Report about fish parasitic diseases

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About diagnostic laboratories for parasitic diseases in the region

Most laboratories participating in the survey (48 out 54) from 13 Mediterranean countries stated that they perform diagnosis of fish parasitic diseases.

Of the 48 laboratories that declared to work on the diagnosis of fish parasitic diseases, 41 stated that they worked with seabream and seabass and 32 with trout. Other species they worked with were eels (13), carps (12), turbot (9) and salmon (9). Many (31 laboratories) stated to also work with other fish species, such as tilapia, mullets, other sparids (*Dentex dentex, Puntazzo puntazzo*, etc.) and other marine species.

This is coherent with available fish production information for the region, and as is the case for the diagnosis of viral and bacterial diseases, most diagnosis is performed on finfish species produced intensively in Mediterranean countries.

The different techniques available in the laboratories performing diagnosis of fish parasitic diseases (Table 1) show that most laboratories based their diagnosis partly or totally on simple techniques, such as observation of clinical signs, macroscopical examination of fish and examination of fresh samples (wet mounts, smears). Less laboratories (26 out 48) can perform histopathology studies, and even less (15 or 16) can perform more specialised techniques such as PCR or immunohistochemistry.

Table 1. Available techniques in the laboratories performing diagnosis of fish parasitic diseases

Diagnostic methods	Available
Clinical signs	48
Microscopical examination of fresh samples	47
Macroscopical examination	46
Histopathology	30
Haematological examination	26
Fluorescent antibody technique	15
PCR	16
Immunohistochemistry	15
Transmission electron microscopy	9
DNA probes	4

The above mentioned capability (available techniques) explains why, as indicated below, for most parasitic diseases the diagnosis is just based on simple techniques (clinical signs, macroscopical examination, examination of fresh samples) and also histopathology. Thus, the possibility of overlooking or misidentification of some parasites cannot be discarded. Efforts should be done to improve the technical level of diagnostic laboratories, in order to obtain more accurate diagnosis and the detection of asymptomatic carriers.

Main reported parasites and diseases

A summary about the incidence (based on answers received) of the different parasites and

diseases found in the laboratories participating in the survey is presented in Table 2. There were reports of about 35 different parasitic diseases, which is significantly higher than reports stated for fish viral diseases (9) and fish bacterial diseases (15).

Table 2. Overview of the fish parasitic diseases covered in the survey

Disease	Status based on survey
AMOEBOZOANS	
Amebiasis	Diagnosed by 6 laboratories from 2 countries (Spain and Greece) from marine fish (turbot, seabass, seabream and salmon) in routine studies and in occasional examinations, sometimes in fish suffering mortalities.
DINOFLAGELLATES	
Amyloodiniosis	Diagnosed in 8 laboratories from 7 countries in seabass and seabream, with mortalities frequently reported in juvenile fish.
FLAGELLATES	
Ectoparasitic flagellates	
Costiasis	One of the parasites most frequently reported, in 13 laboratories and 8 countries in different freshwater and marine fish species. Found in routine examinations and with mortalities in hatcheries.
Cryptobiasis	Some cases were stated in 3 laboratories from 3 countries, mainly in marine fish.
Endoparasitic flagellates	
Trypanosoma sp.	Only reported by 1 laboratory (Croatia) in different freshwater fish.
Hexamitiasis	Diagnosed only in 2 laboratories of 2 countries (Croatia and Spain) in salmonid, cyprinids, *Scardinius* and exotic fish.
CILIATES	
Chilodonellosis	Stated by 6 laboratories in 4 countries (Croatia, France, Greece, and Spain) in freshwater fish. A possible misidentification with *Brooklynella* may have occurred in a case in marine fish.
Cryptocaryosis	Diagnosed in 7 laboratories from 4 countries, mainly in sparids and seabass both in routine samplings and associated to mortalities.
Trichodiniasis	The most frequently found parasitosis in the survey with reports from 22 laboratories from 10 countries in both freshwater (eel and carp) and marine (seabass and seabream) fish. Found in routine examinations and also associated to mortalities, and sometimes associated to other pathogens, mainly bacteria.
White spot disease	Reported by 13 laboratories from 6 countries in routine samplings and associated to mortalities in different freshwater fish, mainly *Oncorhynchus mykiss*.
Scuticociliatida	Diagnosed in only 2 laboratories from Spain in marine fish (turbot, seabream and seabass) in routine examinations and mortality cases.
ENDOPARASITIC PROTOZOANS	
Coccidiosis	Reported by 11 laboratories from 4 countries, mainly in marine fish (turbot, seabream, seabass and puntazzo). Some reports indicate the disease without identification of the species or genus.
Microsporidiosis	Stated in 6 laboratories from 3 countries, mainly in routine examinations in turbot and seabream, and on one occasion in other sparids, mugilids and ornamental fish.
MYXOSPOREA (Myxosporidiosis)	
Whirling disease	Reported only by 1 laboratory (Croatia) in *Oncorhynchus mykiss*.
Proliferative kidney disease (PKD)	Diagnosed by 5 laboratories from 3 countries, in mortality episodes and routine examination, in *Oncorhynchus mykiss* in most cases.
Spaherospora renicola	Reported by 1 laboratory from Croatia in cyprinids
Ceratomyxa spp.	Stated commonly in 11 laboratories from 6 countries, mainly in routine or occasional studies, seldom in mortality cases, in seabass and seabream.
Enteromyxum leei	Diagnosed by 16 laboratories from 7 countries in mortality cases (sometimes also in routine studies) in cultured sparids, mainly seabream and puntazzo but also in *Diplodus sargus, Pagrus pagrus* and *Pagellus erythrinus.*
Enteromyxum scophthalmi	Stated only by 2 laboratories from 1 country, in mortality cases and in routine studies in turbot.
Kudoa spp.	Only reported by 1 laboratory (Portugal) in routine examination of *Sardina pilchardus*.
Sphaerospora dicentrarchi	Diagnosed by 5 laboratories from 4 countries usually in routine studies in the type host seabass. Also some mortality cases. The parasite is also indicated in seabream.
Sphaerospora testicularis	Reported only in 2 laboratories from Greece in seabass, the type host. An increased susceptibility to bacterial diseases is indicated.

Table 2 (cont.). Overview of the fish parasitic diseases covered in the survey

Disease	Status based on survey
MONOGENEA	
Monopisthocotylea	
Gyrodactylus spp.	Diagnosed by 9 laboratories from 5 countries, mainly in *Oncorhynchus mykiss*, and also in other samonids, some cyprinids, *Scardinius* and tropical exotic fish. Also reported in seabream and seabass. *G. salaris* is included in the OIE list of other significant diseases and in the list III of the EU.
Dactylogyrus spp.	Reported by 11 laboratories from 6 countries, mainly in freshwater fish: carps, trout, eel and tropical fish.
Diplectanum spp.	Stated in 3 laboratories from 3 countries (France, Spain and Turkey) in routine and mortality cases in the usual host seabass, but also found in seabream.
Furnestinia spp.	Diagnosed by 5 laboratories from 4 countries in routine and mortality cases in the usual host seabream, but also found in seabass. Sometimes found in mixed infections.
Polyopisthocotylea	
Microcotylosis	Diagnosed by 10 laboratories from 5 countries. Most of them in mortality cases in the most frequent host, seabream. Also diagnosed associated to bacterial infections or just in routine examinations.
TREMATODA DIGENEA	
Diplostomum spp.	The parasite was recorded by 3 laboratories from 3 countries in numerous freshwater fish (mainly cyprinids, but also salmonids and others) in routine examinations.
Sanguinicolosis	Diagnosed by 4 laboratories from 3 countries (Croatia, Greece and Spain) in a marine fish (seabream) in routine examinations and in mortality cases. One record in freshwater fish (carp).
CESTODES	
Caryophyllidea	Only one report (*Khawia* sp.) from Croatia.
Pseudophyllidea	Recorded by 2 laboratories (France and Croatia). *Triaenophorus* was reported in *Perca fluviatilis*. Other findings of pseudophyllidean cestodes include those of *Bothriocephalus*, *Eubothrium*, and *Ligula* in several freshwater fish.
Protecephalidea	Stated by only 1 laboratory (Croatia) in different feshwater fish.
CRUSTACEANS	
Copepoda	Sea lice disease was recorded by 5 laboratories from 5 countries, involving different copepoda of freshwater (salmonids and cyprinids) and marine fish (seabream, seabass and *Seriola*). Records were not frequent and, when stated, they were associated to both routine examination and mortality cases.
Isopoda	Isopod infections were only reported by 3 laboratories of 3 countries (Greece, Turkey and Portugal) in *D. labrax* (2 laboratories), *Sparus aurata* (2 laboratories) and other sparids (*Dentex dentex*, *Pagrus pagrus*, *Diplodus puntazzo*) (1 laboratory).

It is highlighted that within the OIE list of notifiable fish diseases there are no parasitic diseases. In fact, of the diagnosed parasitic diseases only gyrodactylosis – *Gyrodactylus salaris* (reported in freshwater and marine fish by 9 laboratories from 5 countries) – is included in the OIE list of other significant diseases and in the list III of the EU regulation.

The main parasitoses or parasites found in the survey, with reports from laboratories in 5 o more countries are: trichodiniasis, costiasis, *Enteromyxum leei*, white spot diseases, *Ceratomyxa*, amyloodiniosis, *Dactylogyrus*, gyrodactilosis, mycrocotylosis, and sea lice disease. The species susceptible to these diseases are of economic importance in the region; however with the received information, the impact and/or /repercussions on aquaculture production of the different diseases is difficult to assess as most of them are diagnosed both in routine examinations and in mortality cases too.

The main parasitic diseases reported in freshwater fish (salmonids, cyprinids and eels) were trichodiniasis, costiasis, white spot diseases, *Dactylogyrus*, gyrodactilosis, and sea lice disease.

The main parasitic diseases reported in marine fish (seabass, seabream, turbot, other sparids, etc.) were trichodiniasis, costiasis, *Enteromyxum leei*, *Ceratomyxa*, amyloodiniosis, mycrocotylosis, sea lice disease.

Other diseases, with reports in 3 or 4 countries, with a significance that is difficult to establish, are: cryptobiasis, chilodonellosis, cryptocaryosis, coccidiosis, microsporidiosis, proliferative kidney disease, *Sphaerospora dicentrarchi*, *Diplectanum* spp., *Furnestinia* spp., *Diplostomum*, and sanguinicolosis.

Therefore, the survey has demonstrated the increasing concern of parasitic diseases in cultured finfish in the Mediterranean area. Most of the obtained information deals with intensively cultured marine fish and several records were obtained from the expanding netpen cultures.

Some parasites can be considered a serious threat for mariculture, as *Amyloodinium* (Dinoflagellates), Scuticociliatida (Ciliates), *Enteromyxum* spp. (Myxosporea) or Mycrocotylidae (Monogenea). Other parasites are seldom reported in mortality episodes, or have been found only in routine examinations. However, their pathological concern should not be neglected, considering their increasing presence in the cultures and their direct or aside deleterious effects, even when they are not the direct cause of high mortality.

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Amoebozoans

Amebiasis

Although different species of amoebas have been associated to amoebic diseases in fish, recent observations point out to *Neoparamoeba* spp. as agents of amebiasis*. N. pemaquidensis* may be of special pathological concern in salmon, in which it produces the amoebic gill disease (AGD). Amoebas may appear in small numbers, probably trapped in the gills, without damage. In heavy infections, the parasites elicit epithelial hyperplasia, resulting in complete fusion of secondary lamellae an subsequent gill disfunction. Fish become lethargic and thin. Diagnosis is mainly based on microscopical examination and histopathology, though immunodiagnosis and isolation in culture, followed by identification by morphology have also been used. Species specific PCR has been developed for *N. pemaquidensis*. The disease has been diagnosed mainly in *Salmo salar*, though recent findings include different sea fish, as *Scophthalmus maximus*, *Sparus aurata* and *Dicentrarchus labrax*. The most widely recommended treatment is the use of freshwater baths, though it is not fully effective in killing amoebae. Hydrogen peroxid and levamisol have also been assayed with variable results.

Current status based on answers received

In this survey of Mediterranean region, amebiasis has been diagnosed in 6 laboratories and 2 countries, 2 laboratories in Spain, 3 in Greece and another finding not located. There is an unique record in *Salmo salar* from Spain. The other findings include three marine species, *S. maximus*, *S. aurata* and *D. labrax*. It has been detected in some routine studies, but it was mostly found in occasional examinations, sometimes in fish suffering mortalities. Thus, prevalence is difficult to establish, but findings were rather occasional. Diagnosis in the survey was mainly based on microscopical examination and histopathology, rarely in electron microscopy or culture. No reference to control was made in the survey.

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Dinoflagellates

Amyloodiniosis

Also known as "velvet disease", the causative agent is *Amyloodinium ocelatum*, an ectoparasite on the skin and gills of different fish species. Appart from the velvet appearance, clinical signs consist of anorexia and scratching. Histopathological lesions include gill inflammation, haemorrhages and hyperplasy. Massive infections are frequently associated to mortalities, both in mariculture and sea aquaria, mainly at high temperatures. The infection is very common in Mediterranean fish, though other fish species are affected, including tropical and aquarium fish. Other dinoflagellates (as *Piscinoodinium* spp.) parasitize different freshwater fish. Diagnosis is mainly based on microscopic fresh and histological examination, though an ELISA test is available. No effective control measures are known for Mediterranean fish. Freshwater (2-4 minutes) or copper sulphate (0.75 mg/l, 12-14 days) baths have been suggested as an aid to control the trophonts or dinospores, respectively. In Pacific threadfin (*Polydactylus sexfilis*) recent findings suggest the suitability of hydrogen peroxide as treatment in juvenile fish. Some evidences suggest the development of immunity against re-infections, and specific antibodies have been demonstrated in the sera of infected fish.

Current status based on answers received

In the survey it was diagnosed in 8 laboratories from 7 countries, covering the main production area of commercial Mediterranean fish. It was found in *Dicentrarchus labrax* and *Sparus aurata* in all the Mediterranean countries and Portugal. Less frequent in *Diplodus puntazzo* (Greece) and mullets (Italy). In addition, *Oodinium* was reported by 1 laboratory (Morocco) in seabream and puntazzo (we asume it is actually *Amyloodinium*). The results of the survey confirmed the pathological concern of this disease, as mortalities were frequently reported, mainly in juvenile seabass and seabream at temperatures over 20°C. Diagnosis was mainly based on microscopic fresh and histological examination. No reference to control measures was made in the survey.

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Flagellates

ECTOPARASITIC FLAGELLATES

Costiasis

Ichthyobodo spp. (also known as *Costia*) are the agents of this disease of the gills and skin. *I. necator* is the species parasitizing salmonids in freshwater, but a different species is considered to be present in marine fish. Affected fish appear thin and lethargic, and may show a grey-whitish pellicle on skin, epidermic erosion or even haemorhages or ulcers, as well as gill hyperplasy and edema. Costiasis is widely distributed in different fish species, mainly in larval and juvenile stages, and mortality can occur in fry or ornamental fish with moderate to severe infections. Besides direct mortalities, indirect damage due to decreased health condition and gill lesions must be considered. Diagnosis is based on microscopical examination and histopathology. Prevention relies on hygienic measures. Costiasis can be treated with formaline 1:4000 or 1:6000 in baths with a good aeration.

Current status based on answers received

It is one of the parasites most frequently found in the survey, both regarding the number of laboratories (13), countries (8) and the number fish species. Besides freshwater species (including *Carassius auratus, Cyprinus carpio, Ictalurus punctatus, Oncorhynchus mykiss, Scardinius erythrophthalmus, Perca fluviatilis, Coregonus* sp*., Salmo trutta* and exotic fish), it has been diagnosed in the marine fish *Dicentrarchus labrax, Sparus aurata* and *Diplodus puntazzo*. Some laboratories have identified marine costiasis as produced by *I. necator*. The findings by countries are representative of the importance of the culture of the different affected fish species. The parasite was reported in association with mortalities in several cases, mainly in hatcheries, though it was also found in routine examinations. Concomitant bacterial infections were found in some occasions. Diagnosis was again based on microscopical fresh and histological examination. No control measures were indicated in the survey.

Cryptobiasis

The other ectoparasitic flagellates found in the survey are the gill *Cryptobia* spp., with a direct life cycle. Marine ectozoic species include *C. branchialis*, from different coastal fish and *C. eilatica*, described from the gills of *Sparus aurata* and *Diplodus noct* in the Red Sea. In heavy infections, the parasites produce gill hyperplasia and epithelial destruction, with subsequent respiratory impairment. External signs are anorexia and skin darkness. In the Mediterranean it is relatively common in cultured seabass and seabream. The infection can produce trickling but persistent mortalities, so loses can reach 10% after several weeks. Diagnosis is based on microscopic fresh and histological examination. Formalin baths (150 mg/l) can be effective for treatment.

Current status based on answers received

In the survey, *Cryptobia* spp. were only found in 3 laboratories from 3 countries. Affected fish include the typical species more representative of the cultures, i.e. *S. aurata* in Spain and Greece, *D.*

labrax in Greece and Turkey and *D. puntazzo* in Greece. Occasionally associated to mortalities in Greece and Turkey, it was also diagnosed in routine examinations. It may appear in mixed infections, mainly with monogeneans. Diagnosis was again based on histopathology, sometimes also on fresh microscopical examination.

ENDOPARASITIC FLAGELLATES

Cryptobia spp*., Trypanoplasma* spp*., Trypanosoma* spp*.*

Some species of these genera parasitize internal organs of fish. *Cryptobia iubilans* is the only pathogenic intestinal species, common in aquaria cichlid fish. *Trypanoplasma spp*. and *Trypanosoma* spp. include parasites of the bloodstream and of tissues, with indirect life cycles (leeches are the main vectors). The best known is *Trypanoplasma salmositica* (frequently referred as *Cryptobia samositica*) producing cryptobiasis of salmonids. Clinical signs consist of exophthalmia, splenomegaly, hepatomegaly, abdominal distension with ascites, anemia and anorexia. Mortality is dependent on fish stocks and species, but may be high in juveniles. The disease has severe impact in salmonid cultures in North America. An experimental protective vaccine has been developed. Other pathogenic species, *Trypanoplasma borreli,* parasitizes mainly cyprinids in Europe and North America. The genus *Trypanosoma* includes numerous species of both freshwater and marine fish. Some freshwater species are pathogenic for cyprinids.

Current status based on answers received

In the survey, only *Trypanosoma* sp. was reported by 1 laboratory (Croatia) in different freshwater fish (cyprinids, *Esox lucius* , *Scardinius erythrophthalmus* and *Salmo trutta*).

Hexamitiasis

Hexamita spp. are parasites of the intestine and gall bladder of freshwater fish, mainly salmonids but also cyprinids and ornamental fish. Hexamitiasis, typical of weak fish, is frequent as a secondary infection. Affected fish can show nervous behaviour, and internally the intestine may appear pale. Mortalities can occur in fry and ornamental fish. Diagnosis is manly based on the direct observation of the flagellate in fresh intestinal scrapings or histopathology study.

Current status based on answers received

In the survey it was diagnosed only in 2 laboratories of 2 countries (Croatia and Spain), in salmonid, cyprinids, *Scardinius* and exotic fish.

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See also general references for fish parasites.

Ciliates

Chilodonellosis

Most *Chilodonella* spp. are free living, but some of them are serious pathogens of freshwater fish, causing heavy loses in aquaria and in cultures. Under conditions favouring their proliferation they invade the gills and skin of affected fish. The gills suffer hyperplasia, degeneration and necrosis, and respiration is drastically impaired. On the skin they may virtually cover the body surface. Diagnosis is mainly based on the microscopical examination of gill or skin scrapings. Formalin baths have been suggested as treatment. Prevention relies on optimisation of fish maintenance and increasing of water quality.

Current status based on answers received

In the survey it was only reported by 6 laboratories (2 from Greece, 2 from Spain, 1 from Croatia and 1 from France) in several fish species, including ornamental freshwater species, barbels, rainbow trout or mullets (considered very sensitive in this study). A Greek laboratory reported it also from marine fish (*Sparus aurata*, *Dentex dentex*, *Diplodus puntazzo* and *Dicentrarchus labrax*). Considering that *Chilodonella* is typical of freshwater, a misidentification with *Brooklynella*, typical from marine fish and morphologically quite similar to *Chilodonella* could have occurred. Diagnosis was mainly based on fresh microscopical and/or histopathological examination.

Cryptocaryosis

Cryptocaryon irritans, parasite of gills and skin, is the causative agent of this disease. External signs consist of white spots and mucous excess or ulcers on the skin and impairment of respiratory function. Gill histopathology consist of inflammation, haemorrhages, hyperplasia and lamellar destruction. This ciliate is a typical marine fish parasite affecting commercial and ornamental fish and producing high mortality in culture conditions. Outbreaks appeared mainly at high temperatures. Some treatment and control measures are similar to those recommended for ichthyophthiriasis, though quinine derivatives and low salinity baths have also been used. Diagnosis is based on macroscopical examination followed by microscopical examination for confirming the presence of the ciliate. The parasite can also be found in histopathological studies.

Current status based on answers received

In the survey it was diagnosed mainly in the Mediterranean cultures of sparids and seabass. Seven laboratories from 4 countries (all of them with the most representative Mediterranean maricultures) reported the disease, both in routine samplings and associated to mortalities. Diagnosis was mainly based on fresh microscopical and/or histopathological examination.

Trichodiniasis

Fish trichodinids include mainly *Trichodina* spp., *Trichodinella* spp. and *Tripartiella* spp. These peritrichid ciliates are more commensals than genuine ectoparasites, but can produce different damages in massive infections. The fish show a grey-blue turbid layer on the skin. Respiratory function can be impaired in gill infections. Trichodinids parasitize a lot of freshwater and marine fish species. Diagnosis is mainly based on microscopical examination of fish or gill scraping preparations. Hygiene in hatcheries and quarantine for ornamental fish are recommended for prevention. This ciliatosis can be treated with formaldehyde in baths. In freshawter ornamental fish and fry, baths of salt solutions can be applied, with variable success.

Current status based on answers received

In the survey, trichodiniasis was the most frequently found parasitosis, as it was diagnosed in 22 laboratories from 10 countries. Considered mainly as opportunistic, it was sometimes associated to other pathogens, mainly bacteria. More frequent in routine examinations, it was also found associated to some mortality cases. Diagnosis was usually made to the genus level (*Trichodina* sp.) and was

mainly based on fresh microscopical or histopathological examination. Surprisingly, a diagnosis based on macroscopic examination is reported, apparently based on the presence of white spots on skin and gills.

White spot disease

Ichthyophtirius multifiliis produces the well known white spot disease or ich. The most characteristic external sign is the presence of white spots on the skin and gills, due to parasite trophonts located under the upper layer of the skin. Affected fish can rub or flash and show breathing problems. In heavy infections the typical white spots are visible with the naked eye. Histopathological damage is more evident in heavy infections and in reinfections, including proliferative response and cell necrosis. The disease is widely distributed in many freshwater fish species, mainly in aquaria and culture conditions, in which it can produce epizootics. Mortality is mostly dependent on fish size and infection intensity. Diagnosis can be made macroscopically in heavy infections (white spots), followed by confirmation at microscope in fresh smears or scrapes. General preventive measures based on lower stocking densities or increased water flow and frequent routine examination, can aid to control the disease. Malachite green is quite effective in treatment of this ciliate, but presently its use is not allowed, and formalin is generally used. Free stages (tomites) are more susceptible but cysts (stages on plants or substrates) and trophonts (in the fish skin) are quite refractory to treatments. Evidence of resistance to reinfection has been observed in survivor fish, and some experimental vaccines have been assayed with different success.

Current status based on answers received

In the survey, it was found in 13 laboratories (of Croatia, France, Greece, Italy, Spain and Turkey) affecting numerous species of cultured fish (cyprinids, salmonids and exotic fish) and several wild fish. It was found both in routine samplings and associated to mortalities. *Oncorhynchus mykiss* was the fish most frequently parasitized. When the number of diagnosed cases is available, the values are similar for the 3 considered years. Diagnosis was mainly based on fresh microscopical or histopathological examination. No reference to control measures was made in the survey.

Scuticociliatida

Several species of the genera *Uronema*, *Phylasterides* and *Miamiensis* have been recorded as facultative parasites of different fish. Clinical signs of scuticociliatosis depend on the parasite location. External signs include skin lesions or ulcers and pigmentation changes, but the parasite frequently invade the body muscle and the internal organs, which become destroyed by this histophagous parasite. Nervous system can also be colonised, which can be accompanied by erratic swimming, equilibrium loss or lethargy. The disease cause severe infections and outbreaks in some cultured fish and mortalities can reach 100% of some affected stocks. Different fish species can be affected, but the most severe cases reported deals with turbot, Mediterranean seabass, several marine aquarium fish, Australian tunnids and Japanese flounder.

Diagnosis is based on the finding of ciliates in ascitic fluid or scrapings of different organs. The typical morphology of Scuticociliatida can be easily observed in fresh preparations at light microscope, in stained smears or histological sections. However, the identification at the genus and species level requires specific staining evidencing the somatic and oral infraciliature and the scutica. There are no efficacious treatment for this parasitosis. Formalin baths have been assayed with certain success only in the initial phase of infection.

Current status based on answers received

In the survey it was diagnosed only in 2 laboratories from Spain, both in mortality cases and routine examination. Affected fish include turbot (both laboratories), seabream and seabass (1 laboratory). Diagnosis was based on fresh microscopical examination and histopathology.

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Endoparasitic Protozoans

Coccidiosis

Different coccidia (Apicomplexa) are known from among freshwater and marine parasites, but their pathological significance for the cultures is very variable. The genera *Eimeria, Goussia* and *Cryptosporidium* include the species more frequently reported from cultured fish. In freshwater fish, *G. carpelli* parasitizes different cyprinids and *E. anguillae* is typical of eels. In marine fish, *E. sparis* and *G. sparis* have been reported from *Sparus aurata* and *E. dicentrarchi* and *E. bouixi* from *Dicentrarchus labrax*. Fish *Cryptosporidium* spp. include species from seabream, seabass, turbot, and aquarium fish, affecting mainly larvae and juvenile, with deleterious effects not always very evident, but resulting in poor condition. *C. molnari* is more frequent in seabream than in seabass. The species of turbot is probably a new one. Diagnosis of fish coccidia is mainly based on histopathology and/or on fresh examination at microscope. Immunodiagnostic methods are available for some human and animal species, but not for fish species. Control of animal coccidians is based on the use of different coccidiostatics or coccidiocides, but information regarding fish coccidia is very scarce. Furazolidone, amprolium chloride and furanace, among others, have been tried to treat different fish cocidia.

Current status based on answers received

In the survey, some reports indicate only coccidiosis without identification at the species or genus level. In other cases, identification at the genus level is provided, including *Cryptosporidium*, *Eimeria* and *Goussia*. Considering all the coccidiosis, they have been reported by 11 laboratories of 4 countries, and another finding not located. The most frequent hosts are marine fish, mainly *S. aurata* and *D. labrax*, followed by *Scophthalmus maximus* and *Diplodus puntazzo*. Freshwater fish include only cyprinids and some exotic fish. *Cryptosporidium* spp. were found in seabream, seabass and turbot from Spain (2 laboratories), and seabream suffering mortality (1 laboratory, location not indicated). The involved species in cryptosporidiosis of seabass and seabream is probably the recently described *C. molnari*.

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Microsporidiosis

Microsporea are represented in fish by different genera, mainly *Enterocytozoon*, *Glugea*, *Loma*, *Pleistophora* and *Tetramicra*. In freshwater fish, *Pleistophora* and *Loma* are relatively frequent. Among cultured marine fish, there have been several reports of *Plesitophora senegalensis* in gilthead seabream, whereas *Glugea* sp. and *Tetramicra brevifillum* have been found in turbot. Pathological concern of microsporidiosis in fish is dependent on location and infection intensity. Variable loses in turbot cultures have been related to *Tetramicra* infections. Diagnosis is based on the direct detection of the parasite at microscope, mainly the spores, but ultrastructural studies are necessary for identification at the specific (or even generic) level. A PCR based assay has been recently developed for *T. brevifillum*. Among chemicals tested for treatment, toltrazuril has apparently given better results than fumagillin and amprolium.

Current status based on answers received

In the survey, microsporidioses have been reported by 6 laboratories from 3 countries. Parasitized hosts include mainly *Sparus aurata* and *Scophtahlmus maximus*, though they were recorded in one occasion in other sparids (*Dentex dentex* and *Pagrus pagrus*), mugilids and ornamental fish. In one case, it was diagnosed in wild *Pagellus acarne*. Usually reported as microsporidiosis, the only case of generic identification is doubtful. Microsporeans were mainly found in routine examinations, only one case was associated to mortalities in cage juveniles. Prevalences (when indicated) are generally low. Diagnosis was mainly based on fresh microscopical examination and histopathology, only in one case in electron microscope. No references to control measures were made in the survey.

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Myxosporea (myxosporidiosis)

The class Myxosporea (phylum Myxozoa) include numerous genera and species, most of them parasites of fish. Some species are well know pathogens for freshwater fish. In the last years Myxosporea have been increasingly reported in cultured marine fish. They are characterised by a spore with one to several valves, one or more infective sporoplasms and one to several polar capsules with a coiled polar filament inside.

The most pathogenic species belong to the genera *Ceratomyxa*, *Myxobolus, Myxidium, Spaherospora*, *Enteromyxum, Kudoa, Tetracapsuloides* and *Sphaerospora*.

In freshwater fish the most significant diseases are whirling disease, PKD, sphaerosporosis and ceratomyxosis (produced by *Ceratomyxa shasta*).

Myxosporea reported from cultured marine fish include species of the genera *Ceratomyxa, Enteromyxum, Kudoa, Lepthoteca, Sphaerospora and Sinuolinea*.

Other myxosporeans reported ocassionally in the survey are *Leptotheca* sp. and *Polysporoplasma spari*s from the kidney of *S. aurata,* and *Sinoulinea* sp. from the urinary bladder of turbot, the three reported only by 1 laboratory from Spain, mainly in routine or occasional samplings. The species of *Leptotheca* has been described as *L. sparidarum* from seabream an dentex (see references).

Whirling disease

The causative agent is *Myxobolus cerebralis* (synonym *Myxosoma cerebralis*). Clinical signs include dark coloration of the posterior part of the body and abnormal swimming in spiral, followed by skull deformation and spinal curvature. Almost all salmonid species can be infected, but susceptibility is very variable according to the species. *Oncorhynchus mykiss* and other *Oncorhynchus* spp. are very susceptible, while *Salmo trutta* is rather resistant. The involvement of an intermediate oligochaete host in the life cycle of this myxosporean was demonstrated 18 years ago. This knowledge has facilitated preventive measures, consisting of the use of concrete or plastic ponds or tanks and their frequent cleaning for avoiding the presence of oligochaetes and thus the transmission of the disease.

This is very important, considering the limited efficacy of treatments assayed till now (fumagillin, toltrazuril) for this myxosporea and other species. In some European countries, the incidence of whirling disease has clearly decreased, whereas in USA whirling disease is widely distributed and is still an important pathological problem. Diagnosis is based on the histological examination of skull cartilage, or their enzymatic digestion followed by microscopical observation of the typical spores. A PCR assay has also been developed.

Current status based on answers received

In the survey it was diagnosed only in one laboratory of one country (Croatia) in *Oncorhynchus mykiss*.

Proliferative kidney disease (PKD)

The causative agent, formerly known as PKX, has been recently identified as *Tetracapsuloides bryosalmonae* (syn. *Tetracapsula bryosalmonae*, *T. renicola*). Extrasporogonic stages of this myxosporean are located in the kidney of different salmonid fish, but spores are formed in a bryoozoan host. The parasite is highly pathogenic and can produce a severe disease in rainbow trout, with 30-50% mortality. External clinical signs are abdominal swelling, darkening and exophthalmos. Internally, a kidney enlargement is observed, accompanied by ascites in advanced cases. In histopathology, the kidney shows interstitial hyperplasia, associated with chronic, granulomatous interstitital nephritis and tubular atrophy. A severe inflammatory reaction is also produced. Poor food conversion and immunodepression are also a consequence of this infection. Macroscopical diagnosis relies on the gross observation of kidney enlargement. Confirmation is achieved by observation of parasitic stages in histological sections or squash preparations (in this case by experienced observers).

Current status based on answers received

In the survey, PKD was diagnosed by 5 laboratories of 3 countries (Croatia, France and Italy) mostly in mortality episodes but also in routine examination. *Oncorhynchus mykiss* was the fish host in most cases but there is one record in *Salmo trutta* in Italy. Diagnosis was mainly based on histopathology, but in two cases, electron microscopy or PCR were also used.

Spaherospora renicola

Spaherospora renicola is widely distributed in intensive cultures of cyprinids, mainly *Cyprinus carpio*. Spores and sporognic stages are located in the renal tubules, but proliferative stages appear in the blood and can reach the swimmbladder, causing inflammation. *S. renicola* may be a serious pathogen. In the kidney tubules it produces dilatation, atrophy and necrosis of the epithelium, with subsequent impairment of renal function. The development of swimbladder stages elicits the swimbladder inflammation of juvenile carps. Fish can show some external clinical signs as locomotory disorders and swimming in circles.

Current status based on answers received

In the survey it was diagnosed only by 1 laboratory from Croatia, in one occasion. However, it must be considered the scarcity of cyprinid cultures in the surveyed countries.

Ceratomyxa shasta

Ceratomyxa shasta is an important pathogen, causing serious loses in cultured and wild populations of salmonids on the west coast of North America. Intestine is the target organ and parasites can be observed in the epithelium, eliciting lymphocytic infiltration, hyperplasia and necrosis. In advanced stages of the infection, parasites spread to other organs an fish become anorexic, lethargic, and show abdominal swelling, ascites and exophthalmia. Significant mortalities can occur, depending on fish species, as susceptibility is variable. An intermediate host (a polychaete) has been demonstrated in the life cycle of this Myxosporean. A PCR assay is available for diagnosis.

Ceratomyxa spp.

This genus include a lot of marine species, though they have been rarely associated with significant pathological problems. The main species recorded in cultured marine fish are *C. diplodae, C. labracis* and *C. sparusaurati*. *C. diplodae* (originally described from *Diplodus annularis*) and *C. labracis* are quite frequent in wild and cultured seabass. They are not usually associated with clinical disease, but they can induce several histopathological lesions in the gall bladder and neighboring tissues. *C. sparusaurati* is a very common parasite of the gall bladder of *Sparus aurata*. It generally causes limited histopathological damage, but in some massive infections it has been associated with clinical signs and mortality.

Current status based on answers received

In the survey *Ceratomyxa* spp. and/or ceratomyxosis have been reported by 11 laboratories of 6 countries (no indication of country is given by 1 laboratory). Records were very common in the 3 years of study. Host fish include *Dicentrarchus labrax* in 8 cases and *S. aurata* or other sparids in 10 cases. They have been mainly diagnosed in routine or occasional studies, though there are some records in mortality cases (though direct association with mortality is not stated). The results indicate the wide dispersion of *Ceratomyxa* spp. in Mediterranean cultured fish, though they are not considered significant pathogens. Diagnosis was mainly based on microscopical (fresh or histological) examination.

Enteromyxum spp.

Two species of this genus have special pathological concern for marine fish of high commercial value, and both parasitize the digestive tract of infected fish.

Enteromyxum leei is the myxosporean previously known as *Myxidium leei*, which has been transferred to the new genus *Enteromyxum* after morphological and molecular studies (see references). It produces the myxidiosis of sparids, which now should be named enteromyxosis, the most significant myxosporidiosis of cultured sparids in the Mediterranean. Not only *S. aurata*, but also other sparids can be affected, including the new cultured species *Diplodus puntazzo* and *Pagrus* spp. The range of susceptible fish is extraordinarily wide, as seabass, mullets, *Sciaenops ocellatus* and different marine aquarium fish, belonging to 25 species of 4 separate orders have been found parasitized. The parasite invades the intestinal tract causing severe chronic enteritis, frequently followed by emaciation and dead. Thus, external clinical signs mainly consist of a extreme thinness ("knife-fish" for some farmers). Losses can reach 80% of some stocks, specially in *D. puntazzo* which seems to be specially susceptible. Horizontal direct fish-to-fish transmission has been demonstrated for this myxosporean, which enhances the pathological importance of the disease in the cultures. Diagnosis is mainly based on histopathological study of the intestine and detection of parasitic stages, though these stages can be also seen in fresh smears by a experienced observer. A PCR assay is being validated in the framework of an European project.

Current status based on answers received

In the survey, *E. leei* (reported as *Myxidium leei* or myxidiosis) was diagnosed by 16 laboratories of 7 countries, all of them important cultivators of sparids. *S. aurata* and *D. puntazzo* were the most frequent fish host, followed by *D. sargus, Pagrus pagrus* and *Pagellus erythrinus*. Diagnosis was based on clinical signs together with microscopical examination of fresh smears and/or histopathology. The survey has confirmed the pathological importance of the disease, as all laboratories have diagnosed it in mortality cases (sometimes also in routine studies) and some laboratories indicate high mortalities and important loses, mainly for *D. puntazzo*.

Enteromyxum scophthalmi is the other species of the recently erected genus *Enteromyxum*, reported up to now only from turbot, *Scophthalmus maximus. E. scophtahlmi* is an important pathogen of turbot cultures, as mortalities can reach 100 % of the affected tanks or stocks, with subsequent economical impact. Clinical external signs include anorexia, caquexia, sunken eyes and a typical prominent bony ridge on the skull. At necropsy, pallor of internal organs, intestinal haemorrhages and the presence of liquid in the intestine are also observed. Histopathological damage is specially evident

in the intestine, with severe enteritis, detachment of the epithelium, haemorrhages and inflammation. As in the case of *E. leei*, direct fish-to-fish transmission has been demonstrated, with subsequent influence on the disease dispersion and impact. Diagnosis is mainly based on microscopical examination of fresh smears and histopathology. A PCR assay is available (restricted use).

Current status based on answers received

In the survey, it is reported as enteric Myxosporea or myxidiosis, and only 2 laboratories from 1 country have diagnosed it in turbot, in mortality cases and in routine studies. Diagnosis was based on histopathology in 1 laboratory, and the other has also used macroscopic and microscope examination, immunohistochemistry, electron microscope and PCR.

Kudoa spp.

These marine myxosporeans (Multivalvulida) infect the muscle of many marine fish forming plasmodial cysts. Heavy infections can cause unsightly white cysts or soft texture in filets, which gives the name to the disease (soft flesh condition), with subsequent lowering of market value. In aquaculture, *Kudoa* infections have been described in salmonids (mainly Atlantic and Pacific salmon), and *Seriola*. Reported findings in gilthead seabream are occasional. The soft texture of muscle fillets and the presence of white cysts can be observed with the naked eye. Diagnostic must be confirmed by microscopic detection of the parasite in fresh or stained squash preparations or in histological sections.

Current status based on answers received

In the survey, it was only reported by 1 laboratory in Portugal in routine examiantion of *Sardina pilchardus*.

Sphaerospora dicentrarchi

Originally described from *Dicentrarchus labrax*, the type host, it can be ocassionaly found in some other fish species, as mullets. This histozoic parasite can be considered systemic, though it has speciall affinity for the connective tissue of gall bladder and intestine. It is usually found in chronic infections, without external clinical signs, though massive infections have been associated with extensive mortalities in juvenile fish. Diagnosis is mainly based on histopathology, though groups of spores can be seen in microscopical examination of fresh smears of gall bladder and intestine.

Current status based on answers received

In the survey it was diagnosed by 5 laboratories in 4 countries (1 laboratory without indication of country), usually in the type host *D. labrax*, but sphaerosporosis is indicated in *S. aurata* from Greece, and *Sphaerospora sparusaurati*, in the same host from Malta, which is a probable misidentificacion with *Ceratomyxa sparusaurati*. Sphaerosporosis was diagnosed more frequently in routine studies, but also in some mortality cases. Diagnosis was mainly based on histopathology and/or microscopical examination of fresh samples.

Sphaerospora testicularis

The only known host is *D. labrax*. This myxosporean has a very specific location in testes, so only male fish are affected. Heavy infections can result in parasitic castration of valuable broodstock fish. Affected testes can appear inflammed or necrotic at necropsy. In very heavy infections, the parasite invades the serosa and other organs, producing abdominal swelling and ascitis, though this situation is rather exceptional. In histopathological study parasite stages are observed filling the seminal tubules. In the reproductive period, the parasite can be easily diagnosed at microscope in fresh smears of seminal fluid.

Current status based on answers received

In the survey, it was only diagnosed in 2 laboratories from Greece. An increased susceptibility to bacterial diseases, but doubtfull direct association with mortality are indicated.

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Monogenea

Members of Monogenea, mostly ectoparasites of gills and skin, can cause different degrees of damage in parasitized fish. Mortalities may appear in moderate or heavy infections, mainly in juvenile fish, increasing with water temperature. Clinical signs include lethargy, anoxia, loss of appetite and scratching. Mucus excess, opacity, and even ulcers or haemorrhages may appear. Gill histopathological signs include focal hyperplasia, lamellar fusion, haemorrhages and inflammatory infiltration. Monogeneans are usually very host specific, though in certain culture conditions some species can be found in unusual hosts. Diagnosis is mainly based on direct methods of fresh examination and histopathology. Identification to the species level usually needs the observation of fresh parasites at microscope and comparison of morphometric features. Different treatments have been assayed against Monogeneans, including organophosphates (though its use is restricted by legal regulations) or formaline (mainly for larval stages). However, some infections are very refractory to treatments, so general preventive measures are strongly recommended, as well as periodic examination of fish.

Monogenea include 2 main groups, *Monopisthocotylea* (with a simple adhesive disc) and *Polyopisthocotylea* (with a complex adhesive disc including clumps and hooks).

Among *Monopisthocotylea* the most significant species for cultured fish are *Gyrodactylus* spp., *Dactylogyrus* spp., *Diplectanum* spp. and *Furnestinia* spp.

Polyopisthocotylea include several species of pathological concern for fish cultures, most of them belonging to the family Microcotylidae, and some to Heteraxinidae.

Gyrodactylus spp.

Gyrodactylus spp., causing gyrodactylosis, are mainly typical from freshwater fish. Some species are very pathogenic for salmonid fish, specially *G. salaris*, considered the most pathogenic species, and included in the list III of the EEC policies.

Current status based on answers received

In the survey, gyrodactylosis has been diagnosed in 9 laboratories from 5 countries. Several freshwater fish are affected, mainly *Oncorhynchus mykiss*, but also other salmonids, some cyprinids, *Scardinius*, and tropical exotic fish. However, gyrodactylosis has also been reported occasionally from marine fish, such as Sparidae (mainly *Sparus aurata*) and *Dicentrarchus labrax*.

Dactylogyrus spp.

This genus include numerous species, mostly parasites of freshwater fish. Cyprinids are the main hosts of these monogeneans, but fish of many other families are also affected. The eel parasites *Pseudodactylogyrus* spp. are included in the same superfamily Dactylogyroidea. The pathologic significance is very dependent on the species and intensity of infection.

Current status based on answers received

In the survey, dactylogyrosis was diagnosed in 11 laboratories of 6 countries. Reported fish hosts include *Cyprinus carpio, O. mykiss*, *Anguilla anguilla* (probably *Pseudodactylogyrus*) and tropical fish from fresh waters. Among marine fish, *Dactylogyrus* has been found in *S. aurata* and *D. labrax*. A misidentification with the respective specific monogeneans, *Furnestinia* or *Diplectanum* could have occurred, specially for juvenile stages.

Diplectanum spp.

The best known species of these genus are *D. aequans* and *D. laubieri*, parasites of *D labrax*. Their dispersion is very wide in the Mediterranean and Atlantic areas, mostly coinciding with seabass distribution. *D. aequans* is considered more pathogenic, mainly for juveniles and brood stocks.

Current status based on answers received

In the survey, *Diplectanum* was reported by 3 laboratories, from 3 countries (France, Turkey and Spain), both in mortality cases and routine examination. Surprisingly, on one occasion it was recorded from *S. aurata*, and not from the usual host *D. labrax*.

Furnestinia spp.

This genus belongs also to the family Diplectanidae. *F. echeneis* is quite frequent in *S. aurata* in different Mediterranean countries. It is not considered very pathogenic, though certain damage can occur in heavy infections.

Current status based on answers received

In the survey, it was diagnosed in 5 laboratories of 4 countries (Greece, Israel, Spain and Turkey), usually in the specific host, *S. aurata*, but in one case in *D. labrax.* Findings are reported in mortality cases, routine and occasional samplings, sometimes found in mixed infections with other monogeneans or with protozoans.

Microcotylosis

In the last years, microcotylosis has affected severely the sparid cultures, causing important loses. Besides the gill dysfunction usually associated with monogenosis, microcotylids produce increased damage due to their haematophagus condition, causing anemia and poor fish condition. These monogeneans are very frequent in Mediterranean cultures of sparids and *D. labrax*. The involved species have been identified as *Sparicotyle chrysophry* parasitizing Sparidae, and *Serranicotyle labracis*, parasite of seabass. In the Heteraxinidae, *Heteraxine* spp. important parasites for Japanese amberjack (*Seriola* spp.) cultures, have been also recorded in Mediterranean amberjack.

Current status based on answers received

In the survey, microcotylidis were diagnosed by 10 laboratories, belonging to 5 countries (Cyprus, France, Greece, Portugal and Spain). *S. aurata* is the most frequent host, though there were some records in other sparids, and one in *Seriola*. The number of cases per year was not generally very high. Most of them were associated to mortalities, though they were also diagnosed in routine examinations and occasional samplings. Some laboratories reported heavy infections and significant mortality, caused by the associated anaemia and anorexia. The condition may be related to bad husbandry, fouled nets or high stock density. Secondary bacterial infections were also reported. Diagnosis was always based on fresh microscopical or histological examination, though macroscopical examination was also indicated. No control measures were indicated for monogenosis in the survey.

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Trematoda Digenea

Diplostomum spp.

The metacercarial phase of these digeneans parasitizes the eye of fish, though it can be occasionally found in other organs, including the brain. Numerous fish species are susceptible to metacercaria of Diplostomatidae. Clinical signs consist of cloudness of eye lens, leading to cristalline opacity and blindness. Dark body coloration can be also observed. Acute mortality is very rare, but poor condition is frequent, probably due to the problems with locating food. Diagnosis is based on macroscopical or estereomicroscopical observation of eye opacity, followed by microscopical detection of metacercariae in squash preparations at low magnification. No efective treatment is known, so prevention based on breaking of transmission chain, avoiding the presence of snails intermediate hosts, is recommended.

Current status based on answers received

In the survey, *Diplostomum* was recorded by 3 laboratories from 3 countries (Croatia, Italy and Turkey) in numerous freshwater fish (mainly cyprinids, but also salmonids and others) and in exotic fish. Records were quite frequent and, when indicated, correspond to routine examinations. Diagnosis (if stated) was based on histopathology and on macroscopical and microscopical examination.

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Sanguinicolosis

Members of the family Sanguinicolidae are parasites of the circulatory system of fish. Adults are located in the heart or kidney. Eggs can accumulate in blood vessels, and are quite frequent in the gills, producing vascular obstruction. Haemorrhages can also occur at miracidial eclosion. Moderate inflammatory response can be produced and the lesions are not extremely severe in moderate infections. Some species are parasites of freshwater fish, particularly salmonids and cyprinids, but there are also Sanguinicolidae of marine fish, as *Sparus aurata* and *Seriola* spp.

Current status based on answers received

In the survey, sanguinicolosis was diagnosed by 4 laboratories of 3 countries (Croatia, Greece and Spain). Records of 1 laboratory (Croatia) corresponded to freshwater fish (cyprinids) and the reamining 3 laboratories found it in the marine fish *S. aurata*, both in routine examination and in mortality cases. In Greece, a serious affection of broodstock and subadults in cages was reported. Diagnosis was mainly based on histopathology.

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Cestodes

Difilobotriosis, bothriocephalosis, eubotriosis, ligulosis, trienoforosis

These platyhelminthes may parasitize fishes in larval or adult stages, sometimes causing diseases in cultured fish with variable economic impact. The life cycle involves at least one intermediate host. Most species causing disease in fish of economic importance fall within three orders: Caryophyllidea (*Caryophyllaeus* and *Khawia*), Pseudophyllidea (*Bothriocephalus, Diphillobothrium, Ligula* and *Triaenophorus*) and Proteocephalidea (*Proteocephalus*).

Cestodes in fish usually do not cause mortality, though poor condition is frequently observed, mainly in heavy infections.

Caryophyllidea

Adult cestodes of the genera *Caryophyllaeus* and *Khawia* parasitize the digestive tract of cyprinids and salmonids, producing different degrees of damage and economic impact in the cultures. In heavy infections, abdominal swelling and poor condition can be observed, mainly in small fish. After necropsy, cestodes can be seen with the naked eye in the intestine.

Current status based on answers received

In the survey, only one member of *Caryophyllidea (Khawia* sp.) was reported by 1 laboratory (Croatia) in cyprinid fish and *Salmo trutta*, though the number of cases diagnosed was low in the 3 years.

Pseudophyllidea

Bothriocephalus spp. and *Eubothrium* spp. are parasites of freshwater and marine fish. *B. scorpius*

has been found in turbot. Some *Eubothrium* spp. are frequent in salmonids. *Triaenophorus* spp. can cause severe pathology in some fish, as pike and trout. Fish are usually the final hosts, but some small fish may act as reservoirs of certain species.

Larval stages (plerocercoids) of *Diphyllobothrium* spp. parasitize the muscle and visceral organs of different freshwater fish, mainly salmonids, esocids, percids and coregonids. Mammals are the final hosts of these tapewormes, which can also infect humans. Thus, diphyllobothriasis in fish must be controlled due to its zoonotic character. Plerocercoids of *Ligula* spp. parasitize the visceral cavity of different fish, mainly some freshwater fish, as cyprinids and catostomids.

Current status based on answers received

In the survey, *Triaenophorus* was reported by 1 laboratory of 1 country (France) in *Perca fluviatilis*. Other findings of pseudophyllidean cestodes include those of *Bothriocephalus*, *Eubothrium*, and *Ligula* reported by 1 laboratory (Croatia) in several freshwater fish.

Proteocephalidea

Fish can act as definitive and intermediate hosts of different *Proteocephalus* spp. Affected fish include salmonids and coregonids, among others.

The only finding of Proteocephalidea in the survey was reported by 1 laboratory (Croatia) in different feshwater fish.

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Crustaceans

The main parasitic crustaceans of commercial fish belong to the groups Copepoda and Isopoda. Among Copepoda, species of the genera *Argulus*, *Caligus, Ergasilus, Lernanthropus, Lernaea, Lerneaocera* and *Lepeophtheirus* (sea lice) parasitize different freshwater and marine fish. They are located on the gills, bucal cavity and skin and produce different degrees of damage, even mortality, depending on the fish species and degree of invasion. Clinical signs include occasional rubbing, decrease of condition and gill damage leading to respiration problems. Inflammed wounds, ulcers and mucous excess can be produced as a consequence of crustacean bites, even affecting muscle. Salmons affected by sea lice may show small white-grey spots. In addition, crustaceans can carry or facilitate other infections.

Different Isopoda have been reported, mainly in sea fish, including *Nerocyla orbygnyi*, *Anilocra physodes*, *Gnathia* and *Paragnathia,* parasitizing seabass, seabream and other sparids, *Seriola* spp., or mugilids. Clinical signs include lethargy, anorexia and respiratory difficulties, as a consequence of gill damage and necrosis. The picture is frequently complicated by secondary bacterial infections. Mortality can appear mainly in juvenile fish.

Some parasitic Crustacea are visible with the naked eye, but generic and specific identification require the observation at microscope or estereomicrocope. Routine examination is recommended as preventive measure. The use of organophosphates for treatment has legal restrictions.

Current status based on answers received

In the survey, sea lice disease was recorded by 5 laboratories of 5 countries, involving different copepoda of freshwater (*S. trutta, O. mykiss, C. carpio* and other cyprinids, *Esox* and *Scardinius*) and marine fish (*D. labrax, S. aurata* and *Seriola dumerili*). Records were not frequent and, when stated, they were associated to both routine examination and mortality cases. One laboratory indicated low to moderate mortalities in heavy infections of *D. labrax* and *S. dumerili*.

Diagnosis in the survey was mainly based on macroscopical observations, less frequently on fresh microscopical examination.

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