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The Situation of Freshwater Fish Disease Epidemic and Sanitary in Thailand

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The Situation of Freshwater Fish Disease Epidemic and Sanitary in Thailand [1]

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Summary

There are many freshwater fish farms in every parts of Thailand, but mainly in the central part. Catfish *(Clarias batrachus)* amd snakehead fish *(Ophicephalus striatus)* are the most important economic species. The serious freshwater fish disease epidemic has been occurred from 1980 to 1985. This condition was firstly found in the southern part and then spread to all freshwater fish of the country. The ulceration is the prominent lesion. Low water quality, particularly sudden drop of temperature in winter, is supposed to be an important predisposing cause. Several pathogens have been isolated; bacteria, particularly *Aeromonas hydrophila*, fungi *(Achlya spp.)*, ecto-and endoparasites. Virus particle has been found by electorn microscopy. The primary cause of this ulcerative disease remains uncertain and should become the subject of an intensive investigation. Bacteria from the moribund fishes are destroyed after passing the process of salted and these salted fishes can be consumpted after cooking.

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Introduction

Aquaculture is one of the main profession for Thai people. In 1983, there were more than thirty-five thousand freshwater fish farms, cover about ninety-three thousand acres (Fisheries record of Thailand, 1983). There are about fourteen species of economic freshwater fish have been cultured. The main carnivorous fishes are snakehead fish *(Ophicephalus striatus)* and walking catfish *(Clarias batrachus)*. These two species are culture mainly in earthen pond and fed with the mixture of trash fish and rice bran. The main source of water supply to the pond is from rivers and irrigation canal. The density of fish in each pond is not certain depend on the farmer. In pond capacity eight hundred cubic metre of water can produce snakehead fish nearly fifteen tons in one year.

The disease has played an important role for many years ago mainly in cultured fish. Saitanu, *et al.* (1979) have isolated *Aeromonas hydrophila* from sand goby *(Oxyeleotris marmoratus)*. This organism has been found in many species of fish; *Clarias batrachus, Oxyeleotris marmoratus, Ophicephalus striatus* and *Fluta alba* (Saitanu and Poonsuk, 1980). A serious epizootic of freshwater fish with ulcerative lesion, both in cultured and wild fish occurred during the end of 1982 to the beginning of 1983. This condition was first observed in the south the Thailand in 1980 and has since spread to all fresh water fish of the country in the following year. In addition, it is now know to occur among populations of fish in Laos, Cambodia and Burma. Economic losses in 1983 were calculated to be 105 million Baht (4.5 million U.S. dollars) (Bhukaswan, 1983). In 1984-1985, this type of epizootic was reported again from many parts of the country, particlarly northern and north-eastern part. The lesions observed were exactly the same as reported previously. Sudden drop of water temperature, poor water quality, bacteria, virus, parasites, herbicide, and pesticide were thought

to be related to disease outbreaks.

The cause of this ulcerative disease remains uncertain and should become the subject of an intensive investigation. This paper reports the present knowledge about the epidemiology, clinicopathology, etiology, therapeutic, control and hygienc of this fish epidemic.

I. Epidemiological studies

1. *Geographic distribution* (Figure 1)

From questionnaire, this disease was firstly reported in the south of Thailand in 1980 at Narativaj and Pattalung provinces. In 1981, it was reported at the Nakornsritammaraj, Pattani and Yala provinces. A serious epidemic was occurred during the end of 1982 to the begining of 1983 mainly at the north-southern part, central and Eastern part of country. This disease still occurred in these regions at the end of 1983. In 1984-1985, the disease spread to all the country but the high mortality was observed at the northern and north-eastern parts. The lesions were same observed from the previous year (Tantrongpiros, *et. al.* 1984).

The serious fish epidemic occurred similar to the other countries of the world. Baudouy and Tuffery (1973) reported the high mortality in middle European salmonids rivers particularly in France. In March 1972, red-spot disease was reported in Australia (Rodgers and Burke, 1981). In Indonesia, Djajadiredja *et al.* (1982) reported the disease outbreak in October 1980.

2. *Species affected* (Table 1)

Twenty-eight species of freshwater and two estuarine fishes, both wild and cultured operation were affected during an epidemic in 1982-1983. Boonyaratpalin *et al.* (1983) reported that 42 species of fishes, shrimps and snails from 25 provinces had necrotic lesions on the upper and lower jaws and on other parts. During an epidemic in 1984, Wongratana (1985) found that only 27 species of fish were affected.

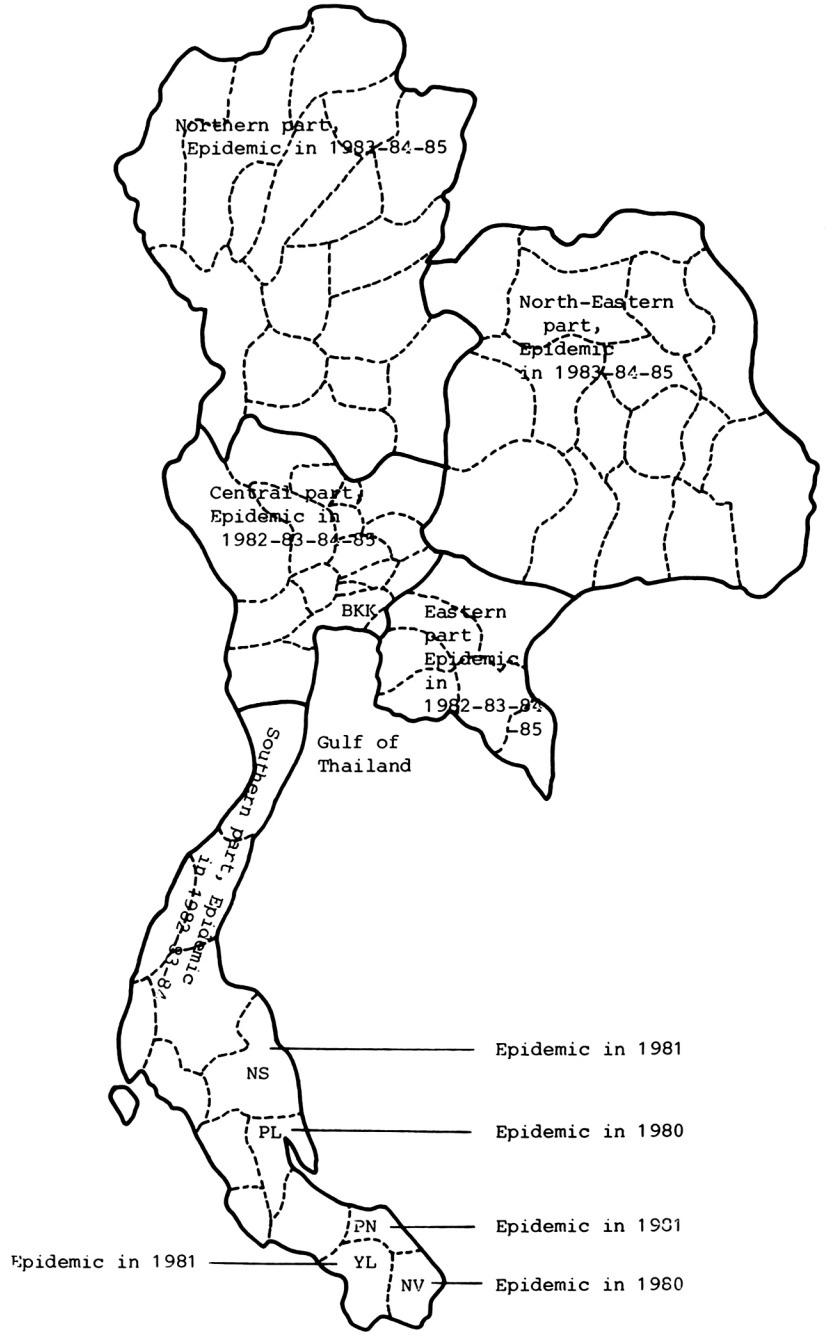


Figure 1 Geographic distribution of fish epidemic from 1980 - 1985

BKK = Bangkok, NS = Nakornsritammaraj, NV = Narativaj

PL = Pattalung, PN = Pattani, YL = Yala

Table 1 List of affected fishes during an epidemic in 1982-1983.
(- = none culutre)

No	Scientific Name	Common Name	Cultured fish	Wild fish
1	Ophicephalus striatus	Snakehead fish	x	x
2	Clarias batrachus	Walking catfish	x	x
3	Oxyeleotris marmoratus	Sand goby	x	x
4	Puntius gonionotus	Carp	x	x
5	Tilapia nilotica	Nile perch	x	x
6	Trichogaster pectoralis	Snake skin Gourami	x	x
7	Pangasius sutchi	Stripped Catfish	x	x
8	Cyprinus carpio	Comman carp	x	x
9	Lates calcarifer	Giant perch	x	x
10	Fluta alba	Swamp eel	-	x
11	Macroganthus aculeatus	Spiny eel	-	x
12	Trichogaster microlepis	Moonbeam Gourami	-	x
13	Morulius chrysophekadion	Black shark	-	x
14	Labiogarbus spp.	Minnow	-	x
15	Anabas testudineus	Walking Fish Climbing Perch	-	x
16	Notopterus chitala	Featherback fish	-	x
17	Ophicephalus micropeltis	Serpent-head fish	-	x
18	Rasbora spp.	-	-	x
19	Mastacembelus armatus	Spiny eel	-	x
20	Trichopsis vittalus	Croaking gourami	-	x
21	Osphronemus goramy	Giant gourami	-	x
22	Tachysurus sciurus	Catfish	-	x
23	Mystus wyckii	Catfish	-	x
24	Demogenys pusillus	Siamese halfbeak	-	x
25	Helostma temmineki	Kissing gourami	-	x
26	Pristolepis fasciatus	The stripped tiger	-	x
27	Mystus cavasius	Catfish	-	x
28	Mugil spp	Mullet	-	x
29	Labeo rohita	Rohu	-	x
30	Puntius orphoides	Red cheek barb	-	x

3. *Environmental studies*

It is clear that the epidemic occurs in the winter season or during the cooler months in Thailand (November-February). The temperature of water was declined more than 10^{o}C. from the normal in some regions. It is well known that this stress diminish the production of antibody in fish. Dorson (1984) reviewed the optimum temperature and other factors in antibody production in fish. He cited that carps could not produce the antibody against bovine serum albumin at 12^{o}C. but it will be produced at 25^{o}C.

As regard to the other environmental parameters, Tamiyavanich (1983) found the high level of total ammonia and low dissolved oxygen (DO) in the pond. This low DO might not be quite effect to catfish and snakchead fish because these two species of fish have the accessory organ. Amorasit *et al.* (1983) analysed the herbicide, pesticide, heavy metals from the water and fish organs. They concluded that herbicide, pesticide and heavy metals might not be the direct effect of this fish epidemic Menasveta *et al.* (1983) showed that paraquat at 1.5-2.5 ppm did not do any harm to three species of freshwater fish i.e. *Poecilia reticulata, Tilapia nilotica* and *Ophicephalus striatus* after 14 days exposure. The exposure of snakehead fish to 1.0 ppm paraquat in combination with *Aeromonas hydrophila* at 10^{6} cells/ml for seven days did not result in the infection (Sihanonth *et al.*, 1983). Paraquat at 2.0 ppm and 3.0 ppm did not shown any significant histological change except at 4.0 ppm showed swelling of gill filaments (Kanchanopas, 1984; Sinhaseni and Tesprateep, 1985).

II. Clinico - pathological studies

1. *Symptom of diseased fish*

Fish, particularly cultured snakehead fish, showed inappetite and slowly movement, sometime it floated. In some pond, high mortality was developed in one week (Fig. 2). The external lesions; in early stage, the petechial hemorrhage and erythema were observed (Fig. 3).

Figure 2. Numbers of dead snake-head fish float at the corner of the pond (D)

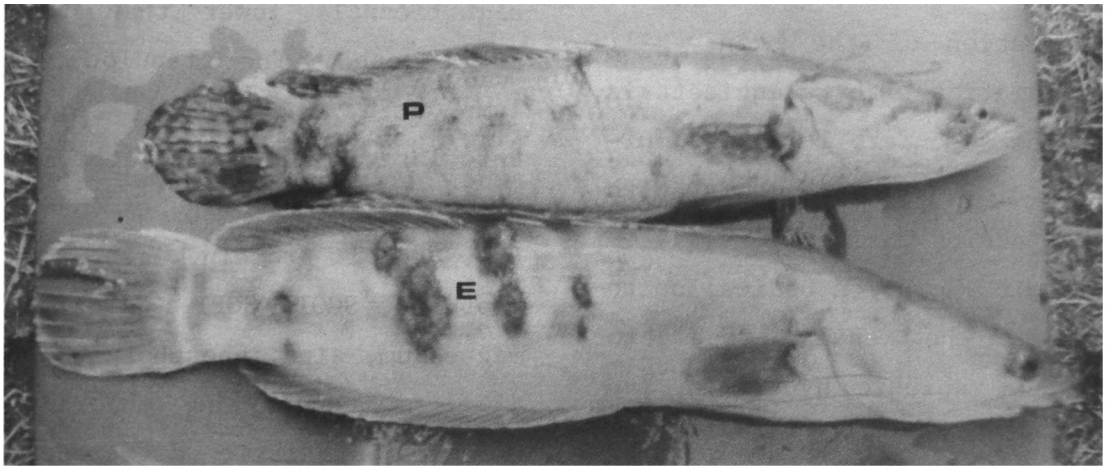


Figure 3. The early stage of disease show petechial hemorrhage (P) and erythema (E).

In chronic lesion, the small to large ulceration had been developed in some part of the body. Some fish lost the upper or lower jaw or peduncle (Fig.4,5) but it can be survived.

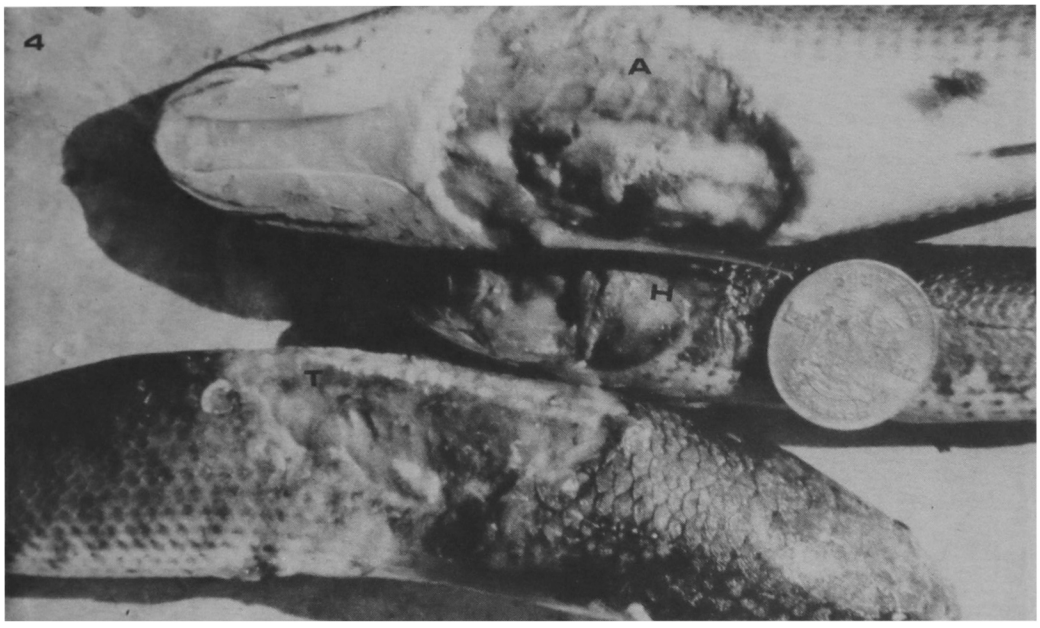
The internal organs looked macroscopicly normal.

2. *Histopathological lesions* (Fig. 6, 7, 8, 9, 10,11)

During an epidemic, the organs from moribund fishes were studied in histopathology by Tesprateep *et al.* (1983). They found Zenker's necrosis and myositis of the skeletal muscle, ulceration of the skin, gill inflamation with telangiectasis of the secondary lemellae, multifocal hepatic necrosis with presence of eosinophilic cytoplasmic inclusion bodies, necrosis of the renal hemopoietic tissue, splenic hyperemia and lymphoid necrosis, myocarditis and catarrhal gastroenteritis. In 1985, they confirmed their previous study by classification based on external lesions; early pinpoint, acute, chronic ulcerative, healed and control groups. They found the variable skin lesion and necrosis of gill lamellae in all groups except the control. The pathobiology of the disease was possibly linked to bacterial toxemia and other external factors. The lesion started from the skin, spread to the underlying musculature, and finally involved the systemic organs.

3. *Hematological values*

During and epidemic in 1982 and 1984 Tangtrongpiros *et al.*, (1983, 1985) found that the hemoglobin of infected fish was significantly lower than the normal ones. The granulocyte, lymphocyte, neutrophil, erythroblast, granuloblast, thrombocyte met-hemoglobin, glucose, BUN, total protein, albumin, globulin, SGOT, SGPT, calcium, magnesium, zinc, phosphorus, sodium, potassium and chloride were not statistic difference between normal and infected group. Gel electrophoresis revealed that serum protein from normal fish indicated 5 or 6 major bands. However, in the



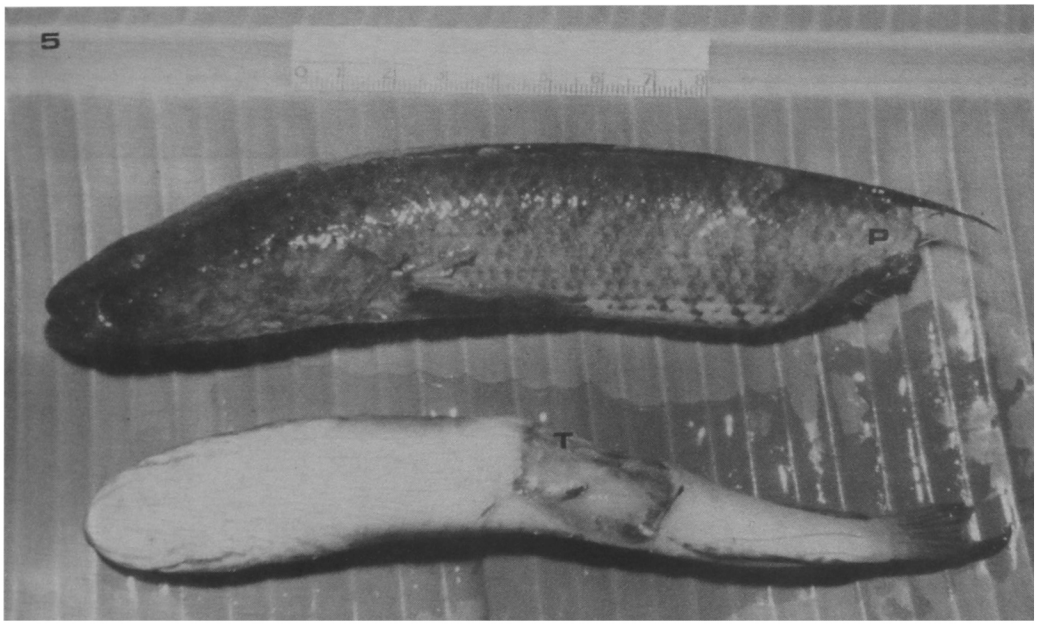


Figure 4,5 The large number of ulcer were developed at the abdomen (A), head (H), trunk (T), peduncle (P).

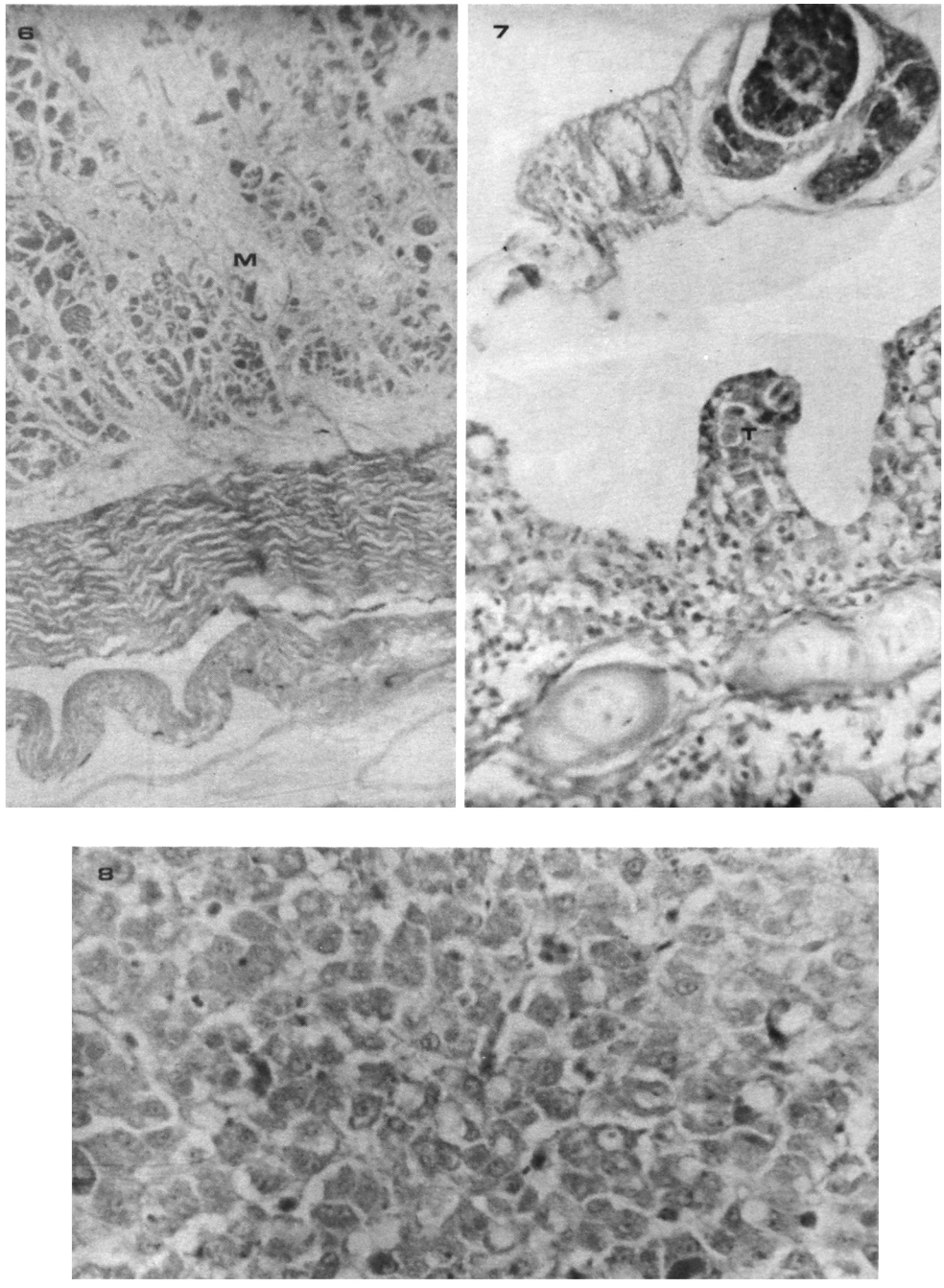


Figure 6,7,8 Show histopathological change : Muscle necrosis (M), telengiectasis (T) of the second lamellae and cytoplasmic inclusion bodies in hepatic cells (H) (Photo by Dr. Tesprateep)

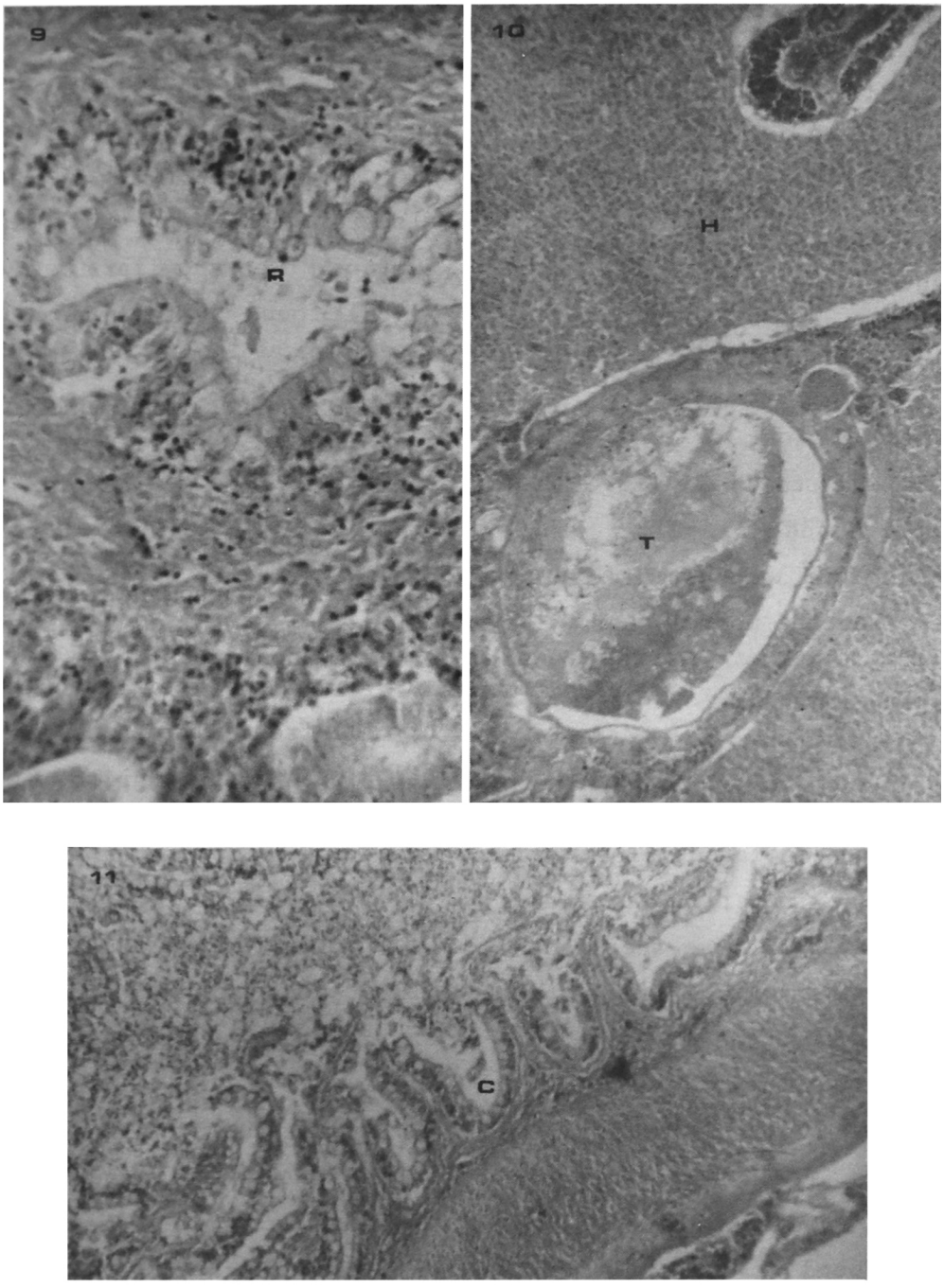


Figure 9,10,11 Shows necrosis of renal tissue (R), splenic hyperemia (H) and thrombus mass in splenic vessels (T), catarrhal enteritis (C) (Photo by Dr. Tesprateep)

severely infected fish the 4^{th} band was faded and the 5^{th} and/or the 6^{th} band was clearly stained. No significant changes in electrophoretic patterns were observed in the sera from slightly infected fish or from those which recovered from the infection (Chaisiri *et al.*, 1985).

Tosukhowong *et al.*, (1985) found also that the levels of vitamin A and C in liver of infected group were significantly lower than the normal ones.

III. Etiological finding

Many causative agents of this ulcerative lesion disease have been isolated. The primary cause is still intensive investigated because the disease is very complicated. Among these disease agents, *Aeromonas hydrophila*, an hemolytic bacteria, was commonly found during the outbreaks, (Poonsuk *et al.*, 1983, 1985). Boonyarataplin *et al.* (1983) found also the *A. hydrophila, A. punctata, Flavobacterium spp., Pseudomonas Pseudomonas sp., Edwardsiella trada, Vibrio parahaemolyticus* and *streptococcus sp.*

The virus has been deeply investigated during the epidemic in many species of fish by electron microscopy and tissue culture. Wattanavijarn *et al.* (1984) found various forms of virus-like particles in the necrotic, degenerative muscle fibers, red and white blood cells, liver and spleen (Fig. 12). These particles appeared to be related to pathogenesis of the disease. The cytopathic effects was occurred after inoculation of cell-free filtrate to the EPC cell line (Tangtrongpiros *et al.*, (1983).

The ecto and endoparasites; *Trichodina sp., Dactylogyrus sp., Spinitectus sp.* and *Camallanus sp.*, were found (Tangtrogpiros *et al.*, 1983, 1985). *Trypanosome sp.* were also identified from 11 sicked snakehead fishes of 264 fishes. During the epidemic in 1982-1983, Pichyangkura and Bodhalamik (1983) found the fungus, *Achlya sp.* (Fig. 13) in every ulcer of 5 fishes but during the epidemic in 1984 the *Achlya sp.*

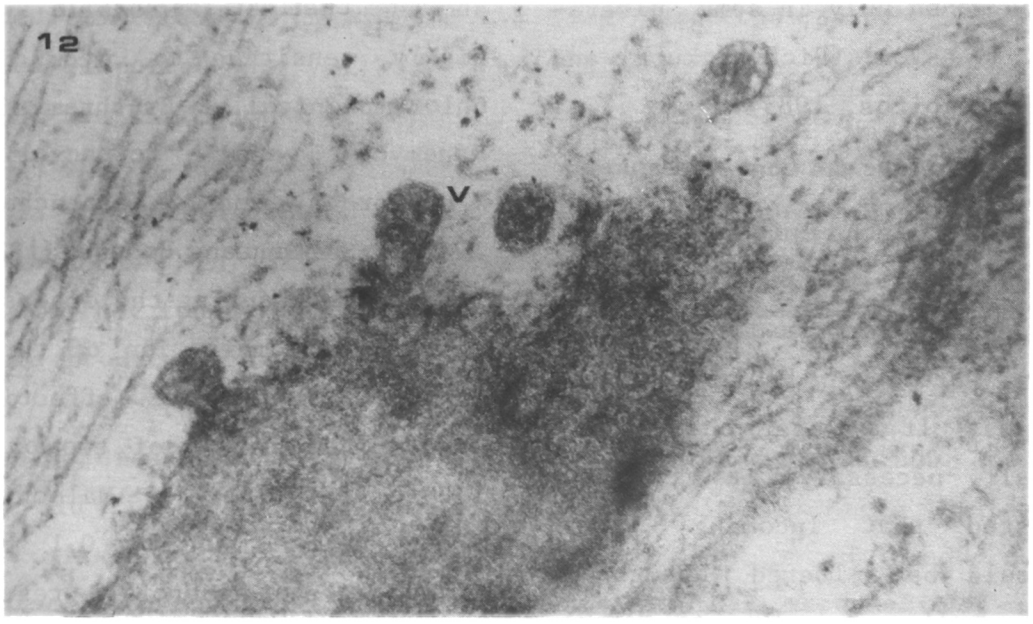


Figure 12 Viruses in the cytoplasm (V) of splenic cell of sick snake-head fish *(O.striatus)* (Magnification 103,000 X) (Photo by Dr. Wattanavijarn,W.)

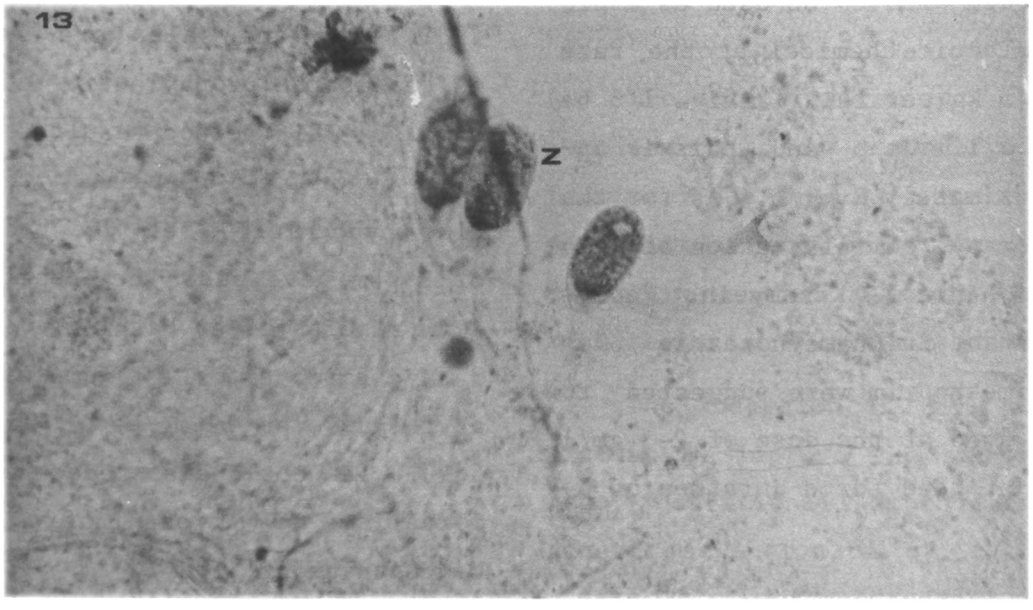


Figure 13 Show zoosporangium of *Achlya sp.* (Z) from ulcer of infected fish *(O.striatus)*

were found only in some ulcerative lesion (Pichyangkura and Tangtrogpiros, 1985).

IV. Therapeutic and Control Measures

During the epidemic in 1983, the Fishery Department recommended the farmers not to change the water in their culture ponds. If a water change was really necessary, it was suggested that incoming water should be filtered through a charcoal bed in order to reduce the amount of pesticides. Potassium permanganate was recommended also to be used as a prophylactic chemical at the rate of 5 kg per rai (1 rai = 1/6 ha) of a 1 m deep pond, this is approximately 3 mg/L. As for the therapy, administration of Chloramphenicol, Kanamycin, Neomycin and Sulphamethoxazole plus Trimethoprim were suggested to be used at the dose of 2-3 gm/1 kg of feed for a duration of 5-7 days.

Tachushong and Saitanu (1983) reported that $KMnO_4$ at 5 mg/L could not inactivate *Aeromonas hydrophila* in fish pond water. The sensitivity test showed that *A. hydrophila* was highly sensitive to Neomycin, Chloramphenicol, Erythromycin, Kanamycin and Tetracycline (Reungprach and Kesornchan, 1983). Saitanu and Poonsuk (1983) reported that curing the diseased fish by a combination of three antibiotics was more effective than a combination of two or a single antibiotic. Malachite green at the rate of 0.1 ppm showed the beneficial result after spraying to the ponds (Tangtrongpiros *et al.*, 1983).

During the outbreak in 1984-1985, lime and salt were suggested as the prophylactic chemicals. This method resulted in some beneficial results. Calcium hypochloride at the rate of 1 ppm gave also the beneficial results after bathing every 3 days for 3 times. Flumequine at the dose of 12 mg/kg of fish for 1 week could also reduce the mortality (Tangtrongpiros and Wongsatayanont, 1984).

V. Hygienic studies

During the disease outbreak, the moribund and dead fishes were preserved by salt-

cured. Saisith and Kasornchung (1983) reported that *A. hydrophila* was killed by this method. The toxin of *Aeromonas hydrophila* was studied by several authors both *in vitro* and *in vivo*. They found that *A.hydrophila* produced cytotoxin at 25 and 37°C. The fresh, heat-treated disease fish, 1 and 15 day old Pra-ra from diseased fish, fresh and heat-treated *A. hydrophila* toxin were fed into mice. The results showed that mice showed no sign of illness after investigation for 72 hours. (Laohaviranit, 1983; Supawat *et al.*, 1983, Tesprateep *et al.*, 1983). This result confirmed by Luangtongkum *et al .*, (1983) by inoculation of heat treatment of cell free toxin and washed-cell suspensions into the rabbit ileal loop.

During the disease outbreak, *Aeromonas hydrophila* was isolated from human (Reinprayoo *et al.*, 1983; Chumkasien, 1983). This infection might be contaminated from the drinking water.

Conclusion

The disease is an important problem for the intensive fish culture. It can occur easily when the pathogens, fish and environment are imbalance. From this epidemic, the predisposing causes appear to be a combination of abnormally low temperature, low DO and high NH_3N. This environmental stress made the fish in low immune response and they are easily susceptible to microorganism. The anemic condition of fishes indicate the destruction of blood corpuscle. This destruction may be the effect of virus or toxin of *A. hydrophila*. The purification of virus is for need this proof.

The ectoparasites and fungus seem to be the secondary infection. The present levels of insecticides and herbicides in natural water had nothing to do with this fish disease epidemic.

The induction of high resistant by improve the management; nutritional improvement, genetic selection or by vaccination is need for future study.

Acknowledgement

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