ENVIRONMENTAL PROTECTION AGENCY
REGION VI
DALLAS, TEXAS

INVESTIGATION OF FISH KILLS

BUREAU OF SPORTS FISHERIES AND WILDLIFE
OKLAHOMA COOPERATIVE FISHERY UNIT
OKLAHOMA STATE UNIVERSITY
STILLWATER, OKLAHOMA

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FOREWORD

Floating dead fish have always been an indicator to the observer that something is not right with the environment. Since fish mortalities may result from a variety of causes, some of natural origin and some man-induced, a prompt, well organized investigation is necessary in order to provide the information needed to determine the cause of a fish kill and the source of the killing agent.

A fish kill investigation can be one of the most complex procedures encountered by agencies concerned with water pollution control. With the usual demand for prompt results, limited opportunities for obtaining proper information, the high degree of variability, and the many unknowns, a fish kill investigation can become a perplexing and frustrating task.

Therefore, Region VI, Environmental Protection Agency and the Oklahoma Cooperative Fishey Unit, Oklahoma State University, cosponsored a seminar dealing with various phases of fish kill investigations. Federal, state, local and industrial personnel from Region VI and nearby states interested in fish kills and their investigation were invited to participate. The purpose of the seminar was to disseminate and exchange knowledge and ideas concerning various phases of a fish kill investigation.

In this manual we have included a collection of papers that were presented during the seminar. In a few cases, papers could not be provided by the speakers. For the purpose of consistency and clarity minor editorial changes were made in some of the papers by the Seminar Director.

John E. Matthews
Seminar Director
Region VI
Environmental Protection Agency

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REMARKS TO THE OPENING SESSION OF THE FISH KILL INVESTIGATION SEMINAR. November 2, 1971, Oklahoma City, Oklahoma

by

Kenton Kirkpatrick

Assistant Director, Air and Water Programs
Environmental Protection Agency, Region VI, Dallas, Texas

It is my pleasure to be here this afternoon representing the Acting Regional Administrator of EPA, Region VI, Mr. Bill V. McFarland. Mr. McFarland asked me to relay his regrets to you that other committments prevent his being here himself. He is participating in the Galveston Bay Enforcement Conference in Houston.

So on his behalf and on behalf of all of EPA, welcome to the Manpower and Training Program's Fish Kill Investigation Seminar.

I feel that the Fish Kill Investigation Program is a vital part of the overall EPA operation. Statistics alone support this view. EPA's Annual Fish Killed Report for 1970, which is now at the printer, states that there were 634 recorded incidents in the United States involving more than 23 million fish. That represents a 36 percent increase over 1969 figures. The 1970 totals bring to 161 million the number of fish killed in 4,500 incidents throughout the country since reporting began in 1961.

This shocking mortality rate provides us with convincing evidence of the degradation of our Nation's waters. It also stresses the importance of investigating each incident thoroughly to determine the nature of the lethal pollutant.

Unfortunately, all too often the cause of the fish kill remains undertermined due to inadequate channels of reporting and methods of investigation. The EPA is only now beginning a serious effort to establish an effective program in cooperation with State and other Federal officials and concerned citizens.

During the next two days you will be discussing some of these deficiencies and the techniques for correcting them.

The EPA is involved in fish kill investigations on two levels. The initial concern is with the field investigation conducted by the Surveillance and Analysis Division.

When we are advised that a fish kill has occurred, a team of technical experts is dispatched immediately to the scene. But inaccurate and untimely reporting too frequently prevents effective investigation. The team obviously cannot function effectively when the cause of the kill has been washed away.

In an effort to improve the notification system, a program is being developed in Louisiana on an experimental basis. A special telephone has been installed in our Baton Rouge Facility and will be manned on a 24-hour basis to receive reports of fish kills. I might as well give you the number now because we are going to publicize it far and wide. It is area code 504, number 926-0188.

This number will be provided to all State and local government agencies which have the potential for obtaining knowledge of fish kills. Fishermen and other private citizens who might be aware of fish kills also will be provided with the number.

When they call, they will be asked a series of questions designed to obtain the basic information. When appropriate, a field team can then be sent out.

Also tied into the system will be the very effective early warning network established several years ago along the Lower Mississippi River by the Louisiana Department of Health and Stream Control Commission. This network is designed to alert operators of drinking water treatment plants taking water from the River when spills of dangerous pollutants occur. The in-take valves are then closed until the contaminated water has passed. Dead fish are a reliable indication that there is trouble.

It is hoped that this system will enable our field teams to reach the scene in time to evaluate the circumstances and determine the cause. This will make our reports, which are relayed to the States, more valuable.

Our analysis attempts to pin down the specific material involved and the source.

At this point our second-level concern is activated. When it is deemed necessary, the report is turned over to the Enforcement Division for whatever action is indicated.

In the event that legal action is to be taken, it is essential that the data be reliable. This again emphasizes the importance of the investigation.

What is the relationship between EPA and the States? The basic foundation of this relationship is established in the Federal Water Pollution Control Act. This law places the primary responsibility for pollution control in the hands of the State agencies. So as far as possible, EPA relies on the States to enforce water quality standards.

However, since expanding use of the 1899 Refuse Act, independent action by the Federal Government has grown. Pending Legislation would give EPA direct enforcement authority over all navigable waters rather than just interstate waters as at present.

When EPA proceeds on its own initiative, the State is kept fully informed of our intentions and activities.

Whether it is the State or Federal Government taking the lead, cooperation is essential to a successful program. But no matter how much cooperation is developed among the various concerned agencies, the program can only really be successful if the information about the incident is precise.

So we regard this seminar as not only an opportunity to exchange knowledge and ideas but as an opportunity to develop and improve our professional skills.

So again, welcome to the seminar and best wishes for a successful program.

A PREFACE FOR THE INVESTIGATION OF FISH KILLS

by

Kenneth M. Mackenthun

Acting Director, Division of Technical Support
Water Quality Office, Environmental Protection Agency

Since the beginning of water pollution investigations, floating dead fish have been a red flag to the observer that something is not right with the aquatic environment. Early in legislative history for water pollution control, many States adopted particular laws to assess damages from those who, through the introduction of foreign materials, would render the States' waterways toxic to fish and other aquatic life. Fish kills have been the subject of litigation in the courts and many times have served a useful purpose to stimulate and focus public concern and attention towards appropriate water pollution control.

The Federal government began a voluntary annual census of fish kills in June 1960. These pollution caused fish kills are reported through appropriate State officials to the Federal water pollution control authority (now the Water Quality Office within the Environmental Protection Agency) where the data are tabulated and analyzed. Since 1960 over 144 million fish have been reported killed in 4,200 incidents.

In 1969 alone an estimated 41,000,000 fish were reported killed in 45 States by identifiable pollution sources. Of the number of reports, industrial operations ranked first as a killer of fish, followed by agricultural, municipal, transportation and other operations in that order. As expected, the months of May through September ranked highest with the number of reported fish kills. August was the highest month with number of reports and December was the least. Indicative as these annual summaries are, there remained an urgent need to determine the specific cause of a particular kill so that the cause could be corrected and water quality enhanced in the future.

In a news release dated August 21, 1969, Commissioner Dominick of the Federal Water Pollution Control Administration said that the present voluntary program of reporting fish kills is no longer adequate in our stepped up overall campaign against water pollution. The old

system did not provide for immediate reporting of fish kill incidents and did not require any counter action by the FWPCA. Because fish kills indicate a violation of water quality standards or enforcement conference recommendations, it is particularly important that any sizable fish kill be reported and scientifically investigated. The Commissioner directed each Regional Director to take an active role in investigating fish kills, in determining their cause, in providing technical assistance on any immediate control measures or warnings that can be taken and in providing technical or other authorized assistance in designing control measures that would prevent a recurrence of the kill. Under this plan the Regional Offices of the FWPCA were to establish procedures with State water pollution control agencies to insure that they receive immediate reports of significant fish kills within each region. It is recognized that the primary responsibility for the investigation of fish kills rests with the States as is the case in all water pollution control matters. The Water Quality Office of EPA shares these responsibilities in interstate waters or in waters where there are Federal-State enforcement actions.

The proper investigation of a fish mortality is through a State-Federal partnership. The role of the Federal government is to offer technical assistance to the State in the investigation of kills to the extent that resources permit. In areas of Federal jurisdiction we are prepared to initiate independent action to achieve a swift and adequate determination of the cause of a kill and of the appropriate control and preventative measures if such action becomes necessary. Fish kill coordinators have been identified for each of our regional offices and in Headquarters. The coordinators are in a position to respond to requests for technical assistance from the States in any aspect of the fish kill investigation and to receive the immediate reports from the States on significant fish mortalities.

Obviously each fish death can not be investigated fully. Only significant fish kills should be reported. The definition of significance is a matter of judgement, but it should include mortalities that are important because of sport or commercial fish values, that result from a suspected negligent discharge or malfunction of a waste treatment device, or that show widespread environmental damage.

The immediate report received by the region and supplied by the State should include sufficient pertinent information to identify the fish kill area, the relative extent of the mortality, the kinds

of fish involved, whether or not technical assistance to investigate the cause has been requested, and who should be contacted to obtain additional information. This information is then transmitted to Headquarters by the Regions.

Follow up information developed in fish kill response investigations or otherwise acquired should be sent to the Regional coordinators and in turn to Headquarters of the Water Quality Office, as soon as possible. We are particularly interested in information concerning the cause of the kill and actions to alleviate the cause, to collect damages, and to prevent future incidents. To make this fish kill investigative partnership effective, we need to receive information on fish mortalities immediately, by telephone.

Recently we evaluated our records relating to the program of reporting and responding to significant fish kills. In this evaluation we choose a fisherman year - that period of time from September 1, 1969 to October 1, 1970. At the onset I will say that our efforts have not been enough and they must be improved. In this period of time 231 fish kills were reported to my office under this program. These reports accounted for approximately 50,000,000 fish; and 860 miles of stream and 10,700 acres of other waters were affected. Of the 231 reported incidents, 194 were investigated in some fashion. The States investigated 170 incidents and the Federal government assisted the States in their investigation of 21 incidents. There is some indication in the record that the States have requested and received more assistance from the Federal government in the investigation of fish kills during the latter half of our fisherman's year than during the first half. We hope that as this program matures and develops that assistance from the Federal government will be requested in a much greater number of investigations and that we will be able to provide the type of assistance that will be meaningful and definitive in determining the cause of fish deaths.

Initial laboratory support for the investigation of fish mortalities should come from any Water Quality Office laboratory within the Region where the mortality occurs. If this support is unable to respond to the needs of a particular investigation, additional special services can be obtained from the Analytical Quality Control Laboratory in Cincinnati, Ohio, the National Water Quality Laboratory in Duluth, Minnesota, and the Southeast Water Laboratory at Athens, Georgia. Analytical capabilities in pesticides and other organics, heavy and trace metals, oil identification, complex organic compounds and certain fish diseases and bioassay analyses are available at these laboratories.

The Bureau of Sport Fisheries and Wildlife maintains a small staff of disease specialists within the Division of Fish Hatcheries to provide diagnostic services to national fish hatcheries. As time permits, these hatchery biologists are available to assist other agencies in diagnosing fish diseases. Emergency inquiries for assistance may be directed to the nearest hatchery biologist. Such hatcheries are located in the States of Washington, California, Montana, Utah, Arkansas, Wisconsin, North Carolina, West Virginia, and Maine. In addition, there are two Bureau research laboratories that specialize in fish diseases. The Eastern Fish Disease Laboratory, Kearneysville, West Virginia 25430 serves the area east of the Mississippi River, and the Western Fish Disease Laboratory, Building 204, Sand Point Naval Air Station, Seattle, Washington 98115 serves the Western States. Except in emergency situations, requests for fish disease diagnostic services should be directed through Headquarters of the Water Quality Office.

As far as the art and science of fish kill investigations are concerned, one might say with truth that we are entering the age of the Renaissance. Historically we have advanced slowly and simetimes it seems but little since the days of Shelford, Richardson, Forbes, Juday, and Birge.

One of the greatest deterrents to the conclusive investigation of fish mortalities is our seeming inability to arrive at the scene soon enough. Speed is all important in the initial phases of any fish kill investigation. Early arrival at the scene, after a fish kill begins, usually determines the success of the investigator's evaluation of the problem. Observations of the actions of moribund fish, and the collection of appropriate samples at this time, are often a vital link to the arrival at a conclusive determination of the cause of death. Dead fish disintegrate rapidly, especially in hot weather and the cause of death may disappear or become unidentifiable within minutes. Thus, the system of early warning followed by speedy investigation is essential to a successful fish kill investigative program.

The degree of success of an investigation will depend to a large extent on the investigative preparedness of the party involved. Planning is an essential component of any successful enterprise and the successful investigation of fish mortalities is not an exception. Planning will decrease the interval of time between notification and arrival at the scene. Appropriate planning will insure the availability of proper equipment and testing facilities and it will decrease the confusion and uncertainty that sometimes is associated

with the analyses of collected samples. Planning will identify specific laboratory capabilities before the need for these occur. Planning will alert reserve resources should these be required by a particular incident.

The investigator should be ever mindful of the potential legal aspects of his investigation. The investigation should be conducted to insure that collected data will be acceptable to the courts and will withstand critical cross examination. Samples collected in the field and transported to a laboratory should be sealed and so identified by the sender that the chain of evidence always remains intact, and that it cannot be broken by opposing attorneys. When the samples are received in the laboratory they should be associated with a continuous log that records the date and exact time of each step in the analytical procedures and the person responsible for the samples during that step. When not in use, samples should be retained in a locked room. As an effective tool of the water pollution control program, the evidence collected by the investigator must withstand the scrutiny of the courts. Preparation of the investigative data in this manner will be an asset even though remedial actions and pollution abatement procedures are arrived at through informal negotiations rather than court action.

There are about six essential steps connected with the investigation of a fish mortality. The first is the collection of preliminary information to identify the problem and its extent in very specific terms. There follows three concurrently attacked phases that include physical observations, the collection of chemical data, and the collection of biological samples. Observations by an experienced investigator are invaluable to define the cause of a given kill. Perhaps no single factor is so important. Particularly noteworthy are the general appearance of the water, its color, clairty and whether sparkly or dull and lifeless. The presence of oil, excessive algae, unusual biotic growths such as sewage fungus, excessive amounts of higher aquatic plants and the color and appearance of rocks, Sticks or debris within the water are important considerations. Notes on the actions of fish prior to death are important, as well as observed actions of other organisms such as crayfish and other benthos. Often much can be learned regarding the severity of the kill to the aquatic environment by lifting a rock from the stream bed or a piece of debris from the water and noting the types of organisms that remain alive. The fifth component of an investigation is the analyses of the many collected samples. The last embodies the tabulation and interpretation of data and the preparation of the report. These are

equally important aspects of an investigation and, if it is to be successful, all must be performed with great care.

The collection of chemical and biological samples and data involve a number of technical considerations. The Division of Technical Support of the then Federal Water Pollution Control Administration published a 21 page booklet entitled, "Investigating Fish Mortalities." This publication considers some of the technical aspects of the sample and data collection. It may be obtained from any of our regional offices or in Headquarters. I commend it to your reading.

RULES OF EVIDENCE

by

Tony Anthony

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Environmental Protection Agency, Region VI, Dallas, Texas

The importance of evidence cannot be overemphasized. The procedure, the investigation, the methods, everything you will be hearing today, everything you probably studied about, everything you will learn in the future is all directed to the one key point of evidence.

In response to the question: how significant does a fish kill have to be before you go out and collect good evidence? If it is worth investigating, it is worth documenting. Those cases that get to court are frequently the ones with the lousiest information possible; the one where some guy goes out and looks at it over the weekend and didn't take any samples or didn't take any notes. When the case gets to court the lawyers are sort of hard put to try to make something out of it. So if it is worth investigating at all, it is worth doing a good job on it.

What is evidence? Evidence is really everything; a mass of information and data which is prepared, preserved, presented to a trier of fact in order for him to supposedly arrive at the truth of various allegations or charges. Now when I say it is everything, naturally I have to immediately start making exceptions. There are probably as many exceptions to what evidence is as there are rules trying to describe it.

One of the most well known exclusions of any evidence that I would point out is hearsay evidence. Here again there are numerous exceptions to the so called hearsay evidence rule. Basically, hearsay evidence is something that you do not know or something that cannot be presented as a hard, cold fact. For instance, I can see a container sitting on the front table. I can testify to the fact that there is a container sitting there. One of the gentlemen sitting there could tell me it was a container full of water or a pitcher of water. At the present time that would be

hearsay evidence as far as my testifying to the fact that there is a pitcher of water on the table. Here again I would immediately run into one of the exceptions to the hearsay rule. If the gentlemen who told me there was water in the pitcher sitting on the front table is also in court and can rebutt my statement, I can testify to the fact that that is water in the pitcher. But if he is nowhere around, and I just say something that someone else told me, it is excluded as admissible evidence.

What are some of the forms of evidence? Most of it, a good deal of it, is oral testimony or personal knowledge of a witness, someone who has seen or knows from his own personal knowledge about a particular fact. If I further investigate the container sitting on the table, it looks like water, it sounds like water, and it probably feels like water. But it may be gin, so I can't, at this point, testify that that is water in that pitcher. Now if I pour a glass of it, drink it and it is water, that is evidence that I can testify to.

Circumstantial evidence is used to a considerable extent by lawyers. Mainly because we do not have the factual evidence that we really need to tie one point to another. We can string a group or a line of individual facts that will lead to a particular assumption or interpretation. My interpretation right now is that there is water in the pitcher. Conceiveably, what I have seen here today could be circumstantial evidence that there is water in the pitcher.

We also have graphic evidence. Photographs are excellent examples of graphic evidence. If you are in an investigation of a particular water body that looks to you to have caused the fish kill, horrible looking, real bad stuff, get a picture of it. You see fish floating in it belly up, get a picture of it. Very recently in a federal court case in which I was participating, one of the witnesses had color slides of a particular body of water. He had put dye in this water and traced it down the waterway, taking pictures now and then. At one point it just so happened the dye was in the same field of view of his camera as three or four floating dead fish. He was projecting these slides on a screen for the judge. The minute that picture of those dead fish hit that projector the judge stopped everything, gently folded up his robes, and traipsed down to where he could get right in front of that screen and see the dead fish. This is beautiful evidence, it is hard to refute. Now there are problems of tying the evidence in with what caused their death. If you can document with photographs what you are investigating, your attorney can probably tie this into a pretty effectual presentation.

Demonstrative evidence is occasionally used in court. This ordinarily is within the discretion of the court itself as to whether it will be permitted or not. If we had, for instance, a case where it was suspected that ammonia was the source of death in a particular fish kill, conceiveably you could bring a small tank of fish in their natural water into court and insert a given amount of ammonia to demonstrate that these fish will be killed, or their gills will start swelling and bleeding. Be sure and check with your attorney so he can check with the court before this type of evidence is prepared or anticipated being used.

Another thing I want to point out is that one of your prime functions in this evidence area is educating your attorney. Some attorneys aren't exactly right up on their toes on all this technical jargon. You may have evidence coming out of your ears, but you must get over to your attorney and educate him as to the value of this evidence. It really is not going to do any good if he can't get it to the trier of facts.

Another particular type of evidence which may be considered is derived evidence, or calculated evidence. When a particular stream or water body is sampled and this sample taken back to the laboratory and analyzed, certain calculations may have to be made, e.g., it has to be measured against certain standards. The end result of these manipulations is more or less a calculated piece of evidence. You can demonstrate the reliability of the evidence if your procedure and method is reliable. If the very fact which you are trying to present is not so self evident that you can bring it in and set it on the court table, you must calculate something to show the trier of fact what you are trying to present to him.

Most witnesses in a court case such as a fish kill case probably will be just plain, straight, factual witnesses. An investigator who counts the fish and calculates the number of dead fish, who samples the water, who has taken photographs, can testify to the fact that he took the photographs, he sampled the water, he counted the fish. He may not be expert enough in a particular area to determine what caused the death of these fish. This is where we arrive at the role of an expert witness.

An expert witness is an expert in his given field. An ichthyologist for instance may be a specialist in a particular phase of the study of fish. He may have studied a considerable amount on the cause of death of certain fish. This man, once he is qualified by education

and experience, is allowed to make an interpretation from other factual data which he may not have personally picked up himself but which has been presented to the court. For instance, the investigator established as a fact that he has sampled a given water body, he has picked up dead fish and he has turned the samples over to a chemist. The chemist gets on the stand and testifies to the fact that he analyzed this particular sample and found x, y, z parameters in it. An expert witness could then get on the stand and from this information could make an interpretation to present to the court that these x, y, z parameters were probably the cause of death of these particular fish. An ordinary lay witness is not allowed to make an assumption or interpretation of this nature. An expert witness in biology is not permitted to make an interpretation in the field of chemistry, a field where he has no expertise.

The problem of what evidence is admissible is also one with which we have a struggle. In every case that comes up something is going to be objected to as being admissible or not admissible. Here again this is not primarily your function, but you should be aware enough of the problems the lawyer faces to be able to help him. As I mentioned before you may have a trunk full of data, but unless you can explain the significance of this data to the attorney who is handling the case he may not know whether that trunk full of data is admissible or not. In order for it to be admissible it must be at least relevant to the issue before the court. By relevance we might illustrate with the quality of water in a stream in which dead fish have been found. This is probably relevant to a fish kill because of the fact that fish ordinarily live in water. It may or may not be material if the fish are dead because they were seined and left on the bank to die. The quality of the water may be completely immaterial, if the quality of the water shows, for instance, certain parameters of a, b, c in the water, and the fish died from x, y, z causes. The amount of a, b, c in the water may be completely immaterial to the issue of where the x, y, z came from or what caused the end result or death from x, y, z. You can't just throw in everything you know about the water quality. If you found a dead horse in the stream, it may be relevant or material to the issue; it may not. You have to use a little of your judgement, and you have to help educate the attorney as to the particular significance of the information you are trying to get to the court.

Once a sample of water has been taken to a laboratory to be analyzed, those of you who are doing this analyses must always use a scientifically recognizable test in order to establish what was in this particular

water. If you have dreamed up a test all of your own when everybody else in the world is using standard methods, it may be the best test in the world but unless it is widely accepted within the profession, it will probably go to the weight of the evidence whether the trier of fact should accept it or not. So the results of your work must depend upon the acceptance by the scientific discipline in which you are endeavoring to establish these facts.

The chain of custody of samples is a very important part of any investigation, and it can't be expressed enough how important it is. If you are an investigator and you collect a sample of water from a particular stream or waste effluent, identify each sample as you take it with the date, the time of day, any witnesses that might have been there the same time and hopefully you have some witnesses, whether there was any preservative put in the sample, very definite information that would identify this particular sample as being taken from that particular spot. Each individual sample must be identified. Don't collect 25 different samples up and down the stream and them put the same tag on all of them. It can completely invalidate the results of any particular one.

Once you have picked up a sample, establish a regular, orderly procedure for transferring that sample to your analyst or to your lab. If you are doing it yourself establish methodical procedures for handling the sample. Do it that way every time. There may be several different ways of transferring this material, or sampling or even analyzing and they may all be equally valid, equally acceptable, but any one particular organization is going to be much better off in a court of law if they can establish that they have done this particular job the same way every time. For instance, I go out to take a sample of water, the parameters I need it tested for make it mandatory that I preserve this sample before I leave the site and take it back to the laboratory. It may be immaterial to the analysis of that sample whether I put the preservative in my container before I left the lab or whether I put the preservative in immediately after I have filled the container with sample. Assuming it is equally valid one way or the other, establish a procedure whereby you either always put the preservative in the container prior to sampling or you always put it into the sample after it has been taken. If one day you do it one way and the next day you do it the other, it is not too difficult a task for an attorney on the adverse side to establish that maybe you put in an extra dose or that you failed to put in any. This can tear up some awfully good evidence. So establish a set pattern. Do it the same way every time.

Once these samples have been collected keep a chain of custody tag with that sample at all times, with its identification on the outside of the container. Once you turn the sample over to your analyst, sign off on the tag, date it, put the time on it that you turned it over to this particular individual, have him sign off on it that he received the sample at that particular time. Then when he turns the sample over to the fish chemist this procedure must be followed through on the chain of custody. Each time the sample leaves one person's hands or custody it should be recorded so that the attorney can trace this sample all the way to the very final results, without having a gap somewhere. Once a gap can be found in a chain of custody it can tear hell out of the results, and you can lose the case. And that is not the name of the game.

If you must, in the transporting of the sample, turn it over to a common carrier, for instance an airline, have the agent of the carrier sign off on the custody tag. Have your custodian at the other end sign that he received the sample. You can't be held responsible for everything the agent does with it. There may be numerous other people who handle it in between, but that is not your responsibility. That is the responsibility of the common carrier, and the attorney can get to the root of that. He can bring the common carrier into court to establish the custody during that period of time.

Once you have analyzed and completed the work on your sample, don't toss it down the drain. Save enough of the sample so that it can be analyzed again for a duplication of results, in case it does come to court. Now I realize this causes trouble. There is not enough warehousing space anywhere in the country to house all the samples that have been taken. You can't keep them forever, but you can keep them or a usuable portion thereof, for a sufficient amount of time, before you destroy or throw them away. The amount of time will vary somewhat with each type of sample. Some types do not lend themselves to storage. So please establish some procedure for retaining samples. The sample that you throw away is frequently the one that is essential in the one case you are called on, and that is the one your attorney is going to be scratching his head trying to figure out, what do we do now?

The main enforcement responsibilities of the Environmental Protection Agency are related to the Water Pollution Control Act of 1965 and amendments. We do have several responsibilities set forth in the Act, one of which is the conduct of enforcement conferences in given

situations. Another is to file civil cases against water polluters under other given situations. The enforcement responsibilities of the Agency itself is to gather information or evidence and prepare it to be presented to the Justice Department, which presently handles all litigation for our Agency, in such a fashion that it is usuable and is as close to a sewed-up case as you can get. The Enforcement Division of the EPA has the final say as to whether an enforcement action will be undertaken or not. Surveillance and Analysis has the discretion of deciding whether there has been a significant enough set of circumstances to warrant, in their opinion, an enforcement action. They will at that time forward this particular incident to the Enforcement Division for review, and further action if it is an acceptable case.

Also, as you are well aware, EPA has the responsibility, not for enforcing the 1899 Refuse Act, but for preparing evidence and determining the water quality, and the extent of refuse which might be a violation under that particular Act. It is up to the Department of Justice to actually make the final determination whether a prosecution under that particular Act will be pursued or not. Once they get that determination they can take either civil or criminal action.

STATE LEGAL CONSIDERATIONS

by

Robert H. Mitchell

Assistant Attorney General
State of Oklahoma

The Attorney General's office is currently trying the first fish kill case in the State of Oklahoma in Payne County concerning a fish kill on the Cimarron River. We are unable to speak directly to all of the issues since many issues remain unresolved and have not been decided by the Court. Eventually the jury will decide the issue once the case goes to trial.

HISTORICAL BACKGROUND

One. Case of State v. Wheatley, which was one of the early pollution cases, wherein the defendant was charged by indictment and information under the criminal statute which provides one year imprisonment and $100 to $500 fine. The appeal was to the Court of Criminal Appeals, 1921. The case is significant because the Court recognized the Constitutional rights of the State to protect fish and wildlife under the policy power of the Constitution. They found that it was valid to charge an individual for the killing of fish under our Constitution.

Two. Pollution in Oklahoma has been a problem through the oil industry since it has been our major industry. In effect, there used to be associations of oil producers who would bind together and obtain easements from all land owners surrounding streams and lakes and oil fields and obtain from them, in effect, an easement to pollute. The land owner would sign an instrument, in return for money, stating that he would forego any legal action he might have in case of damage by salt water or crude oil or other deleterious substances.

Three. There of course are other cases, civil in nature, where individual land owners can sue for damage to domestic animals or property due to pollution.

Four. In 1968 the next move was made by the Oklahoma Legislature in setting up the Pollution Control Coordinating Act.

The Oklahoma Statutes, 82 O.S. 931 to 942 hold the composition of the Department of Pollution or the Pollution Coordinating Control Board.

PERTINENT STATUTORY AUTHORITY FOR LEGAL ACTION IN FISH KILL CASES

Title 82, O.S. 931 to 942 - Organization of a pollution control coordinating board and legal enforcement measures.

Composition of the Department of Pollution or the Pollution Control Coordinating Board, nine members, (a) Commissioner of Health, (b) President of the State Board of Agriculture, (c) Director of the Water Resources Board, (d) Director of Department of Wildlife Conservation, (e) Chairman of the Oklahoma Corporation Commission, (f) Director of Industrial Development and Park Department, (g) Executive Director of Soil Conservation Board, and (h) Two members appointed by the Governor with the advice and consent of the Senate ... (i) the Attorney General of the State of Oklahoma shall serve as the legal counsel for the Department of Pollution Control and shall assist the Board in the performance of its powers and duties designated by the Act.

First example of legal authority in 932.3 - Any duly authorized representative of the Board may enter and inspect any property to ascertain the state of compliance with the law and rules and regulations of the Board.

Title 82 O.S. 936. A Board has hearing on charges of violations and authority to issue orders on notice.

Title 82 O.S. 937. Penalties and Injunctions.

Violation of an order - $500.00 fine per day first ten continuous days and $1,000 for each day thereafter or imprisonment for a term of not more than 90 days or both such fine and imprisonment. Each and every day the violation occurs will constitute a separate violation.

The offender shall be liable to pay the State an amount equal to the sum of money reasonably necessary to restock such waters or replinish such wildlife and all costs incurred in investigating, locating or establishing the responsible person, firm or corporation as determined by the Oklahoma Wildlife Conservation Commission and approved by the Board. Such amount recovered by the Board on behalf

of the State in a civil action brought in the District Court shall be divided among the agencies in accordance with expenses incurred as determined by the Board.

It shall be the duty of the Attorney General to take any action, including an action for injunction.

This Act does not obviate other statutory provisions such as criminal provisions for pollution of land.

The State of Oklahoma Ex Rel Pollution Coordinating Control Board v. Kerr McGee.

A. Measure of Damages.

B. Punitive Damages.

C. Injunction.

A task force has been formed from this Act composed of individuals from each department with individual duties to perform in the case of a fish kill alert. For example, the chemists from the Health Department are to analyze water samples, the game rangers from the Department of Wildlife are to count the number of fish, and the other duties are separated out as will be noted by the head of the Task Force, Glen Sullivan, in a later speech at the Seminar.

From the lawyer's standpoint, important evidence that we want in addition to the chemical analysis of the water and fish, is an area inspection, including any possible back water areas where fish may have been effected, aerial photos and photographic maps, both aerial and otherwise. We want statements from the inhabitants of the area and employees of the possible pollution source. We need weather reports from the area and temperature reports from the stream or lake. We want samples taken from the riverbed or the banks of the lakes. In any event, we want all of the evidence prepared as if we were going to court on every case.

In the Cimarron River Fish Kill Case we are attempting to sue for punitive damages, although it is not provided for in the statute, on the theory that gross negligence of the polluters was as such as to amount to an intentional act; therefore, they are liable for punitive damages. Otherwise, we are following the statute which allows us to sue for the cost of replacement of the fish.

The end result of these lawsuits are not for political magnification or for monetary gain, but to insure cooperation between city, state

and federal agencies and private industry to protect our environment. The real reason, and the only reason for enforcing pollution laws is to ensure that our environment will be protected and that eventually through the cooperation of all of us, its existence will be insured for generations to come.

STEPS IN ESTABLISHING GOOD PUBLIC RELATIONS WITH THE MEDIA AND THE PUBLIC

by

Diane Norvell

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Communication is not one-sided. In order to be a complete model, it must be viewed from both ends. And when acting in the capacity of a state or public agency, it is most important that the agency sees how its publics view its efforts as well as how the agency sees itself.

How does John Q. Public measure the accomplishments? Does he visit with the various agencies to see its facilities, staff and latest programs? Does he peruse the latest annual reports to bring him up to date? Likely not, as much as we hate to admit. Instead, he usually gets a capsule of news from reading the headlines and first paragraphs of his morning newspapers, or hearing the spot news on the radio, or watching the 10 o'clock news before retiring to bed. Consequently, media does play a strong role in influencing Mr. John Q. Even the politicians, our representatives of federal, state and local governments admitted its strength by spending millions of dollars in the last general election in advertising to win Mr. Public.

With the population explosion comes the fight for brotherhood, population zero, and last but not least pollution. These are all examples of man's need to be socially responsible. Man wants to breathe clean air, drink clean water and most of all enjoy recreational activities. An outgrowth of these needs comes regulations pertaining to restrictions on air emissions, solid waste, and discharges of waste into the streams.

Since Mr. Public is not a professional environmentalist but is concerned, he wants to point the legal finger at those people who take away his wildlife. He wants polluters to pay for causing fish kills.

And what is your role in reporting the latest results of fish kills? As I see it, your responsibility is:

1. Reporting the fish kill.

2. Reporting it in such a manner as he will understand.

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*Former reporter with the Oklahoma Environmental Reporter, Oklahoma City, Oklahoma.

3. Letting him know in some situations there is no criminal but Mother Nature herself, as in the case of the Oklahoma droughts.

4. In the process, let Mr. Public know what you are doing about preventing and abating fish kills.

This is also a good time to put in a plug about your facilities and if you need a more sophisticated lab or investigation tools let him know. It would mean more dollars and cents when appropriation considerations come before Congress and the state legislature.

Here is an outline in establishing good public relations:

I. ORGANIZATION OF PUBLIC INFORMATION SOURCES

A. Set up a central public information system.

1. Name a public information officer (PIO) to organize and write all press releases for your agency.

2. In the case of fish kills and other incidents which require the efforts of several state agencies, an expedient way of releasing information is to appoint one PIO to funnel information to the press. This system would save overlapping stories and man hours to boot.

3. If the coordinated system is adopted, make sure participating agencies refer the press corps to a particular spokesman who will speak for the group.

4. If the coordinated system is not used, only release information on your particular aspect of the fish kill investigation and try to keep from making comments to the press which would cast a shadow over the other agencies.

5. By all means don't keep your PIO man out in the cold. Keep him up-to-date on the latest investigation results and give him guidelines on what information he is to release. An old story is not NEWS.

6. Keep a file on all fish kill stories. This collection will aid your agency in handling other fish kills. You can use the file for composing a thorough annual report.

7. Give your PIO man feedback. Criticize his stories and give him ideas for writing better releases. Don't forget to compliment him when he writes 'Pulitizer Prize Winning' articles.

B. Set up a working relationship with the press corps.

1. Know the reporters who cover conservation activities, for they write stories read by a large number of subscribers.

2. Abide by newspaper deadlines so your story will be timely.

3. Don't make the article too technical. If the reporter doesn't understand the concept, neither will the reader.

II. LEGAL LIMITS

Premature fish kill investigation reports to press may harm the court suit. Careful analysis should be made when releasing information on the following:

A. Fish kill losses. Don't get locked in with a specific number.

B. Give possible pollution sources but be leary in naming alleged polluters until formal charges are brought.

As you can see, communication is being able to relate your ideas effectively and in such a manner that those people who receive transmission will understand your thoughts.

ALABAMA PROGRAM FOR THE INVESTIGATION OF FISH KILLS

by

Sam L. Spencer
Chief, Fisheries Section, Game and Fish Division
Alabama Dept. of Conservation

In Alabama, we have a two phase program for the investigation of fish kills. This consists of the Alabama Water Improvement Commission (AWIC) and the Game and Fish Division of the Alabama Department of Conservation.

AUTHORITY TO CONTROL POLLUTION

The authority to investigate and control water pollution is vested primarily in the AWIC; however, the Game and Fish Division has been designated to assist, and damages collected are to be used by Game and Fish for "improving fishing in the affected area".

The AWIC has a central office in Montgomery and a Mobile lab for extended studies. At present they have no field offices. The Game and Fish Division has approximately 130 field personnel and are better situated to investigate fish kills. We have 105 Conservation Officers, 12 field Game Biologists and 13 field Fisheries Biologists. We have two-way radio contact with these field personnel through a state-wide radio network. In addition, an airplane is usually available when needed. The fisheries biologists have the primary responsibility for investigating the fish kills, however, other field personnel are available if needed.

REPORT OF FISH KILL

In Alabama fish kills are normally reported in one of four ways.

1. Reported by citizens to Game and Fish personnel.
2. Reported by citizens to AWIC personnel.
3. Reported by industries and municipalities to AWIC.
4. Observed by field personnel.

We have plans for a leased telephone line so individuals can report fish kills at any time of day or night. Upon receipt of notice of a fish kill, the informants name, telephone number and location of kill is reported to the AWIC, the Alabama Department of Conservation officials and to the district fisheries biologist nearest the kill. Fish kills are our highest priority item and every effort is made to reach the kill as soon after it is reported as possible.

INVESTIGATION OF FISH KILLS

Ideally a fish kill is investigated by both the AWIC and the Game and Fish Division. The AWIC technical personnel would collect water and fish samples, conduct basic water chemistry and attempt to determine the cause of the kill. The fisheries biologist would determine number, size, species and location of dead fish or other aquatic organisms. They would check for possible causes unrelated to pollution such as disease and parasite problems. When this work is completed, they would assist AWIC personnel with their investigation. Assistance is often rendered by other agencies such as the Tennessee Valley Authority, and the Environmental Protection Agency. The Environmental Protection Agency has been most helpful in analyzing samples collected during fish kills. Often the Game and Fish personnel is the only person on the kill and it falls his lot to conduct the entire investigation.

REPORTS

After the kill, a written report is prepared by the fisheries biologists and the AWIC personnel that conducted the investigations. A copy of each report is submitted to the State Attorney General's office for review. The reports are reviewed at regular meetings of the AWIC board.

LAW SUITS

Potential law suits are usually referred to the Attorney General for decision and legal action. Such suits are handled by the State Attorney General's office. Under a new law the Attorney General has the power to independently enforce provisions of the pollution law if the AWIC fails to. Suits may be filed for compensatory damages for the value of the fish in cases where negligence is suspected. Punitive damages may be pursued in cases of willful or wanton destruction of fish. This is almost impossible to prove. Under our new law the penalty for violation of water pollution laws is from $100 to $10,000 and each act or day is a separate offense.

Another significant change in our new law is the provision that "If the pollution has caused damage to fish and/or other wildlife in excess of $5,000, as determined by the Department of Conservation, the damage shall be presumed to have been the direct and proximate result of negligence of the person shown to be responsible for such pollution and the burden shall then be upon such person to prove freedom of negligence in causing the pollution in such cases."

THE AWIC - PAST AND PRESENT

In the last session of our State Legislature the make-up of the AWIC was drastically changed. The old commission was originally set up by law in 1949, changed slightly in 1953 and again in 1965. The commission was composed of 14 members as follows:

1. Chairman - State Health Officer
2. Vice Chairman - Director of Conservation
3. Commissioner of Agriculture and Industries
4. State Geologist
5. One representative of municipal government
6. One representative of county government
7 & 8. Two representatives of wildlife conservation
9. One representative of the mining industry
10. One representative of the textile industry
11. One representative of the chemical industry
12. One representative of the lumber industry
13. One representative of the paper industry
14. One representative of the metals industry

It has often been stated that in Alabama we had the "foxes guarding the hen house."

The commission also included a technical staff of merit system employees composed mainly of engineers, chemists, and aquatic biologists.

This commission was abolished effective October 1, 1971, by our State Legislature. The members of the new commission are as listed below:

Non-Voting Members:

1. Chairman - State Health Officer

2. Vice Chairman - Director of Conservation

Voting Members:

1. Physician qualified in water-borne diseases

2. Professional engineer qualified in water resource management and water supply

3. Attorney qualified in fields of water supply and riparian rights.

4. Two residents of the State for two years with no specialized experience required.

It was further specified by law that the members of the commission could be from industry but not from industries that discharge effluent into the public waters of the State. The voting members serve four year staggered terms, are chosen by the Governor and serve at his pleasure. He has the power to replace any member at any time.

PRESENT NEEDS IN ALABAMA

We feel we have a good setup for investigating fish kills in Alabama. The main items in my opinion where improvements are desired are as follows:

1. The appointment of commission members that favor clean water. At this time, the members have not been appointed yet.

2. The AWIC needs additional funds to establish district offices. This would improve detection and investigation of the kills.

3. A few more successful law suits may be helpful in reminding these few industries that are slow to clean up that the State of Alabama plans to protect its aquatic resources.

ARKANSAS FISH KILL INVESTIGATION PROGRAM

by

Neil Woomer

Arkansas Dept. of Pollution Control and Ecology

Officially, the responsibility and authority for investigating pollution caused fish kills in Arkansas rests entirely with the Arkansas Department of Pollution Control and Ecology. As a practical matter, such investigations are carried out with the aid of the Arkansas Game and Fish Commission, particularly in the areas of reporting of fish kills and making damage evaluations.

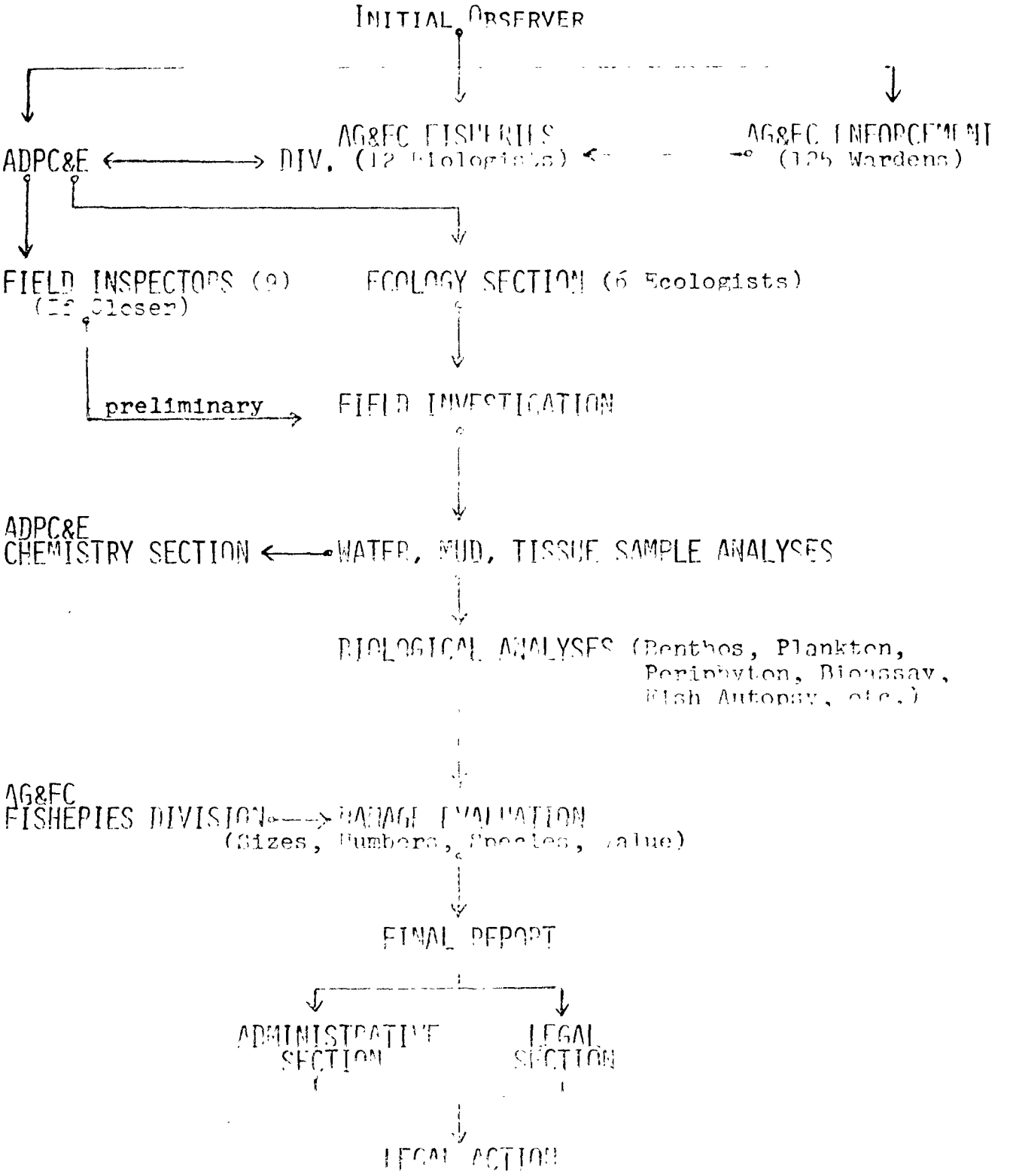
Referring to the attached flow diagram, fish kill investigations are carried out in the following way. Beginning with the initial observer, the fish kill is nearly always reported to either a Game and Fish Enforcement Officer, of which there are 125 spread throughout the State; to the Game and Fish Commission Fisheries Division, which has 12 District Fisheries Biologists; or directly to the Department of Pollution Control and Ecology, which has 12 Field Inspectors throughout the State and 6 Ecologists in the Central Office at Little Rock. If an Enforcement Officer receives the report, he radios the Fisheries Division which then contacts Pollution Control. If Pollution Control gets the report first, the Fisheries Division is immediately notified.

Once the report has reached the ADPC & E Ecology Section, an attempt is made to contact the nearest District Field Inspector. If this can be done and he can reach the site of the kill more quickly than someone from Little Rock, he makes a preliminary investigation, gathering pertinent information and taking the all-important first set of samples. If no Inspector is available a Biologist is immediately dispatched from Little Rock. All fish kill reports are investigated. If the preliminary investigation shows that the fish kill is substantial, the Inspector immediately calls Little Rock and a team of biologists go to the scene to do a complete work-up. After the field investigation is completed, all water samples are turned over to the ADPC & E Chemistry Section, which is capable of doing all necessary analyses, and all biological samples are analyzed by the Ecology Section. Meanwhile, the Arkansas Game & Fish Commission Fisheries Biologists make a complete damage evaluation which is submitted to the investigating Pollution Control Biologist who then writes a complete final report that is turned over to the legal and administrative people who then proceed with appropriate legal action.

Violation of the Arkansas Pollution Control Law is a misdemeanor and is punishable by a maximum fine of $250.00 or up to one year in prison. Arkansas has no law enabling the State to recover damages caused to its natural resources by pollution, although the industries involved in several fish kills have more or less voluntarily restocked fish equal in value to the fish killed by their discharges. Such a law is very badly needed in Arkansas.

The only other major problem that lessens the effectiveness of the Arkansas program is that of communication. We have no radios and the only way Field Inspectors can be contacted is when they are in their offices, which they seldom are. We need radios for the Inspectors and for the Ecology Section vechicles to tie in with the Arkansas Game and Fish Commission radio network which is Statewide. If this can be done and an adequate damage law passed, Arkansas will have an ideal fish kill investigation program.

ARKANSAS FISH KILL INVESTIGATION PROCEDURES



INVESTIGATION OF FISH KILLS IN MISSOURI

by

Frank Ryck

Missouri Department of Conservation

The Missouri Department of Conservation is the Wildlife regulatory agency and the Missouri Water Pollution Board is the agency having legal responsibility for pollution control. Within the Conservation Department, the Water Quality Branch of the Fisheries Research Section is most directly involved in fish kill investigations. At the present time, the Water Quality Branch is composed of a Chemist - Supervisor and two Fisheries Biologists.

There are basically 5 sources of original information of fish kills that occur in Missouri. Most fish kills are reported to the Water Quality Branch by Conservation Agents. The Conservation Department has 128 Agents - one in each county. The Agent has an intimate knowledge of his county and is in close personal contact with concerned, conservation-minded citizens. These citizens provide the Agent with most of the initial reports of fish kills. The Department of Conservation has 8 Fisheries Management Biologists whose duties bring them in close contact with the Conservation Agents and concerned citizens. We frequently receive notification of fish kills from the Management Biologists. The Water Pollution Board sometimes receives the initial report of fish kills from industries, municipalities, or other state agencies. In a number of fish kills, the first report has come from direct observations by members of the Fisheries Research Section.

When notification of a fish kill is received by the Water Pollution Board or the Conservation Department the other agency is immediately notified. The Water Pollution Board notifies the Environmental Protection Agency, the Missouri Division of Health, and any other agencies that may be concerned. In cases of very large fish kills the Conservation Department also notifies the Environmental Protection Agency. After this process of notification a joint investigation is initiated by the Conservation Department and the Water Pollution Board.

A typical fish kill investigation begins with the notification of Water Quality personnel. The person reporting the fish kill is expected to have roughly determined the distance of stream affected, the seriousness of the kill, and possible sources of the pollution.

He is also expected to have collected water samples above, in, and below the kill zone. If possible, a sample of the pollutant is collected. Water Quality and Water Pollution Board personnel proceed to the kill site and meet with the reporting personnel. Coordination of movement is facilitated through the use of radio-equipped cars and airplanes. County files are consulted before any investigation is initiated to help pin-point possible sources of pollutants.

Once Water Pollution Board and Water Quality personnel are at the scene of the kill a full scale investigation is begun. Additional water samples are collected, and field analyses are run to determine the following parameters: temperature, pH, NH_3, dissolved oxygen, turbidity, acidity or alkalinity, conductivity. Concurrently, sample counts of dead fish are made, and estimates of the number of fish killed are based on these counts. The size and species of dead fish in the counts are established and monetary damage estimates are calculated by consulting the publication of the Pollution Committee of the Southern Division of American Fishery Society relating to Monetary Values of Fishes. Whenever possible, post-kill benthos and fish population studies are made to determine the types and numbers of organisms that survive. Water samples are transported to the laboratory where Water Quality personnel conduct additional analysis, and conduct bioassays. Samples suspected of containing heavy metals, or pesticides are turned over to the Water Pollution Board or the Fish Pesticide Laboratory for analysis. When the investigation is completed a report is written. Copies are filed with the Water Pollution Board, Conservation Agents, and the Attorney General's office. When possible, criminal charges are filed with the county Prosecuting Attorney against the pollutor for violation of the Wildlife Code of Missouri. The Attorney General's office is routinely filing for damages to the State whenever fish are killed.

NON-TOXICANT RELATED FISH MORTALITIES

Introductory Remarks

by

Dr. Robert C. Summerfelt

Leader, Oklahoma Cooperative Fishery Unit
Oklahoma State University, Stillwater, Oklahoma

This seminar is designed to serve as a forum for dissemination of knowledge and for exchanging views on the complex problem of fish kills for State and Federal agencies responsible for protecting our health, environment, and fish and wildlife resources. The focus of the conference is fish centered, however, not because most agency representatives attending the conference have the responsibilities to protect fisheries as valuable natural resources, but, because dead and moribund fishes are conspicuous indicators (barometers) of the state of an environment which needs monitoring for our survival, livelihood and general well-being.

We have heard it mentioned many times that a fish kill is a conspicuous red flag but the real value of this signal is often overlooked even by communities with intakes on major rivers susceptible to environmental contaminants. Keeping an eye out for a fish kill on a water supply is vital to the community because our rivers are arteries of a vital life support system for which dead and dying fish serve as an early warning system. Investigations of fish kills should be as vital to a municipality as caged canaries were to coal miners. The latter carried canaries into labyrinths of the subterranena tunnels they worked, and, so long as the canaries were alert, the men did not fear asphyxiation for the dreaded coal gas because they knew the birds would collapse from the coal gas before the concentration became fatal.

Our conference deals head-on with the causes of fish kills but few attending this seminar subscribe to a view that the absence of conspicuous fish kills is an indication of a healthy environment. An analogy will clarify the point:

"If a tree falls in the forest but unseen by man, does it make a sound?" No! By definition, sound is a vibration that stimulates the auditory nerves and produces sensations of hearing. If man is not there to hear, sound is not produced, and vis-a-vis

the environment, an absence of a fish kill means losses have not occurred and all is well. This line of reasoning mimics the expression, "What you don't know won't hurt you."

In looking only at fish kills we are looking at only the part of the iceberg which is above water. More radical changes may be associated with gradual attrition and changes in species composition of communities than the changes associated with a fish kill.

It seems that more profound environmental factors affecting fish populations operate in a slow or not easily perceived manner.

Evolution, for example is a process of extinction and speciation resulting from gradual changes and replacement. The most well-known example of the effect of human perturbation on evolution is the case of industrial melanism in the "peppered moth" (Biston betularia) which occurs in light (peppered) and dark (melanic) forms. In 1848 the dark type was very rare near Manchester, England, but in years thereafter, the relative frequency of the dark form was observed to increase until today, only the dark forms are found near Manchester. Prior to industrialization, lichens covering the trees give the surface a variegated surface against which the lightly peppered moth was hard to see and the dark form quite conspicuous, but with industrialization based on consumption of fossil fuels, tree trunks blackened by soot, made the light moth very conspicuous but concealed the dark moth. Industrialization resulted in a change in gene frequencies in the population because it reversed natural selection (H. B. D. Kettlewell). Carried far enough, differences between the first and last series of hereditarily related organisms can lead to speciation, but more importantly, it leads to changes in species composition in a population because just as the survival of the dark form was competitively better than the white under the altered conditions, changes in the environment can favor other species, species which are more resistant.

In natural lakes and reservoirs, gradual eutrophication favors carp, buffalo, gar and bullheads and a concomittant reduction or elimination of bass, bluegill, walleye, and other desirable species. Survival and development of pesticide resistance often is associated with a partial loss of other elements of physiological vigor, or less fit for natural stress.

My next examples relate to problems of reproduction, although it is obvious that every species requires successful reproduction and survival of at least two young from every set of parents, reproductive

failures resulting from environmental conditions are often overlooked as important problems. Poor recruitment of largemouth bass in a certain Oklahoma reservoir, barely enough to sustain the population, has been found to be due to wind induced turbulence and lack of suitably protected spawning sites. Poor recruitment of largemouth and smallmouth bass in a certain Oklahoma river has occurred because releases from a hydroelectric plant, with penstocks located in the hypolimnion, maintains a water temperature too low to allow spawning. Fish have a sufficient high fecundity and have evolved with natural stresses and survived natural catastrophes, but persistent, sublethal effects can change and destroy natural ecosystems and fisheries resources with far greater effect than the natural stress.

The examples are those of insiduous environmental induced changes caused by man's perturbation of natural ecosystems. No fish kills need to be involved, yet the effects are very pronounced. I know that you are aware of similar cases of environmental alterations which have not produced fish kills, but need critical assessment. Legal liability for insiduous changes, although as I believe are of greater magnitude than the fish kill, are not adequately covered by the existing administration-legal framework which operates to assess losses related to costs of dead fish observed in massive kills.

Small or reoccurring kills are often neglected, receive only a cursory investigation, and the concerned agencies are by necessity forced to concentrate on the major kills. Habitat degredation is rarely investigated and there are few legal precedents for compensatory charges for habitat alteration. Although it is a violation to throw or allow to run into waters any substance injurious to fish, the laws usually provide penalties based on the number of fish killed. In some cases, it allows a penalty for a condition which would kill fish even where no fish are killed, however, this still doesn't apply to the situation of habitat degeneration.

Regarding penalties and compensatory payments for damages it may be biologically unsound to replace the fish except that these costs are symbolic or representative of the environmental degredation.

NATURAL VS MAN-INDUCED MORTALITIES

Fish mortalities result from a variety of causes, some of natural origin and some man-induced, a prompt, well organized investigation is necessary in order to provide the information needed to determine the cause of the fish kill and the source of the killing agent as well as to alleviate the possibility of reoccurrence. Fish kills

as used in this conference refers to sudden mortalities, not chronic losses, and mortalities of sufficient magnitude because most small fish kills are inadequately investigated. Moreover, fish kills in this conference refers to the mortalities, large or small, due to natural or man-induced. Unfortunately, some investigators make a distinction between epizootics and other "natural" mortalities as fish "die-offs", and mortalities which are man-induced as "fish-kills". This leads to a rather confusing situation in an investigation where occurrence of dying fish cannot be called a fish kill unless you know it is man-induced, and you do not call it a fish-die-off unless you know it is attributable to natural causes. I personally favor use of fish kills as a general term for both types.

NATURAL VARIATIONS IN FISH POPULATIONS

by

Austin K. Andrews

Bureau of Sport Fisheries and Wildlife
Oklahoma Cooperative Fishery Unit
Oklahoma State University, Stillwater, Oklahoma

The intent of this discussion is basically a plea for recognition of the need for in-depth study of our fish stocks before obvious fish kills occur and especially before occurrence of those insidious changes that alter species and stock composition just as inexorably and possibly more permanently than the spectacular kill.

To address the question of natural variations in fish population size without being merely anaectdotal is a real challenge because although we have an extensive literature concerning the timing, extent and consequences of these fluctuations, we know very little of the mechanisms involved with causality.

The fact that fish population size and structure varies without the interference of man has intrigued and excited fishery biologists from the time of the first investigations, because variation indicates controlling factors that are not static and intimates that once these factors have been identified, they may be modified and in so doing allow us to manipulate or "manage" the resource. Therefore, let us examine our approaches to identifying and dealing with these fluctuations and how our knowledge may then be applied to accessing responsibility for these variations.

THE FISHERY BIOLOGISTS' VIEW OF MORTALITY

Fishery biologists are primarily concerned with making fishermen happy. Now the surest way to have happy fishermen is to be sure that they are able to catch a reasonable number of fish of the right size and species on each fishing trip. Therefore, in order to achieve this goal (i.e., happy fishermen) our fishery biologist will attempt to manipulate the various fish stocks for which he is responsible. When he performs these manipulations he must have some way of knowing whether he is adjusting his stocks in the right direction and perhaps even more important, he must be able to predict the stock composition several

years in advance. To determine the current status of the fish populations (with respect to its contribution to the fishery), he can sample fisherman catch; however, to predict, he must look directly at the fish stocks and the population dynamics that will determine the relative size of any population when it first begins to contribute to the catch. Therefore, the essence of management of fish stocks is the study of population dynamics and population dynamics consists of the balance between natality, growth and mortality. Mortality is often studied because it is relatively easy to estimate and allows us to examine stock composition under the basic assumption that the decline in stock density from year to year is due to mortality and the ratio of stock densities can thus be used to estimate mortality (Cushing, 1968).

SOURCES OF MORTALITY

This brings us to the crux of the whole matter: when a fishery biologist looks at mortality he traditionally (and almost automatically) divides it into 1) fishing mortality and 2) natural mortality (unusually gotten by difference and includes all fish losses not attributable to fishing). Now our category of natural mortality contains a very mixed-bag of sources of death, and to reach a definition of what we will consider as "natural" and "unnatural" mortality that would be satisfactory to us all becomes an exercise in semantics. For instance, if we can consider as "natural" only those systems unaltered by man, then we're fresh out--but serious attention must be directed to how much of man's casual influence we can accept before he is the unintentional controlling factor in the aquatic ecosystem.

As I mentioned earlier, fish populations did and do vary, sometimes drastically, without the aid of man. We give tacit acknowledgement to this by the very title we give to these studies of ... population dynamics. What, then, are some of the factors that contribute to these natural variations? Alphabetically we can quickly list:

Behavior;

Catastrophies in the environment, both immediate and evolutionary in scope;

Chemical, again our attention is usually directed to seasonal or yearly fluctuations, but geologic time references are applicable;

Critical life stages, although not a source of mortality in itself, moving from one to the next is a singularly precarious time in the life of a fish and one should examine these tranistional periods to determine if a disproportion amount of mortality is occurring (Kramer, 1969, has assembled a nice bibliography dealing with early mortality in freshwater fish);

Disease, Dr. Meyer will discuss the role of diseases in fish kills later in this seminar;
Food, or really the lack of the right kind or size at the right time;
Genetic;
Light;
Parasites, I believe Dr. Meyer will also touch upon this subject;
Predation, even discounting man this is reputed to be the source of the greatest amount of mortality in most fish stocks, however, with the possible exception of mortality occurring early in life, I will argue most vigorously against this assumption;
Radiation;
Senility, this actually accounts for very little of the total mortality suffered during the tenure of a given year class;
Siltation;
Water temperature;
Water level fluctuations;
Wind and wave action;
and on and on.

The point to be made here is that fish (being ectothermic, relatively stenothermic and, in the case of our freshwater fishes, trapped in a very limited environment), are completely at the mercy of the vagaries of nature.

BALANCE SHEETS AND MORTALITY

We still are no closer to accessing responsibility for our mortality because although we are able to be quite precise in our estimates of the magnitude and timing of fish deaths, we are still, in the vast majority of cases, most uncertain as to the actual cause(s) of mortality.

Regier and Robson in their chapter on Estimating Population Number and Mortality Rates in Shelby D. Gerking's Freshwater Fish Production point out that:

> "Close monitoring of the dynamics of a single freshwater fish population is often frustrating and discouraging because of the occurrence of violent and inexplicable fluctuations. Pattern and regularity in freshwater fish production will emerge more clearly by combining observations made at different locations and years over a geographic region, as is done in the analysis of agricultural production." pg 32.

I don't really believe that it is quite as black a picture as they paint. Although it is true that these violent fluctuations do occur, to write them off as inexplicable is only defeatism. However, the charge is clear. We must know the fish stocks with which we are dealing!

More often the variations that we may expect are much more limited. If we accept, a priori, the concept of carrying capacity being representative of the productive capacity of a body of water, then we would expect only minor yearly fluctuations in productive capacity and consequently in total fish biomass. What then are the clues to change within our system? (The theory is present and continually being refined.) We must look to the dynamics of the individual populations and examine the structure and patterns of age and growth together with species diversity indices to tell us of major shifts in system structure. In this way, we can gain insight into not only the timing of fluctuations (our predictive element) but into casuality and control within the system. Once an understanding, of the types and magnitude of factors driving and steering the system has been gained, we will be in a position to sort and access the effects and fill in our balance sheet for mortality.

AN OVERVIEW OF THE TECHNOLOGY

The way in which we describe community stability is to determine relative abundance or relative total biomass of the population of all the fish species in the system. Since the relative abundance of the species is a condition of dynamic balance resulting from the way in which the environments productivity has been divided, then, even if total productivity remains constant a change in one species' numbers (even if only temporary) will cause a series of oscillations throughout the rest of the system. Therefore, once the structure of the system has been defined, change in one component or species allows us to predict the changes, both in respect to time and magnitude, in the rest of the components (species).

We are able to use the same general procedure in describing the relative stability of a single species. Relative size of the individual year classes over time does tend to be constant. Mayhew (1956) found that bluegills in an Iowa lake exhibited a fairly stable population over a period of 14 years, and McFadden and Cooper (1962) stated that brown trout recruitment in six different populations appeared stable with only occasional variations caused by flood or drouth. Nikolski (1969) in his book Theory of Fish Population Dynamics sums it all up stating that each species has its own mortality rate with its own distribution over age groups.

This information now allows us to draw some generalizations about age structure in our fish stocks. First, we expect stability or equal sizes of year classes over time. Second, we expect variation that does occur to happen early in life, usually the first year is when the relative year class size is established. Even though no year class operates independently within a population, the magnitude of the variation should be identifiable throughout the tenure of that age class (Figure 1). Note in the figure that if we had made a point estimate of the mortality characteristics in this population at year 3 or year 4, we might attribute the apparent increase in mortality (from 0.5 to 0.75) to the wrong cause, for example a logical explanation might be that this was the time that this species first became vulnerable to fishing. Extreme care must be exercised in interpretation of data gathered from a population over a short period of time. Last, these characteristics of population structure allow us to formulate a set of assumptions for support of our theory of mortality estimation. These are:

1. The age groups in question were equal in numbers at the time each was recruited to the fishery. (Probably least often satisfied and the proximate cause of most natural population fluctuations.)

2. The mortality rate is uniform with age, over the range of age-groups in question.

3. Since mortality is composed of fishing and natural mortality, each of these, individually, is uniform. (Some authors have seriously questioned whether this is ever satisfied.)

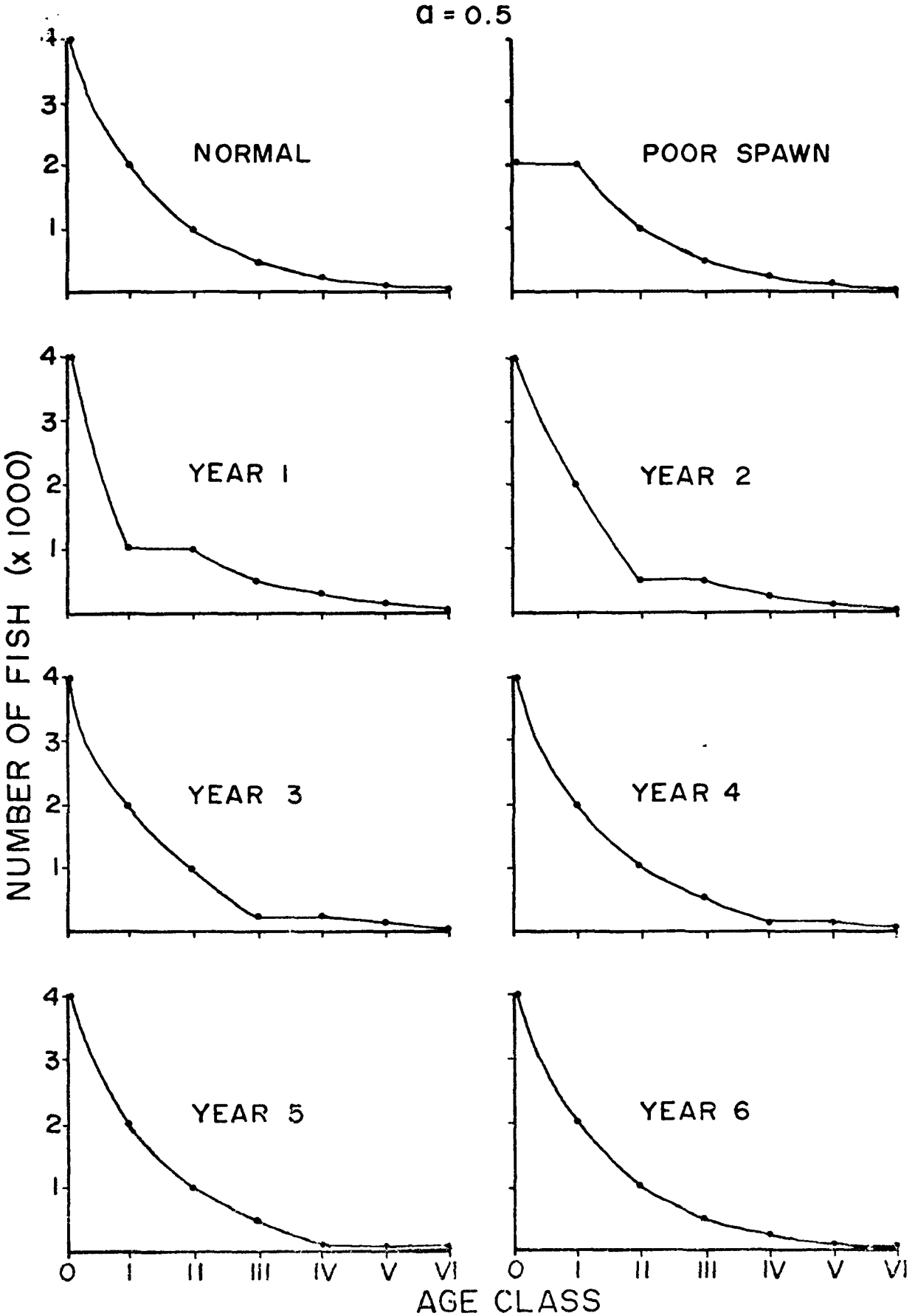
4. There is no change in the population mortality rate with time.

5. Samples are taken randomly from the age groups involved.

Of course, in practice you never expect to completely satisfy these assumptions. However, we do have some idea of the effects of failure to satisfy them on our estimates. The outgrowth of this is that while fluctuation in individual year classes is expected, we don't expect general depression of all year classes within a short time span. If we do observe this latter type of population change, we should be able to identify a climatic catastrophy or begin to look for the possibility of man's influence.

Another point to bear in mind is that natural mortality is highly seasonal. We expect loss in a new year class to occur predominately within the first few months of life. Older fish will usually succumb,

Figure 1. A classical, hypothetical example of the effects, over time, of a poor year class.


a = 0.5
NORMAL
POOR SPAWN
YEAR 1
YEAR 2
YEAR 3
YEAR 4
YEAR 5
YEAR 6
NUMBER OF FISH (x 1000)
1
2
3
4
0
I
II
III
IV
V
VI
AGE CLASS

providing the major losses are not due to predation, during periods of stress which are usually associated with either reproduction or the period when the aquatic environment reaches the farthest point from optimum levels. This usually may be referenced by temperature; therfore, the critical period for a warmwater fish such as a bluegill will probably occur during the latter part of the winter while the critical period for one of the trouts may occur during mid-summer.

DISCUSSION

In most freshwater systems (and I say most as a broad generalization that certainly has many exceptions) the external or physical variables exerting the greatest effect on a system are temperature and water level both operating within a critical time vector. According to most analyses in the literature concerned with the question on natural variation, if we add the internal control of predation (and I would rather substitute disease and parasitism), especially in the first few weeks or months of life, we have identified those factors most responsible not only for the ultimate size and structure of a population, but also for the fluctuation and variation that we observe.

So let us turn from consideration of those few freshwater species that are typically cyclical with impressive displays of "boom and bust" that have intrigued us and stolen much of our investigative energies and examine the subtleties of the much more common, and in the overview, more important minor fluctuations and "creeping" change that indicates overall system stability or direction of system movement.

CONCLUSION

So, while I urge you to continue with the important monitoring programs, that have been described earlier in the seminar, as a viable means of preventing future catostrophic losses due to man's activities, I would also urge you to look also to the potentially more dangerous problem (in the long run) of insiduous changes that are not normal geologic and evolutionary change. These are the ones that will be responsible for total change and in some cases total loss of the recreational/ commercial fishery resource.

In conclusion, since we, upon entry into the realm of scientific inquiry have, at least tacitly, acknowledged and accepted a deterministic philosophy, let us get about the business of sorting out the effects and identifying the proper causes without losing awareness of the role of proximate and ultimate causality. Then and only then may we assign responsibility to man for his actions and address ourselves to the questions of direction and type of control within the aquatic ecosystem that he may be allowed to exercise.

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THE ROLE OF DISEASES IN FISH KILLS

by

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INTRODUCTION

Mortality due to natural causes is probably the largest single cause of death in a fish population. It has been shown that unless killed by some introduced factor (usually due to man) or predation, the most likely cause of death of fish will be old-age (Ricker, 1954). Although many fish may die of disease during a year, seldom does the problem develop to epizootic proportions. Epizootics in nature caused by disease are often induced by a deterioration of the environment in which the fish live (Reichenbach-Klinke and Elkan, 1965; Petrushevski and Shulman, 1959). In this respect, it might be noted that the same situation exists in the managed water areas on commercial fish farms (Meyer, 1970). Stress plays an important role in the initiation of disease and may often be overlooked unless the observer is alerted to the possible effects of low oxygen, sudden temperature changes, or pollution of any kind (Wedemeyer, 1970). It should also be noted that many of the organisms which cause serious problems on fish hatcheries and fish farms are present in low numbers in natural habitats.

The host-parasite relationship between the fish and their parasites has developed to the point where the parasites or pathogens should feed only upon surplus tissues and energy of the host. Successful parasitism requires that the parasite species does not endanger the survival of the host. Should the parasites feed excessively upon a host, either because of excessive numbers or due to a reduced vitality of the host brought about by other factors, the net result is death of the host, a loss of habitat for the parasitic organisms, and a subsequent reduction in the population of parasites (Chandler, 1955). If, on the other hand, host populations have developed proportions beyond their normal food supply and beyond the carrying capacity of the habitat, their resistance may be sufficiently reduced that disease will remove the excess and help bring the host population back in balance with its environment. Thus, under normal circumstances

of adequate food and favorable environmental conditions, disease will seldom be a major factor in the elimination of a species since the host-parasite relationship is self-limiting on both the host and the parasitic organism. Epizootics must then occur only whenever some factor has entered the picture to disrupt the balance (Baer, 1952) and observers must be alert to identify the causative agent, be it over-population, starvation, pollution, or other environmental change.

RECORDS OF DISEASE-INDUCED FISH KILLS

While the foregoing emphasizes the infrequent occurrence of epizootics in natural habitats, records are available describing situations in which it was determined that disease was indeed the primary factor. It is also likely that disease plays a significant role in many other losses but that it has been overlooked. The following list indicates that nearly all major groups of parasitic organisms have the potential of initiating epizootics in natural waters. In addition, readers should keep in mind that those organisms which cause epizootics on hatcheries and fish farms have the potential of doing so in lakes and streams.

Fish kills due to bacteria are frequently overlooked since few observers are trained to recognize symptoms of bacterial disease and even fewer have the specialized facilities at hand to make the necessary isolations and cultures needed to verify the cause. Records at the Fish Farming Experimental Station Diagnostic Laboratory, Stuttgart, Arkansas contain numerous records of fish kills in natural waters caused by bacteria. Lakes with high populations of stunted black bullheads or crappies are frequently hit with epizootics of myxobacterial infections (Columnaris disease). A case of particular interest concerns a reported loss of largemouth bass in Lake Maumelle (Arkansas) in which only bass over 2 pounds were dying, with a preponderance of dead fish over 4 pounds. Field observations revealed that a high number of stunted, emaciated, 6" gizzard shad present in the lake were also dying. These had gone unnoticed by the sportsmen whose only concern was that the large game fish were lost. Gizzard shad in varying stages of dying were evident over much of the lake. Large bass were feeding actively on the moribund shad. A study of both species indicated a heavy infection of Columnaris disease in the shad which was, in turn, transmitted to the bass when they fed on the distressed fish.

Pacha and Ordal (1970) report that numerous outbreaks of Columnaris disease have occurred in natural and hatchery populations since 1944 in both warmwater and coldwater environments. Epizootics have been

reported in Atlantic salmon (Elson, 1968; Pyefinch and Elson, 1967); Sockeye salmon (Colgrove and Wood, 1966); stonerollers (Lennon and Parker, 1960) and among "coarse fish" (Bottomley and Holland, 1966).

Snieszko (1958) reports that high water temperatures reduce the resistance of fish to Columnaris disease and Pippy and Hare (1946) indicated that copper and zinc pollution induced conditions which rendered the fish susceptible to Aeromonas liquefaciens.

The incidence of bacterial hemorrhagic septicemia caused by Aeromonas liquefaciens has shown a stress-related pattern similar to that of Columnaris disease. On fish farms, outbreaks are a common occurrence and are closely correlated to stresses of low oxygen, high temperature, and handling. Epizootics in natural waters are also common. Major epizootics have been reported from a wide variety of warmwater fishes (Haley, Davis and Hyde, 1967; Reed and Toner, 1942; Rock and Nelson, 1965; Wagner and Perkins, 1952; and Meyer, 1964). Rock and Nelson noted that low levels of oxygen were prevalent during the time of the epizootic and cited additional evidence, including high temperature, debilitation of the fish, and very low water flows as factors in the reported and other outbreaks of the disease.

Further evidence of the role of bacteria as etiological agents in epizootics can be cited. Vibrio sp. (Ross, 1970; Cixar and Fryer, 1969; Smith, 1961; Anderson and Conroy, 1970); Aeromonas salmonicida (Fryer and Conrad, 1965; Lange and Ljungberg, 1962); Pseudomonas sp. (Ross, Nordstrom, Bailey and Heaton, 1960; Meyer and Collar, 1965; Seaman, 1951; Wolf, 1937; Van Duijn, 1938); Pasterurella sp. (Snieszko et. al., 1964); Streptococcus (Robinson and Meyer, 1966) and Nocardia sp. (Snieszko, et. al., 1964) have all been shown to be pathogens of fish. With the intensified interest in the culture of marine and freshwater fishes, it is certain that additional organisms will be added to the list.

Outbreaks of bacterial disease are seldom the result of a single factor. Three factors are involved in every potential disease situation ... susceptible hosts, pathogenic organisms, predisposing environmental conditions. All must be present if an outbreak is to occur. Snieszko (1964) lists decreased immunological resistance, poor genetic resistance, temperature stresses, pollution, unfavorable water chemistry, and unfavorable environmental conditions as some of the predisposing factors. In the latter, I would include such factors as crowding, inadequate food supply, spawning activity, storms, and seasonal changes. While bacteria may be the ultimate cause of death in a particular situation, it is not unlikely that

some other factor is more important. Consider, for example, the massive losses of threadfin shad and Tilapia which occur when the water temperatures fall below the optimum for these species. A sudden cold wave may result in massive losses of these fish. Survivors or moribund individuals will often yield heavy cultures of bacterial pathogens and, unless the observer is alert to the circumstances involved, a diagnosis of a bacterial epidemic might be given. Intrusions of salt water into freshwater environments (or vice versa) can cause similar situations.

Fungi have been indicated in a great many epizootics. Because of the obvious nature of fungal growths, many workers are tempted to list the fungus as the cause of death. It is my opinion that, for the most part, such growths are secondary infections feeding upon necrotic tissue induced by bacterial infections, wounds, or death. One species, Ichthyophonus hoferi, has reportedly caused major losses of marine herring (Sinderman, 1958). Since this organism is highly contagious, its introduction into new environments would be expected to lead to epizootics in other natural waters.

Protozoan parasites have been implicated in a number of epizootics but the occurrence is relatively rare. One of the most spectacular outbreaks was that of Ichthyophthirius in the Coosa River of Alabama in which many species of fish were killed along a 200 mile stretch of the river (Allison and Kelly, 1963). Myxosporidia of the genus Thelohanellus have caused severe mortalities of Coregonus and Rutilus sp. in Europe (Kudo and Neglitsch, 1971). Microsporidia of the genus Glugea were implicated by Putz, Hoffman, and Dunbar (1965) in an epizootic in gizzard shad in Ohio. Additional cases can be cited, but, for the most part, either Ichthyophthirius (Elser, 1955; Hopkins, 1959; Kudo, 1934) or sporozoans (Sanders, Fryer and Gould, 1970; Hoshina, 1952; Shulman, 1966) have been involved. Other protozoans, including Costia, Chilodonella, Trichodina, Trichodinella, etc., cause major problems on fish farms and hatcheries (Meyer, 1970). Since these organisms have their greatest impact on young of the year and small-sized individuals, it is likely that most epizootics induced by these organisms go unnoticed.

Metazoan parasites are occasionally involved in epizootics. As a rule, however, documented cases are infrequent and this group can not be considered as a major factor in the initiation of epizootics even though this group possesses adequate potential for doing so. Those cases in which they have been involved have usually involved peculiar conditions which especially favored survival of parasites far beyond normal circumstances.

Petrushevski and Shulman (1958) reported that the introduction of a monogenean, Nitzchia sturinois, caused major losses of sturgeon in Russia. In fish culture, monogenetic trematodes are a serious problem, posing a threat to the survival of small fish. It is therefore also possible that the effect of this group of parasites is over-looked in natural waters. Large numbers of young of the year fishes could die without being noticed by most field biologists and fishermen.

Adult digenetic trematodes have not been identified as causative agents of mortalities among fish. However, cercariae, the larval stages of such flukes, have caused epizottic losses. Hoffman (1954) and Krull (1934) both reported losses of fish caused by the penetration of excessive numbers of cercariae. Experimental evidence (G. L. Hoffman, personal communication) suggests that fry and other small stages of fish may be killed by cercarial penetration but no studies of such effects in natural waters have been reported.

Cestodes, nematodes, and acanthocephalans are often found in large numbers in fish. Often, heavily-infected fish will appear in good health and show no outward signs of ill effects. In other instances, infected fish will be emaciated and debilitated. Larval cestodes have caused losses when excessively abundant (Petrushevski and Shulman, 1958; Moore, 1925) but they are not likely causes of epizootics in natural waters.

Parasitic copepods have caused major epizootics in a number of areas. Argulus biramosus killed large numbers of fish in lakes in South Dakota (Allum and Hugghins, 1959). Ergasilus arthrosis was the cause of a heavy infestation of centrarchid fishes stocked in a brackish water lake in Alabama (Kelly and Allison, 1965). Ergasilus sieboldi annually kills large numbers of fish in Russia (Petrushevski and Shulman, 1958). Although Lernaea cyprinacea causes major economic losses on fish farms, it is seldom a cause of death in natural waters (Meyer, 1966).

Leeches have not proved to be a serious problem to date. The only reported epizootic in natural waters due to leech infestation concerns a report by Rupp and Meyer (1954) where brook trout were killed by an unusually heavy leech population in a lake in Maine.

Occasionally, the mutual occurrence of heavy infections of several parasites on a particular species of fish may prove fatal. Petrushevski and Shulman (1958) report an instance in which they felt an epizootic in sticklebacks was cuased by a double infection of Gyrodactylus (a monogenetic trematode) and Glugea (a microsporidian protozoan parasite).

PROCEDURES FOR IDENTIFYING THE CAUSE OF A FISH KILL

Field Observations: Care should be taken to record as much information concerning the kill as possible ... seemingly unimportant items about the hours of the day when it occurred, the weather, water conditions, and other environmental factors may provide valuable insight as to the WHY of a kill. Changes in water color, algal blooms, vegetation, odor, or flow rates should be noted. Keep in mind that stress of some kind may have been the predisposing force.

Observers should study the species make-up of a fish kill with care. Note the species affected, life stages affected, size of fish affected, general condition of the affected population, age affected, number affected, loss pattern, and any behavioral patterns exhibited by moribund fish.

Collection of Samples: In selecting specimens for study, it is always desireable to use moribund fish, if possible. If not, choose freshly-dead specimens. Live fish from an affected population are *always desireable to fish which have been dead for some time* since parasites may leave a dead fish and saprophytic bacteria rapidly infiltrate dead tissue. If live fish cannot be transported to the laboratory, the best procedure is to place individual fish in plastic bags and refrigerate them on wet ice. If necessary, the specimens can be frozen but many parasites may be lost during the freezing and thawing process. If neither refrigeration or freezing is possible, the fish may be preserved in 10% formalin or a suitable fixative as a last resort. Preservation destroys the usefulness of the specimens, however, for bacteriological or virological culture so this procedure should be avoided if possible.

Procedures for Viral Study: Since few laboratories are equipped with qualified personnel or equipment for virological studies, it is adviseable to send suspect specimens to laboratories which have the necessary facilities. Frozen specimens are adequate for viral studies. The Eastern Fish Disease Laboratory, Kearneysville, West Virginia; Cooperative Fish Disease Project, Auburn University, Auburn, Alabama and the Western Fish Disease Laboratory, Seattle, Washington are some of the stations to which suspect fish may be sent. In each case, however, it is adviseable to call prior to shipping specimens.

Procedures for Bacteriological Study: When collecting the fish, examine the external surface of the fish for lesions such as ulcers,

fin rot, or patches of fungi. If these are found, a scraping of tissue from the margin of a lesion should be prepared as a wet mount (a piece of tissue in a drop of water and covered with a cover slip) and studies under 440X magnification. If myxobacteria are present, they may be observed as long as flexible bacteria are attached to the tissue by one end. Frequently, they can be observed moving actively. See Davis (1953) for a good description.

Material for culture should not be taken until the external surface of the fish has been sterilized with a good disinfectant such as household bleach, Roccal, Hyamine, or a similar product. Using sterile instruments, the body cavity should be carefully opened and the internal organs exposed and moved to one side. Care should be taken to avoid puncturing or cutting the intestine since leakage may contaminate the body cavity. A sterile loop should then be used to collect tissue from the kidney for inoculating culture medium. The kidney is the organ of choice but isolates may also be taken from the liver, blood, and directly from the site of a lesion. The most commonly used medium for the initial isolation is Tryptic Soy Agar (TSA), although Beef Heart-Infusion Agar, Blood Agar or similar rich medium would suffice. If myxobacteria are suspected, it is desireable to also inoculate some Ordal's medium since myxobacteria do not grow readily on TSA. Other specialized media may be required if the pathogen involved is not a standard warmwater fish pathogen. If bacteria are the likely cause of an epizootic, workers should expect to find large numbers of a single type of colony. If this is the case, it is desireable to transfer the organism to fresh media (screw cap test tubes filled with slants of the isoldation medium) and then take or ship the culture to a bacteriological laboratory, where biochemical and serological tests required for the identification of bacteria can be run. Do not tighten the cap for shipment.

Procedures for Parasitological Study: Fish specimens should be examined for parasites as soon as possible. External parasites often leave the body soon after a fish expires, others will die quickly if the fish is allowed to dry or if the specimen is frozen. Examine the external surface of the fish for lesions, cysts, or large organisms which may or may not be moving. Study scrapings from lesions, cyst contents, and large organisms in wet mounts beginning with the lowest magnification available and work up to the highest magnification that the thickness of the slide will permit. For cyst contents, it may be necessary to use 440X magnification.

In the case of small fish, an entire gill arch may be prepared as a wet mount. In large fish, a scissors may be used to remove a

small number of gill filaments for making the preparation. Study all organisms, cysts, and cyst contents as above.

Fin tissue and/or scrapings from the dorso-lateral area of the body should also be prepared as wet mounts and studied in a similar manner. Observers should watch especially for movement since most parasites are active. If they are the causative agent, large numbers of them should be present on a high percentage of the affected fish.

After examining the external surface of the fish, open the body cavity and examine the internal organs. If the number of specimens is limited, the following order of procedure should be observed: (1) collect tissues for the detection of external parasites, (2) sterilize the external surface of the fish, (3) open the fish with sterile instruments and collect material for bacterial cultures, (4) perform the examination of internal organs for possible parasitism. A failure to observe this sequence may render the material unfit for a complete examination.

Examine all internal organs for cysts, hemorrhagic areas, blood clots, lesions, or gross abnormalities. Carefully remove all suspect tissues and organs and place them in physiological saline (0.85%) to prevent them from drying out. Then examine each under a dissecting microscope, using the compound microscope if needed to check cyst contents, organisms, or peculiar structures. Preserve specimens and tissue for later study.

If the supply of fish permits, preserve additional specimens to supply material for further study if needed.

Fixatives and preservatives are available for most of the major groups of parasites. Unfortunately, few of them are universally acceptable or available. I suggest that field workers carry as many of the following solutions with them as possible: 10% formalin, Bouin's solution, AFA (FAA), and 70% ethanol. Material for the study of possible protozoan parasites should be dropped in Bouin's or AFA. Worm parasites should be flattened under a cover slip and then placed in AFA, parasitic copepods should be placed directly in 70% ethanol. Tissues which are suspected of containing parasites of unknown identity should be placed in Bouin's or AFA. If only one fixative or preservative can be carried, 10% formalin may be an acceptable compromise. This solution, however, is a very strong dehydrating agent and may cause excessive constriction or hardening of specimens.

For identification purposes, workers need not go beyond genus identification in most cases. Hoffman (1967) provides a useful tool for the general identification of fish parasites. If precise identification is desired, smears of the affected organs should be made. These smears, small chunks of affected tissue, and specimens of the parasites should be placed in an appropriate fixative and preserved for later study. In many instances, species identification will be possible only by experienced workers who may use special stains or equipment in their work. Most field biologists will find it adviseable to send the material to some specialist for identification. However, I strongly advise against sending every specimen you encounter since the time of such men is limited and often too valuable to be requested except when you suspect the organism to be the cause of a major problem. Advice on where to send specimens is available from our laboratory, the Eastern Fish Disease Laboratory, Western Fish Disease Laboratory, Auburn University, and zoology departments of most universities.

DISCUSSION

Before attempting to ascribe an epizootic to a particular cause, every observer should study the entire environmental picture to acquaint himself with as much information about the outbreak as possible. Seemingly insignificant factors of weather, water flow, nutrition, pollution, and water chemistry may play important roles in the true cause of the epizootic. As an example, I would like to describe a unique situation which occurred during the winter of 1963. Weather conditions showed an abrupt change from a daily temperature of 25° F to 85° F. White catfish being held in a 0.1 acre pond at the Fish Farming Experimental Station fed heavily on chironomid larvae on the third day of warm weather. During the following night, the temperature fell from 83° during the day to about 24° by the following morning. The change was accompanied by a brisk wind which speeded cooling of the pond waters. During the next day, large numbers of catfish fingerlings were observed belly-up on the water surface, some in rigor, others making feeble attempts to swim. All had distended abdomens and were unable to dive below the surface. Losses had begun. Preliminary symptoms suggested dropsy (a bacterial disease) but a more complete examination revealed that the stomach was filled with gas ... the fish were in fact bloated. The sudden drop in water temperatures was apparently so rapid that the fish were unable to digest the contents of the stomach and gut. Bacterial decay then began and generated sufficient gas to bring the fish to the surface. Transfer of the fish to warmer

water eliminated some of the problem but both parasitic and bacterial infections followed the stress of the sudden temperature change and handling. Similar relationships are common in natural environments.

Frequently the first indication that something is wrong in natural waters is the appearance of dead fish. Such evidence is "after the fact" and workers are then faced with re-constructing the situation which led to the kill. Fish killed by pesticide will appear much the same as fish killed by an oxygen depletion. As a consequence, no evidence should be overlooked.

An epizootic due to parasites can be expected to have an extended course with fish dying over a prolonged period. Losses affect a limited number of species. Specimens may appear weak or listless and often will be emaciated. Small fish would be expected to die first. Generally, the die-off will affect a few fish at the outset and the mortality rate will climb with time. Other aquatic animals are not affected.

Bacterially-induced epizootics will follow a similar course of action. All sizes of fish will be affected but will usually be limited to one species. Occasionally tadpoles and frogs may also be killed. As a rule, however, the observer should expect to see lesions on the body of the fish. Should an unusually virulent strain of bacteria be the cause of the epizootic it is possible that the course of the disease will be very short, highly lethal, and lack the typical lesions. Such cases are rare.

Kills resulting from the depletion of oxygen generally occur during the early morning hours of darkness, but the die-off usually ends with sunrise. The mortality will be abrupt and the large specimens will die first. Frequently there will be conspicuous changes in the odor and color of the water. Vegetation or algal blooms frequently will also die but zooplankton will be present. Depending upon the level to which the dissolved oxygen has dropped, there is often a species selectivity in an oxygen depletion with those species having the highest oxygen requirements dying first.

Kills due to toxicants are likewise abrupt. Mortalities may occur at any hour and will continue through daylight and darkness. Zooplankton, snakes, turtles, clams, and frogs may also be affected. Small fish will die first, usually exhibiting signs of convulsions, loss of equilibrium, and toxicosis. Species selectively will be evident during the early stages of the kill but, in the case of highly toxic substances, may progress through an entire population. Some insecticides cause the fish to exhibit a forward thrusting of

the pectoral fins (Meyer, 1965), others may cause curvatures of the spine (Meyer, 1966). A lethal toxicant such as cyanide may cause no outward signs but an examination of the internal organs will reveal massive blood clots. Water conditions may appear normal with no apparent color change or odor.

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MORTALITIES FROM MISCELLANEOUS CAUSES AND IMPORTANT ENVIRONMENTAL CONSIDERATIONS

by

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I. Fish Kills vs. Natural Mortalities

A. Natural Mortalities - Those which are caused through natural phenomena such as: acute temperature change, storms, ice and snow cover, decomposition of natural materials, salinity change, spawning mortalities, parasites, and bacterial or viral epidemics.

B. Man caused fish kills - Produced by environmental changes through man's activity, and may be attributed to municipal wastes, industrial wastes, agricultural activities and water control activities.

1. Direct

2. Indirect

II. Organism Interactions

A. Pollution is often studied as a factor affecting the biota, but it is equally important to recognize the environmental changes produced by the biota. According to Westlake, under many conditions ".... the environment is almost as much a product of the community as the community is of the environment."

B. Changes in ecosystems are occurring continuously.

1. Myriad interactions take place at every moment of the day as plants and animals respond to variations in their surroundings and to each other. Evolution has produced for each species, including man, a genetic composition that limits how far that species can go

in adjusting to sudden changes in its surroundings. But within these limits the several thousand species in an ecosystem, or for that matter, the millions in the biosphere, continuously adjust to outside stimuli. Since interactions are so numerous, they form long chains of reactions.

2. Small changes in one part of an ecosystem are likely to be felt and compensated for eventually throughout the system. Dramatic examples of change can be seen where man has altered the course of nature. It is vividly evident in his well-intentioned but poorly thought out tampering with river, lake, and other ecosystems.

   a. The Aswan High Dam

   b. The St. Lawrence Seaway

   c. Lake Kariba

   d. The Great Lakes

   e. Valley of Mexico

   f. California earthquake (Scientific American 3981, p. 333)

   g. Everglades and the Miami, Florida Jetport

   h. Copperhill, Tennessee (Copper Basin)

3. Ecosystem Stability

   a. The stability of a particular ecosystem depends on its diversity. The more interdependencies in an ecosystem, the greater the chances that it will be able to compensate for changes imposed upon it.

   b. The least stable systems are the single crops - called monocultures - created by man. A cornfield or lawn has little natural stability. If they are not constantly and carefully cultivated, they will not remain cornfields or lawns but will soon be overgrown with a wide variety of hardier plants constituting a more stable ecosystem.

c. The chemical elements that make up living systems also depend on complex, diverse sources to prevent cyclic shortages or oversupply.

d. The oxygen cycle, which is crucial to survival, depends upon a vast variety of green plants, notably plankton in the ocean.

e. Similar diversity is essential for the continued functioning of the cycle by which atmospheric nitrogen is made available to allow life to exist. This cycle depends on a wide variety of organisms, including soil bacteria and fungi, which are often destroyed by pesticides in the soil.

4. Some environmental examples:

a. Diatom Blooms

"... Asterionella (an oil storage alga) produces an autotoxin (autoantibiosis) which will inhibit itself, may stimulate or inhibit other species that store oil, but always stimulate algae that store starch. For example, Asterionella may produce a bloom and inhibit itself, but stimulate a population of Synedra. These oil storage algae produce a substance that stimulates a starch-storing alga, Coelastrum, which may stimulate another starch-storing organism, Cosmarium, and they, in turn, stimulate an oil storing species of Dinobryon." Patrick.

b. The altered structure of the plankton community due to the introduction of the alewife.

c. Fecal deposit feeders in the estuarine environment.

III. Biological Evaluation

A. Biological evaluation of an aquatic environment includes a comparison of the living community at one location with that of another. It also calls for an understanding of the physical conditions impinging upon that population.

B. The inability of man to adequately predict or control his effects on the environment is indicated by his lack of knowledge concerning the net effect of atmospheric pollution

on the earth's climate. Similarly, there are large scale fish mortalities, which are associated with red tide organisms (dinoflagellates), that may, or may not, be related to the discharge of nutrients.

C. Levels of Biological Integration

1. From higher to lower we have:

a. Biosphere

b. Ecosystems

c. Communities

d. Populations

e. Individuals

f. Organ Systems

g. Organs

h. Tissues

i. Cells

j. Protoplasm

2. When we observe phenomena at some level, the causes (or explanation of mechanisms) are usually to be found at a lower level of biological integration and the effects (or significance) at a higher level.

IV. Possible Natural Causes of Fish Kills

A. Oxygen depletion due to ice and snow cover on surface waters.

B. Oxygen depletion at night because of plant respiration or at anytime during the day because of natural occurring organics in the water.

C. Abrupt temperature changes.

D. Epidemic and endemic diseases, parasites, and other natural occurring biological causes.

E. Lake water inversion during vernal or autumnal turnover which results in toxic material or oxygen-free water being brought to the surface.

F. Interval seiche movement in which a toxic or low DO hypolimnion flows up into a bay or bayou for a limited period of time, and later returns to normal level.

V. "Natural Populations"

A. Man's activities have left few, if any, control (reference) populations. As an example, pesticides and combustion products are now all globally dispersed.

B. Most populations are continuously or intermittently stressed by man's activities. If these stresses produce populations susceptible to various parasites or diseases, should we call this a natural mortality?

VI. Pollution Comes in Many Packages

A. The sources of air, water, and land pollution are interrelated and often interchangeable.

1. A single source may pollute the air with smoke and chemicals, the land with solid wastes, and a river or lake with chemical and other wastes.

2. Control of air pollution may produce more solid wastes, which then pollute the land or water.

3. Control of wastewater effluent may convert it into solid wastes, which must be disposed of on land.

4. Some pollutants - chemicals, radiation, pesticies - appear in all media.

B. "Disposal" is as important and as costly as purification.

VII. Persistent Chemicals in the Environment

Increasingly complex manufacturing processes, coupled with rising industrialization, create greater amounts of exotic wastes potentially toxic to humans and aquatic life.

They may also be teratogenic (toxicants responsible for changes in the embryo with resulting birth defects, ex., thalidomide), mutagenic (insults which produce mutations, ex., radiation), or carcinogenic (insults which induce cancer, ex., benzopyrenes) in effect.

A. Metals - current levels of cadmium, lead, and other substances whose effects on humans and fish and wildlife are not fully understood constitute a mounting concern. Mercury pollution, for example, has become a serious national problem.

B. Pesticides

1. A pesticide may move through an ecosystem in many ways. Hard (pesticides which are persistent, having a long half-life in the environment includes the organochlorines, ex., DDT) pesticides ingested or otherwise borne by the target species will stay in the environment, possibly to be recycled or concentrated further through the natural action of food chains if the species is eaten. Most of the volume of pesticides do not reach their target at all.

2. Biological magnification

Initially, low levels of persistent pesticides in air, soil, and water may be concentrated at every step up the food chain. Minute aquatic organisms and scavengers, which screen water and bottom mud having pesticide levels of a few parts per billion, can accumulate levels measured in parts per million - a thousandfold increase. The sediments including fecal deposits are continuously recycled by the bottom animals.

a. Oysters, for instance, will concentrate DDT 70,000 times higher in their tissues than its concentration in surrounding water. They can also partially cleanse themselves in water free of DDT.

b. Fish feeding on lower organisms build up concentrations in their visceral fat which may reach several thousand parts per million and levels in their edible flesh of hundreds of parts per million.

c. Larger animals, such as fish-eating gulls and other birds, can further concentrate the chemicals. A survey on organochlorine residues in aquatic birds in the Canadian prairie provinces showed that California and ring-billed gulls were among the most contaminated. Since gulls breed in colonies, breeding population changes can be detected and related to levels of chemical contamination. Ecological research on colonial birds to monitor the effects of chemical pollution on the environment is useful.

C. "Polychlorinated biphenyls" (PCB's)

PCB's are used in plasticizers, asphalt, ink, paper, and a host of other products. Action has been taken to curtail their release to the environment, since their effects are similar to hard pesticides.

D. Multifactoral Mortalities

When some stress is placed on a population they become particularly susceptible to a pathogen and/or parasite. Thus, we have one or more primary stress events which lead to secondary causes of death.

1. A serious disease of salmonids in the United Kingdom is Ulcerative Dermal Necrosis (UDN). A strain of Saprolegnia has been indicated as cause for death, but investigators are divided as to the real causitive factor(s).

2. Factors involved would include:

   a. Species

   b. Age

   c. Breeding condition

   d. Pre-disposition to the disease

   e. Chemical environment

   f. Physical environment

   g. Immunological barriers

   h. Exposure to pathogens

VIII. Other Sources of Stress in Populations - Biological Pollution

Contamination of living native biotas by introduction of exotic life forms has been called biological pollution by Lachner et al. Some of these introductions are compared to contamination as severe as a dangerous chemical release. They also threaten to replace known wildlife resources with species of little or unknown value.

A. Tropical areas have especially been vulnerable. Florida is referred to as "a biological cesspool of introducted life".

B. Invertebrates

a. Asian Clams have a pelagic velier larvae, thus, a variety of hydro installations are vulnerable to subsequent pipe clogging by the adult clams.

b. Melanian snails are intermediate hosts for various trematodes parasitic on man.

C. Vertebrates

a. At least 25 exotic species of fish have been established in North America.

b. Birds, including starlings and cattle egrets.

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TOXICANT CAUSED FISH KILLS
COLLECTION, PRESERVATION, AND ANALYSES OF INORGANIC SAMPLES

by

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INTRODUCTORY REMARKS

As a representative of the primary analytical laboratory for water analyses in Oklahoma, I should like to point out that we are aware that, in the particular case of fish kills, we are only looking at the top part of the iceberg. We realize that although fish may not be actively and openly dying in a particular body of water, the quality of that water may be changing in such a way as to cause a deleterious effect on the fish populations.

When we were asked to participate in this seminar, we were told that we would primarily be speaking to administrators as opposed to actual investigators. This will prejudice our remarks somewhat.

Up to this point, you have heard about sophisticated teams and sophisticated methods to investigate fish kills. You have heard references to teams composed of biologists, chemists, and engineers. It might be adviseable to note that even the EPA does not have such a team in this region. Neither do we have such a team in Oklahoma. This talk is, therefore, on a somewhat lower plane than have been some of the previous talks.

SAMPLE COLLECTION

On numerous occasions throughout this seminar, time has been emphasized as one of the most important factors in the investigation of a fish kill. It is essential that representative samples be taken from active problem areas. Without samples from active problem areas, it is extremely difficult, and frequently impossible, to determine the cause of a fish kill. Samples should also be taken from all potential sources within the area that could contribute to the problem. Each tributary should be sampled. It is also extremely desirable to have samples which are above and below the problem area as well as above and below the probable source or sources of the problem. The purpose of the above and below samples is to provide comparative data for the determination of why a particular water is killing fish.

A natural question that arises is relative to the number of samples which should be taken. I do not feel as if a set number of samples should be taken. It is extremely important that your investigator be a capable man who will utilize good judgement and take those samples which are absolutely necessary to define the problem and eliminate potential sources so that the actual primary offender may be determined. We have found that it is adviseable for the analyst to be included on this survey team. (Of course, in our case, frequently the entire team is the analyst). It is extremely important that your investigators only take those samples which are necessary. It is not infrequent that inexperienced investigators will take unnecessary samples merely to prove that they were at a particular point. This does, of course, help to justify travel expenses. However, we are not in the business of justifying travel expense, rather, we are attempting to investigate a fish kill.

I would like to reemphasize the point that samples should be taken where and while the fish are actually dying. Samples which are taken after the fact, are generally taken after the problem has washed downstream. An early morning dissolved oxygen test is frequently highly desirable. In any case where a depletion of oxygen is suspect as the cause of the kill, or in cases where a marginal dissolved oxygen content is noted, useful information may be derived from a dissolved oxygen determination immediately prior to sunrise.

SAMPLE PRESERVATION

When one considers the amount of money which is invested in each sample, it becomes apparent that all steps must be taken to insure that those samples be as representative as possible of the conditions existing at the time of sampling. Hopefully, containers for the sample will be supplied by the laboratory. Unfortunately, in Oklahoma, we do not have sufficient funds to provide such containers to all personnel who might possibly take samples associated with fish kills. Of course, sample bottles are supplied for our own personnel. If a sample bottle supplied by the laboratory is not available, one should utilize as clean a bottle as possible to take the sample. In the past, we have suggested that a bottle of distilled water be purchased for use as a sample container. Whatever container is used, the container should be flushed with the water to be sampled several times. Our preferance is for plastic containers with deep-sealing air-tight non-metallic caps. We ask for at least a half gallon sample, due to the fact that frequently we do not know the chemical cause of the fish kill and must make numerous analytical determinations.

If the sampler has a fairly accurate idea of which chemical parameter is causing the fish kill, it is highly desirable to preserve the sample according to the enclosed chart taken from the 1971 Environmental Protection Agency Manual for Chemical Analyses of Water and Wastewater. Unfortunately, such preservatives frequently are not available. Should this be the case, one should refrigerate all samples and deliver those samples immediately to the analyst. In cases where samples are preserved, additional samples which are not preserved should be taken and refrigerated.

It is imperative that in all cases the samples be delivered as quickly as possible to the analyst.

SAMPLE ANALYSIS FOR INORGANIC CAUSES

A great deal of the analytical work surrounding a fish kill is determined by the field investigation. It is extremely important that dissolved oxygen be determined at the site at the same time as the samples are taken. It is also highly desirable that temperature, pH, total alkalinity, and other parameters (such as ammonia) be determined dependent on the type of kill. If the field investigator is able to get a rough idea of the number of fish and the numbers of each species which are affected, the types of industries, and whether or not a municipality discharges in the area, much analytical time can be saved. (As a general rule of thumb, if the fish kill tends to involve a single species, we tend to believe that a kill of a biological nature is in progress.) If a municipality is on the water shed and a high dissolved oxygen is noted, we tend to rule out sewage as a cause of the kill, and so on. In other words, the field investigation rules out or rules in various analytical approaches.

All methodology should follow Standard Methods as outlined in the EPA manual for analytical techniques. Dependent on the type of kill, parameters such as COD, BOD_5, ammonia, pH, total alkalinity, chloride, metals, and other analyses are determined, again, dependent on the field observations. If the field investigation does not tend to single out a particular type of waste as being the cause, as a last resort, we run everything and hope to find some causative agent.

CONCLUDING REMARKS

It might be wise to point out that we are not in the business of soliciting samples. We have sufficient samples at any given time. Additionally, samples which are or are not taken properly, or

which are or are not necessary, but which relate to a fish kill, have a very high priority. As a result, it is necessary to run these samples in lieu of other samples. Generally, the other samples would be drinking water supplies. It is important to point out that samples should not be taken unless an extremely good reason exists for analytical results of that sample.

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Table 2 - Sample Preservation*

Parameter	Preservative	Maximum Holding Period
Acidity-Alkalinity	Refrigeration at 4°C	24 hours
Biochemical Oxygen Demand	Refrigeration at 4°C	6 hours
Calcium	None required	7 days
Chemical Oxygen Demand	2 ml H_2SO_4 per liter	7 days
Chloride	None required	7 days
Color	Refrigeration at 4°C	24 hours
Cyanide	NaOH to pH 10	24 hours
Dissolved Oxygen	Determine on site	No holding
Fluoride	None required	7 days
Hardness	None required	7 days
Metals, Total	5 ml HNO_3 per liter	6 months
Metals, Dissolved	Filtrate: 3 ml 1:1 HNO_3 per liter	6 months
Nitrogen, Ammonia	40 mg $HgCl_2$* per liter - 4°C	7 days
Nitrogen, Kjeldahl	40 mg $HgCl_2$* per liter - 4°C	Unstable
Nitrogen, Nitrate-Nitrite	40 mg $HgCl_2$* per liter - 4°C	7 days
Oil and Grease	2 ml H_2SO_4 per liter - 4°C	24 hours
Organic Carbon	2 ml H_2SO_4 per liter (pH 2)	7 days
pH	Determine on site	No holding
Phenolics	1.0 g $CuSO_4$/1 + H_3PO_4 to pH 4.0 - 4°C	24 hours
Phosphorus	40 mg $HgCl_2$* per liter - 4°C	7 days

*Disposal of mercury-containing samples is a recognized problem; research investigations are under way to replace it as a preservative.

Table 2 - Sample Preservation

(Continued)

Parameter	Preservative	Maximum Holding Period
Solids	None available	7 days
Specific Conductance	None required	7 days
Sulfate	Refrigeration at 4°C	7 days
Sulfide	2 ml Zn acetate per liter	7 days
Threshold Odor	Refrigeration at 4°C	24 hours
Turbidity	None available	7 days

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TOXICANT CAUSED FISH KILLS
COLLECTION, PRESERVATION, AND ANALYSIS OF ORGANIC SAMPLES

by

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COLLECTION

Fish kills can result from a variety of causes such as:

1. Low dissolved oxygen concentrations
2. Toxic chemicals
3. Excessive water temperatures or rapid changes in water temperatures
4. Extreme pH levels
5. Excessive suspended solids
6. The effects of diseases and parasites, etc.

When investigating a fish kill, all of the possible causes should be kept in mind. Any factors that might cause a change in water conditions should also be considered. For example, a heavy rainfall shortly after a pesticide application may wash sufficient pesticides into a stream to cause a fish kill, or an extended drouth with high temperatures might reduce the flow of dilution water producing toxic results from a normally acceptable industrial effluent.

Sampling sites and the number of samples to take depends on several factors. In a flowing stream it is necessary to take water samples in the area of the fish kill, above it, and below it. Since the polluting substance may have been present in a section of the stream, it may be advisable to collect samples at a distance downstream from the central kill area which corresponds to the elapsed flow time since the kill was first noticed. When fish kills occur in lakes or impoundments, the depth of sampling is important because of stratification.

An important aspect in sample collection is that of the on-site investigation. Information concerning an industry's products as well as by-products could be valuable to help narrow the possibilities and cut the work load of the analyst. If pesticides are suspected, a list of possibilities would be appreciated. The usual procedure is to approach the chemist, with samples in hand, and ask for - yea demand - a complete pesticide analysis. It is important to realize that, even though proper samples were taken and preserved correctly, if the on-site investigation was not conducted properly, generally no amount of laboratory investigation can solve the fish kill. The amount of useful laboratory work that can be done is often limited by what was not done during the on-site investigation.

Before the actual collection, all containers to be used for organic samples, should be prepared by rinsing with running tap water, distilled water and several times with a suitable redistilled solvent (e.g., acetone, hexane, petroleum ether, chloroform). Caps and liners are washed with detergent, rinsed with tap water, distilled water and solvent. Some also recommend heat treatment at 300°C overnight to destroy any trace of organic matter.

Assuming competent sampling personnel, from one liter to one gallon of water is collected in glass bottles equipped with screw caps fitted with Teflon liners. Plastics, such as polyethylene, are not suggested because traces of plasticizer can be leached from the plastic by the water and can be a source of analytical interference. If Teflon liners are not available, clean solvent rinsed aluminum foil may be used. Some plastics may actually absorb pesticides and other organic materials, so its use is certainly discouraged. Many investigators avoid the use of glass sample bottles because breakage in shipment frequently causes loss of sample. This can be overcome by the use of expanded polystyrene foam shipping containers molded to fit the bottle.

PRESERVATION

Whatever the method of preservation used for organic samples, it is generally intended to:

1. retard biological action
2. retard hydrolysis of chemical compounds and complexes and
3. reduce volatility of the constituents.

Although a universal method or additive for the preservation of samples would be nice, it is a practical impossibility. At best, the various procedures for preservation can only slow the inevitable chemical and biological changes which occur when a sample is removed from a stream or lake.

Techniques for preserving organic contaminated samples are usually limited to solvent addition, refrigeration, and freezing. Needless to say, the length of storage should be as short as possible. Even properly preserved samples often have short holding times. EPA's Methods for Analysis recommends the following holding times:

Oil and Grease	24 hours
Phenolics	24 hours
Organic Carbon	7 days

The wide variety and large number of pesticides in use today makes it impossible to specify a time beyond which all analyses are worthless or at best questionable. However, it can be stated that a practical time limit should be measured in days and in some instance to hours. Some of the phosphates, carbamates, and 2-4-D esters will even undergo slow hydrolysis at a neutral pH and low temperature. Because pesticide samples are subject to change, it is important that dates of analyses be included in any report so proper interpretations can be made. Although not applicable to all types of samples, immediate refrigeration at temperatures near freezing or below is the best preservation technique available, at least until the advice of laboratory personnel can be obtained.

ANALYSES

Realizing that most laboratories are searching for the minimum amount of time and effort required to complete a project, one should consider two questions:

1. Must the laboratory investigation start from scratch, or did preliminary work show that there was one or more major suspects responsible for the fish kill?

2. How well must the probable cause be established?

It is quite difficult to determine the number of samples that can be handled, the cost involved, and the time required for analysis for a particular laboratory because of the many and varied organic analyses available. Private laboratories prefer to quote a price and time required as determined on an individual basis rather than across-the-board quotations for every parameter.

When attempting to identify and quantitate organic chemicals in water, it is necessary to be familiar with some of the techniques used in the identification of organic compounds in general. Many physical constants such as:

A. Melting points

B. Boiling points

C. Refractive indices

D. Density

E. Optical rotation

F. Molecular weights

can be determined in organic analyses.

Expensive instruments such as the NMR and the mass spectrometer can also be valuable in organic identification. However, in many cases infrared spectrometry and chromatographic procedures prove adequate.

Gas-liquid chromatography is an analytical method for the separation and identification of a mixture of volatile (usually organic) components in a sample. GLC can be extremely sensitive, but because of this sensitivity it is often necessary to apply extensive clean-up techniques. Also, while GLC is ultra-sensitive and does an excellent job of separation, it still lacks the specificity necessary for positive identification.

Infrared analysis after separation and possible identification by GC has found extensive use in water analysis. Because no two compounds will have identical spectra, an infrared spectrum of a pure compound presents a definite method of identification. The greatest use for IR is to detect functional groups that are most commonly encountered in organic compounds.

COLLECTION, PRESERVATION, AND ANALYSIS OF ORGANIC SAMPLES

by

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The most important aspect of monitoring fish kills is speed. The field biologist should make every effort to do three things as quickly as possible: get to the site of the fish kill, make the collections, and get the samples to the laboratory. If the fish kill is in a river, samples should be collected above and below, as well as at, the site of the kill. At least several species should be collected. Comparisons can be made only between fish of similar size classes of the same species. If possible, 20 fish of a particular species and size class should be collected at each sampling site. Only whole fish should be collected (unless otherwise specified). Representative bottom organisms also should be collected. Much the same procedure should be followed in collecting samples in lakes. Fish and other organisms should be collected at the site of the kill and, if the lake is large enough and the kill is localized, from several areas where they have not been affected. Again, every effort should be made to collect the same species and sizes from the different locations. Dead, dying, and apparently healthy fish of each species collected from each site should be packaged and labeled separately.

There is some disagreement among analysts as to the best way to package fish. Some types of plastic bags may contaminate samples with plasticizers or PCB compounds. Probably the safest procedure is to wrap the samples in acetone-washed aluminum foil before placing them in plastic bags.

All samples should be frozen, if possible, or at least iced, immediately after collection. Analysis of a rotten fish is messy at best, and because of the possibility of bacterial action, the results of the analysis often are of little value.

Proper lines of communication between the field biologist and laboratory personnel are extremely important in the monitoring of

fish kills. Each sample must be properly labeled to show species and the locality, date, and method of collection. Too often, field people feel they are sending samples off into some unknown void and laboratory personnel feel that that is where their samples are coming from. This feeling is eliminated if the biologist, in addition to preparing a written report describing the conditions of the fish kill, discusses the kill directly with the analyst. Unfortunately, no two fish kills are ever exactly alike; consequently the sampling and analytical problems differ with each situation. A field biologist who can communicate with the laboratory people will better understand the analytical problems and will be better able to collect samples that provide the most information to the analyst.

INTERPRETATION AND USE OF DATA

by

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The purpose of a fish kill investigation is to accumulate information (evidence) that can be used to determine the exact cause of a fish kill. Often some of the necessary information is not available and cannot be readily obtained.

Evidence concerning fish kills basically falls into three categories:

1. On-site evidence: collected during the investigation at the scene as soon as possible after the kill. Includes routine analysis of water samples.

2. Miscellaneous evidence: collected by various means.

3. Laboratory evidence: collected during subsequent laboratory investigation. Includes field toxicity tests. Lab investigations supplement field investigations.

The ability to diagnose the cause of a fish kill is highly dependent on the manner in which samples are collected and the amount and reliability of information recorded at the time of sample collection.

Although the investigation is a team effort and normally involves numerous individuals in the collection and analysis of samples, data should only be appraised by scientists or administrators competent to do so.

"In general, the job of the scientist is to invent a story which accounts for a set of observations and then to decide how likely the story is." Our starting point should be a set of conclusions, and our purpose should be to find which of these conclusions are most nearly correct and which are clearly not correct. It is not unusual for all preset conclusions to prove to be incorrect.

Use of chemical data from analysis of fish and water samples to provide answers as to the cause of a fish kill involves not only a familiarity with conditions under which samples were collected, including visual observations of dead and dying fish as well as water in the fish kill area, but also an awareness of basic principles of fish toxicology. There is no substitute for training, experience, judgement and familiarity with toxicity and fish kill literature in interpretation of data.

Even then data is not easy to interpret. There is a tremendous volume of toxicity literature, much of which is contradictory. Determination of a concentration of a material that is equal to or greater than that stated to be lethal to fish in some reference is not conclusive evidence that the material caused the observed fish kill. Environmental variables such as temperature, pH, calcium, DO, species and age of fish, and many others influence the toxic actions of many materials, sometimes over one or two orders of magnitude. In addition most toxicity studies involve exposures to continuous concentrations; whereas water samples may reflect a concentration that existed for only a short time.

Problems encountered in interpreting and using chemical data are usually inversely proportional to the complexity of the effluent and the number of possible sources.

There is also more to interpreting data than finding the presence of a toxicant in fish samples. The question is was there enough of the toxicant present to kill the fish. What constitutes enough may vary from species to species and even from population to population within a species. Often it may be necessary to find enough of the toxicant in certain tissue(s) of dead fish. What constitutes the right tissue must be determined for each individual toxicant. For example blood is the right tissue for several pesticides; the gill for cadimium; the gill and opercular bone for zinc. The Japanese have conducted considerable research in the use of whole fish bodies and critical tissues for post mortem identification of fish killed by toxicants. Results of this research is published in the Japanese Society of Scientific Fisheries Bulletin and is referenced in the Annual Literature Review issue of JWPCF.

The key to using the critical tissue concept is collecting proper samples. Three kinds of samples should be collected for tissue analysis:

1. Live unexposed fish. (Control)

2. Dead fish.

3. Live exposed fish.

The number collected of each is dependent on what you are looking for. Twenty (20) of each would be good but this may be impractical. At least ten (10) of each is recommended. Preferably all fish of one species should be about the same size.

A lethal critical level is said to exist when the lowest level of the toxicant found in certain tissues in fish killed by the toxicant is higher than the highest level of the toxicant that can be found in the same tissues in fish exposed to, but not killed, by the same toxicant.

By using appropriate samples and analytical techniques, one can attempt to determine if there appears to be a critical level of the suspected substance in any tissue for the sample fish. This technique possibly overcomes complications for hypersensitive fish due to other stresses, resistant fish, and prior exposure.

This is probably the only kind of approach that should be used at the present time to try to prove the cause of a fish kill from tissue analyses. It cannot be used unless proper samples (kind and numbers of fish) are collected and preserved during on-site investigation. Tissue analyses of only dead or moribund fish, however, may be used to place possible suspected toxic agents at the scene or to eliminate them all together.

The toxic agent and source cannot be positively identified unless stream samples can be shown to be lethal. After collection of a sample assumed to contain the toxic substance or complexes causing the kill, the sample must be proven toxic. This can be done by routine bioassay in the majority of cases. If concentrations are of sufficient magnitude, a bioassay may not be necessary.

The relationship of other chemical conditions to the toxicity of pure compounds has been studied intensively. These contributions are important but not too helpful in finding who, or what, killed the fish. In fact it is almost impossible to establish the cause of a fish kill if positive identification and determination of the conditions, chemicals, or complexes contributing to death are required.

The addition of any chemical or waste that can be shown to change a previously nontoxic complex to a toxic one should be considered to cause the kill, even though the substances in the water act to increase the toxicity greatly. This approach now forms the basis for the use of bioassays to establish the source of a kill when additional supplementary evidence is not obtained.

In summary I would say that the ideal situation is to observe conditions under which the fish are dying and actions of dying fish; to examine freshly dead fish; to make a reconnaissance of the area for all possible sources; and from this information, come up with a list of possible toxic agents. Interpretation of analytical data then, in some ways, becomes a process of elimination. Again the interpreter must be familar with the toxic actions of all suspected agents.

One thing to keep in mind is that it should not automatically be assumed that subsequent fish kills in a waterway are caused by the same reasons as preceding ones. When investigating any fish kill all possible causes should be kept in mind.

It is the responsibility of the scientist to interpret the data in light of his experience, training, judgement and all related conditions and circumstances. It will then be the decision of a lawyer to decide if the evidence is strong enough for litigation action and a judge and/or jury to decide if the interpretation as to what killed the fish is correct.

FISH KILL INVESTIGATIONS AND TECHNIQUES

by

Ed Sorrels

Arkansas Dept. of Pollution Control and Ecology

Fish kill investigative procedures involve:

A. Preplanning

B. Information to be gained from informant

C. Preparation at office and laboratory

D. Area observation and

E. Area sampling

F. Trace of causative agent

G. Documentation of events

H. Post-investigation procedures; bioassay, benthos monitoring

Speed of response has been stressed by a number of participants in this Seminar. Preplanning is vital for speedy response to a report of a fish kill. It requires provision of a prompt fish kill reporting network, trained personnel who will make the investigation, and the necessary equipment for thorough procedures and facilities for all pertinent analyses.

The Arkansas Game and Fish Commission has 125 enforcement officers stationed over the State and 12 biologists in district offices, all in radio contact with the central office in Little Rock. In turn, this is linked by telephone through the State governmental exchange to the Department of Pollution Control and Ecology. A specific section of the Department has the responsibility for investigating pollutional-caused fish kills. The Ecology Section of the Water Division of the Department comprises 5 ecologists and 2 technicians who are involved in routine biological monitoring, annual stream

basin water quality surveys and periodic special surveys; however, 1 or more is ready to drop current activity and proceed with a fish kill investigation at instant notice. In addition, the Department maintains 9 field inspectors and 7 district offices, as well as 2 mobile laboratory units located for current water basin surveys. Ordinarily any area of the State can be reached by a trained investigative team within 3 hours of first receipt of report of a fish kill.

The Department Chemistry Section is continually engaged in analyzing routine water samples collected throughout the year from over 70 stations of the Interstate Stream Water Quality Network. Chemists furthermore operate from the 2 mobile laboratories during specific water basin surveys. The section is capable of performing any chemical analysis described in "Standard Methods for the Examination of Water and Wastewater". The section also has facilities for extraction and gas chromatography of pesticides in water, sediment and tissue samples.

As much pertinent information as possible should be obtained from the observer reporting the fish kill. A copy of a suggested information form is appended to this paper. Where possible the investigator should arrange to meet the reporting individual and have him accompany the investigator to the fish kill area. By all means obtain precise directions to the fish kill area. Determine when the kill first began or was first observed, whether or not the kill is continuing at present; if not, when it stopped. Get an estimate of the extent of the kill as to miles of stream affected or size of pond or lake covered. What percentage of the estimated fish population was affected? What are the species and size ranges involved? Estimated number of dead fish per yard of bank? What is the opinion of the reporting individual(s) as to cause? What recent activities have taken place in the drainage basin, such as crop dusting, industrial spills, start-ups or change-overs? What has been the weather pattern preceding the kill? Has there been heavy rainfall? When? Duration? Or a long extended period of cloudy overcast? Highwinds, wind direction, temperature, sudden temperature changes, etc.; all of these questions may bear upon the problem. What is the nature of the watershed and predominant land uses--whether it drains municipal areas or areas of considerable population density? Have similar fish kills occurred before? If so, is there a periodicity or an association with other events, weather conditions or time of year?

On gaining this information, the investigator pinpoints the location of the kill on the map, obtains needed maps of the area from files,

determines possible sources of pollution and decides whether any non-routine sampling and analysis may be required. For example, a pole-preserving (creosoting) plant may be expected to have in its wastes phenolics (cresols), pentachlorophenol, and possibly a complex oily carrier. An electroplating industry uses cyanide as a complexing agent. In water, hydrogen cyanide is the major toxic factor, so that a cyanide concentration which is innocuous at pH 8 may be lethal at pH 6. Preservation of cyanides, in addition to icing, is accomplished by adding 20 ml of 1 N NaOH per liter of sample. Ammonia may be found in the discharge of industrial wastes from chemical or gas plants, from ice plants, or from scouring and cleaning operations where "ammonia water" is used. Preservation of ammonia, in addition to icing, is accomplished by adding 40 mg H_2SO_4 per liter of sample. The investigator notifies the chemistry laboratory operator of the estimated number of samples and kinds of analyses to be expected, the estimated time of return to the laboratory, and of any special analyses that may be required.

A suggested checklist of equipment needed to make the investigation is appended. A good portion of this necessary equipment can be kept in portable cases designed for the purpose in the vehicle reserved for the fish kill investigations so that the investigator can get under way readily by gathering up an armload or so of equipment and sampling containers.

Once in the field, discerning observation is the keynote of a thorough fish kill investigation. If the investigator arrives in the area while fish are still dying and can make the necessary observations, collect and analyze the required dying samples, the cause of the kill can be determined. Results depend primarily on the field investigation. One cannot depend on subsequent laboratory analysis to "tell the story", because it is on the physical basis of what has happened while on the scene that each decision made, each sample collected should proceed from a simple question, yes or no. (It may be that some questions cannot be answered directly.)

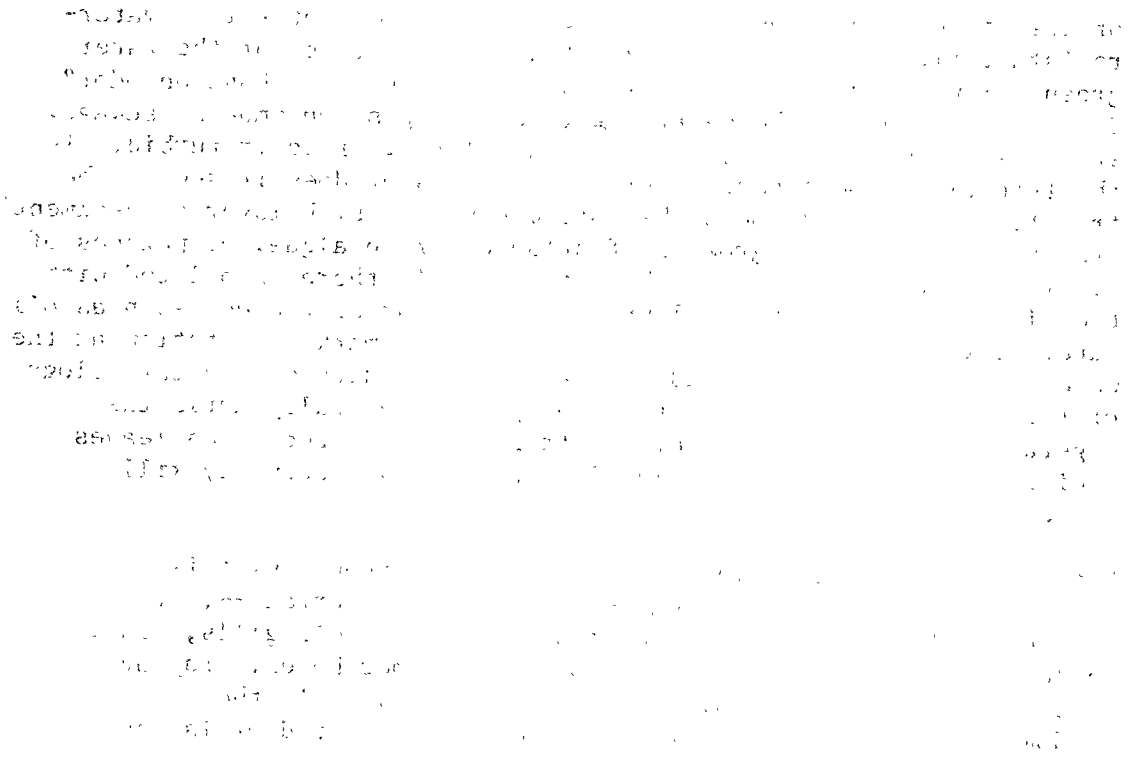
Some rough stream diagrams are appended to illustrate the overall physical survey involved, indicating points or areas where these questions are repeated in order to locate possible sources of pollution and tracing the extent of damage.

Are there dead fish? Are there dying fish? Are there live fish? (In the case where someone has observed dead fish, but none are apparent on investigation, make searches. Check the bank for remains and dredge the bottom.) Immediately observe the water and the dying

or dead fish. Note your observations. Penciled notes in a water-resistant field logbook are suggested. Is the color of the water green, brown, reddish, or black and septic? Does it have an odor? Try to characterize the odor; e.g., rotten eggs, cucumbers, sewage, etc. Is the water clear, even though colored, or is it turbid? Is the turbidity from grossly visible particles or does it seem to be from fine particles? Does the bottom have a thick layer of sediment? Are there conspicuous growths of attached green algae, or patches of light brown sewage fungus (Sphaerotilus)? Is there associated with the kill some other physical presence that can be traced, such as oil material that creates a skim or "rainbow" and marks vegetation at the water's edge? The banks and vegetation can indicate some chronology of the water level. A rise that has passed generally marks the vegetation to the highest level obtained. Submerged green leaves indicate higher than normal levels. Some agents actually kill susceptible vegetation reached by a rise.

Note the behavior of dying fish. Are they nervous, skittish, convulsive? Are they "piping"--gasping at the surface--, or listless? Note the appearance of dying fish, their gills, fins, scales, abdomen. Look for discoloration, hemorrhages, fraying, sores or cysts, whitish slime, swollen abdomens. Is there a predominance as to size of fish and species affected or is the distribution random? Tabulate size and species distribution of one or more collections of affected fish with appropriate notes as to representativeness, etc. A finding that only the smaller fish died or that large fish died but small fish survived is important in distinguishing between oxygen depletion and toxic causes. The key difference is gill surface ratio to total body mass. Small fish, being relatively more rapid in absorbing water solutes, withstand marginal oxygen depletion but succumb first to toxic materials. Observation that animals other than fish are killed--snakes, turtles, etc.--will certainly indicate that an extremely toxic factor other than dissolved oxygen depletion is involved. The observations made according to this section should enable the investigator to estimate whether the kill is an acute situation or a continuing chronic problem.

Record the temperature of the water. If very deep, check it at several depths. Test the dissolved oxygen, always! On a small stream five feet deep or less collect the sample at mid-depth, midstream. On deeper streams collect the samples at 0.2- and 0.8- depth. On broad streams collect these samples at quarter-points also. The absolute minimum of samples to be taken are:



(1) In at least one point downstream, preferably where fish are actually dying; or where kill was heaviest, if over. (2) In source effluent, if any. (3) Above kill area or suspected source. On ponds and lakes sampling on several transects may be required; across, above and below the fish kill area. Sample in source effluent, if any. Also sample on windward side and on leeward side.

If the dissolved oxygen is adequate at all depths and all areas, B.O.D. samples are omitted. If oxygen deficit is a possible factor, a B.O.D. sample from the area is to be collected. If there is a heavy growth of algae, a plankton sample also is collected. If there appears to be an algae die-off with a surface scum and/or clear brownish water, a skimming of the surface also should be collected.

If a source of organic pollution is a factor, a grab sample and B.O.D. sample from the effluent must be collected. If there is a suspected source of municipal waste, cannery waste, or feedlot waste, a routine bacteriological sample (total coliform, fecal coliform, fecal streptococcus) from the kill area may also help in establishing the causal connection.

Measure the pH. The appearance of the water and nature of suspected sources of pollution are clues to whether the reaction may be a factor.

Each sample collected should be labeled immediately with the name of the stream or lake, the sample point number by noted map or diagram, the date and time of collection, the analysis required, the bottle numbers of D.O. and B.O.D. samples collected at the same point and time, the air and water temperature in degrees centigrade, and the initials of the investigator. Samples should be sealed and iced immediately.

Collect only distressed or dying fish for lab examination and/or blood and tissue analysis. Collect at least one individual preferably two, of each species affected. Place in plastic bag, tag with date, time, stream and location, condition of fish when collected, as well as suspected causative factor. Ice, and freeze as soon as possible. Take blood samples if pesticides or toxic chemicals are suspected. Cut through the body of the fish ahead of the caudal fin and "milk" blood from the dorsal vein and aorta into a prepared vial. Collect at least 5 ml from each species.

Make an estimate of the size and extent of the fish kill; the species, sizes, numbers and weights, and the miles of stream or acres of lake affected. In Arkansas the State Game and Fish Commission will make an extensive and thorough evaluation on large kills.

In the field the investigator is working against time. His critical samples should be back in the laboratory for analysis within 24 hours of collection. This is usually readily accomplished with some overtime, perhaps. Even if he spends the entire day in the field he can have his critical samples in the laboratory the next morning.

Answers to the questions indicated in the paragraphs on observation and sampling, at the key points in his physical survey, will enable the investigator to trace the causative agent to its source, ruling out other possibilities in turn. Judiciously-scened photographs can aid in documentation of the circumstances of the kill. Once the agent is traced to its source, the investigator may require entrance and inspection of the source waste facilities and storage facilities. He should inform plant management that he is investigating the specific kill, that he has traced the agent to the plant, and request they accompany him on the desired inspection. In Arkansas, such entrance for the purpose of pollution investigation is specifically provided for in the State Pollution Control laws.

The investigator may find it appropriate to take statements, signed and witnessed, from observers of events pertinent to the investigation.

In the case of complex and poorly-defined wastes, documentation of toxic effects and levels of toxicity may be well-served by bioassay procedures. These procedures should always be carefully planned, defined and recorded along the lines of standard scientific methods. The benthic community of a body of water is subject to the effects of the materials that pass through. A profile of the number and types of organisms according to their sensitivity will serve to document the unaffected portion, the zone of degradation and the zone of recovery or of marginal effects. In situ monitoring can be accomplished by diverse means such as caged fish, rock basket benthos samplers and floating anchored periphyton samplers.

Remember that information flows both ways: Water Quality Criteria/ Field Determinations. A well-documented fish kill investigation with levels of the toxic agent established and definition of conditions obtaining prior to and during the fish kill will also serve as a bench mark for future work.

APPENDIX

1. Standard Initial Information Form for Fish Kills.

2. Checklist for fish kill investigation equipment.

3. General diagram of stream and tributaries to emphasize importance of checking and ruling out possible physical sources as the investigator moves upstream in tracing the causative agent of any fish kill.

4. Illustration of specific fish kill. Heavy rainstorm washed-over inadequately free-boarded holding pond causing a total fish kill in some thirty-five miles of Little River. Total poundage 394,000; total value $46,382. Previous bioassays had shown that even treated waste from this plant was highly toxic (TLm_{48} = 0.05% dilution). Forms of life other than fish were killed, including some vegetation. Water quality parameters (D.O., pH, etc.) were normal except for turbidity. Agent was traced to source by dead fish (where any alive preceding kill), by phenol content of samples and by marking of water, banks and bank vegetation by the oily carrier.

   Some three weeks later a partial kill occurred in Old River Lake. Sources of other toxic factors were ruled out, a residual of unemulsified material from the original spill was demonstrated, and it was shown that movement of some of the residue had occurred with reversing of normal flow in channels with dispersion of some of the material over a portion of the lake by wind and rain.

5. Illustration of specific fish kill. A number of fish were killed on east side of lake. Wildlife officer found empty herbicide cans at the water's edge near the kill area. Other findings were consistent with conclusion of indirect kill from oxygen depletion due to algae kill.

6. Illustration of specific fish kill. Trace of causative agent showed that acid mine waste had been pumped from a reopened pit some fifteen stream miles from the kill area. Spraying of power-line right-of-way was ruled out as a significant factor.

FISH KILL

INVESTIGATION FORM

1. Commission information obtained from:

   Name_____ Phone_____
   Address_____ Directions to home or office -
   _____
   _____

2. Initial observation made by:

   Name_____ Phone_____
   Address_____ Directions to home or office -
   _____
   _____

3. Location of kill: (Specific directions to point of kill). County - T-R-S- and nearest town or major Hwy._____
   _____
   _____

4. Duration (A) First notice (time_____ Date_____)

   (B) Continuing at present. (yes or no) if no, when stopped_____

5. Extent of kill (A) Area covered - (miles of stream or size of pond or lake)._____

6. Approximate amount of fish affected - (% of estimated population of area affected)._____

   A. Species - affected_____
   B. Size_____
   C. Estimated number per yard of bank_____

7. Opinion of reporting individuals as to cause._____
   _____
   _____
   _____

8. Recent activities in area (dusting, etc. to include recent weather)_____

9. Possible [illegible] in area. (Known to reporting individual)_____
   _____
   _____

10. Supplementary statement of observer and Commission personnel who [illegible].
    _____
    _____
    _____

CHECKLIST FOR FISH KILL INVESTIGATION EQUIPMENT

D.O. and B.O.D. bottles; alkaline azide and manganous sulfate solutions; A.P.H.A. sampler.

Liter-size plastic grab bottles; 2- or 3-liter pesticide grab bottles with foil-lined caps; benthos jars, formalin, string tags, pencils. Thermometer.

Ice chest and ice.

pH meter, kit or fresh pH paper; portable D.O. probe; Hach Field Laboratory Kit.

Dip net, plastic bags for fish, dissecting kit. Vials for blood samples.

Needed maps, county and quadrangle.

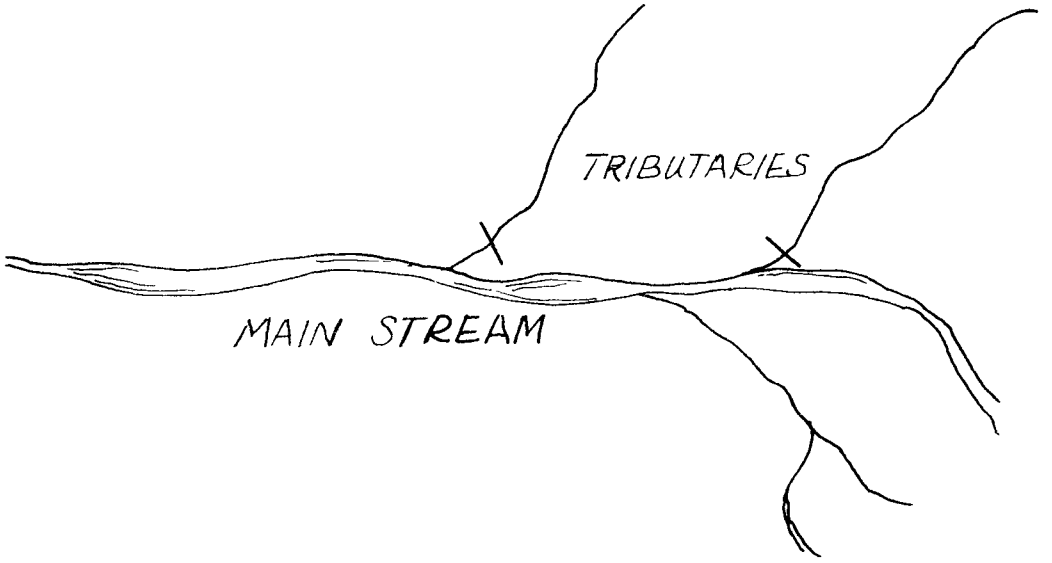
Dredge, spade and screen.

Vehicle, boat and outboard motor.

Camera and film.

Paper towels, distilled water; lantern; appropriate clothing and boots. Flash light and batteries.

PHYSICAL SURVEY 2



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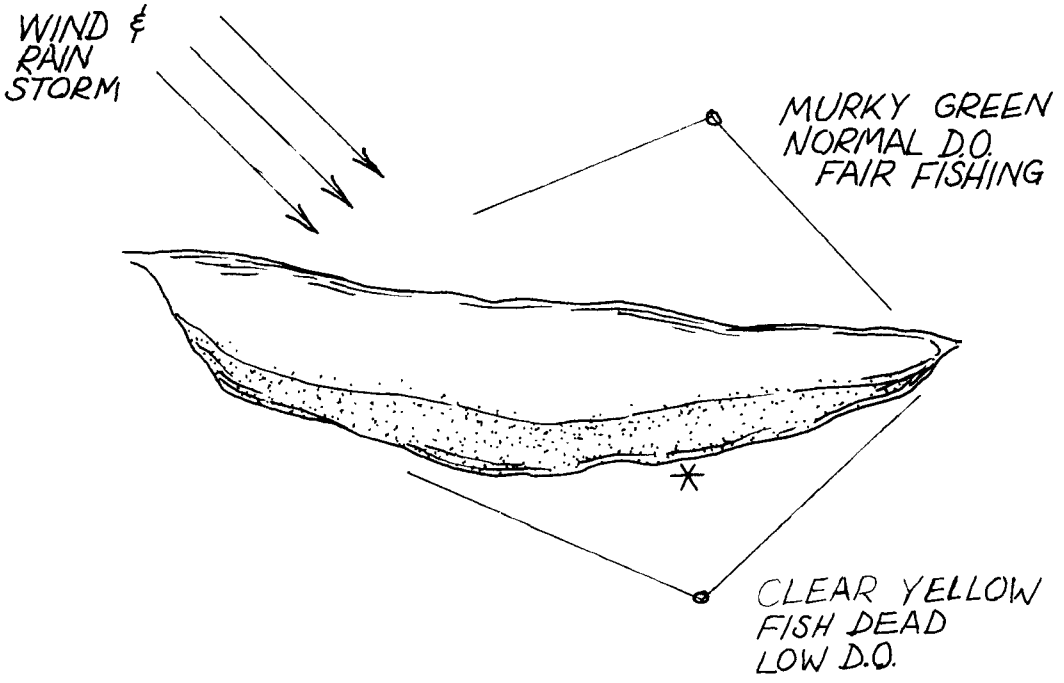

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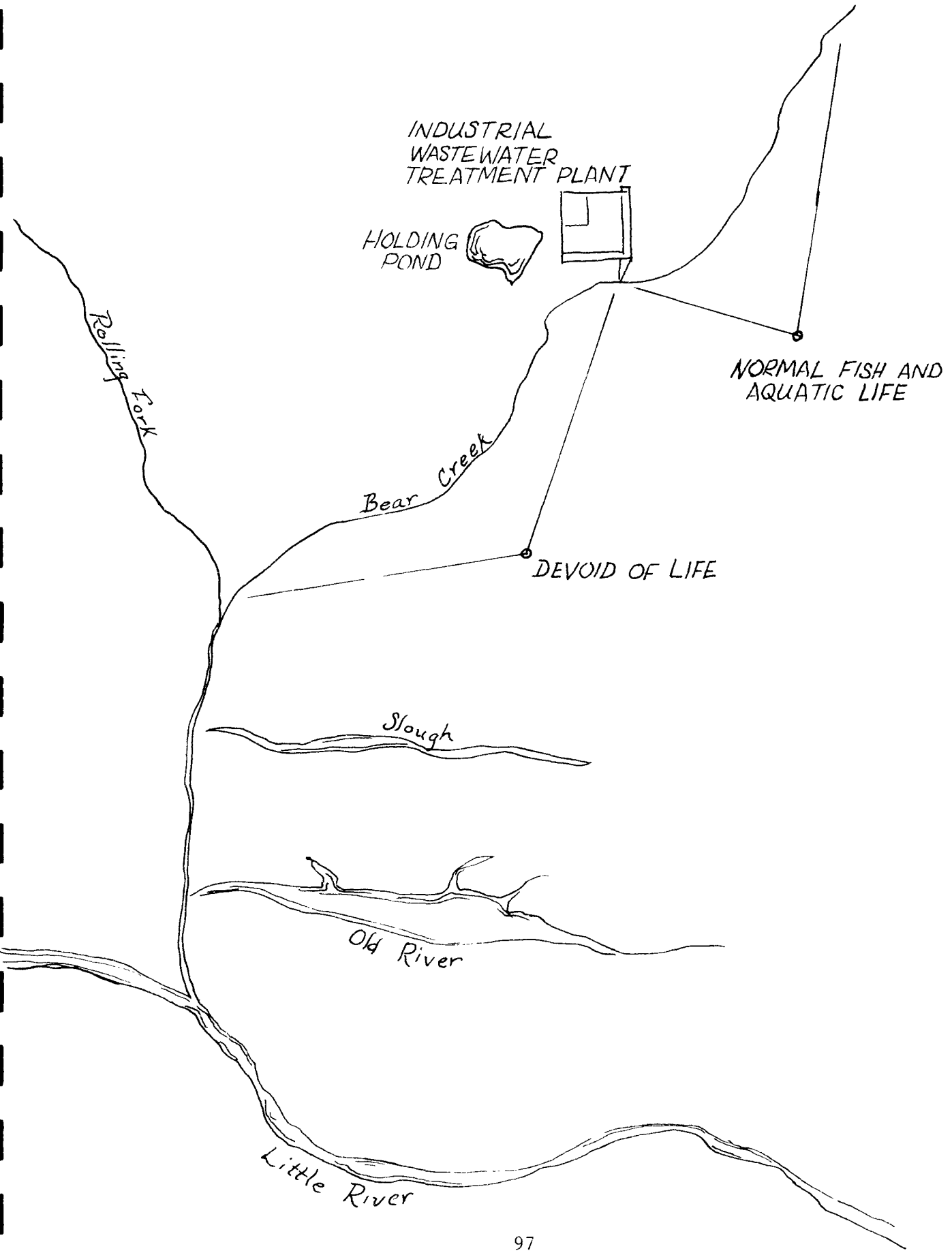
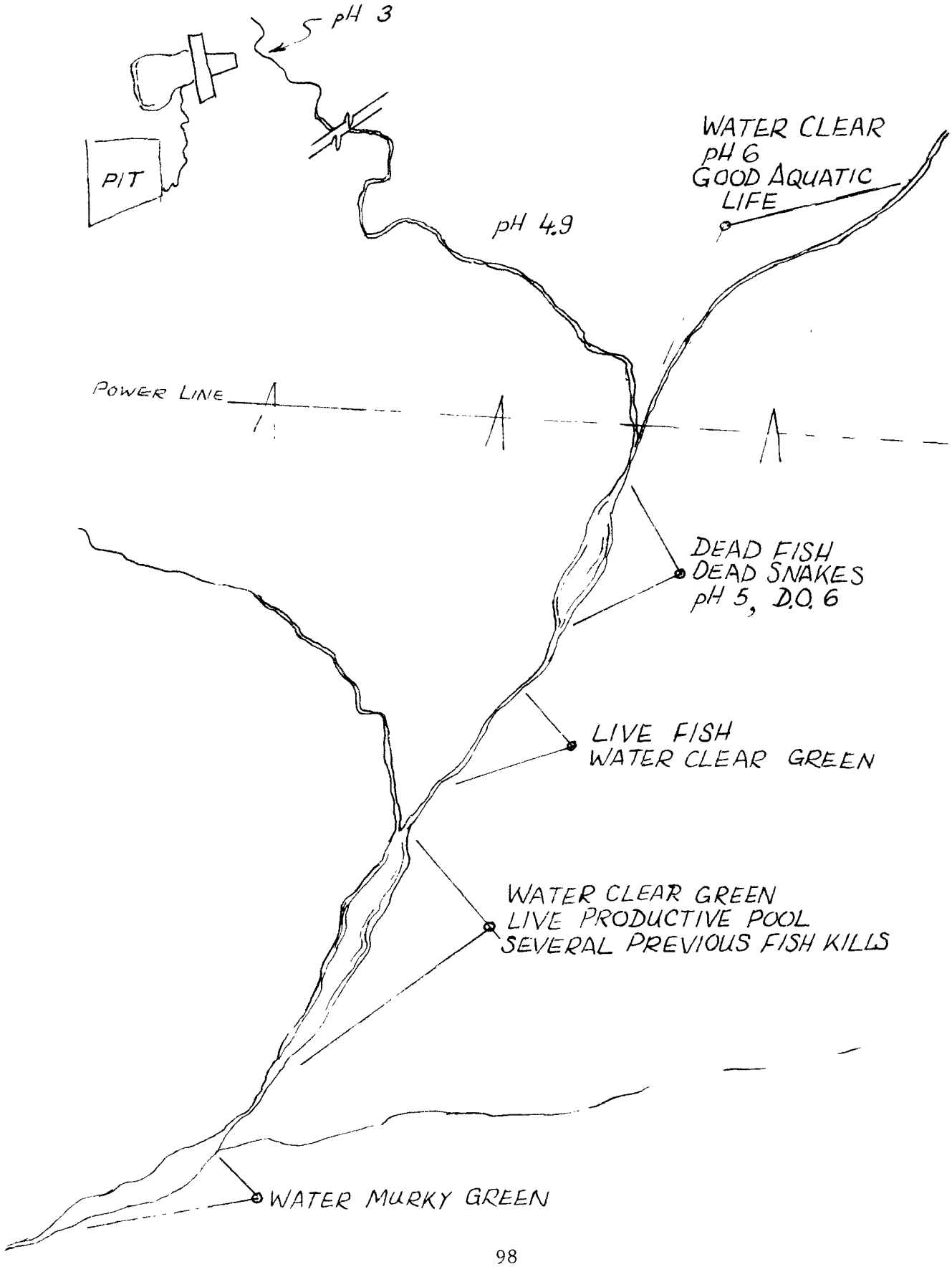


PHYSICAL SURVEY 5



KILL NUMBER ESTIMATES

by

Byron Moser

Oklahoma Dept. of Wildlife Conservation

The following report prepared by the Pollution Committee, Southern Division, American Fisheries Society, 1971, was used as a background for the presentation by Mr. Moser.

BASIC PROCEDURES

FOR INVESTIGATING FISH KILLS

IN STREAMS

PREPARED BY

THE POLLUTION COMMITTEE, SOUTHERN DIVISION

AMERICAN FISHERIES SOCIETY

1971

Billy J. Grantham, Chairman

COMMITTEE MEMBERS

Sam Spencer	Alabama
Jim Collins	Arkansas
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Howard Zeller	Federal Water Quality Administration

BASIC PROCEDURES

FOR INVESTIGATING FISH KILLS

IN STREAMS

PREPARED BY

THE POLLUTION COMMITTEE, SOUTHERN DIVISION

AMERICAN FISHERIES SOCIETY

1971

Billy J. Grantham, Chairman

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INTRODUCTION

The Pollution Committee, Southern Division, American Fisheries Society recognized that the most paramount problem following the establishment of the monetary values of fish by the 1970 pollution committee was to establish guidelines and procedures for determining the size of fish kills. Only two of the states in the Southern Division have written standard procedures. The methods employed by other states varied considerably.

It was also recognized that in all cases the primary responsibility of the Conservation Agency was to delineate the kill by species and magnitude and not to identify the exact toxicant. This latter responsibility is in all cases designated to the Pollution Control Agency. Although the Conservation Agency is not charged with identification of the toxicant every effort is made to do this to offer supportive and corroborative evidence to the Pollution Control Agency. Furthermore, detailed field notes, photographs, etc., are made to lend added support. In addition to the types of data acquisition mentioned above, all of the Conservation Agencies examine the fish for disease conditions and if necessary seek the services of disease specialists if there is a doubt.

BASIC PROCEDURES FOR INVESTIGATING FISH KILLS IN STREAMS

Before any plans can be formulated for making a count of dead fish, it will be necessary to determine the miles involved in the kill. This area should then be delineated on topographic or other suitable maps. The most desirable method for determining the number of dead fish is for all to be counted, but this is often impossible. If the investigator deems it necessary to make only a partial count, this must be done in a manner that will produce unbiased results. The committee felt that an appropriate procedure would be as follows:

> Make 100 yard counts every one half mile starting with the first dead fish observed. The additional counts should then be made at each half mile interval throughout the region of the kill.

Each count will consist of the following:

1. Identify, count, and determine inch groups of all fish in each 100 yard segment.

2. Calculate the total number of fish killed by species in each count area.

3. Multiply the total number of each species by 8.8 (the number of 100 yard segments per 1/2 mile).

4. The answer obtained in item 3 represents the number of each species killed.

In order to facilitate use of this method an example is included below:

	Species	Number	Inch group
1st 100 yards	Blue gill	140	1
	"	120	2
	"	60	3
	"	30	4
	"	25	5
	"	30	6
	"	10	7
	"	5	8
		420	

	Species	Number	Inch group
2nd 100 yards	Blue gill	100	1
	"	80	2
	"	40	3
	"	20	4
	"	15	5
	"	15	6
	"	5	7
	"	5	8
		280	
3rd 100 yards	Blue gill	40	1
	"	30	2
	"	20	3
	"	15	4
	"	25	5
	"	20	6
	"	10	7
	"	5	8
		165	
4th 100 yards	Blue gill	0	1
	"	10	2
	"	5	3
	"	10	4
	"	15	5
	"	5	6
	"	0	7
	"	0	8
		45	
5th 100 yards	End of Kill		
Total fish counted	910		
Number of miles	2		

910 X 8.8 = 8,008 Blue gills killed

Counts such as those illustrated should be made for each species and the total number of fish killed calculated. Since the kill will be assessed by use of the monetary values, it is necessary to have a breakdown to the various inch group catagories.

In the event of excessive large kills over many stream miles the investigator may deem it necessary to make counts at one mile intervals.

VALUE OF FISHES

by

Sam L. Spencer

Chief of Fisheries, Game and Fish Division
Alabama Department of Conservation and Natural Resources

If you plan to go to court to recover compensatory damages for dead fish, you must have a value to place on the fish that were killed. Prior to 1970, only four of fourteen Southern Division states of the American Fisheries Society had a list of monetary values for their fish. These values varied considerably among these four states. For example, a 12" bass in the Tennessee River could have had a different value depending upon whether it was killed in the Tennessee, Alabama, Mississippi or Kentucky portion of the river. What would have happened if a fish kill occurred near the state line and fish were killed in two or more states and these states used different sets of fish values?

In 1970, the Southern Division, American Fisheries Society, formed a Pollution Committee with one member from each of thirteen states and one member from the Federal Water Quality Administration. The states included were Alabama, Arkansas, Florida, Georgia, Kentucky, Louisiana, Maryland, Mississippi, Oklahoma, South Carolina, Tennessee, Texas and Virginia. The first task undertaken by this committee was the development of a set of fish values to be used as a guideline. At the end of 1970, the committee published their "1970 Monetary Values of Fish". I would like to read a few lines from the Introdcution of this booklet which may help explain how the values were derived.

"Every fish should have a monetary value placed on it. The purpose of a monetary value list for fish is for a fair and reasonable assessment of a fishes' value for collecting compensation for the destruction and/or loss of fish due to water pollution. The establishment of a list of monetary values for the many species of fish was a complex task which required the experience and judgment of the various members of the Pollution Committee. Several hundred commercial fish hatcheries were contacted to determine price and availability of any and all sizes and species of fresh water fish. Some problems were immediately encountered such as the non-availability

of certain sizes and species. Another problem was the fact that a "native" or wild fish specimen is usually considered a more valuable resource than a hatchery-reared specimen. Consideration was given to establishing values of fish based upon what it is worth to a fisherman to catch a given species and size of fish. The Committee did not feel this approach was practical or valid for the several following reasons:

(1) Many species are important in the ecological balance, yet are seldom pursued or captured by anglers. Under this type evaluation such fish would have no value.

(2) The value for a given fish would vary greatly according to the skill, experiences, likes and dislikes of individual anglers.

(3) In many cases only a small percentage of sport fish are ever harvested by anglers. Only this percentage could be considered to have a value. For example, if only 10 percent of the bass are normally caught, then the remaining 90 percent that die of other causes could have no legal value assigned to them.

"Therefore, where possible, the best assessment of this value was based upon what it would actually cost to purchase or replace or restock given fish specimens. This was determined from the price lists received from the many hatcheries contacted. Certain other fish, such as the sturgeons, were considered to be a non-replaceable resource. Thus, a price commensurate with the loss of this rather unique fish was assigned. Where hatchery produced replacement stock was not available, the Committee had to judge a replacement value on the role that this species plays in the ecological balance of a body of water and not on what it would cost to rear and stock a given number of specimens. The Committee's judgment of these values was based upon approximately 200 years total experience and training in the fisheries profession.

"Admittedly, at times, certain fish may be purchased live at lower prices. However, this list attempts to take into consideration the availability and nonavailability of numbers of some species. Also, these values do not reflect the almost immediate loss of some fish that occurs when hatchery-reared fish are stocked into public waters.

"We recognized that the laws of several states did not allow the assessment values for fish killed by pollution. It is hoped that this report and list may be helpful in changing this. We also recognized that certain Southern Division states already had or were developing their own list of values. The adoption of this list by all Southern Division states will present a more united front to the pollution abatement effort which must be a goal of highest priority of all fishery scientists."

As pointed out, some values may be considered arbitrary; however, this will always be the case. There is no formula you can use to put in the fishes' scientific name, add some magic factor and out will come an exact value that cannot be questioned. We have to accept the fact that there is no precise way to determine the value of a sturgeon, madtom or darter and there will always be regional differences in how a particular fish is valued.

For these reasons, the list prepared by the Pollution Committee was designed to be used as a guide; however, it was hope of the Committee that all Southern Division states would individually adopt the list, thus giving it more legal strength. At present, a majority of the Southern Division states and several other states have officially adopted the list. We have been informed that the values have been used successfully in several law suits. We have found that when opposing lawyers ask how the values were determined, if you show him this booklet that was prepared by 14 professional fishery workers with over 200 years total experience, he usually has no further questions on the values.

The fish are listed by species and most species in the South are covered. The fish and values are listed by each inch group up to the length at which the species reaches one pound; then the values are listed per pound.

The usefulness of such a list depends upon how the law in your state is written. For example, our law in Alabama states that the values are to be determined by the Alabama Department of Conservation. We have adopted the Southern Division list and the Alabama Water Improvement Commission (AWIC) has officially adopted the list and approved its use.

Another different situation is the New York law which states that each fish killed may be valued at $10.00 each. Therefore, New York is not too interested in our list, and I don't blame them.

The Pollution Abatement Committee of the American Fisheries Society hopes to prepare a guide for all fifty states next year. If such a list could be prepared and adopted by the Society, it would give considerable strength to fish values in court.

We heard mention yesterday of "significant" fish kills as perhaps those involving game fish. The Committee's feeling on this is summed up by the first sentence in the introduction: "Every fish should have a monetary value placed on it". Many of us feel that today's rough fish may be tomorrow's game fish. We see this happening in Alabama now. There is a move underway by sportsmen to have catfish designated as game fish. Catfish are the fourth most sought after species by Alabama sport fishermen, yet, it is classified as a rough fish. In addition, many of our fishermen are enjoying fishing for carp because they can be attracted in large numbers and caught on sporting tackle when other fish won't cooperate. We even sponsored our first carp fishing rodeo in Alabama this year, and it was very successful. We haven't learned how to enjoy eating them yet, but they are fun to catch and we have a very good supply.

If you find you must prepare a monetary list of values for your state, I would suggest that you gather together as much information on fish prices as you can, get as much good professional help as you can, then be prepared to make a lot of tough decisions. Many hours can be spent arguing about a few cents on the value of a fish. You must accept the fact that there are very few absolute values that cannot be questioned.

In court you may possibly encounter an opposing lawyer that wants to question your values; however, they usually prefer to question the number of fish you reported. The lawyer may state, for example, that menhaden only bring $30.00 per ton and you are asking $.15 per pound or $300.00 per ton. You can counter this with the fact that you are referring to the value of the live fish and he is talking about fish his client killed. Dead fish floating on the surface of a public stream or lake are a liability, not an asset.

As I mentioned previously, usually an approved published list will prevent further discussion of such values. However, if the opposing lawyer continues to argue the point, I would suggest that his client be given the choice of either paying your values which are based upon expert opinion or replace the fish with the exact number, size and species killed and somehow compensate fishermen for lost fishing opportunities that occurred as a result of the pollution. If the defendant has killed a few sturgeon, or striped bass, or pirate perches that are not available from commercial hatcheries, the lawyer may decide your values weren't so unreasonable after all.

OTHER ECONOMIC DAMAGES

by

Gene F. Forbes

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Bureau of Sport Fisheries and Wildlife

A fish kill is indicative of much more than a loss to a single resource. It is an indication that the integrity of our water has been violated and that all values dependent upon its quality have been reduced.

When appraising environmental damage the total effect on all the resources in question should be considered. In order to evaluate this damage the value of the "potential uses" must be known.

The potential uses of a resource are both consumptive and nonconsumptive with tangible and intangible values.

Consumptive uses such as irrigation, municipal and industrial waters have tangible values. The water quality required, the volume needed, and the value of these waters for both the present and future generations are relatively easy to determine.

Nonconsumptive uses have both tangible and intangible uses and values. Tangible uses include such things as fishing, hunting, swimming, boating, and many other outdoor oriented activities. The magnitude of these are determined in man-days use. These may be calculated by actual counts, by sampling, or by other various methods of estimation.

The value of the tangible uses may be determined by ascertaining the price an outdoor recreationist would be willing to pay, the net profit which a private operator would make, or by the value to the local or state economy.

The quality of the experience and the other psychological reactions which are an inherent part of outdoor activities have intangible values. Other intangible values include such things as protection of wild and primitive areas, areas of unequaled beauty, scenic, historical and scientific interest, and the preservation of rare and endangered species and their habitat.

Intangible values are much more difficult to define monetarily. Those associated with active participation should be reflected in the value assigned per man-day use. Other monetary values might be estimated using the cost necessary to protect the intangibles in question, or by using the single-purpose alternative method. In this latter method the cost of developing a similar area to replace the intangibles "in kind" is equal to the value of the area in question.

A separate evaluation should be made for each area with the significant physical, biological, or geographical differences defined. This inventory should include the amount, type, and quality of the waters and the demand for the resources available with these waters.

With this basic data on supply and demand the needs of the area may be determined. From these needs the potential uses may be derived.

The potential uses may be based on todays needs, but I suggest that they be evaluated on projected needs. A projection may be made using population projections in conjunction with long-range programs and anticipated public demands.

There are numerous advantages to using this method but I believe the following are primary:

1. Long-range projections require updating as new data become available but it is not necessary to conduct an annual survey or census.

2. This method will standardize evaluations from year to year and insure consideration of all applicable uses.

3. When repeated or continuous pollution damage occurs it is much easier to evaluate the total effect.

4. In addition to using this information to determine pollution damage it may be used to establish long-range goals and act as a guideline in evaluating current programs.

Once the values of the water and its related resources have been established the cost of a loss due to pollution may be equated to the value of the potential uses that were lost or reduced.

Thus in a single case of pollution with a short term effect the value of the loss would be minor. But, if the pollution was cronic or of a lasting nature the cost to the resource would be considerable.

Any form of damage assessment or fines will be a deterrent to violation of the water quality, but this is only treating a symptom and not the cause.

People must be made aware that immediate economic gain at the expense of good environmental quality is not good business. In the long run continued pollution will cost the nation more in both tangible and intangible values than the cost of prevention.

THE FISH KILL INVESTIGATION REPORT

by

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Every fish kill investigation that is worth making is worth reporting. Even the "wild goose chase" should be documented in some manner for possible future reference.

The report should be a systematic recording of organized thought to convey the fact findings of the investigation accurately, completely, and understandably to the reader. The final report should be such that it can be introduced as evidence into court and withstand critical scrutiny and cross-examination.

The final fish kill investigation report is largely dependent upon the investigator's compilation of a thorough and complete set of notes about the contacts made, visual observations, and field and laboratory analyses conducted. All field notes, laboratory notes, and data forms must be maintained in the agency files until there is assurance that they will not be needed further in court actions or elsewhere.

The report should be prepared as soon as possible after completion of the field work, while the details are still fresh in the mind of the report writer. Details of the situation start to fade from the memory in a relatively short time regardless of how complete field notes may be.

Reports of findings in fish kill investigations should be made only by competent personnel (biologists) thoroughly qualified to judge the nature and completeness of the data collected. The writer should have participated in the field work ensuring familiarity with the waterway, sampling locations, water sources, and details of the field operation. This knowledge is essential for the most intelligent and complete interpretation of the data. Reasons for apparent pecularities of the data that would be totally baffling

to one unfamiliar with details of the situation and study may be quite obvious to one who was involved in the work. The person who has been there can bring far more conviction to writing of the report than one to whom the waterway is merely a picture on a map.

When final reports are prepared, they should be filed with the agency authorizing the investigation which will, at its discretion, make them available to other agencies and groups as deemed to be in the public interest.

The rules and techniques of good technical writing apply the same as for all survey reports.

1. Write in a clear, concise manner.

2. The who, what, why, when, where and how questions must be answered at every opportunity.

3. Non-committal language should be avoided.

ESSENTIAL ELEMENTS OF A FISH KILL INVESTIGATOR'S REPORT

1. An Introduction that briefly tells where the kill occurred, when and by whom it was reported, who made the investigation and when, and the authority for the investigation.

2. A Summary description of the findings brought out by the investigation, including extent of the kill, and the numbers and species of fish involved.

3. Conclusions that detail the cause of the fish kill.

4. Recommendations that specify how similar kills may be prevented in the future. Recommendations should be included only with sanction of, and reviewed by, administrators of conservation and/or pollution control agencies.

5. A Description of Study Area section that includes an area location map showing the extent of the kill, principal population centers, major waste sources and sampling stations.

6. A Fish Mortality Discussion section that details in a logical manner the steps taken in the investigation and the knowledge gained from each.

7. A Water Uses section detailing the uses and value of the affected waterway.

8. A Damages section that assigns a monetary value, when practical, to the fish kill.

9. An Appendix that includes all tabular data; describes the manipulations used to estimate the numbers of fish killed and how their value (damage claim) was determined; and that further describes any nonstandard biological and chemical analytical procedures used. If the investigation is prolonged and many contacts are made and investigative steps taken, it may be helpful to include a chronological schedule of all activities of the investigator pertaining to the fish kill from the initial report of the kill until the writing of the Investigation Report. This should include all contacts made (when, where, how, who) as well as other steps in the investigation process.

Sometimes it may be necessary to prepare an addendum to the original report if additional information is uncovered that alters conclusions drawn or recommendations made.

SUMMATION

1. We must recognize that the report is an important and essential element of any fish kill investigation. Take as many pains with the report as with other phases of the investigation.

2. Follow-up steps to be taken (litigation proceedings) may be completely based on the content of the report.

3. A thorough and well-written report may save the investigator many headaches especially if litigation proceedings do occur and the investigator is called upon to testify.

4. Even if no court action is taken the report may prove to be useful in the event of subsequent fish kills or water quality investigations in the same area.

INVESTIGATION OF PETROLEUM CAUSED FISH KILLS

by

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This presentation on petroleum caused fish kills covers a broad spectrum of chemical substances most of which have not yet been isolated and identified. Crude oil generally is not considered to be directly toxic to fish unless it contains a high percentage of aromatic compounds (Shelton, 1971). Indirectly, crude oil may smother aquatic vegetation and non-motile, benthic organisms with subsequent indirect deleterious effects on fish through the food chain. Refined products such as diesel fuel, gasoline, etc. are much more toxic than crude oil. Many of the first surfactants or oil dispersants used on oil spills were more toxic to aquatic organisms than the oil; however, new products are now available which are not acutely toxic to aquatic organisms. Infrared spectrophotometry (Kawahara and Ballinger, 1970, Kawahara, 1969), atomic absorption spectrophotometry (Moore, Miller, and Glass, 1966) and spectrophotometry combined with gas chromatography (Stuart and Branch, 1970) may be useful in identifying sources of oil spills.

The contaminants most commonly associated with the petroleum industry are oil, ammonia and phenols. The standard analytical methods for ammonia and phenol are well known and widely used. The toxic threshold values for these substances are often a subject of debate, especially in the case of ammonia since its toxicity to fish is closely related to other environmental conditions such as pH, dissolved oxygen levels, hardness, etc.

Other toxic substances are present in petroleum wastes but they have not been adeuqately characterized nor determined to be present in all crude oils or waste products from crude oil, an inadequacy which is apparent in many aspects of fish toxicology. The field of human toxicology has been established for many years and autopsy techniques have been developed for isolation and characterization of toxic drugs and poisons in humans, but the field of fish toxicology is relatively new. The fish toxicologist's

problem is made more difficult by the tremendous diversity of natural and synthetic chemical compounds in the aqueous environment. A fish may absorb many compounds from the surrounding media, not all of which are toxic.

It is a difficult task to isolate and characterize the toxic substances from fish, and perhaps of utmost importance, determine the toxic threshold concentration of the substance in fish. For example, isolation and detection of .1 ug/g (parts per million) of endrin in the blood of a catfish would not be positive proof that endrin caused the death of the fish although this level has been considered to be toxic (Mount, 1966). Analyses of acute levels of toxins in fish is a research area of vital national importance. Japanese scientists have actively investigated autopsy techniques and perhaps will develop new methods for fish toxicologists (Kariya, 1968).

We have performed many bioassays with Fathead Minnow (Pimephales promelas) on oil refinery process waste waters and final effluents in a cooperative research program sponsored by Oklahoma Oil Refiner's Waste Control Council and USDI, Office of Water Resources Research to identify ichthyotoxins.

We have analyzed test waters and fish tissues for organic toxins by gas chromatography - mass spectrometry and for inorganic toxins by atomic absorption spectrophotometry. We have observed large variation in heavy metal content of minnows purchased from commercial dealers as compared to minnows spawned and raised in our laboratories. Over 400 minnows have been analyzed for heavy metal content. A typical sample of these analyses is shown in Table I, illustrating the individual variation.

The lack of background data on heavy metal content in native fish creates difficulties when interpreting data from fish suspected to have been killed by a heavy metal. Many investigators have not been aware of the individual variability of fish and have misinterpreted heavy metal content of fish. Mount (1964) utilized a ratio of zinc content in soft tissue to skeletal tissue as an index to zinc caused fish kills. Such a ratio might reduce variability among fish and permit an objective interpretation of data.

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Table I

Comparison of Heavy Metal Content in Commercial and Laboratory-Raised Fathead Minnows (*Pimephales promelas*)

Element	Commercial Minnow			Lab-Raised Minnow		
	Number Analyzed	$\bar{X}$ mg/kg	Std. Dev.	Number Analyzed	$\bar{X}$ mg/kg	Std. Dev.
Cd	20	0.83	0.56	20	0.74	0.33
Cu	20	9.80	3.90	20	13.32	3.74
Cr	20	16.15	13.42	20	3.27	1.97
Ni	20	6.39	5.03	20	12.43	4.77
Pb	20	9.46	6.26	20	4.48	1.61
Fe	20	127.20	42.73	20	73.21	13.49
Zn	20	286.66	142.26	20	129.27	19.64

TOXICANT CAUSED FISH KILL IN ALABAMA - A CASE HISTORY

by

Sam L. Spencer

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During the period 1953 to June, 1968, sixteen fish kills were reported and investigated on the Tombigbee River near McIntosh, Alabama. This area is sixty river miles upstream from Mobile in southwest Alabama. These fish kills ranged from only ten fish found up to 8,000. In each case dead fish were found in the immediate vicinity of discharges from two industries, Olin Mathieson and Geigy Chemical Companies. We were finally able to determine the reason for the lower number of fish found in some kills. One of the industries had employees picking up and disposing of the dead fish to help cut down on complaints from fishermen.

On June 19, 1968, a fish kill involving 50,000 fish valued at $10,299.00 was investigated over a sixteen-mile stretch of the river. The kill involved six species of fish; however, bluegill comprised 78% of the number of dead fish found. The bluegills exhibited reverse pectoral fins and an organo-phosphorus insecticide was suspected. Geigy Chemical Company manufactured Diazinon, an organo-phosphorus insecticide. Water samples collected after the kill by the Alabama Water Improvement Commission (AWIC) showed progressively higher concentrations of Diazinon downstream. However no sample was found to contain levels of Diazinon considered toxic to fish.

One factor that complicated the kill was extreme water level manipulation by the U. S. Corps of Engineers at a dam sixty miles upstream. The ten-year, seven-day low flow for this area of the river has been reported to be 1,100 CFS. The flow was reduced on June 17-18 to approximately 725 CFS. It was documented that all other industries except Geigy were notified by the Corps that the river flow was being reduced.

It was generally agreed by our Alabama Water Improvement Commission that this kill was caused by the low water and no attempt was made to recover damages.

However, nine days later at approximately 9:00 a. m. on June 28, 1968, two of our conservation officers observed dead fish on the Tombigbee River below McIntosh and notified our Central office. Because I had worked that area previously, I immediately flew in our Game and Fish Division airplane from Montgomery to investigate the kill. The airplane was used to determine the exact location of dead fish and to observe discharges of all other industries on the river near the kill. A visual observation of Geigy Chemical Company's discharge was made at tree top level before landing to conduct the ground investigations.

Two members of the Technical Staff of the AWIC were visiting the plant at 10:00 a. m. in relation to the previous kill. When they arrived at the plant, they were greeted with the news of the new kill. We jointly investigated the kill while it was in progress. With two boats we collected water samples and samples of distressed fish throughout the kill area.

The water samples were iced down and fish samples were frozen with dry ice. Later that afternoon, after alerting the lab, the samples were flown to the EPA Lab at Athens, Georgia.

All bluegill exhibited reverse pectoral fins and Diazinon was again suspected. At that time the dead fish were estimated at 14,000. However, they were still dying. The following day's investigation by the district fishery biologists revealed 51,246 dead fish valued at $5,857.00.

Water samples revealed Diazinon concentrations up to 200 parts per billion. Diazinon was found in Geigy's ditch also. All samples taken from the kill area exceeded the 48-hour TLM of 30 PPB and the Federal Laboratory concluded "There was a sufficient concentration of Diazinon in the area of the fish kill to kill fish."

Samples of control fish were collected several miles upstream from the kill area. Cholinesterase inhibition studies were conducted on both batches of fish by EPA. The average of specific activities of both sets of samples were 0.48 (distressed fish) and 1.55 (control fish). This gave an average inhibition of 69%. The EPA Lab reported that "Dr. Charles Weiss of the School of Public Health at the University of North Carolina reported that inhibition levels of 50.5% caused death in sunfish when exposed to organo-phosphorus compounds in water. Therefore, it is our belief that the cause of death of the fish in the Tombigbee River was inhibition of cholinesterase activity." After considerable opposition by certain

members of the AWIC, a suit for compensatory and punitive damages was filed for $250,000 against Geigy Chemical Company by the State of Alabama.

Since Geigy was the largest single employer in Washington County, the suit was filed in Mobile County. This was possible because the investigators found dead fish in Mobile, Baldwin, Clarke and Washington Counties and our law allows suits to be placed in any county where damage occurs.

The trial was held on August 17, 1970, more than two years after the kill occurred. Each side took pre-trial depositions of all persons known to have knowledge of the fish kill. My deposition involved two hours of questions by the opposing lawyer and resulted in 94 type written pages of testimony.

Pre-trial testimony obtained from Geigy personnel revealed that a plant employee had apparently opened the wrong valve and Diazinon was allowed to enter the cooling water ditch. This was the first time the employee had handled this particular operation which consisted of draining water out of a 15,000 gallon wash vessel containing a Benzene-Diazinon mixture. The Diazinon drained onto the ground, travelled through a retaining dike through an open pipe onto a gravel bed. It travelled fifty feet through this gravel and entered the cooling water ditch that by-passed the treatment facility and went into the Tombigbee River where the kill occurred.

After the law suit was filed, Geigy performed several activities to help prevent similar occurrances in the future.

1. They put operating instructions on the wash vessels for the operators to follow. None were there prior to the kill.

2. They closed off the open pipe through the retaining dike.

3. They changed the pipe and drainage system so all spills would be routed through the treatment basin.

4. Constructed piping system so cooling water could be immediately diverted into a five day retention basin if necessary.

5. Installed a underwater diffuser pipe and thus prevented us from seeing what goes into the river.

In the interim period between the kill and the trial two reports were received and checked out.

1. Geigy attempted to burn 500,000 quart aerosol insecticide containers just across the state line in Mississippi. This was true.

2. It was reported by several persons that seven tank cars of Diazinon were returned to Geigy just prior to the two fish kills and were emptied into the treatment basin resulting in an overload. No evidence could be found in the railroad shipping records to support this rumor.

The fish kill and suit resulted in considerable publicity unfavorable to Geigy. Immediately prior to the trial Geigy offered a $25,000 settlement which we declined. We felt we had a good case and might possibly end up with a $250,000 award.

We employed Dr. Charles Weiss as an expert witness to testify on the effects of Diazinon upon bluegill. This was during the time of considerable concern over the leaking containers of nerve gas being dumped into the ocean. Dr. Weiss testified that Diazinon acted upon bluegill in a fashion similar to nerve gas upon humans.

Our district fishery biologist was questioned very closely on how he arrived at this estimate of the number of dead fish. Fortunately he still had his complete set of field notes which contained the actual counts of fish in each 100 yard segment and the location of each sample point. He referred to his notes and quickly convinced the opposing lawyers that his count was probably conservative.

At the close of testimony around noon on the second day the trial took a strange course. The Judge stated that in his opinion, the State had proven negligence and was entitled to the value of the fish; however, he felt the State had failed to prove willful or wanton destruction of the fish and, therefore, was not entitled to punitive damages. He stated this was probably the most interesting case he had ever presided over; thanked the jury for listening to the trial and directed that Geigy pay the State for the value of the fish plus 6% interest since the date of the kill.

The award amounted to approximately $6,000. We figured that this probably paid for the cost of investigation, witness fees and legal fees. This brings up the question "Was it worth it?" We feel it was. In the first place, we would have investigated the

kill anyway. No fish kills have been reported below Geigy during the three years since the suit was filed. We are now able to collect largemouth bass in our fish samples immediately below their discharge.

Earlier this year Geigy announced the planned construction of additional treatment facilities that will cost ten million dollars to install and one million annually to operate. There is no doubt that the adverse publicity of the fish kill suit had a lot to do with this industry deciding to construct this new treatment facility. We feel it was worth our time and effort. We also feel this industry will be one of our "good neighbors".

AN ENDRIN-CAUSED FISH KILL IN ARKANSAS - A CASE HISTORY

by

Neil Woomer

Arkansas Dept. of Pollution Control and Ecology

Arkansas is primarily an agricultural state. The very fertile Delta and Grand Prairie regions support "big" agriculture, and all of the alteration and degradation of the natural environment that is implied in that phrase is found in these regions. Wholesale clearing of bottomland forests, drainage of wetlands, channelization of streams, and contamination of aquatic ecosystems by the unrestricted and largely indescriminate use of pesticides and herbicides - all of these activities have been carried on to the extent that no stream or lake in the region has escaped some deleterious effects.

In the area of acute fish mortalities in Arkansas, those resulting from the use or misuse of pesticides by far outnumber those from any other cause. An estimate of the number of fish killed by pesticides is not possible because many of these fish kills go unreported. Many of the people who live in these areas simply expect this sort of thing to happen and pay little attention when it does. Lately, however, an increasing number of people are learning that they should become alarmed when this happens. We are beginning to get better reporting of fish kills in agricultural areas. Efforts are also being made to instigate greater controls over the use and application of pesticides in Arkansas.

In the past two or three years we have investigated a number of pesticide-caused fish kills, some of which were more extensive and perhaps more spectacular than the one I'm going to talk about today. As any of you know who have been involved in pesticide-caused fish kills, they are notoriously difficult to pin down, and we managed to wrap this one up quite nicely.

The fish kill occurred in Plum Bayou in mid-July 1971. Plum Bayou is a typical dredged stream of some 60 miles length having its headwaters in Pulaski and Lonoke Counties in central Arkansas. The stream meanders back and forth between these counties south through Jefferson County to its confluence with the Arkansas River north of the City of Pine Bluff. It was a typical fish kill in

that it was reported on Saturday. A Game and Fish Biologist contacted me at home about 10:00 A. M. with the report that a fish kill was occurring on Plum Bayou. The report to him was from an individual who was concerned that his livestock which were pastured along the Bayou would be harmed by the water that was killing the fish. There were rather vague references to sick and dead cows and horses which on later investigation proved to have no substance.

We went by the Dept. of Pollution Control laboratories in Little Rock to get the equipment we needed for the investigation. By about 2:00 P. M. we were on Plum Bayou at a point about a mile below what was later discovered to be the source of the fish kill. The original complainant had not left his name so we had no contact with anyone who knew the area.

We noted the dead fish in the area, which were fairly numerous and had been dead about a day. We noted the condition of the water, took dissolved oxygen, temperature, pH and chemical grab samples. Everything looked good - in fact, too good for this type of stream, which is typically muddy and sluggish. The water was clear and rather swift running with thick growths of attached algae attesting to the fertility of the rice irrigation water that was later found to be contributing to the strong flow of clear water.

We headed upstream to the next access point where we found the likely source. At this point a pump was at the water's edge with a plastic hose leading up the bank to a clear spot along the road where a number of discarded pesticide containers were strewn about. A gulley was found leading over the bank to the stream which contained a puddle of a milky yellowish substance. This material was sampled. The whole area was permeated with the strong odor of insecticides, and the ground all around was saturated with spilled material. The empty containers were examined. There were a number of gallon containers of Monosodium methane arsonate (MSMA), a herbicide; some 5-gallon containers of Dinitro, also a herbicide; some 5-gallon containers of about 50% Methyl Parathion; and several 5-gallon containers of an aptly named poison called Kill-Plenty, which is a mixture of Methyl parathion and endrin.

The stream was checked upstream of this point and no dead fish were found. Live fish were seen. Just below this point for perhaps 1/4 mile no dead fish were found; also no live ones were seen. The stream was flowing rapidly. The flow is straight for almost a mile

from this point. It is dredged so that no limbs or rocks or other objects are present to catch floating fish. We assume that dead fish floated downstream or perhaps the live ones fled ahead of a heavy slug of toxicant some distance before succumbing.

We talked to some people in the area including an investigator from the Jefferson County Sheriff's Dept. who had been called by the people who feared that their stock were endangered. We also talked to two men who had been fishing in the Bayou the previous day below the suspected source point and had seen the water become cloudy and had seen fish in convulsions. They had traced the cloudy water back to the point where a farmer was filling the tanks of his spray rig with the pesticide and using the Bayou water for dilution. Spillage occurred in this operation and also back-siphoning occurred through the hose and into the stream. We got the name and address of the farmer who was leasing the land. There were also several children in bare feet running around. They were informed that playing in puddles of methyl parathion was likely to be quite dangerous. The dermal toxicity of organophosphates to humans, as you probably know, is quite high.

The next task was to try to determine the extent of the kill and if possible, to head off the slug of insecticide and find dying fish. At a point two miles below the spill hundreds of dead fish were found piled up behind a bridge pier. All of the representative species were noted and all sizes. Some live fish were seen at this point - very small sunfish which couldn't be captured. They were very skittish. You could see them lying just under the surface, with rapid opercular movements, but when you moved quickly they darted off, zig-zagging away. Very nervous, very scary, but they didn't try to hide, just darted away. The next access point was 6 3/4 miles below the spill. Here we hit pay dirt. Large carp and buffalo were found swimming at the surface, convulsive, very distressed. They would gulp at the surface, some would swim in circles on their sides, some would swim rapidly to the shore and lay at the edge, quivering and jerking. Several of these large fish were collected. Blood samples were taken by the prescribed method of cutting the fish behind the dorsal fin and collecting blood from the dorsal aorta in clean screw-top vials, which were immediately plunged in ice.

A water sample was taken for pesticide analysis and of course the other standard samples. The next access point was 8 miles below the spill (1 1/4 miles below the last sample point). Very few

dead fish were found, only very small ones and more of the small, skittish fish. The water was muddy at this point and if one smacked the water surface, several small fish would jump out of the water. The fact that small fish were dying first was, of course, further evidence of a toxicant caused kill.

It appeared that the kill was bracketed. We then contacted the owner of the spray rig who was instructed to cease the mixing operation and to clean up the spilled pesticide and empty barrels. We had collected and tagged one of the Kill Plenty containers for evidence. The label clearly stated that the material was highly toxic to fish and aquatic life and was not to be used near lakes or streams.

Game and Fish personnel went back Monday and checked to be certain that the kill had stopped and found that it had extended to about 12 miles below the point of the spill. No more fish were found dying. They made counts and estimated the value of the dead fish, finding that approximately 15,000 fish were killed which were valued at $2,05[illegible].

Analysis of the samples showed endrin to be present in the water 6 3/4 miles below the spill to the extent of 5.82 ppb and methyl parathion 8.00 ppb. The water sample obtained 8 miles below the spill contained 0.7 ppb endrin and 0.77 ppb methyl parathion. The literature showed threshold toxicity of endrin to be in the range of 0.1-0.2 ppb and TLM to be in the 0.5-1.0 ppb range depending on species. Methyl parathion is toxic in the range of 8 ppm. The blood collected from the dying fish contained 531 ppb endrin and 54.7 ppb methyl parathion.

Very little information on blood levels of fish acutely intoxicated with pesticides is available. The study done by the FWPCA on the Lower Mississippi fish kill of 1966 gives 250-280 ppb endrin as the level in blood indicative of acute intoxication in channel catfish, and about half that for gizzard shad. So it appeared that endrin was the cause of the fish kill. The report was prepared, and turned over to our attorney who filed charges against the farmer. The farmer chose not to contest our conclusions and he was fined $250 by the judge with $200 suspended. No attempt was made to recover the value of the fish killed.

This fish kill received very wide local coverage in the press and even though the penalty was inconsequential I feel quite sure that

many farmers were much more careful with their pesticide application activities in Arkansas last summer than would otherwise have been. Perhaps other fish kills as serious or moresc were prevented. If this is true it certainly was worth the effort.

POLLUTION CAUSED BY STRIP MINING IN MISSOURI - A CASE HISTORY

by

Everett H. Fuchs

Missouri Department of Conservation

Fish kills which cause the most problems and frustrations are not those caused by some unknown factors, but are those which we expect. I would like to consider situations where we are able to predict fish kills but are powerless to prevent them; situations where an industry announces its intentions not to cause problems, but nevertheless, does; and situations where the cause of a fish kill is well known and easily proven, but prosecution and collection of damages is still difficult.

This case history starts in August, 1961, on Rocky Fork Creek, 3 miles north of Columbia, Missouri, near the geographical center of the state. At that time, a water quality survey of the creek was conducted by the Missouri Department of Conservation and the Missouri Water Pollution Board, along with representatives of Peabody Coal Company. The reason for the survey was the announced intention of the coal company to start a strip mining operation in the headwaters of the creek.

In August, 1961, Rocky Fork Creek was an unpolluted Ozark border stream with a high aesthetic value. It alternated between shallow riffles and deep pools, and contained clear water and a clean gravel bottom. Rocky Fork contained a diverse fish and benthic fauna. Some of the major components of the fish fauna were largemouth bass, green sunfish, bluegills, channel catfish, white suckers, golden redhorse, and northern redhorse, along with assorted minnows and darters. Rocky Fork could be classed as a "wading creek". In Missouri, a "wading Creek" is the type that is fished with light spining gear or a light fly rod, and in an easy afternoon you expect to catch 15 - 20 pan-size sunfish and 1 or 2 keeper-size bass. Rocky Fork was heavily fished and constituted a valuable natural resource in a densely populated area.

The Department of Conservation, being familiar with strip mining operations, feared the destruction of Rocky Fork Creek. However, representatives of the coal company calmed a somewhat concerned

segment of the local population in 1961 by claiming, even in the newspapers, that they planned to mine without causing any damages. There was no reason to doubt they had anything but the best of intentions. The mine was even named after one of Missouri's most famous citizens, Mark Twain.

Despite the best of intentions, economic considerations apparently won out over environmental ones as the mid 1960's were marked by poor mining practices at Mark Twain Mine. This resulted in acid producing spoil banks and the accumulation of acid mine water in abandoned strip pits. The acid water in the pits had pH values near 3.0 and acidities near 1,000 ppm. Also, lack of reclamation work resulted in severe erosion from the unvegetated spoil banks.

On August 23, 1970, 9 years after the initial investigation, a fish kill occurred in Rocky Fork Creek. The cause-acid mine water flushed from several strip pits by a heavy local rain.

An estimated 17,000 fish were killed in 4.5 miles of the creek. Their monetary value, based upon the publication "Monetary Values of Fish", was $1,028.61. The degradation of Rocky Fork resulting from erosion off Mark Twain Mine was obvious during the fish kill investigation. Two to six inches of clay and silt covered the gravel bottom of many pools.

After the fish kill, the coal company brought in heavy equipment and took some corrective action to prevent further loss of acid mine water from their property. Needless to say, the fish were already dead.

In September 1970, Peabody Coal Company appeared in Magistrate Court and pleaded guilty to the charge of violating the Wildlife Code of Missouri, Section 252.210, which reads, "It shall be unlawful for any person to cause any deleterious substances to be placed, run or drained into any of the waters of this state in quantities sufficient to injure, stupefy or kill fish . . . . ." Causing a fish kill is a criminal offense in Missouri and is classified as a misdemeanor under Missouri Law. The fine was $500 plus costs, something less than 3 cents per fish. The $500 fine was the maximum allowed under the law.

Seven months later, in April, 1971, a break in the dam of a coal slurry pond blanketed 4 miles of Rocky Fork Creek with coal particles. Officials of the coal company claimed this was an accident. Yet,

it was known by mine personnel that the dam was leaking and they had even installed a pump to relieve pressure on the dam. After the dam broke and the coal slurry had flowed into Rocky Fork Creek, large amounts of heavy equipment were used to reconstruct the dam.

During the latter part of 1970 the strip mining operation was extended into the nearby Hinkson Creek watershed. This fact was first realized by most Columbia citizens when the coal company filed an application with Columbia city officials to strip mine about 70 acres of land in the recently annexed northeast corner of Columbia. Coming after the problems on Rocky Fork Creek, and in the new era of environmental awareness, this request to strip mine within the city limits was viewed with alarm by the local citizens. After a number of meetings between city and company officials, permission to strip mine within the city limits was finally granted, but only after elaborate reclamation plans were agreed to by the coal company.

On July 31, 1971, a fish kill occurred in Hinkson Creek. The cause-acid mine water. Approximately 500,000 gallons were drained from a strip pit into Hinkson Creek when an employee of the coal company intentionally cut the dam. The pit water had a pH of 2.3, an acidity of 4,420 ppm and an iron concentration of 270 ppm. About one mile below the pit outflow Hinkson Creek had a pH of 2.6, an acidity of 1,900 ppm and an iron concentration of 120 ppm. Above the pit outflow Hinkson Creek had a pH of 7.4, an alkalinity of 115 ppm, and an iron concentration of 0.2 ppm. An estimated 12,600 fish were killed in 1.5 miles of the creek. Their monetary value was $1,691.90. Heavy deposits of clay and silt were present in all the pools and the smaller riffles in the kill zone. On August 6, 1971, charges were filed against Peabody Coal Company. The case has not yet come to trial.

On August 26, 1971, the Missouri Department of Conservation received a letter from the Attorney General's Office of Missouri asking for an estimate of damages to Rocky Fork Creek and Hinkson Creek by pollution from the Mark Twain Mine. Under the Missouri water pollution law polluters are liable for damages to the state caused by water pollution.

The assessment of damages caused by pollution is a task in which we had little prior experience. Various approaches are possible. First, there is the monetary value of the pollution-killed fish. One method would be to charge for the value of the dead fish. But what

about the other organisms which were killed such as crayfish, aquatic insects, and other invertebrates? What is their value? Do they have no value? What about the loss of the use of the stream itself? Does the stream have a value? What about the environmental degradation, the acid water, the iron precipitates that blanketed the bottom?

Also, long term population studies in Missouri, on Courtois Creek, have shown that the annual production of fish can often equal or surpass the instantaneous standing crop. For example, we expect the standing crop of smallmouth bass in a stretch of stream in September, 1969, would have been 10 pounds per acre. In September, 1970, we would still expect the standing crop to be 10 pounds per acre. However, we also expect the harvest from September, 1969 to September, 1970 to equal or exceed 10 pounds per acre. It them becomes clear, in the fish kills under consideration, that in addition to losing the fish that were directly killed, there was also a loss of fish production for at least a year. This is true for a number of reasons.

1. Not only fish, but fish food organisms such as crayfish, aquatic insects, and even algae and diatoms were killed. This hinders repopulation even by fish movement from other sections of the stream.

2. Consider the time of year in which the kills occurred, the last day of July and the third week in August. Reproduction of fish was essentially completed for that year and there was little chance of a successful spawn and establishment of a strong year class.

3. Also, consider the break in the coal slurry pond which occurred in April. This not only wiped out benthic organisms but prevented reproduction of fishes for at least another year.

Based on the above factors the monetary damages to Rocky Fork Creek and Hinkson Creek were calculated.

Pollution of Rocky Fork Creek from Peabody Mark Twain Mine:

August 1970	Acid pollution.	
	Monetary value of fish killed	$1082.61
	Loss of fish production for one year	1082.61

April 1971	Coal dust slurry pollution. Prevention of repopulation of fish for one additional year	1082.61

Pollution of Hinkson Creek from Peabody Mark Twain Mine:

July 1971	Monetary value of fish killed	1691.90
	Loss of fish production for one year	1691.90
	Total Damages	$6631.63

I would like to consider, is $6,631.63 really compensation for the damages that were caused? I think not. The amounts calculated by the above method will be only a fraction of the actual damage because it is determined for only one segment of the stream ecosystem. Rather than trying to collect actual damages, we really need to assess a penalty which will be an incentive to prevent damage. Possibly the best way of doing this has already been adopted by the State of New York. The legal value of fish killed by pollution has been set by the legislature as $10 per fish regardless of size or species. The damage assessment on the two fish kills in Rocky Fork Creek and Hinkson Creek, based on the New York method would be $296,000. This would provide a strong incentive to prevent future problems, which is the only real solution.

REFERENCES

"Monetary Values of Fish", 1970, Pollution Committee, Southern Division, American Fisheries Society.