

2010

Fish Disease Manual



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Hamish D. Rodger

FISH DISEASE MANUAL

Hamish D. Rodger, BVMS, PhD,
Vet-Aqua International, Oranmore, Co. Galway, Ireland

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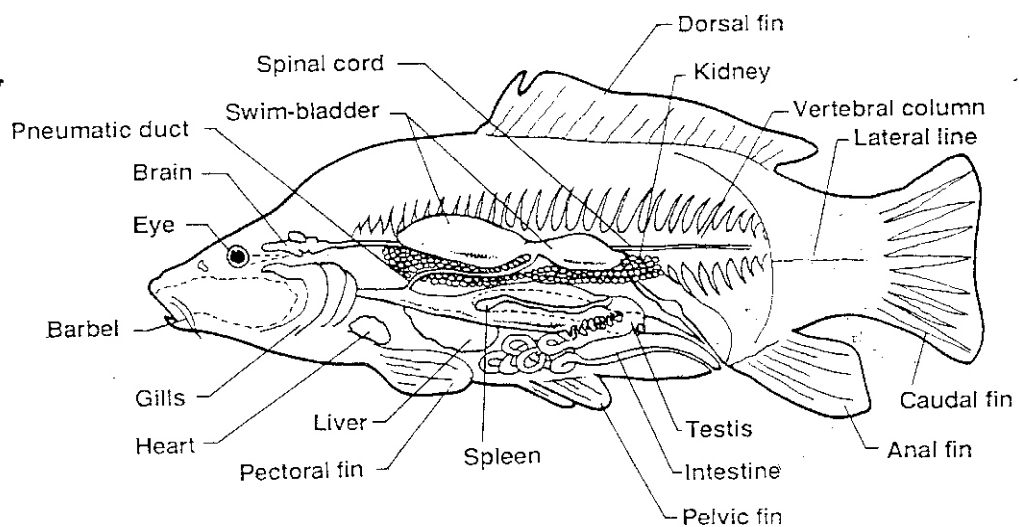
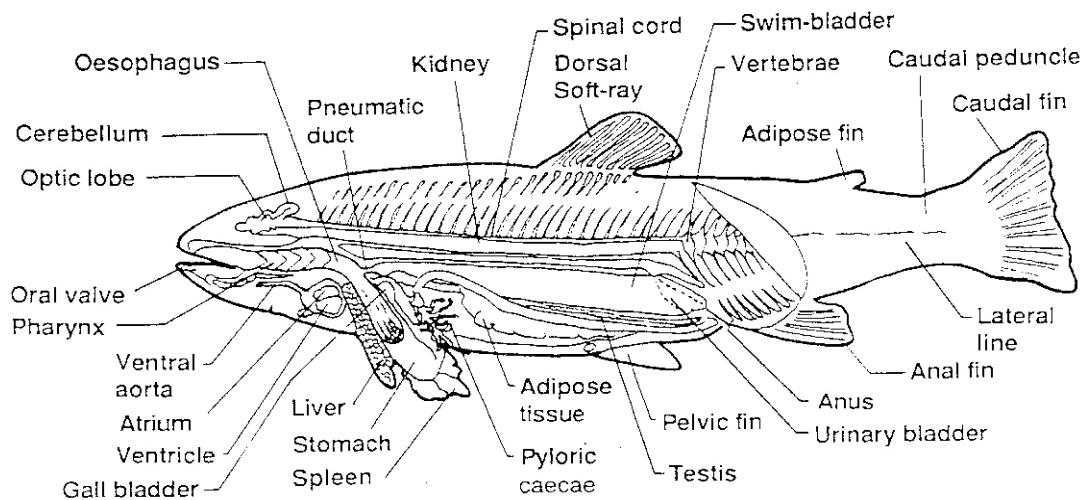
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BASIC FISH ANATOMY & DISSECTION GUIDE

Fish are cold-blooded or poikilothermic animals, their body temperature varying passively in accordance with the temperature of the surrounding water. Although fish as a group are tolerant of a wide range of temperatures, individual species have a preferred range or optimum and changes in this range, significantly affects the biology of fish with the rates of all chemical reactions and processes within their bodies showing 50% increases with each 5°C rise in temperature.

The body shape of fish is usually streamlined, an important prerequisite for successful aquatic life, due to the 800-fold higher density that water is to air, and most predatory fish (such as bass and salmonids) are ovoid in cross section and torpedo-like or fusiform in shape.



Organ systems of trout (upper) and carp (lower).

Fish examination and dissection guide

1. Locate various fins (dorsal, adipose[if present], tail, anal, pelvic and pectoral) and note whether fin rays present and if any degree of erosion or shortening.

2. *Locate nostrils, operculum (gill cover), lateral line, eyes and anus/urogenital opening.*
3. *Examine inside mouth and feel for teeth and gill rakers.*

The **dorsal fin** and **tail fin** are vital for locomotion, but can be subject to erosion or damage in a crowded farm situation. Dorsal fin rot or erosion is often seen where stocking levels are too high, nutrition is marginal or water temperatures have been at the low end of normal ranges, for that fish species, for a prolonged period. Aggression, especially at feeding time, can result in fin and tail nipping, which will result in erosion or tail rot. Too high a stocking level may also damage pectoral fins or when the tank/pond sides are constructed from an abrasive material. The adipose fin acts like the spoiler of a car and is only present in some fish species. Clipping of this fin in salmonids has been used for identification purposes.

The **nostrils** have epidermal flaps in some species and are blind ending pits, which house nerve endings and mucus cells. The nerves run directly to the forebrain. Pollutants can damage these sensitive surfaces, which are relied on greatly by migratory species such as salmon and eels. The **lateral line** is the main vibration sense organ in fish and can be damaged by pollutants, chemicals or parasites. It runs as a paired canal along the flanks, has an integumental cover, which is punctuated by sequential pores along its length. The mechanoreceptors or neuromasts are located basally in the canal and are stimulated by changes in the external *milieu*, in terms of displacement or vibration.

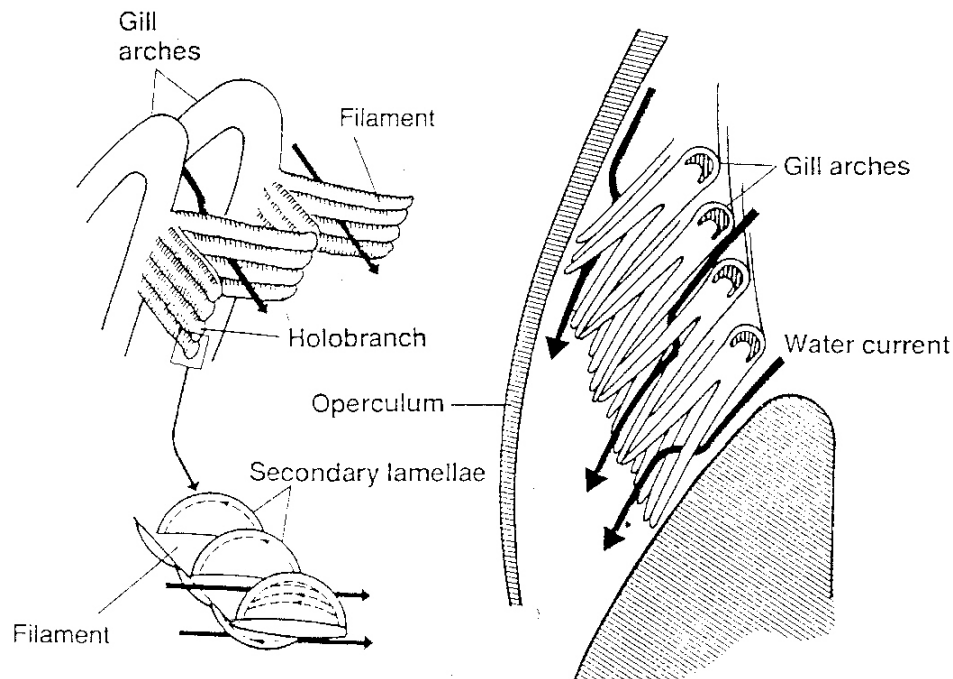
The **operculum** or gill cover provides physical protection for the gills but is also an actual component of the respiratory mechanism. Foreshortened opercula are a problem in many species and can be either genetic or environmental in origin.

The **eyes** have fixed, spherical lenses, which are virtually free floating and are vulnerable to parasites, environmental damage and nutritional deficiencies. The lens protrudes partially through the iris to provide a very wide angle of view and the iris is limited in reaction to light intensity, having a poorly developed sphincter and dilator muscle. The cornea may be tinted in some species.

The **gills** are located beneath the opercula and consist of four white bony or cartilaginous arches and the red or pink gill lamellae. The 5th gill or **pseudobranch** is an embryonic red gill-like structure located on the underside of the operculum but is not present in all species (e.g. eels). Its function remains to be defined in full but it has an endocrine and regulatory function as well as a hyperoxygenation function for the retinal blood supply. The gills undertake the tasks of the uptake of oxygen and associated loss of carbon dioxide. Secondary lamellae branch off the primary lamellae and the numbers present reflect the fishes lifestyle i.e. slow moving bottom dwellers may have only 10 lamellae per mm of filament, whereas fast swimming predators will have 30 to 40. A complex of capillary channels is present in the secondary lamellae and the thin lamellar walls (usually only one cell layer thick) readily allows for respiratory exchange between the blood and the surrounding water. Blood flow is arranged so that the direction of flow is opposite that of the water crossing the gills thereby increasing the efficiency of respiratory exchange. The spikes on the gill arches are the **gill rakers** and these prevent food materials entering the gill chambers. They are particularly well developed in plankton or filter feeding fish.

As well as a respiratory function, the gills are also responsible for regulating the exchange of salt and water and play a major role in the excretion of nitrogenous waste products

(ammonia). Even slight structural damage can thus render fish very susceptible to osmoregulatory as well as respiratory difficulties.



Diagrams of gill structure (adapted from Lagler 1982)

With low oxygen levels in the water column or gill damage that reduces the respiration efficiency, a direct consequence will be an increased ventilation rate, which can be observed clinically as an increased rate of opercular movement. This can be observed as water temperature rises due to the fact that less oxygen can dissolve in the water at a higher temperature. Gill epithelium is very prone to damage from parasites, water borne irritants or toxins, and high levels of suspended solids.

4. Examine scales under the microscope and look for chromatophores.

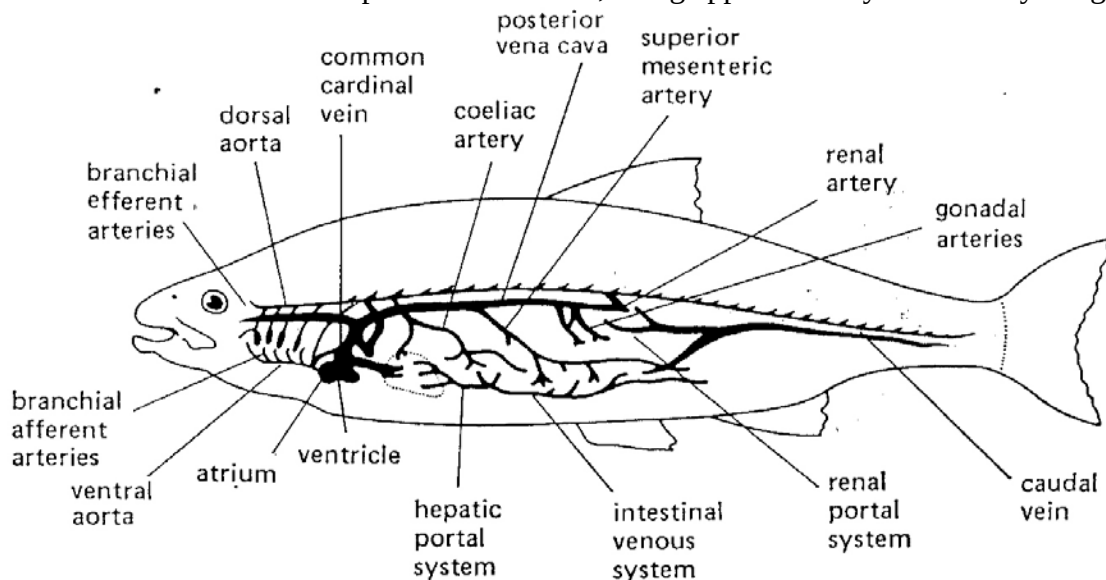
The **skin** of fish is very important from various aspects, and there are more losses of fish through failure to look after the skin than any other system. Fish skin can be viewed as having two main layers; the outer **epidermis** and the underlying **dermis**. Outwith the epidermis is the cuticle or **mucous** layer, which in addition to providing lubrication, makes the skin less permeable and prevents entry of pollutants and microorganisms. The mucous is secreted from mucus cells which reside in the fragile epidermis; the epidermis is composed of living cells to the outermost layers. The **scales**, which are calcified flexible plates, grow out from the dermis and in higher teleosts have spicular processes from their external posterior edge. Growth rings or annuli are visible on scales of wild fish, similar to the rings seen in the main trunks of trees.

Pigment cells or chromatophores are highly developed in fish and the melanophores are the dark brown/black pigmented cells, iridophores are silver and there are a range of lipophores which contain the organic solvent-soluble pigments (reds, yellows, etc.). The decision on the color the fish should be depends to some extent on what it sees outside and further on its health status; if it is sick or starving the fish releases the melanin concentrated in the melanophores and this results in a dark fish.

5. Lay fish on side and remove abdominal body wall, operculum and integumental wall over cardiac cavity.
6. Expose viscera and identify oesophagus, liver, gall bladder, swim bladder, spleen, stomach, pyloric caecae (if present), intestine and kidney.
7. Remove heart and identify three main chambers (triangular ventricle, darker red soft atrium and white elastic bulbus arteriosus).

In circulation the venous deoxygenated blood enters the thin walled cardiac **atrium**, is then pumped into the muscular **ventricle**, and from there into the fibroelastic **bulbus arteriosus**. Coronary vessels run over the outside of the ventricle, supplying the compact muscle. Heart rates vary considerably according to temperature, from 15/min in trout at 5°C to 100/min at 15°C. The ventral aorta runs from the heart and distributes blood to the gills via the afferent branchial arteries.

For **blood sampling** the preferred sites are the caudal vein from either a ventral or lateral aspect, cardiac puncture (ventrally) or the brachial plexus (caudal to the gills). Blood volumes in fish are small compared to mammals, being approximately 5% of body weight.



Schematic representation of the circulation of a typical teleost fish.

Haematology and blood biochemistry of fish is an area that has been utilized to only a limited extent for clinical investigations. Normal values for many species remain to be established and these will vary according to season, age, temperature, genetic strain, physiological status, nutrition and sampling methodology.

Haemopoietic tissue in the fish is predominantly located in the stroma of the spleen and the interstitium of the kidney. To a lesser extent it is also found in the periportal areas of the liver, the intestinal submucosa and the thymus. The **kidney** of fish is usually located in a retroperitoneal position up against the ventral aspect of the vertebral column. It is usually divided by function and histology into the anterior or head kidney, which is predominantly haemopoietic, and posterior or excretory kidney. In salmonid kidneys the corpuscles of Stannius can be seen as paired white nodules in the mid-kidney. These are endocrine glands and appear to be involved with calcium metabolism, although their exact role is unclear.

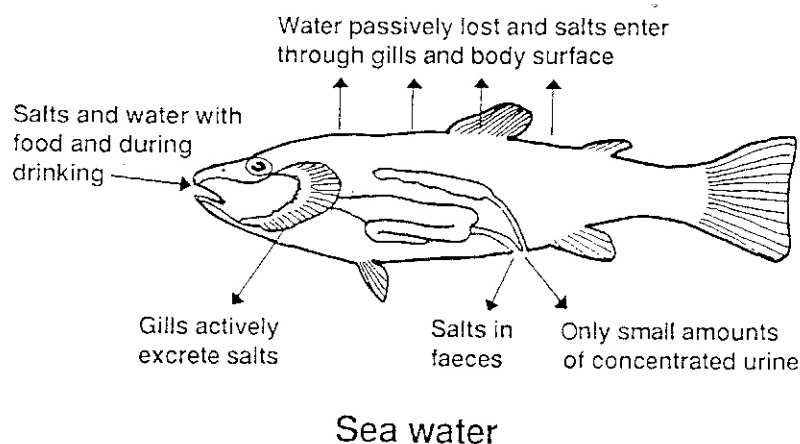
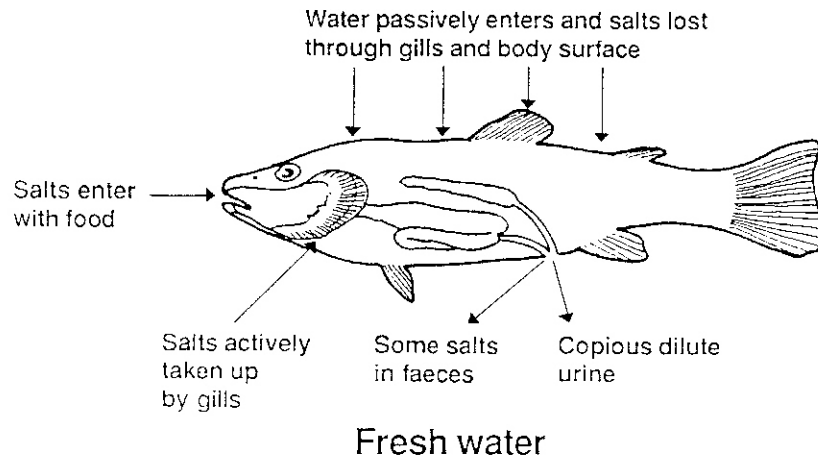
The **spleen** is located in the peritoneal fat, near the greater curvature of the stomach or the first flexure of the intestine. It is usually single, although in some species it may be paired. In

some species the pancreas is located as a subcapsular layer in the spleen, but in most species the main elements of the spleen are the ellipsoids, the pulp and the melanomacrophage centers (MMC). The **thymus** is a paired organ, an ovoid pad of primary lymphoid tissue located subcutaneously in the dorsal commissure of the operculum.

The excretory section of the fish kidney varies dramatically depending on whether the fish is marine, euryhaline or freshwater, reflecting the significant differences in their respective functions. The major work on this field is that of Hickman & Trump (1969)¹. In **freshwater**, fish drink very little but produce copious amounts of dilute urine; few salts appear in the urine because the kidneys reabsorb them. Salts are also gained from the surrounding water by the active uptake through the gills by special chloride cells found at the base of the secondary lamellae. In **seawater**, fish drink a lot (up to 15% of bodyweight per day), selectively excrete monovalent ions (Na^+ , Cl^-) through the gills and produce small amounts of concentrated urine. The chloride cells in the gills are responsible for removing the excess salt from the blood and passing them out to the water.

Salt and water regulation as well as excretion require that gills and kidneys are in healthy condition; damage to either or both organs will result in an inability of the fish to respond to osmotic change. For this reason kidney damage, from diseases such as bacterial kidney disease (BKD) or nephrocalcinosis, may not be apparent in salmonids until such fish are moved to seawater where they will suffer high mortalities.

¹ Hickman, C. P. & Trump, B. F. (1969) The kidney. In *Fish Physiology*, ed W. S. Hoar & D. J. Randall, vol. 1, pp 91 – 239. New York and London: Academic Press



Osmoregulatory and ionic factors and their regulation in the freshwater (upper) and seawater (lower) environments.

The digestive systems of fish vary in a number of areas in accordance with species; herbivorous fishes have long intestines and little or no stomachs (grass carp, silver carp) and carnivorous fish having larger stomachs and short intestines (salmonids, striped bass). Other variations include dentition and presence and numbers of diverticula. The **stomach** is usually sigmoid and highly distensible. **Pyloric caecae**, blind ending diverticula from the distal, pyloric valve region of the stomach and from the anterior intestine, are found in many species, most notably salmonids where they may number 70 or more. Histologically these resemble intestine.

The teleost **liver** is relatively large and the color depends on diet; in wild fish it is usually reddish brown in carnivores and lighter brown in herbivores but seasonal variations can occur with yellow or cream also observed. In farmed fish the color is usually a reflection of the dietary lipid levels, and is normally lighter than wild fish. In some species the liver can be a compound organ, known as the **hepatopancreas**, where exocrine pancreatic tissue is located around hepatic portal veins. The fish liver is not lobulated like those of mammals. The **biliary** system also differs in that intracellular bile canaliculi occur which eventually anastomose to form typical bile ducts. The bile ducts fuse and ultimately form the **gall-bladder**. The **pancreatic tissue** is more variation in location in fish than other abdominal viscera, but the most common site is in the mesenteric fat interspersed between the pyloric caecae.

The gas filled **swim bladder** is a characteristic feature of many teleost fish, although absent in bottom dwelling fish and some fast swimming pelagics. Its primary function is a buoyancy mechanism, but it is also used for sound and pressure reception and in some species is equipped with drumming muscles for sound production. The embryonic connection between the gut and the swim bladder is retained as a pneumatic duct in the more primitive fish species (**physostomes**) but has been lost in most of the spiny rayed fish (**physoclists**). In many of the physostomes the swim bladder is two chambered, separated by a diaphragm, with the anterior chamber associated with gas reception and retention and the posterior chamber involved with gas reabsorption.

Teleost fish show more diversity in their reproductive patterns than any other group in the animal kingdom. Although most species have male and female sexes, hermaphroditism and bisexuality occur and both parthenogenesis (development from an unfertilized ovum) and gynogenesis (development from an ovum stimulated to divide by penetration from a sperm which does not contribute genes) are also recorded. The **gonads** develop as paired organs lying just below the kidneys. In immature fish these are rudimentary thread-like structures and in mature fish the ovaries can constitute up to 70% of the body weight. The **testes** have a vas deferens or collecting duct, which conducts mature spermatozoa to the excretory meatus at the urinary papilla, and the **ovaries** pass the ova to the outside via the oviduct or into the abdominal cavity in the more primitive species, for evacuation via the genital opening. Live bearers store eggs in a pouch referred to as a uterus, but this is in essence a simple storage space.

8. *Cut a cross section through the fish, anterior to the caudal peduncle and examine for red and white muscle, vertebral column and spinal cord.*
9. *Carefully remove the top of the cranium and expose the brain. Section through the anterior olfactory nerves and the spinal cord so the entire brain can be removed.*

Most fish swim by passing a wave of increasing amplitude along the body and this is generated by sequential contraction from head to tail of the muscle blocks or myomeres. Histologically and biochemically the **muscle** can be divided into two types:

- a) the red, aerobic, slow contracting muscle fibers and
- b) the white, anaerobic, fast contracting muscle fibers.

In some species of fish they are also pink fibers, which are sandwiched between the two types and these appear to be intermediate in function as well as location. The well vascularised red muscle is best observed as the triangles of darker muscle located over the lateral line and midline dorsally. The majority of the body muscle is the white muscle and is usually only used for escape or chase situations.

There are two types of **bone** in teleosts: cellular as in other vertebrates and acellular, which is unique in vertebrates, and found only in the advanced teleosts such as perch and sunfish. The majority of fish species have no haemopoietic tissue in their bone spaces and vascular canals.

The piscine **brain** is similar in its basic components to the brain of higher animals; however, there are significant differences in form and complexity. It can be divided simply into five main areas:

- 1) the telencephalon or forebrain (olfaction, color vision, memory)
- 2) the diencephalons (thalamus, epithalamus and hypothalamus)
- 3) the mesencephalon (optic lobes)
- 4) the metencephalon or cerebellum and
- 5) the medulla oblongata, which merges with the **spinal cord**.

The **pituitary gland**, which incorporates the neurohypophysis, can be located at the ventral base of the brain in a bony cupula. Its function is similar to that in other vertebrates in that it conducts the body orchestra.

Further reading

Brown, L. (1993) *Aquaculture for veterinarians*. Pergamon Press, Oxford, UK

Stoskopf, M. K. (1993) *Fish medicine*. W.B. Saunders, Philadelphia, USA

SAMPLING FOR DISEASE DIAGNOSIS

The ideal specimens for disease investigation or health monitoring are live fish. Samples can be taken from these either on site or transported live to the appropriate laboratory.

Transport of live fish

Place the fish, representative of the problem, in a plastic bag filled to approximately a third with water and two thirds oxygen. Seal the bag with cable ties or equivalent and place in another bag and seal again. Then place the bag on ice or cool-packs in an insulated box e.g. polystyrene, place more ice on top and seal the container.

The maximum transport time depends on water temperature, and the ratio between biomass, water volume and oxygen. As a rough guide the transport time should not exceed 12 hours and the biomass should not exceed one third of the water volume. Transport time is significantly reduced if oxygen is not used.

Live fish weighing more than approximately 300g should not be sent by normal goods transport (air, rail or road), but should either be sampled on site or sent via specialised forms of transport.

Transport of fresh material

Unopened fish, reproductive products, virology samples, fish heads (for Myxobolus) may be dispatched for laboratory investigation in the fresh state.

All samples must be chilled to as close to 0°C without freezing. Pack samples in ice and in an insulated container and dispatch. The maximum transport time is 24 hours.

PARASITOLOGICAL SAMPLING OF FISH

- a) Examination of skin: first stun the fish with a sharp blow to the head. Then take scrapings for microscopic examination using a scalpel and scrape from front to back of the fish or around the fins (Figure A). Place scraping on a clean glass slide with a drop of water from the holding facility and cover with a coverslip.
- b) Examination of gills: following gross examination of the gills, clip a small portion of gill lamellae with sharp scissors and place on glass slide (alternatively scrap the gill lamellae with a scalpel), add a drop of the holding tank water and cover with a coverslip. Examine under low power with high contrast or phase.
- c) Examination of other organs: any other organs suspected of having parasitic infection can have squash preparations made from small sub-samples of tissue and examined similarly using light microscopy.
- d) Record and/or draw your findings.



Figure A. Skin scrape of salmon parr using a scalpel.

HISTOLOGICAL SAMPLING OF FISH

Histology encompasses the scientific area concerned with the structure of tissues and histopathology the relevant branch of pathology. Histopathology can therefore provide information on the processes and changes occurring in tissues and in many cases form the basis for disease diagnosis and prognosis.

Accurate sampling of tissues for histology is a vital part in the diagnostic procedure and to follow are guidelines for onsite sampling. Before sampling any fish note any behavioural abnormalities or visible external lesions.

- 1) As with other diagnostic procedures, a mixture of sick (moribund) and healthy fish should be sampled. Dead fish (recent mortalities) provide little accurate information and are virtually useless for histology.
- 2) Sacrifice the chosen fish by stunning the fish by a blow to the head or through anaesthesia.
- 3) Phosphate buffered formalin is the tissue fixative of choice for the majority of samples. 10% formalin is the usual concentration (formalin being 40% formaldehyde). Care should be taken as formalin is irritant, especially to eyes.
- 4) Following killing of the fish, samples should be immersed in formalin as quickly as possible (all tissues should be sampled within five minutes of killing) as post-mortem changes will occur rapidly in these cold blooded animals.
- 5) Remove a small piece of tissue ($<1\text{cm}^2$) from the organs and include any areas showing gross abnormalities.
- 6) Sample gill, heart, spleen, liver, pyloric caecae/pancreas, kidney, section of skin and muscle (at lateral line) and brain. Small fish or fry may be sampled whole with the abdomen opened to allow proper fixation of the internal organs.
- 7) Ensure amount of formalin in pot is approximately 10 times the amount of tissue.

8) Record any details about fish on paper e.g. body condition, weight, length, feeding, internal appearance, etc..

9) Label the sampling pot with a water proof marker and ensure that each pot will not leak.

10) Dispatch/deliver samples with relevant details and clinical history and contact the diagnostic service by telephone or fax.

Phosphate Buffered Formalin Solution (PBFS)

Formula:	Formaldehyde (40%)	100ml
	Water (tap OK if distilled unavailable)	900ml
	disodium hydrogen orthophosphate	6g
	sodium dihydrogen orthophosphate	5.5g

Dissolve dry powder in water, then add formaldehyde.

THE GRAM STAIN

This is a differential staining method and is the first characterisation test applied to all bacterial isolates. It demonstrates bacteria of different types:

Gram positive - those which resist decolourisation
- stain blue/purple

Gram negative - those which are decolourised
- stain red/pink

The difference in colour reaction is due to the different chemical composition of the cell wall and membrane. This stain will also reveal the general shape of the bacteria.

It can be useful in the diagnosis of Bacterial Kidney Disease (BKD) and rainbow trout fry syndrome (RTFS) using kidney material taken directly from the fish.

METHOD

1. Take a clean slide and using a sterile loop, emulsify a small amount of material in a drop of sterile saline or distilled water.
2. Allow the slide to air dry, then heat fix it by passing it through a flame three times. Allow slide to cool.
3. Flood the slide with crystal violet for one minute.
4. Holding the slide at a steep angle, wash off the stain with Grams iodine and allow to sit for one minute.
5. Tip off the iodine and pour the alcohol/acetone mixture over the slide from the upper end, so as to cover its whole surface. Repeat until no more colour runs off, then wash gently in water.

6. After shaking off the excess water, flood the slide with the safranin counterstain and leave for approximately two minutes.
7. Wash the slide in water and dry.
8. Examine under X40 objective, then under oil immersion with X100 objective.

BACTERIOLOGICAL SAMPLING OF FISH

1) Examination and direct inoculation of solid media

Before examining a fish internally, the external body surface, including gills, tail and fins should be examined for the presence of any lesions. Observations should always be recorded on paper. Samples from these sites can be taken by searing the surface with a hot scalpel blade followed by insertion of a sterile bacteriological loop or swab. Material from the loop/swab is then plated out onto suitable agar medium by the spread plate technique.

Once external examination or sampling has been carried out, the body surface is opened to expose the internal organs. Care must be taken not to puncture the gastrointestinal tract. In the absence of any visible internal lesions a sample of kidney is taken and inoculated onto suitable agar medium. The surface of the kidney (or other organ) should be seared with a hot scalpel blade before insertion of the sterile loop (Figure B).

Agar plates containing the streaked out samples should be incubated and examined daily for any evidence of growth. The majority of bacterial fish pathogens will grow on Tryptone Soya Agar (TSA) within seven days. Media such as Marine Agar or TSA plus 1.5% salt (NaCl) can be used for marine pathogens.



Figure B. Bacteriological sampling of salmon kidney using a hot wire loop.

2) Kidney smears

Direct examination of Gram stained kidney smears may give an indication of bacterial septicaemia and is especially useful in the examination of fish for Bacterial Kidney Disease (BKD).

A small portion of kidney is emulsified in a loopful of sterile physiological saline on a microscope slide, allowed to air dry then fixed by passing the slide through a bunsen flame several times.

Direct observation of the slide is carried out using the X40 and X100 (oil immersion) objectives after being stained using Gram's method.

3) Bacteriology plates or swabs can be dispatched to the lab for investigation but must be clearly identified and labelled. A written note regarding the history and clinical appearance of the fish and samples taken is important.

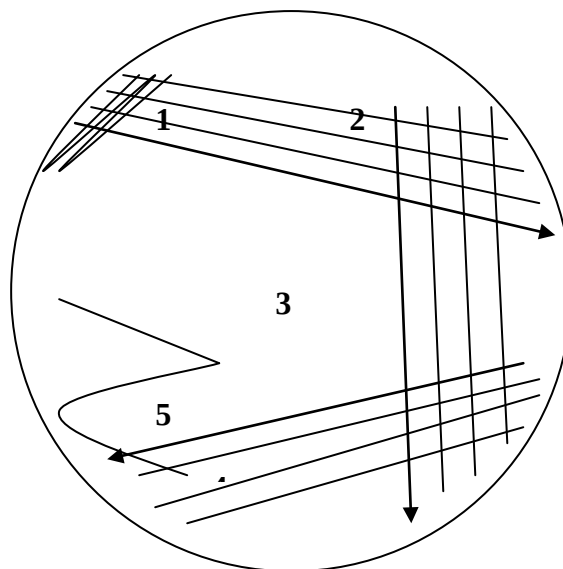
INOCULATION OF AN AGAR PLATE

Petri dishes containing solid medium (agar) are used to provide a large surface of media for the cultivation of micro-organisms. Inoculation of an agar plate is often carried out using the **streak plate technique**. This involves diluting the culture or other sample, e.g. kidney material, by smearing it across the surface of the agar. Organisms present in the sample will be separated and after suitable incubation each organism present will give rise to a colony. Although this colony contains many millions of organisms they will have all originated from one, and therefore all organisms in one colony are identical.

By using this method organisms can be cultured in the laboratory and if a mixed culture is present it will become apparent on plating out. This is essential before starting identification procedures as methods are only valid when carried out on pure cultures, i.e. cultures containing one type of organism.

Care should be taken that plates used for this purpose are dry. Also avoid unnecessary exposure of the agar surface to potential contamination from the environment throughout the procedure.

STREAK PLATE TECHNIQUE



1. Sterilise a bacteriological loop and allow to cool.
2. Pick up a small amount of sample using the sterile loop.
3. Inoculate the sample on a segment of the surface of the culture medium (1).
4. Sterilise the loop, allow to cool and touch edge of uninoculated area of medium to ensure coolness.

5. Spread part of the sample over about a quarter of the plate by making 3-4 parallel streaks with the loop (2).
6. Repeat streaking procedure as shown (3, 4 & 5), sterilising and cooling the loop between each sequence.
7. Label the underside (not lid) of the plate, writing only at the edge, and incubate or dispatch with related clinical notes and history. Ensure dispatched samples are adequately padded to protect against physical damage.

BLOOD SAMPLING

Fish blood may be sampled for disease monitoring or health status analyses in the following areas:

- i) haematology (examination of blood cells and blood cell indices), e.g. red blood cell count, haematocrit
- ii) blood biochemistry e.g. hormones, enzymes, etc.
- iii) plasma parameters e.g. plasma chlorides (for salt water challenge tests in salmon)
- iv) serology for pathogen antibodies i.e. salmon pancreas disease virus antibody screening
- v) virology and bacteriology: some micro-organisms can be screened for using frank blood.

For biochemistry and haematology an anticoagulant should be used in the blood collection tube and this is normally heparin (heparinised tubes can be purchased), however, the laboratory where samples are being submitted should be consulted for their normal requirements. For serology, bacteriology and virology blood should be taken without anticoagulants.

Where fish are large enough, blood samples can be taken under anaesthesia i.e. non-lethal sampling. The normal sampling site is from the tail vein and the easiest approach is ventrally at midline towards the vertebral column.

VIROLOGICAL SAMPLING

For virological sampling the fish organs and transport medium/conditions will be determined by the virus/es suspected. The laboratory where samples will be submitted should be contacted prior to sampling.

Further reading

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O.I.E. (2009) Manual of Diagnostic Tests for Aquatic Animals, 6th edition. O.I.E., Paris, France.

VIRAL DISEASES

Pancreas Disease (PD)

Definition

Pancreas disease (PD) is a viral disease that affects marine stage farmed salmon, usually in their first season at sea, and results in mortalities and/or poor growth.

History

PD first observed in Scotland in 1976, but subsequently recognized throughout Europe and North America. First confirmed in Ireland in 1985 and is now considered endemic.

Clinical signs of disease

The initial sign is usually a drop in feeding response, which rapidly proceeds to complete anorexia. Fish become unable to hold their body position in the water and may “hang” in the water column (Fig. 1). As the disease progresses, affected fish appear emaciated or mortalities occur. Internally there may be focal haemorrhage in the caecal fat, and yellow casts in the intestine.

Histopathology

Histopathology is most obvious in the exocrine pancreatic tissue where widespread necrosis occurs, leading to rapid loss of all acinar tissue. This is then replaced by either fibrous tissue or healthy pancreatic tissue. Focal to severe cardiomyopathies and more extensive skeletal myopathies are a further manifestation of this disease.

Aetiology

Salmonid alphavirus (SAV) of which there are six subtypes (SAV 1 to 6) with SAV 1, 4 and 6 present in Ireland. SAV is an alphavirus within the Togavirus family.

Diagnosis

Clinical signs and histopathology have been relied on for confirmation of the disease, however, cell culture, serology (VN) and PCR are also utilised.

Host and geographic range

Atlantic salmon and rainbow trout appear to be the susceptible species, and natural outbreaks have only been recorded in seawater. Scotland, Norway, Ireland, France, Spain and Washington State have all reported the presence of this disease. Rainbow trout farmed in freshwater are affected by “sleeping disease” due to SAV 2.

Source

Clinically affected fish and carrier fish, which have survived a previous outbreak. The original source is considered to be marine, probably wild fish or invertebrates, however, limited investigations have been undertaken to screen for the virus in wild species.

Immunity

A good antibody response is mounted to the virus and fish can be screened by serology for evidence of exposure to the virus. A commercial vaccine is in use in Northern Europe.

Treatment and control

Good management practices such as single bay management, fallowing and single year classes in single sites have all proven beneficial in controlling the disease. During an outbreak, a period of starvation (7 – 10 days) appears to reduce the impact of the disease. Health diets, low stress management and lice control are all important in reducing the impact of PD.

References

- McLoughlin, M. F. *et al.* (1996) Experimental pancreas disease in Atlantic salmon *Salmo salar* post-smolts induced by salmon pancreas disease virus (SPDV). *Diseases of Aquatic Organisms*, 26, 117 – 124.
- McVicar, A. H. (1987) Pancreas disease of farmed Atlantic salmon, *Salmo salar*, in Scotland: epidemiology and early pathology. *Aquaculture*, 67, 71 – 78.

Rodger, H. D. *et al.* (1994) Myopathy and pancreas disease in salmon: a retrospective study in Scotland. *The Veterinary Record*, 135, 234 – 235.

Ruane, N., Graham, D. & Rodger, H. (2008) Pancreas disease in farmed salmon: health management and investigations at Irish farm sites 2005 – 2008. *Marine Environment and Health Series*, No. 34, Marine Institute, Oranmore. 58pp.



Figure 1. Salmon affected by pancreas disease (PD) exhibiting lethargy and occupying an elevated position in the water column (left) and internally exhibiting yellow intestinal casts (right).

Viral Haemorrhagic Septicaemia

Definition

Viral Haemorrhagic Septicaemia (VHS) is an infectious disease caused by coldwater rhabdovirus which is of clinical and economic importance in rainbow trout farming in Europe. VHS is notifiable in Ireland.

History

Early history of Egtved virus or VHS virus is conjectural, but it may have existed in Europe for eons. More recent evidence indicates that the virus is also widespread in wild marine fish and that this may be the original source is still debatable. There was an outbreak of VHS in a turbot farm in Cape Clear in Ireland in 1997 (also in turbot in Scotland in 1994). An outbreak occurred in England in a freshwater trout farm in 2006.

Clinical signs of disease

Typical outbreaks result in acute to chronic disease among fingerling rainbow trout at temperatures generally below 14°C. A wide range of possible disease signs are recorded including a profuse haemorrhaging, but in many fish a less dramatic pathology is noted. Fish may be lethargic and congregate at tank/ponds sides or outlets, have pale gills, dark body color, exophthalmos and in some cases intermittent periods of erratic spiraling swimming. Haemorrhage may be visible in the eyes and skin, within the muscle and internally in the viscera and intestine. In more chronic cases some of the above signs may be obvious with abdominal distension due to oedema in visceral organs and ascites (Figure 2).

Histopathology

Severe glomerular changes recorded resembling a membranous glomerulonephritis with focal necrosis and degeneration in the kidney. Liver sinusoids become engorged with blood with widespread necrosis.

Aetiology

Enveloped RNA virus belonging to the family *Rhabdoviridae* (Figure 3), genus *Novirhabdovirus*. There are four major genotypes (I to IV) and these appear more associated with geographic origin than fish species.

Diagnosis

Direct methods for detection using cell culture (epithelial papilloma of carp - EPC, bluegill fry - BF-2 and rainbow trout gonad - RTG-2), followed by immunological identification such as neutralization, immunofluorescence, ELISA. PCR and immunoperoxidase techniques also developed.

Host and geographic range

Rainbow trout in freshwater (and seawater) are the most susceptible group, however, other salmonids and non-salmonids e.g. turbot, may become infected in both fresh and seawater. Outbreaks have occurred throughout Europe, Japan, Taiwan and North America. A large outbreak is on-going in wild fish in the North American Great Lakes.

Source

VHSV has been isolated from Atlantic cod, haddock, rockling, sprat and herring in the Atlantic ocean and Baltic sea which appears indistinguishable from normal fresh water isolates. A genetically distinct strain of VHSV has also been isolated from Pacific cod, Pacific herring and Pacific salmon and appears to be of low pathogenicity.

Immunity

Antibody response mounted to VHSV and fish serology could be of importance for detecting the carrier state among fish stocks, but has yet to be validated. Vaccination is at an experimental stage.

Treatment and control

Control methods for VHS currently lie in official health surveillance schemes coupled with control policy measures, such as stamping-out procedures, and have resulted in eradication of the disease from several parts of Europe. Genetic approaches to selection of disease resistant stock and intergeneric hybridisation are also being pursued.

References

- Bruno, D. W. & Poppe, T. T. (1996) A colour atlas of salmonid diseases. Academic Press, London, UK.
- Dale, O., *et al.* (2009) Outbreak of viral haemorrhagic septicaemia (VHS) in seawater-farmed rainbow trout in Norway caused by VHS virus Genotype III. *Diseases of Aquatic Organisms*, 85, 93 – 103.
- Wolf, K. (1988) Fish viruses and fish viral diseases. Cornell University Press, Ithaca.



Figure 2. Juvenile turbot affected by VHS displaying abdominal swelling due to ascites.

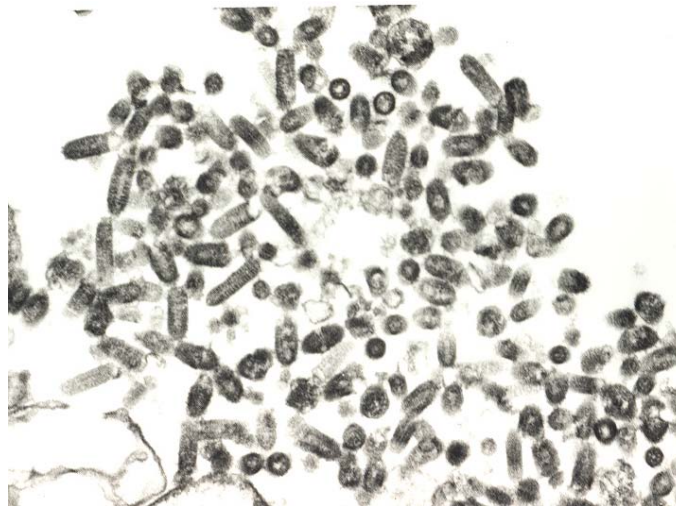


Figure 3. Transmission electronmicrograph of viral haemorrhagic septicaemia virus isolated from turbot in Scotland. Note the classical bullet shaped virions (70 – 180nm in size) (x 43,000).

Spring Viraemia of Carp (SVC)

Definition

Spring viraemia of carp (SVC) is an acute haemorrhagic and contagious viral infection, typically of cyprinids and more specifically of the common carp, in which the disease usually erupts in spring and causes mortality of the adults as well as the young. SVC is notifiable in Ireland.

History

Name first coined by Fijan *et al.* (1971) when virus isolated from carp affected by what had previously been known as acute infectious dropsy of carp. From early times in Europe, the most serious cause of carp mortality in pond culture has been attributed to infectious dropsy.

Clinical signs of disease

Viral outbreaks often complicated with secondary bacterial infections, however, signs attributed only to the virus are as follows: lethargy, distended abdomen, petechiation on gills and skin and around eyes, oedematous vent and trailing mucoid casts, exophthalmia and internally ascites with focal haemorrhages in swimbladder and other visceral organs.

Histopathology

Incomplete descriptions, but perivascularitis and oedema in blood vessels of the liver, and ultimately focal liver necrosis and areas of hyperaemia. Multifocal necrosis present in the pancreas.

Aetiology

Rhabdovirus named spring viraemia of carp virus (SVCV).

Diagnosis

Confirmatory diagnosis by isolation on cell culture (FHM, EPC or BF-2 cell lines) followed by immunological identification using virus neutralization, immunofluorescence or ELISA tests.

Host and geographic range

Overt infections recognized in common carp (*Cyprinus carpio*), grass carp (*Ctenopharyngodon idella*), silver carp (*Hypophthalmichthys molitrix*), bighead carp (*Aristichthys nobilis*) crucian carp (*Carassius carassius*), goldfish (*C. auratus*), tench (*Tinca tinca*) and sheatfish (*Siluris glanis*). Geographic range was restricted to countries of the European continent that experience low water temperatures during winter, however, there have been recent outbreaks in the USA and the UK.

Source

Clinically affected fish and covert virus carriers from either cultured, feral or wild fish. Virus shed via faeces, urine, sexual fluids and probably gill and skin epithelia.

Immunity

Carp can mount an immune response to SVC, but response influenced by temperature, route of immunization, quantity and strain of virus, and age and condition of host. Vaccine developed but not in commercial use.

Treatment and control

Control methods for SVC currently lie in official health surveillance schemes coupled with control policy measures, such as stamping-out procedures and have resulted in eradication of the disease from several parts of Europe.

References

Fijan, N. *et al.* (1971) Isolation of the viral causative agent from the acute form of infectious dropsy of carp. *Vet. Arch. Zagreb*, 41 125 – 138.

Wolf, K. (1988) *Fish viruses and fish viral diseases*. Cornell University Press, Ithaca.

Infectious Salmon Anaemia (ISA)

Definition

Infectious salmon anaemia (ISA) is an infectious disease of Atlantic salmon (*Salmo salar*) caused by an orthomyxovirus. ISA is notifiable in Ireland.

History

First described in 1984 in farmed salmon in Norway where it was named Infectious Lax Anaemia then Salmon Anaemia Syndrome and finally ISA. Outbreaks recorded in salmon farms in New Brunswick, Canada (1997), Scotland (1998), Faeroes (2000), USA (2001),

virus isolated but no outbreak in Ireland (2002) and most recently Chile (2008) and virus isolated again in Scotland in 2009.

Clinical signs of disease

Mortalities, pale gills, lethargy may be the only external signs of disease, but internally dark, almost black livers with petechiae in visceral organs (Figure 4). Haematocrits can be as low as 2 – 4%.

Histopathology

Multifocal haemorrhagic hepatic necroses may become confluent giving the changes a zonal appearance, leaving areas around large veins intact (Figure 4). Focal congestion and dilation of hepatic sinusoids and focal haemorrhage or congestion in the kidney parenchyma can also feature.

Aetiology

Orthomyxovirus (enveloped, ss RNA virus).

Diagnosis

Clinical signs, histopathology and cell culture (Salmon Head Kidney [SHK] or CHSE – 214 cell lines). Cell culture difficult and CHSE only supports some isolates. Monoclonal based tests and PCR also utilized.

Host and geographic range

Clinical disease only confirmed in Atlantic salmon, however, experimentally rainbow trout and sea trout shown to act as asymptomatic carriers. Virus also isolated from various wild fish (eels, sea trout) and detected by PCR in wild Atlantic salmon. ISA virus detected, in the absence of clinical disease, in farmed marine rainbow trout in Co. Mayo in 2002.

Source

Clinically affected fish, but original source may be wild salmonids.

Immunity

Antibody response to the virus and killed vaccine in use in Canada, Færoes and Chile. USDA issued notice in 1999 that autogenous vaccines could be prepared for ISAV in the event of an outbreak in US.

Treatment and control

Incidence of ISA greatly reduced in Norway through the use of regulatory measurements regarding the movement of fish, mandatory health control, slaughterhouse and transport regulations, as well as specific measures on affected farms.

References

Falk, K. *et al.* (1997) Characterization of infectious salmon anaemia virus, an orthomyxo-like virus isolated from Atlantic salmon (*Salmo salar*). *Journal of Virology*, 17, 9016 – 9023.



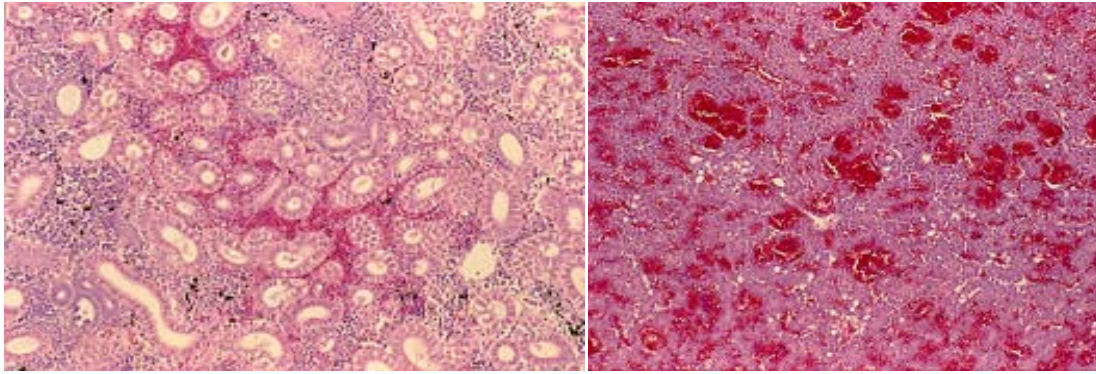


Figure 4. Atlantic salmon affected by ISA exhibiting dark red liver, pale gills and congested caecae (top left) and dark red liver with petechiae in the caecal fat (top right). Histopathology sections of ISA affected salmon showing multifocal haemorrhagic hepatic necrosis (bottom right) and focal congestion in the kidney parenchyma (bottom left) (H & E x 100).

Infectious haematopoietic necrosis (IHN)

Definition

Infectious haematopoietic necrosis (IHN) is an infectious disease, caused by a rhabdovirus, of salmonids. It is of concern due to its clinical and economic consequences in trout and salmon farming and its effects in wild stocks. IHN is notifiable in Ireland.

History

First reported in the early 1940s in North America (Pacific rim states) but later spread to central and eastern USA, Canada, Japan and southern Europe. Early name was sockeye salmon virus.

Clinical signs of disease

Natural outbreaks of IHN are rare above 15°C. Diseased fry are usually lethargic and hang at the areas of low water current. Whirling or flashing may also be seen. In older fish these signs may not be seen. Pale gills, dark skin, swollen abdomens, haemorrhages at the fin bases and opaque faecal pseudocasts trailing from the vent are frequently reported. Caecal fat petechiae and peritoneal haemorrhages may also be seen (Fig. 5). Subdermal haemorrhage between head and dorsal fin and in surviving sockeyes spinal deformities are quite common.

Histopathology

Multifocal degeneration and necrosis are usually seen in the spleen and interstitial tissue of the kidney. Necrosis of the eosinophilic granular layer in the digestive tract is considered pathognomic.

Aetiology

Enveloped RNA virus belonging to the family *Rhabdoviridae*.

Diagnosis

Direct methods for detection using cell culture (chinook salmon embryo – CHSE 214), followed by immunological identification such as neutralization, immunofluoresence, ELISA. PCR and immunoperoxidase techniques also developed.

Host and geographic range

Affects rainbow trout, sockeye, chinook, chum, Atlantic and coho salmon as well as amago and yamame. Historically disease in NW USA but now into southern Europe and Far East.

Source

Reservoirs of IHNV are clinically infected fish and covert carriers from either cultured, feral or wild fish. Transmission of IHNV is horizontal and possibly vertical or egg-associated.

Immunity

Strong antibody response in survivors mounted to IHNV. Vaccination is widespread in salmon farming in British Columbia, Canada using a nucleic acid based vaccine.

Treatment and control

Control methods for IHNV currently lie in official health surveillance schemes coupled with control policy measures. Thorough disinfection of eggs and incubation of eggs and rearing of fry and alevins in virus-free water supplies in premises completely separated from those harboring possible virus carriers and free from possible contact with fomites, are critical for preventing the occurrence of IHNV in a defined fish production site.

References

- Bruno, D. W. & Poppe, T. T. (1996) A colour atlas of salmonid diseases. Academic Press, London, UK.
- Wolf, K. (1988) Fish viruses and fish viral diseases. Cornell University Press, Ithaca.



Figure 5. Salmon affected by IHN virus exhibiting peritoneal and caecal fat haemorrhage, ventral congestion and pale gills (Photos courtesy of Dr. Diane Morrison and Mr. Brad Boyce).

Nodavirus

Definition

Piscine neuropathy nodavirus (PNN), otherwise known as viral encephalopathy and retinopathy (VER) or viral nervous necrosis (VNN), is a distinctive syndrome of vacuolating encephalopathy and retinopathy of many farmed marine fish species. The causal agent is a betanodavirus.

History

VNN was first described in Japanese parrotfish in 1990 and then appeared in many species in mariculture around the world, most notably in Japan and the Mediterranean, where hatcheries infected by the disease experienced 90 – 100% mortalities. The disease has recently been confirmed in various marine species (farmed and wild) in the UK.

Clinical signs of disease

Uncoordinated swimming, corkscrewing, whirling, dark coloration, darting across water surface and anorexia are all signs seen during an outbreak. Fish also appear blind. Internally the only abnormalities may be overinflation of the swim bladder and empty intestine.

Histopathology

Extensive vacuolation in nervous tissue including the brain, spinal cord and nervous layer of the retina (Figure 6). Malacia and gliosis are also observed and occasionally intracytoplasmic inclusions are observed.

Aetiology

Genus *Betanodavirus*, family *Nodaviridae* (non-enveloped, icosahedral, RNA virus, 25 – 30nm in diameter) (Figure 7).

Diagnosis

Cell culture (SSN – 1 [striped snakehead cell line]), immunohistochemistry and PCR all in use for confirmation, but clinical signs and histology will give presumptive diagnosis.

Host and geographic range

Numerous species of marine fish confirmed with natural outbreaks of the disease, including sea bass (*Lates* and *Dicentrarchus* sp.), turbot, halibut, grouper, striped jack, tiger puffer, Japanese flounder, etc. Disease confirmed in Japan, Norway, Denmark, Mediterranean (Greece, Malta, Turkey, Croatia), the UK and California. The disease has not been recorded in Ireland at the time of writing.

Source

Clinically affected fish and covert virus carriers from either cultured, feral or wild fish. Live feed (rotifers and artemia) were suspected as a source, but no natural isolation confirmed.

Immunity

Antibody response to the virus but no vaccine developed.

Treatment and control

Sterilization of water coming into hatcheries and disinfection of hatcheries after an outbreak. Screening of juveniles prior to purchase and avoidance of wild fish as broodstock. Broodstock screened by PCR in Japan (striped jack), however, not 100% reliable.

References

Frerichs, G. N., Rodger, H. D. & Peric, Z. (1996) Cell culture isolation of piscine neuropathy nodavirus from juvenile sea bass, *Dicentrarchus labrax*. *Journal of General Virology*, 77, 2067 – 2071.

O. I. E. (2003) *Manual of Diagnostic Tests for Aquatic Animal Diseases* (4th edition). O. I. E., Paris, France.

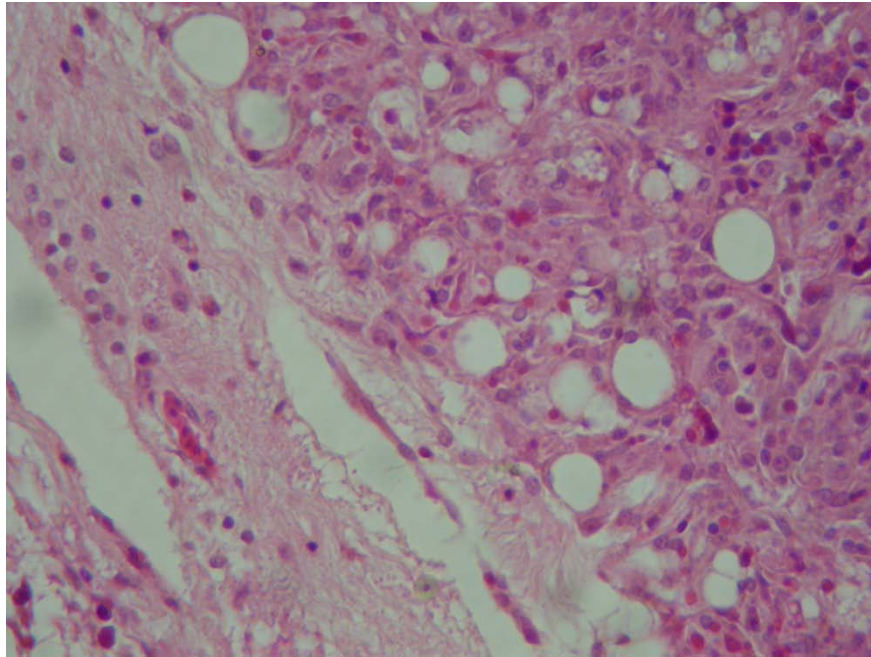


Figure 6. Histological section of brain of sea bass (*D. labrax*) affected by nodavirus and exhibiting vacuolation of the telencephalon and focal gliosis (H & E x 400).



Figure 7. TEM of sea bass brain showing nodavirus virions (arrows) (x 36,000).

Infectious Pancreatic Necrosis (IPN)

Definition

Infectious pancreatic necrosis is a highly contagious viral disease of young fish of salmonid species held under intensive rearing conditions. Susceptibility generally decreases with age, with resistance to the disease being reached at 1500 degree days except for Atlantic salmon smolts.

History

In early fish husbandry in North America, acute enteritis among lots of newly feeding hatchery trout fry was associated with a rise in mortality and although initially this catarrhal enteritis was thought associated with an intestinal protozoal agent or nutritional problems, it was finally demonstrated to be viral in origin in the 1950's. In 1957 Wolf and Dunbar made the first fish virus isolation – infectious pancreatic necrosis virus (IPNV).

Clinical signs of disease

First sign in salmonid fry is frequently a sudden and usually progressive increase in daily mortality, particularly in the faster growing individuals. Clinical signs include darkening pigmentation, a pronounced distended abdomen and a corkscrewing/spiral swimming motion. Cumulative mortalities may vary from less than 10% to more than 90% depending on a combination of various factors such as virus strain, host and environment. Internally the fish can display swollen intestine and catarrhal exudates in the lumen (Figure 8). There may also be petechiae on the caecal fat and a pale liver.

Histopathology

Focal necrosis of the acinar pancreatic tissue with necrotic areas replaced by a loose fibrous network and fat degeneration. Macrophages and leucocytes may infiltrate pancreatic and hepatic tissues. There may be necrosis and sloughing of the caecal endothelium.

Aetiology

Double stranded RNA virus of the family *Birnaviridae* (Figure 9).

Diagnosis

Histopathology and clinical signs can be diagnostic with confirmation conducted by cell culture (CHSE – 214 cell line) and/or PCR. Cell culture also useful for detection of subclinical carriers.

Host and geographic range

Initially described in brook trout but now confirmed in most salmonids throughout the world and further in many non-salmonids and shellfish. Several serotypes confirmed. The disease has been present in Ireland since the 1970s and has a global distribution.

Source

Infected farmed salmonids, but also in wild fish and shellfish.

Immunity

Good antibody response, although in young fish this is limited. Commercial vaccines available and utilized in Norway, Scotland and Chile. There has been some use of IPN vaccine in Ireland under an AR16 license.

Treatment and control

Prevention can be achieved by avoidance of fertilized eggs originating from IPN virus carrier broodstock and the use of protected water supply (e.g. spring or borehole). In outbreaks, a reduction in the stocking density may help reduce the overall mortality or alternatively a short period of increased water temperature (>18°C) also appears to be of benefit. Certain fast-growing strains of fish may also be more susceptible and IPN marine sites appear to experience the disease year after year.

References

- Smail, D. A. *et al.* (1992) Infectious pancreatic necrosis (IPN) virus Sp serotype in farmed Atlantic salmon, *Salmo salar* L., post-smolts associated with mortality and clinical disease. *Journal of Fish Diseases*, 15, 77 – 83.
- Wolf, K. & Dunbar, C. E. (1957) Cultivation of adult teleost tissues in vitro. *Proc. Soc. Exp. Biol. Med.*, 95, 455 – 458.



Figure 8. Salmon parr affected by IPN displaying a swollen intestine filled with mucus and catarrhal exudate.

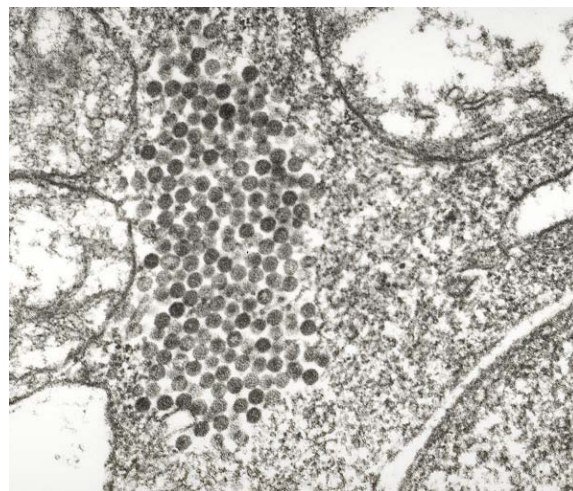


Figure 9. TEM of the birnavirus, IPNV (x 40,000)

Koi herpesvirus disease (KHV)

Definition

Koi herpes virus disease (KHV) is a herpesvirus infection capable of inducing a contagious and acute viraemia in common carp and varieties such as koi and ghost carp. KHV is notifiable in Ireland.

History

The first reports of the disease were in Israel and Germany in the late 1990s, since when the disease is now known to have spread globally, predominantly with the trade in koi.

Clinical signs of disease

All age groups of fish can be affected although younger fish are most susceptible and mortalities can be very significant. The skin appears pale or congested with a roughened appearance and gills can appear pale. Gills may have necrotic patches present. Eyes appear sunken (enophthalmia) and haemorrhages may be obvious on the skin and base of fins with fin erosion. Fish appear lethargic, lose body coordination and may also show signs of hyperactivity.

Histopathology

Inflammation and necrosis of gill tissue is a consistent feature, as is hyperplasia and hypertrophy of gill tissue. Branchial epithelial cells and leucocytes may have prominent nuclear swelling, margination of chromatin to give a signet ring appearance and pale diffuse eosinophilic intranuclear inclusions are commonly observed. Inflammation, necrosis and nuclear inclusions have also been observed in other visceral organs.

Aetiology

Koi herpes virus (KHV) in the family Herpesviridae. Also known as cyprinid herpesvirus 3 (CyHV-3).

Diagnosis

Histopathology and clinical signs can be diagnostic with confirmation via PCR, IFAT or cell culture.

Host and geographic range

Affects common carp and varieties such as koi, ghost carp and hybrids of these varieties. KHV present throughout Europe, including the UK, Asia and USA.

Source

Reservoirs are clinically affected fish and covert virus carriers among cultured, feral or wild fish.

Immunity

A vaccine is currently licensed in Israel and is widely used in carp farms there.

Treatment and control

Methods to control and prevent disease should mainly rely on avoiding exposure to the virus coupled with good hygiene and biosecurity. Disinfection of eggs can be achieved with iodophor disinfectants (200mg/l for 30 seconds at 15°C).

References

O. I. E. (2009) Manual of Diagnostic Tests for Aquatic Animals. 6th Edition, O. I. E., Paris, France.

Epizootic haematopoietic necrosis (EHN)

Definition

Epizootic haematopoietic necrosis is considered to be a systemic clinical or subclinical infection of finfish with the EHN virus. EHN is notifiable in Ireland.

History

EHN virus first recognized in Australia in 1986 in perch and transmitted to farmed rainbow trout.

Clinical signs of disease

The disease causes low level mortalities in rainbow trout but high mortality rate in perch. Moribund fish may exhibit loss of equilibrium, may have flared opercula and be dark in colour. Lesions may be present on skin, fins and gills.

Histopathology

Focal, multifocal or locally extensive coagulative or liquefactive necrosis of the liver, kidney and spleen are commonly seen. A small number of basophilic intracytoplasmic inclusion

bodies may be seen (Figure 10), particularly in areas immediately surrounding necrotic areas in the liver and kidney. Necrotic lesions may also be seen in heart, pancreas, gastrointestinal tract, gill and pseudobranch.

Aetiology

EHNV is a member of the genus *Ranavirus* in the family *Iridoviridae* with the type species Frog virus 3 (FV3). Ranaviruses have been isolated from healthy and diseased frogs, salamanders and reptiles in America, Europe and Australia. Ranaviruses are large (150 – 180nm), icosahedral, ds DNA viruses. EHNV can be distinguished by genomic analysis from the similar European catfish virus (ECV) and FV3.

Diagnosis

Suspect cases where finfish present with typical histopathology and confirmation in such fish where EHNV has been demonstrated by CPE in cell culture (BF-2), ELISA or PCR.

Host and geographic range

Natural EHNV infections have only been reported in perch and rainbow trout, however, other fish species have been shown to be susceptible experimentally. EHNV has only been reported from Australia.

Source

The virus is very resistant and may be transferred by fomites and vectors and is presumed to persist in farms and water for prolonged periods but there are no known wild aquatic reservoirs.

Immunity

No vaccine available.

Treatment and control

Disease control in rainbow trout at the farm level relies on reducing the impact of infection by maintaining low stocking densities and adequate water quality.

References

- O. I. E. (2009) Manual of Diagnostic Tests for Aquatic Animals. 6th Edition, O. I. E., Paris, France.
- Whittington, R. J. *et al.* (2010) Iridovirus infections in finfish – critical review with emphasis on ranaviruses. *Journal of Fish Diseases*, 33, 95 – 122.

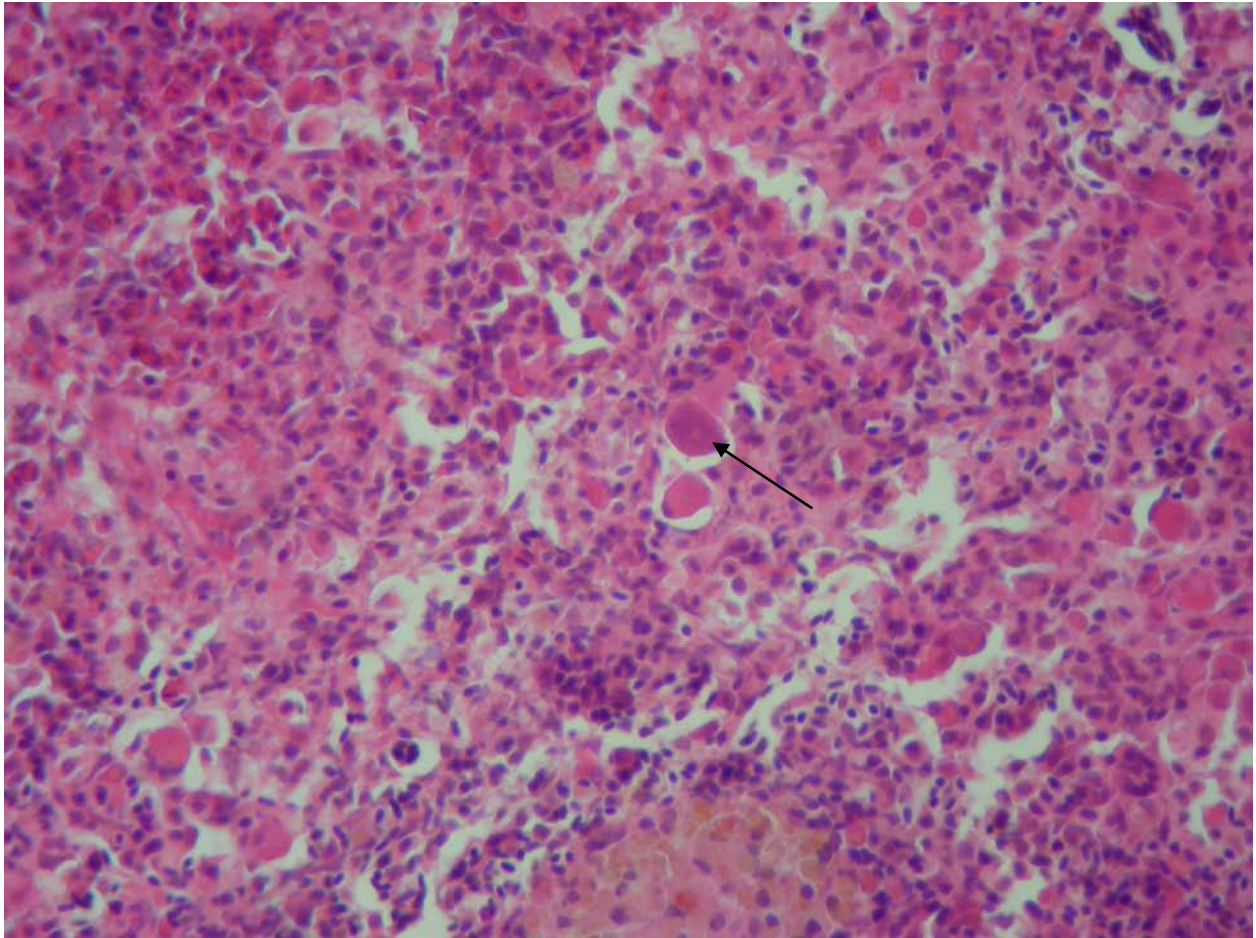


Figure 10. Histopathology section of angelfish spleen infected with iridovirus showing some cells with basophilic intracytoplasmic inclusions (arrow) (H & E x400).

DISEASES CONSIDERED TO BE VIRAL IN ORIGIN

Cardiomyopathy syndrome (CMS)

Cardiomyopathy syndrome (CMS) is a condition of grower stage Atlantic salmon which was first described in Norway in 1985 and subsequently in the Faroes and Scotland. The cause is considered to be infectious, probably viral, and mortalities due to the condition can be significant, especially in terms of biomass loss. Clinical signs and pathology observed are pop-eye, skin oedema, congestion of skin and internally ascites, enlarged hearts and congested livers (Figures 11). Histopathology reveals extensive myodegeneration of the cardiac muscle fibres with inflammation. Control is via high levels of biosecurity and accelerated harvest and fallowing in an affected site. This disease has not been diagnosed in Ireland.

References

Ferguson, H. W. *et al.* (1990) Cardiomyopathy in farmed Norwegian salmon. *Diseases of Aquatic Organisms*, 8, 225 – 231.



Figure 11. Salmon affected by CMS exhibiting skin congestion and oedema (top left), exophthalmia (top right) and internally bloody ascites (bottom).

Heart and skeletal muscle inflammation (HSMI)

Heart and skeletal muscle inflammation (HSMI) was recorded for the first time in 1999 in Western Norway in farmed marine stage Atlantic salmon. The disease outbreaks normally start 5 to 9 months after transfer to sea water. Affected fish have been reported to be lethargic, pointing into the current and mortalities occur (<10%). Internally ascites, a pale and soft heart and sometimes a white layer of fibrin on the liver. Histologically there is necrosis of the heart muscle fibres and a massive inflammatory response (myocarditis and epicarditis). Red muscle inflammation follows the same pattern but is not a consistent finding. The disease appears infectious and a reo-like virus is the subject of investigation. The disease has only been confirmed in Norway although there is one report of a suspect case in Scotland. This disease has not been diagnosed in Ireland.

References

Kongtorp, R. T. *et al.* (2004) Heart and skeletal muscle inflammation in Atlantic salmon, *Salmo salar* L.: a new infectious disease. *Journal of Fish Diseases*, 27, 351 – 358.

BACTERIAL DISEASES

Mycobacteriosis

Definition

Mycobacteriosis in fish is a chronic progressive disease caused by certain bacterial species within the genus *Mycobacterium*. The species in fish are non-tuberculous mycobacteria (NTM) and do not cause major disease in healthy humans.

History

First described in carp in 1897 but has been recorded in over 167 species of marine, freshwater, wild and farmed fish. There have been reports of outbreaks in farmed salmon in British Columbia, Canada and Scotland, as well as in farmed trout and tench in Italy.

Clinical signs of disease

Emaciation, skin inflammation, pop-eye, skin lesions or ulceration can all be observed. Internally grey-white nodules may be obvious in liver, kidney, spleen, heart and muscles. Ascites and peritonitis may also be observed. Low level mortalities may also occur.

Histopathology

Granulomas in visceral organs with acid-fast micro-organisms present (Ziehl-Nielsen positive) in centre of suspect lesions (Figure 12). Granulomas have central necrotic areas.

Aetiology

Mycobacterium marinum, *M. fortuitum*, *M. salmoniphilum* and *M. chelonae* are all considered pathogenic for fish. All are aerobic, acid-fast, Gram positive and non-spore forming.

Diagnosis

Clinical signs, histopathology (ZN), growth of organisms on Löwenstein-Jensen or Middlebrook 7H10 media but this can take 2 – 3 months.

Host and geographic range

Global distribution and many different fish species. The disease is commonly observed in ornamental fish.

Source

Many of these bacteria occur naturally in the aquatic environment. Contaminated feed (based on uncooked trash fish) is a common source.

Treatment and control

There is no fully effective treatment therefore the best course is to cull and disinfect premises. 10,000ppm chlorine or 60 – 85% alcohol do kill mycobacteria.

Zoonotic potential

Transmission from infected fish to humans is rare but “fish handler’s disease” or “fish tank granuloma” is a condition that can occur in humans as a result of the skin infection from these bacteria. Gloves should be worn when handling suspect or infected fish or when cleaning contaminated surfaces.

References

- Bruno, D. W., *et al.* (1998) Pathology attributed to *Mycobacterium chelonae* infection among farmed salmon and laboratory infected Atlantic salmon *Salmo salar*. *Diseases of Aquatic Organisms*, 33, 101 – 109.
- Jacobs, J. M., *et al.* (2009) A review of mycobacteriosis in marine fish. *Journal of Fish Diseases*, 32, 119 – 130.

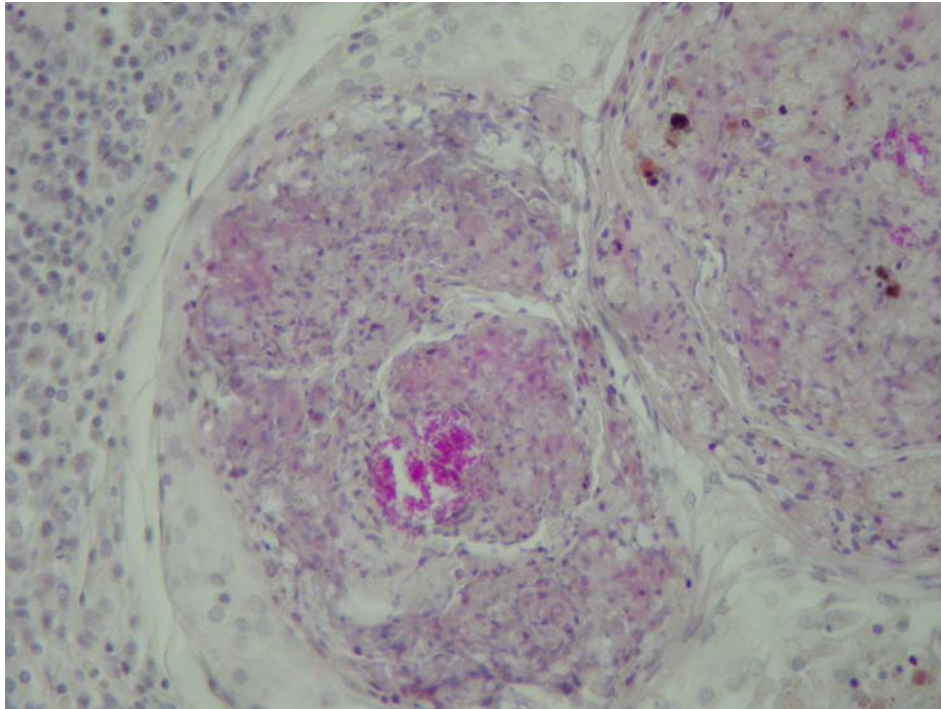


Figure 12. Histopathological section of angelfish spleen affected by mycobacteriosis showing acid fast bacteria in the centre of the granuloma (ZN x 400).

Coldwater disease & Rainbow Trout Fry Syndrome (RTFS)

Definition

Bacterial coldwater disease (CWD) is a serious septicaemic infection of hatchery reared salmonids, especially young coho salmon, which has also been referred to as peduncle disease and is prevalent in northwest American hatcheries during colder months of the year. Rainbow trout fry syndrome (RTFS) or rainbow trout fry anaemia is a freshwater systemic disease affecting trout (and to a lesser extent salmon) in Europe, that results in high mortalities. The same bacterium (*Flavobacterium psychrophilum*) is the causal agent of these diseases in both continents.

History

CWD described in US since mid 1940s in rainbow trout, however, juvenile coho salmon appear most susceptible. RTFS described throughout Europe in the 1990s where due to its level of impact and persistent nature has risen to become the most important disease problem for freshwater rainbow trout farming in Europe.

Clinical signs of disease

CWD – haemorrhage at the base of fins, pale gills, haemorrhagic ulceration in muscle and tail rot. Disease usually in spring with water temperatures 4 - 10°C. If alevins affected by yolk-sac erosions mortalities can be 30 – 50%. Coagulated yolk-sac may precede the disease.

RTFS – high mortalities in trout fry, pale gills, swollen spleens with blood tinged caecal fat around spleen, lethargy, darkened skin, ascites, exophthalmos. Skin ulcerations or eroded/dissolving jaw in older fish (Figure 13).

Histopathology

CWD - necrosis and bacterial presence in visceral organs. Where skin ulceration present, oedema and localized inflammation can be observed, but not well described.

RTFS – epidermal hyperplasia, characteristic proliferation of the splenic capsule and inflammation around spleen, necrosis and depletion of red pulp in spleen.

Aetiology

Flexibacter psychrophilus = *Cytophaga psychrophila* = *Flavobacterium psychrophilum* are all terms that have been used for the causal agents of these diseases. Most recent classification work indicates that *Flavobacterium psychrophilum* is the correct name for these bacteria. These bacteria are Gram-negative, filamentous and require extended growth (14 days) on Anaker and Ordal's media (or equivalent low nutrient agar) at 15°C.

Diagnosis

Clinical observations, fresh microscopy and histopathology with biochemical or serological characterization of the isolated bacteria

Host and geographic range

RTFS is widespread in Europe affecting primarily farmed rainbow trout and increasingly Atlantic salmon. CWD was reported in the USA in Pacific salmon until the 1980s and now appears widespread in rainbow trout in Europe, Australia, Japan and Chile. RTFS affects farmed trout in Ireland.

Source

Natural reservoirs are uncertain. The disease can be transmitted vertically and horizontally and the bacterium is very robust, resisting some disinfectants which are normally used for egg cleaning (iodophors).

Immunity

Protection has been demonstrated using a bacterin administered by injection or immersion. Autogenous vaccines are in use in some farm sites in the UK.

Treatment and control

Broad spectrum antibiotics have been partially ineffective in controlling an outbreak, but improving the environment and using 3 – 4 times recommended doses of antibiotics have shown benefits. Florfenicol also appears effective at recommended dose regimes. Autogenous vaccines are in use in the UK.

References

- Holt, R. A., Rohovec, J. S. & Fryer, J. L. (1993) Bacterial cold-water disease. In: Bacterial Diseases of Fish (Ed. by Inglis, V. *et al*), Blackwell, Oxford, London, pp3 – 22.
- Nematollahi, A. *et al.* (2003) *Flavobacterium psychrophilum* infections in salmonid fish. Journal of Fish Diseases, 26, 563 – 574.
- Rangdale, R. E., Richards, R. H. & Alderman, D. J. (1999) Histopathological and electron microscopical observations on rainbow trout fry syndrome. Veterinary Record. 144, 251 - 4.



Figure 13. Rainbow trout affected by *Flavobacterium psychrophilum* infection displaying the swollen spleen typical in RTFS (left) and dissolving mandibles which occurs in some cases (right).

Bacterial Kidney Disease (BKD)

Definition

A serious disease of fresh and seawater, farmed and wild salmonids, that results in an acute to chronic systemic granulomatous disease. This disease is notifiable in Ireland.

History

First described in early 1930's in wild salmon in Scotland. Then observed in three species of farmed trout in Massachusetts and California. Described in cutthroat trout in Canada in 1937. There is no history of this disease in Ireland.

Clinical signs of disease

Those fish severely affected by the disease may show no obvious external signs or may show one or more of the following: pale gills, exophthalmia, abdominal distension (due to ascites), skin blisters (filled with clear or turbid fluid), shallow ulcers (the results of broken skin blisters), haemorrhages (particularly around the vent) and more rarely, cavitations in the musculature, filled with blood tinged caseous or necrotic material. Internally, there may be fluid in the abdomen, varying haemorrhage on the abdominal walls and viscera, a membranous layer on one or more of the visceral organs, and most characteristically, creamy-white granulomatous lesions in the kidney and less frequently in the liver, spleen and heart (Figure 14). Pacific salmon seem more susceptible to BKD than Atlantics and the granulomas are well encapsulated in Atlantics but less so in Pacifics.

Histopathology

Lesion is chronic granulomatosis, principally of haematopoietic tissue, but extends to liver, cardiac and skeletal muscle or indeed any organ. The granuloma is often large, with a central caseous zone bounded by epithelioid cells and infiltrating lymphoid cells. Presence of a capsule is variable and lack of encapsulation is often associated with more aggressive infections.

Aetiology

Renibacterium salmoninarum is a small, Gram-positive diplococcus that grows best at 15 - 18°C and not at all at 25°C. Has requirement for cysteine and serum or serum substitutes in bacteriological media.

Diagnosis

Clinical signs, Grams smear, ELISA, FAT, IFAT, histopathology, isolation (2 –3 weeks at 15°C) in cysteine-enriched media such as kidney disease medium (KDM2), or selective kidney disease medium (SKDM) with agglutination tests and PCR.

Host and geographic range

Salmonids are clinically susceptible, especially the genus *Oncorhynchus* (Pacific salmon and rainbow trout). Disease reported in North America, Japan, Western Europe and Chile.

Source

Probably salmonid fish.

Immunity

No vaccines commercially available, but are greatly needed. There is evidence that under some conditions the pathogen elicits an immune response in fish, and there are some reports of vaccination. The protective ability of the vaccine is questionable, however, and one of the problems is the intracellular nature and vertical transmission of the agent.

Treatment and control

Chemotherapy (erythromycin) provides limited and only temporary relief. The bacteria can survive and multiply within phagocytic cells. Screening of farmed broodstock and regular testing of growing stock for agent combined with disinfection and movement controls have proven effective in Europe.

References

- Evelyn, T. P. T. (1993) Bacterial kidney disease – BKD. In: Bacterial Diseases of Fish (Edited by V. Inglis, R. J. Roberts & N. R. Bromage), Blackwell Scientific Publications, Oxford, UK, p177 – 195.
- Fryer, J. L. & Saunders, J. E. (1981) Bacterial kidney disease of salmonid fish. Annual Review of Microbiology, 35, 273 – 298.

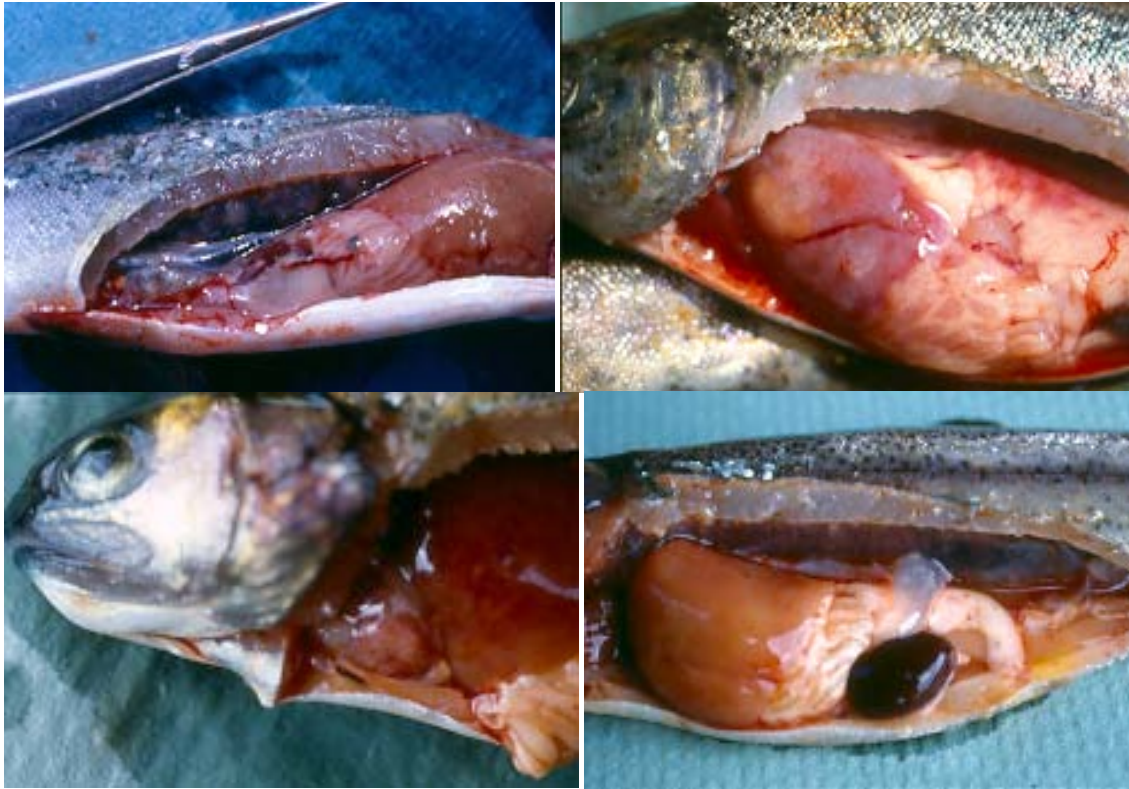


Figure 14. Rainbow trout affected by BKD exhibiting kidney and liver granulomas (top left), diphtheritic membrane on liver (top right), heart granulomas (bottom left) and swollen spleen, ascites and petechiae in visceral fat (bottom right).

Enteric Redmouth Disease (ERM)

Definition

Bacterial septicemic condition of farmed salmonids, in particular rainbow trout. Recent reports in channel catfish.

History

First associated with losses in rainbow trout in Hagerman valley, Idaho in the 1950s. Killed cell vaccine was in use before the organism was assigned a formal name in 1978. The disease is widespread in European trout farms, including Ireland.

Clinical signs of disease

Gross external signs first described were lethargy, skin darkening and congestion around the mouth and operculum, and at the base of the fins. Other signs seen include exophthalmos, ulceration and cutaneous petechiae. Internally the fish show signs of haemorrhagic septicemia with congestion and petechiae throughout the peritoneum and visceral organs, in particular the caecal fat. Splenomegaly and fluid filled stomach and intestine are also observed (Figure 15).

Histopathology

Necrosis of the haematopoietic tissues in kidney and spleen following bacterial invasion are most commonly seen and bacteria may be observed in any organ throughout the body. With bacterial spread to the gills, musculature and liver there is capillary dilation and haemorrhage, tissue oedema and further necrosis.

Aetiology

Yersinia ruckeri is the causal agent and the Gram-negative, motile rod-shaped bacterium is catalase positive and oxidase negative. Several serotypes have been identified.

Diagnosis

Gross and histological signs are helpful but confirmation requires isolation on general nutrient agar (24 hours at 22°C) such as TSA or BHI. FAT and ELISA tests have also been used but isolation usually necessary for antibiotic sensitivity.

Host and geographic range

Principally a problem for young farmed rainbow trout, however, all salmonids susceptible and outbreaks confirmed in North America, Europe, South Africa, Chile and New Zealand.

Source

Feral fish, imported baitfish or even ornamentals have been suggested as sources of ERM. *Y. ruckeri* also isolated from bird faeces and sea gulls.

Immunity

A bacterin produced using *Y. ruckeri* was the first commercial fish vaccine, and a considerable amount of work in vaccine development and field testing has involved oral, injection and immersion products against ERM. Authorised vaccines in use in US and Europe. Protection with dip or immersion vaccines only protects for up to six months.

Treatment and control

Broad spectrum antibiotics effective in controlling an outbreak, but increasing antibiotic resistance is observed and sensitivity should be tested.

Reference

Stevenson, B., Flett, D. & Raymond, B. T. (1993) Enteric Redmouth (ERM) and other enterobacterial infections of fish. In: Bacterial Diseases of Fish (Edited by V. Inglis, R. J. Roberts & N. R. Bromage), Blackwell Scientific Publications, Oxford, UK, p80 - 106.

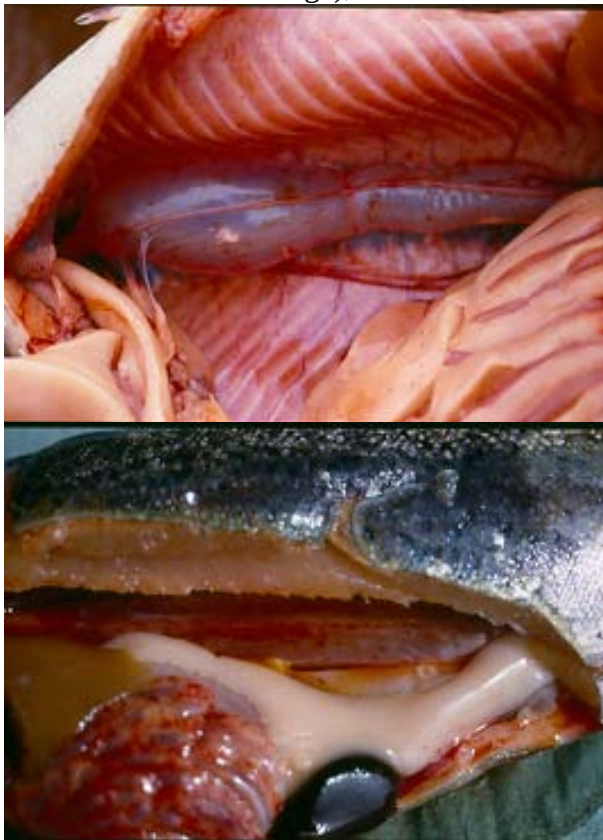


Figure 15. Rainbow trout affected by ERM presenting with petechiae on the swim bladder (left) and in the pyloric caecae (right) with a swollen spleen.

Furunculosis

Definition

Furunculosis is a fatal epizootic disease, primarily of salmonids, caused by the bacterium *Aeromonas salmonicida*. This organism can also cause clinical disease in other fish species where it is named ulcer disease or carp erythrodermatitis.

History

First authentic report in a trout hatchery in Germany in 1894, and known for over 100 years as a disease in wild salmon in freshwater (Furunculosis Committee 1930, 1933, 1935 in UK). With the growth of salmon farming, particularly in Scotland and Norway, its effects have been described in the marine environment too. The disease is endemic in some water bodies in Ireland.

Clinical signs of disease

In a population affected by typical furunculosis there will be examples of both chronic and acute forms of the disease. High mortalities, without external signs of infection are often associated with acute furunculosis, although anorexia may be present. Other fish may appear dark in colour, lethargic with reddening at the fin bases or head (Figure 17). Internally there may be widespread petechiae in the viscera and a swollen spleen. In chronic furunculosis, usually seen in older fish, there may be similar clinical signs to a subacute form but with attempts at repair in the tissues. Liquefactive, haemorrhagic lesions may be present in the musculature with bloody discharge from the vent and splenomegaly also present (Figure 16). Atypical furunculosis may cause lower level mortalities and small skin ulcers with a dark, pigmented periphery.

Histopathology

Focal localization of bacteria in the dermis, gills, kidney, spleen, heart, liver or most visceral organs with little host response in some foci may be the most common finding, but associated haemorrhaging, oedema, macrophage infiltration and liquefactive necrosis may also be seen with some bacterial foci.

Aetiology

Aeromonas salmonicida is a Gram-negative, non-motile short rod and most strains produce a brown diffusible pigment on agar containing tryptone. Grows best at 22°C or less. Atypical furunculosis is caused by a slower growing non-pigmenting isolate *A. salmonicida achromogenes*.

Diagnosis

Gross and histological signs (H & E) are helpful but confirmation requires isolation on general nutrient agar (24 - 48 hours at 22°C) such as TSA or BHI. Isolation vital for antibiotic sensitivity.

Host and geographic range

Salmonids principally affected in Europe and North America in both fresh and seawater. Cyprinids (carps) and ornamentals also affected in Europe and US where the disease manifests as skin ulceration.

Source

Salmonids (wild and farmed) can carry the organism and when these fish are stressed, such as with high water temperatures or low oxygen levels, then clinical disease can break.

Immunity

Pioneering work started in 1940s on vaccines has now resulted in effective injectable oil-based vaccines, which are widely used by the salmon farming industry. Presmolts usually injected 6 – 10 weeks prior to transfer to sea and these vaccines provide protection for up to 12 months.

Treatment and control

Broad spectrum antibiotics effective in controlling an outbreak, but increasing antibiotic resistance is observed and sensitivity should be tested.

Reference

Bernoth, E-M., Ellis, A. E., Midtlyng, P. J., Olivier, G. & Smith, P. (1997) Furunculosis; multidisciplinary fish disease research. Academic Press, London, UK.



Figure 16. Salmon parr affected by atypical furunculosis presenting with small skin ulcers (left) and post-smolts affected by typical furunculosis with dermal haemorrhage and liquefactive haemorrhagic lesions in the muscle (right).



Figure 17. Farmed turbot affected with furunculosis exhibiting haemorrhage and congestion around the mouth.

Piscirickettsiosis

Definition

Piscirickettsiosis is a disease of salmonids caused by *Piscirickettsia salmonis* and is a significant disease problem in farmed marine salmonids.

History

First reported in coho salmon in Chile in 1989, and still the main disease problem there. Now also confirmed in northern hemisphere salmon farming countries (Norway, Canada, Scotland and Ireland).

Clinical signs of disease

Skin lesions, dark skin, lethargy, anorexia, nervous signs in some cases, and internally petechiae, peritonitis, ascites, white nodules in liver and kidney (Figure 18).

Histopathology

Extensive necrosis of the haematopoietic tissues particularly the kidney with oedema, some fibrosis and influx of inflammatory cells. Haemorrhage in visceral organs, musculature and intestinal tract. Meningitis also reported in various fish species. *Rickettsia* may be observed within membrane bound vacuoles using H & E or Giemsa in visceral organs or white blood cells.

Aetiology

Piscirickettsia salmonis is a Gram-negative, acid-fast, non-motile, spherical to coccoid, non-capsulated (although often pleomorphic) organism.

Diagnosis

Confirmation via immunohistochemistry, isolation in cell culture (CHSE-214) without antibiotics. PCR also developed.

Host and geographic range

Salmonids, particularly Pacific salmon vulnerable. Chile has the most serious problem with this disease and in northern hemisphere countries the mortalities associated with the pathogen are usually of a low level.

Source

Wild fish, shellfish, crustaceans all reported to harbor rickettsia but the true source not established. Vectors (lice, isopods) may also be involved.

Immunity

Vaccines are used in Chile.

Treatment and control

Broad spectrum antibiotic therapy used, although some resistance developing. Outbreaks usually associated with stressful event, such as algal bloom, sudden change in environment or grading.

References

Fryer, J. L. et al. (1990) Isolation of a rickettsiales-like organism from diseased coho salmon (*Oncorhynchus kisutch*) in Chile. *Fish Pathology*, 25, 107 – 114.

Rodger, H. D. & Drinan, E. M. (1993) Observation of a rickettsia-like organism in Atlantic salmon (*Salmo salar* L.) in Ireland. *Journal of Fish Diseases*, 16, 361-369.

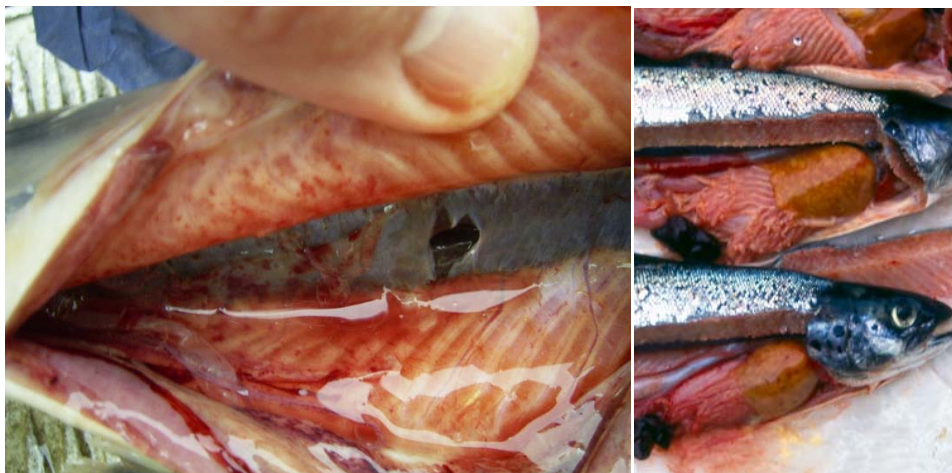


Figure 18. Atlantic salmon affected by piscirickettsiosis exhibiting haemorrhage on the swim bladder and peritoneum (left), swollen spleens, congested caecae and mottled livers (right).

Bacterial Gill Disease

Definition

Bacterial gill disease is an important disease in farmed freshwater salmonids. The bacterium *Flavobacterium branchiophila* causes a chronic, proliferative response in gill tissue.

History

Since the first report in an experimental salmonid hatchery in 1922, most reports of this disease have been in intensively reared salmonids.

Clinical signs of disease

Gill is the only target organ and clinical signs include lethargy, dyspnea, coughing and flared opercula. Strands of mucus may trail from the gills and gill themselves may exhibit pale and/or swollen areas (Fig. 19).

Histopathology

BGD is primarily an epithelial disease with bacteria colonizing the tips of the secondary lamellae, spreading inwards and then resulting in a proliferative bronchitis that causes epithelial hyperplasia. Fusion of the secondary lamellae may occur distally enclosing the bacteria, sloughed epithelial cells and mucus, or may obliterate any lamellar space. Fusion of the primary lamellae will also occur in severe long-standing cases.

Aetiology

Flavobacterium branchiophila, is a Gram-negative, long, thin, filamentous rod.

Diagnosis

Wet gill smears or histopathology. Isolation on Cytophaga Agar (Anacker & Ordal) at 18°C.

Host and geographic range

Global distribution and most reports are in farmed salmonids, although most teleosts are probably susceptible.

Source

Probably wild fish and aquatic ecosystems where it may be present naturally.

Immunity

Vaccination not considered viable at present, although agglutinating antibodies to the organism have been recorded in older fish.

Treatment and control

BGD usually responds well to antiseptic and surfactant baths such as chloramine T and benzalkonium chloride. Providing adequate oxygen is useful supportive therapy.

References

- Brocklebank, J. R. (1999) Atypical bacterial gill disease in Atlantic salmon hatcheries in British Columbia. *Canadian Veterinary Journal*, 40, 54.
- Ostland, V. E., Lumsden, J. S., MacPhee, D. D. & Ferguson, H. W. (1994) Characteristics of *Flavobacterium branchiophila*, the cause of salmonid bacterial gill disease in Ontario. *Journal of Aquatic Animal Health*, 6, 13 - 26



Figure 19. Salmon fry affected by bacterial gill disease and presenting with mottled and swollen gills.

Vibriosis

Definition

Vibriosis is the term most commonly used to describe infections associated with *Vibrio anguillarum*, but *V. ordalii* and other *Vibrio* sp. may cause similar clinical signs in wild and farmed fish in many parts of the world.

History

Vibrio anguillarum was the first *Vibrio* isolated from fish, these being from eels in the Mediterranean. It is now known that many different *Vibrio* species in different parts of the world, and different species can cause significant and similar disease problems. In mariculture, *V. anguillarum*, *V. ordalii* and *V. salmonicida* have proven to be the most seriously pathogenic.

Clinical signs of disease

Acutely affected fish are usually anorexic with pale gills and occasional periorbital oedema, and these signs correspond with rapidly rising mortalities. The fish may appear dark in color and dermal or subdermal skin lesions (Figure 20) may ulcerate and release haemorrhagic fluid which contains numerous bacteria. Internally ascites with petechiae in the musculature and viscera are common.

Histopathology

Multifocal necrosis and haemorrhage in the visceral organs, most notably the liver is commonly observed as is necrosis and haemorrhage in the skeletal musculature. Bacterial foci may or may not be present. Meningitis also observed with *Vibrio* sp. infection.

Aetiology

The genus *Vibrio* consists of Gram-negative, straight or slightly curved rods which are motile. Colony morphology, biochemical tests and the use of diagnostic keys will confirm the family *Vibrionaceae*. The organisms can be isolated on general nutrient agar plus sodium chloride (1.5%).

Diagnosis

Clinical observations and biochemical or serological characterization of the isolated bacteria (48 hours at 20°C for *V. anguillarum*, or 15°C or less for *V. salmonicida*)

Host and geographic range

Most marine species susceptible to *Vibrio* sp., wild fish carry organisms and outbreaks in farmed fish closely related to rearing environment conditions. Vibriosis is reported globally.

Source

Vibrio sp. ubiquitous in marine and brackish water but more frequent where static water and soft benthos occur in combination with a high organic load. Their frequency is greatly reduced in areas where rocky shores or high energy beaches combine with well-aerated waters.

Immunity

Killed vaccines available for *V. anguillarum*, *V. salmonicida*, *V. ordalii* and *V. viscosus* (causal agent of winter ulcer in salmon farms in Northern Europe also known as *Moritella viscosus*). The injectable, oil-based vaccines are most widely used and have been demonstrated to be effective. Also effective in cod and eels.

Treatment and control

Broad spectrum antibiotics effective in controlling an outbreak, but increasing antibiotic resistance is observed and sensitivity should be tested. Vaccines widely used, including autogenous vaccines for farmed cod.

Reference

Hjeltnes, B. & Roberts, R. J. (1993) Vibriosis. In: Bacterial Diseases of Fish (Ed. By Inglis, V. *et al*), Blackwell, Oxford, London, pp109 – 121.



Figure 20. Farmed cod affected by *V. anguillarum* O2ß presenting with with dermal ulceration (left) and farmed summer flounder affected by vibriosis presenting with abdominal distension due to ascites (right).

Epitheliocystis

Definition

Epitheliocystis is a chronic and unique infection by a chlamydial organism that results in hypertrophied epithelial cells – typically of gills but sometimes also skin – or certain freshwater and marine fishes.

History

Described in Germany by Marianne Plehn (1922) in carp and in US in bluegills, white perch and striped bass in the 1960s. Now recorded globally and in over 11 families of fishes (fresh water and marine).

Clinical signs of disease

Lightly infected fish behave normally, whereas heavy infections can be dramatically obvious with small white cysts present (Figure 21). Terminally ill fish are lethargic, gill covers flared and respiration rate rapid.

Histopathology

Young branchial cysts are small and contain a central inclusion surrounded by foamy cytoplasm and a thickened host epithelium. Mature cells are surrounded by layers of normal epithelium and remnants of the host cell and numerous basophilic uniformly sized chlamydia.

Aetiology

Based on electronmicroscopy the epitheliocystis organism is currently placed within the *Chlamydiales* as a species of Chlamydia. Some studies suggest that there are different forms, but those within the salmonid family have a similar morphological appearance, although

more recent evidence of different species in freshwater and seawater is emerging for Ireland and Norway.

Diagnosis

Wet mounts, histology and PCR. Transmission electronmicroscopy needed for definitive diagnosis.

Host and geographic range

Global distribution in 11 families of fishes in freshwater and marine.

Source

Exclusively a pathogen of fish.

Immunity

No available information.

Treatment and control

Broad spectrum antibiotics have been used with some degree of success, avoidance of infected fish should be adhered to at all cost.

Reference

Nylund, A. *et al.* (1998) A morphological study of the epitheliocystic agent in farmed Atlantic salmon. *Journal of Aquatic Animal Health*, 10, 43 – 55.

Wolf, K. (1988) Epitheliocystis. In: *Fish Viruses and Fish Viral Diseases*, Cornell University Press, Ithaca, NY.



Figure 21. Atlantic salmon gills affected by epitheliocystis showing numerous small white-grey spots on the gills.

Tenacibaculosis

Infection of marine fish by *Tenacibaculum maritimum* is common in farmed fish or many species. The bacteria appears opportunistic, commonly infecting fish after minor epidermal or epithelial trauma or irritation, and they can rapidly colonise such tissue. These bacteria are toxigenic (previously known as *Flexibacter maritimus*), are Gram negative, slender bacilli which multiply in mats on the damaged tissue. A yellowish colouration on lesions or damaged areas is characteristic of infection with this bacteria (Figure 22). Mouth rot, tail rot, skin and fin lesions as well as gill, gill raker and intestinal colonization are often seen. Oral treatment with broad spectrum antibiotics is generally successful if fish are maintained in a low stress environment. No commercial vaccines are available although experimental inocula have been utilized.



Figure 22. Salmon affected by tenacibaculosis in fins (left) and gill rakers (right, arrow) showing yellow pigmentation of affected tissues.

Streptococcosis

Streptococcosis in fish due to β -haemolytic strains causes clinical disease in trout, tilapia as well as some marine species e.g. mullet. *Streptococcus iniae* is Gram positive (Figure 23) facultative anaerobe which occurs in long chains of cocci. Affected fish have exophthalmia, petechiae and congestion at fin bases. Histologically lesions are principally intravascular leading to meningitis, peritonitis and pericarditis. The disease has become particularly significant in recirculation units and has not been recorded in Ireland.

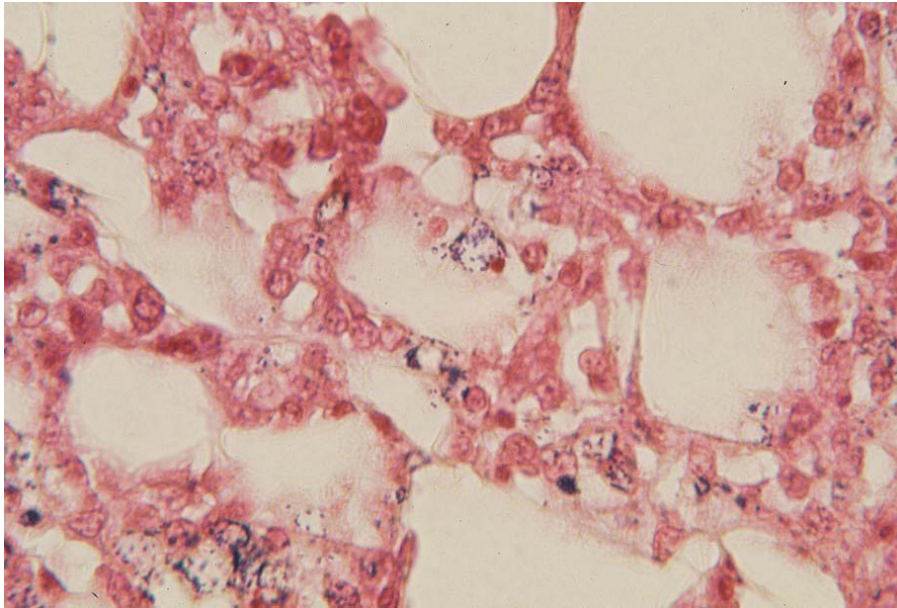


Figure 23. Histological section of retrobulbar tissue in tilapia affected by streptococcosis showing Gram positive bacteria scattered throughout (Grams x 400).

Francisellosis

Definition

Francisellosis is the term used to describe infection associated with *Francisella philomiragia* subspecies *noatunensis* which has emerged as a major pathogen of farmed cod. *Francisella* spp. are also a major pathogen of farmed tilapia.

History

A novel granulomatous disease was first described in farmed cod in Norway in 2005 as the cod farming industry developed. This disease was demonstrated to be caused by *Francisella* spp. bacteria. A similar condition, originally considered to be caused by a rickettsia-like organism was described in farmed tilapia in many countries and has now been confirmed as also being caused by *Francisella* spp.

Clinical signs of disease

Affected fish appear emaciated and may have raised haemorrhagic nodules in the skin, uni- or bilateral ocular pathology including opacity and corneal perforation. Internally there may be minor or extensive white, partly protruding nodules in the spleen, heart, kidney and liver (Figure 24). Kidney and spleen may be swollen and a thickened intestinal mucosa may also be present. Sero-haemorrhagic ascites may also be present.

Histopathology

Extensive chronic granulomatous inflammation with multiple granulomas in visceral organs and heart, white muscle, gills and eye. Predominantly cell type within granulomas are hypertrophied, foamy macrophages similar to epithelioid cells. Some macrophages may contain spherical, bacteria-like cells.

Aetiology

Francisella philomiragia subspecies *noatunensis* is the proposed name for the novel subspecies of these weakly Gram negative, intracellular coccobacilli. The bacteria can be grown on cysteine heart agar with 5% sheep blood added and appear low convex, whitish and mucoid.

Diagnosis

Clinical signs, pathology and histopathology give presumptive diagnosis and confirmation is by culture and PCR.

Host and geographic range

Farmed cod have been affected in Norway and Ireland and tilapia have been affected worldwide, including the UK. Atlantic salmon have been affected by a *Francisella* spp. bacteria in freshwater in Chile.

Source

Wild marine fish (cod) have been detected with *F. philomiragia* in the Norwegian sea.

Treatment and control

No effective treatment due to the intracellular nature of the infection. Experimental vaccines are in development. Removal of affected fish and disinfection of premises/equipment and fallowing.

References

Mikalsen, J. *et al.* (2007) *Francisella philomiragia* subspecies *noatunensis* isolated from farmed Atlantic cod (*Gadus morhua*). International Journal of Systemic and Evolutionary Microbiology, 57, 1960 – 1965.

Olsen, A., *et al.* (2006) A novel systemic granulomatous inflammatory disease in farmed Atlantic cod, *Gadus morhua* L. associated with a bacterium belonging to the genus *Francisella*. Journal of Fish Diseases, 29, 307 – 311.



Figure 24. Atlantic cod affected by francisellosis exhibiting numerous granulomas in the spleen (left) and on the skin (right) (Photos courtesy of Dr. Louise Henry).

DISEASES CONSIDERED TO BE BACTERIAL IN ORIGIN

Rainbow trout gastroenteritis (RTGE)

Definition

Rainbow trout gastroenteritis (RTGE) is an enteric syndrome of freshwater farmed rainbow trout reported in several European countries, which results in significant economic loss and daily mortalities of 0.5 to 1.0% are common.

History

RTGE was described in France in 2001 and subsequently in the UK and Spain. There has been one case in Ireland in 2008.

Clinical signs of disease

Lethargy, discolouration and slight swelling (oedema) of body giving a hunched up appearance. Internally, congestion and oedema of the intestinal wall with a yellow catarrhal faecal cast (Fig. 25). Mortalities can persist for many weeks and the condition is associated with warmer water temperatures in the summer months.

Histopathology

Congestion and oedema of the intestinal wall with large numbers of segmented filamentous bacteria (SFB) within the digestive tract (Fig. 25).

Aetiology

Not fully established and the role of the SFB remains unclear as they are also found in apparently healthy fish. However the “*Candidatus arthromitis*” may have some role to play in the disease. This bacteria has not yet been cultured in vitro.

Diagnosis

Clinical signs, pathology and histopathology.

Host and geographic range

Rainbow trout in freshwater are the affected species, although there have been a low number of suspect (unconfirmed) cases in Atlantic salmon in freshwater. Disease is present in France, Croatia, UK, Ireland, Italy and Spain.

Source

Unknown, however, infected farmed trout will be a significant reservoir.

Treatment and control

Changing the diet type or the addition of salt to the diet as well as broad spectrum antibiotics all appear to be effective once the disease is present, however, none appear to be preventative. Biosecurity is important in preventing the disease entering a farm.

Reference

Branson, E. (2003) Rainbow trout gastro-enteritis (RTGE) – first diagnosis in the UK. Fish Veterinary Journal, 7, 71 – 76.

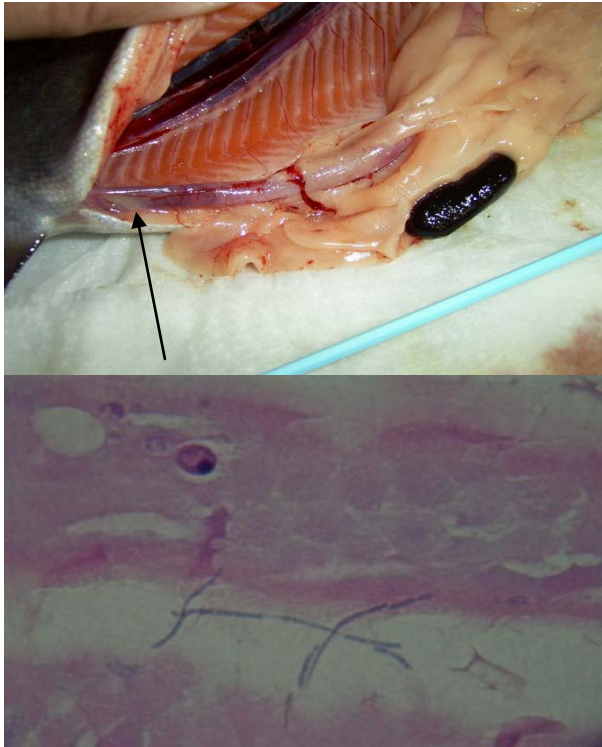


Figure 25. Rainbow trout affected by RTGE presenting with congested and swollen intestine with catarrhal intestinal contents (black arrow) (left) and histopathology section of intestine with SFB present (white arrow) (H & E x 400).

Red mark syndrome (RMS) or Cold water strawberry disease

Definition

Red mark syndrome (RMS) is an infectious dermatitis of rainbow trout which does not cause mortality but presents as dramatic haemorrhagic marks on the skin.

History

RMS or cold water strawberry disease was first observed in the UK in 2003 and is also present in mainland Europe (1985 in France). The disease has not been observed in Ireland. Strawberry disease (which appears identical pathologically) as described in the USA, has been reported there since the 1960s. The disease causes significant economic impact for trout farms in Europe.

Clinical signs of disease

Red, haemorrhagic marks on the flanks of trout can appear suddenly and then resolve within a few weeks (or months) without treatment (Figure 26). There are no mortalities or internal abnormalities associated with the disease.

Histopathology

Lesions present with a massive inflammatory reaction in the dermis with dissolving scales.

Aetiology

Not fully established although *Flavobacterium psychrophilum* and rickettsia-like organisms have been associated with the condition in Scotland and the USA respectively. An adeno-like virus was observed from two cases in France.

Diagnosis

Clinical signs and histopathology.

Host and geographic range

Rainbow trout in freshwater are the affected species, although there have been a low number of suspect (unconfirmed) cases in brown trout in freshwater and rainbow trout in seawater. Disease is present in France, Germany, UK and an identical condition known as strawberry disease is present in the USA.

Source

Unknown, however, infected farmed trout are a significant reservoir.

Treatment and control

The lesions will resolve eventually without treatment, however, broad spectrum antibiotics do induce a rapid healing of the condition. Avoidance of any livestock from infected farms reduces the chances of introduction of RMS onto a site.

Reference

Verner-Jeffreys, D. W., *et al.* (2008) Emergence of cold water strawberry disease of rainbow trout *Oncorhynchus mykiss* in England and Wales: outbreak investigations and transmission studies. *Diseases of Aquatic Organisms*, 79, 207 – 218.



Figure 26. RMS affected rainbow trout exhibiting three typical lesions.

FUNGAL DISEASES

Saprolegniosis

Definition

Saprolegniosis is the term most commonly used to describe infections in fish and fish eggs associated with the water mould of the fungus *Saprolegnia* spp.

History

Epizootics of wild Atlantic salmon with fungus were described at the end of the 19th century in the UK. A further wave of extensive mortalities in the rivers of Western Europe in the 1970s and '80s in the ulcerative dermal necrosis (UDN) outbreak were probably almost all ultimately caused by secondary saprolegnia infections.

Clinical signs of disease

Lesions are focal, grey-white patches on the skin or gills of fish which, when examined underwater have a cotton-wool-like appearance where the hyphal filaments extend out into the water (Figure 27). The early lesions are often almost circular and grow by radial extension around the periphery until lesions merge. At this later stage the patches are often grey or brown in colour as mud or silt becomes trapped by the mycelium. Gills, mouth or branchial cavity can also be affected. Internal infections in the peritoneum or gastrointestinal tract in younger fry can also be seen and results in high mortalities. Freshwater fish eggs are also very prone to infection.

Histopathology

The fungus usually establishes itself focally, invading the dermis and extending laterally over the epidermis eroding it as it spreads. In haematoxylin and eosin stained sections, infected with saprolegnia, numerous hyphae are seen on the skin surface, beneath which are areas of degenerating tissue ranging from superficial dermal necrosis and oedema to deep myofibrillar necrosis and extensive haemorrhage.

Aetiology

Saprolegnia parasitica-diclina complex is both a saprotroph and a necrotroph, typically feeding on waste from fish or other dead cells. Where they inhabit a live animal the fungal infection is known as a mycoses. Saprolegnia fungi are with the Oomycete Class that has aseptate hyphae. They are widely distributed in the freshwater aquatic habitat, produce motile biflagellate spores and undergo asexual reproduction by means of zoospores, produced in zoosporangium.

Diagnosis

Clinical signs, fresh smears for microscopy and histopathology (Fig. 27).

Host and geographic range

Global and ubiquitous distribution, affecting many temperate, freshwater fish species and their eggs.

Source

Saprolegnia sp. fungi are ubiquitous in freshwater and will tolerate brackish water and even moist soil.

Treatment and control

It has long been considered that the fungi responsible for saprolegniosis are secondary pathogens and lesions are commonly seen after handling or trauma which abrade the skin and allow invasion by fungi. Overcrowding and poor water quality may also give rise to infection. Affected fish are difficult to treat although formalin, formalin with bronopol (Pyceze) and salt as bath treatments show some benefit. Liquid paraffin in the feed is of benefit for fry which have fungal infections in the gastrointestinal tract.

Reference

Roberts, R. J. (2001) The mycology of teleosts. Chapter 9. In: Fish Pathology, 3rd edition (edited by R. J. Roberts), W. B. Saunders, London.

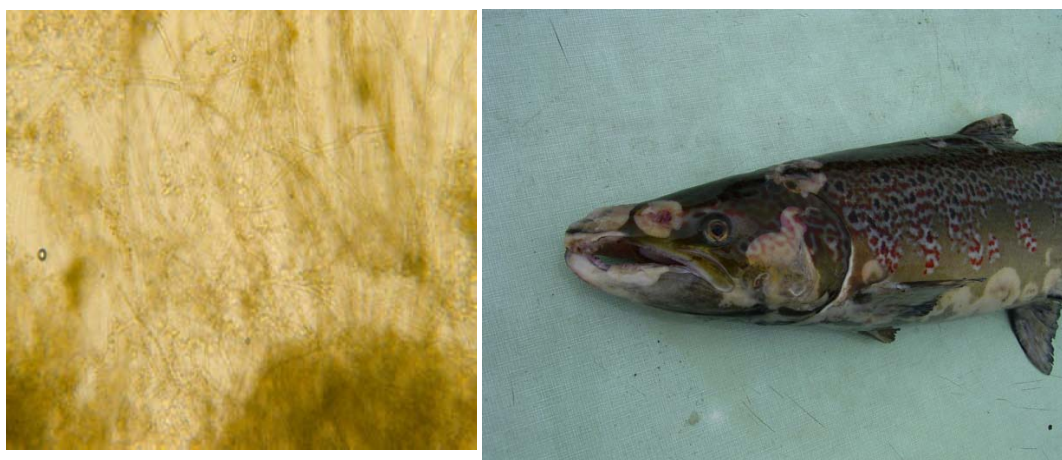


Figure 27. Fresh mount of aseptate saprolegnia fungal hyphae (left) and wild adult Atlantic salmon with fungal plaques obvious as grey-white marks.

Epizootic ulcerative syndrome (EUS)

Definition

Epizootic ulcerative syndrome (EUS) is considered to be an infection with the oomycete known as *Aphanomyces invadens* or *A. piscicida* and characterized histologically by penetrating hyphae surrounded by granulomatous inflammation. It is an epizootic condition of wild and farmed freshwater and estuarine fish. It is a notifiable disease in Ireland.

History

EUS is also known as red spot disease (RSD), mycotic granulomatosis (MG) and ulcerative mycosis (UM). The first outbreak is considered to have been in Japan in 1971 and at least 24 countries in four continents are now known to be affected.

Clinical signs of disease

Red spots may be observed on the body surface, head, operculum or caudal peduncle. Large, red or grey shallow ulcers, often with a black - brown necrosis are observed in the later stages. Most species other than striped snakeheads and mullet will die at this stage. In species such as the snakehead, the lesions become more extensive and can lead to complete erosion of the posterior part of the body or to necrosis of both soft and hard tissues of the cranium (Figure 28).

Histopathology

Early EUS lesions are caused by erythematous dermatitis with no obvious oomycete involvement. Fungal hyphae are observed growing in muscle as the lesion progress from a mild chronic active dermatitis to a severe locally extensive necrotizing granulomatous dermatitis with severe floccular degeneration of the muscle. Granulomas are formed around the penetrating hyphae (Fig. 28).

Aetiology

Aphanomyces invadens has an aseptate fungal-like mycelia structure and this oomycete has two typical zoospore forms.

Diagnosis

Clinical signs, fresh smears for microscopy and histopathology. PCR is also used.

Host and geographic range

EUS causes mortality in farmed and wild fish worldwide. Around 76 species of fish have been confirmed by histopathological diagnosis to be naturally infected by EUS. Carp, tilapia and milkfish are considered resistant to the disease. The disease is widespread in Asia, North America, Southern Africa and Australia.

Source

It is not clear how the fungus survives outside of host fish, although if the motile zoospore does not find a suitable substrate it will encyst.

Treatment and control

There is no protective vaccine and no effective treatment although to minimise fish losses in infected ponds water exchange should be stopped and lime or hydrated lime and/or salt be applied.

Reference

O. I. E. (2009) Manual of Diagnostic Tests for Aquatic Animals. O.I.E., Paris, France.



Figure 28. Indian major carp affected by EUS exhibited advanced dark brown/black lesion with tail necrosis (middle) and earlier stage haemorrhagic lesions (right). Histopathology of the lesion in muscle of rohu showing *Aphanomyces invadens* hyphae staining up black (left) (Grocotts x 200).

PARASITES

Amoebic and protozoan infections

Costiasis

Definition

Costiasis is the term used for infection of fish by costia or *Ichthyobodo necator*, a protozoan parasite of skin and gills.

Clinical signs of disease

Mortality in fry, flashing, thickened mucus on skin giving an opaque blue-grey appearance to body. Respiratory distress and mortality with gill infestation.

Histopathology

Epidermal or epithelial (gill) necrosis and sloughing (Fig. 30).

Aetiology

Ichthyobodo necator has two stages in its life cycle, a) a free swimming kidney shaped stage and b) a sessile pyriform stage which penetrates the epithelial cells. Both stages are approximately 10µm in length and have two flagellae (Fig. 29).

Diagnosis

Clinical signs and light microscopy.

Host and geographic range

Global and not host specific.

Source

Fish become infected by the free swimming forms. Free living and parasitic forms presumed to encyst under adverse environmental conditions. Cysts in the water become an additional source of infection and contribute to the spread of the organism to new areas.

Immunity

It is generally assumed that some protective immunity is acquired on recovery from an infection.

Treatment and control

Formalin baths or flush treatments are effective. Salt may also help as a bath in some cases. The parasite multiplies rapidly between 10 and 25°C and encysts at about 8°C.

Note

Ichthyobodo spp. have been found on marine salmonids and other marine species and has been associated with high mortalities in young fish, with pathology similar to that in freshwater.

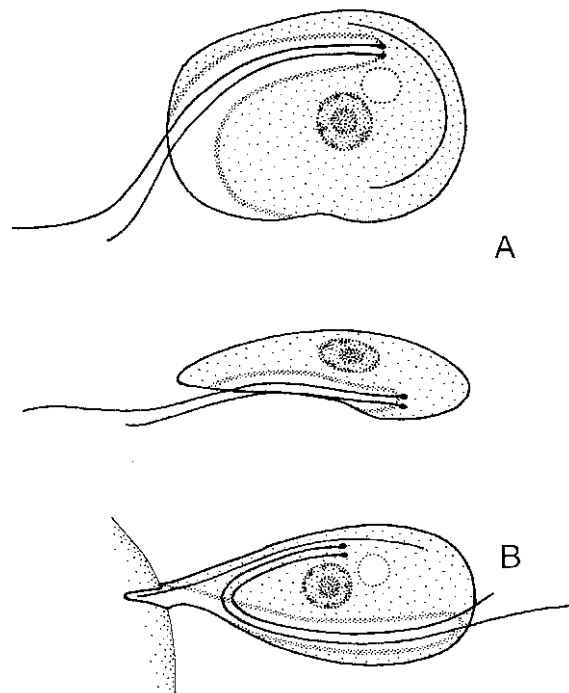


Figure 29. *Ichthyobodo necator*, free living (A) and attached stage (B) forms.

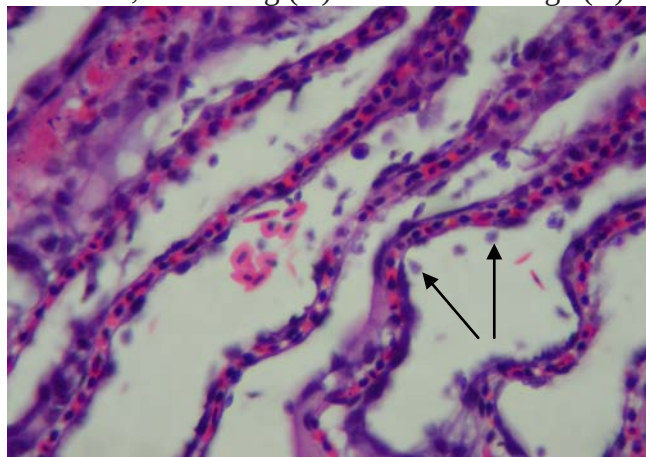


Figure 30. Gills from marine stage Atlantic salmon with attached stage costia present (arrows) (H & E x 400).

Chilodonella and *Trichodina* spp.

Trichodina spp. and *Chilodonella* spp. are ciliated protozoans that can give rise to mortalities in young fish due to skin and/or gill damage (Figs. 31 & 32). Treatment for chilodonella is with formalin and/or salt and *Trichodina* sp. do not usually require treatment unless infestation is heavy. Improving the tank or pen environment will usually help the situation with regard to any parasite infestation.

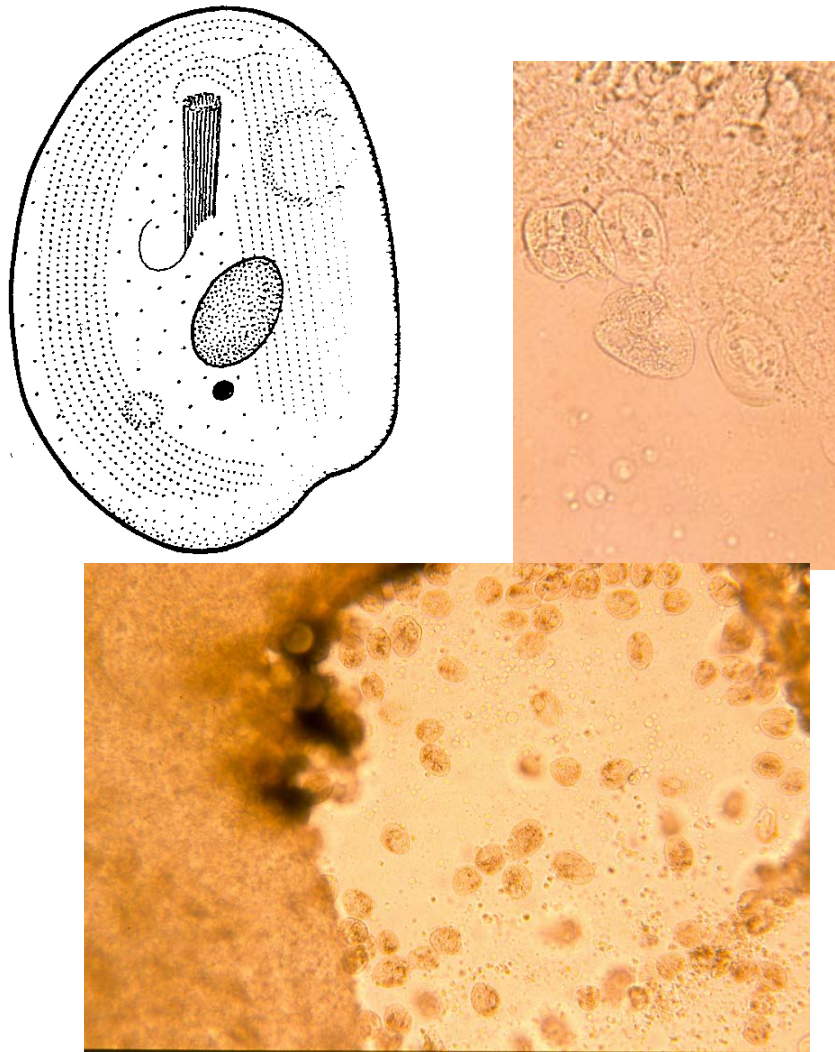


Figure 31. *Chilodonella* spp. (line drawing – top left and fresh gill smears (right) showing distinct heart shape (top right) and a heavy infection on gills (bottom).

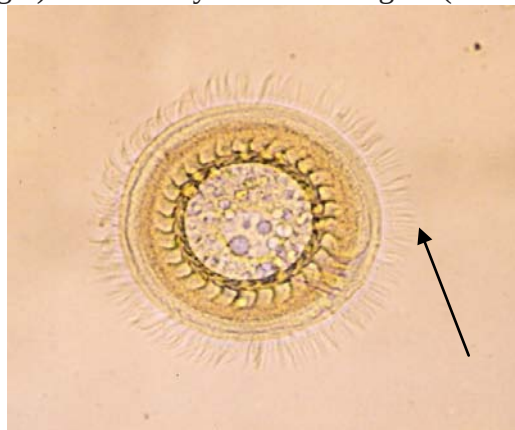


Figure 32. *Trichodina* sp. from skin of salmon with cilia obvious (arrow)

Hexamita sp.

Hexamita sp. are flagellated protozoans often observed in the digestive tract of fish. Trophozoites of *H. salmonis* can be found in the lumen of the intestine and heavy infestations can give rise to anorexia, emaciation (Figure 33), abdominal distension, faecal pseudocasts,

catarrhal enteritis and mortality. Oral sulphonamides have been effective in controlling this parasite in the past.



Figure 33. Salmon fry: the lower fish in poor condition and suffering a heavy infestation with *Hexamita* sp..

Amoebic gill disease

Definition & History

Amoebic gill disease (AGD) of farmed salmonids is a significant disease which results in respiratory distress and mortality. Amoebic infections of wild urchins and crabs were described in the 1960s and 1980s but AGD emerged with the development of salmon farming and was first described in Tasmania in 1985.

Clinical signs of disease

Lethargy, respiratory distress (fish nearer the water surface and breathing rapidly) and mortalities. Affected gills exhibit multifocal patches of white to grey swollen gill tissue with associated excess mucus (Fig. 34).

Histopathology

Hyperplasia and hypertrophy of gill epithelium with lacunae development and amoeboid organisms obvious (Fig. 34).

Aetiology

Neoparamoeba sp. (originally described as *N. pemaquidensis*, but now *N. perurans*) which has one or more parasomes in the cytoplasm (15 - 40µm diameter) and transitional forms have up to 50 digitate pseudopodia.

Diagnosis

Clinical signs, light microscopy and histopathology.

Host and geographic range

Atlantic salmon and rainbow trout are most susceptible to AGD, however, turbot, Pacific salmon, sea bass and other marine species have also been affected. AGD is most significant in Tasmania, however, it has also been reported in Ireland, France, Spain, Norway and Scotland.

Source

A single smolt with AGD may have several hundred thousands of amoebae, therefore the risk from a pen of infected fish is tremendous. Dead fish have live amoeba for up to 3 days post mortem. The parasite can survive in the water and maintain its infectivity for at least 14 days.

Immunity

Atlantic salmon can develop resistance to AGD. Serum antibodies have been detected in previously infected fish, but these do not correlate with resistance. All efforts to immunize fish against AGD have not yet been successful.

Treatment and control

The most effective and frequently used treatment is freshwater baths. Briefly fish are bathed in freshwater close to zero salinity for 2 to 6 hours.

Reference

Zilberg, D. & Munday, B. L. (2006) Phylum amoebazoa. In: Fish Disease and Disorders Vol. 1 (ed. By P. T. K. Woo), CAB Int., UK

Note

Systemic amoebiasis is also observed in freshwater fish species and can present as a granulomatous disease.



Figure 34. Salmon affected by amoebic gill disease showing thickening of gill lamellae as pale areas (left) and on histopathology fusion of filaments with hyperplasia and lacunae development (right) (H x E x200).

Ich or white spot

Ichthyophthirius multifiliis is a pathogenic protozoan ciliate which infects freshwater fishes. Affected fish are lethargic, dark in colour, may flash or rub themselves exhibit respiratory distress and then mortality. White spots (0.5 to 1mm diameter) may be obvious (Fig. 35) on skin and gills and under light microscopy the characteristic large, brown ciliate with a horseshoe shaped nucleus will be obvious (Fig. 36). Salt, formalin and Virkon flowing treatments all appear to help to control the parasite, although moving fish from severely affected tanks to clean tanks is also of benefit.



Figure 35. Salmon parr affected by white spot and life cycle of parasite.

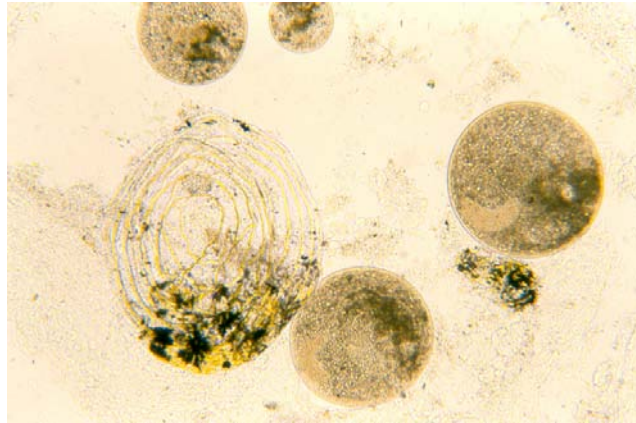


Figure 36. Fresh skin smear showing fish scale and *Ichthyophthirius multifiliis* with distinct horseshoe shaped nucleus.

Metazoa

Kudoa

Definition & History

Kudoa sp. are myxosporidean parasites of marine fish. The parasite has been responsible for significant economic losses in the wild fisheries (hake, whiting, mackerel) sector by causing post mortem myoliquefaction which results in the fish being unmarketable. More recently it has become a pest for farmed salmon (and other marine species).

Clinical signs of disease

Signs of infestation only become apparent at post mortem, with the exception of very heavy infestations, when the flesh softens either in discrete patches or extensively (depending on the level of infestation) (Figure 37).

Histopathology

The aggregates of parasite spores, the pseudocysts, are invested within individual muscle cell sarcolemmae, replicating the sarcoplasm with a mass of spores. In some species the cyst is eventually invested by a pigmented capsule, but in others proteolytic enzymes secreted by the parasite lead to extensive myodegeneration producing the softened, whitish, mushy muscle.

Aetiology

Kudoa thyrsites is the myxosporidean parasite (Phylum: Myxozoa) which infects farmed salmonids and the spores are stellate in shape, with four valves and four polar capsules (Figure K).

Diagnosis

Post mortem signs should be investigated using light microscopy. Fresh tissue mounts with or without stains (Giemsa, Methylene Blue) or histopathology (Giemsa) will allow visualization of the distinctive spores. PCR screening has been used to some extent in Canada.

Host and geographic range

K. thyrsites has a global distribution and has been reported in many fish species. Farmed salmonids have been affected in British Columbia, Canada, Ireland and France.

Source

Wild fish and farmed salmonids carry the mature spores. The life cycle is not established, however, myxosporideans have complex life cycles using more than one host. They usually use fish and an oligochaete or polychaete worm, and in one case a bryozoans. Direct transmission of *K. thyrsites* failed when naïve fish were fed fresh myxospores.

Immunity

Fish can respond to myxozoan infections through a number of specific and non-specific mechanisms. No vaccines available or in development.

Treatment and control

No treatment available. Control by monitoring and early, accelerated harvest with rapid processing if present.

Reference

Feist, S. W. & Longshaw, M. (2006) Phylum Myxozoa. In: Fish Disease and Disorders Vol. 1 (ed. By P. T. K. Woo), CAB Int., UK

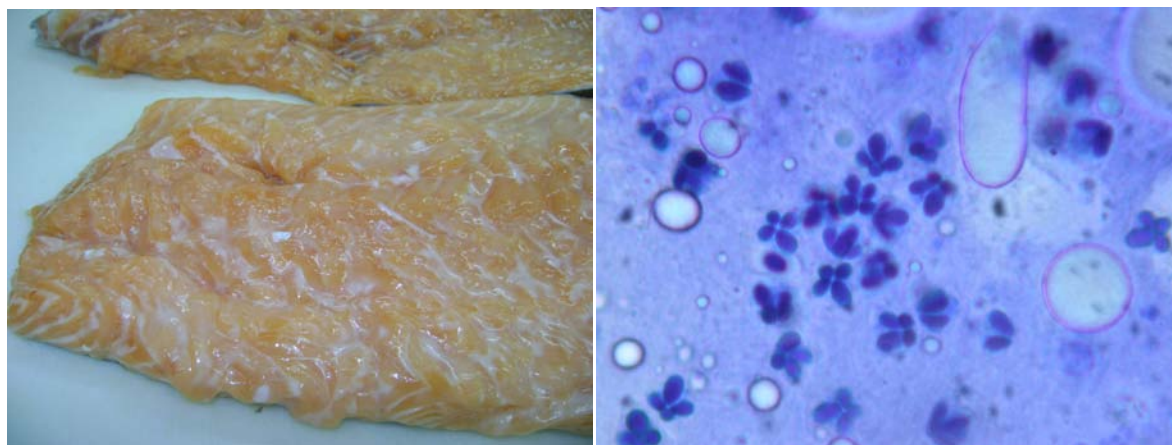


Figure 37. Salmon fillet presenting extensive myoliquefaction as a result of *Kudoa* sp. infestation (left) and the parasitic myxosporidiean *Kudoa* sp. spores can be seen in a fresh squash of affected tissue (right) (methylene blue, x 1000).

Proliferative kidney disease (PKD)

Definition & History

Proliferative kidney disease (PKD) is a condition affecting primarily rainbow trout in freshwater, but all salmonids can be affected, and presents with a swollen kidney and causes mortality. The disease was originally termed PKD in 1974 in the UK but was observed for many years prior to this. The disease was originally associated with the PKX cell and the agent is now known to be a myxozoan called *Tetracapsuloides bryosalmonae*.

Clinical signs of disease

Dark colouration, abdominal swelling, exophthalmia, pale gills, loss of normal body position and respiratory distress. Internally the fish may exhibit granulomatous renal swelling, splenomegaly and generalized pallor (Fig. 38).

Histopathology

Renal haematopoietic hyperplasia followed by granulomatous interstitial nephritis. The extrasporogonic or sporogonic stages of the parasite may be observed.

Aetiology

Tetracapsuloides bryosalmonae is the causal agent and is a myxozoan with four distinct polar capsules which also parasitizes freshwater Bryozoa (moss animals).

Diagnosis

Clinical and gross pathological signs combined with history and season of occurrence (warmer months) are characteristic. Histopathology and IHC will confirm.

Host and geographic range

Disease is endemic in Western Europe and North America and affects all salmonids, but rainbow trout are particularly susceptible.

Source

Wild fish and farmed salmonids carry the mature spores.

Immunity

Fish can respond to myxozoan infections through a number of specific and non-specific mechanisms. No vaccines available although research continues.

Treatment and control

No treatment available although malachite green and fumagillin had been used in the past. Control lower summer water temperature (using bore hole water), delaying transfer of naïve stocks to endemic waters until later in the year (to allow acquired immunity but not clinical disease to develop), eliminating secondary infections and reducing feeding rates have been implemented to limit impact.

Reference

McGurk, C. (2005) Microscopic studies of the link between salmonid proliferative kidney disease (PKD) and bryozoans. *Fish Veterinary Journal*, 8, 62 – 71.



Figure 38. Rainbow trout affected by PKD exhibiting a swollen granulomatous kidney (Photo courtesy of Prof. R. Richards).

Gyrodactylosis

Definition & History

There are approximately 20,000 species of the monogenean *Gyrodactylus*, however, one species, known as *G. salaris*, has affected wild Atlantic salmon in more than 44 rivers in Norway resulting in heavy losses of wild fry. This one species is exotic to, and notifiable in Ireland. Other gyrodactylids are observed on wild and farmed fish in Ireland.

Clinical signs of disease

Increased flashing, fish rub on substrate, then grayish colouration due to increased mucus on the skin and eventually fin erosion. Diseased fish are lethargic and are found in slower moving water.

Aetiology

G. salaris is an ectoparasite mainly on Atlantic salmon, but can survive and reproduce on several salmonids, including rainbow trout, Arctic char and brook trout. The parasite is viviparous and is an obligate parasite (Fig. 39).

Diagnosis

Observation of gyrodactylids on salmon or trout either in skin scrapings or on fins will require molecular and/or morphological identification of species (Fig. 39).

Host and geographic range

G. salaris is an ectoparasite mainly on Atlantic salmon, but can survive and reproduce on several salmonids, including rainbow trout, Arctic char and brook trout. The parasite has been found on wild fish in Russia, Sweden and Norway. In 2006 it was reported from fish farms in

Italy (trout) and in 2007 in Poland and Macedonia. The UK and Ireland appear free of the parasite.

Source

Salmonids can act as carriers of the parasite and the parasite has spread between rivers and farms mainly by restocking and transport of live fish.

Treatment and control

G. salaris is sensitive to chemical changes e.g. high salinity, formalin and halogens as well as slightly acidic solutions of aluminium sulphate. This latter chemical has been used to eradicate the parasite in moderately acidified waters in Norway.

Reference

O. I. E. (2009) Manual of Diagnostic Tests for Aquatic Animals. O.I.E., Paris, France.

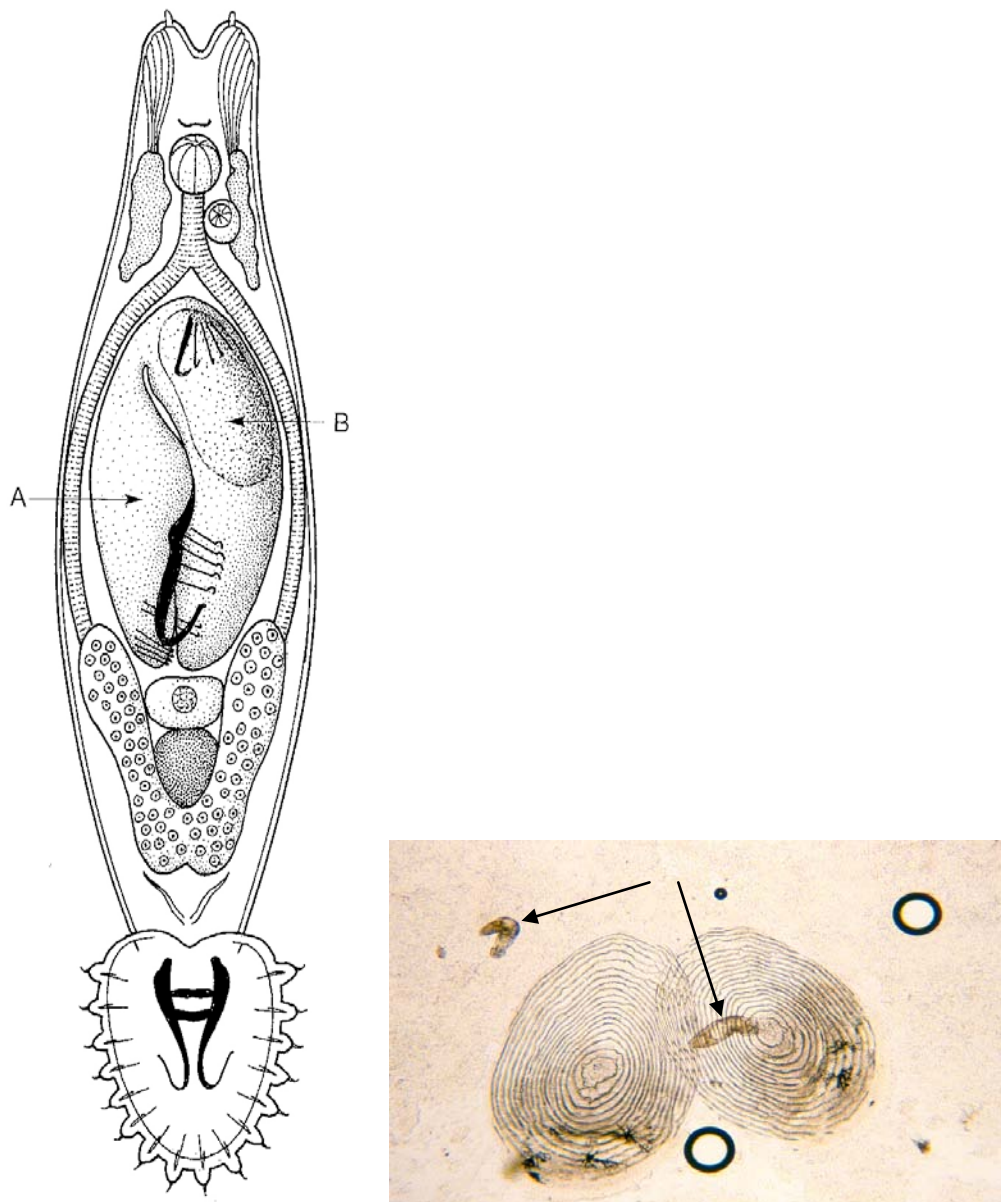


Figure 39. Line drawing of *Gyrodactylus* sp. with developing embryo (A) and secondary embryo (B) (left) and fresh skin scrape of salmon showing scales and *Gyrodactylus* spp. (arrows).

Sea lice

Definition & History

Sea lice are copepods which are marine ectoparasites that feed on the mucus, epidermal tissues and blood of host marine fish. The levels are monitored on marine fish farms in Ireland by regular inspections. Sea lice have been recorded on wild marine fish since records began and are now a significant pest on marine salmonid farms globally.

Clinical signs of disease

Lice are obvious to the eye on the skin of the fish and when present in high numbers give rise to whitening of the skin on the heads and backs (Figure 40), haemorrhagic marks on the ventrum and eventually dermal perforation and ulceration leading to exposure of head bones/cartilage in worst cases. Heavily infected fish become lethargic, lose their appetite and mortalities eventually ensue.

Histopathology

Mechanical damage caused by copepodid and attachment and feeding chalimus stages leads to epidermal hyperplasia. Lesions develop with dermal fibrosis and inflammatory infiltration.

Aetiology

Lepeophtheirus salmonis and *Caligus elongatus* are the two species of most significance in Northern Europe and *C. rogercresseyi* in Chile. Generation time for *L. salmonis* is 8 – 9 weeks at 6°C, 6 weeks at 9 to 12°C and 4 weeks at 18°C. Egg strings from one adult female *L. salmonis* can bear a total of 700 eggs. *C. curtus* have been reported on cod.

Diagnosis

Observation of parasites and clinical signs. Larval stages can be examined by light microscopy.

Host and geographic range

L. salmonis is host specific infecting only salmonids. *C. elongatus* infects salmonids and other marine species. *L. salmonis* in Northern hemisphere countries and *C. elongatus* is a cosmopolitan species and has been found on over 80 species of fish.

Source

Wild and farmed salmonids are source of *L. salmonis* and marine fish are source of *C. elongatus*.

Immunity

Humoral antibodies produced when salmon immunized with crude extracts of *C. elongatus* or *L. salmonis*. Non-specific cellular responses suggested as well. A sea lice vaccine remains a focus for many research groups.

Treatment and control

Farm management through regular lice counts, effective treatment, fallowing, synchronous treatments of all farms in bay, rotation of treatments and monitoring of resistance are all important. Medical treatments include oral emamectin benzoate, teflubenzuron, diflubenzuron and baths with pyrethroids (cypermethrin, deltamethrin), azamethiphos and hydrogen peroxide. Cleaner fish (wrasse), genetic selection and feeding of immunostimulants also are a focus for control. Resistance of lice to some medicines is increasingly reported.

Reference

Wootten, R., *et al.* (1982) Aspects of the biology of the parasitic copepods *L. salmonis* and *C. elongatus* on farmed salmonids, and their treatments. Proceedings of the Royal Society of Edinburgh, 81B, 185 – 197.



Figure 40. Salmon infested with sea lice (*L. salmonis*) (left) and adult sea lice (right) with an ovigerous female at top.

Argulus

The freshwater louse or *Argulus* sp. (Fig. 41) is common on wild fish in Europe and Asia. Heavily infected fish are lethargic, anorexic and mortalities eventually ensue. The skin damage caused by the parasites is similar to that with sea lice. In a farm situation or small ponds, treatments can be undertaken as for sea lice, however, prevention of introduction and biological controls (substrate for eggs with removal weekly) are also important.



Figure 41. *Argulus* sp. adult.

