are caused by bacteria (Mawdesley - Thomas and Jolly, 1968

The most common agent is Chondrococcus columnaris, but other organisms have also been considered for such diseases (Bullock, and Mc Laughlin 1970; Marks et al.,1980). But their exact pathogenic role in disease production is not clear. According to Ghittino (1970, 1972) they may play the part of pure opportunists. Marks et al.,(1980) isolated Corynabacterium sp. and Flexibacter columnaris from lesion of catfish associated with Columnaris disease. He has further noticed that the exposure of pure culture of these bacterium fails to develop Columnaris disease; but when catfishes are exposed to mixed culture of these two bacteria they develop sign of Columnaris as well as mortality occur. Kumar et al. (1986 c) have reported Columnaris disease in Labeo rohita caused by Flexibacter columnaris .

Roberts (1972) has reviewed the ulcerative dermal necrosis of Salmon, Salmo salar which had spread to most of the British rivers. The disease is characterized with various skin lesion from small oval greyish rough area of skin to large shallow haemorrhagic ulcer. The disease more or less resembles the columnaris disease caused by Chondrococcus columnaris. But exact aetiological agent for the disease is still controversial.

(ix) Red Mouth Disease

It is a disease characterised with inflammation of buccal cavity, reddening of mouth, throat and opercula. Internal organ like liver is usually pale, the spleen and kidney are congested and become dark in colour. In certain cases septicemia is found. The disease is caused by Aeromonas hydrophila (Mawdesley-Thomas and Jolly, 1968). Other bacteria such as A. liquefaciens and Yersinia ruckeri are also isolated from internal organs and blood (Snieszko and Bullock, 1965; Grawinski,1990).

(x) Other bacterial infection

Several other bacteria such as Streptococcus, Mycobacterium sp., Hafnia alvei are also common pathogen among marine and fresh water fishes. Streptococcal disease in fishes have been reported by several authors (Kusuda et al., 1976; 1978; Barham et al.,1979; Miyazaki, 1980, 1982; Kitao, et al., 1982; Rasheed et al. 1985; Michel, 1989). Miyazaki et al.,(1984) studied the histopathology of Streptococcal infection in cultured tilapia Sarotherodon niloticus. The symptoms of the streptococcal disease in tilapia was corneal

opacity, exopthalmous, destruction of eye balls, dark body colouration and cutaneous haemorrhage; internally the disease fish showed dropsy, epicarditis, peritonitis, pale colour of liver and splenomegaly.

Tuberculosis of fishes caused by acid-fast, gram-positive rod, Mycobacterium sp. is well documented (Mawdesley-Thomas and Jolly 1967, 1968 Ghittino, 1970). According to Ghittino (1972) Fish Tuberculosis, Fish Mycobacteriosis and Fish Nocardiosis (caused by Nocardia sp.) belong to the same group of disease as all have similar characteristics.

Miyazaki (1980) has described N.kampanchi infection in yellowtail. Based on external and anatomical views the disease has been classified into four manifestation types.

Kusuda et al., (1987) isolated fish pathogenic bacterium, Mycobacterium sp. from an epizootic occured in cultured yellow tail in Kochi prefecture, Japan. The sign of the disease were haemorrhage and abdominal ascities with hypertrophy of the spleen and kidney with tubercles. Bragg et al.,(1990) also isolated Mycobacterium fortuitum from three species of fresh water fish in South Africa.

Gelev et al.(1990) identified a bacterium Hafnia alvei responsible for epizootic haemorrhagic septicamia in rainbow trout. This organism exhibited some antigenic similarities with Brucella abortus and Yersinia ruckeri, but further studies have shown that the strain was H. alvei.

Kusuda and Takahashi (1970) have reported outbreaks of scale protrusion disease in carp fishes frequently observed in fish farms in Japan. They have identified Aeromonas liquefaciens as the causative agent and successfully induced the disease with the bacterium.

VIRAL DISEASE

Recently, piscine viral diseases have become a serious problem in aquaculture. (Ghittino et al.,1984; Trust, 1986, Meguro et al., 1991). Infectious carp dropsy or viral haemorrhagic septicemia needs special attention.

## Infectious carp dropsy or viral haemorrhagic septicemia

Different European countries, as for example, German Democratic Republic (Schäperclaus, 1930,1965, 1969), Rumania(St.Nicolau, 1951), USSR(Goncharov,1965), Yugoslavia (Tomasec, 1951; Tomasec and Fijan, 1965), Czechoslovakia (Volf and Havelka, 1965), France (Bellet, 1958, 1965), Italy (Ghittino, 1962, 1965), Poland (Kocylowski, 1965; Miaczynski, 1965), have witnessed haemorrhagic ulcer disease of fishes. The disease have been named differently by different authors, e.g. infectious carp dropsy (Schäperclaus, 1965), hydropigenic neurovirosa (St.Nicolau, 1951), rubella (Goncharov, 1965), popeye sickness, viral haemorrhagic septicemia (Ghittino, 1965; Bellet, 1965), carp septicemia (Kocylowski,1965) etc. At the conclusion of the first European symposium on fish disease, Turin, Italy, October 20-24, 1962, a committee of experts have opined that the disease should be named "Viral haemorrhagic septicemia" (VHS). The symptoms of the disease are external lesions, ulceration, exopthalmia and dropsy.

There are two conceptions regarding the etiology of infectious carp dropsy : bacterial and viral, Schäperclaus have started his investigations on infectious carp dropsy in 1920's in Germany and regarding

the etiology of the disease the bacterial conception has been founded by him (Schäperclaus, 1930). He has opined from his experimental work on infectious carp dropsy that A. punctata is the etiological agent of infectious carp dropsy and Pseudomonas fluorescens in a virulent form may act as predisposing agent (Schäperclaus, 1965). Volf and Havelka (1965) have mentioned that the infectious dropsy has been during the last three decades, the most trouble some disease of carp in Czechoslovakia and ascertained that besides P. punctata ascitae, P. fluorescens also is involved in infectious dropsy of carp. Goncharov (1951) has believed that the disease is caused by a virus, as the disease is induced by bacteria free filtrates prepared from the skin of diseased carp and presence of eosinophilic spherical inclusion bodies in the cytoplasm of epithelial cells and brain cells of sick carp. St. Nicolau (1951) has found intracellular inclusions in the brain. Ghittino (1965) also has reported presence of inclusion bodies in some nuclei and cytoplasm of hepatocytes of diseased fishes. Negative results of experiments in growing the virus of infectious dropsy on carp tissue have been published by Tec and Jakovleva (1962). Kocylowski (1965) has mentioned that the virus has a rather polyorganic affinity and low virulence and that healthy fishes infected with a filtrate of diseased carp tissue does not show specific changes

characteristics for the pathological entity. Bellet (1965) has also mentioned that tissue preparations for diseased fishes has caused no infection when filtered. Tomasec and Fijan (1965) have claimed development of cytopathogenic effects in cultured tissue by a filtrate of diseased carp tissue and transmission of disease into healthy fishes. Schäperclaus (1969) has mentioned that A. punctata, P. fluorescens and a virus may be involved in infectious carp dropsy. Fijan (1972) has mentioned that infectious dropsy in carp is a disease complex and it covers Spring Viremia of Carp (SVC), Carp Erythrodermatitis (CE) and several other diseases.

Recently, several workers have been able to isolate piscine viruses. The important isolates are : a birna virus, infectious pancreatic necrosis virus (IPNV) (Dobos, 1976; Dobos et al., 1979), three rhabdovirus, viral haemorrhagic septicemia virus (VHSV), infectious hamatopoietic necrosis virus (IHNV), the spring viremia of carp virus (SVCV) (Hill,1975), a harpes virus, channel catfish virus (CCV) (Pilcher and Fryer, 1980). All are responsible for systematic infections, often most fatal and preferentially produce disease early in the life of the fish (Ghittino et al. 1984). IPNV, VHSV and IHNV are significant pathogen of

salmonids. SVCV is a pathogen of the common carp and CCV (herpes virus) has an extremely restricted host range producing disease in young cultured channel catfish in the southern United States (Pitcher and Fryer, 1980). Herpes virus have been found in many fresh water fishes such as channel catfish Ictalurus punctatus (Wolf and Darlington, 1971), Kokanee Oncorhynchus nerka (Sano, 1976), rainbow trout (Wolf et al. 1978), Oncorhynchus masou (Kimura et al., 1981), wall eye Stizostedion vitreum (Kelly et al., 1983) and common carp Cyprinus carpio (Sano et al., 1985), Larvae and Juveniles of the Japanese Flounder Paralichthys olivaceus (Iida et al., 1989).

For detection of viral pathogens among fishes several workers established fish cell lines (Chen and Kou, 1981; Chen et al. 1982; 1983a,b; Yasushi et al., 1991).

MYCOTIC DISEASE

Reports are there regarding wide spread losses due to mycotic disease of fishes. Fungal parasites attack eggs, fingerling and adult fishes. They are probably secondary invaders following physical or

physiological injuries brought about by rough handling or attack by primary pathogen. In India, the incidence of mycotic disease, mainly due to the genera Saprolegnia, Brachiomyces and Achlya has been observed in major carps, catfishes, murrels and several other common species in different parts of the country (Gopalakrishnan, 1963; 1964 and 1966; Jhingran, 1974; Srivastava and Srivastava, 1976, 1977 a,b; Prabhuji and Srivastava, 1977; Kumar and Dey, 1991).

In Japan, Hatai and Kubota (1989) reported the visceral mycosis caused by Ochroconis. Some visceral mycoses in fish due to other fungi have also been reported in Japan (Hatai and Egusa, 1975; 1977; Hatai et al., 1986). In Kenya, a single type of systematic mycosis has been reported in tilapia farms; two species of Aspergillus, A.flavus and A.niger were involved (Olufemi et al., 1983).

## METAZOAN DISEASE

Disease caused by parasitic worms, leech and crustaceans are grouped under the category of metazoan disease. Although a large number of trematodes, cestodes and acanthocephlans have been described, in most cases

infestations are not severe enough to cause significent problem. However, in certain environmental condition these parasites can lead to epizootic. The disease due to metazoan parasite have been described by several authors (Fryer, 1968; Paperna, 1970, 1974, 1980; Rai and Pande, 1965; Sarig, 1971; Jhingran, 1974; Kabata, 1985; Morand, 1985).

## PROTOZOAN DISEASE

Protozoan parasite of fishes have been described by many workers (Chaudhuri and Chakravarty, 1970; Seenappa and Monohar, 1980 a,b; Mishra et al., 1982; Kabata, 1985; Landsberg and Paperna, 1987). The common diseases caused by protozoan parasites are ichthyophthiriasis, trichodinosis, costiasis, myxosporidiasis, and microsporidiasis.

## THERAPEUTIC USE OF DRUGS AND VACCINE AS CONTROL MEASURES FOR FISH DISEASES

Time to time many workers have used and suggested various drugs for controlling fish diseases.

Schäperclaus (1965) have tested the sensitivity of chloramphenicol, streptomycin, terramycin against different strains of Aeromonas and Pseudomonas isolated from carps suffered from Infectious carp dropsy. Volf and Havelka (1965) have mentioned that chloramphenical is used for controlling infections by A. punctata ascitae and in mixed infections (A. punctata ascitae and P. fluorescens) streptomycin is used and concluded that with full use of this treatment for 5 years in carp culture, the losses of fish are reduced from 20 percent to 5 percent. Bellet (1965) have concluded that sulpha drugs and antibiotics have not given valid results except in a very few cases e.g. chloramphenicol in the case of very young fry. Ghittino (1965) also mentioned about the failure of any specific treatment, antibiotics and sulfonamides for dropsy.

Wakabayashi and Egusa (1972) have studied the sensitivity of Pseudomonas sp. from pond cultured eel to different antimicrobial agent. Jo (1978) have concluded that oxonilic acid, nalidixic acid and piromidic acid administered by bathing or orally are found to be effective to control the disease. Wiklund and Bylund (1990) have shown sensitivity of Finish strain of P. anguilliseptica to different chemotherapeutants and have concluded that ampicillin and trimethoprim/sulphamethoxazole are most active against the pathogen.

Treatment methods by drugs for aeromonas disease have been investigated by Takahashi (1984 b) and concluded that dipping 20 ppm of chloram-phenicol or oxytetracycline for 30 minutes and oral administration of 100 mg/kg.b.w./day of sulfamonomethoxine or 10 mg/kg/day of oxolinic acid for 5 days are effective.

Kumar et al.(1991) have used antibiotic terramycin for controlling the ulcerative disease but did not show any effectiveness on the disease. Jhingran (1990) have recommended to use of antibiotics such as erythromycin or nalidixic acid or oxytetracycline or terramycin for ulcer disease. Antibiotics such as erythromycin, oxytetracycline/sulphamethoxazole have been recommended by Jain (1990) for preventive measurement of ulcer disease. Saha et al., (1990) have also recommended the use of bactrim, an antibiotic for ulcer disease.

The search for a successful vaccine has had a long history. A lot of work has been done on furunculosis vaccine.

Duff (1942) has observed that prolonged feeding with chloroform-killed Aeromonas salmonicida whole cells give protection to cutthroat trout (Salmo clarkii),whem

challenged artificially. Krantz et al., (1964 a) have reported that intraperitoneal injection of formalin-killed whole cells in a mineral oil adjuvant gives protective agglutinating antibodies, but a feeding technique is unsatisfactory (1964 b). Spence and Fryer(1965) have observed the serum from vaccinated rainbow trout (Salmo gairdneri) passively protect coho salmon (Onchorhynchus kisutch),but oral vaccination of formalin-killed whole cells fails to induce protection in the laboratory or the field. Paterson and Fryer (1974) have shown that a single parenteral vaccination with formalin-killed A. salmonicida whole cells in Freund's complete adjuvant gives protective immunity against furunculosis. Hara et al. (1976) have found slight protection in field trials with a soluble antigen administered in the food. Antipa and Amend (1977) have noticed that formalin killed I.P. and hyperosmotic (H.I) application did induce production of serum antibodies. Austin and Rodgers (1981) have reported good protection by formalin-killed whole cell vaccine when given orally. Rodgers and Austin (1984) have concluded that whole cell vaccines coupled with toxoid subcellular components give protection in rainbow trout against furunuclosis. Rodgers (1990) has shown that immersion vaccination fry of rainbow trout can be protected with a 3-component vaccine consisting of

whole cells, 'toxoided' extracellular product and lipopolysaccharide and that the protection level is significantly enhanced when the vaccine contains liposomes.

Nakai and Moroga (1982) have shown that vaccine consisting of Pseudomonas anguilliseptica produces a persistent and protective immunity in the eels. On the otherhand, the fish vaccinated by immersion with a sonicated bacterin does not develop agglutinating antibodies or protection.

Vaccination has proved to be highly effective against infections with Vibrio anguillarum (Evelyn, 1984) and Yersinia ruckeri (Bullock and Anderson, 1984). Lelillchaug (1990) has shown that vaccination by immersion against cold-water vibriosis in Atlantic salmon provides a high level of protection.

Takahashi (1984 b) has detected the antibodies in serum of carp, after inoculation with heat and formalin-killed cells of pathogenic Aeromonas. The antibodies have been detected for atleast 27 weeks after inoculation. Takahashi and Kusuda (1971) have studied about immune response of carp in relation to scale protrusion disease. Baba et al., (1988a) studied the cell mediated protection in carp, Cyprinus carpio against A. hydrophila. Baba et al., (1988b) have

observed that vaccination with crude lipopolysaccharide (LPS) has induced better protection against infection with A. hydrophila in carp than vaccination with formalin-killed cells. They also have mentioned that dipping in vaccine is more effective than intraperitoneal injection and antibodies are not detected in the dip method. They have concluded that the protection against A. hydrophila infection in carp is not dependent on humoral immunity.