FISH HEALTH SECTION
AFS
NEWS LETTER

Volume 16, Number 2 April 1988

INTERNATIONAL FISH HEALTH CONFERENCE

The International Fish Health Conference sponsored by the Fish Health Section of the American Fisheries Society is proceeding on schedule. As most Section members are aware, this will be the first international meeting hosted by the Section and is being held in Vancouver, B.C. Canada July 19-21, 1988. Trevor Evelyn appears to have done his usual efficient job of lining up the facilities at the Holiday Inn-Vancouver Harbourside and BioMed Laboratories has generously offered to assist with the costs of a reception. With the urging and efforts of Ron Hedrick, the AFS Executive Committee has approved the establishment of the North American Journal of Fish Health and it appears that papers presented at the meeting will form the first volume.

While the program will not be set until after the receipt of abstracts on May 15, the present plans are to have sessions in the following areas: Bacterial and Fungal Diseases, Viral Diseases, Protozoan Diseases, Parasitic Diseases caused by Metazoans, Nutritional and Environmental Diseases, Neoplasms and Other Proliferative Disorders, Immune Repsonses, Stress, Diseases of Molluscs and Crustaceans, Diagnosis and Detection, and Disease Control. Interest in the meeting appears to be growing rapidly and it may be necessary to establish concurrent sessions for some topics, although, the organizers will try to avoid this option. A poster area is planned for those desiring this form of presentation.

If you have not yet made reservations it may be necessary to do so soon. Registration materials and abstract forms can be obtained from Trevor Evelyn, Department of Fisheries and Oceans, Pacific Biological Station, Nanaimo, B.C., Canada V9R 5K6, phone (604) 756-7066 and room reservations should be made directly with the Holiday Inn-Vancouver Harbourside, 1133 West Hastings St., Vancouver, B.C. Canada, V6E 3T3, phone (604) 689-9211 (toll free: 1-800-663-8882) or telex 04-507763. You should mention you are attending the meeting as a block of rooms at reduced rates is being held until June 18, 1988. For those presenting papers, abstracts must be sent to Trevor by May 15, 1988 and final copies of manuscripts will be due at the meeting. Registration fees will be $50.00 (Canadian funds) until July 1, after which they will be $55.00.

NEW FISH HEALTH JOURNAL

Dr. W.A. Rogers
Auburn University
Auburn, AL

The new fish health journal has been approved and will be published by the American Fisheries Society. It will be titled, "The Journal of Aquatic Animal Health." Manuscripts will follow the guide for authors published in Transactions of the American Fisheries Society. This guide will be available from Dr. W.A. Rogers. We are soliciting manuscripts in all areas of aquatic animal health. Papers may concern any marine or fresh-water species. We propose to publish papers in two major categories· articles and communications. Manuscripts from the International Meeting in Vancouver in July will comprise the initial publication and we will begin accepting manuscripts Sept. 1, 1988. Manuscripts should be sent to Dr. W.A. Rogers, Dept. of Fisheries and Allied Aquacultures, Auburn Univ., Auburn, Alabama 36849. Publication costs will be $50 per page. Subscription to the Journal will be $25 for AFS members and $105 for non-members.

WESTERN FISH DISEASE WORKSHOP

Wayne Brunson has generously offered to host the annual Western Fish Disease Workshop on July 18 at the Holiday Inn-Vancouver Harbourside in conjunction with the International Fish Health Conference which will insure a good attendance. The meeting was originally scheduled to be held in the San Juan Islands and Wayne was kind enough to modify his plans. If you are planning to attend the one-day Western Fish Disease Workshop, you should notify the Holiday Inn as soon as possible. Reigstration for the meeting will be $7.00 and further information can be obtained from Wayne Brunson, 4912 - 192nd St. SW, Lynnwood, WA 98036, phone (206) 776-9353. If you are planning on presenting a brief talk, you should contact Wayne immediately.

Be a PRO — Recruit a New Member

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PASSAGES

Tom Wellborn has left Mississippi State University and is now at the University of Florida. His new address is Department of Fisheries and Aquaculture, University of Florida, P.O. Box 434, Blountstown, FL 32424. His new phone number is (904) 674-8353.

M.H. "Spike" Beleau has moved from Mississippi to Abbot Laboratories where he is now a senior research and development specialist. His new address is Abbot Laboratories, Department 453, 14th and Sheridan, North Chicago, IL 60064. His new phone number is (312) 937-6088.

INFECTIOUS HEMATOPOIETIC NECROSIS VIRUS NOT DETECTED IN SPAWNING SOCKEYE SALMON MAINTAINED IN FRESHWATER NET PENS

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Spawning sockeye salmon were examined for the presence of IHN virus as part of the National Marine Fisheries Service sockeye salmon enhancement project in Washington State. It is suspected that the virus may be transmitted vertically from the parents in reproductive fluid or directly within the egg (Mulcahy, D. and Pascho, R.J. 1985. J. Fish Dis. 8:393-396). Although not proven by definitive studies, it is widely believed that most, if not all, of adult sockeye salmon are infected with IHN virus at the time of spawning.

This belief has been based primarily on examinations of sockeye from their natural spawning grounds. In the present study, potential sockeye brood stock were captured before they reached their spawning grounds on tributaries of Wenatchee Lake, Washington during the summer of 1987. The fish were captured at the Dryden Dam, approximately 15-20 miles from where the Wenatchee River runs into the Columbia River. The fish were transferred to freshwater net pens at Lake Wenatchee, Washington, where they were maintained until spawning. This was done to enhance their survival and to determine if this procedure would reduce the prevalence of the virus in spawning fish. Contrary to the expectations of many fishery biologists who reviewed the project before its inception, minimal mortalities occurred in the net pens; approximately 10 of 250 fish died prior to spawning.

Fish in the net pens underwent normal maturation and were spawned in late September through early October 1987. Kidney and spleen tissues, and reproductive fluids were collected from all brood stock sockeye salmon from 24 September to 14 October 1987. Samples were collected shortly after spawning, placed in tissue culture medium containing 8X penicillin and streptomycin (800 ug/ml) and delivered to the Battelle Marine Research Laboratory within 24 h. The ovarian fluid or semen, kidney and spleen of all spawning males (n=97) and females (n=129) were assayed for the presence of IHN and IPN virus on EPC and CHSE-214 cell lines using standard techniques as described by Amos, K.H. (1985. Procedures for the detection and identification of certain fish pathogens. 3 ed. Fish Health Sec., Am. Fish. Soc. Corvallis, Oregon). All fish were assayed individually. Tissues were homogenized and incubated at 4°C in tissue culture medium with 8X penicillin/streptomycin solution for 1-3 days prior to culture inoculation. After centrifugation, the supernatant was inoculated onto cells in 24 well plates at the following concentrations: 1:5 and 1:20 for reproductive fluid and 1:20 and 1:50 for spleen and kidney tissues. Cultures were incubated at 15°C, and suspect cultures were blind passed after 14 days. For confirmatory diagnosis, 37 of these fish were also assayed at the fish pathology laboratory, University of California, Davis using similar methods.

No cytopathic effects indicative of IHN or IPN viruses were observed from any of the individual samples at either dilution on either the CHSE-214 or EPC cell lines in fish tested at Battelle and U.C. Davis. The lack of detectable virus in the brood stock examined indicates that these fish were likely free of active IHN and IPN infections at the time of spawning.

No significant mortalities were observed in the fry. Sixty of these fry were collected for virus examination on 8 March 1988 and assayed for virus on CHSE-214 and EPC cell lines in 12 pools of 5 fish each as using standard methods as described above. No CPE indicative of viral infection was observed in these samples.

These results indicate, contrary to commonly held beliefs, that not all returning sockeye salmon are infected with IHN virus. It is not known if salmon are carriers of IHN throughout their life, or if they become reinfected upon reaching their respective spawning grounds. Why the sockeye salmon in the present study were free of active virus at the time of spawning was not determined, but it appears capture of the fish prior to reaching the spawning ground prevented infection.

IRIDOVIRUS ASSOCIATED WITH GILL NECROSIS SYNDROME IN WHITE STURGEON (*ACIPENSER TRANSMONTANUS*)

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The artificial culture of white sturgeon (*Acipenser transmontanus*) in California has been growing steadily and presently 10 farms are actively engaged in rearing juveniles obtained from feral broodstock. Growth rates obtained in captivity have been exceptional and these fish have been marketed both for the aquarium trade and as food fish. Diseases encountered in the early-rearing phases and broodstock development are the major difficulties now encountered by this growing industry.

The principal disease problems occur during egg incubation, with fungal problems and in juveniles under 12.5 cm length. In the latter case, many of the problems are associated with poor adaptation to artificial diets. Gill infections caused by *Flexibacter* spp. and liver diseases of unknown etiology are commonly encountered during this rearing phase. Periodic bouts with adenovirus infections of the gut epithelium are also observed in certain stocks.

For the past two years, fish at several farms have suffered from a gill disease characterized by an initial proliferative phase followed by necrosis of the epithelial cells. Gills in the later stages were often free of bacterial agents and periodic treatments with nitrofurans failed to control the disease. Fish with the gill condition often had livers with abnormal color and texture. An examination of the gills at the light microscopy level showed several cells in the epithelium with slightly enlarged nuclei and a strongly basophilic cytoplasm. An examination of these enlarged and rounded cells by electron microscopy showed abundant hexagonal and enveloped virion with a mean diameter of 258 nm (between opposite sides, n=20) present in the cytoplasm (Fig 1). The virions (206 nm in diameter) possessed a prominent internal electron dense nucleoid. Many incompletely formed or partially filled particles were also observed.

The virus particles observed in the white sturgeon gills are very similar in size and morphology to members of the iridoviridae. Perhaps the best known representatives of this group of viruses among fishes are lymphocystis virus and erythrocytic necrosis virus.

The source and significance of the iridovirus in sturgeon is unknown although similar viruses have been reported as causes of gill necrosis in cyprinid fishes. Attempts to culture the agent on sturgeon cell lines and to transmit the disease by cohabitation with healthy juvenile sturgeon failed.

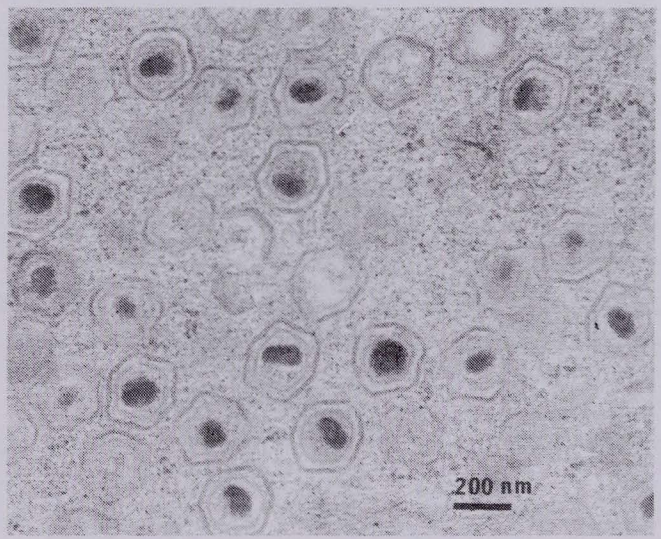

Figure 1. Iridovirus from white sturgeon.

WATER TEMPERATURE SIGNIFICANTLY AFFECTS FIELD SURVIVAL OF MARKED FISH

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Marks and tags are useful tools for fishery managers and researchers and are used to obtain information for stock assessments, behavior and movement patterns, age validations, mortality rates, and population estimates. Minimal marking-induced mortality is a critical assumption for valid interpretations of data resulting from mark/recapture. Of the many techniques available, fluorescent-pigment marking has been shown to result in minimal mortality from the handling and marking procedure. Pressures needed for fluorescent tagging, species tolerances, and the effects of temperatures on survival were evaluated in laboratory settings to develop procedures that would cause minimal mortality. However, work undertaken at the National Fisheries Center-La Crosse observed significantly different survival patterns in the field from those observed in the laboratory.

Fluorescent-pigment marking techniques were used in a study to examine habitat specificity of juvenile sunfishes and adult minnows in backwaters of the upper Mississippi River. The literature and confirmatory studies in the laboratory, where test individuals were held in 12°C well water, indicated that our target groups—juvenile bluegills (40-60 mm total length) and adult minnows—were quite capable of withstanding the 90 PSI and 180 PSI application pressures, respectively, with little mortality. Field marking of juveniles and minnows was initiated in mid-summer and resulted in virtually 100 percent mortality. We repeated marking of juvenile bluegills in August (25.6°C) and experienced a 98 percent mortality, including 84 percent of control fish (handled but not marked). High water temperatures and associated lower tolerances of stress are believed to have been the causal factors in our field mortalities. A series of temperature-related field studies was initiated to mark and hold fish under "natural" conditions in backwater areas for three to four days to assess the combined impact of handling and marking on survival as river temperatures declined.

For the field study, small bluegills (30-60 mm) were marked at 90 PSI while larger fish were marked at 140 PSI. Fish were divided into three groups: (1) small bluegills (30-60 mm), (2) minnows (35-70 mm), and (3) larger centrarchids and percids (75-155 mm). Handling stress was minimized by immediately placing fish in small floating mesh-bottom trays after collection by seine. They were transported, marked, and placed in large holding cages for monitoring. Control fish were treated in the same manner as marked fish, except for the pigment application. The percent mortality in all lots was determined on day 3. These field studies were conducted from September through mid-October, when river temperatures were declining from 19.5° to 10.0°C.

Water temperature significantly influenced the degree of mortality observed in our field studies. In addition, the various groups responded differently to the capture/marking process. Mortality among small bluegills and the larger centrarchid/percid group was directly related to temperature and decreased noticeably below 18.5°C. Minnows did not show this decrease in mortality with cooling river temperatures; mortality was high at all temperatures and survival was never greater than 50 percent. Mortalities among controls were slightly lower but showed similar trends. Within the larger centrarchid/percid group, individual species varied in their tolerance to capture and marking. Seven different species were tested and most showed no mortality. Mortalities occurred in bluegills only in the 19.5°C treatment but black crappies died at all temperatures. Black crappies appeared to be particularly sensitive to marking stresses; mortalities of 100, 67, and 50 percent occurred at temperatures of 19.5, 18.5, and 13.6°C, respectively. At 10°C, only 17 percent of the crappies died, but this was still greater than for any of the other species in the larger centrarchid/percid group.

We have found that the collection and marking of small fishes, and some larger ones, during the summer causes a variable and unacceptable level of mortality. When water temperatures were above 19°C, seining and the removal of the fish from the net alone caused greater than 75 percent mortality. Marking of small fish only in spring and fall may reduce mortality.

Our data also indicate considerable variation in mortality by size and taxa. When study objectives permit, those taxa more tolerant to high temperatures and handling stresses should be used in marking studies.

LAKE TROUT MORTALITIES

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A serious problem has developed over the past several years that could severely impact the lake trout restoration program in the Great Lakes. Several lake trout rearing stations have incurred excessive losses of fingerling fish due to unexplained causes.

In 1983, essentially all production fish being propagated in outside raceways were lost at the Pendills Creek, Hiawatha Forest National fish Hatcheries and the Marquette State Fish Hatchery in Michigan. No known fish pathogen could be detected and losses were attributed to nitrogen gas supersaturation. Further losses occurred in 1984. In January 1986, a major die-off of lake trout occurred at the Iron River Wisconsin National Fish Hatchery. A task force of USFWS researchers assembled at the station in an attempt to isolate the problem. The group's conclusion was that an infectious agent was present and had spread from outside raceways to fish being held inside the hatchery building. Approximately 20% of the yearling fish outside were lost, as well as, the entire lot of over three million fingerling fish in the hatchery building.

Attempts at isolating an infectious agent have continued at the National Fisheries Center-Leetown, West Virginia; LaCrosse Fish Disease Control Center, Wisconsin; and at the University of Rhode Island, with little success. Losses at Iron River continue although stringent precautions have been taken to minimize the risk of infection by separating broodstock from production unit.

A further review of previous lake trout mortalities revealed that excess mortalities have also occurred in New York state hatcheries, as well as, hatcheries in Tennessee, Illinois, and Wisconsin. Again, known fish pathogens were not found in moribund fish. The magnitude of this problem is such that the entire Great Lakes Basin may be affected and, therefore, remedial action is necessary to identify and control the causative factor or factors.

On October 27-28, 1987, a group of research and operations personnel (list attached) met in Twin Cities, Minnesota to discuss this issue and needed actions to solve the problem and restore lake trout production to normal. Following is a summary of the findings of this group and a chronological action plan with estimated budget needs.

In response to a conclusion by a special task force in June 1986 that an infectious agent was the primary factor in losses of lake trout at the Iron River National Fish Hatchery, the USFWS initiated a number of health management practices. First, resident brood fish were isolated from egg incubation and fry-rearing areas, and the effluent from the broodstock section of the hatchery was directed to the waste treatment lagoon. Strict control measures for disinfection were instituted, including foot baths, disinfectant hand washes, and separate sets of equipment restricted to use in the broodstock area. Secondly, eggs were disinfected before being taken from the broodstock area. All production facilities and equipment, other than the broodstock raceways, were disinfected before eggs were placed in the incubators. Fish in the waste treatment lagoons were eradicated.

Susceptible young lake trout were held in live cages placed in the effluent from broodstock raceways. In June, the disease was detected in one of five cages. This indicated that carriers were present among the brood fish.

Despite rigorous isolation and disinfection efforts, the disease reappeared in outside production raceways in early September and reached epizootic proportions in a number of raceways. To date, over 420,000 fish from the 1986 egg take have died. More losses are expected.

Electron microscopy has detected the presence of several unknown organisms in epithelial tissues of diseased lake trout fingerlings. Although the organisms could not be identified, they are suspected of being a virus or a possible chlamydia.

Therapeutic measures were attempted. In human medicine, chlamydial infections are treated with tetracycline or erythromycin. Erythromycin was fed to fish in affected raceways for 21 days with no effect on the course of the disease. Subsequent attempted therapy with tetracycline and Romet-30 is in progress with no evident beneficial effect. The disease has continued to spread in the raceways and it is evident that the etiological agent is highly virulent, very contagious, and very dangerous to lake trout. Other species of salmonids appear to be refractile to the disease but could be carriers without developing acute infections.

Possible explanations for failure of the rigorous sanitation/disinfection program were considered. Although human factors could not be completely ruled out, this was considered to be unlikely. It was noted that there was considerable bird activity between the lagoons and the lower raceways. The primary predators involved were gulls and great blue herons. These birds are suspected of being active or passive transmittors of the infective agent and could have carried the organism from the effluent lagoons. Birds have been known to transfer fish diseases and are also known hosts for chlamydial infections. Escaped fish from the raceways have repopulated the lagoons and would have been exposed to the disease agent via effluent from the brood fish raceways. Any transfer of fish, dead fish, or bird excreta to the production raceways would thus have provided a possible route for introduction of the pathogen.

It is now evident that the disease poses a very real and major threat to lake trout culture throughout the entire nation. Unless control measures can be developed quickly, the disease could place lake trout restoration efforts and a major portion of the Great Lakes sport fishing industry in jeopardy. Emergency action is warranted.

Although the causative agent is, as yet unknown, two courses of action require immediate attention. Failure of the attempts to prevent transfer of the pathogen between carrier brood fish and production fish strongly indicates that the presence of both age groups of fish at the same station is incompatible to successful lake trout culture. It will be necessary to temporarily depopulate the Iron River National Fish Hatchery of all fish to accomplish complete disinfection of the station. Alternate housing of the present brood fish or alternate sources of eggs must be developed.

Repopulation of the station is recommended only through the introduction of disinfected eggs or by fish from sources known to be free of the disease.

Although the question of possible transfer of the pathogen within eggs has not been answered conclusively, hatchery experiences associated with shipments of eggs from known infected hatcheries to clean stations suggest that this route of transfer is unlikely. Until research can explore the matter in detail, the issue will have to remain an open question.

Concurrently, and equally important to disinfection of the facility, is the need to identify the etiological agent and to develop methods to detect presence of the organism. This information is vital to determine the incidence of the disease, to determine which stocks harbor the pathogen, and to evaluate the success of eradication efforts.

Three possible types of infective agents have been suggested in studies of the disease. These include a viral agent, a chlamydia-like organism, and a possible highly virulent myxobacteria. Until one of the organisms has conclusively been demonstrated to be the cause of the disease, all three possibilities must be checked. Each requires a different approach. Electron microscopy will be required. Tissue culture capability and special lake trout cell lines are likely to be needed.

Possible viral agents are being investigated at the National Fish Health Research Laboratory and at the University of Rhode Island.

The question of possible chlamydia or related organisms will be investigated by the National Veterinary Services Laboratory (USDA) at Ames, Iowa.

At present, a possible myxobacterial agent has been detected only at a single location (New York) that is geographically distant from the endemic area of the lake trout syndrome (upper Midwest). As a consequence, it was recommended that this organism be investigated by the Region 3 Fish Disease Control Center in cooperation with the state of New York fish pathologist.

Editor's note: The previous article is an epizootiological description of occurrence of a disease in lake trout. Subsequent to that description, work has continued in some laboratories to determine the etiological agent. The following article gives some evidence that a viral pathogen might be responsible for the lake trout mortality.

VIRAL EPIZOOTIC EPITHELIOTROPIC DISEASE OF LAKE TROUT (*SALVELINUS NAMAYCUSH*)

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During the past 4 years cumulative mortalities of unknown etiology of hatchery reared juvenile lake trout in the Great Lakes Region have exceeded 15 million individuals. Epizootics have been reported at 7 hatcheries in 4 states. Investigations of environmental conditions, ectoparasites and known bacterial, fungal, and viral agents failed to reveal the cause of mortalities. Lesions associated with the epizootics include: hyperplasia and hypertrophy of branchial and skin epithelium, renal tubule degeneration and dilation of glomeruli.

Recently, we have isolated a previously undescribed virus from moribund lake trout. The virus, which measures 110 nm in diameter (Fig. 1), has not been characterized completely. Purified virus, as described by isopycnic centrifugation and electron microscopy, has been utilized in infectivity studies to fulfill Koch's postulates and verify that the virus is the etiological agent. Horizontal transmission has been demonstrated. Uninfected juvenile lake trout exposed to moribund fish at 10°C exhibited clinical signs of whirling, ataxia and hyperexcitability and died 10-15 days post contact. Infectivity studies with rainbow trout, brook trout, and brown trout indicate that these species are not susceptible.

In vitro propagation of the agent has not been accomplished. The virus does not multiply in EPC, RTG-1, FHM and CHSE-214 cell lines. Primary lake trout cell cultures are being evaluated presently.

The epidemiology of the disease and the impact on fish in the great Lakes is unknown. Development of an *in vitro* assay is a prerequisite for answering these questions and for successful management of this disease.

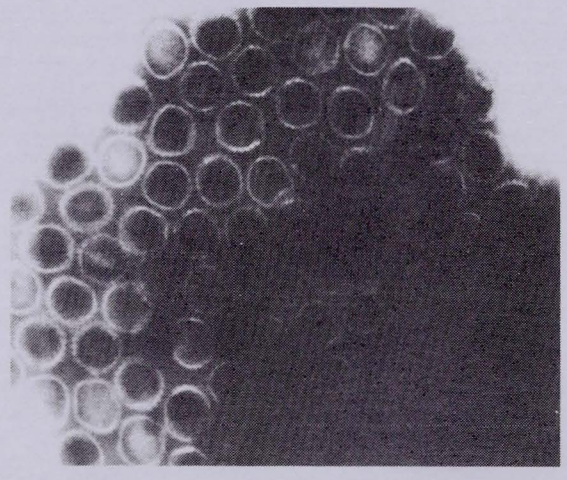

Figure 1. Electron photomicrograph of negative stained particles of epizootic epitheliotropic virus.

WHIRLING DISEASE IN IDAHO

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Myxobolus (Myxosoma) cerebralis has been confirmed histologically at three facilities in Idaho. Pat Chapman made the first and second identifications in August 1987. These were at a state operated hatchery on the upper Salmon River in juvenile spring chinook and in juvenile summer chinook at a state operated hatchery on the Pahsimeroi River. Fish at both locations exhibited minor whirling behavior, and some fish at the Pahsimeroi River facility also had characteristic black tails.

Rainbow trout at a private hatchery in the Lost River drainage were found positive in December 1987. No external signs were seen initially in these fish, and only very minor whirling behavior was seen later. All three findings have been confirmed histologically.

Sampling effort to evaluate the distribution of *M. cerebralis* in Idaho is continuing. Salmonids from Clearwater River, Snake River, and Salmon Falls Creek have tested negative to date.

Additional wild, hatchery, and returning anadromous fish from a number of sites throughout Idaho have been examined; from this testing, *M. cerebralis* has been confirmed only at a small, private trout pond on Rapid River. Samples from the Jarbidge River, from a small farm pond in northern Idaho, and a state hatchery in southern Idaho contained myxosporidan spores with similar *M. cerebralis* morphology, but histopathological tests have either not provided confirmation or have not yet been conducted.

The Idaho Department of Fish and Game (IDFG) is reviewing all shipment records from the affected hatcheries to evaluate the source and distribution. The IDFG and USFWS will continue testing fish at unaffected hatcheries and watersheds for the same purpose.

THE EFFECT OF DIFFERENT SALT SOLUTIONS ON THE SURVIVAL OF *AMYLOODINIUM OCELLATUM*

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With an increasing interest in culturing warmwater marine and estuarine species such as striped bass and redfish, the dinoflagellate ectoparasite *Amyloodinium ocellatum* has become an increasing problem in aquaculture. Consequently, we adapted the parasite to cell culture in order to facilitate the study of this pathogen (Noga, E.J. 1987. *Science* 236: 1302-1304). In developing this propagation system, we tested a number of different salt solutions and found that the parasite was capable of growth in completely defined media that contained only major mineral salts (i.e., Na, K, Cl, Mg, Ca, SO_4, PO_4, and CO_3). Trace minerals normally present in seawater or other nutrients (amino acids, vitamins, etc.) were not needed.

Interestingly, not all the simple salt solutions tested were equally as good in supporting survival of the parasites. Some salt solutions, even though they were high in salinity (osmolarity), did not allow prolonged survival of *Amyloodinium*. While the types of salts in the permissive and nonpermissive solutions were identical, the ***proportions*** of different salts varied somewhat, suggesting that manipulation of the salt composition (e.g., by adding certain salts) might be used to treat this disease. We are initiating studies to examine how manipulation of the salt composition may inhibit growth of this parasite. This information may hold promise not only for treating amyloodiniosis but may allow prevention of infections because of the stability of these mineral salts and (presumably) their relatively low toxicity compared to conventional chemotherapeutants. Hopefully, this information may also be useful in controlling other ectoparasitic infections.

CONTROL OF FURUNCULOSIS WITH ROMET®

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A field trial to investigate the value of early treatment in preventing summer mortalities in juvenile coho salmon (*Oncorhynchus kisutch*) due to *Aeromonas salmonicida* septicemia (furunculosis) was carried out the Puntledge Salmonid Enhancement Program facility on Vancouver Island, British Columbia. Furunculosis is an annual problem in coho parr at the site following the increase of water temperature in the early summer.

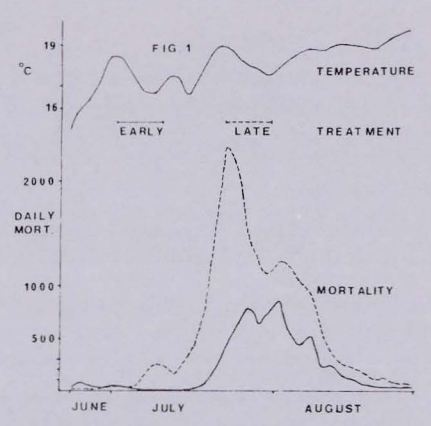
The rearing channel is supplied with unfiltered surface water. It is divided serially into four sections, each of which was stocked in early June 1987, with 300,000 coho fingerlings. To compare the efficacy of advance treatment with that of treatment following diagnosis of the disease, two of the sections received 10 day oral treatment with a potentiated sulfonamide, Romet®, beginning July 1. The two remaining sections received the same treatment when mortality due to *A. salmonicida* had reached 1,000 fish per day in each of these sections. The drug was administered, premixed, in a commercial diet (Oregon Moist Pellets) that was fed at 3.5% of body weight daily. During the period of the study (June 23-August 28, 1987), mean daily temperatures and mortality counts were recorded. Samples of moribund fish were taken from each section weekly to document whether *A. salmonicida* or PKD was accounting for the losses. Antimicrobial sensitivies of strains isolated were measured by culturing peptone-saline suspensions on Mueller-Hinton agar in the presence of drug-impregnated discs.

Mean daily mortalities and temperatures smoothed over 3 days are presented in Figure 1. The early-treated sections received the drug from July 1 to 10 (solid bar in Figure 1), and the other two sections received it from July 21 to 31 (dashed bar). One of the early-treated sections received a second 10-day treatment starting July 24, when mortalities in that section approached 1,000 per day. The cumulative mortality figures for the study period are summarized .

Section	Treatment	Cumulative Mortality	Cumulative % Mortality
1	Late	29444	9.8
2	Early	7450	2.5
3	Early	21635	7.2
4	Late	46772	15.6

Mean cumulative % mortality of the early-treated sections was significantly lower ($p < 0.001$, Student's t test) than that of the late-treated sections. The pattern of mean daily mortalities observed in the different treatments (Figure 1) suggests that early treatment gave nearly complete protection for approximately 10 days following cessation of treatment. Although furunculosis subsequently occurred in the early-treated groups, daily mortality counts never reached the levels observed in the late-treated groups. Romet resistance was not observed in the *A. salmonicida* strains isolated during this study.

The most common diagnosis in the weekly samples of moribund fish prior to July 27 was furunculosis. The PKX organism was observed in Diff-Quick®-stained kidney impression smears, but gross pathologic changes typical of the disease were not observed until the week of July 27. Following this date, proliferative kidney disease and fungal growth on the skin or gills were the predominant diagnoses. These occurred independently in some of the fish examined and concurrently in others. Isolation of *A. salmonicida* in the occasional fish suggested that this organism continued to be responsible for a few mortalities. Many of the fish examined histologically had metacercariae of the trematode *Diplostomulum spathaceum* present within their retina, and an occasional fish had a mild epitheliocystis-like infection of branchial epithelial cells. If the latter can be confirmed as epitheliocystis, this would be the first record of this condition in salmonids in British Columbia.



ISOLATION OF VIRUSES FROM CYPRINID FISHES

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Golden shiner virus (GSV) was the most common agent isolated from populations of golden shiners (*Notemigonus crysoleucas*) and grass carp (*Ctenopharyngodon idella*) either directly from California state waters or from fish bound for import. The virus induced rapid cytopathic effects (CPE) characterized by syncytium formation in the EPC cell line in 1 - 2 d at 25°C. This type of CPE is exhibited by all members of a recently described group of reoviruses from aquatic poikilotherms (includes catfish, chum salmon, American oysters). All of the viruses in our study exhibiting this type of CPE were readily neutralized by anti-GSV rabbit serum and had identical electrophoretic patterns of the RNA genome segments to that of GSV (reference strain from Dr. J.A. Plumb, Auburn University).

The virulence of one virus isolated from grass carp was further investigated by exposing young grass carp (1.2 g) and golden shiners (0.5 g) by immersion to $10^{7.3}TCID_{50}$/ml of virus for 30 min before water (25°C) flow was resumed to the aquaria. There were no mortalities in either species of the experimental group or among control groups held under the same conditions but not exposed to the virus. Additionally, there were no indications of microscopic changes induced by the virus as judged from histological sections although concentrations of $10^{5.5}TCID_{50}$/g were detected in pooled samples of kidney, liver and spleen of golden shiners 1 wk after exposure. Concentration of virus in pooled kidney, liver and spleen of grass carp were considerably lower ($10^{3.3}TCID_{50}$/g) after one week. Four weeks following exposure, virus was detected in only one pool from golden shiners and the concentration was below the level needed to establish a 50% endpoint.

In a second study, juvenile grass carp were injected (intraperitoneally) with $10^{8.2}TCID_{50}$ of the same virus used in the study with small fish. Again there were no indications of disease although virus was easily recovered from fish at 3 wks and again at 3 mo. following injection. Mean water temperature was 25°C during the course of the study.

A second virus isolated from two populations of golden shiners induced a unique CPE characterized by rounded clusters of cells that first appeared at 1 wk after inoculation of EPC cells. The virus which was not neutralized by anti-GSV serum, was sensitive to chloroform treatments. In initial trials, the virus has failed to replicate or cause disease in golden shiners exposed to the agent by immersion but further studies are warranted. Electron microscopy of infected cells or virus concentrated from the culture media, showed numerous round to hexagonal particles with a mean diameter of 81 nm. There is also evidence of a tightly fitted membrane around the virion.

ASSOCIATION OF *AEROMONAS SOBRIA* WITH MORTALITIES OF ADULT GIZZARD SHAD

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Motile *Aeromonas* strains have been associated with a wide range of infections occurring in fish and other poikilothermic and homeothermic animals as well. The majority of fish infections by *Aeromonas* spp. have been due to *A. hydrophila* and *A. salmonicida*. We report here the association of *A. sobria* with an epizootic in a natural population of gizzard shad (*Dorosoma cepedianum*). In June, 1987, large numbers of shad were found dying in the C and O canal which parallels the Potomac River in Maryland. Specimens were brought to us by National Park Service personnel. No external or internal lesions were evident. Pure cultures of *A. sobria* were isolated from the kindey, liver and spleen of all 6 fish examined. The role of disease in the decline of shad populations in the Chesapeake Bay is unknown since little has been done on the microbial pathogens of this species. We plan to do transmission studies this summer to determine if *A. sobria* can cause shad mortalities under controlled laboratory conditions.

BOOKS OF INTEREST

Fish Immunology. J.S. Stolen, D.P. Anderson, and W.B. van Muiswinkel, Eds. Elsevier Science Publishing Co. Inc., P.O. Box 1663, Grand Central Station, New York, NY 10163. 485pp. $72.75.

This book contains the proceedings of an International Meeting on Fish Immunology, held in Sandy Hook, New Jersey, September 1985. The comprehensive collection includes the sessions: 1) Fish immunoglobulins, lymphocyte populations, MHC, non-specific factors, soluble substances, 2) Lymphocyte and macrophage structure and function; ontogeny, lymphoid organs, 3) Cellular and humoral responses, 4) Modulation of the immune response, and 5) Immunity to disease agents. The 47 scientific papers, which also appear in volume 12 of VETERINARY IMMUNOLOGY AND IMMUNOPATHOLOGY are indexed by author and subject.

Fish immunology is rapidly advancing and dividing into specialized areas. This collection of papers gives an outstanding presentation for updating the pathologists and biologists in fish health in examining new areas of disease diagnostics, immunization against diseases, and technical features of the fishes immune response. This book is highly recommended also in graduate courses. D.P. Anderson

* * * * *

Immunology and Disease Control Mechanisms of Fish. T.C. Hunt and A.R. Margetts, Eds. Published in the Journal of Fish Biology, 31 (Supplement a). Inquires for obtaining copies should be directed to *Attention* Jill Kettley, Subscriptions Department, Journal of Fish Biology, Academic Press Limited, High Street, Foots cray, SIDCUP, Kent DA14 5HP, U.K. 1987.

Convenor, Professor M.J. Manning, assisted by Drs. M.F. Tatner and C.J. Secombes, gathered a group of internationally recognized fish immunologists at Plymouth, England in 1987. The results of this Symposium, supported by the Fisheries Society of the British Isles, are published in this supplement containing 32 full papers and 14 brief communications. The papers are grouped according to the broad subject headings of Immunoglobulins and antibodies, Lymphocyte populations, Immune system, Immunogenetics, Phagocytic responses, Immunomodulation, Immune response to disease agents, Immunoparasitology, Immunization and vaccines. The index of authors and subjects is included as a separate flyer.

Since the Plymouth meetings were held 5 months prior to the appearance of this collection of published scientific papers, the editors are certainly to be congratulated for their organizational abilities. This edition also reflects the increasing international importance of aquaculture and fish farming in promoting research and development in fish immunology. The series of papers is an excellent contribution to this growing field. D.P. Anderson

FUTURE EVENTS

June 16-18, 1988 - ***Eastern Fish Health Workshop.*** University of Maine, Orono. For information: Dr. Paul Reno, University Maine, Department of Microbiology Mitchner Hall, Orono, Maine 04469-0131. (207) 581-2810.

July 17-18, 1988. ***Western Fish Disease Workshop.*** Holiday Inn - Vancouver Harbourside, Vancouver, British Columbia. For information: Wayne Brunson, Washington Department of Wildlife, 4912-192nd St. SW, Lynwood, WA 98036. (206) 776-9353.

July 19-21, 1988 - ***Fish Health Section of the American Fisheries Society. International Fish Health Conference.*** Holiday Inn-Vancouver Harbourside Vancouver, British Columbia. For information: Dr. Trevor Evelyn, Department of Fisheries and Oceans, Pacific Biological Station, Nanaimo, B.C. Canada, V9R 5K6. (604) 756-7000.

July 24-29, 1988 - ***International Society of Developmental and Comparative Immunology.*** University of Nottingham, England. For information: Dr. Michael Balls, Department of Human Morphology, Medical School, University of Nottingham NG7 2UH.

August 22-25, 1988 - ***International Symposium on Viruses of Lower Vertebrates.*** University of Munich, Germany. For information: Dr. W. Ahne, Institute of Zoology and Hydrobiology, University of Munich, Kaulbachstrasse 37, D-8000 Munchen 22 Fed. Rep. Germany (089) 2180-2785 (2687)

September 6-9, 1988 - ***Aquaculture International Exposition.*** Vancouver, British Columbia. For information: Project Coordinator, Aquaculture International, Box #202, 999 Canada Place, Vancouver, B.C., Canada V6C 3C1.

October 2-6, 1988. ***Third International Colloquium on Pathology in Marine Aquaculture.*** This meeting will be held in Gloucester Point, Virginia at the Virginia Institute of Marine Science and will include topics on viral, microbial, parasitic and chemical diseases of mollusca, crustacea, finfish and other marine and estuarine animals. For further information please contact: Dr. Frank O. Perkins, Virginia Institute of Marine Science, College of William and Mary, Gloucester Point, Virginia 23062, (804) 642-7102.

February 12-16, 1989 - ***Aquaculture '89***. The Westin Bonaventure Hotel, Los Angeles, California. For information: Conference Headquarters, Crest International, 940 Emmett Avenue, Suite 14, Belmont, California 94002.

April 18-21, 1989 - ***The Second Asian Fisheries Forum.*** Fisheries Science and Communities: Partners in Development. Tokyo, Japan. For information: The Secretariat, The Second Asian Fisheries Forum, Department of Fisheries, Faculty of Agriculture, The University of Tokyo, Yayio 1-1-1, Bunkyo-ku, Tokyo 113, Japan.

May 9-12, 1989. ***Riparian Management Symposium.*** This symposium will be co-sponsored by the Montana Chapter and The Western Division of the AFS. The workshop will be held at the Montana Convention Center, Billings, Montana. For a preliminary program and symposium details contact: Glenn Phillips, Publicity Chairman, Montana Department of Fish, Wildlife and Parks, Capitol Station, Helena, Montana 59620, or call: (406) 444-2406 (Marilyn Goetzinger).

September 12-13, 1988. ***Mysid-Fisheries Symposium.*** This will be a one-day special session within the Annual AFS meeting held n Toronto, Ontario. For information: Eric Bergersen, Colorado Cooperative Fish and Wildlife Research Unit, 201 Wagar Building, Colorado State University, Fort Collins, Colorado 80523, (303) 491-6942 or Tom Nesler, Colorado Division of Wildlife, 317 West Prospect, Fort Collins, Colorado 80526, (303) 484-2836.

ERRATUM

The new telephone number for BioMed Research Laboratory was erroneously reported in Volume 16(1). The correct number is (206) 882-0448.

FISH HEALTH NEWSLETTER

The Fish Health Newsletter is a quarterly publication of the Fish Health Section of the American Fisheries Society. Submissions of any length on a topic of interest to fish health specialists are encouraged and should be addressed to one of the editorial staff or to a member of the publication committee.

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