

Antiparasiticides

- Agents against parasites are commonly divided into ecto- and endoparasitic substances.
- The prefix ecto means external and refers to parasites on skin or gills, whereas endo means internal, and refers to parasites in the gastro-intestinal system of the fish.

Ectoparasitic agents

- Ectoparasitic agents may be applied in baths (acetylcholinesterase inhibitors, pyrethrines and pyrethroids) or by addition to medicated feed (ivermectins and chitin synthetase inhibitors).

1. Acetylcholinesterase inhibitors

- Acetylcholinesterase is an enzyme that specifically cleaves acetylcholine to acetate and choline.
- It is located both in the pre- and post-synaptic nerve terminal, bound to the membrane.
- An acetylcholinesterase inhibitor indirectly provides a cholinergic action by prolonging the lifetime of acetylcholine produced at the cholinergic nerve endings.
- This results in the accumulation of acetylcholine in the synaptic space.
- One among several effects of this accumulation is paralysis of motor function (skeletal muscles) and convulsion.

- Organophosphate is an example of Acetylcholinesterase inhibitors.
- Organophosphates bind covalently to a serine OH group at the active site of the acetylcholinesterase molecule.
- When this occur the enzyme is permanently inactivated and unless a reactivator like pralidoxime (PAM), a synthetic pyridinium compound which can reactivate inhibited acetylcholinesterase by displacing the organophosphate, is administered, restoration of the activity require synthesis of new enzyme molecules.
- Organophosphates are administered by bath.
- At present the only organophosphate in use is azametiphos.
- Azametiphos is effective against adult and pre-adult lice whereas the larval stages are unaffected.
- Resistance to organophosphates has developed and requires an increase in dose and exposure time to achieve acceptable results.

2. Pyrethrines and pyrethroids

- The pyrethroid insecticides have their origin in the natural insecticide pyrethrum, an extract of *Chrysanthemum* sp.
- The extract contains six esters, pyrethrines, all with insecticidal properties.
- The naturally occurring pyrethrines degrade quickly when exposed to light, and synthetic analogues have been developed (pyrethroids) to enhance both insecticidal activity and reduce the excitability and finally paralysis of the parasite.
- Furthermore, it has been hypothesised that the observed neural action of pyrethroids can also be related to interference with the calcium regulation in the cell.
- Direct effects on the release of neurotransmitters have been observed, by blocking of the calcium channels.

- The pyrethroids are relatively safe for mammals due to lower receptor sensitivity and poor absorption from the gut.
- In fish, however, the safety margin is less.
- The pyrethroids have a broader spectrum of activity than the organo- phosphates since chalimus, pre-adult and adult stages of the lice are affected.
- The pyrethroids in use today are cypermethrin and deltamethrin.
- Pyrethroids are administered by bath using an emulsifier due to low water solubility.
- Pyrethrum is administered using a layer of oil on the surface of the water in which the drug is dissolved.
- The fish must pass the oily layer in order for the drug to have effect either by active jumping or in association with handling.

3. Avermectins

- Originally isolated from the actinomycete, *Streptomyces avermitilis*, the avermectins represent a most recent developed group of insecticides.
- The mechanism of action in invertebrates, as described in the nematode *Caenorhabditis elegans*, is by binding to glutamate-gated chloride channels leading to an influx of chloride ions, thus giving a hyperpolarised cell.
- A second mechanism of action of avermectins is by increasing the production of the inhibitory neuro-transmitter GABA at nerve endings and prolonging the binding of GABA to the receptor.
- Avermectin used in fish farming is emamectin benzoate under the trade name Slice.
- Emamectin benzoate is highly effective against pre-adult and adult lice and prevents the maturation of chalimus to motile stages.
- Given orally incorporated in feed emamectin gives protection for reinfestation for an extended period compared to the bath-administered compounds.

4. Chitin synthetase inhibitors

- While the pyrethroids, organo-phosphorus compounds and avermectins all act on the nervous system of the parasite, the benzyl-ureas have an entirely different mode of action.
- Chitin is a major part of the exoskeleton (cuticle) of insects and crustaceans.
- The orally administered delousing agents Ektobann Vet. (teflubenzuron) and Lepsidon Vet (diflubenzuron) act by inhibiting chitin synthetase, thereby interfering with the formation of cuticle, thus exerting an effect at the moulting stage in the life cycle of exposed organisms.
- Chitin synthetase inhibitors have effects on all stages except adult lice that already have formed their exoskeleton.
- Diflubenzuron is moderately absorbed from the gut of the fish with a bioavailability of 31%.
- The drug enters the lice via the blood and skin of the fish.

Endoparasitic agents

- Since endoparasites are by definition found in the stomach or intestine of fish, agents against such parasites are only given orally.

1. Praziquantel and the benzimidazoles

- Praziquantel is rapidly and reversibly absorbed by the parasite.
- It is believed to have two primary and immediate actions in susceptible organisms.
- At the lowest therapeutic concentration the drug causes increased muscular activity followed by contraction and spastic paralysis.
- It is probably this effect that causes the parasite to lose their attachment to the host tissue.
- At a higher but still therapeutic concentration, praziquantel induces vacuolisation and vesiculation of the parasite tegument.
- This leads to osmotic and nutritional imbalance and release of the contents of the parasite, followed by activation of the host defence mechanisms and finally destruction of the worm.

2. Anticoccidial drugs

- Anticoccidials are generally given to poultry in the feed to prevent acute disease and the economic loss often associated with subacute infection.
- Prophylactic use is preferred because most of the dosage occurs before signs become apparent, and delayed treatment may not benefit the entire flock.
- There are a lot of drugs developed, including amprolium, clopidol, halofuginone, ionophores, nicarbazin, tetracyclines and sulphonamides, but only a few are used in fish for infections of parasites producing similar spores (i.e. Myxosporea and Microspores)
- **Fumagillin:** Fumagillin, an antibiotic isolated from *Aspergillus fumigatus*, was used primarily for treating *Nosema apis* (Microsporea) infections in honey bees *Apis mellifera*, and for treating patients with amoebiasis.

- In microsporidian *Octosporea muscaedomesticae*, fumagillin treatment caused a decrease in total RNA, suggesting that fumagillin inhibited RNA synthesis.
- In fish, fumagillin was effective against the microsporean *Pleistophora anguillarum* in eels *Anguilla japonica* and against *Myxidium giardii* infections by blocking the development and preventing the formation of new spores in European eels *Anguilla anguilla*.
- It is used to control *Enterocytozoon salmonis* and *Loma salmonae* infection in Chinook salmon (*Oncorhynchus tshawytscha*).
- It is effective against haemorrhagic thelohanellosis caused by *Thelohanellus hovorkai* and *Sphaerospora renicola* in common carp (*Cyprinus carpio*, L.) when administered during the infective period, early intracellular trophozoites and more developed plasmodia of *Hoferellus carassi*, *Myxobolus cerebralis* in rainbow trout and against the myxosporean *Sphaerospora testicularis* in sea bass.

- **Toltrazuril:** Toltrazuril displays both anthelmintic and antiprotozoal activity.
- It is a symmetric triazine derivative, and is capable of entering the host cell as well as the intracellular or tissue-inhibiting parasitic stages in chicken-parasitizing coccidia *Eimeria* sp.
- Incubation of *Glugea anomala*, which parasitizes the connective tissue of sticklebacks (*Gasterosteus aculeatus*), led to complete destruction of multinucleate meronts and to fragmentation of sporogonial plasmodia, suggesting an inhibitory effect of the drug on nuclear division possibly via inhibition of enzymes involved in pyrimidine synthesis and nuclear division.
- It is active against pre-spore stages of *Myxobolus* sp. in the gills of bream (*Abramis brama*), as well as against developmental stages of *Henneguya* sp. parasitic on the gills of the tapir fish (*Gnathonemus petersi*).

- It is also effective against parasitic ciliates, such as *Ichthyophthirius multifiliis*, *Trichodina* spp. and *Apiosoma* spp. and fish-infecting members of the Coccidia, Microsporidia and Myxozoa.
- It has been suggested that in Mediterranean fish, it is effective for treatment of Myxosporean infections including *Myxidium leei*.
- It is also known to be effective against the monogenean *Gyrodactylus aculeatus*.
- **Amprolium:** Amprolium is a structural analogue of thiamine (vitamin B1) causing competitive inhibition of thiamine utilization by the parasite.
- It acts upon the first generation in the cells of the intestinal wall, preventing differentiation of the metozoites.
- It may also suppress the sexual stages and sporulation of the oocysts.
- Amprolium has been shown to be ineffective against *Hexamita salmonis*, *Gyrodactylus* sp. and *Ichthyobodo necator* in rainbow trout.

- **Quinine:** Quinine acts on skin-inhabiting trophozoite stages of *Ichthyophthirius multifiliis* in ornamental fish and causes severe alterations in the parasites structure, mainly consisting of an enlargement of the alveolar sacs, a partial destruction of the nephroplasm, and a sloughing off of the membranes bordering the contractile vacuoles.
- It has also been shown to be effective against the plasmodial developmental stages of *Henneguya sp.* in tapir fish.
- Quinine administrated orally led to malformation of the presporogonic and pansporoblastic stages of *Henneguya sp.* parasitizing the gills of tapir fish.
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Treatment for Protozoan infections

- **Formalin:** Formalin (= a saturated 37% aqueous solution of formaldehyde gas) is a widely used chemical for parasite treatments or disinfections in both freshwater farms and marine hatcheries.
- It is also used as an anti-bacterial and anti-fungal agent and has become popular because of its recent licensing as an approved therapeutic for farmed finfish in the USA and Canada.
- **Hydrogen peroxide:** Hydrogen peroxide has been used as a bactericidal agent and as a fungicidal for eggs, but its main use has been against sea lice in the treatment of chalimus and mobile stages.
- It is found that under commercial farming conditions lice treated with hydrogen peroxide at 1500 ppm for 20 minutes at 10 °C did not reattach to salmon.
- Treated egg strings did not produce viable copepodites.
- Hydrogen is a strong oxidizing agent and causes the parasite to separate from its host, possibly by generating lethal oxygen emboli inside the sea lice.

- **Chloramine T:** Chloramine-T is an organic chlorocompound, which slowly breaks down to release chlorine.
- It is most commonly used as an antibacterial disinfectant and as a mucous stripping agent.
- It is also used for treatment of protozoa and for skin and gill flukes in a variety of fish species.
- Mixed infections can simultaneously be treated with this chemical.
- Treatment using low doses (2.5-10 ppm) for up to one hour does not normally lead to problems and is not stressful for either the freshwater or the marine environment.
- **Potassium permanganate (KMnO₃):** Potassium permanganate is very effective against cichlidogyrasis of tilapia fish (*Oreochromis hornorum*) and an ectoparasitic ciliate (*Ambiphrya ameiuri*) of channel catfish (*Ictalurus punctatus*).

- **Copper sulphate (CuSO₄):** It is used as a source of copper ion (Cu⁺⁺) for the treatment of marine fish diseases caused by the protozoan parasites *Amyloodinium (Oodinium) ocellatum* and *Cryptocaryon irritans*.
- It is also used for controlling fresh water fish diseases caused by *Costia*, *Chilodonella*, *Epistylis* and *Trichodina*, and ichthyophthiriasis.
- Total suspended solids are normally correlated to the efficacy of the chemical and it is usually toxic to fish, especially in soft water .
- **Iodophore:** Iodine is an effective disinfectant for most egg pathogens and has been widely adopted by the aquaculture industry.
- Iodophor treatment may prevent *Loma salmonae* infections of the endothelial cells of Chinook salmon by reduction of viability of spores.
- However, some spores survive even when high concentrations of iodophor are used.