

STUDIES ON GILL PARASITOSIS OF THE GRASSCARP (CTENOPHARYNGODON IDELLA) CAUSED BY DACTYLOGYRUS LAMELLATUS ACHMEROW, 1952.

III. THERAPY AND CONTROL

By

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Authors of textbooks on fish pathology (SCHÄPERCLAUS, 1954; KOCYLOWSKI-MYACZYŃSKI 1966; REICHENBACH-KLINKE, 1966; BAUER, MUSSELIUS and STRELKOV, 1969) have generally recommended the traditional bathing solutions (NaCl and NH₃) treatment of *Dactylogyrus* infections. Certain organic phosphoric acid esters however have increasingly gained favour in examinations of SARIG, LAHAV and SHILO (1965), who successfully employed Dipterex at a low concentration for long-term bathing treatment in the pond.

GRABDA and GRABDA (1966) have reported that Neguvon, which contains the same active substance as Dipterex, eliminated *Dactylogyrus* infection in a shorter time when applied at a higher concentration than the latter.

Specific treatment for *Dactylogyrus lamellatus* infection has not yet been dealt with in the literature except in a preliminary report (MOLNÁR, 1966) on attempts at therapy.

This paper is a report of extensive experiments conducted with the aim of establishing reliable control measures against *D. lamellatus* infection of grasscarps, which has become an economically important parasitic disease in pond farms in Hungary. In lack of specific information, the control schemes have been developed on the basis of the general veterinary hygienic measures set forth in the textbooks by SCHÄPERCLAUS (1954), BAUER (1959) and KOCYLOWSKI-MYACZYŃSKI (1963).

Previous studies (MOLNÁR, 1971) have shown that *D. lamellatus* is highly pathogenic and that it causes a considerable loss of condition even in sublethal infections. The main aim of the experiments was therefore on the one hand to find some means of preventing the clinical manifestation of *D. lamellatus* infection under pond farming conditions, and on the other hand to find a chemical agent suitable for killing the *D. lamellatus* parasites of massively infected hosts.

Control measures in breeding technology

Effective prevention of clinical dactylogyrosis begins already at the breeding phase and the technology of pond rearing should be established with due consideration to the developmental cycle of *D. lamellatus* and to the epizootiological features of the infection.

Control requirements are different in ponds for breeding and for table-fish production. In the latter the incidence of dactylogyrosis is low and it

generally becomes important only during the winter season. Treatment of the fishes by bathing before removal to, or from, the wintering pond is therefore as a rule sufficient to prevent economically important dactylogyrosis infection.

Prevention of infection in fry rearing ponds may be achieved in two ways:

1. The water supply to fingerling and nursery ponds should be provided from sources free of older grasscarp populations producing *D. lamellatus* larvae. The fry can in this way be reared free of dactylogyrosis, because artificial hatching eliminates contamination by infected mother fishes in the hatchery. A "clean" water supply to fry rearing and fingerling pond can even prevent the establishment of dactylogyrosis among fry already invaded by the larval parasite. *D. lamellatus* eggs or larvae which may occasionally be present even in "clean" water usually die through being unable to find a susceptible host before the water supply reaches the fish ponds. In a pond farm grasscarp fry were reared free of dactylogyrosis for four years by providing a "clean" water supply from a reservoir pond placed far enough away from the rearing ponds to ensure that parasites would die during their passage through the water-conducting channels. When (in 1970) the water supply had not been conducted through reservoir pond for some days for technical reasons, infection with *D. lamellatus* was already demonstrable among the fry only a few days later. This observation stresses the importance of waterborne larvae in the spread of dactylogyrosis.

2. If, as sometimes happens, breeders insist on keeping adult grasscarps in the water-conducting channels to prevent excess growth of vegetation, or if a supply of water can not be arranged dactylogyrosis-free rearing is impossible. Nevertheless, infection can be kept at a low level without drug therapy provided adequate breeding technological measures are taken.

Both practical and experimental observations have shown that acute dactylogyrosis develops only if the fishes are maintained at a high population density for a long time. Fry placed in the nursery pond at a density of 50–100 fishes/m² were invaded by only a few parasites during the short 20-day period of maintenance in this particular type of pond. Drug therapy only becomes necessary when the nursery period is prolonged beyond 20 days, or when the population density attains 200–300 fishes/m² under such conditions, the parasite counts increase rapidly, if no measures are taken.

Chemotherapy

This becomes necessary if the breeding technological methods are in themselves insufficient for an effective control of *D. lamellatus* infection.

In lack of literary data on the drug therapy of grasscarp dactylogyrosis, both the conventional bathing solutions and the recently applied organic phosphoric acid esters were tested for activity against the parasite.

A 2.5% solution of NaCl easily killed *D. lamellatus* under both experimental and field conditions. The fry tolerated this concentration well for fifteen minutes, and even severely infected hosts seemed unaffected by the bathing solution on a 10-minute application. This treatment can practically eradicate the parasite, but sometimes a few young stages may survive and later become sources of reinfection.

Organic phosphoric acid esters also proved to be very active against *D. lamellatus*. Five such preparations were tested: crystalline trichlorphon, Ditrifon 50, Flibol E, Nuvanol and Gardona. The active substance of Ditrifon 50 and Flibol E is trichlorphon [dimethyl-(1-hydroxy-2, 2,2-trichlorethyl phosphonate)], that of Nuvanol is iodophenphos (dimethyl-dichloro-iodophenylthiophosphate), and that of Gardona is 2-chloro-1-(2,4,5-trichlorophenyl) vinyl dimethyl-phosphate. All preparations were tested for activity during short-term and long-term bathing.

Ditrifon 50 was used to examine the efficiency of phosphoric acid ester on short-term bathing treatment. Fishes infected with *D. lamellatus* were placed in Ditrifon 50 solution containing the active substance at a concentration of 100 ppm for 20 min, 30 min, 1, 2, or 4 hours and they were examined one or three days later for viable parasites (Table I). Parasites were still present after one day, regardless of the duration of bathing but after three days all fishes except those treated for 20 minutes were negative.

Table I

Therapeutic results of bathing 3–5 cm long grasscarp fry* infected with 40–80 *D. lamellatus* parasites in Ditrifon 50 solution

Concentration of active substance in bathing solution	Time of examination after beginning of treatment (hours)	Time of bathing				
		20 min	30 min	1 hour	2 hours	4 hours
100 ppm	24	+	+	+	+	+
	72	+	–	–	–	–

* Six were tested for each treatment and six each served as controls; the latter harboured 38–78 (average: 57) *D. lamellatus* stages

Preliminary experiments for long-term treatment were performed in aquaries. The compounds were dissolved at a concentration of 1 or 0.1 ppm and experimental fishes and untreated controls were examined one day before and one day after bathing for 48 hours. As can be seen from Table II some of the fishes bathed in Gardona and Flibol E solutions still harboured viable parasites after 24 hours; all fishes bathed for 48 in ppm Gardona and Ditrifon 50 solutions were negative one day after treatment and those bathed in 0.1 ppm solutions harboured only a few *D. lamellatus* parasites.

Table II

Therapeutic results of bathing 3–5 cm long grasscarp fry* infected with 20–40 *Dactylogyrus lamellatus* parasites in organic phosphoric acid ester solutions

Time of bathing: 48 hours

Concentration of active substance in bathing solution	Time of examination after beginning of treatment (hours)	Preparations tested				
		Cryst. tri-chlor-phos	Ditri-fon 50	Nuva-nol	Car-dona	Flibol E
1 ppm	24	—	—	—	+	+
1 ppm	72	—	—	—	—	—
0.1 ppm	24	—	—	—	+	+
0.1 ppm	72	—	+	—	+	—

* Ten tested with each solution, and ten each served as controls; the latter harboured 25–38 (average: 32) *D. lamellatus* stages. Symbols used: — no parasite; + parasite(s) present

Long-term bathing trials in ponds were with Ditrifon 50 and Flibol E at a concentration of 1 ppm. The solution was poured into the pond and the water supply was stopped for two days after which it was re-started to remove the drug. This treatment managed to keep the population entirely free of parasites for a few days, but after one week masses of young *D. lamellatus* stages appeared on the gills of the hosts. From this it was concluded that the organic phosphoric acid esters did not kill the parasite eggs and thus reinfection by hatching stages began as soon as the concentration of the chemotherapeutic fell below the active level.

Aquary and field trials have shown that both NaCl and organic phosphoric acid esters are active against *D. lamellatus* when applied at an appropriate concentration and on an appropriate time schedule.

Short-term bathing in NaCl solution kills the parasites rapidly without affecting the flora and fauna of the pond, but as the fishes have to be taken from their habitat for treatment this may impose a severe, sometimes lethal stress on the host, or if performed sparingly may leave parasites unaffected. The method is in any case not simple enough for field use.

Bathing at a high (100 ppm) concentration of Ditrifon 50 also has the advantage of high activity against the parasite without damaging the host, but it too has the drawback that fishes have to be taken from the pond, and furthermore its action develops slowly, so that dying parasites removed with their hosts after treatment to a new pond may still produce viable eggs.

Long-term bathing in the pond with an organic phosphoric acid ester solution appears to be an effective and convenient method of control. Nevertheless, it does have the disadvantage of destroying the plankton, and furthermore reinfection may occur if the bathing is not repeated at the proper time(s).

Control of *D. lamellatus* infection by combined breeding technological and chemotherapeutic methods

D. lamellatus infection of various degrees of severity often becomes established in fry rearing or wintering ponds if neither a "clean" water supply nor the optimal breeding technological measures are employed. Under such conditions chemotherapeutic treatment is imperative to prevent greater losses, but it is in itself unsatisfactory for effective control. Under field conditions, successful chemotherapy is preconditioned by the elimination of adverse environmental factors: after treatment, the fishes should be removed to a new pond as free as possible from parasite larvae to prevent reinfection, and the population density should be kept at a level not favouring the spread of dactylogyrosis.

Fishes bathed in NaCl solution in the proper way and removed afterwards to a new pond are practically free of parasites, because this treatment kills several non-monogenean parasite species as well.

Field observations have shown that treatment with Ditrifon 50 or Flibol E for 48 hours is remarkably active in the face of a severe outbreak, but the parasites reappear after a few days unless the hosts are removed to a new pond. In the original pond, damage of the plankton by the phosphoric acid ester results in the reduction of the natural feed supply, and the consequent growth retardation of the population — which is usually kept at a higher than optimal density — favours parasite growth, especially that of protozoa. A recurrence of dactylogyrosis may also occur, sometimes taking a more severe course than originally, particularly if prior to treatment equilibrium had been reached between host and parasite. Treatment with phosphoric acid esters should therefore, always be followed by removal of the cured fishes to a new if possible parasite-free pond.

Short-term bathing in 100 ppm solutions of Ditrifon 50 or Flibol E appears to be a simple practicable method for field purpose, despite the disadvantage that the parasites deteriorate only after removal of the hosts to a new pond. HÁMORI (1971) reported that owing to their relatively broad therapeutic range, the above solutions are suitable for the treatment of fishes in the tank during transport inside or outside the pond farm (0.5–5 hours).

Field trials launched by the author have shown that the breeding technological and chemotherapeutic measures outlined in the foregoing provide effective control of dactylogyrosis among grasscarp fry. The same applies to chronic or semi-acute *D. lamellatus* infection of older fishes, particularly if the technological measures are combined with drug therapy. Since exposure is greatest in the densely populated wintering ponds, it is recommended that fishes be treated prior to stocking of these ponds in order to reduce the number of dactylogyrus parasites to a minimum. Since fishes do not feed during the winter, this chemotherapy does not cause nutrition biological damages. Treat-

ment in the pond is particularly effective if the NaCl or phosphoric acid ester applied against dactylogyrosis is combined with antiprotozoan and antifungal preparations.

Proper timing of the removal of the fishes from the wintering ponds is undoubtedly the most effective control measure against both chronic and acute dactylogyrosis, but if infection cannot be prevented in this way, it can be kept at a reasonably low level by treatment on one or more occasions. To ensure parasite-free population in the production ponds, fishes in the wintering pond should be treated prior to their removal or during the subsequent transport.

Summarizing the above observations, the principles of the control of grasscarp dactylogyrosis can be outlined as follows:

1. The water supply to fry-rearing ponds should be taken from a source not populated with older fishes carrying *D. lamellatus*;
2. High population density should be avoided by removal of fry to larger ponds at the proper time;
3. Antiparasitic treatment should be preferably applied prior to removal of fish to a new pond;
4. The establishment of chronic dactylogyrosis in the densely populated wintering ponds should be prevented by drug treatment prior to removal of the fishes to the winter habitat, and an outbreak occurring in the latter should be checked by treatment in the pond;
5. Prophylactic treatment should be applied if removal from wintering to production ponds is delayed.

SUMMARY

Experimental and field observations have shown that the establishment of *D. lamellatus* infection among grasscarp fry can be prevented by supplying the water from reservoir ponds not populated with older infected fishes and by removal of the growing fry to larger ponds before the population density becomes too high.

Bathing of the hosts in a 2.5% NaCl solution kills *D. lamellatus* and the organic phosphoric acid ester solutions (Ditrifon 50, Flibol E, Nuvanol, Gardona) tested for long-term action were 100% effective when applied at a concentration of 1 ppm for 48 hours. Ditrifon 50, also tested for short-term action at a concentration of 100 ppm, killed all parasites in 0.5–4 hours.

The combination of breeding technological and chemotherapeutic methods ensures effective control of grasscarp dactylogyrosis.

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