FISH HEALTH SECTION AFS NEWS LETTER

Volume 12, Number 4 October, 1984

FROM THE PRESIDENT

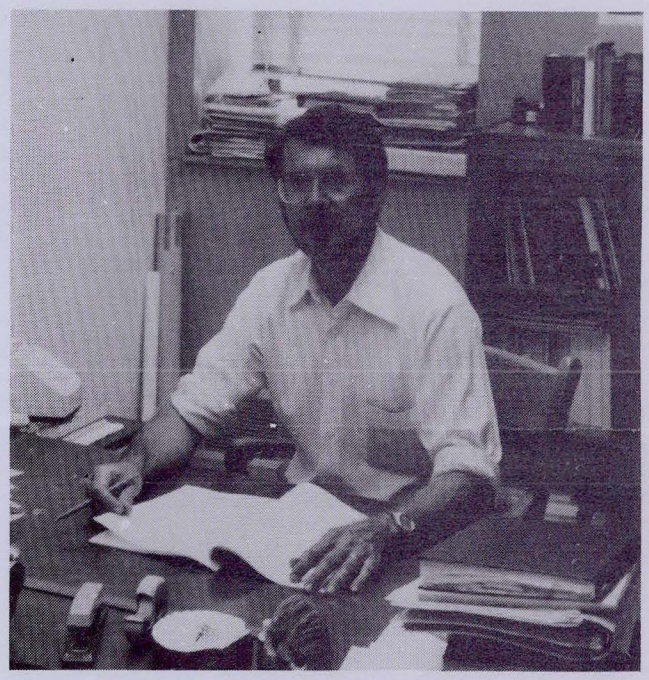
Trevor P.T. Evelyn
Pacific Biological Station, Nanaimo, B.C., Canada

By the time this issue of the Newsletter is published, my wishes to you for a "Happy New Year" will be somewhat belated because the Fish Health Section (FHS) new year starts on September 1. I apologize for this and hope that your resolution for the new year will be to renew your memberships without too much delay and to urge badly lapsed colleages to join the section once again! I trust that in 1984-85 we can bring to completion some important projects currently underway and that we can initiate and foster others that will add to the vitality and luster of the section.

I am pleased to announce that committee appointments for 84-85 are virtually complete (see page 2) and I am very grateful to the committee members for their willingness to serve. The make-up of some of the committees has changed, usually to give long-serving members a well-deserved rest. However, other committees remain essentially unchanged because I considered that change might unduly hamper progress on work that I know we would all like to see successfully completed in 84-85 (e.g. the Blue Book, the Directory and the Certification Examinations for Fish Pathologists), or because the quality of the work being done was so good (e.g., the Newsletter) that it would have been foolish to "rock the boat". Regretably, one committee (Fish Health Evaluation Committee) died a natural death, apparently because of a lack of sufficient interest to sustain it. However, another committee, the Funds Raising Committee, has risen to take its place, and it is my hope that this committee can act to maintain the financial integrity of the Section.

In my view, the most serious problem facing the Section is a financial one. Factors contributing to it are a dwindling membership and the increased costs of doing business. (The annual cost of producing the Newsletter for example, now exceeds our annual income!). If there are no drastic demands of the Section, we could survive this situation for another year or two. However, it is clear that something should immediately be done to prevent us from having to dip into our reserves. Obviously we need to increase our membership. But, it is not likely that we will be able to manage this through significant reductions in the cost of membership, at least, not as long as we wish to remain members of the American Fisheries Society. The alternatives, therefore, are to publicize ourselves more widely and tailor our projects to appeal to more people. In addition, there may be other ways for raising money. I have therefore asked Dave McDaniel to put together a committee (Funds Raising) to look into ways of keeping us solvent and for coordinating efforts for accomplishing this. If any of you have suggestions, or are willing to help on this important matter, I hope you will contact Dave without delay.

Another pressing problem that should be resolved as soon as possible is the question of a meeting site for the International Meeting planned for 1986. At the FHS Executive Committee meeting held last July in Little Rock, the decision was made to find an alternative site to the one (Snowbird, Utah) first proposed for the meeting. The general feeling was that the meeting would have greater appeal if it were situated closer to a major international airport and to the "bright lights" of a large city. I have asked John Fryer, Chairman of the Time and Place Committee to see whether a suitable alternative site and



willing host can be located. In the meantime, Dick Heckmann deserves a special vote of thanks for his willingness to act as host for the Snowbird site. In my view, hosting a world-class scientific conference will be good for the vitality of the FHS and, should an alternative site not be located, it would be far better to hold the meeting at Snowbird than not to hold it at all. In addition, the initial concept was that the meeting be sponsored by the FHS and the European Association of Fish Pathologists and that it be devoted to problems related to fish health certification. However, it would probably be preferable not to restrict the program to certification problems, and to encourage the widest possible participation in the meeting. For example, the Japanese and other non-European participants could be expected to make significant contributions to the success of the meeting.

There are other topics that might legitimately be raised here but to avoid making this too long a harangue I will mention only that the next FHS meeting is to be held in Nanaimo (on Vancouver Island), B.C., tentatively in the second week of July. The hope is that the meeting program can be fairly evenly split between infectious and non-infectious problems in fish. Perhaps if the Section shows a genuine interest in the fact that non-infectious problems are an important constraint in fish production we may be able to attract some new members.

Finally, my thanks to all who willingly stood for the various FHS offices and to the membership for electing me President. I know that I speak for both the elected and appointed officials when I say that we shall try, mightily, to make 1984-85 a successful year. Please feel free to contact us with any ideas that you consider might contribute to the health of the Section.

FHS OFFICERS AND COMMITTEES 1984-85

EXECUTIVE COMMITTEE

Voting Members
Trevor Evelyn, Chairman and President, FHS
John Rohovec, President-Elect
Glenn Hoffman, Immediate Past President
Doug Anderson, Secretary-Treasurer
Diane Elliot, Chairman, Nominating Committee

Non-voting Members (Chairmen of Standing Committees)
John Rohovec, Newsletter and Publications Committee
Emmett Shotts, Awards Committee
Steve Leek, Membership and Ballotting Committee
Paul Janeke, Professional Standards Committee
Kevin Amos, Technical Procedures Committee

STANDING COMMITTEES

Nominating
Diane Elliott, Chairman (elected)
John Schachte
John Grizzle

Newsletter and Publications
John Rohovec, Chairman
Dave Ransom
Jim Winton
Tom Wellborn
Jack Gratzek

Membership and Balloting
Steve Leek, Chairman
Ray Brunson

Technical Procedures
Kevin Amos, Chairman
Emmett Shotts
Ray Brunson
Ken Johnson
Ellis Wyatt
Dave Groman, Ex Officio

Professional Standards
Paul Janeke, Chairman
Jim Carlisle
Doug Mitchum
John Cvitanich
Bev Larson

Finance
Doug Anderson, Chairman
Steve Leek (Membership)
John Rohovec (Newsletter)

Awards
Emmett Shotts, Chairman
Dennis Anderson (two years)
John Rohovec (three years)

BOARD OF CERTIFICATION
(Elected)

Doug Mitchum, Chrm. (2 years)
Jim Warren (1 year)
Gary Camenisch (1 year)
Kevin Amos (2 years)
Joe Lientz (3 years)

AD HOC COMMITTEES
(Appointed)

Archives
Joe Sullivan, Chairman
Roger Herman
John Grizzle

Bylaws
Jim Warren, Chairman
Fred Meyer

Directory
Rowan Gould

Fund Raising
Dave McDaniel, Chairman
Doug Anderson
Roger Herman

International Meeting (1986)
Chairman to be selected
Bill Rogers
Leo Margolis
Barry Hill
Dave Conroy

Program (1985 meeting)
Trevor Evelyn
Ron Goede

Time and Place (86-89)
John Fryer, Chairman
Charles Suppes
Bill Rogers

PASSAGES

Ms. B.J. Lee has become Dr. B.J. Lee and has moved from College Station, Texas to take a position with Fritz Chemical Company. Her new address is Research and Development-Aquaculture, P.O. Drawer 17040, Dallas TX, 75217.

Vicki Blazer has moved to the Cooperative Fisheries Unit, School of Forest Resources, University of Georgia, Athens, GA 30602.

Michael Mensi has moved from Illinois to 2309 6th Ave., Gulfport, MS 39501.

MICROBIAL STANDARDS FOR FISH NEEDS, AN IMPOSSIBLE DREAM, OR AN URGENT NECESSITY?

Gordon Bell, Pacific Biological Station, Department of Fisheries and Oceans, Nanaimo, B.C., Canada V9R 5K6

Some years ago Trevor Trust (1971, J. Fish. Res. Board Can. 28:1185-1189) drew attention to the high numbers of bacteria in commercial fish feeds. Counts ranged from 10^3 to 10^7 bacteria per gram of diet. The bacteria included aerobes, anaerobes, spore-formers and enteric species. Recently, Dr. Higgs (a nutritionist at our West Vancouver Laboratory) and I became concerned about the microbial quality of the OMP used in our hatcheries. At our request, our Inspection Laboratory examined random samples of OMP and found counts of *E. coli* (MPN/100g) ranging from 50 to $> 24 \times 10^3$, and total viable bacteria from 9×10^5 to 4×10^7/g. No salmonellas were found in 200g samples. Clearly, the microbial quality of fish needs examination and possibly control.

There are three principal reasons for concern:
1. The high loads of bacteria might affect fish health directly. Some species might be fish pathogens or potential pathogens.
2. Certain bacteria might affect food handlers, fish handlers or downstream water users.
3. High loads of bacteria may lead to loss of nutritional quality (vitamin C, rancidity, etc.)

Should we try to set standards for microbial quality of fish foods? I think we should before they are imposed on us by public health authorities already concerned with the microbial quality of hatchery effluents. (I was surprised to find that in Canada at least, there are apparently no such standards for animal feeds). It would no doubt be difficult, even fruitless, to set standards for total counts (maybe the bacteria add nutrients!) but perhaps we could require industry to produce feed free of organisms of public health significance and free of named fish pathogens.

Is anyone else worked up about this problem, or do readers feel it's a storm in a fish pond? Take a kick at the cat(fish) by expressing your opinions in the next issue.

POSITION ANNOUNCEMENT

Faculty Position: Aquatic Animal Pathologist

The person in this position will be expected to develop and carry out independent and collaborative programs dealing with aquatic animal species, principally fish and shell-fish. Responsibilities include helping to operate the departmental Fish Diagnostic Laboratory (30%), assisting in the teaching programs at both undergraduate and graduate student levels (20%), and conducting basic and applied research on diseases of economic importance with aquatic animals (50%).

The position is open to graduate veterinarians or others with relevant training and experience in aquatic animal pathology. It should be particularly attractive to those interested in developing and applying special skills in new and fertile areas.

Appointment to this tenure-track position will be made at a rank and salary commensurate with training and experience. Starting date will be as soon as possible.

Interested persons should send a curriculum vitae, personal bibliography, and references to:

Dr. Bruce W. Calnek, Chairman
Department of Avian and Aquatic Animal Medicine
N.Y.S. College of Veterinary Medicine
Cornell University
Ithaca, New York 14853

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REPORTS FROM PAST MEETINGS

S.F. Snieszko Commemorative Fish Disease Workshop

Midwest Fish Disease Group was held in Little Rock, Arkansas on July 10-12, 1984. Over 89 persons attended and the 49 papers presented covered items of importance in virology, bacteriology, parasitology and the environment. There were four panel discussions on Proliferative Kidney Disease, Microbiological Problems of Current Interest, Ceratomyxosis and Inspections. At the banquet, Dr. Fred Meyer was presented the S.F. Snieszko Distinguished Service Award (see accompanying article). The Presidential Address was given by Dr. Glenn Hoffman. Copies of the program and abstracts may be obtained from The Fish Farming Experimental Station, P.O. Box 860, Stuttgart, AR 72160. Please make checks for $1.50 payable to Fish Health Section/AFS. The Time and Place Committee is considering Nanaimo or Vancouver, B.C., Canada for the next meeting site for the FHS. The Midwest Fish Disease Workshop will be held in Peoria, IL. G.L. Hoffman, FFES, P.O. Box 860, Stuttgart, AR 72160.

Sixth International Congress of Virology

The Sixth International Congress of Virology was held September 1-7 in Sendai, Japan with over 2500 virologists from around the world in attendance. One of the workshop sessions, chaired by Dr. T. Sano and Dr. J.L. Fryer, was titled Viruses of Aquatic and Marine Animals. In this session, eight papers and seven posters were presented, all involving viruses of fish or shellfish. Topics covered included molecular biology of lymphocystis and IHNV; the herpesviruses of carp, salmon, catfish and pike; characteristics of reoviruses from grass carp; a proposed new genus of fish reoviruses; use of the coagglutination assay for viruses and the creation of monoclonal antibodies to IPNV. At the Congress, the International Committee on Taxonomy of Viruses met and accepted the name Birnavirus to include all the IPN-like viruses of fish and shellfish. Photocopies of the abstracts of this session may be obtained from J.R. Winton, Oregon State University, Marine Science Center, Newport, Oregon 97365.

IR-4/FDA Workshop on Minor Use of New Animal Drugs

The second IR-4/FDA Workshop for Minor Use of New Animal Drugs was held in Gaithersburg, MD, August 21 and 22, 1984. Aquaculture was one of four work sessions and included representatives from most aquacultural industries. Careful critique of the FDA guidelines for minor species resulted in recommendations for further revision of the document. Guidelines for food fish will provide a format for obtaining FDA approval of drugs to be used in fish. The guidelines are intended to be flexible and allow scientific discretion in designing safety and efficacy studies on new drug products. M.H. Beleau, College of Veterinary Medicine, Stoneville, MS 38766.

Twenty-Fifth Annual Western Fish Disease Conference

The twenty-fifth annual Western Fish Disease Conference was held on the campus of Oregon State University in Corvallis on June 27-29. Approximately 100 people participated and 25 papers were presented. Panel discussions and informal sessions concerning virology and field pathology gave attendees an opportunity to share ideas, experiences and problems. The highlight of the meeting was a presentation by R.R. Rucker, the founder of the meetings. This year's meeting had an international flavor as individuals from the USA, Canada, Chile, Hungary, Taiwan, France and Japan participated. The next meeting will be hosted by the Washington Department of Fisheries. J.S. Rohovec, Dept. of Microbiology, Oregon State University, Corvallis, OR 97331.

Eastern Fish Health Workshop

The Ninth Annual Fish Health Workshop was held on June 12-14, 1984 on the campus of Auburn University. Sessions were titled "Cold Water Diseases", "Warmwater Diseases", "Diseases of Marine Fish and Shellfish", and "Immunology of Fish." The last session was a "Panel Discussion on Fish Immunology" in which the relationships of applied and basic immunology were discussed. This panel discussion pointed out the problem of the application of basic immunology research to the fish culture industry. It was apparent that there is a need of some mechanism for field immunologists to assimilate and put to use the information generated by the basic researcher. During the workshop, attended by approximately 75 fish culturists, students and scientists, 30 papers were presented. A highlight of the workshop was a fish fry at the "ponds" at the Fisheries Research Unit of Auburn University. The National Marine Fisheries Services, Oxford, Maryland will host the 1985 Eastern Fish Health Workshop. A limited supply of abstracts of the 1984 workshop are available at no cost by writing: John A. Plumb, Dept. of Fisheries & Allied Aquaculture, Auburn University, AL 36849.

Third International Seminar on Fish Pathology

The twentieth anniversary meeting of The Japanese Society of Fish Pathology was held in Tokyo, Japan, September 8-10, 1984. Over 120 scientists from around the world attended and 48 papers were presented. The meeting drew a broad range of topics including fungal, protozoan, bacterial and viral diseases, drug resistance, toxins, immunization and chemotherapy. A highlight of the meeting was the presentation of the S.F. Snieszko Distinguished Service Award to Dr. Syuzo Egusa, Professor Emeritus, University of Tokyo (see accompanying article). The meeting was held on the campus of the Tokyo University of Fisheries and was hosted by Dr. T. Sano and the staff of the University. J.R. Winton, Dept. of Microbiology, Oregon State University, Corvallis, OR 97331.

EXAMINATION DATE APPROACHES

Beginning January 1, 1985, it will be necessary to pass a written examination for partial fulfillment of the requirements for AFS/FHS Certified Fish Pathologist. Until January 1, 1985, a "grandparent clause" will continue to be in effect and no written exam is necessary. Application forms and information concerning required qualifications can be obtained from the Chairman of the Board of Certification, Doug Mitchum. His address is: University Station, Box 3312, G & F Lab, Laramie, WY 82071. And for further information contact Paul Janeke, Chairman of Professional Standards Committee, Fish Disease Control Center, P.O. Box 8, Ft. Morgan, CO 80701.

1984 S. F. SNIESZKO DISTINGUISHED SERVICE AWARD

The 1984 S. F. Snieszko Distinguished Service Award of the American Fisheries Society was presented to two outstanding scientists this year. Dr. Fred Meyer, Director of the National Fisheries Research Center at La Cross, Wisconsin was given the award at the banquet of the Joint Workshop of the Fish Health Section and Midwest Fish Disease Group.

The second recipient was Dr. Syuzo Egusa, Professor Emeritus, University of Tokyo. Dr. Egusa was presented his award during the Third International Seminar on Fish Pathology in Tokyo, Japan. Below are the texts of the presentations made at those meetings.

DR. SYUZO EGUSA

The S. F. Snieszko Award was created by the Fish Health Section of the American Fisheries Society to recognize distinguished service through outstanding contributions and leadership in the field of fish health. As many of you know, Dr. S. F. Snieszko was for many years Director of the U.S. Fish and Wildlife Service Eastern Fish Disease Laboratory and recognized as the dean of modern fish pathology. Unfortunately, he died January 12, 1984. For those of us who knew him personally or through his many publications, he was a great intellect, a scholar, a fine human being and someone we all miss. It is appropriate that the only such award presented in North America bears his name, and it is further appropriate that Dr. Snieszko himself was the first recipient of this award.

Thus far, those honored have all been from the United States. However, for the first time, the Award is being given to an outstanding scientist from another country. This individual was nominated for his years of creative and scholarly activities in the field of fish health. He is internationally known for his work and continues to exert a major impact on the fish health field. He is a man admired and respected by all who know him for his many contributions to our understanding of the diseases of fish.

The recipient of the Award is Dr. Syuzo Egusa, Professor Emeritus, University of Tokyo. On behalf of the Fish Health Section of the American Fisheries Society, its President, Dr. Glenn Hoffman, and the Awards Committee, I should like to present to Professor Egusa the S. F. Snieszko Distinguished Service Award.

Dr. J.L. Fryer, Department of Microbiology
Oregon State University, Corvallis, OR 97331

DR. FRED MEYER

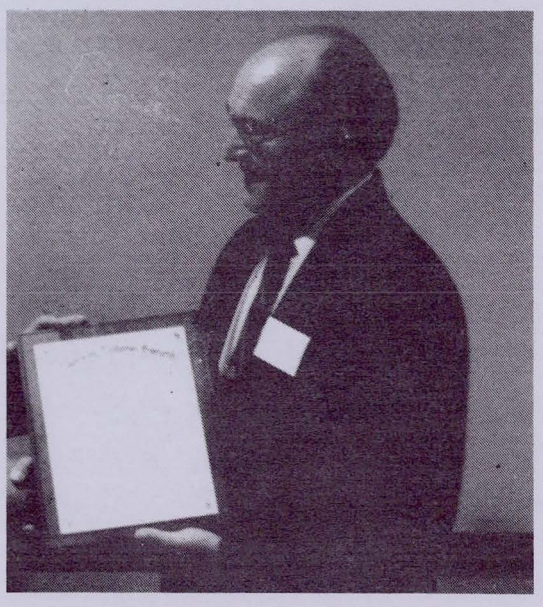
As we all know this workshop is dedicated to the memory of Dr. S.F. Snieszko, a friend and teacher to many of us. Thus, it is highly appropriate that the person being honored this evening with the S.F. Snieszko Distinguished Service Award exemplifies everything for which Dr. Snieszko stood. This person is an outstanding teacher, a dedicated scientist and a good friend to many of us here this evening. Like Doc, this individual is truly a good and gentle person and is most deserving of the highest honor that the Fish Health Section of the American Fisheries Society can bestow on one of its members.

The recipient has been active in the field of fish health for more than 20 years as a teacher, researcher and diagnostician. The recipient has more than 80 scientific publications in the field of fish health and aquaculture and has been a member of the Board of Certification FHS and the Professional Standards Committee FHS. In addition this individual belongs to many professional and honor societies, including Phi Kappa Phi and Sigma Xi.

I could go on for a long time about the recipient's professional accomplishments and honors received. But everyone in the field of fish health is aware of the tangible and significant contributions made by the recipient. However, the really significant contribution made by this individual to the field of fish health is not the number of scientific articles published, although numerous, the new diseases described, the disease treatments developed, nor the thousands of fish kills diagnosed. But rather, like Doc Snieszko, it is the inspiration the recipient has been to countless students and fellow workers. Like Dr. Snieszko, the recipient has had great influence, directly or indirectly, on practically everyone working in fish health. These intangibles, the willingness to be a counselor, to be a teacher, to help someone solve a problem, the dedication shown through the years, are to me the most significant contributions made by the recipient of the S. F. Snieszko Distinguished Service Award.

It is with great pleasure that I ask Dr. Fred P. Meyer to step forward to receive the S. F. Sneiszko Distinguished Service Award.

Thomas L. Wellborn, Jr. for Dr. Thomas E. Schwedler,
Chairman Awards Committee, FHS/AFS



ISOLATION OF CHANNEL CATFISH VIRUS FROM BROODFISH

Paul R. Bowser, College of Veterinary Medicine
Drawer V, Mississippi State, MS 39762

Channel catfish virus disease (CCVD) is a herpesvirus infection that can cause significant losses of fry and fingerling channel catfish. The virus can be isolated from young fish during the course of an active epizootic. Adult catfish have been considered a potential source of virus, which could be transmitted to the young, susceptible fish. A carrier state for the virus in adult fish has been supported by the presence of serum antibodies against CCV. However, this provides only circumstantial evidence of a carrier state.

During the winter of 1983-84, CCV was successfully isolated from 10 of 22 brood channel catfish. The first isolation was made from a moribund male broodfish that was presented to our laboratory for examination. The fish had a moderate infestation of *Trichodina* and *Ambiphrya* on the gills. CCV was isolated from a cell-free filtrate from the posterior kidneys of this fish. The virus was neutralized by a reference CCV antiserum provided by the National Fish Health Research Laboratory, USFWS, Leetown, WV. The remaining 21 fish in the pond were collected and complete necropsies were performed. The fish were divided into two groups: (1) fish #1-14 were necropsied on day of collection, (2) fish #15-21 were transported to the Aquatic Medicine Laboratory, College of Veterinary Medicine MSU, where they were immunosuppressed with Dexamethazone and necropsied 7 days later. Virus isolation procedures included: cell-free filtrates from (1) posterior kidneys, (2) gonad and (3) leucocyte co-cultures with channel catfish ovary (CCO) cell cultures. Channel catfish virus was isolated from fish #2 (female, posterior kidneys and leucocyte co-cultures), fish #3 (female, gonad) and fish #15-21 (3 male, 4 female, leucocyte co-cultures).

Several aspects of this investigation are worthy of note. First, this is the first account, to our knowledge, of the isolation of CCV from broodfish. All fish, with the exception of the original fish, appeared healthy. There have never been complaints of CCVD associated with fry or fingerlings produced by these broodstock. The role of CCV in the moribund condition of the original fish is open to speculation. The degree of parasitism could have contributed to the moribund condition of the fish. Work is continuing with the goal of perfecting a reliable method of screening broodfish for channel catfish virus.

MICROPOSRIDIAN: CAUSE OF PROLIFERATIVE GILL DISEASE (HAMBURGER GILL DISEASE) OF CATFISH

W.A. Rogers, Department of Fisheries and Allied Aquaculture, Auburn University, AL 36849

T. Miyazake, Faculty of Fisheries, Mie University 2-80, Edobashi, Tsu, Mie, Japan

Proliferative gill disease or hamburger gill disease is a condition in channel catfish where the gills are swollen, hemorrhagic, and have been compared to raw hamburger meat. Several causes of the condition have been postulated including the Myxosporidian *Henneguya* and poor water quality, i.e. ammonia.

Sections of gills of fish with the disease were found to have cysts of an unknown microsporidian primarily in the gills, but the cysts were also in the liver, spleen and kidney. Gills were most markedly affected with cysts. The organism has been observed in tissue sections of diseased channel catfish from Alabama, Mississippi and Arkansas. At first it seemed that the parasite belonged to the genus *Plistophora*, but it is apansporoblastic without sporocysts which does not fit *Plistophora*. It seems most closely related to the genus *Microsporidium*, but further work is needed to confirm its exact classification.

MONOCLONAL ANTIBODY AGAINST IHN VIRUS

C.L. Schultz, Department of Microbiology
University of Maryland, College Park, MD 20742

B.C. Lidgerding and P.E. McAllister, U.S. Fish and Wildlife Service, National Fish Health Research Laboratory
Kearneysville, WV 25430

F.M. Hetrick, Department of Microbiology
University of Maryland, College Park, MD 20742

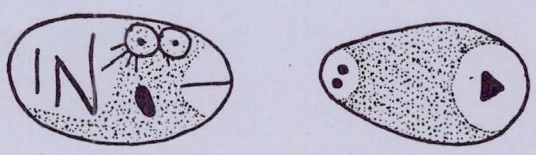
Immunological techniques that require defined antibody preparations as reagents are increasingly being used in the identification and detection of fish viruses. To date, antisera to fish viruses have been limited to the incompletely defined products made in animals, and, as anticipated, these preparations are less suitable than defined reagents for use in immunologically based assay systems. This is particularly evident when using antisera made against infectious hematopoietic necrosis virus (IHNV). To have a readily available source of antibody to IHNV, we have established á hybridoma cell line that secretes an IHNV specific IgG antibody. Concurrently, procedures were developed to purify and concentrate the virus specific antibody from the hybridoma culture fluids.

Fusion of IHNV primed BALB/c mouse splenic lymphocytes and the P2-X63-Ag8.653 continuous mouse myeloma cell line resulted in only one population of antibody producing cells being formed from the 562 cultures initiated. This antibody producing cell population has been cloned four times, resulting in numerous antibody secreting hybrid cell populations. Five of these populations have been established in continuous culture. The hybrid cells are grown in 10% CO_2 at 37C and require subcultivation every 4 to 7 days. The cells can be readily recovered from frozen stocks without loss of antibody production or growth potential.

The cells continually produce antibody and large volumes of culture fluids can be collected weekly. A protocol using affinity chromatography and ultrafiltration is used to routinely purify and concentrate the IgG from 400 - 500 mL of cell culture fluid. In ELISA tests, the purified IgG did not produce a reaction above background values with IPN and viral hemorrhagic septicemia viruses. In neutralization assays, the concentated IgG had a titer of only 1:50 against 50 $TCID_{50}$ of IHNV. The IgG from numerous hybrid cell populations was shown to be universally subtype 2b with a kappa light chain. These data indicate that the hybridoma IgG is highly specific for IHNV, binds rather than neutralizes the virus, and is of monoclonal origin.

Although the hybridoma produced IgG is predominately a non-neutralizing antibody, it is useful in ELISA type tests because neutralization is not required to indicate the presence of the virus. At present, the multilayer ELISA test for IHNV has reproducible detection limit of 30 - 300 ng of viral protein which is not as sensitive as cell culture assay. However, ELISA can be used to rapidly detect samples having high viral titers and thus reduce the number of samples that require cell culture assay for the detection of virus. The use of the ELISA is particularly advantageous when large numbers of samples must be assayed.

We are currently refining our avidin-biotin based multilayered ELISA system to lower the detection limits and further reduce the immune specific background that can occur during the processings of fish tissue samples.



HIGHLIGHTS OF U.S. FISH AND WILDLIFE SERVICE REGISTRATION ACTIVITIES INVOLVING FISHERY-USE CHEMICALS AND DRUGS* (APRIL—JUNE 1984)

David F. Walsh, Registration Liaison Officer, Division of Fishery Research, USFWS, Washington, D.C.

In early May 1984, the Food and Drug Administration (FDA) approved the Service's request to renew, for the 1984 salmon spawning runs, the Investigational New Animal Drug (INAD), #2573. The INAD allows the use of and requires data collection on malachite green at selected National Fish Hatcheries. Ten west- and four east-coast hatcheries will use approximately 300 pounds of malachite green to treat adult salmon spawners and their eggs. The increase of 60 pounds and the addition of eggs to the INAD reflects an increase in the need for control of fungal infections in our hatchery operations and an increase in the data we must gather. The clinical trial forms, used to tabulate information gathered under the INAD, were revised to enhance our data collection efforts. As with earlier permits, all hatcheries using and discharging malachite green are required to notify appropriate Federal or State offices regarding effluents associated with any potable water supply downstream or the disposal of spent activated carbon (the latter used in certain filtration systems). FDA indicated that the extension of this INAD is consistent with the public health and emphasized that all other aspects of the original authorization (May 1981) remained unchanged. We also submitted a report on a mutagenicity study and a publication on teratology studies performed on malachite green. These documents will become part of the complete record on malachite green.

Several documents have been added to the Service's rotenone files at the Environmental Protection Agency (EPA). In May, we submitted reports for studies on "Aerobic Metabolism of 14-C-Rotenone" and "Anaerobic Aquatic Metabolism of 14-C-Rotenone." These studies should meet requirements in the aquatic metabolism area. We requested a review of two protocols for studies in lactating dairy animals and we also requested a review of study protocols for determining accumulation of rotenone in rotational crops and the quantitation and characterization of 14-C-residues in crops irrigated with 14-C-rotenone treated water. The Service is interested in maintaining that portion of the label that permits the occasional use of rotenone-treated waters by livestock or for irrigating crops.

This office has been advised that makers of medicated fish feed have apparently been using generic sulfamerazine rather than purchasing the product American Cyanamid developed and labelled specifically for fishery use. The American Cyanamid product is the **only** formulation for which FDA approval exists. American Cyanamid advised us that they have sold no sulfamerazine under the fish NADA in the past four years. As a consequence, the company is questioning whether there is any reason to maintain the NADA. Should the company drop the NADA, fish food producers would no longer have sulfamerazine available. Apparently, this same practice has occurred in other animal husbandries. In a recent (July 5, 1984) Federal Register issue, FDA indicated that it was terminating certain approvals for sulfamerazine but, fishery uses were not included on the list. As a result, users of generic sulfamerazine were suddenly no longer able to obtain the drug, including makers of medicated fish feed. The American Cyanamid product will carry a higher price. This is inevitable because of costs incurred in obtaining the label, quality control expenses, and the small volume of sales. This tariff is merely a cost of doing business in today's regulatory climate. Attempts to circumvent the system seriously undermine the Service's efforts to obtain private industry's cooperation in developing needed chemical and drug registrations.

In another situation related to the cost of registered products, staff at the LaCrosse Laboratory have been negotiating with FDA and Natchez Animal Supply Co. to amend the NADA on formalin to permit a reduction in quality control requirements. If FDA approves of the proposed amendment, the cost of the labelled product would be reduced. Currently, the formulator is required to use stainless steel tanks, lines, and pumps in the packaging process and to use plastic lined barrels for shipment. The proposed amendment seeks approval for the presence of inert ingredients in the form of iron oxides at levels less than 0.5 percent in the marketed product.

**Use of trade names does not imply U.S. Government endorsement of commercial products.*

RECENT DEVELOPMENTS IN PKD RESEARCH

G.W. Klontz, Dept. of Forestry and Range Sciences, University of Idaho, Moscow, ID 83843

The 1984 PKD episode at the Hagerman Trout Hatchery, IDFG has presented some unexpected observations. To put it mildly, my theory about the sea gull (or other fish-eating migratory waterfowl) being involved has had some holes shot in it. I am not saying that I was wrong but my thinking certainly has been modified by this year's episode.

First, the episode began right on schedule. The organisms were first detected in late May on the Riley Creek side of the facility. There were no infected fish on the Tucker Springs side. Part of the Riley Creek side had bird wires which had some effect on keeping the gulls out. They hit the station pretty hard in late March and early April. By late July all ponds on the Riley Creek side had infected fish. The prevalences ranged from 5% to more than 30%. However, the mortalities to date have been very slight even though this year the *Sanguinicola* prevalence is very high.

Second, the fish in the head ditch supplying Riley Creek water to the facility were sampled by electroshocking. The majority of the fish were steelhead escapees from the Hagerman, NFH - identified by their LV clip. They had been in the head ditch for some time, presumably, because of their very low condition factor. None had eaten in some time. All were positive for the PKD organism. Now, how did they become infected? The head ditch is covered with mesh and the fish stay fairly much inside the culvert leading from the weir to the hatchery.

Third, we had 200 or so fish known to be harboring the organism transferred to the Moscow campus. They were transported in 16C water which had increased to 20C by the time they arrived. They were put into 10C water. The post-transport mortality was slight—12-15 fish died within a week after arrival—they were all positive for the organism. But, subsequent examination of these fish has indicated that the infection is waning. The organisms are undergoing a marked degeneration in the "hot" water. We inoculated some chicks (16 days old) via the esophagus and the cecum. This was on 31 July. To date none has any detectible PKD organism in the cecal contents or the feces.

Fourth, based upon these observations I enlisted the aid of an immunopathologist at the veterinary college at WSU. We both agree that the temperature-related decline in prevalence of the organism and the lack of organisms from the chicks could be influenced by the degree of immune response occurring in the affected trout. In our opinion the organism is being affected by the host's immune response. This belief comes from the fact that the beta-2-globulins are elevated in PKD-infected fish and also the macrophages "parasitized" by the organism are surrounded by small lymphocytes (perhaps these are T-cells).

Fifth, and most exciting, Cyzine (aka Enheptin-A) fed at the rate of 4 mg/kg feed for 7 days will markedly reduce the PKD organism population. The tests are currently being further evaluated for dose: response curves. The treated fish do appear to be less depressed than their untreated counterparts. We hope to have some definitive results by next Newsletter time—that is if the disease doesn't undergo remission before we finish. At this time we have no sure-fire way to keep the infection going past mid-to-late September.

THE EFFICACY OF PRAZIQUANTEL AND OTHER PHARMACEUTICALS AGAINST THE EYE FLUKE OF FISH, *DIPLOSTOMUM SPATHACEUM*

Richard Heckmann, Zoology Dept., 153 WIDB
Brigham Young University, Provo Utah, 84602

Praziquantel (Droncit®) was tested on mottled sculpin, *Cottus bairdi*, infected with the metacercarial stage of *Diplostomum spathaceum* during 1982 and 1983. Two other life stages of the eye fluke were also tested during the 1982 trials.

During 1983 eight other drugs—Mebendazole, Clorsulon, Ronidazole, Rafoxanide, Oltipraz, Oxamniquine, Thiabendazole, and Nicarbazin—were checked for efficacy against *Displostomum spathaceum* in fish.

Infected mottled sculpin were collected from the upper Salmon River, Obsidian, Idaho, and injected IM with 0.1 ml Droncit® (56.8 mg/ml injectable) and the other pharmaceuticals. Then 2 to 4 fish were killed 1, 2, 4, 6, 8, 10, and 24 hours after injection. The treated fish were compared with controls. Snails, *Physa* and *Lymnae*, were also collected from the same area for experimental trials with Praziquantel.

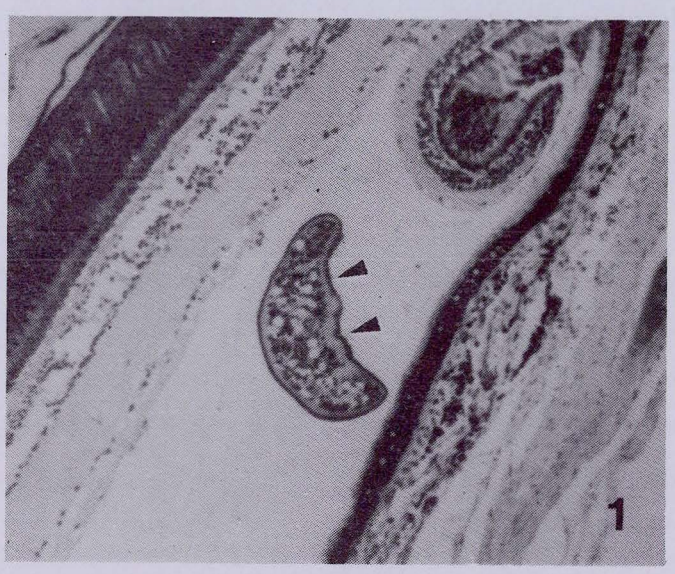
Droncit® is defintely an effective drug for destroying metacercariae (Figures 1 and 2), sporocysts, and cercariae of *D. spathaceum*. Droncit® has limited FDA approval which does not include food fish.

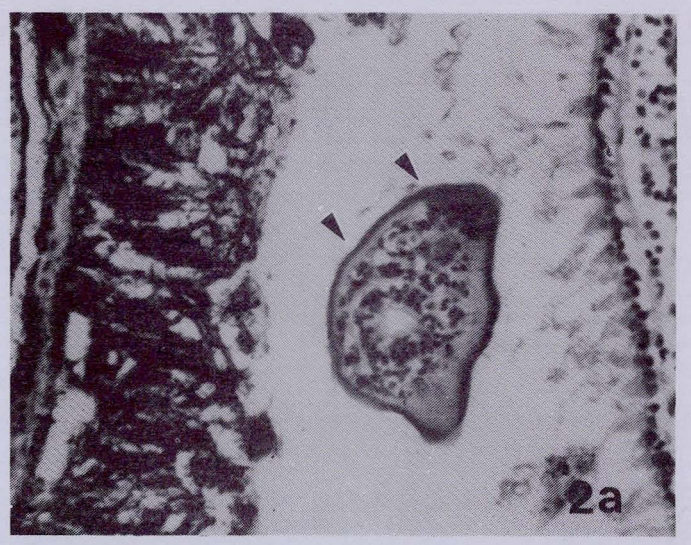
Fish exhibited excellent tolerance for the experimental drugs. Mebendazole and Oxamniquine are also quite effective in killing the metacercariae. The least effective drugs tested were Clorsulon and Nicarbazin.

Fish were also subjected to 100 times the amount of drug they would receive in the feed. No fish died during this experiment; thus, fish can tolerate all nine compounds.

Sections of metacercariae subjected to Praziquantel for 1, 2, 4, and 24 hours were prepared for electron microscopy and viewed with a high resolution transmission electron microscope. From the EM sections it was noted that the nucleus becomes condensed and the organelles become necrotic. There is excessive water and tissue loss from the integument and inner confines of the worm. This explains the shrinkage depicted by the light microscopy done previously for this study.

The use of pharmaceuticals should be the last resort for the management of diplostomatosis. Praziquantel and Mebendazole are efficacious in alleviating the disease in fish and snails. The major problem with both drugs is the expense of application and that neither drug has been cleared through the FDA for use with fish.





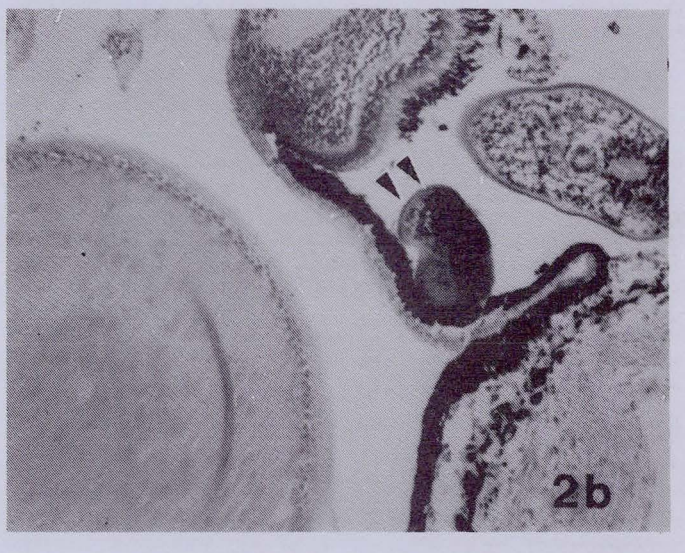


Figure 1: Metacercariae (arrows) of ***Diplostomum spathaceum*** *behind the retina of this infected sculpin. This is the control, no drug treatment.*

Figure 2: This figure shows the effect Droncit® has on the metacercariae 4 hours (2a) and 8 hours (2b) after injection. The worms (arrows) are shrinking and dying.

USE OF CALCIUM TO INCREASE SURVIVAL OF STRIPED BASS AND STRIPED BASS HYBRIDS DURING TRANSPORT

Dr. John M. Grizzle, Dept. of Fisheries and Allied Aquacultures, Auburn University, AL 36849

During 1983, 83% of the striped bass and all of the *Morone* hybrids from two Georgia hatcheries died during removal from rearing ponds and stocking in reservoirs; addition of NaCl and MS-222 to the water did not increase survival. In 1984, $CaCl_2$ was used to increase the total hardness of some ponds from 20 to 45-100 mg/l as $CaCO_3$ 5 days before harvest. $CaCl_2$ was used also to increase the hardness of the water from 10 to 70-200 mg/l as $CaCO_3$ for holding and transportion some of the fish after harvest. All groups of fish in water containing additional calcium had 80-99% survival compared to 16% survival for a group of fish without additional calcium in the water before or after harvest.

BRIEF REPORTS

Bacterial gill disease caused severe losses of rainbow trout sac fry at Norfork NFH, Arkansas in May-June 1984. This is only the second time that BGD has been observed in rainbow trout sac fry in Region 4. Jimmy E. Camper, Rt. 3, Box 71, Heber Springs, AR 72543.

In a 48-h toxicity test, Nitrofurazone was four times more toxic to channel catfish than to goldfish (19 mg/l compared to 76 mg/l). The channel catfish that survived exposure to 17 mg/l for 100 h had necrotic skin lesions similar to those previously reported for channel catfish exposed to Nifurpirinol. Goldfish that survived Nitrofurazone exposure had no necrotic lesions. John Grizzle, Dept. of Fisheries and Allied Aquaculture, Auburn University.

Glenn Hoffman had reported that the USFWS had Schaperclaus' 1979 2-volume book on fish diseases translated from German to English. The publisher, Akademie-Verlag of Eastern Berlin has made arrangements for the English version to be published by Amerind Publishing Company of India. No available date was issued. Fish Farming Experiment Station, USFWS, P.O. Box 860, Stuttgart, AR 72160.

The FDA sponsored a workshop on drugs in aquatic species, August 20, 1984 in Gaithersburg, Maryland. The conference objective was to present the status of current scientific knowledge on drug metabolism and disposition in aquatic animals. Presentations included overviews on *in vitro* and *in vivo* drug metabolism, effects of stress, drug fate and disposition, and pharmacokinetcs of specific drugs. FDA plans to publish the proceedings of the workshop. Dr. M.H. Beleau, College of Veterinary Medicine, Stoneville, MS 38776.

About 5-10% of the lake trout hatched this year at Dale Hollow NFH, TN, had an extra "dorsal" fin on the caudal peduncle. Most of the affected fish lacked an adipose fin. These lake trout eggs came from the Jordan River NFH, MI. This condition is believed to be genetic anomaly. Jimmy E. Camper, Rt. 3, Box 71, Heber Springs, AR 72543.

Glenn Hoffman has translations of the following books which are available free of charge on a first come basis: **Tapeworms of the Genus Triaenophorus** by Kuperman, **Ichthyology** by Poznanin, **Camallanata of Animals and Man and Diseases Caused by Them** by Folitarek, **Aquatic Oligochaeta Worms, Taxonomy, Ecology and Faunistic Studies in the USSR** by Belyaev. Fish Farming Experiment Station, P.O. Box 860, Stuttgart, AR 72160.

Dick Heckmann has a list of 19 abstracts presented at the 1984 American Society of Parasitologists Meeting held at Snowbird, UT. Dept. of Zoology, Brigham Young University, Provo, UT.

The new edition of Miscellaneous Special Publication 31, "Fish Health Protection Regulations: Manual of Compliance" (the Canadian Blue Book) has just been issued. It is available at no charge from: Department of Fisheries and Oceans, Fisheries Research Directorate, Aquaculture and Resource Development Branch, Ottawa, Ontario, Canada K1A OE6.

PKD has been found at a second salmon hatchery in B.C., in coho and steelhead at Robertson Creek on the west coast of Vancouver Island. Also, as before, fish were suffering from PKD and furunculosis. Losses were low but typical PKD could readily be found in random samples. A programmed drop in temperature was associated with a drop in mortality and typical PKD became more difficult to find. Gary Hoskins, Pacific Biological Station, Nanaimo, B.C.

We have produced a strain of *Renibacterium salmoninarum* resistant to 500 µg/ml erythromycin. The strain, originally susceptible to 0.5 µg/ml, retained its resistance after 6 passages on antibiotic-free medium. Work is continuing to determine the nature of this resistance and other properties of the resistant strain. Gordon Bell, Garth Traxler, Pacific Biological Station, Nanaimo, B.C.

REMEMBER TO FILL OUT THE MEMBERSHIP DIRECTORY INFORMATION AND RETURN IT TO ROWAN GOULD.

The grandparent clause concerning certification by the AFS/FHS as Fish Health Inspector or Certified Fish Pathologist will no longer exist after January 1, 1985. This is another reminder that after this date a written examination will be an additional requirement.

Paul Janeke, Fish Disease Control Center, Box 917
Ft. Morgan, CO 80701

(continued from page 9)

It is also my hope that reconsideration of the model program will involve some discussion of the necessity for equivalent levels of scrutiny and reporting of findings among all cooperators in the Great Lakes Fish Disease Control Program. We would hope that all hatcheries are being monitored in accord with recommended procedures and would expect that when agencies such as the Fish and Wildlife Service conduct diagnostic surveys for private sector hatcheries—either through sampling of production fish or through the use of "sentinel fish" of susceptible species placed in water supply or effluent—that the resulting information would be made readily available and freely exchanged among GLFDC members.

In closing, I appreciate the opportunity to propose amendment of the model program and look forward to a prompt and objective consideration of removing whirling disease caused by *M. cerebralis* from the categories of "Emergency" and "Certifiable" diseases. In the meantime, Fish Commission staff will, in accord with the monitoring procedures recommended by the GLFDC, continue surveillance of the Tylersville Station and, as a further measure, will conduct assessments in the drainage and effluent areas of both Tylersville and the Lamar unit of the National Fisheries Research and Development Laboratories. No stockings of Lake Erie will be conducted from the Tylersville unit until we are satisfied we meet the guidelines of the GLFDC program.

SPECIAL CONTRIBUTION

Ralph W. Abele, Executive Director
Pennsylvania Fish Commission, P.O. Box 1673
Harrisburg, PA

Recently the Pennsylvania Fish Commission stocked a tributary of Lake Erie with coho salmon that had been reared at the Tylersville Station. Tylersville is a former Fish and Wildlife Service facility which was turned over to the Pennsylvania Fish Commission due to the presence of *Myxosoma cerebralis*. Subsequent to this stocking, Tim Carey, Chairman of the Great Lakes Fish Disease Control Committee, called Vincent Mudrak of the Pennsylvania Fish Commission and expressed the Committee's concern about stocking fish from Tylersville into the Great Lakes. As a result of Tim Carey's concern, stockings were postponed and a meeting of Disease Control Committee members and Fish Commission staff was held in Erie, Pennsylvania, on April 12. (I believe minutes of that meeting will be available from Tim Carey should they be needed.)

After discussion of that meeting and of the general opinion of my technical staff toward the Model Fish Disease Control Program, I have reached several conclusions which I wish, via this letter, to formally communicate to the Secretariat, the Commission, and the Great Lakes Fish Disease Control Committee.

1. The remaining coho salmon at the Tylersville Station will not be stocked in the Great Lakes Basin.

   This action is taken voluntarily in support of the concept of consensus as expressed in the Joint Strategic Plan for Management of Great Lakes Fisheries. My staff did not view the stocking of these coho as a substantive change from existing practice nor did they consider these to be diseased or disease carrying fish. Samples of coho salmon and brown trout from Tylersville had been subjected to appropriate diagnostic procedures for detection of myxosporidian spores and no spores were found. The techniques used were those of O'Grodnick as outlined in "Fisheries and Environment Canada," Miscellaneous Publication 31, *Fish Health Protection Regulations, Manual of Compliance*. A follow-up analysis of the Tylersville coho samples was conducted by staff at the Fish and Wildlife's Leetown Diagnostic Center utilizing the techniques of Markiw and Wolf. No spores were found. The decision to not stock Tylersville coho in the Great Lakes Basin is made entirely on the basis of interagency cooperation and recognition of concerns of fellow signators to SGLFMP and should not be interpreted as any indication that the salmonids reared at the Tylersville Station are anything other than a healthy product entirely suitable and appropriate for use in any hatchery supported fishery.

2. The Fish Commission staff should have been more specific in calling the proposed stocking of fish from Tylersville to the attention of the Great Lakes Fish Disease Control Committee and we apologize for any distress or inconvenience this oversight may have caused.

   The presence of coho salmon at the Tylersville Station was included in the January report submitted to the Great Lakes Disease Control Committee. The coho had been checked for the presence of whirling disease. No one intended to deceive any other agency nor to stock fish in a clandestine or surreptitious manner. The failure to specifically "flag" the Tylersville coho as being destined for the Lake Erie drainage was an oversight, not a malicious nor intentional act. The distress and concern expressed by the Chairman of the Great Lakes Fish Disease Control Committee is understandable and we do apologize.

3. The Fish Commission proposes an amendment to the Model Fish Disease Control Program which would reclassify whirling disease caused by *M. cerebralis* from the categories (a) Emergency Diseases and (b) Certifiable Diseases to (c) Reportable Diseases.

The Model Fish Disease Control Program currently lists infectious pancreatic necrosis (IPN), bacterial kidney disease (BKD), and furunculosis as diseases which shall be monitored for observational and hatchery classification purposes. Given the body of knowledge now available on whirling disease, it seems entirely appropriate that whirling disease be categorized similarly to IPN. The classification of whirling disease as an emergency disease with a recommendation of eradication or, in the case of National Fish Hatcheries, closure of stations is reflective of the attitudes extant at the time the model program was conceived; but in the face of current knowledge about the disease and its effect on hatchery operations and free-living salmonid populations, this is a most unrealistic classification and attitude.

My staff felt that some members of the Disease Control Committee may have been surprised that the Fish Commission was not in agreement with the manner in which the model program addresses whirling disease.

Please accept my assurance that this is not a last-minute thought offered in rationalization for the rearing of coho at Tylersville. I call your attention to *A Guide to Integrated Fish Health Management in the Great Lakes Basin*, Special Publication 83-2 of the Great Lakes Fishery Commission, April 1983, specifically the section on whirling disease, page 226, and the quote from Joseph O'Grodnick:

> "Since *M. cerebralis* has already become established in certain geographic areas, policies of eradication are not practical and abandonment of existing facilities is not economically justifiable. An acceptable management alternative may be the rearing of proven resistant salmonids in contaminated hatcheries. State agencies could arrange production schedules so that resistant species which do not develop clinical whirling disease are substituted for susceptible rainbow trout or brook trout. Coho salmon, brown trout, and lake trout can be reared in contaminated hatcheries with no whirling disease development. The number of spores developed by these species is quite low and it would be unlikely that establishment of whirling disease in large water areas would be possible by stocking these fish. Also, hatchery effluent-receiving streams would receive fewer spores from very resistant production fish and contamination would be reduced. Since the value of the coho salmon and lake trout to the Great Lakes fishery is well documented, a careful evaluation of the policies regarding rearing these resistant species in known *M. cerebralis* contaminated waters should be made."

Mr. O'Grodnick's comments about the rearing of resistant species and an evaluation of policies relative to *M. cerebralis* reflect the official attitude of the Pennsylvania Fish Commission and are the basis for a formal request for modification of the model program.

It is my hope that amendment or modification of the Model Fish Disease Control Program will include recognition that *M. cerebralis* is already present in the Great Lakes Basin. (At least two tributaries to Lake Huron and one tributary to Lake Erie have been identified as having *M. cerebralis* present.) This recognition of the relatively long-term presence of the protozoan which causes whirling disease in tributaries to the Great Lakes should be a factor in the further recognition that this is not a disease which spreads rapidly and causes devastating or serious losses in modern, well managed fish hatcheries and in Great Lakes fish populations. We are not minimizing the need for concern about and reasonable precautions to prevent the spread of whirling disease. We are asking for a realistic appraisal of the disease, its actual impact on free-living and hatchery fish, and full recognition that the disease is present in some portions of the Great Lakes Basin.

(continued on page 8)

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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