

INFECTIOUS DISEASES OF SILVER CARP, *Hypophthalmichthys molitrix*  
(Valenciennes, 1844)

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## Introduction

Species of minnows and carp worldwide belong to the Order Cypriniformes, Family Cyprinidae. The order is usually classified in two suborders, 33 families, and about 5000 species. Cyprinids are one of the largest and most important groups of fish in the world, and they dominate fresh waters. The minnows and carp of Africa, Europe, Asia and North America include some 275 genera and over 1500 species, making the Cyprinidae the largest of all families of fish. Three families and 68 species of cyprinids occur in the fresh waters of Canada (Scott and Crossman, 1979).

Silver carp (*Hypophthalmichthys molitrix*) are large, laterally compressed cyprinids of a uniform silver color, and occur naturally in temperate waters in China. They have been introduced as a food fish and for water management strategies, in temperate waters of Asia, Europe and the USA. They were imported into Arkansas, USA in 1972.

Silver carp are similar to bighead carp (*Aristichthys nobilis*), but differ in having a longer ventral keel which extends from the insertion of the anal fin to the isthmus (as opposed to the pelvic insertion in bighead carp), in lacking the small irregular dark blotches possessed by bighead carp, and in having their gill rakers fused into a sponge-like porous plate (Robison and Buchanan, 1988). Specimens of 126 cm (57.27 inches) in length and up to 10 years of age have been reported from some waterways in Asia (Anon, 2003a).

Adult silver carp consume mainly phytoplankton and detritus, and for this reason, the species, along with bighead and grass carp (*Ctenopharyngodon idella*), has been used in aquaculture not only as a food fish, but also in water management to improve water quality. Typically, silver carp are unable to ingest phytoplankton smaller than 10  $\mu\text{m}$  (Herodek *et al*, 1989; Smith, 1989); they passively select for phytoplankton over zooplankton, based on particle size and the mechanical effects of their feeding apparatus. They feed actively only when

high concentrations of phytoplankton, detected through chemical cues, are present (Smith, 1989). Silver carp have a characteristic feeding behavior, swimming rapidly gulping water, closing their mouth, and pumping it out through the opercula. Ingested food is ground by pharyngeal teeth against a cartilaginous plate (Robison and Buchanan, 1988). Beveridge *et al* (1993) reported that silver carp can detect toxic strains of cyanobacteria (blue-green algae) by the presence of low levels (<250 ng/L) of microcystins (toxins) or related substances, and by modifying their behavior, avoid ingesting these algae.

In Canadian waters, the quillback (*Carpoides cyprinus*), an indigenous cyprinid species, is sometimes called a silver carp, among other names (Scott and Crossman, 1979).

As an aside, it is interesting to note that in September, 2002, two letters, one from the Council of Lake Communities, and the other from The Great Lakes Fishery Commission (GLFC), to the US Fish and Wildlife Service in Arlington, Va, asked that the black (*Mylopharyngodon piceus*), bighead and silver carp be listed as injurious species, to protect the Great Lakes from harmful invasive species, including other Asian species of carp (Appendix 1). Further, it is the view of the GLFC that all black carp, including triploids and eggs, should be destroyed, and that co-operation should be sought from Canada and Mexico to implement a seamless North American strategy for preventing invasion and range extension by Asian carp. If implemented and carried to their ultimate conclusion, these measures could have a major impact on US sources of silver and bighead and possibly, grass carp.

### Known Viral, Bacterial, Parasitic and Mycotic (Fungal) Diseases Associated with Silver Carp

Like other species of fish, silver carp are susceptible to a number of infectious diseases, several of them in common with other species. From a philosophical perspective in terms of infectious disease, I have taken the broad

view (perhaps in error) that many cyprinids may share similar susceptibilities to a number of infectious diseases. In other words, my point is that certain infectious diseases that occur in one family of cyprinids may occur in others of the 33 families of cyprinids, given the circumstances and opportunity for exposure to these various agents. Hence, I have included here, information on infectious viral, bacterial, parasitic and mycotic diseases of a number of cyprinids, and haven't limited the discussion to silver carp alone. For example, a number of infectious diseases of common carp (*Cyprinus carpio*) are included in the discussions.

It should be noted that a number of references are made to Lio-Po and Lim (2002) whose chapter in a larger publication refers to infectious diseases of (often caged) warm-water fish in fresh waters of south east Asia. Despite the focus of their work on warm-water fish, the principles of exposure, infection and the development of lesions associated with these infections are likely applicable to fish reared in other environments. Also in the same volume is a chapter by Brown and Bruno (2002) who deal with infectious diseases of cold-water fish in fresh water; reference to some of these conditions is made herein.

Disease does not happen in isolation. In order for disease to begin, the classical configuration of the interaction of the host, the agent and the environment must come into play. This idea suggests that it is only the opportunity for exposure (or lack of) to these various agents that determines whether these specific infections will occur. The interplay of the host, the infectious agent and the environmental conditions certainly affects the final outcome of these biological encounters (Hedrick, 1998; Reno, 1998).

In part, this document is based on the format, data and some references from a previous report dealing with infectious diseases of the European eel (*Anguilla anguilla*) (Chalmers, 2003).

## A. Viral Diseases

In general, systemic viral infections of fish are common and important. General characteristics of these infections include: 1) pathogenicity (the ability of a microorganism to produce disease) is often temperature-dependent; 2) the disease is frequently host-specific; usually younger fish become ill, older fish become carriers; 3) medications are not available – for control, only disinfection and quarantine can be tried (Noga, 1996). Viral infections can cause mass mortality especially in fry or fingerlings, whereas older fish may develop resistance or are scarcely affected. Most viral infections occur in fish held at low temperatures – which may explain the low level of viral infections recorded in fish in warm fresh water. Stress from handling, poor water quality, temperature, age of fish, high stocking density, and poor nutrition are factors that influence the development of viral diseases (Lio-Po and Lim, 2002).

In many parts of the world, cage culture is an increasingly favored method of rearing fish (Beveridge, 1996; Lio-Po and Lim, 2002) and in this environment, many of the factors just mentioned come into play. Among viral infections in fish, disease caused by the Channel Catfish Virus has the most impact on cage culture. In grass carp, the Grass Carp Hemorrhagic Virus, and in tilapia, the Spinning Tilapia syndrome are potential viral problems (Lio-Po and Lim, 2002).

### 1. Spring Viremia of Carp and Swim Bladder Infection

Spring viremia of carp (SVC) is caused by *Rhabdovirus carpio*, a virus that has been responsible for high mortality in common carp, the species of carp most susceptible to this infection. The disease occurs in geographic regions of the world with temperate climates, ie, continental Europe and the former USSR. Spring viremia of carp is primarily a disease of yearling common carp, but is

known to produce disease in many species of cyprinid fish, including koi (a strain of common carp), grass, silver, bighead, and crucian carp (*Carassius carassius*), goldfish (*Carassius auratus*), tench (*Tinca tinca*) and sheatfish (*Siluris glanis*). Very young fish of various pond species, including northern pike (*Esox* spp.), perch (*Perca* spp.) (Gaylon and Roth, 2003), and guppy (*Lebistes reticulatus* = *Poecilia reticulata*) (Crane and Eaton, 1997) are also susceptible.

In North America, known susceptible species include common, silver, bighead and grass carp, as well as goldfish, koi, plus two species of baitfish (fathead minnow, *Pimephales promelas*, and golden shiner, *Notemigonus crysoleucas*) (Goodwin, 2002). It is interesting to note that the virus of SVC was once suggested as a biological agent for the control of common carp in Australia (Crane and Eaton, 1997).

It has been suggested that the establishment of SVC in North America in the past may have been prevented because the virus causes disease only at cool temperatures (15-20°C), and that the infection is cleared efficiently by the immune system of fish when temperatures are higher. For example, most ciprinid production in the USA occurs in the southern part of the country, where water temperatures are higher than 30°C for prolonged periods of time, thus protecting these stocks (Goodwin, 2002).

Typically, this infection is characterized by darkening of the body, sluggish breathing, tilting to one side, exophthalmia (bulging eyes), anemia, reddening and swelling of the anus, mucoid casts trailing from the vent, abdominal swelling that is caused by ascites (the presence of excess abdominal fluid, also called “dropsy”), and tiny hemorrhages of the skin and gills (Brown and Bruno, 2002; Harper, 2002; Petty *et al*, 2002; Gaylon and Roth, 2003). The causative virus was first isolated by Fijan *et al* (1971) from common carp affected with infectious dropsy in Yugoslavia.

In 2002, Goodwin reported the first outbreak of SVC in North America, in a commercial operation rearing koi in Kernersville, North Carolina on the east coast of the USA (APHIS, 2002). The producer had noted moderate mortality,

characterized by severe ascites and external hemorrhages, in koi from several ponds. These signs were consistent with those of SVC, but could also have been associated with common bacterial infections and severe parasitic infestations (Jeney and Jeney, 1995). Of 150,000 koi in four ponds of infected fish, 15,000 died, and the remaining 135,000 fish in these ponds were destroyed (APHIS, 2002)

Goodwin (2002) reported that common carp, koi, goldfish and golden shiners, held at 20°C, and injected with fluids containing SVC virus isolated from this outbreak, died in 3-7 days. Mortality varied from 100% for koi and common carp, to 80% for other species tested. Common carp were most susceptible, with 100% mortality in 4-10 days at 15-25°C, but no mortality at 30°C. Mortality of goldfish and golden shiner was 50-66% at 15 and 20°C, and zero at 25 and 30°C.

According to Harper (2002), infection of cyprinids can occur at 20-22°C, but because there is a good immune response at these temperatures, clinical disease does not occur. In water temperatures <10°C, the activity of the immune system is reduced and viral replication is slowed, but the pathogenicity (the ability of a microorganism to produce disease) remains high, and the disease can become fatal. Once SVC is established in a pond, it can be very difficult to eradicate unless all forms of aquatic life at the site are destroyed (Gaylon and Roth, 2003).

Swim Bladder Infection (SBI) is caused by one of several subtypes of *Rhabdovirus carpio*. In this infection, there is pronounced inflammation and hemorrhage of the swim bladder (Moeller, 2001). Fijan (1976) detailed the lesions found in the swim bladder: initially the inner wall may be turbid and slightly thickened ventrally; as the disease progresses there are a wax-like deposit, congestion, hemorrhage and post-hemorrhagic pigmentation; the anterior, posterior or both chambers may be enlarged or reduced in size and deformed; necrotic foci develop on the wall; the swim bladder may contain a

serous, bloody or pus-like fluid; the swim bladder may be completely destroyed, with a cyst formed around it.

It has been suggested that cyprinids that are indigenous to Alberta should be examined for evidence of SVC (Johnson, 2003).

## 2. Infections Caused by Herpesviruses

Herpesviruses are the most abundant group of DNA viruses found among fish, but only one member of Herpesviridae, *Herpesvirus cyprini* (cyprinid herpesvirus, CHV) has been isolated from cyprinids. It was isolated from and later demonstrated to be the cause of carp pox (epithelioma papillosum, papillosum cyprini), a seasonal (and generally believed to be non-fatal) disorder of the skin, and characterized by mucoid to waxy white to yellow epidermal tumors on common and koi carp; healed lesions are usually black (Plumb, 1999; Hedrick *et al*, 2000; Moeller, 2001). This agent was once thought to be a benign condition of older carp only, but Sano *et al* (1991) showed the lethal nature of this virus in koi and common carp under two months of age. Fry of other carp species, including crucian and grass carp, and the willow shiner (*Gnathopogon elongatus caerulescens*) were resistant to experimental infection.

Hedrick *et al* (2000) reported the isolation of a herpesvirus associated with mass mortalities in juvenile and adult koi in the USA and Israel. This agent was determined to be different from *H. cyprini* on the basis of fluorescent antibody tests with anti-*H. cyprini* serum. It had the capability of inducing severe branchial (gill) and systemic disease in koi. Hedrick *et al* (2000) found that this herpesvirus in koi (KHV) had certain characteristics in common with CHV, but the two agents could be distinguished by differences in the specificity of cell lines affected, type of cytopathic effect (CPE), and antigenic properties. In laboratory trials, in contrast to CHV, KHV caused disease in older fish. Severe branchial necrosis (death and destruction of the gills) was the main external sign in affected fish from both the USA and Israel. At the electron microscopic level,

however, the systemic nature of the disease became evident, with viral particles evident in two main sites: branchial epithelium and liver of affected fish. As well, with use of a newly developed cell line (KF-1) from koi, this herpesvirus was isolated from gills, liver and other internal organs from many fish in both countries.

However, Dawes (2002) noted that research in Israel (manuscript apparently *in press*) indicated that the causative agent of this condition may not be a herpesvirus but rather, a double-stranded DNA virus with icosahedral morphology that makes it resemble a herpesvirus. Hedrick (2003) has some reservations about the veracity of this claim. The debate continues.

### 3. Infectious Pancreatic Necrosis

Although it induces disease primarily in salmonids, the causative virus of Infectious Pancreatic Necrosis (IPN) has been isolated from a number of non-salmonid species of fish. According to Noga (1996), IPN virus can infect at least one member of 20 families of fish, including carp, pike, poeciliid (guppy), etc., and has been isolated from oysters, crabs and digenean trematodes (flukes). In almost all cases, the isolates from these crustaceans and trematodes have not been shown to be pathogenic for the host, although on occasion, they have been pathogenic for trout, findings that indicate that these species are most important in acting as non-susceptible reservoirs of the virus. Citing several authors, Ahne and Thomsen (1985) noted the isolation of IPNV from eels, tilapia, and goldfish, among other species.

### 4. Golden Shiner Virus

This agent belongs to the Family Reoviridae. In recent years, the genus *Aquareovirus* was created to include reoviruses isolated from fish and shellfish. Golden Shiner Virus (GSV) is the type species for this new genus. In general, aquareoviruses have low or no pathogenicity for fish. However, GSV and

several others are capable of causing significant mortality. This disease appears to be confined to the southeastern area of the USA where golden shiners are reared commercially. The virus has also been isolated from grass carp, but other species of fish are refractory (Plumb, 1999).

#### 5. Grass Carp Hemorrhagic Disease

This disease was seen first in China in the 1980s. The causative virus is currently classified in the Family Reoviridae, genus *Aquareovirus*. Although it usually affects grass carp, the disease can also infect black carp, and can replicate in silver carp without producing clinical signs. Outbreaks occur in southern China during the summer when temperatures range from 24-30°C. Acute infections cause significant mortality of more than 80% in fingerlings and up to 70% in yearlings. Signs include exophthalmia and severe hemorrhages of the gills and the base of fins. Internally, hemorrhages occur in muscles, oral cavity, liver, spleen, kidneys and intestines. Disease and mortality are seen within 1-2 weeks of exposure of fish to the virus in water temperatures of 25°C or more. Experimental vaccination with inactivated virus induced 80% protection by Day 4 at temperatures above 20°C, by Day 20 at 15°C, and by Day 30 at 10°C (Plumb, 1999; Lio-Po and Lim, 2002).

#### 6. Other Viral Agents of Carp and Minnows

As reported in Plumb (1999), an iridovirus and a herpesvirus have been reported from goldfish. In addition, in Hungary, a rhabdovirus was isolated from two-year-old apparently healthy grass carp but its role in the health of grass carp is unknown.

## B. Bacterial Diseases

### 1. Diseases Caused by *Edwardsiella* spp.

Two members of this group of organisms infect fish: *Edwardsiella tarda* [formerly called *E. anguillimortifera* and *Paracolobactrum anguillimortiferum* (Noga, 1996)] and *E. ictaluri*. These bacteria produce two different diseases.

*Edwardsiella tarda* causes septicemia (invasion and multiplication in the bloodstream) in warm water fish, particularly in eels and catfish (*Ictalurus punctata*). It is widely disseminated in aquatic animals, pond water and mud, occurrences that provide ready opportunities to re-infect cultured fish. Infected fish processed for human consumption are a source of this organism, which can cause gastroenteritis in humans.

*Edwardsiella ictaluri* causes a septicemia in catfish, and is a highly contagious disease with serious effects on the commercial culture of catfish (losses from 10-50%) in the southern USA (Inglis *et al*, 1993; Noga, 1996).

*Edwardsiella* sp. septicemia is a mild to severe systemic disease of mainly warm water fish in the USA and Asia. It is caused by *E. tarda* and is also called fish gangrene, emphysematous putrefactive disease of catfish, and Red Disease of eels. Catfish and eels, notably Japanese eels (*Anguilla japonica*) [but not reported from American (*Anguilla rostrata*) or European eels (*Anguilla anguilla*)], and catfish, are the most commonly infected species. However, the organism has been isolated from a variety of species of fish, including goldfish, common and grass carp, tilapia, etc. (Noga, 1996).

In the USA, *E. tarda* has been isolated from 75% of water samples holding catfish, 64% of mud samples from ponds holding catfish, and 100% of frogs, turtles and crayfish from ponds containing catfish. The source of this organism is likely intestinal contents of carrier animals. Catfish and eels, as well as amphibians and reptiles, are likely sources of infection. Although environmental stressors don't appear to be essential for infection to occur, high temperature,

poor water quality and crowding are likely contributing factors. Infections caused by *E. tarda* are not confined to fish, but are also found in snakes, alligators, sea lions, birds, cattle, swine and humans (Inglis *et al*, 1993; Noga, 1996).

It is an important zoonotic disease of humans in which it is a serious cause of intestinal disease. In humans, it has also been implicated in meningitis, liver abscess, wound infections; most commonly, however, this organism causes gastroenteritis. Catfish fillets in processing plants are often contaminated with this organism that may spread to humans by the oral route (Noga, 1996). In an earlier study, Brady and Vinitnantharat (1990) injected live catfish with *E. tarda* or *E. ictaluri*, *Aeromonas hydrophila*, and *Pseudomonas fluorescens*, and when the injected fish died or were moribund, froze them at  $-20^{\circ}\text{C}$ . They found that *E. tarda* could be recovered on culture for 50 days, *E. ictaluri* for 30 days, *A. hydrophila* for 20 days, and *P. fluorescens* for 60 days, in these frozen fish.

## 2. Diseases Caused by *Yersinia* spp..

*Yersinia ruckeri* is the cause of enteric redmouth (ERM) disease, a condition that has been known since the 1950s. ERM has been a problem mainly for cultured rainbow trout (*Oncorhynchus mykiss*), but all salmonids and some other species of fish are affected. Losses may be relatively low in chronic infections or can become much higher if water conditions are poor, or if fish are exposed to stresses such as handling. As fish become asymptomatic carriers relatively easily, ERM remains a constant threat to salmonid culture (Inglis *et al*, 1993). The organism is widespread in fresh-water environments (Noga, 1996.)

A table indicating various sources of isolation of the ERM organism was provided by Inglis *et al*, (1993). These sources included 1) salmonids, 2) diseased fish such as emerald shiners (*N. atherinoides*) and fathead minnows (*P. promelas*), 3) apparently healthy fish including goldfish, common carp and European eels, along with 4) other sources such as muskrats, feces of predatory and carrion-feeding birds, crayfish, gulls, humans, sewage and river water. The

first outbreak of ERM in Canada (Wobeser, 1973) followed the importation of apparently healthy fish from Idaho, an observation that suggested the presence of asymptomatic carriers in the shipment.

There are six serovars of the organism, Types I to VI, with Type I the most common, although not all Type I serovars are pathogenic. High environmental temperatures (15-18°C) can induce carriers to begin shedding, and signs in susceptible fish can become evident within several days of the stress. Mortality may be as high as 70% initially (Noga, 1996).

In its early stages in salmonids, ERM disease resembles aeromonad and vibrio infections, with darkening of the dorsum, lack of appetite and lethargy. Internally, there are tiny hemorrhages on the viscera, enlargement of the spleen, and necrosis of the inner surface of the intestine and a mucoid exudate. In the more chronic condition, the abdomen is distended, there maybe unilateral or bilateral exophthalmos, and hemorrhage (blood spot) in the eyes. The latter causes darkening of the fish because of the induced blindness which leads to lack of control of melanin pigment (Noga, 1996; Brown and Bruno, 2002).

### 3. Diseases Caused by *Flexibacter (Cytophaga) spp.*

Bacterial gill disease (BGD) is caused by a variety of bacteria including *Flexibacter columnaris*, *F. psychrophilus*, *Cytophaga psychrophila*, and various species of *Flavobacterium* (Moeller, 2001). Usui (1991) described bacterial gill disease (BGD) caused by an organism he identified as *Chondrococcus columnaris* -- more recently called *Flexibacter columnaris* -- in cultured Japanese eels. Currently, a suggested name for this agent is *Cytophaga columnaris*, although it has also been called *Flexibacter columnaris* (Inglis *et al*, 1993; Noga, 1996). In common terms, the disease it causes is often called Columnaris Disease or BGD. Whereas *F. columnaris* infects fish only in fresh water environments, *F. maritimus*, an organism that seems to be less infectious than *F. columnaris*, infects fish in marine environments, where it produces salt-water

columnaris disease. *Flexibacter maritimus* has an absolute requirement for sea water (Inglis *et al*, 1993).

Farkas (1985) reported the presence of a filamentous *Flavobacterium* sp. from sheatfish, silver carp and rainbow trout affected by gill disease in cold-water environments.

*Flexibacter columnaris* can likely infect most fresh water fish and usually causes disease when water temperatures are higher than about 15°C. Crowding seems to be a factor in the occurrence of this disease. Experimentally, crowding hastened infection and mortality in fish exposed to *F. columnaris*, by reducing resistance of the exposed fish, and by increasing the chances of contact between fish and this agent (Inglis *et al*, 1993).

According to Inglis *et al*, (1993), and Plumb (1999), known cyprinid species affected by this organism include goldfish, common and grass carp, and other Asian carp, tench and sheatfish, shiners and fathead minnows, along with non-cyprinids such as Japanese (*Anguilla japonica*) and European eels, tilapia (*Oreochromis* sp.), pike (*Esox* sp.), and rainbow (*Oncorhynchus mykiss*) and brook trout (*Salvelinus fontinalis*).

Plumb (1999), noted that cyprinids are extremely susceptible to infections by columnaris organisms. Both mortality and acuteness of disease increase with temperature (Noga, 1996).

The main effects of this organism in fish occur in epithelial tissues, ie, skin and gills, where it causes erosions that may even ulcerate and become systemic. It is often seen as whitish plaques that may have a red edge on head, back and/or fins (fin rot). The typical “haystack” appearance of the colonies, plus the characteristic gliding motion of these organisms can be seen on microscopic examination of smears of affected gills or other tissues (Noga, 1996).

#### 4. Diseases Caused by *Aeromonas* spp.

These bacteria occur widely in fresh water and sewage. For example, Henebry *et al* (1988) found that the most common bacterium in the gut of young silver carp fed alternately on manure-silt and algal sources of food was *A. hydrophila*. Some species of *Aeromonas* are pathogenic to fish, and occasionally, to humans.

According to Noga (1996), motile aeromonad infection (MAI) is likely the most common bacterial disease of fresh-water fish, all of which are probably susceptible. Motile aeromonads can also inhabit brackish water, but they decrease in prevalence with increasing salinity. MAI has been associated with several of the *Aeromonas* sp. group which are widespread in fresh water environments. However, only a few of the aeromonads have been strongly documented as true pathogens of fish.

In cyprinids, including common, grass, silver and bighead carp, and golden shiners, fathead minnows and goldfish (Plumb, 1999), MAI may be peracute, with few signs, or acute with hemorrhages associate with ulcerative lesions of the skin, and may also be seen on the surface of organs or deep within tissues. In the chronic condition, there may be large, long-standing ulcers, and often associated with ascites (Fijan *et al*, 1971; Inglis *et al*, 1993) .

By far the most important pathogen of fish is *A. hydrophila* (synonyms: *A. liquefaciens*, *A. formicans*). This group is often described as the *A. hydrophila* complex. *Aeromonas hydrophila* is one of the agents involved in Carp Dropsy which is caused primarily by a virus (Inglis *et al*, 1993). Motile aeromonads are common on the mucosal surfaces and internal organs of clinically normal fish, and are often secondary invaders in infections such as those caused by *A. salmonicida*. Kumar and Dey (1985) reported on septicemia caused by *A. hydrophila* in silver carp.

*Aeromonas salmonicida* causes a fatal outbreak of disease called furunculosis in salmonids. "Furunculosis" is a term borrowed from a human condition. However, the changes seen in salmonids affected by this condition do not resemble the pus-filled swellings on the skin of humans affected by classical furunculosis. Despite these differences, the designation persists because it is too well established in scientific literature to be changed (Inglis *et al*, 1993).

According to older literature, infection by *A. salmonicida* subspecies *nova* has been indigenous in cyprinids, especially in common carp. Likely more than salmonids, cyprinids and other ornamental fish and baitfish are transported live around the world, and thereby provide a mechanism for the transfer of *A. salmonicida* to naïve populations of other species of fish. In cyprinids, this infection starts as a skin lesion that progresses to multiple lesions in the skin, and then becomes systemic. Very often, other bacteria become involved, making unclear, the actual cause of death (Inglis *et al*, 1993).

i) Ulcer Disease is the most common form of *A. salmonicida* infection in nonsalmonids, but salmonids can also be affected. Unlike furunculosis, ulcer disease is typically localized to the skin, and becomes systemic only late in the disease. In this condition, the lesions range from whitish discolorations to shallow hemorrhagic ulcers to deep lesions that expose the muscle or bone. These lesions are often secondarily infected by water molds, protozoa, and other bacteria. There may be hemorrhage on the body and at the base of the fins (Noga, 1996).

ii) Carp Erythrodermatitis (CE), a skin infection, is an important disease in cultured carp in Europe, and is caused by atypical *A. salmonicida*. It is part of the infectious dropsy of carp (IDC) complex which involves two diseases in cultured carp. The acute form of IDC is now known to be caused by *Rhabdovirus carpio*, whereas the chronic form of IDC is caused by atypical *A. salmonicida* (Fijan, 1976; Noga, 1996). Carp Erythrodermatitis generally occurs in spring following stocking of production ponds, with less severe outbreaks in autumn. Negligible losses occur in the summer. This condition is associated with

optimum water temperatures (15-20°C) and with spawning activities that that can lead to injury to the skin. When temperatures fluctuate between 6 and 20°C losses of carp caused by this agent can be greater than 50%. Young carp are highly susceptible to CE, but the severity changes from an acute infection to a subacute and/or chronic infection as fish become older (Plumb, 1999).

Further information on furunculosis and other diseases caused by *A. salmonicida* in fish may be found in the paper by Cipriano and Bullock, 2001.

As a point of interest, it is useful to note that infections caused by *Aeromonas* sp. in humans have been known since the early 1950s (Mathewson and Dupont, 1992). The most common manifestation of *Aeromonas* sp. infections in humans is bacteremia (the presence of bacteria circulating in the bloodstream). As well, wound infections in humans are becoming more commonly reported in the scientific literature. Their importance is related to the fact that they can have fatal or seriously debilitating results, such as amputations of affected limbs (Musher, 1980). Accordingly, wound infections should not be washed in river or pond water!

##### 5. Diseases Caused by *Pseudomonas* spp..

*Pseudomonas* spp. are commonly associated with fish, and are found on eggs, skin and gills, and in the intestines. In fish, the presence of this organism, among others, likely reflects the microbial population of the environment, and is influenced by factors such as bacterial load of the water and salinity. Because *Pseudomonas* spp. are so widespread and numerous, they may become involved in disease and act as secondary invaders if fish that are affected by pathogens or other factors). It seems likely that all species of fish are susceptible to *Pseudomonas* spp. under adverse environmental conditions, or when they are compromised by other factors. High quality of the environment, reduced stocking densities and other management strategies are important in preventing these infections (Inglis *et al*, 1993).

Some *Pseudomonas* spp., such as *P. anguilliseptica* and *P. fluorescens* have been reported to be primary pathogens (Inglis *et al*, 1993). Csaba *et al* (1984) and Petrinic *et al* (1986) described septicemia caused by *P. fluorescens* in silver and bighead carp in Hungary. Lio-Po and Lim (2002) reported epizootic outbreaks of infection caused by *P. fluorescens* in grass, silver and bighead carp, and Nile tilapia. Signs include ascites, exophthalmia, septicemia (multiplication of bacteria in the blood stream) and ulcers. The infection may be acute or chronic; the latter is often associated with skin lesions. *Pseudomonas fluorescens* is known to be part of the normal flora in the intestines of tilapia (Lio-Po and Lim, 2002).

*Pseudomonas anguilliseptica* was first described as the cause of a severe septicemia (called Sekiten-byo --means "red spot disease") of cultured Japanese eels in Japan (Usui, 1991). Japanese eels are more susceptible to this infection than are European eels, but the disease has occurred in other areas of the world. This disease has been controlled in Japan and Scotland by raising the temperature of the water to 26-27°C for about two weeks. An effective bacterin (= vaccine) has produced protective immunity to this disease in eels (Inglis *et al*, 1993). Experimentally, this organism was found to cause disease in common and crucian carp, goldfish, and several other species of fish (Inglis *et al*, 1993).

## 6. Epitheliocystis (Mucophilosis)

This condition may be associated with *Chlamydia* or *Rickettsia* spp., which are obligate intracellular organisms. Moeller (2001) has given a brief description of the condition as a chlamydial infection characterized clinically by respiratory distress or the secretion of excess mucus. Signs may also be completely absent. In affected fish, multiple white cysts are seen on the skin and gills. Microscopically, these cysts consist of distended epithelial cells filled with numerous organisms.

Epitheliocystis appears in several forms related to the number of cysts present and the response of the host. The response to cysts may be benign with no reaction by the host, or there may be a limited localized proliferative reaction. As well the host can react with a proliferative response involving a widespread severe proliferation of epithelial cells. In common carp, the proliferative response may be seen when only a few cysts are present (Paperna and de Matos, 1984). Differential diagnosis includes infestation by the parasite *Ichthyophthirius multifiliis*, and lymphocystis disease. However, unlike lymphocystis disease, epitheliocystis can also infect salmonids, catfish and cyprinids (Noga, 1996). Along with various salmonid species and tilapia, common, bighead and grass carp have been affected by epitheliocystis (Molnár and Boros, 1981; Inglis *et al*, 1993).

#### 7. Diseases Caused by *Streptococcus* spp..

Streptococcal septicemia has occurred sporadically and as epizootics among cultured fresh-water and salt-water fish in many parts of the world. The streptococcal species most commonly involved is *Streptococcus iniae*. It is mainly a disease of tilapia, hybrid striped bass (*Morone saxatilis* x *M. chrysops*) (Stoffregen *et al*, 1996), and rainbow trout. The known cyprinid species that are affected include golden shiner and blue minnow (*Fundulus grandis*) (experimental). Those species that don't appear to be affected by this agent include common carp, big-mouth buffalo (*Ictiobus cyprinellus*), goldfish, and some species of tilapia (eg., *Sarotherodon mossambicus*, *Tilapia sparrmanii*) (Inglis *et al*, 1993). However, Johnson (2003) noted that *S. iniae* is a serious problem in some operations rearing tilapia.

Signs of this illness vary among different species of affected fish. Erratic swimming, darkening of the body color, exophthalmos, corneal opacity, hemorrhages on the opercula and the base of fins, and ulceration of the skin are the most common signs (Inglis *et al*, 1993). Because *S. iniae* is a common

concern in rearing tilapia, and is a zoonotic disease (one that is transmissible to humans), knowledge of this organism is important (Johnson, 2003).

#### 8. Diseases Caused by *Salmonella* spp.

Several species of aquatic animals, including snails, clams, oysters, newts, frogs, crayfish, turtles, alligators, crocodiles and fish, have been known to carry *Salmonella* spp. (several references cited by Bocek *et al*, 1992). Souter *et al* (1976) found enteric (intestinal) bacteria in a study of common carp and white suckers (*Catostoma commersoni*) from five locations, four in Ontario and one in Quebec. As well, *Salmonella enteritidis* serotype Montevideo was cultured from the intestines of fish netted in the St Lawrence river at Montreal. It is notable that in 1975, the International Joint Commission reported that four of the five areas sampled did not meet water quality objectives in 1974 (Souter *et al*, 1976).

An experimental study by Bocek *et al* (1992) determined that silver carp could retain a streptomycin (antibiotic)-resistant strain of *Salmonella typhimurium* in their intestines. However, other internal organs such as kidney were not affected. Isolations of this organism from intestine occurred for 14 days after exposed fish had been placed in clean water. These findings and those of Souter *et al* (1976) indicate the potential for the contamination of aquatic environments, and the transmission of *Salmonella* spp. and other enteric pathogens of humans to other species of fish in the same environment, and by extension, to humans consuming these fish. Many species of *Salmonella* may infect humans.

#### 9. Diseases Caused by *Mycobacterium* Spp..

Mycobacteria, consisting of a single genus, *Mycobacterium*, are currently represented by at least 54 recognized species of organisms. Most of these agents are free-living in soil and water, and some species cause disease in animals and humans. Mycobacterial infections of fish are really tuberculosis of a

number of species. The disease affects a wide range of fresh water and marine species of fish, and particularly aquarium fish, especially the fresh-water families *Anabantidae* (climbing gouramies), *Characidae* (piranhas, tetras, etc.) and *Cyprinidae* (Noga, 1996). However, it seems likely that any species of fish may be infected. Mycobacteriosis is a chronic systemic disease, with lesions (granulomas) developing externally and throughout internal organs.

The species of *Mycobacterium* that are pathogenic for fish are *M. marinorum*, *M. fortuitum* and *M. chelonae*. Treatment is not satisfactory, and diseased stock should be destroyed, especially since these agents can infect humans as well as fish (Inglis *et al.*, 1993).

*Mycobacterium marinorum* represents the largest proportion of all mycobacteria isolated from fish. Tropical fresh water and tropical marine fish may be infected, but natural infection in a temperate-water species has not been reported (Inglis *et al.*, 1993).

The isolation of *Mycobacterium fortuitum* has been documented less frequently than that of *M. marinorum*, but the prevalence of infection by *M. fortuitum* is likely more widespread than is suspected. This organism infects fish from both tropical and temperate waters, but is most common in fresh-water fish, although infection is known to occur in marine species (Inglis *et al.*, 1993).

So far, infection by *M. chelonae* has been identified only in cold-water salmonid species. This infection has been specifically linked to fresh-water hatchery environment, but once established, it seems to persist throughout both fresh and salt-water phases of the cycle (Inglis *et al.*, 1993).

The main signs of this illness depend on the species of fish involved and the existing ecological conditions (Inglis *et al.*, 1993). The common findings are listlessness, lack of appetite, emaciation, difficult respirations, exophthalmia, skin discoloration and external lesions that range from loss of scales to nodules, ulcers and necrosis of fins as signs of advancing infection (Inglis *et al.*, 1993). In cold-water salmonids, there may be no external sign of the disease other than mortality, or variable degrees of skin coloration. Internally, lesions are similar in

tropical and cold-water fish. Visible or microscopic tiny gray-white changes may be found scattered in any tissue, but especially in spleen, liver and kidney.

Mycobacteria that are pathogenic for fish can infect humans, in which the lesions are usually localized, non-healing ulcers (fish tank granuloma, swimming pool granuloma) that may be difficult to treat because of resistance by the causative organisms to most anti-tuberculosis drugs. Although the risks to healthy humans is low, infections caused by *M. marinum* have been reported from HIV-infected individuals. Accordingly, gloves should be worn by individuals who are at risk when cleaning aquaria or handling fish (Noga, 1996). Johnson (2003) too has warned of the zoonotic dangers to workers working with species of carp.

#### 10. Miscellaneous Bacterial Diseases

In several species of carp, including grass and silver carp, Katoch *et al* (2001) reported a variety of bacterial species recovered on the culture of tissues: *Aeromonas hydrophila*, *A. caviae*, *A. salmonicida*, and *Staphylococcus*, *Pseudomonas*, *Citrobacter*, *Yersinia*, and *Vibrio* spp..

Noga (1996) listed a number of miscellaneous bacterial diseases that can affect cyprinids. These diseases include:

- 1) the important streptococcal infections;
- 2) infections caused by *Staphylococcus aureus* (reported once in silver carp in India);
- 3) nocardiosis (resembles tuberculosis);
- 4) pseudokidney disease (*Lactobacillus piscicola* = *Carnobacterium piscicola*;
- 5) also *Lactococcus piscium* and *Vagococcus salmoninarum*) in salmonids, common carp, etc.;
- 6) fin rot caused by *Pseudomonas fluorescens*, *P. putida*, *P. putrefaciens*, *P. pseudoalkaligenes*, *Pseudomonas* sp. in goldfish, silver, grass, bighead and black carp, tench and many other species;

7) Sekiten-byo caused by *Pseudomonas anguilliseptica* in Japanese and European eels, and experimental infections in common and crucian carp, goldfish, tilapia, etc.;

8) *Citrobacter freundii* in carp, rainbow trout, etc.;

9) *Proteus (Providencia) rettgeri* in silver carp. Bejarano *et al* (1979) described mass mortalities caused by *P. rettgeri* in silver carp after handling. This organism is normally found in the gut of poultry; feces of poultry were used extensively to fertilize ponds containing carp.

10) Also, *Mycoplasma mobile*, the only *Mycoplasma* sp. recovered from fish, was isolated from the gills of tench, but has not yet been shown to be pathogenic to fish, even though it can damage gill tissues cultured *in vitro*.

### C. Parasitic Diseases

Several species of parasites affect marine and fresh water species of fish. These agents include protozoa (microscopic one-celled organisms), trematodes (flukes/monogenean/digenean parasites), cestodes (tapeworms), nematodes (roundworms), acanthocephalans (thorny-headed worms), and crustaceans (a large class of creatures with hard outer shells, such as lobster, crab, shrimp, woodlice, water fleas, barnacles, etc.). A number of those parasites that affect species of carp include the following:

#### 1. Protozoa

These parasites are the most primitive animals on earth, and are comprised of a single cell that is capable of metabolism, reproduction and individual existence. Over 65,000 protozoan species have been described and named, and have adapted to every possible ecological existence (Post, 1987).

## Phylum Sarcomastigophora

### i) Trypanosoma spp.

Although uncommon in cultured fish, these parasites are found commonly in the blood of free-living fresh and salt-water species of fish over the world. They are found especially in cold-water species such as European carp and tench (Noga, 1996). They can cause anemia, damage to the red cell-producing tissues, and death. Common pathogenic trypanosomes include *T. carassi* (*T. danliewski*) that infects goldfish, common carp, and some non-cyprinids. *Trypanosoma borreli* causes anemia (sleeping sickness) in goldfish, koi, and other cyprinids. Trypanosomes are transmitted by leeches; the only known method of treatment is to control the population of leeches. (Noga, 1996).

### ii) Hexamita (Octomitis) and Spironucleus spp.

*Hexamita* spp. and the closely related flagellate, *Spironucleus* have been associated with gastrointestinal disease in salmonids and aquarium fish. Flagellates are often found in the gut of cyprinids such as grass carp, and occasionally in bighead and common carp. Predisposing stress appears to be a triggering factor, since *Hexamita*, *Spironucleus* and similar flagellates often reside in the intestinal tract of clinically normal fish (Noga, 1996). *Spironucleus* also infects grass carp and other cyprinids (Molnár, 1974). *Spironucleus* and *Hexamita* spp. have similar morphology and one is often mistaken for the other (Lom and Dykova, 1992). In grass carp, losses caused by an organism described initially as a *Hexamitus* sp., and later classified as *Spironucleus*, were characterized by reddening of the posterior two thirds of the gut. In one affected grass carp confined to an aquarium, there were ascites, exophthalmia, lifting of the scales, brown discoloration of the liver, and the gut was teeming with *Spironucleus* sp. (Molnár, 1974).

Through continued studies in Hungary, Molnár (1971) determined that, among the parasitic fauna of the fry of grass, silver and bighead carp, protozoa played the most important pathogenic role and could cause considerable losses among these fry. The identities of the protozoan species he found in these carp are listed in Table 1:

Table 1.

<u>Protozoan Parasite</u>	<u>Grass carp</u>	<u>Silver carp</u>	<u>Bighead carp</u>	<u>Cyprinids in Habitats in Hungary</u>
<i>Cryptobia branchialis</i>	+	+	+	
<i>Spironucleus</i> sp.	+			
<i>Chloromyxum nanum</i>	+			
<i>Sphaerospora carassi</i>	+			
<i>Myxobolus dispar</i>	+			+
<i>Myxobolus pavlovskii</i>		+	+	
<i>Myxobolus drjagini</i>		+		
<i>Myxidium</i> sp.	+			
<i>Chilodonella cyprini</i>	+	+	+	+
<i>Ichthyophthirius multifiliis</i>	+	+	+	+
<i>Balantidium ctenopharyngodonis</i>	+			
<i>Trichodonella epizootica</i>	+	+	+	+
<i>Trichodina</i> sp.	+	+	+	+
<i>Apiosoma cylindriciformis</i>	+	+	+	
<i>Trichophrya sinensis</i>	+	+		

### Phylum Ciliophora

Certain species of the genera *Ichthyobodo*, *Hexamita*, *Oodinium*, *Amyloodinium*, *Trypanosoma*, *Trypanoplasma* and *Cryptobia*, etc., are potentially

pathogenic. All *Ichthyobodo*, *Oodinium*, *Amyloodinium* and some species of *Cryptobia* are ectoparasites that occupy the skin, fins or gills of fish (Post, 1987).

Other pathogenic organisms in this group include *Chilodonella*, *Trichodina*, *Tripartiella*, and *Trichodenella* spp.. Large numbers of this group of parasites affect goldfish, common, grass, silver and bighead carp, and were introduced into Israel and Southeast Asia from China. According to Lio-Po and Lim (2002), parasites in this group commonly cause mortality in warm-water hatcheries and may continue to cause problems after fish are transferred to cage-culture systems. They are found commonly on grass, silver and bighead carp in hatcheries in Vietnam and China (Lio-Po and Lim, 2002).

Pathological effects of these parasites depend on the response of the host, the intensity of the infection and environmental conditions, since stressful conditions can affect the ability of the host to respond. Some of these parasites live specifically on body surfaces or the gills, whereas others are found on both skin and gills. When the skin is involved, the preferred sites are the base of fins.

i) *Ichthyophthirius* spp.

*Ichthyophthirius multifiliis* is well known to the aquarium industry as the cause of «Ich» and is one of the most common diseases of fresh water fish, virtually all of which are likely susceptible to this infection. Outbreaks of this condition are common at 15° to 25° C, but may develop at low (<10° C) temperatures in spring when fish are stressed from overwintering. «Ich» cysts appear as small white nodules that produce a salt-like dusting, but in heavy infestations can coalesce to form mucoid masses on the skin. Fish so affected are unlikely to survive (Noga, 1996). Molnár (1971) found grass, silver and bighead carp affected by this parasite in Hungary (Table 1).

ii) Chilodonella spp.

*Chilodonella* spp. are known to affect grass carp (AAFRD records) and to cause infections of the skin and gills; in large numbers they can be pathogenic. Most *Chilodonella* spp. are free-living, but two species, *C. piscicola* (formerly *C. cyprini*) and *C. hexasticha*, are pathogenic for fish. Molnár (1971) found grass, silver and bighead carp affected by *C. cyprini* in Hungary (Table 1). *Chilodonella piscicola* infests virtually all fresh water fish, mainly fingerlings, and can also infest fish in brackish waters. *Chilodonella hexasticha* is less widely distributed but produces similar lesions, mainly in older fish (Noga, 1996).

Chilodonellosis is more insidious than «Ich» and can cause severe damage before visible changes are evident. Advanced infestations are sometimes associated with ulcers in the skin ; these ulcers may have a tattered appearance. High numbers of organisms may cause secondary bacterial infections and considerable mortality (10% per week).

Chilodonellosis has a wide temperature tolerance: in cold-water species of fish, outbreaks often occur at 5° to 10°C, whereas tropical varieties of fish are affected when the temperature drops to 20°C ; outbreaks can also occur at high temperatures (Noga, 1996). Mass mortalities caused by *C. hexasticha* have occurred in free-living species of fish (Langdon *et al*, 1985). Some free-living species of *Chilodonella* can damage weakened fish in polluted waters.

Horwath *et al* (1978) found that copper oxychloride at a concentration of 4 mg/L was effective in eliminating *Trichodina* and *Chilodonella* spp. from infected grass carp, and *Trichodina* sp. from silver carp. Salt-water baths are also effective in removing *Chilodonella* spp. from affected fish.

iii) Amyloodinium spp.

*Amyloodinium ocellatum* is the cause of Marine Velvet Disease and can also affect certain species of fish such as tilapia in brackish water (Noga, 1996). *Piscinoodinium pillulare*, an organism that is closely related to *A. ocellatum*, is the cause of Velvet Disease, Rust Velvet Disease, Gold Dust Disease, Pillularis Disease and fresh water *Oodinium* Disease. Like its marine relative, *A. ocellatum*, *P. pillulare* is found on a wide range of host species, and is known to cause mortality in warm-water fish. It has been reported on 14 species of tropical ornamental fish as well as on carp and cultured cyprinids, including grass and bighead carp in pond culture. In cyprinids, it causes Velvet Disease, and was first reported in pond-reared fish, but is now also found on cage-reared species. Skin and gills are the sites of infection. Affected fish have a covering of excess, dense mucus as well as a yellow to rusty (velvety) appearance to the skin. Signs of the illness include darkened skin, respiratory distress and ulcers of the skin (Lio-Po and Lim, 2002).

Phylum Apicomplexa

i) Coccidia

The most important coccidian parasites of fish are those that affect solid tissue. Virtually all of these coccidian parasites belong to the Family Eimeriidae and include *Eimeria*, *Epieimeria*, *Goussia*, *Crystallospora*, *Calyptospora*, *Octosporella* and *Isospora* spp.. Certain species affect only the intestines; others affect both intestinal and extraintestinal tissues; as noted, some species affect extraintestinal tissues only. Nearly 200 species of eimeriids have been identified in fish, and it seems likely that their prevalence is underestimated. They are uncommon problems in most cultured fish, but have caused serious disease in some species such as common carp (Noga, 1996).

a) Intestinal Forms:

*Goussia subepithelialis* is found in common carp in which it is a common cause of nodular coccidiosis (Studnicka and Siwicki, 1990, cited by Noga, 1996). Another *Goussia* sp., *G. carpelli*, is a parasite of the intestines of cyprinids, particularly common and crucian carp in Europe and the USA. It is most severe during overwintering stress, and can also cause high mortalities in fry of goldfish after the stress of transport. Changes include necrosis (death of tissue), ulceration and diffuse hemorrhagic enteritis. Two other *Goussia* spp., *G. cheni* and *G. mylopharyngodon* are similar, and are found in herbivorous east Asian cyprinids (Noga, 1996).

Molnár (1971) found two species of coccidia, *Eimeria cheni* and *E. sinensis*, in both silver and bighead carp in Hungary (Table 1). In 1978, Molnár reported a study of infection by *E. sinensis* in silver and bighead carp cultured in ponds. The main gross changes were swelling of the inner surface of the intestine, accompanied by a coating of viscous mucus, in which many “yellow bodies” containing oöcysts were found; hemorrhage or inflammatory lesions were not seen, even in massive infections.

*Goussia iroquoina* is found in the intestines of fry of *Notropis*, *Pimephales* and other species of minnows in Canada (Paterson and Desser, 1982).

b) Intestinal and Extraintestinal Forms:

*Calyptospora funduli* affects the viscera and skin of the cyprinids, topminnow (*Fundulus* sp.) and silverside (*Richardsonia* sp.) in the coasts of the USA Atlantic and Gulf of Mexico. In heavy infections, white or black foci of oöcysts are found in liver; it also infects pancreas, fat, mesentery, ovary, gall bladder and skin (Noga, 1996).

### c) Extraintestinal Forms

*Eimeria rutili* infects the kidneys of roach in Eurasia, and causes tubular necrosis (Noga, 1996).

### Phylum Myxozoa

#### i) *Sphaerospora* spp.

A *Sphaerospora* sp. has been suggested to be the cause of proliferative kidney disease (PKD) of salmonids, since fish that are recovering from this infection often have sporoblasts resembling those of a *Sphaerospora* sp. (Kent and Hedrick, 1985). These findings provide evidence that PKD may be caused by the prespore stage of a myxosporean parasite (Noga, 1996). There is speculation that stickleback (Family Gasterosteidae) and common carp may be the definitive (final) hosts (Kent and Hedrick, 1986).

*Sphaerospora renicola* affects the swim bladder, kidneys and blood of common carp and goldfish in Eurasia. This parasite causes inflammation of the swim bladder, with hemorrhage and thickening, plus locomotor dysfunction, peritonitis (inflammation of the body cavity), and swelling of the kidneys (Lom and Dykova, 1992).

*Sphaerospora molnari* infects the skin and gills of common carp and goldfish (Europe, Israel and USA). This infection causes proliferation and necrosis of the epithelium, and can be fatal (Paperna, 1991).

*Sphaerospora tincae* affects the kidneys of tench and results in enlarged kidneys that are visible externally (Lom and Dykova, 1992).

ii) *Myxobolus* spp.

*Myxobolus* is the most species-rich genus of myxosporean parasites, and contains about 450 described species, most of which are relatively nonpathogenic. However, some species may be severe pathogens in either wild or captive fish. Cyprinids are host to about 40 species of *Myxobolus*, of which *M. cyprini* is among the most potentially pathogenic species that infect cyprinid fish (Kent *et al*, 1996). *Myxobolus cyprini* has been reported from many species of cyprinids throughout Europe and the former Soviet Union, and in China. Included are the following affected species: common carp, loach (*Rutilus rutilus*), chub (*Leuciscus cephalus*) and tench.

Kent *et al* (1996) provided the first report of *M. cyprini* in the western hemisphere, in peamouth (*Mylocheilus caurinus*), a cyprinid species from the lower Fraser river in British Columbia. (Its range also extends into western Alberta.). The source of this infection is unknown. Whether this parasite is endemic to North America or was imported to the Fraser river with exotic cyprinids from Europe or Asia, is not known. The latter is a possibility since thousands of cyprinids such as koi carp and goldfish are imported to Vancouver annually. As the Fraser river traverses metropolitan Vancouver, it is probable that some of these cyprinids have been released into the river; populations of common carp and goldfish are now established in the Fraser river system. In addition, it is possible that the organism from peamouths is very closely related to, but distinct from *M. cyprini* from Europe. Molecular comparisons of ribosomal RNA sequences in organisms from both sources could establish this point (Kent *et al*, 1996).

*Myxobolus cyprini* was described in common carp by Molnár and Kovács-Gayer (1985) who noted that this parasite is seen frequently in common carp, and less frequently in other species of carp. Most authors have regarded *M. cyprini* to be of pathological significance in cyprinids. It is a parasite of striated

(skeletal) muscle where it causes rupture of the fibres it invades. Spores from the ruptured muscle are released into the adjacent connective tissue, after which most gain entry to the blood stream. Those reaching the skin and gills are released directly to the environment; those from the intestines, liver and kidneys reach the external environment via the intestines, kidneys or bile ducts. Spores without direct access to the exterior are destroyed by the host.

Hoffman (1990) published a review of whirling disease of salmonids and included a list of *Myxobolus* spp. that affect a variety of species of fish. Two of these *Myxobolus* spp. found in silver carp are *M. drjagini*, which affects the central nervous system (but not the cartilage), operculum and oral cavity, and *M. pavlovskii* which affects the gills.

Akhmerov(1954) (in Russian, cited by El-Mansy and Molnár,1997) provided a description of *M. drjagini* in silver carp from the Amur river in China. Mass mortalities caused by infection of the brain, nervous system and sensory organs in yearling and older silver carp have been reported from China (Odening, 1989). Molnár (1971) (Table 1) reported this parasite in silver carp introduced into Hungary, where it has caused severe infections in populations of yearlings of this species. El-Mansy and Molnár (1997) reported on their experimental studies in which they infected the oligochaete, *Tubifex tubifex*, with mature spores of *M. drjagini*. Like *M. cerebralis* in salmonids, *M. drjagini* in silver carp has a two-host life cycle involving the fish and tubificid worms.

*Myxobolus pavlovskii* is found in the gills of bighead and silver carp in Asia and Europe. Despite the fact that it causes destruction of the gill arches, *M. pavlovskii* seems to be a well host-adapted parasite (Molnár, 1971 [Table 1]; Molnár, 1979). This parasite was imported to Hungary in 1963 in 2-3 cm silver and bighead carp from natural habitats in China and was distributed country-wide with the movement of fry (Molnár, 1979). Severe outbreaks of this condition in farm ponds often accounted for massive losses among populations of silver and bighead carp. However, a direct lethal effect of *M. pavlovskii* could not be confirmed because it occurred in mixed infections with other pathogenic

parasites, especially *Eimeria sinensis*, and the gill parasites *Cryptobia branchialis*, *Chilodonella cyprini* or *Trichodina* spp.. After treatment for these parasites, even massively infected fish of both species survived for several months in aquaria under good conditions of feeding and ventilation. However, death invariably occurred if the parasitic cysts filled at least half of the spaces between gill lamellae. Cysts adhering to the lamellae reduce the available respiratory surface area and thereby, reduce oxygen consumption. Although the infection is most massive in fry, it has been found in all age classes of silver and bighead carp (Molnár, 1979).

In their discussion of *M. corneus* in the eyes of bluegills (*Lepomis macrochirus*), Cone *et al* (1990) referred to *M. ellipsoides* which forms cysts in the cornea of tench in Europe.

*Myxobolus koi* has been found in goldfish and koi, in which it infects connective tissues of the subcutaneous tissues of the head and gill filaments where it causes proliferative branchiitis; it can be a fatal condition (Noga, 1996).

*Myxobolus exiguus* is known to infect cyprinids and mullet (Family Mugilidae) in Africa, Asia and Europe. It infects the skin, gills, stomach and pyloric ceca, and has caused mass mortalities in mullet (Noga, 1996).

Two other known myxobolid parasites of common carp are *M. basilamellaris*, which is found at the base of gill filaments or in the gill arches, and *M. dispar*, located in the tips and medial areas of the gill filaments (Kovács-Gayer and Molnár (1983). Molnár (1971) reported this parasite in grass carp in Hungary (Table 1).

Cone and Frasca (2002) reported on the revised diagnosis, site of development, and lesions caused by *M. hyborhynchus* in fathead minnow in Minnesota. These authors found that lesions caused by this parasite in fathead minnow resembled those reported for *M. cerebralis* in salmonids. Further, they also noted that natural infections by *M. hyborhynchus* in fathead minnow more closely resembled those of *M. cerebralis* in brown trout than they do similar infections in rainbow trout.

In the USA, *Myxobolus notemigoni* affects the skin of golden shiner, in which the pseudocysts lift scales and produce a bristled appearance that reduces their commercial value as bait fish (Noga, 1996). As well, Mitchell (1989) reported that cyprinids and salmonids in the Columbia river system in western Montana harbored five members of the Myxobolidae in several species of fish, including two species of cyprinids, peamouth and red shiner (*Richardsonius balteatus*).

Other myxozoan parasites of common carp include :

*Myxobolus encephalicus* (encephalitis, locomotor disturbances and emaciation) (Lom and Dykova, 1992);

*Myxobolus artus* which was found in the skeletal muscles of common carp in Japan (Ogawa *et al*, 1992) ;

*Thelohanellus nikolskii* (*T. cyprini*) (pseudocysts affect the rays of the fins which may break off, thus allowing secondary infections and impaired locomotion) (Molnár, 1982, cited by Noga, 1996);

*Thelohanellus kitauei* (pseudocysts occlude the intestines and cause emaciation and pressure atrophy of adjacent viscera) (Lom and Dykova, 1992).

*Hoferellus cyprini* affects the kidneys of koi and common carp which become infected by eating infested tubificid worms (Anon, 2003b). In carp this condition produces distension of the abdomen and exophthalmos. Another *Hoferellus sp.*, *H. carassii*, produces similar lesions in goldfish and gibel carp (*Carassius auratus gibelio*) (Molnár *et al*, 1989; Noga, 1996).

In addition, *Chloromyxum cristatum* (*C. cyprini*?) causes necrosis of the liver in cyprinids (Lom and Dykova, 1992); *C. barbi* causes infection of the gall bladder of silver carp, and has also been found in grass, black and bighead carp. In China, toward the end of their first year of life, there was 90% mortality of silver carp infected with this parasite (Odening, 1989).

*Zschokkella nova* affects the liver of goldfish and other cyprinids, in which the organisms distend the bile ducts and cause atrophy of the liver (Lom and Dykova, 1992).

Podosporosis caused by *Podospora hypophthalmichthydia* Chen and Hsieh, 1984, was found in the intestinal wall of silver carp in China (Odening, 1989).

Spirosuturiosis caused by *Spirosuturia hypophthalmichthydis* Chen and Hsieh, 1987, was described in silver, bighead and grass carp in China. In silver and bighead carp, the kidneys and especially the muscles are affected. Infection of the muscles in two-year-old silver carp can be serious and can lead to massive losses (Odening, 1989)

Molnár (1993) discussed the role of chemotherapy, including the use of fumagillin and toltrazuril in the treatment of myxosporean infections in fish. Prolonged feeding of fumagillin appears to be the only effective method that can be used successfully against several myxosporean infections: sphaerosporosis of common carp, hoferellosis of goldfish, myxidiosis of eels, and proliferative kidney disease, whirling disease and ceratomyxosis of salmonids.

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*Dermocystidium* spp. are a poorly studied group of organisms that produce varying sized (0.1 to 4.0 mm) white nodules on the skin or gills of many species of fish, including carp. These organisms are likely related to protozoa or fungi but their actual classification is unclear (Noga, 1996 ; Anon, 2003b). Some species produce hypha-like (branch-like) structures, and have caused mortality in salmonids and other species of fish (Noga, 1996).

## 2. Phylum Acanthocephala

In practical terms, acanthocephalan (thorny-headed worms) infections may be found in wild-caught fish, but in cultured fish they are rare because of their complex life cycle that requires one or two intermediate hosts. Fish may be intermediate or final hosts, depending on the acanthocephalan species involved (Noga, 1996).

Taraschewski (1998) reported on the prevalence rates of *Acanthocephalus anguillae* in several species of fresh water fish in Germany. He found a prevalence of 84% in chub (*Leuiscus cephalus*), 44% in tench, a cyprinid species, 34% in bream (*Abramis brama*), 30% in the European eel, 16% in roach (*Rutilus rutilus*), and 15% in bleak (*Alburnus alburnus*), and described the light microscopic and electron microscopic changes that occurred in these species

## 3. Phylum Cestoda

One of the most serious adult tapeworms affecting fish is the Asian tapeworm (*Bothriocephalus acheilognathi*, formerly known as *B. gowkongensis*), a cyprinid parasite. Because it has an unusually wide host range which includes minnows, golden shiner, various carp species including grass, silver and bighead carp, channel catfish, and possibly aquarium fish, the Asian tapeworm has the potential to be a very serious problem (Lio-Po and Lim, 2002). This tapeworm is not confined to imported carp, but has spread to native fish in warm waters of Asia. It was introduced into Israel with grass, silver and bighead carp (Lio-Po and Lim, 2002), and into the USA with grass carp (Hoffman, 1980), where it has caused serious problems with producers of bait minnows. It is widely distributed in Europe (Odening, 1989). The adult is large and can cause up to 90% mortality in grass and juvenile common carp (Noga, 1996). Worms accumulate in the

anterior intestine where they can cause obstruction or perforation, and high mortality.

The completed life cycle requires about one year in warm water, but development stops at <12°C. Worms mature in about 21 days at 28°C and in two months at 15°C. The larval stages (plerocercoids) are transmitted by several genera of copepods (intermediate hosts), and distribution depends on the abundance of these intermediate hosts (Noga, 1996).

In addition to *B. gowkongensis*, adults of *Khawia* sp. (*K. sinensis*) are found in common and grass carp (Odening, 1989). The intermediate host for *Khawia* sp. is tubificid worms. Both parasites can be devastating in young fish (Anon, 2003b).

In general terms, fish infested with adult tapeworms have retarded growth, erratic swimming behavior, a distended abdomen, become emaciated, cease to feed, and develop a hemorrhagic enteritis as a result of destruction of the surface epithelium of the intestines, and heavily infested fish develop excess fluid (ascites) in the body cavity (Lio-Po and Lim, 2002).

#### 4. Phylum Trematoda

The most important monogenean parasites (flukes) in cultured fish are the viviparous (live-bearing) gyrodactylids and the oviparous (egg-laying) dactylogyrids. They are external parasites. Monogenean parasites usually affect the gills, oral cavity, body surface and fins where they browse on debris from the skin and gills (Post, 1987; Lio-Po and Lim, 2002). The gyrodactylids are primarily parasites of the skin and gills; the dactylogyrids are parasites of the gills (Lio-Po and Lim, 2002; Noga, 1996). It is likely that all fresh water and many marine teleost fish of the world act as hosts to at least one species of *Dactylogyrus* or *Gyrodactylus*. The two species may be found together on cultured fish, especially if fish are crowded and unhealthy, or in water of poor quality (Post, 1987). Although they rarely cause disease in wild stocks, these parasites are

important pathogens in intensive culture operations. Disease caused by these parasites is usually more debilitating than fatal, and fatalities are usually associated with secondary bacterial or viral pathogens (Lio-Po and Lim, 2002).

Most monogenean parasites are specific to a group of related host species, eg, *Dactylogyrus* are found on cyprinids. The translocation of monogenean parasites along with their hosts, has been well documented for the various *Dactylogyrus* species on imported Chinese carp (Lio-Po and Lim, 2002).

With a few exceptions, *Dactylogyrus* spp. are specific to the cyprinids; they are often listed as a disease-causing agent since cyprinids are the most cultured group of fish. Some species of *Dactylogyrus* flourish at low water temperatures (16-17°C), whereas others prefer warmer temperatures (20-24°C). Common, grass, silver and bighead and other southeastern Asian species of carp are affected. *Dactylogyrus* spp. have been shown to cause mass mortalities in fry, small fish and brood fish (Lio-Po and Lim, 2002).

Anchoring and feeding on surface epithelium, these parasites cause severe destruction of the gills, and result in hemorrhage and significant changes to epithelium of the gills. In turn, these changes interfere with the normal respiratory function of the gills. Secondary bacterial infections usually occur and result in death of the fish. *Dactylogyrus* spp. usually affect the gills, but in severe infestations they can affect the oral cavity as well (Lio-Po and Lim, 2002).

Chemical treatments are effective if caged fish are reared in ponds, but not if cages are in rivers or large bodies of water (Lio-Po and Lim, 2002). Complete elimination of these parasites is usually impossible; only a reduction in numbers is likely to be achieved (Post, 1987).

*Gyrodactylus* spp. are easily differentiated from the dactylogyrids because they are viviparous, with developing embryos in the uterus. They can be found on the skin and fins of tilapia reared in brackish and fresh water (Lio-Po and Lim, 2002).

Lucky (1981) reported the existence of the monogenean fluke, *Diplozoon homoin* in pond-reared fry of silver carp in Hungary. This finding represented a

new host record for this parasite. Retarded growth of fry appeared to be the major effect of the parasite.

Digenean trematodes are internal parasites with a complex life cycle involving two to four hosts. In general, these species are not a serious threat to cultured species (Avault, 1996). Infection by *Diplostomum volvens* was noted in silver carp and to a lesser extent in common carp, and resulted in lower increments in weight in affected fish (Pietrzak, 1978). As well, some metacercariae of *Diplostomum* sp. (eye fluke) can be dangerous, causing blindness and a resulting inability to find food (Noga, 1996). Molnár (1974) reported an epizootic of *D. spathaceum* in three-week-old fry of equal numbers of grass and silver carp in the same pond. Although 90% of the fry of grass carp died, none of the silver carp died, even though both species were infected to the same degree. Szekely and Molnár (1991) found that praziquantel (Droncit) was 100% effective against metacercariae of *D. spathaceum* that parasitized the lens of grass and silver carp.

*Sanguinicola* spp. (digenean blood flukes) infect salmonids, and have caused mild to heavy mortalities in hatcheries in the western USA. This parasite has also been found on exotic cultured grass and bighead carp in the Far East (Lio-Po and Lim, 2002), and in common and other carp in Europe (Odening, 1989; Noga, 1996). *Sanguinicola* spp. live in major blood vessels of carp; eggs lodge in blood vessels of the gills, where they hatch and cause hemorrhage and extensive damage (Anon, 2003b).

## 5. Phylum Nematoda

Nematodes (roundworms) are common in feral and food fish. Except for *Capillaria* and *Camallanus* spp., parasitic nematodes are normally not a serious threat in aquaculture, although they may make the flesh wormy and unsuitable

for commercial value (Avault, 1996). In common carp, *Philometroides cyprini* is an important nematode species.

## 6. Phylum Crustacea

Parasitic copepods are becoming more serious in cultured fish, and can also have an impact on indigenous populations of fish. Most of the approximately 10,000 copepods are free-living, and about 1700 species are parasites. Most of these parasites affect marine fish, but some species are important parasites in fresh water systems. Most are parasites of the skin and gills, and a few penetrate deeply into tissues of the host, eg, the heart. Three major types are most commonly found on cultured fish: ergasiliform, caligiform, and lernaeiform types. As well, *Argulus* spp. (fish lice) may affect a number of hosts (Noga, 1996).

### i) *Ergasilus* spp..

Ergasilids are similar to free-living copepods since the body is divided into a distinct cephalothorax and abdomen, and the presence of paired locomotory structures. In small numbers they seem to cause few problems. However, their feeding activity does severe local damage, and heavy infestations can be damaging. They are usually <2 mm long, and affect mainly the gills of fresh water fish. Nie and Yao (2000) studied the seasonal dynamics of populations of *Sinergasilus polycolpus* and *S. major* in farmed silver, bighead and grass carp, and found that higher levels of these parasites were seen in summer; reproduction of these copepods occurs from spring to autumn, as indicated by the higher ratio of gravid copepods. Wang *et al* (2002) reported mortalities caused by *S. polycolpus* in farmed silver and bighead carp in a reservoir in China.

ii) *Lernaea* spp..

Lernaeids are anchor worms. The word “anchor” is used for these highly modified copepods that have anchor-like processes for attachment to the host. Goldfish, koi, and grass, silver, bighead and black carp, or wild native fish are most commonly affected (Lio-Po and Lim, 2002). *Lernaea* is the most important genus of lernaeid copepods, but other genera such as *Opistolernaea* and *Lernaeagiraffa* are important in tropical environments. *Lernaea* and related genera affect fresh water fish. It is most likely to be seen in summer when reproduction occurs, eg, *L. cyprinacea*, a cosmopolitan species that infects a wide range of species (now, 45 known species of cyprinids, as well as other orders of fish) (Lio-Po and Lim, 2002), and even tadpoles, does not reproduce at temperatures lower than 14°C. At optimal temperatures (>25°C), a female may produce several hundred larvae about every two weeks for up to 16 weeks.

Single lernaeids are usually not life-threatening unless they infect a small fish or if they penetrate near vital organs. Heavy infections may lead to debilitation and secondary bacterial or fungal infections. Hemorrhage at the site of attachment is common. Sometimes, considerable proliferation of tissue or scarring may develop at the site of attachment, and can result in sufficient disfiguring that consumers will reject the fish.

iii) *Caligus* and *Lepeophtheirus* spp..

These species are sea lice that infest the skin. Species are usually restricted to certain groups such as salmonids. They can survive at least one week in fresh water if attached to the fish.

iv) *Argulus* spp.

These are branchiuran parasites, of which there are about 140 species. According to Noga (1996), virtually the only genus encountered is *Argulus*, also called the fish louse. It is uncommon in fresh-water aquarium fish but may occur if wild or pond-reared fish are introduced; it is especially common on goldfish and koi, and is prevalent on many wild fresh-water fish. Many fish lice affect a wide range of hosts. *Argulus* feeds by inserting a stylet into the host and sucking body fluids. Fish can display violent erratic swimming or other behavioral abnormalities as a result of the irritation caused by the stylet. The skin is damaged by repeated piercing by the stylet; hooks and spines on the appendages may also cause mechanical damage. *Argulus* spp. can also transmit viral or bacterial infections.

#### D. Mycotic (Fungal) Diseases

i) *Saprolegnia* spp..

This well-known fungal agent is a member of the *Oomycetes*, and is by far the most commonly found fungal condition on the skin and gills of fish. The *Saprolegnia parasitica-diclina* complex (called saprolegniasis) is of most importance in the pathology of fish. Several predisposing factors to the infection by *Saprolegnia* spp. which are common secondary invaders, include handling and any trauma to the skin in crowded conditions and in conjunction with pollution or bacterial or viral infections in the spring and fall when water temperatures are in the range of 15-20° C (Usui, 1991). Lesions of this condition are focal gray-white patches that have a cotton-wool appearance under water. Early lesions are almost circular and grow by radial extension from the periphery

until contiguous lesions merge. At this time, the lesions are often dark gray or brown as the fungal mycelia trap mud or silt (Roberts, 1989; Noga, 1996).

The distribution is usually random, but certain parts of the body may be especially involved, eg, the head area in secondary infections of ulcerative dermal necrosis of salmonids, and "sure" of eels. The skin and gills are most commonly infected (Roberts, 1989; Noga, 1996).

Another member of the *Oomycetes* is *Aphanomyces piscicida* which produces granulomas in some species of fish; it is also highly pathogenic for goldfish and crucian carp (Lio-Po and Lim, 2002).

## ii) Branchiomycosis

This is a fungal disease, sometimes called Gill Rot, that has caused acute, often high mortality in a number of fresh water fish, including the American and European eel. There are two species, *B. sanguinis* which affects common carp, tench, and three-spined stickleback in Europe, and *B. demigrans* which infects large-mouth bass, northern pike, tench and striped bass in Europe, Taiwan, or the USA (Noga, 1996). There is speculation that this condition is a type of water mold infection, but there are too few data by which to classify the causative organism. Gills may be mottled because of thrombosis (plugging of blood vessels with clots) and resulting death of the tissue which produces the appearance of light and dark areas. There is no treatment. Surviving fish are carriers of the infection and should not be transferred into *Branchiomyces*-free geographical areas. Dead affected fish should be burned and/or buried (Lio-Po and Lim, 2002). Other miscellaneous mycotic agents, including *Fusarium culmorum*, which has produced infection of the skin and eyes in common carp, have been reported from Europe.

A *Cryptococcus* sp. (a yeast) has been recovered from tench affected with bilateral exophthalmos in Europe (Noga, 1996).

Appendix 1. Letters from the Council of Lake Communities and the Great Lakes Fishery Commission concerning Asian carp.

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