SOME ASPECTS ON THE BIOLOGY AND CONTROL OF THE ANCHOR WORM *(LERNAEA CYPRINACEAE)* IN INDONESIA

Final Technical Report

Submitted to

International Development Research Centre (IDRC)

Prepared by

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PROJECT PERSONNEL

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1. INTRODUCTION

The future development of fisheries in Indonesia will have more focus on aquaculture, which has been known in this country for centuries but still practiced on a modest level of subsistence.

Thus, in this respect considerable effort has been carried out to improve the cultivation and breeding of freshwater fish through better technology and methods.

However, it is evident that aquaculture, like other agrobusiness is subject to many biological risks and environmental constraints. Furthermore, the introduction of the improved technology is believed to have given rise to many new problems, and certain old problems such as fish parasites and diseases may become more critical.

Fish parasite infection in aquaculture is a very common occurence in Indonesia, although mostly localized and generally of no serious economic importance. This fact may be accountable to the traditional natur of the culture practice, involving low stocking density, short period of rearing, and the availability of good quality of water.

However, in the past epizootic of certain fish parasites were known to inflict serious damages to fish breeding practice, namely *Ichthyophthirius* sp and *Myxobolus* sp respectively in 1932 and 1951, both occured in West and Central Java (Sachlan, 1952).

Furthermore, Sachlan in 1952 described the occurence of about 12 fish parasites which include Ciliate, Sporozoa, Trematoda, Centodes, Acanthocephala, Nematode, Clitella and Crustacea (Appendix 1).

In the last decade, there has been an alarming indication that the damage inflicated by fish parasite and disease to fish production in this country, could be no longer under estimated or neglected.

The outbreak of a copepode parasite *Lernaea cyprinacea*, commonly known as the Anchor Worm, in 1970 exerted a nation wide damage to common carp and Java carp hatcheries. About 30 percent of fry production of the above two fish species was loss due to the outbreak of this parasite. This means that in Java a total 1.48 billion fish fry was loss during that time, equivalent to a value of 7.4 billion rupiahs. It is difficult to ascertain how much damage has been brought exactly by this parasite to fry production in the whole country during the occurence of the outbreak.

It is a wellknown fact that the existing prophylactic and curative control measures of fish parasite and disease can only be effectively and economically applicable to modern aquaculture industries. It is sometimes difficult to be carried out in traditionally extensive fish culture system such as practiced in this country.

The overall objective of the Project described in this report was to conduct a series of investigations on the problem of fish parasites affecting aquaculture, with special reference to *Lernaea*.

Within the framework of the term of reference of the Project the following scope of studies have been conducted :

(a) Studies on the epidemiological and distribution aspect of *Lernaea* and other fish parasites in fish culture area of Java and North Sumatera .

(b) Studies on the biological and ecological aspect of *Lernaea* in this country's climatic and environmental condition.

(c) Studies on the control methods of *Lernaea* in fish culture practice

During the early period of the Project (August 1976 to December 1978), the main activities were focused on the instalation of a fish parasite laboratory, survey works on the epidemiological aspect of fish parasites in Java and North Sumatera, and preliminary works on the second and third scope of studies mentioned above.

Most of the main field and laboratory works were carried out from August 1979 to February 1981.

The operation of the Project was momentarily stopped from October 1980 to January 1981, accountable to the occurence of a serious disease outbreak caused by the bacteria *Aeromonas hydrophyla*, affecting aquaculture industries in West Java, which rapidly spread through out Central and East Java.

The disease inflicted great damage to common carp culture, causing mass mortality to big size fishes and brooders. A loss of 119.4 tonnes of consumption size carp (half to one kilogram), and 56.4 tonnes of brooders was recorded in West Java, during the first eight months of the disease outbreak.

The average time alloted by the research workers for the operation of the Project is 3 to 5 man month a year.

During the course of the Project three progress reports have been submitted, respectively in August 1977, September 1979 and March 1981.

This final technical report is a compilation of the results of the studies conducted in the Project, as described in the three above mentioned reports, after several minor revisions have been made.

2. EPIDEMIOLOGICAL AND DISTRIBUTION ASPECTS

2.1. Observations on the occurence of fish parasites in freshwater fish culture areas in Java.

2.1.1. West Java.

Field survey to identify fish parasites occuring in freshwater ponds, was carried out on August 18-22, 1976, in three main important fish culture district areas, i.e. (a) Cianjur, (b) Bandung, and (c) Tasikmalaya.

(a) At the district of Cianjur, assorted samples of fish species indicate the following order of parasitic infection occuring in freshwater ponds: Monogenea (Dactylogyrus and Gyrodactylus), Trichodina, Lernaea, Myxobolus and Epystilis.
Prevalence of parasitic infection in 69 assorted fish samples indicate that Myxobolus (and other Myxosporadian parasites), Trichodina and Lernaea were the most common parasites infecting culture fish (Table 1 and 2).

Table 1. Occurence of fish parasites in fish ponds, located at four villages in the district of Cianjur.

Fish parasite			
Lernaea	carp, tilapia	rice field	Kemangmanis Sayang
	Java carp, sepat siam, nilem carp, P. binotatus	pond, cage	Sukamantri, Cijati
Trichodina	carp, kissing gouramy, nile tilapia, tilapia, nilem carp, sepat siam.	rice field pond, cage	Kemang manis, Sayang, Sukamantri
Gyrodactylus Dactylogyrus	carp, kissing gouramy, nilem carp, P.binotatus	pond, cage rice field	Kemangmanis, Sayang, Sukamantri Cijati
Myxobolus	common carp, kissing gouramy	pond, rice field	Kemangmanis, Sayang
Epystilis	P. binotatus	rice field	Kemangmanis

Occurance and prevalence of fish parasite in the sample is presented in Table 4.

Table 3. Prevalence of fish parasites on fish samples from four sub district areas in the district of Bandung

Sub district	No. of fish sample	Fish species	Prevalence (%)					
			Myxobolus	Lernaea	Trichodina	Monogenea	Epistilis	Cestodes
Ciparay	90	carp Java carp	26.7	21.1	18.9	12.2	11.0	10
Ciwidey	30	carp	83.3	20.0	50.0	3.3	3.3	-
Majalaya	30	carp	10.0	23.3	23.3	33.3	3.3	-
Bojongloa	60	carp Nile tilapia	1.7	1.7	28.3	-	-	-

Table 4. Occurence and prevalence of fish parasites found on fish samples from district area or Tasikmalaya

Sub district	No.of fish sample	Prevalence (%)				
		Trichodina	Monogenea	Myxobolus	Lernaea	Nematodes
Leuwisari	150	20.7	19.3	8.0	8.7	-
Cibereum	30	-	-	-	-	6.7
Cisayang	150	3.3	9.3	2.0	1.3	-
Singaparna	150	48.7	35.3	2.0	5.7	-

2.1.2. Central Java.

Survey on the occurence of fish parasites in fish culture area of Central and East Java was conducted on October 17-26, 1978. In Central Java fish samples were collected from four districts, representing the main fish culture centres of this province.

The composition of fish parasites infecting freshwater ponds in the survey areas, were found to be quite similar to that in West Java (Table 5 and 6). Infection of Myxobolus was observed and reported primarily in government hatcheries. Lernaea infestation caused less problem to fish farmers, relative to the latter.

At Sida Boa Hatchery located in the district of Purwokerto, fry of giant gouramy were reported to be frequently infected with Clinostomum," a parasitic digenetic trematode, which could effect human, cattle and birds. This parasite is also believed to affect the growth rate of the host fish.

Table 5. Composition of fish parasites infecting freshwater ponds located at four fish culture areas in Central Java.

Location	Number of sample	Fish species	Fish parasites
Purwokerto	46	common carp, java carp, giant gourамy, tilapia	Lernaea, Trichodina, Monogenea, Clinostomum, Myxobolus.
Banjarnegara	32	common carp, java carp, nile tilapia, giant gouramy sepat siam.	Lernaea, Monogenea, Myxobolus, Thellohanellus, Glossotella, Trichodina.
Ngrajek	23	common carp, java carp	Lernaea, Monogenea, Trichodina, Myxobolus, Nematode.
Cangkringan	30	common carp, java carp.	Lernaea, Monogenea, Trichodina, Myxobolus, Thellohanellus.

Table 6. Prevalence of fish parasite infecting culture fish at Ngrajek, Government Hatchery, Central Java.

Fish parasite	Number of sample	Number of fish infected	Prevalence (%)
Trichodina	49	18	36.7
Lernaea	49	7	14.0
Monogenea	49	8	16.3
Myxobolus	49	5	10.0
Ichthyopthirius	49	2	4

Fish species : common carp, java carp, kissing gouramy, silver carp and bighead.

2.1.3. East Java

Fish samples collected from four fish culture areas in East Java (Malang, Blitar, Kediri and Nganjuk), showed similar infestation of fish parasites as those found in West and Central Java (Table 7).
However, at the time of the sampling, no infection of Myxobolus could be detected. Lernaea was still reported to be of some problem, producing mortality rate of ten to thirty percent to fish fry and fingerlings.

Table 7. Parasites identified from fish samples collected at four districts in East Java.

District	Number of samples	Fish species	Fish parasite
Malang	35	common carp	Trichodina, Lernaea, Monogenea, Cestode, Ichthyophthirius, Epystilis.
Blitar	42	common carp, java carp, nile tilapia, giant gouramy	Lernaea, Monogenea, Trichodina, Nematode.
Kediri	11	java carp, common carp	Lernaea, Monogenea, Trichodina.
Nganjuk	30	java carp, common carp, giant gouramy	Lernaea, Monogenea, Trichodina

2.2. <u>Observations on the occurence of fish parasites in freshwater fish culture in North Sumatera.</u>

Fish culture areas in North Sumatera were known to be seriously inflicted by <u>Lernaea</u> epidemic in 1970/1971.

Surveys to conduct investigation on the occurence of fish parasites to this area were carried out on October 19-28, 1976 and December, 28-29, 1978. <u>Lernaea</u> was still regarded as a problem in hatcheries. However, according to fish farmers the rate of infection was much less and do not cause significant economical losses.
<u>Myxobolus</u> could not be found in any of the samples collected from this area. The occurence of fish parasites in four districts of this province is listed in Table 8 and 9.

Table. 8. Parasites occuring in fish ponds located at four districts in North Sumatera..

District	Fish species	Fish parasite
Deli Serdang	tilapia, common carp Lebistes, sepat siam	Lernaea, Monogenea, Saprolenia, Epystilis.
Tanah Karo	common carp	Epystilis
Simalungun	common carp, jawa carp, tilapia	Lernaea, Epystilis, Monogenea Ichthyophthirius, Trichodina
Tapanuli Utara	common carp	Lernaea, Monogenea, Trichodina.

Table 9. Occurence and prevalence of fish parasites in two district areas of North Sumatera.

Prevalence (%) District	Deli Serdang (33 samples)	Simalungun (34 samples)
Lernaea	48.5	52.9
Monogenea	24.2	52.9
Epystilis	3.0	17.8
Saprolegnea	6.1	-
Ichthyophthirius	-	11.8
Trichodina	-	14.7

3. BIOLOGICAL AND ECOLOGICAL ASPECTS

3.1. The life cycle of Lernaea cyprinacea on common carp (Cyprinus carpio) and the guppy (Lebistes reticulatus)

According to Lahav and Sarig (1964), the copepode parasite Lernaea cyprinacea needs 18 to 21 days to complete its life cycle at temperature level of 25^{0} Celcius.

Yeshouv (1959) on the other hand reported that the Anchor Worm requires only 12 to 14 days to complete its life cycle at temperature level of 27^{0} to 28^{0} Celcius.

In Indonesia the life cycle of Lernaea cyprinacea on common carp is known to require 21 to 23 days at temperature level of 24.5^{0} - 25.5^{0} Celcius (Rukyani, 1975).

The purpose of the present study is to determine the life cycle of Lernaea, and its relative time to develop on two species of fish as its hosts. The data obtained from the experiment is to be used as a basis for further study on the means of control of this parasite.

Egg sacs of Lernaea are removed from specimen collected from naturally infected common carp and the guppy. The egg sacs are then placed in pairs in petri dishes containing well water (pH 7). Each petri dish contains one pair of egg sac from one fish host. The total number of eggs obtained from Lernaea attached to common carp and the guppy are 180 and 200, respectively.

The eggs are then incubated at 26^{0} to 31^{0} C, till they hatch and develop into successive stages of nauplii and copepodid.

First copodid are placed in 40 litres tanks to be exposed to 20 test fishes, which are dipped preciouslyin 200 ppm formalin for 15 minutes. Test fishe s used are common carp (of 3-5 cm in length) and the guppy (2-3 cm in length).

Each stage of development of Lernaea at room temperature is then observed and recorded.

Lernaea eggs hatch and develop into free swimming nauplius stage. There are three stages of nauplii, after which they moulted and developed into six copepodid stages and finally into adult Lernaea. Adult female penetrates the host body, either in the skin, fin or gills and becomes parasitic.

The time required for each stage of development, with respect to two different fish species as hosts, is indicated in Table 10.

It is difficult to ascertain from the experiment, whether there is any significant variation in the life cycle pattern of Lernaea obtained from Lebistes and common carp, as their respective hosts, particularly in terms of its maturing time.

Table 10. Development of Lernaea cyprinacea obtained from two different fish species (C. carpio and L. reticulatus) as their respective hosts.

Stage of development	Time required (hours)	
	L. reticulatus	C. carpio
Egg to nauplius I	22.5	25.5
Nauplius I to nauplius III	61.5	58.5
Nauplius II to copepodid I	6	6
Copepodid I to copepodid VI	264	360
Copepodid VI to adult	48	72
Development of agg sacs	96	96
Egg to adult female Lernaea with egg sacs	498 (21 days)	594 (25 days)

3.2. The effects of water temperature on the life cycle of Lernaea.

Water temperature is known as the significant factor which influence the duration of Lernaea's life cycle; the higher temperature, the shorter period is required for the parasite to complete its life cycle. The favourable temperature range is between 14^{o}C to 32^{o}C (Nakai and Kokai, 1931). According to Putz and Bowen (1964), the optimum temperature range between 23^{o} to 30^{o}C.

Effective treatment of Lernaea with various chemicals is usually based on its life cycle. Informations of its life cycle under specific local conditions are needed. Experiments on this aspect were carried out at the laboratorium of the Inland Fisheries Research Institute and in experimental ponds, located respectively at Bogor and Garut.

A number of common carp fingerlings were placed in a series of aquaria Three different water temperatures of 25°C, 30°C and 34° were used. Temperature of 30°C and 34°C were obtained by means of a water heater apparatus (RENA Model A1). Mature Lernaea eggs were cultured in vitro and observed at the above mentioned temperatures, until the nauplius stage developed into copepodids. Common carp fingerlings were artificially infested with the first stage copepodids. Twenty to thirty infested carp fingerlings were then placed in aquaria. Three replications were afforded.

Following the above study, a field study was conducted in experimental ponds located in Garut. Fish ponds with various water temperatures, naturally obtained from hot springs are available in this area. Three fish ponds with three different temperatures of 22° - 26°, 32° -34° and 34°-38°C respectively, were used for this experiment.
In each pond three 1 x 1 x 1 cubic meters cage nets were placed. Similar to the laboratory experiment, copepodid larvae of Lernaea were obtained from room temperature culture-dishes. The common carp fingerlings were infested artificially by first stage copepodids and about 100 of the infected fish were then introduced into the net cages. They were fed daily.

Samples of larvae in both laboratories and field experiments were taken initially from the time of hatching from egg sacs, and observations on its development were made daily. Specimens of each stages were fixed in 4% formalin, for determination of morphological details in the laboratory.

The result in this study is summarized in Table 11. The data shows that the rate of Lernaea development tends to increase as the temperature raises. The duration of its life cycle, is therefore, affected by temperature.

In laboratory experiments where the temperatures were made constant at 25° C the life cycle was accomplished in 20-21 days; at 30° C it required only 17.5 days, while at 34°C the life cycle could not be completed.

Table 11. Effects of temperature on the duration of various development stages (in days) of Lernaea in laboratory and field conditions.

Development stages	Duration (in days) Laboratory			Field		
	25°C	30°C	34°C	22-25°C	32-34°C	34-38°C
Nauplius	3	2.5	2.0	3 1/	3 1/	3 1/
Copepodid	9-10	8	0 2/	11-12	0	0
Adult	8	7	0	8	0	0
Complete life cycle	20-21	17.5	NA 3/	22-23	NA	NA

1/ incubated at room temperature

2/ no copepodid or adult Lernaea were found

3/ not applicable, the life cycle could not be completed.

In field experiment the life cycle was completed in 22-23 days at 22°-26°C, while at higher temperature levels of 32° - 34°C and 34°-38°C, the life cycle of the parasite could not be completed.

At high temperature levels of 32°-34°C (in laboratory condition) and 34°-38° C (in field condition), the copepodid stage of Lernaea fils to develop into adult Lernaea. No copepodid larvae was=found on fish (presumably died) 3 days after the nauplii developed into copepodid stage.

On the other hand, at the above temperature level the fishes were found not to be affected, and no fish mortality occured.

Since Lernaea development can be inhibited by high water temperature (34°C and above), this finding may be taken into consideration as a possible alternative measure for the control of Lernaea.

3.3 Host specificity of Lernaea to six species of freshwater fish

It has been reported from field survey that a wide range of culture fish species are noted as host of the Anchor Worm.

Rukyani (1975) observed in an experimental study of the host specificity of Lernaea on three local wild fish species, i.e. the guppy, java barb and sword tail, that none of these wild fishes exhibit a clear different specificity to the parasite.

This laboratory study represents an approach towards the establishment of relative susceptibility of six fish species to Lernaea through artificial exposure of the fish to the parasite.

Five culture fish species (common carp, java carp, kissing gouramy, giant gouramy and tilapia) and one species of wild fish (the guppy), were used in this experiment.

Lernaea eggs were collected and placed in petri dishes filled with well water and incubated until the eggs hatch to obtain Lernaea larvae. Experiments on artificial infection were carried out in 40 litres glass aquaria. Thirty fingerlings from respective fish species were introduced separately and exposed to approximately 600 infective stage of Lernaea larvae per aquarium (20 copepodids per fish). Three replications were afforded for each fish species. Seven days after the infection, 10 infected fish were sampled and examined for the number of copepodid larvae attached to their bodies. When the parasite attained adult stage, the remaining experimental fishes were also examined for the number of adult Lernaea infected the fish.

The result indicated that varying degree of Lernaea infestation can be found among the fish species used in the experiment (Table 12). High mortality of experimental fish due to the infection of pathogenic bacteria or parasites and unfavourable condition in the aquaria occured during the experiment. Therefore, kissing gouramy and giant gouramy were not examined for adult Lernaea infestation since most of these fish died after the first examination. Consequently uncomplete data were obtained of the adult Lernaea infestation.

From the data obtained in this experiment, it is difficult to conclusively ascertain the relative host specificity . of Lernaea of the six speciales of fish tested.

Table 12. Incidence and intensity of larva and adult - Lernaea on six species of fish.

Fish species	Larvae			Adult		
	No fish examined	Incidence (%)	Intensity	No fish examined	Incidence (%)	Intensity
Common carp	30	97.0	6.1	35	25.7	3.2
Java carp	26	94.3	4.9	43	4.7	0.7
Kissing gouramy	21	86.1	14.0	2	-1/	-
Giant gouramy	25	95.8	3.6	3	-1/	-
Nile tilapia	25	11.6	1.0	57	0	0
Guppy	23	96.3	10.3	45	29.6	1.0

1/ not examined

However, different degree of specificity . was noted clearly between nile tilapia and other fish species. Only 11.6 percent of nile tilapia was infected, with an average of 1 copepodid larva per fish and no adult Lernaea was found. Thus, nile tilapia appears to be the leastsusceptible to Lernaea infestation.

In general, the findings in this study show that Lernaea has a wide range of host specificity on those fish species except nile tilapia. Guppy with its high number of prevalence (larva, 96,3%, 29.6%) and average number of parasite per fish (larva, 10.3; adult 1.0) can be - considered as a potential carrier of Lernaea.

Lernaea cyprinacea is still a problem in common carp culture ponds in Indonesia, but has been hardly reported in tilapia ponds. The above experiment shows that tilapia is the least susceptible host of Lernaea.

Since common carp is commonly reared with other cultured fish species it is important to investigate the possible method of reducing Lernaea infestation on common carp by polyculture with tilapia.

Three experimental ponds, 20 square meters each, were stocked with 200 common carp, 100 common carp with 100 nile tilapia and 200 nile tilapia respectively. The experimental fishes were exposed to approximately 20 infective stage of Lernaea larvae per fish in 100 liter fiber glass before stocking them into the ponds. There were three replications for each experimental pond. Samples of each fish species were examined to determine the incidence and intensity of Lernaea larvae infection.

As indicated in previous laboratory experiment, the infestation of Lernaea on nile tilapia was much lower than other species such as common carp (97% to 11.6%). In the present study, not even a single nile tilapia was found to be infected by the copepodid (Table 13). On the other hand, high incidence and intensity of copepodid were found on monoculture of common carp (91.6% and 2.5%).

The data also indicates that carp reared with nile tilapia shows lower degree of infestation relative to common carp reared as a single species. Reduction of copepodid Lernaea infestation on common carp stocked with nile tilapia was almost doubled (53.3 to 91.6% and 0.9 to 2.5).

This experiment was unable to provide a conclusive data on the adults Lernaea infestation. Heavy rain which frequently occured during the course of the experiment, caused considerable lost of the parasite when they are still at a free living larva stage. Therefore, the degree of Lernaea infestation were found much lower than expected.

Table 13. Incidence and intensity of Lernaea infestation on common carp, reared with and (or) without nile tilapia.

Fish species	Larva		adult	
	Incidence (%)	Intensity	Incidence (%)	Intensity
Common carp	91.6	2.5	1.9	1
Common carp + Nile tilapia	53.3	0.9	0	0
Tilapia	0	0	0	0

To conclude, a possible biological control of Lerneae in ponds by rearing nile tilapia together with other cultured fish species, should be considered.

3.4. Investigation on the infestation of Lernaea on common carp reared in pond versus irrigated rice field.

Rice field fish culture in Indonesia, mainly in West Java, are commonly practiced. More than 70.000 hectares of the irrigated rice field area are utilized for fish culture practices (Directorats General of Fisheries, 1973).

It is known that fish fry up to fingerling are subject to man fish parasites and more susceptible to diseases than larger fish. It is well known fact according to fish farmers that common carp fingerlings obtained from irrigated rice field are relatively healthier and larger than those obtained from culture pond.

Therefore it is a common practice of fish farmers in this country, that instead of transfering one to two weeks old fry to rearing ponds, they are introduced in irrigated rice fields and reared there for about one month before transfering them back into fish ponds to be reared up to marketable size.

However, to our knowledge, research data of fish parasite infestation in such culture system is not available so far.

An investigation was conducted to evaluate the degree of Lernaea infestation on young common carp reared in pond versus irrigated rice field. A number of artificially infested fish with Lernaea were stocked in 20 square maters earthen ponds and irrigated rice field. In this experiment, fish samples were taken, and examined to determine the incidence and intensity of Lernaea infestation.

There were six irrigated rice fields used in this experiment
The observation was carried out in two steps i.e.:

1st Fish were sampled and examined for parasties when Lernaea was still at larval stage (7 days after the fish were stocked). All fish in three ponds were then transfered into rice field and vice versa.

2nd About two weeks after the first examination, all fish were collected and held in separated tanks. Examination was done visually for adult Lernaea specimens.

The incidence and intensity of Lernaea on common carp fingerlings reared in pond versus irrigated rice field are presented in Table 14. The data indicate that both larva and adult Lernaea infestation on common carp reared in pond were higher than that reared in rice field (larva, 93% to 66,7%; 2.7 to 2.2; adult, 10.3% to 4.1% and 1.2 to 1.1).
It is also found that the incidence and intensity of the parasite on untransfered fish in pond were relatively higher than the transfered one. A slight different degree of Lernaea infestation was found between the transfered fish (pond to rice field or vice versa .

It appears that the environment of the pond seems to be more favourable for Lernaea development than irrigated rice field. By transferring the fish from pond to irrigated rice field, the degree of Lernaea infestations is apparently reduced.

Table 14. Incidence and intensity of Lernaea infestation on common carp reared in pond and irrigated rice field.

Locations	Larva		Adult	
	Incidence (%)	Intensity	Incidence(%)	Intensity
Pond	93	2.7	10.3	1.2
Rice field	66.7	2.2	4.1	1.1
Rice field to pond 1/	73.3	3.4	7.6	1.3
Pond to rice field 2/	66.7	1.9	9.3	1.1

1/ The fish transfered from rice field to pond

2/ The fish transfered from pond to rice field

4. CONTROL METHODS

4.1. Evaluation on the efficacy of filter system in preventing parasites infestation in fish ponds.

Some of the main important government hatcheries in Java and North Sumatera have been provided with up welling filter units. The primary uses of this filter system are :

(i) to assist in improvement of water quality, (ii) to prevent wild fish from entering fish pond, (iii) and to prevent the entry of certain fish pest and parasites into the pond (Djajadiredja and Martono, 1976).

They are used primarily to prevent Lernaea infestation. However, the effectiveness of this filter system as a control measure of Lernaea and other parasites in fish ponds has not been satisfactory proved through experimentations. A study on this aspect has been carried out by Hadidjah Parsono (IFRI) in 1978 (personnal communication). However, the data she obtained were not conclusive.

The present study was conducted at the IFRI Experimental Station, at Cibalagung, Bogor. Its main objective is to evaluate the efficacy of an up-welling filter system in preventing and reducing Lernaea or other parasites infestation in outdoor fish tanks.

A series of 1 x 1 x 0.3 cubic meters fiber glass containers were filled separately with filtered and unfiltered pond water. The lay-out of the experiment and the filter system is shown in Figure 1. Pond water was kept flowing through the containers at one liter per minute velocity. Each container was stocked with uninfected fry of common carp obtained from uncontaminated spawning pond. There were three replications for those filtered and unfiltered tanks. Six days after stocking, samples of fish fry were taken at every three to four days and examined for fish parasites until the fish reached fingerling size (5-6 cm) or two months old.

The degree of the parasites infestations were expressed as the percent of infected fish (prevalence) at the number of parasites per fish (intensity) (Table 15).

Table 15. Prevalence and intensity of parasites infection on fish in filtered and unfiltered water.

Parasites	Prevalence (%)		Intensity	
	Filtered	Unfiltered	Filtered	Unfiltered
1. Trichodine	62.6	83.9	20.7	59.1
2. Gyrodactylus	32.3	54.6	5.5	7.5
3. Dactylogyrus	18.4	32.5	2.7	4.0
4. Lernaea	0	11.1	0	1.7
5. Myxobolus	0	7.2	0	numerous

From a number of 337 experimental fishes in unfiltered fibre glass tank and 343 fish in filtered fibre glass tank, only five genera of fish parasites were found to infect the experimental fish i.e. Trichodina sp, Gyrodactylus sp, Dactylogyrus sp, Lernaea cyprinacea and Myxobolus sp.

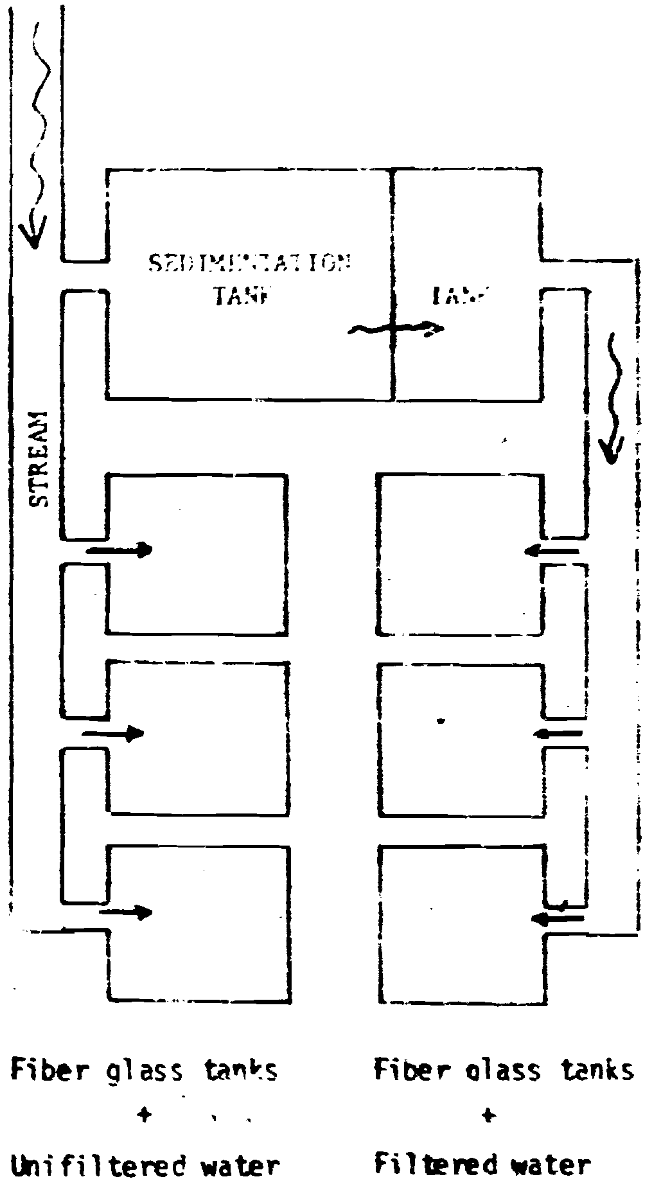
Lernaea cyprinacea and Myxobulus two major important fish parasites which commonly infect young common carp, were not found to infect the fish in filtered water. Three other parasites such as Trichodina sp., Gyrodactylus sp, and Dactylogyrus sp were found on fish either in filtered or unfiltered tanks.

However, the prevalence and the intensity of these parasites were found higher in unfiltered water, relative to the filtered water i.e. Trichodina sp. (83,9% to 62.6% and 59.1 to 20.7), Gyrodactylus sp (54.6% to 32.3% and 7.5 to 5.5) and Dactylogyrus (32.5% to 18.4% and 4.0 to 2.7). It implies that the filter reduces the number of infested fish in ponds as well as the parasites load on the fishes.

This study also suggested that the fish reared in filtered water are protected from being infected by Myxobolus sp. Although the spores size of Myxobolus are relatively smaller than any other fish parasites no spores seemed to be able to pass the filter. This is probably due to the nonactive movement which characterize the parasite, and makes it possible to be cought in the settling or sedimentation tanks of the filter unit.

Myxobolus sp which caused "swollen gill disease" on young common carp has been known recently as the most important parasite effecting nursery and hatchery ponds.

Satisfactory control measure for this parasite is at present still unknown. Therefore, the use of filter system as a possible method for controlling Myxobolus as well as Lernaea has to be considered.



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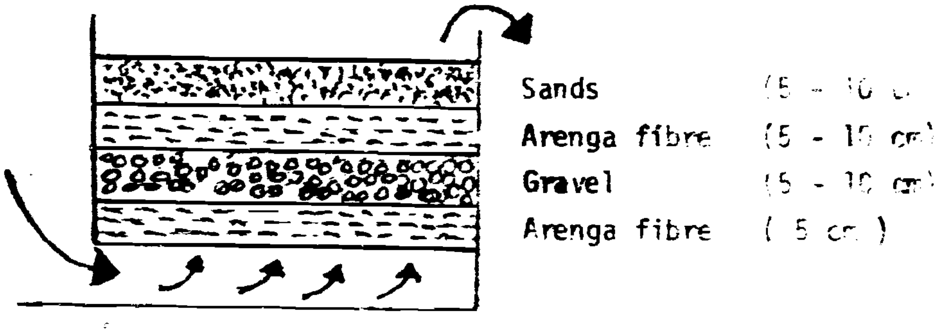

B

Figure 1. Schematie diagram of experiment (A) and filtering device showing upwelling flows (B).

4.2. <u>Evaluation of chemicals used for bath treatment to control Lernaea.</u>

The control and eradication of Lernaea commonly involve the use of various chemicals for dipping and bath treatments.

Salt solution (sodium chloride) can be used for fish species which are able to tolerate salt concentrations required to kill the parasite (Kabata, 1970). It is however of no use for adult female Lernaea attached to the fish, and effective only againsts its free swimming developmental stages.

Further Kabata stated that the only chemical capable of killing the adult Lernaea attached to fish is potassium permanganate. However, potasium permanganate does not effect young,sexually matured stages of the free swimming stages of the parasite (Kabata, 1970).

Putz and Bown (1964) reported the use of 30 to 60 minutes bath of formalin at concentration level of 250 ppm. Formalin bath of 200 ppm was used in Israel (Kabata, 1970).

Experiments were carried out in the Project to find out the range of effective concentrations of ammonium chloride, sodium chloride, potasium permanganate and formalin, against nauplius and copepodid stages of Lernaea.

A series of bioassay was conducted using one to four days old nauplii and one to three days old copepodid stages of Lernaea. Effect on the infective stage of the copepodid (four days old) was also observed, employing artificial infection on carp fingerlings.

The tests were carried out using five concentrations in logarithmic series for each chemicals, employing three replications. For each concentration a number of 30 nauplii or copepodid was tested, while in the experiment using infective stage of copepodid the test was carried out using 15 fishes per concentration.

Period of exposure adopted for the tests were 5, and 10 minutes.

Test results were reported as Effective Concentration Values (EC90) and their confidence limit interval ($p = 0.05$). The Effective Concentration is concerntration of the chemical which is effective and kill or immobilized 90 percent of test animals at a certain period. Thus, the test results were indicated as LC90 - 5 minutes and LC90 - 10 minutes.

Result of the tests were given in Table 16 and 17.

In order to determine the safe level of concentration used for bath treatment, the toxicity of the chemicals to four most important cultured fish species in this country, were also defined.

Table 16. Estimated EC90 values and their 95% Confidence Limit Interval of sodium chloride and ammonium chloride to nauplius and copepodid stage of *Lernaea*.

Chemical	Lernaea stage		Effective Concentration (EC90) in ppm	
			5 minutes	10 minutes
Sodium chloride	Nauplius	I	13000(8300-20100)	9000(4700-17100)
		II	12300(6900-21800)	
		III	12400(9500-16100)	9200(6900-12200)
	Copepodid	I	12500(1000-15600)	7500
		II	10200(8900-11700)	7500
		III	8900(7400-10700)	7500
		IV 1/	20100(16700-24200)	14000(10800-18200)
		V 1/	21000(16700-24100)	14500(10800-18200)
Ammonium chloride	Nauplius	I	8000(5900-10800)	2100
		II	7800(5700-10700)	4400
		III	4900	4000(3200-5000)
		Iv	4900	2100
	Copepodid	I	21oo	2100
		II	2100	2100
		III	2100	2100

Note : 1/ Effect on copepodid IV and V are evaluated from artificially infected carp fingerling.

Teble 17. Estimated EC90 values of formalin and potassium permanganate to nauplius and copepodid stage of Lernaea.

Chemical	Lernaea stage		Effective Concentration (EC90) in ppm.	
			5 minutes	10 minutes
Formalin	Nauplius	I	190	14o
		II	210	145
		III	220	235
Pottasium permanganate	Nauplius	I	540	380
		II	650	350
		III	650	650
	Copepodid	I	470	295
		II	300	180
		III	290	160
		IV 1/	350	225
		V 1/	360	220

Note : 1/ Effect on copepodid IV and V is evaluated from infected carp fingerling.

A series of bioassay was conducted, employing fish fingerlings of 3 to 5 cm in lenght or 1 to 2 grams in weight. The species used were carp (Cyprinus carpio), java carp (Puntius gonionotus), nilem carp (Osteochilus hasselti) and tilapia (Tilapia mossambica).

Five to seven test concentrations were employed for each chemicals, using two replications. Number of test fish per bioassay vessel are ten fishes in ten litre of test media (aliquots of chemical in water to make certain concentration). Twenty test fishes were afforded for each concentration tested.

The test were carried out for 96 hours. Test results were reported as Median Lethal Concentration values and their 95 percent confidence limit intervals, i.e. concentration which kills 50 percent of the test fishes in 96 hours exposure time (Table 18).

4.3. The efficacy of Lebaycid 550 EC as control agent for Lernaea cyprinacea.

Lebaycid 550 EC is a trade name of an insecticide, containing fenthion or O,O-Dimethyl-O (4 methyl-mercapto, 3-methyl phenyl) thiophate, as its active ingredient.
This chemical is known in the United States to be effective for Lernaea control.

The purpose of this experiment is to determine the dosage level of fenthion which is effective against Lernaea under tropical condition. Tests were performed on the effectiveness of the pesticide against Lernaea egg, nauplius and fish.

A number of mature egg sacs were exposed to a series of fenthion concentration, and observed every 6 hours for its development into nauplius stage. The number of nauplii survived was recorded, as presented in Table 19. The result of this experiment gave indications, that a concentration, level of fenthion higher than 1.5 ppm may exert certain effect to the development of the nauplius stage of Lernaea (Table 19).

A number of Lernaea nauplii, obtained from hatching Larnaea eggs in petri dishes containing well water at temperature level of 28^{o} $\pm$ 3^{o}C, were exposed to a series of fenthion concentration (ten nauplii per concentration in three replicates), for a period of six hours. Cummulative mortality of nauplii in each concentration was recorded.

Effective concentration values of fenthion against nauplius (EC50 and EC90), are respectively estimated to be 1.20 (1.05 - 1.37) ppm and 2.00 (1.60 - 2.50) ppm. (Table 20).

Static bioassays were performed to determine the lethal toxicity of fenthion to two most common culture fish species, i.e. common carp and java carp. Fingerlings of 3 to 5 cm were used as test fishes. The test was performed under the following conditions, temperature: 26^{o}C, water pH: 7.14 and alkalinity 50-60 ppm. A series of eight concentrations were used in each test. Ten fishes for each concentration were tested in 3 or 2 replications.

Table 18 : Estimated LC50 values and confidence limit interval (p = 0.05) of four chemicals to four fish species.

CHEMICAL	EXPOSURE TIME	MEDIAN LETHAL CONCENTRATION (LC50) IN PARTS PER MILLION			
		Common carp	Java carp	Nilem carp	Tilapia
SODIUM CHLORIDR	24	14500(12946-16240)	20500(13758-30545)	13800(13018-14628)	27000(26470-27540)
	48	12400(11370-13516)	15000(11278-19950)	12800(12075-13568)	26500(25980-27030)
	96	9500(7983 -11305)	12000(9600 -15000)	10500(9545 -11500)	26000(25490-26520)
AMMONIUM CHLORIDE	24	950.0(6419 - 1406	370.0(310.9-440.3)	375.0(300.0-468.8)	760.0(603.2-957.6)
	48	450.0(338.3-598.5)	273.0(237.4-314.0)	220.0(170.5-283.8)	620.0(500.0-768.8)
	96	245.0(197.6-303.8)	185.0(156.8-218.3)	167.5(131.9-212.7)	490.0(392.0-612.5)
FORMALIN	24	106.0(97.25-115.5)	8.90 (7.95 - 9.97)	16.20(12.00-21.87)	22.00(19.64-24.64)
	48	100.7(92.38-109.8)	8.40 (7.43 - 9.49)	13.50(10.89-16.74)	17.50(15.63-19.60)
	96	98.00(89.91-106.8)	7.30 (6.70 - 7.96)	12.00(10.08-14.28)	17.00(15.18-19.04)
POTASIUM PERMANGANAT	24	19.50(16.81-22.62)	5.60 (4.52 - 6.94)	4.95 (4.38 - 5.59)	6.62 (6.13 -7.15)
	48	16.75(13.62-20.60)	3.95 (3.26 - 4.78)	3.90 (3.42 - 4.45)	4.35 (3.95 -4.83)
	96	11.01(10.48-11.56)	3.00 (2.48 - 3.63)	2.90 (2.61 - 3.22)	3.00)2.61 -3.45)

The Median Lethal Concentration (LC50) for 24, 48 and 96 hours exposure time, and their 95% Confidence Limit Intervals, were obtained by means of the Litchfield Wilcoxon method (Table 21 and 22).

The toxicity of fenthion to the two fish species were found to be not significantly different.

From the result of the experiment on the efficacy of fenthion against Lernaea, the following informations can be summarized :

Effective against	Estimated level of concentration
(1) egg	higher than 1 ppm
(2) nauplius I	2.0 - 2.5 ppm
(3) fish	higher than 3 ppm

Table 19. Number of *Lernaea* nauplii surviving from eggs exposed to different concentrations of fenthion solution

Time of observation (hours)	Fenthion concentration (ppm)											
	0		0.01		0.1		1.0		1.5		2.0	
	Alive	Died	Alive	Died	Alive	Died	Alive	Died	Alive	Died	Alive	Died
30	77	0	123	0	0	0	129	0	0	0	0	0
36	77	0	123	0	137	0	129	0	0	0	0	0
42	77	0	123	0	137	0	129	0	0	0	0	0
48	77	0	108	15	95	42(?)	129	0	0	0	0	0
54	60	17(?)	78	30	95	0	88	41	0	0	0	0
60	51	9(?)	78	0	95	0	88	0	0	0	0	0
66	51	0	78	0	95	0	88	0	0	0	0	0
72	51	0	78	0	95	0	88	0	0	0	0	0

Table 20. Mortality of Lernaea nauplii and Effective Concentration values of Lebaycid 550 RC (fenthion) in 6 hours exposure time.

Concentration (ppm)	Number of nauplii	Number of nauplii died in 6 hours (%)
2.5	30	100
2.0	30	100
1.5	30	63.3
1.0	30	43.3
Control	30	0
Median Effective Concentration (EC50) in ppm formulation		1.20
95-percent Confidence Limits		1.05 - 1.37
Effective Concentration (EC90) in ppm		2.00
95-percent Confidence Limits		1.60 - 2.50

Table 21. Result of toxicity test of Lebaycid 550 EC (fenthion) to common carp.

Concentration (ppm)	Number of test fish	Number of test fish died (%)		
		24 hours	48 hours	96 hours
Control	30	0	0	0
2.4	30	0	0	0
3.2	30	16.0	23.1	46.2
3.7	30	19.8	26.4	56.1
4.2	30	23.1	33.0	69.3
4.9	30	89.2	89.2	95.7
5.6	30	100	100	100
Median Lethal Concentration (in ppm formulation)		4.1	3.8	3.6
95-percent Confidence Limit Interval		3.5 - 4.8	3.2 - 4.4	3.3 - 3.8

Table 22. Result of toxicity test of Lebaycid 550 EC (fenthion) to java carp

Concetration (ppm)	Number of test fish	Number of test fish died (%) 24 hours	48 hours	96 hours
Control	20	0	0	0
3.0	20	0	5	20
4.1	20	15	30	55
4.7	20	30	55	85
5.4	20	60	80	85
7.3	20	100	100	100
Median Lethal Concentration (in ppm formulation)		5.2	4.6	3.9
95-percent Confidence Limit Interval		4.7 - 5.7	4.2 - 5.1	3.5 - 4.3

4.4. Evaluation on the efficacy of potassium permanganate and the insecticides Orthodibrom 80E and Eumulthion TM for the treatment of Lernaea in fish ponds.

Pesticides have been widely used for large scale treatment against Lernaea in fish ponds. A good example is the use of Dipterex 90 SP, an insecticide containing trichlorfon as its active ingredient. It is known in many trade names e.g. Dylox, Masoten and others. The effectiveness of this insecticide to larval stages of Lernaea has been demonstrated by Lahav, Sarig and Shilo (1964) in Israel.

The use of ordinary chemicals such as potassium permanganate for pond treatment was also conducted experimentally in Israel (Shilo et al. 1964).

Suyanto and Koesoemadinata (1969) demonstrated that the application of the insecticides Dipterex 90 SP and Sumithion 50 EC were both effective against Lernaea infestation in ponds in West Java. This is further confirmed by Rukyani (1980) who investigated the efficacy of three formulations of insecticides i.e. Dipterex, Agrothion (an insecticide trade mark containing fenitrothion as its active ingr.dient the same as Sumithion 50 EC) and Abate.

Pond treatment is prefered in most cases because of its efficient and practical use as control measure in fish culture practice. This is particularly true in Indonesia, where fish culture is still largely conducted in traditional and extensive level, employing variety of local types of fish ponds.

Experiment on the efficacy of potassium permanganate and an organophosphate insecticide i.e. naled (Orthodibrom 80E) were carried out during the later phase of the Project.

Field trial is performed using a randomized design with five treatments and three replications. The size of each experimental pond is 50 square metres, with average water depth of 60 centimetres.

The experimental ponds were stocked with artificially infected carp fingerlings, with stocking density of 20 grams per square metre (100 fishes per pond).

Incidence and intensity of Lernaea larvae infection were recorded one day after application and at the termination of the experiment. The survival rate of experimental fishes was also recorded at the end of trial (Table 23 and 24).

The insecticide Orthodibrom 8E (naled) is found to be effective at concentration level of 0.5 ppm in pond treatment against Lernaea larvae.

The lethal effect on fish is also found to be considered not harmful, as could be seen from the number of fish recovered at the end of the trial (47,4%) which was similar as in control pond (48.4%).

On the other hand potassium permangante is found to be ineffective against Lernaea larvae at all concentration level tested as indicated from the incidence and intensity of Lernaea infection. The recovery rate of experimental fishes in ponds treated with 1.0 and 1.5 ppm suggests the susceptibility of the fishes to this chemicals.

Treatment of potassium permanganate with higher concentration level of 2.0 ppm however showed a recovery rate of 48.4% (similar to control). This irregularity was accountable probably to some technical difficulties in controlling experimental fish from natural death (predations etc).

However, the incidence of Lernaea larvae was found to be still as high as 6.94%.

Other insecticide formulation evaluated against Lernaea larvae is Eumuithion-TM, an organo phosphorus insecticide containing a mixture of two active ingradients, i.e. trichlorfon (0.0-dimethyl-l-hidroxy-2,2,2-trichloroethyl phosphonate) and azinphosmethyl (0,0-dimethyl-s-4 oxo, 1,2,3-benzotriazin- 4-gramme-l-methyl phosphorothoate).

Bioassay was conducted in laboratory to define the effective level of Eumulthion concentration against Lernaea larvae.

A series of Eumulthion-TM concentration were prepared in one litre baker glass, each containing two artificially infected carp fingerlings. A replication of three for each concentration was afforded.

The number of Lernaea larvae attached to fish body and those found in the solution after 24 hours exposure time, were recorded.

Table 23. Incidence and intensity of *Lernaea* larvae infection one day after first application of chemicals

Treatment	No. of fish sampled	Incidence (%)	Intensity
Orthodibrom 8E			
0.25 ppm	5	40.0	2.00
0.50 ppm	5	0	o
$KMNO_4$			
1.00 ppm	5	86.7	3.00
1.50 ppm	5	80.0	3.20
2.00 ppm	5	80.0	2.25
Control	5	100.0	3.70

Table 24. Recovery of experimental fish, incidence and intensity of Lernaea larvae infection at the end of the trial.

Treatment	Fish stocked	Fish recorvered number	percent	Lernaea incidence (%)	Larvae intensity
Orthodibrom 8E					
0.25 ppm	95	44	46.3	3.30	1
0.50 ppm	95	45	47.4	0	0
$KMNO_4$					
1.00 ppm	95	19	20.0	5.88	1
1.50 ppm	95	25	26.3	9.10	1
2.00 ppm	95	46	48.4	6.94	1
Control	95	46	48.4	2.17	1

Test results were indicated as Median Effective Concentration value (EC50-24 hours) and Effective Concentration 90 (EC90-24 hours) value and their 95 percent Confidence Limit Interval (Table 25).

Table 25. Mortality of Lernaea larvae and Effective Concentrations values of Emmulthion-TM in 24 hours exposure time.

Concentration	Number of test fish	Number of Lernaea larva died in 24 hours (%)
1.80 ppm	6	100
1.55 ppm	6	92.5
1.35 ppm	6	75.0
1.15 ppm	6	33.3
Control	6	0
Median Effective Concentration (in ppm formulated product)		1.20
Confidence Limit 95 percent		1.08 - 1.33
Effective Concentration 90 (EC-90) (in ppm formulated product)		1.45
95-percent Confidence Limit Interval		1.21 - 1.74

Bioassay in laboratory was also performed to define the lethal toxicity of Eumulthion-TM to java carp (Puntius gonionotus), tilapia (Tilapia mossambica) and common carp (Cyprinus carpio)

The bioassays were conducted in 50x50x30 cm fibre glass bioassay vessels containing 20 litres test media (aliquots of Eumulthion in water to make certain concentration). Ten test fishes were introduced in each vessels. At least five concentrations of Eumulthion-TM were tested in each trial.

Mortality of test fish was observed and recorded every twenty four hours for a period of 96 hours. The data were plotted against concentration in a log-probit graph paper, and evaluted according to the Litchfield and Wilcoxon method (1949), to determine the values of Median Lethal Concentration and their 95% Confidence Limit Interval, i.e. concentrations which kill 50 percent of test fish during certain period of exposure (24, 48 and 96 hours).

Similar bioassays were performed to determine the lethal toxicity of Orthodibrom (naled) to three fish species mentioned above.

The insecticide Eumulthion-TM was found to have selective toxicity against tilapia. This fact may be useful in practice in addition to its efficacy as Lernaea control agent, since tilapia is a predatorous fish which may cause damage in fry and fingerling ponds.
Test results were indicated in Table 26 - 31.

Table 26. Results of toxicity test of Eumulthion-TM to java carp

Concentration (ppm)	Number of test fish	Number of test fish died (%)		
		24 hours	48 hours	96 hours
Control	20	0	0	0
7.5	20	5	15	30
10.0	20	10	25	50
13.5	20	35	50	70
18.0	20	50	75	90
24.0	20	65	95	100
Median Lethal Concentration (ppm formulated product)		18.50	14.00	10.00
95-percent Confidence Limit Interval		15.55-22.01	11.29-17.36	8.47-11.80

Table 27. Result of toxicity test of Eumulthion-TM to tilapia

Concentration (ppm)	Number of test fish	Number of fish died (%)		
		24 hours	48 hours	96 hours
Control	20	0	0	0
0.24	20	0	5	15
0.32	20	0	20	55
0.42	20	30	55	100
0.56	20	35	80	100
0.75	20	90	95	100
Median Lethal Concentration (ppm formulated product)		0.56	0.41	0.32
95-percent Confidence Limit Interval		0.50-0.63	0.36-0.46	0.28-0.36

Table 28. Result of toxicity test of Eumulthion-TM to common carp.

Concentration (ppm)	Number of test fish	Number of fish died (%)		
		24 hour	48 hours	96 hours
Control	20	0	0	0
1.80	20	5	10	15
2.40	20	15	20	40
3.20	20	25	35	65
4.20	20	35	55	80
5.60	20	65	75	95
Median Lethal Concentration (ppm formulation)		4.80	3.80	2.70
95-percent Confidence Limit Interval		3.93-5.86	3.22-4.48	2.33-3.13

Table 29. Result of toxicity test of Orthodibrom BE to java carp

Concentration (ppm)	Number of test fish	Number of fish died (%)		
		24 hours	48 hours	96 hours
Control	20	0	0	0
2.40	20	0	5	15
2.80	20	5	40	70
3.20	20	80	95	100
4.20	20	100	100	100
Median Lethal Concentration (ppm formulation)		3.10	2.80	2.70
95-percent Confidence Limit Interval		2.98-3.22	2.69-2.91	2.60-2.81

Table 30. Result of toxicity **test** of Orthodibrom 8E to tilapia

Concentration (ppm)	Number of test fish	Number of fish died (%)		
		24 hours	48 hours	96 hours
Control	20	0	0	0
4.2	20	0	5	20
5.6	20	0	25	50
7.5	20	20	65	85
10.0	20	60	90	100
13.5	20	95	100	100
Median Lethal Concentration (ppm formulation)		9.25	6.60	5.40
95-percent Confidence Limit Interval		8.33-10.27	5.94-7.33	4.65-6.26

Teble 31. Result of toxicity test of Orthodibrom 8E to common carp

Concentration (ppm)	Number of test fish	Number of fish died (%)		
		24 hours	48 hours	96 hours
Control	20	0	0	0
2.4	20	0	0	10
3.2	20	5	15	30
4.2	20	20	35	55
5.6	20	40	60	85
7.5	20	65	85	95
Median Lethal Concentration (ppm formulation)		6.10	4.90	3.80
95-percent Confidence Limit Interval		5.30-7.01	4.26-5.63	3.30-4.37

Field observation was conducted to determine the efficacy of Eumulthion-TM and Orthodibrom 8E for the control of Lernaea larvae in carp fry ponds. The dosage of application were 2 ppm and 0.5 ppm respectively. The chemicals were applied three times at ten days interval, i.e. 4, 14 and 24 days after pond stocking.

The observation ponds were stocked with 5000 small fry each and evaluated at the end of rearing period of 40 days.

Fish yield, i.e. number of fish recovered , and incidence of Lernaea were recorded (Table. 32).

Table 32. Number of fish recovered and the incidence of Lernaea in fry ponds treated with Eumulthion-TM and Orthodibrom 8E

Treatment	Number of fry stocking	Fish recovered		Lernaea incidence (%)
		number	percentage	
Orthodibrom 8E 0.5 ppm	5000	575	11.50	0
Eumulthion-TM 2.0 ppm	5000	365	7.30	0
Control	5000	378	7.56	4.18

Table 33. Number of fish infected with adult Lernaea in fingerling ponds treated with Eumulthion-TM and Orthodibrom 8E.

Treatment	Pond No.	Number of fish stocked	Number of fish infected
Orthodibrom 8E 0.5 ppm	1	764	0
	2	387	0
	3	575	0
Eumulthion-TM 2.0 ppm	1	417	0
	2	219	0
	3	731	0
Control	1	356	20
	2	400	11
	3	756	31

The above results suggest that the insecticides Orthodibrom 8E and Eumulthion-TM are both effective against Lernaea in pond treatment.

However, the environmental effect of the use of these pesticides to fish ponds remain to be determined further.

This concern is particular to the longterm effect on fish growth and reproduction, and to the adverse effect on the population of aquatic organisms in freshwater pond which are the main source of fish food, particularly in fry and fingerling ponds in this country.

Safety factors of pesticides used for Lernaea control in the above experiments should be considered in choosing which chemical to be used to minimize their harmful effect to aquatic environment. According to Sprague (1971) recent application factor for pesticides lies between 0.1-0.01 toxic units, mostly the lower figure.

Toxic units is the ratio between actual concentration used and the lethal threshold concentration or LC50-96 hours for non persistent pesticides. Thus. 1 toxic unit = LC 96 hours, which means that half of the fish will be killed.

Table 34. Lethal toxicity of four pesticides to Java carp related to their application dosage for Lernaea control in fish ponds.

Trade name	Common name	LC50-96 hours	Application dosage in ppm	Application dosage in toxic unit
Dipterex 90SP	trichlorfon	31.0 ppm	0.5	0.016
Sumithion 50EC	fenitrothion	5.8 ppm	0.5	0.086
Orthodibrom 8E	naled	2.7 ppm	0.5	0.185
Eumulthion-TM	trichlorfon + azinphosmethyl	10.0 ppm	2.0	0.200

Dipterex 90SP (trichlorfon) was found to be potentially safe to be used in ponds relative to its dosage of application, followed by Sumithion 50EC or Agrothion 50EC (fenitrothion) (Table 30).

Eumulthion-TM may be considered as potentially safe if the dosage of application can be reduced to 1 ppm.

5. CONCLUSIONS

The most important findings and conclusive results obtained in this study are summarized as follows :

5.1. The copepode parasite Lernaea cyprinacea constitutes a latent threat to fish breeders in Java and North Sumatera.
This parasite inflicts economical damage to carp breeding practice, notably during the dry season when the availability of adequate quality and quantity of water is poor.

5.2. The life cycle of Lernaea involves three stages of nauplius which develop into six stages of copepodid and finally into adult. In this country's climatic conditions (temperature: 26°C) the life cycle is found to be completed in 21 to 25 days. Water temperature is known to effect the duration of the life cycle, both in laboratory and field conditions. Water temperature higher than 34°C is found to inhibit the life cycle of Lernaea. This fact may be further applied under certain condition, as a mean of control of this parasite.

5.3. It has been demonstrated that Lernaea has a wide host specificity among important culture fish species, including common carp, java carp, kissing gouramy, giant gouramy and nile tilapia. Nile tilapia is found to be the least susceptible to Lernaea infection. On the other hand, common carp is highly susceptible to this parasite. It may be suggested that under certain condition where Lernaea infestation is found to be difficult to control, a polyculture practice involving nile tilapia should be carried out, in order to reduce the intensity and the prevalence of the parasite infection. Some wild fishes, notably the guppy (Lebistes reticulatus), is found to be highly susceptible to Lernaea infection, and potentially can act as a carrier of this parasite.

5.4. Lernaea infestation on common carp fingerling reared in irrigated rice field is found lower, both in prevalence and intensity, relative to those reared in ponds. Irrigated rice fields are most suitable for fry rearing in fish culture areas where fish parasites infestation vide expertise and training to local staff . It will also be necessary to improve research facilities to facilitate the work in the field of bacterial and viral fish diseases.

6. RECOMMENDATIONS.

There is an immediate need to develop and establish a sound and effective fish health control management to protect aquaculture practice in this country.

The programme should essentially involves water management for disease control, prophylaxis and therapy measures, and regulations on the transportation and distribution of live fish, with special reference to fish seed.

Water management control is very important to maintain the credibility of fish pond condition for the production of healthy fish. This involves the management of the sites of water source, water supply and the pond itself.

Prophylaxis and therapy measures for the control of fish parasites and diseases, are largely known and should be applied as a routine procedure in the management of the fish culture industries.

There is a great need for regulations on the traffic of live fish to prevent the introduction of communicable diseases and parasites to aquaculture practices in this country.

It has been pointed out in the Workshop on Tropical Fish Diseases held in Cisarua, Bogor, Indonesia 28 November 1 Desember 1978, that the immediate priority on the aspect of regulation on the traffic of live fish is the establishment of quarantine and health certificate issuance procedures at national and international levels.

Control regulations at the national level (i.e. inter provincial or interinsular levels) should be executed through convention among local government There should be the establishment of a certificate of health, place of issue and the agency or agencies authorized to issue the certificate. It might be more efficient, less expensive and more practical to issue certification of health to private and governmental aquaculture industries, notably hatcheries, which act as fish suppliers and/or distributors. The health certificate should be issued based on the result of periodical examination and evaluation of fish health at the named Aquaculture industries or hatcheries, executed by governmental officials acting as Fish Health Controller.

There is also a great need for the establishment of standard diagnostic techniques or procedures for the examination of fish health. Initial preparation for this kind of standard diganostic techniques and procedures has been carried out by the Research Institute for Inland Fisheries.

Likewise, there is a need the establishment of effective and non expensive treatment procedures for transported fish.
It is recommended that transported fish should be treated for diseases and parasites, both at the place of origin or exit and at the place of recepient or entree.

To conclude, it can never be emphasized too much, that attention mustbe given to further research and training programme on fish diseases in this country.

Further implementation on research activities should be look for through technical assistance, to improve the capability of local counterpart research staffs and to facilitate research works.

Training should be provided to extension workers on fish parasites and diseases control, in order to improve their capabilities in dealing with routine local problems at their respective fish culture areas.

BIBLIOGRAPHY

Baudouin, M. 1905. Nouvelles observation sur le Lernaeanicus. Copepoda parasite de la sardine. Bulletine du Museum Nationale d'Histoire Naturalles. 1:299-300.

Basset, Smith, P.W. 1899. A systematic description of parasitic Copepoda found on fishes, with an enumeration of the known species. Proceeding of the Zoological Society of London. 2 : 438 - 507.

Boxshall, G.A. 1976. The host specificity of Lepeophtherius pectoralis (Muller, 1976) (Copepoda : Caligidae). J. Fish. Biol. 8 (3) : 255 - 264.

______________. 1976. A new genus on two new species of copepod parasitic on freshwater fishes. Mascellanis (Bulletin of the British Museum (Natural History - Zoology). 30 (6) : 209 - 215.

Cressey, R.F. and Collette, B.B. 1971. Copepods and needle fishes : a study in host-parasite relationship. Fishery Bulletin 68 (3) : 347 - 432.

Djajadiredja, R. and Martono. 1976. Filtration and sedimentation tanks of waste-water. IFRI. Contribution No. 1 : 5 pp, (in Indonesia).

Enders, H,E. and S.A. Rifenburg. 1929. A preminary report on Lernaea copepod parasitic on the gold fish. Proceeding of Indian Academy of Science. 38 : 333 - 334.

Fernando, C.H. and H. Hanek. 1973. Some parasitic copepoda from Sri Lanka (Ceylon) with a synopsis of parasitic Crustaces from Ceylonese fresh-water fishes. Bulletin of Fisheries Research Station Srilanka (Ceylon). 24 (1/2) : 63 - 67.

Fryer, G. 1958. A note on Lernaea bisticornis Harding, a parasitic copepod from lake Tanganyika. Reveu de Zoologie et de Botanique Africaines. 58 (3-4): 214 - 216.

Fryer, G. 1968. The parasitic Crustacea of African Freshwater Fishes; Their biology and distribution. Journal of Zoology Proceeding of Zoological Society of London. 156 : 45 - 95.

Grabda, J. 1963. Life cycle and morphogenesis of Lernaea cyprinacea L. Acta Parasitologica. Vol XI : 169 - 199.

Harding, J.P. 1950. On some species of Lernaea Crustacea. copepoda: Parasites on Freshwater fishes. Bulletin of the British Museum (Natural History) Zoology. 1:1-7.

________. and A.C. Wheeler, 1958. Heavy infestation by the parsitic Lernaeanicus of sprats in the river Crouch. Nature, 182 (4634) : 542 - 543.

Ho, Ju-Shayand Masahiro Dojiri. 1976. Parasitic copepods on the fishes of the Great Barrier Reef, Australia. Part I. Cyclopoda. Publication on Seto the Marine Biological Laboratory. 23 (3/5) : 257 - 273.

Hu, Yung-Tsu. 1946. Studies on the parasitic copepods of China. Part 3. The Far Eastern Allies of Lernaea cyprinacea L. with a desrciption of two new subspecies on L. rhodei sp. nov. Sinensia. 29 (1-6) : 86-98.

Izawa, K. 1975. A new and a known chonracanthid copepods parasitic on fishes from Tanabe Bay. Annotations Zoologicae Japonenses. 48 (2) : 108 - 118.

Johnson, S.K. 1969. Sodium Hypochlorite : Use on parasitic copepoda for Identification. Repr. : Trans.Amer.Micros. Soc. 88 (4) : 591 - 592.

Kabata, Z. 1964. Copepoda parasitic on Australian fishes. II. Mappates alter sp. nov. (Caligidae). Ann.Mag. Nat.Hist. 13 (7) : 641 - 649.

___________.1965. Copepoda parasitic on Australian fishes. III. Genera Dentrigrips, Henocophilus and Pseudanuretes (Caligidae) Ann.Mag.Nat.Hist. 13 (8) : 19-31.

________. 1965. Copepoda parasitic on Australian fishes. IV. Genus Caligus (Caligidae). Ann.Mag.Nat.Hist. 13 - (8) : 109 - 126.

Kabata, Z. 1966. Copepoda parasitic on Australian fishes. V.Genus Dissonus (Dissonidae). Ann.Mag.Nat.Hist, 13 (9): 211 - 226.

________. 1966. Copepoda parasitic on Australian fishes. VI.Some Caligoid species. Ann.Mag.Nat.Hist. 13 (9) : 563 - 570.

________. 1967. The gunes Haemobaphes (Copepoda Lernaeo caridae) in the waters of British Columbia. Canadian Journal of Zoology. 45 : 853 - 875.

________. 1968. Two species of Caligus (Caligidae) from New Caledonia Crustaceana, Supp. 1 : 1 - 10.

________. 1968. Copepoda parasitic on Australian fishes. VIII. Shiinoa occlusa. gen.et.sp.nov. J.Nat.Hist. 2 : 497 - 504.

________. 1969. Copepoda parasitic on Australian fishes. IX. Family Chondrocanthidae. J.Nat.Hist. 3: 497 - 507.

________. 1970. Crustacea as enemies of fishes. T.F.H. Publication London.

Kirtisinghe, P. 1934. Parasitic copepoda of Ceylon. Spolia Zeylanica, 18 (2) : 269.

Lahav, M; and S. Sarig. 1964. Observation on the biology of Lernaea cyprinacea L. in fish ponds in Israel. Bamidgeh, 16 (3) : 77 - 86.

________ and M. Shilo. 1964. The eradication of Lernaea in storage ponds of carps through Destruction of the copepodidal stage by Dipterex. Bamidgeh, 16 (3): 86 - 94.

Leigh-Sharpe, W.H. Comensal and parasitic Copepoda. Siboga-Expeditie H. 123 (Mon. 29b). 123 p.

Markewitsch, H.P. 1936. Cardiodectes hardenbergi In never parasitischer Copepode aus der Java see. Treubia, 15:407-411.

________________. 1956. Parasitic copepodes on the fishes of the USSR, Kiev, 259 pp.

Matsui, Y. and Asao Kumada. 1928. Studies on fish disease I. "I Kari-Leigh-Sharpe, a new parasitic copepod of Japanese eel. Journal of the Imperial Fisheries Institute Tokyo, 24(4):101-107.

Mayer, F.P. 1966. A new control for the parasites Lernaea cyprinaces L. The Progressive Fish Culturist, Jan. 33-39.

Nakai, N. 1927.. On the development of a parasitic Lernaea Elegans Leigh-Sharpe, infesting on Cyprinus carpio L. Journal of Imperial Fisheries Institute Tokyo, 23(3):39-59.

__________. and E. Kokai. 1931. On the biological study of a parasitic Copepod Lernaea elegans Leigh-Sharp, infesting on Japanese freshwater. J.Imp.Fish.Exp. Tokyo, 2:93-128.

Okada, Y.K. 1927. Copepode parasite des amphibiens, nouveau parasitisme de Lernaea cyprinacea L. Annotationes Zoologicae Japonensis, 11(2):185-187.

Partasasmita, S. and S. Koesoemadinata. 1978. The life cycle of Lernaea cyprinacea L. on Lebistes reticulatus and Cyprinus carpio. The first Workshop on Tropical Fish Diseases, Bogor, Indonesia, 6 pp.

Pilai, K.N. 1964. Miscellaneous collection of copepods parasitic on South Indian Fishes. Journal of the Marine Biological Association of India, 6(1):61-83.

Putz, R.E. and J.T. Bowan. 1964. Parasites of freshwater fishes IV. Miscellaneous. The anchor worm (Lernaea cyprinacea) and related species. US Bureau of Sport Fisheries and Wildlife, Fishery Leaflet. (575):4.

Rogers, W.A. 1968. The biology and control of the Anchor worm Lernaea cyprinacea L. FAO Fish.Rep. 5(44):393-398.

Rukyani, Akhmad. 1975. Some aspects on the biology of *Lernaea* sp.: The identification, life cycle and effect of organic manure on the degree of *Lernaea* infestation in relation to the total length on common carp *Cyprinus carpio*. Thesis, Lambung Mangkurat University, Banjar Baru. 72 pp. (in Indonesian).

Rukyani, Akhmad. 1980. The use of Dipterex, Agrothion and Abate for control of the anchor worm *Lernaea cyprinacea* L. Thesis, Departement of fisheries Auburn University. . Alabama USA : 23 pp.

Sanderson. J.M. 1974. A new occurence of the parasitic copepod *Lernaea cyprinacea* L. J.Fish.Biol., 6(1):77-78.

Schaperclaus, W. 1954. Fischkrankheiten. Berlin. 489 p.

Sachlan, M. 1952. Notes on parasites of freshwater in Indonesia. Indonesia, Jakarta-Bogor. Contr.Ini.Fish.Res.Sta. No. 2:1-59.

Shen, C.L. 1957. Parasitic copepods from fishes of Chine, part I. Cyclopoda (I). Acta Zoologica Sinica, 9(4):314-327.

Shields, R.J. and W.M. Tidd. 1968. Effect of temperature on the development of larval and transformed females of *Lernaea cyprinacea* L. (Lernaeidae). Crustacean International Journal of Crustacean Research Supplement I. Studies on Copepoda. Leiden. 87-95 pp.

Shilo, M. Sarig. S. Shilo, M and Zeev, H. 1955. Control of *Prymnesium* parvum in fish ponds with the aid of Copper sulfate. Bamidgeh, 6(3):99-102.

Sproston, N.G., W.J. Yin and Y.T. Hu. 1950. The genus *Lamprolegna* (Copepoda parasitica). The discovery of the life histories and males of two Chinese species from food fishes, revealing their relationship with *Lernaea* and of both to the Cyclopoidae. Sinensia, Shanghai (n.s) 1:51-84.

Soejanto, S.R. 1965. On the occurence of new species of parasitic copepod of the genus Lernaea on a freshwater fish in Indonesia. Treubia, 26(4):207-209.

__________. and Santosa Koesoemadinata. 1971. Treatment and prevention measures of Lernaea of fish (in Indonesian. Pemberitaan Lembaga Penelitian Perikanan Darat No. 4.

Sprague, J.B. 1971. Measurement of pollutant toxicity to fish. Rev. Paper III. Sublethal effect and "safe" concentration. Water Research, 5:p. 245-266.

Tidd. W.M. 1931. A list of parasitic copepods and their fish host from lake Eire. Ohio Journal of Science, 31(16): 453-454.

__________. 1933. A new species of Lernaea (parasitic copepoda) from the gold fish. Ohio Journal of Science, 33(6): 453-454.

Velasquez, C.C. 1951. An unreported Philippines species of parasite copepod. Natural and Applied Science Bulletin, 11(3/4) : 243-255.

Vervoort, W. and F. Remirez. 1968. Parambobochus glociceps nov. spec. (Copepoda, Gyclopida) from the gills of (Austroatherina smitti (Lahille) (Pisces, Atherinide). Zoologische Model ingen, 43(-1):k4k-154.

Wilson, C.B. 1910. The classification of the Copepoda. Zoologische Anzeiger, 35(20):600-620.

__________. 1944. Parasitic Copepods in the United States National Museum Proceeding of the United State National Musium, 94:529-582.

Yamaguti, S. and Yamasu, T. 1960. Two new species of Copepods parasitic on Japanese fishes. Publication of Seto Marine Biological Laboratory, 8(1):137-141.

________________________. 1959. Parasitic copepods from fishes of Japan, species., Biological Journal of Okoyama University, 5(3-4):89-165.

Yashouv, A. 1959. On the biology of Lernaea in Fish ponds. Bamidgeh, 4(11):80-89.

APPENDIX 1

Common Parasites of Freshwater Fishes in Indonesia and Their Occurences 1/

Class	Name of parasite	Remarks
Ciliata	Ichthyophthirius multifiliis	First encountered in Bogor in 1928, Probably "imported" from Eurepe and the U.S. Caused a serious epidemic in 1932.
Ciliata	Cyclochaeta domerqui	First encountered on Lebistes reticulatus in November 1951. Later also found on Irichogaster pectoralis. No serious epidemic occurence so far.
Sporozoa	Myxobolus sp.	Thousand of Puntius gonionotus fry died et the government-operated hatchery in Central Java in 1951. However, on the whole, epidemics are rare.
Trematode	Dactylogyrus sp.	Serious epidemic has never been reported Infections were found on Puntius sp., Helostoma temmincki, Irichogaster trichopterus, Osphronemus goramy and Clarias batrachus.
Trematode	Gyrodactylus sp.	Occurence of this parasite has been reported on variety of common carp in West Java.
Trematoda	Distomidae	Found in the guts of various Indonesian fish without any symptoms of distrees. Epidemics reported in 1952 from different locations of Sumatera and East Java.
Cestoda	Tetrarhychidae	In November 1951 some people in South Kalimantan were reported to have died after Consumption of diseased Ophiocephalus
Hirudinea	Piscicola sp.	A number of fish died in South Sulawesi, presumably because of this parasite, but has not been confinmed.
Crustacea	Argulus indicus	Very common in cultured species, espesially on Trichogaster pactoralis, Helostoma temmincki, Anabas testudineus and Osphronemus goramy. No. serious epidemic has ever been recorded. It has been ascertained that infected fishes do not ordinarily die.
Crustacea	Lernaea cyprinacea	Caused serious epidemic in Java, South Sulawesi and North Sumatera in 1971. Still a serious threat to hatcheries, affecting most species.