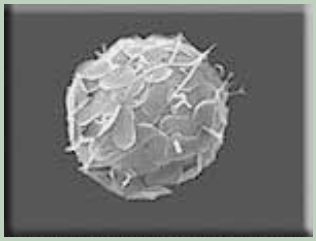
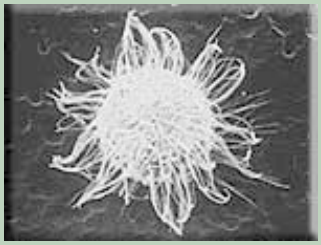
Protocols for Monitoring Pfiesteria and Related Health and Environmental Conditions in U.S. Coastal Waters





National Ocean Service
National Centers for Coastal Ocean Science
Center for Coastal Monitoring and Assessment

Protocols for Monitoring Pfiesteria and Related Fish Kills and Environmental Conditions in US Coastal Waters

by Donna D. Turgeon, Jamison L. Higgins, Danielle Luttenberg, Harris White, Marcia Orencia, Nancy Craig, and Karen Bushaw-Newton.

-EXECUTIVE SUMMARY-

State and Federal agencies need comparable and reliable data to detect bloom development and improve the ability to forecast bloom formation of species within the toxic *Pfiesteria* complex (TPC: see Glossary) in US coastal waters. NOAA's National Ocean Service held two workshops that brought together resource managers and scientific experts from Federal and State agencies and academic institutions to seek consensus on standard protocols for monitoring fish kill events that may have been caused by toxic *Pfiesteria piscicida* and for routine monitoring in waters known to support *Pfiesteria* and *Pfiesteria*-like organisms. The first, a Workshop to Standardize *Pfiesteria* Monitoring Protocols, was held 14-15 December 1998. The second, a Workshop to Standardize Fish Health Monitoring Protocols at Suspected *Pfiesteria* Events, was held 22-23 June 1999. The objective for both workshops was to establish common procedures for collecting and analyzing samples.

Deliberations by the more than 50 invited specialists on harmful algae, fish pathology, and other disciplines, including several experts on *Pfiesteria,* resulted in a set of recommendations for a multidisciplinary, monitoring and assessment program. Workshop participants agreed to make agency data sets available for an integrated electronic database that would support regional and national assessments and recommended the following.

1. A three-tier, national program to monitor water quality, fish health, and phytoplankton with concomitant goals and strategies for:

   - *Rapid Event Response* monitoring to be done by agencies during or just after significant fish kill events that may be related to toxic strains of TPC species;
   - *Comprehensive Surveys and Assessments* to be carried out in areas known to have supported toxic *Pfiesteria* outbreaks and in areas deemed to be at high risk for toxic events; and
   - *Routine Monitoring* of water quality, fish health, and phytoplankton to be conducted in areas that could support toxic strains of TPC species.

2. That phytoplankton, fish health, and a suite of water quality parameters be monitored simultaneously at sites known to support *Pfiesteria*-like species or sites at risk for toxic strains of *Pfiesteria piscicida*.

3. A set of standard parameters and methods for monitoring ambient water quality conditions, plankton, and fish health at sites suspected of having toxic strains of *Pfiesteria*.

The following report integrates the recommendations from both workshops into a set of standard protocols that could have substantial and lasting benefit to the nation's understanding of harmful algal bloom dynamics and mitigation of their impacts in coastal waters. States with *Pfiesteria* concerns held training courses and implemented these protocols in 1999.

INTRODUCTION

***Pfiesteria* and Harmful Algal Bloom Problems.** Harmful algal blooms, HABs (see Glossary: HAB), regularly threaten US coastal living resources, restrict local harvests of fish and shellfish, divert public funds to health and environmental monitoring programs, and depress local recreational and service industries. HABs occur naturally in our coastal waters, but their frequency, intensity, and distribution appear to be increasing. Blooms of familiar and previously unknown species have occurred in coastal areas, and HABs have now been identified in almost every coastal State from the Gulf of Maine through the Gulf of Mexico and north to Alaska. A recently discovered HAB species, the dinoflagellate *Pfiesteria piscicida*, was first identified in 1988 in fish cultures (Smith et al., 1988), and in 1991 at a fish kill in the Pamlico Estuary in North Carolina (Burkholder et al., 1992). *P. piscicida* has been implicated in the death of millions of fish in the nation's second largest mainland estuary, the Albemarle-Pamlico, with toxic outbreaks occurring nearly every year since 1991 (Burkholder & Glasgow, 1997). During 1997, a fish kill of 1.2 million fish was linked to a toxic *Pfiesteria* outbreak in North Carolina waters. That same year, the natural resources and the local economy in Maryland were also threatened when a fish kill of 30,000 was associated wih a toxic *Pfiesteria* outbreak in three Chesapeake tributaries on the bay's Eastern Shore.

Recently, JoAnn Burkholder and Howard Glasgow reported a second *Pfiesteria* species, *Pfiesteria shumwayae* sp. nov. (Glasgow, 2000), that also has ichthyotoxic properties. At present, these are the only two species with demonstrated ability to produce bioactive compounds that cause fish distress, disease, and death, in the toxic *Pfiesteria* complex, TPC (see Glossary: TPC). Studies are underway to test if a number of other, yet unnamed, *Pfiesteria*-like organisms, PLOs (see Glossary: PLO), found in East Coast tidal areas have similar icthyotoxic characteristics.

Additionally, dead and dying fish collected during PLO events often have skin ulcers that harbor a pathogenic and highly invasive *Aphanomyces* fungus associated with an intense granulomatous cellular inflammatory response. In controlled laboratory experiments, *P. piscicida* semi-purified toxin has been shown to destroy fish skin so that the fish become

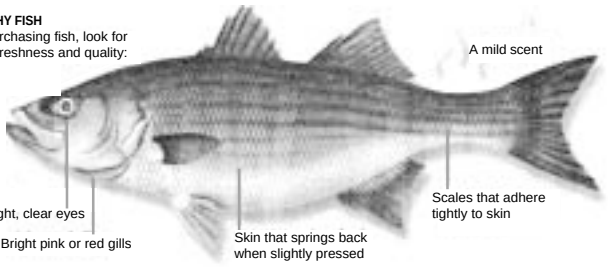
A supermarket featured in 1997 this sign that sought to ease customers' fears of Pfiesteria.

vulnerable to attack by opportunistic fungi and bacteria, and open bleeding sores develop. Research is underway to resolve the cause of such sores or ulcers in estuarine conditions, and the possible interactions of TPC species, other HAB species, and various bacterial and fungal species in lesion formation and development.

The National Oceanic and Atmospheric Administration (NOAA), along with the Environmental Protection Agency (EPA), the Centers for Disease Control and Prevention (CDC), the United States Department of Agriculture (USDA), and other Federal agencies responded in 1997 to the Maryland outbreaks of *Pfiesteria piscicida* by providing immediate support to supplement the State's monitoring and response activities. This effort combined Federal and State resources to monitor environmental conditions and assess immediate watershed land use and nutrient loadings as potential contributing factors to fish kills. The Federal government is continuing its partnership with the States by providing funding for *Pfiesteria piscicida* monitoring and assessment efforts in coastal Atlantic and Gulf of Mexico States. Additionally, a substantial program of research on the TPC and *Pfiesteria* look-alike species has been added to the multi-agency federal program that studies the Ecology and Oceanography of Harmful Algal Blooms (ECOHAB).

Finally, coastal regions that experience periodic HAB outbreaks know first hand that immediate monitoring and assessment of an event is but the first step in a longer process needed to effectively control and manage resource damage and threats to the public. For these reasons, agencies must continue to sponsor long-term programs to monitor water quality, fish health, and HABs, to analyze existing data, and to share results.

Maryland Fish and Seafood Tips—



Do not eat fish or shellfish with sores or lesions or fish that appear unhealthy.

Do not harvest or eat fish or shellfish from areas where fish are dead or dying. Maryland does not require sellers to label the source of fish, but buyers should ask if they are concerned.

Some sores, injuries, and discolorations in fish are normal from contact with fishing gear and predators. Such fish are routinely discarded by fishermen and dealers before reaching the market. But when in doubt, throw it out.

***PFIESTERIA* FACTS**

Food and Drug Administration scientists have found no evidence of *Pfiesteria* toxins in seafood, even when it was taken from waters where the microbe was blooming.

FDA scientists have examined crabs taken from the Pocomoke River after the toxic *Pfiesteria* outbreak there, as well, as oyster's placed in tanks with toxic *Pfiesteria*. In both cases, no toxins were found in the edible parts of the animals.

For information on how to handle and prepare Maryland seafood, contact the **Maryland Department of Agriculture's Seafood Marketing Program at 410-841-5820.**

In response to consumer fears about Pfiesteria piscicida *and the safety of Chesapeake Bay seafood, state and federal officials offer safe-seafood guidelines.*

Workshops for Gaining Consensus on Monitoring Protocols. NOAA, EPA, and the Interagency *Pfiesteria* Working Group agreed there was a need to standardize monitoring protocols among the many agencies that collect information on water quality, fish health, plankton, and environmental conditions associated with PLO events. NOAA proposed holding a workshop to gain consensus among State and Federal agency representatives and scientific

experts. NOAA's first Workshop to Standardize *Pfiesteria* Monitoring Protocols was held December 14-15, 1998. A second Workshop to Standardize Fish Health Monitoring Protocols was held June 22-23, 1999. Conducted by the Center for Coastal Monitoring and Assessment (CCMA), one of the National Centers for Coastal Ocean Science (NCCOS), both workshops were convened in Silver Spring, MD.

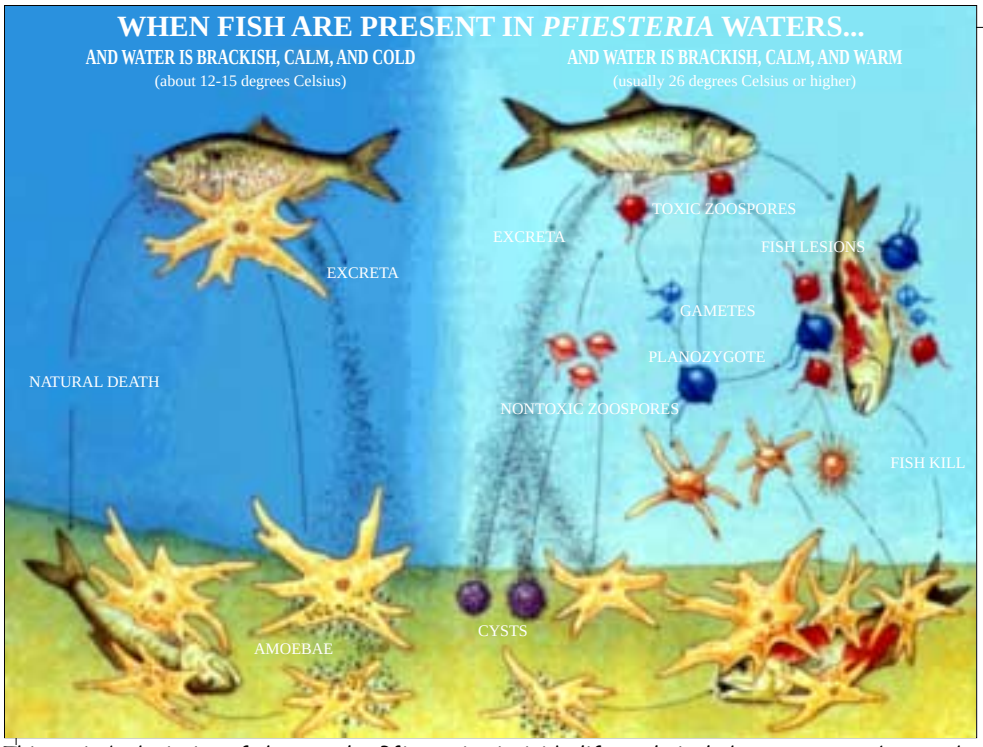
Workshop goals were three-fold. First, reach agreement on protocols for rapid-response assessments of toxic *Pfiesteria* outbreaks. Second, recommend a suite of standard parameters that should be measured (e.g. water quality, fish health, and phytoplankton) when responding to events. Third, discuss the integration of state and federal agency data sets for regional and national assessments. This report presents the deliberations of participants from both workshops.

Workshop I. Coastal state administrators from New Jersey through Texas were asked to designate two state experts who had responsibility for monitoring water quality, fish health, and phytoplankton. Representatives from other Federal agencies with interest in these topics also attended, i.e., the Environmental Protection Agency (EPA), the Centers for Disease Control and Prevention (CDC), the Food and Drug Administration (FDA), the US Department of Agriculture (USDA), and the US Geological Survey (USGS). Of the 51 attendees, 21 were from State agencies, 21 from Federal, and 9 from universities and other non-governmental organizations (see Appendix 1 for the list of attendees of Workshops I and II).

Workshop I opened with Bud Cross of NOAA and Craig Vogt of EPA reporting on their agencies' current efforts related to monitoring and research, as well as the *Federal Event Response Plan for Harmful Algal Bloom Events*—an interagency plan for a coordinated national response to toxic HAB outbreaks. The workshop focused on monitoring phytoplankton, fish health, and water quality. Over the course of two days, there were three two-hour working sessions, one for each topic, during which three break-out groups independently explored the issues related to the topic and developed recommendations through facilitated discussions. Experts introduced each topic and set the stage for the break-out group deliberations that followed. Patricia Glibert made an oral presentation on measuring water quality parameters relevant to PLO, Wolfgang Vogelbein spoke about assessing fish health, and JoAnn Burkholder presented information on field sampling and phytoplankton analytical protocols. Supporting materials that were provided to participants for this workshop included information on the water quality parameters that were then being monitored by State agencies and on how and in what electronic formats their data were stored.

After each two-hour working session, the facilitators (Water Quality - Patricia Glibert, John Pennock, and Tracy Villareal; Fish Lesions/Mortalities – Andrew Kane, Mac Law, and Helen Schurz-Rogers; Phytoplankton – Donald Anderson, Howard Glasgow, and Karen Steidinger) summarized their break-out deliberations to the full assembly. On the last day, all of the presenters, facilitators, and discussion recorders (Karen Bushaw-Newton, Nancy Craig and Danielle Luttenberg), assembled a single set of recommendations for each topic. The integrated recommendations were then reported to all participants in plenary session. Their recommendations were unanimously accepted as presented.

Workshop II. At the first workshop, the fish health break-out groups con-



This artist's depiction of the complex Pfiesteria piscicida *life cycle includes cysts, amoebae, and toxic zoospores that may cause massive fish kills in coastal waters.*

cluded that a second workshop was needed to specifically focus on the parameters and protocols needed to monitor fish health related to *Pfiesteria* events. The experts from the first workshop were asked to provide a list of candidate invitees for this focused fish health workshop. The resulting list included fish pathologists and/or field personnel from the states that had existing *Pfiesteria* programs, as well as academicians and representatives from Federal agencies (e.g., EPA, FDA, USDA, USGS).

Over the course of one and a half days, there were three two-hour working sessions, one for each of the following topics: fish collection procedures; laboratory analyses; and data storage, management, and sharing. Experts introduced each topic and set the stage for the group discussions that followed. Mac Law made a presentation on fish collection, Andy Kane on laboratory analyses, and Joe Macknis on data management.

RECOMMENDATIONS FROM WORKSHOPs I & II

Strategies for an Interagency *Pfiesteria* Monitoring Program.

Participants at both workshops recommended that water quality, fish health, and phytoplankton be monitored simultaneously at sites suspected of supporting problematic PLO. Participants outlined the following three-tier interagency program to monitor for *Pfiesteria* and *Pfiesteria*-like organisms:

1. *Rapid Event Response*
2. *Comprehensive Surveys and Assessment*
3. *Routine Monitoring*

Participants proposed a suite of standard parameters for monitoring ambient water quality conditions at sites at risk from toxic strains of *Pfiesteria*. They agreed to make agency data sets available for an integrated electronic database that would support regional and national assessments. Workshop participants reviewed and commented on this report.

1. Rapid event response **monitoring done by state agencies during fish kills or lesion events that may be related to PLOs.**

> ***Goal:*** Characterize water quality, fish health, and the phytoplankton community at sites of fish kills or fish lesion events that may be due to toxic *Pfiesteria* activity.

When responding to a potential ongoing toxic *Pfiesteria* outbreak, safety precautions including protective clothing and respirators should be used until all samples are sealed and decontaminated (e.g., North Carolina's prescribed protocols). Nitrile gloves (not latex) should be worn when touching the equipment, water, and bottles. Bottles and equipment can be decontaminated fairly well by washing in a dilute Clorox® solution. If possible, samples for monitoring should be collected along transects outward from the epicenter to beyond the kill zone. In addition to sampling phytoplankton, to confirm the presence of toxic dinoflagellates, water samples should be collected for different types of bacterial, viral, and fungal pathogens; organic chemicals (pesticides and herbicides); and toxic blue-green algal species. Laboratory samples for presumptive counts and for fish bioassays should be analyzed "blind" to guard against bias. When presumptive counts (see Glossary: Presumptive Count) are positive for PLOs in concentrations high enough to be of concern, then samples should be split and analyzed by two different laboratories to enable cross-corroboration of findings about the presence of actively toxic strains of TPC species and, potentially, newly detected toxic PLOs. It is extremely important that toxicity be verified using fish bioassays because this is the only technique at present that can be used to verify the presence of actively toxic strains of TPC species.

Water Quality. Water samples should be filtered. Minimally, measure the following:

- Station latitude and longitude
- Date and time
- Tidal stage and water depth
- Weather conditions
- Current speed and direction
- Light penetration/Secchi disc/ turbidity
- Temperature (±1° Centigrade)
- Salinity (± 0.1 ppt)
- pH (± 0.2)
- Dissolved oxygen (± 0.5 ppm)
- Dissolved ammonia
- Dissolved organic nitrogen, carbon, and phosphorus
- Dissolved nitrate plus nitrite
- Dissolved phosphate
- Dissolved silicate
- Chlorophyll *a*

Rapid response assessment during a suspected bloom of Pfiesteria piscicida.

Fish monitoring. If possible, trained fish health response personnel, including a fish pathologist, should be on-site for fish health events. Evaluate fish behavior, make overall environmental observations, and take a photographic record of the event and typical fish condition. At each site, estimate the mortality and prevalence of lesioned fish for all species collected. Subsample diseased, fresh dead, and healthy fish for pathogen analyses. Necropsy fish samples for histopathology, parasitology, and microbiology (bacteriology, mycology, virology): live/moribund* fish are preferred for these analyses but in the absence of live/moribund fish, fresh dead fish* should be collected. Moderately decomposed fish* can be used for toxicological analyses only. Severely decomposed fish* are unsuitable for any analyses.

Dead and lesioned fish from a toxic Pfiesteria event.

Live fish should be euthanized with MS222 or by severing the spine following American Veterinary Medical Association (AVMA) protocols (J. Am. Vet. Med. Assoc., 1993). Archive tissues and blood and make a photographic record of pathologies.

- To determine the geographic extent of the fish kill, use American Fisheries Society procedures (AFS, 1982; Thoesen, 1994; Meyer & Barclay, 1997).
- On a randomly selected subsample of 100 fish affected per species, "map" on a diagrammatic fish the location, type (i.e., loss of scales, ulcers, reddened/ discoloration, raised masses, normal, other), and prevalence of lesions.
- Give each fish a unique accession number on a separate data sheet; the data sheet, photographs, and jars should all be labeled with the same accession number. If possible, use a microscope in the field and examine fresh fish for gill, skin, and gut parasites.
- For histopathological analyses, collect a subsample of 25 fish per species (10 moribund lesioned, 10 moribund normal, and 5 fish from sites near but outside the kill and upstream). Either conduct a full field necropsy on 20 fish (including bacterial analysis), a 3-minute necropsy (see Appendix 2), or chop the whole fish into two or three pieces, depending on size of fish. A serological collection should be done before the necropsy. Preserve the organs for each fish in separate, labeled vials containing 10% buffered formalin (10 parts formalin to 1 part fish).
- If there is a pathologist or other trained personnel on site, then histological and microbiological samples should be field-processed and preserved with appropriate fixative and microbiological transport media for further laboratory study. For viral assays, dissect liver,

* Criteria for selecting fish for histological, microbiological, virological, and toxicological analysis are based on gill and body coloration, integrity and odor: (1) *Live/Moribund* (gills firm, body color vibrant, tissue firm, no odor); (2) *Fresh Dead* (gills firm, body color/markings still apparent, tissue firm, no odor); (3) *Moderately Decomposed* (gills pale pink, body color/markings faded, tissue spongy, slight odor); and (4) *Severely Decomposed* (gills white, body color/markings indistinguishable, tissue mushy, strong odor).

kidney, spleen (excluding intestine) and/or muscle lesions and store in Hank's Balanced Salt Solution with 10% fetal calf serum, 500 IU/ml of penicillin, and 500 mg/L of streptomycin and ice or refrigerate. These samples must be assayed within 72 hours. In the event there is no pathologist to conduct field processing, then individually bag (in plastic) and immediately place on wet ice another 25 moribund fish and 10 normal fish for laboratory bacteriological and virological analyses. If the fish to be used for histology are also to be used for microbiological analyses, then collect an additional 15 moribund fish. It is **imperative** to deliver iced samples to the laboratory for analysis within 24 hours. Microbiological analysis should be conducted according to the procedures outlined in the Fish Health Section of the American Fisheries Society "Bluebook" (Thoesen, 1994).

- For toxicological analyses, collect and flash-freeze 25 fish. For retrospective analyses, archive 25 (10/10/5) whole fish in a freezer from each kill.

<u>*Phytoplankton Monitoring*</u>. During an in-progress fish kill or fish lesion event, or when fish are acting erratically without signs of disease, collect phytoplankton samples from the immediate vicinity and preserve with acid-Lugol's solution for presumptive counts of PLO's. Additionally, collect enough fresh, unpreserved water to conduct fish bioassays, algal assays (see Glossary: Algal Assay), and for molecular probe identifications.

- Complete presumptive counts by light microscopy of plankton samples preserved with acid Lugol's solution.
- Conduct fish bioassays with fresh samples to test for toxicity; if positive, then use scanning electron microscope (see Glossary: SEM) on specially prepared cells (suture-swollen or membrane-stripped) to identify the dinoflagellates to species.
- Perform algal assays for information on other mixotrophic/heterotrophic dinoflagellate species present that eat algal prey. Among these, the PLO species that have increased in abundance in response to the presence of this algal food have all been found, thus far, to be incapable of producing bioactive compounds in enough concentration to harm or kill fish.
- In the future, as assays become available that can be used reliably to detect TPC species toxins, such assays should be used to confirm the presence of TPC toxins. Also, whenever possible, use the molecular probes that have been tested and found reliable to detect the two TPC species. Samples (water and sediment) should be taken for analysis by probes.

***2. Comprehensive surveys and assessments* conducted in areas known to have supported toxic *Pfiesteria* out-**

Biohazard III facilties are used at NCSU when conducting fish bioassays with Pfiesteria.

breaks, and in areas deemed at risk for such outbreaks. Comprehensive survey and assessment data are of particular value in guiding future management efforts.

> ***Goal:*** Determine and predict the presence and distribution of potentially toxic strains of *Pfiesteria* and *Pfiesteria*-like species.

Monitoring should be conducted at carefully selected sites (i.e., historically known fish kill/disease areas; quiet, nutrient-rich waters in deposition areas with organic sediments) during late summer and early fall.

Species identification is confirmed through scanning electron microscopy (SEM).

<u>*Water Quality*</u>. Make the same measurements that are recommended for a Rapid Response to a toxic outbreak.

<u>*Fish Monitoring*</u>. Follow the same procedures recommended for a Rapid Response to a toxic outbreak but, in addition, conduct routine monitoring pre- and post-season based on historical observations of problem times/areas. Depending on state or local needs and concerns, additional comprehensive sampling studies can be adopted. Collect fish samples from each site and necropsy for histopathology, microbiology, parasitology, bacteriology, mycology, and virology. Sampling should take place a minimum of three times a year. Make a photographic record of significant pathologies, then archive tissues and blood.

<u>*Phytoplankton Monitoring*</u>**.** Follow the same procedures recommended for a Rapid Response to a toxic outbreak.

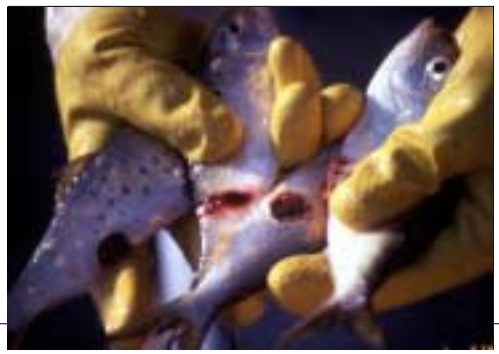
3. Routine monitoring **conducted in areas that could support toxic strains of TPC species, especially where the water quality conditions may be conducive to blooms of potentially toxic *Pfiesteria and Pfiesteria*-like species.** This broad-scale monitoring can enhance existing State monitoring activities. Note: greater efficiency, enhanced communication, and better coverage could be achieved by integrating the efforts of state agencies and academic specialists who currently monitor conduct such activities independently.

> ***Goal:*** Characterize the long-term dynamics of phytoplankton communities, indicate the presence/absence of TPC and PLO species, and determine their relationship to other members of the phyto- and zooplankton communities, water quality, and fish health.

<u>*Water quality monitoring.*</u> Measure the following set of physical and chemical water quality parameters at all sites when monitoring for suspected PLO:

- Station latitude and longitude
- Date and time

Fish health problems like these lesions may indicate the presence of TPC organisms.



- Tidal stage and water depth
- General weather conditions, wind speed
- Current speed and direction
- Light penetration/Secchi disc
- Water temperature (±1° Centigrade)
- Salinity (± 0.1 ppt)
- pH (± 0.2)
- Dissolved oxygen (± 0.5 ppm)
- Dissolved ammonia
- Chlorophyll *a*

Buoys can be used to record in situ *environmental information related to HABs.*

Fish monitoring. Note abnormal fish behavior, general health condition, and the frequency and type of external anomalies (e.g., reddened spots on the skin, lesions) when routinely monitoring for fish health at sites that might support toxic strains of *Pfiesteria* and *Pfiesteria*-like species.

- Enhance existing state programs that routinely monitor coastal waters for environmental quality, fish, and plankton populations by sampling fish populations with cast nets to capture 1-2 inch fish (10-20 casting attempts). Map fish lesion prevalence and location. Collect a subsample of these fish (5 normal and 5 abnormal fish), perform a 3-minute necropsy (see Appendix 2) or chop fish and preserve in 10% buffered formalin.
- Do histopathology processing when appropriate.

Phytoplankton monitoring. Collect integrated water-column samples (or pooled discrete depth samples), preferably at least in duplicate; or surface and bottom samples at each site routinely monitored for TPC and PLO species.

- Characterize phytoplankton community composition (to species level if possible, if not, then to dominant group and size class).
- Use the Utermöhl method (UNESCO 1978a, 1978b; i.e., with Chesapeake Bay Program's or Massachusetts Water Resources Authority's quality assurance protocols) for plankton quantification (include picoplankton but do not miss larger forms). Note that the Utermöhl method generally is not acceptable for quantifying picoplankton, so that other techniques such as Palmer cells (UNESCO 1978a, 1978b) should be used.
- Quantitative probes that enable determination of the cell number present are being tested for *Pfiesteria* spp. As their use becomes more routine, complete counts should be made for *Pfiesteria piscicida* and other members of the TPC known to produce bioactive compounds that hurt or kill fish. It is best to collect and process cells for DNA extraction: filter into a 2.5 cm glass fiber filter and immerse the filter in CTAB buffer or some other solution designed for DNA extraction. Otherwise, a sample (50-100ml) should be preserved with acid Lugol's solution and archived for later analysis. For rapid response events, DNA extraction is preferable; for routine monitoring, the second option may be acceptable. However, PCR analysis of any samples collected for *Pfiesteria* spp. should be conducted within six months.

Additionally, because *Pfiesteria* spp. have been found in the sediments at fish kill sites after the event, even when it has not been present in the water column, collection and analysis of surface sediment material is also recommended. Small samples of sediment (1gm or cc) can be assayed for the presence of *Pfiesteria* spp. using gene probes. However, these probes cannot detect toxicity. Samples should be assayed as soon as possible after collection as there is as yet no established method for sample preservation.

Integrated Data and Assessments. Both workshops included experts (Lowell Bahner, Bill Fisher, and Joe Macknis) who gave presentations on how to integrate now disparate agency data sets into a national database of more reliable and accessible data. Participants generally agreed that quality-assured data on this topic would be desirable for agency assessments and natural resource and human health decision-making. The participants agreed that they would make their data sets available and work towards assuring that spatial and temporal data were comparable if the national database were to provide user-friendly access, integrate with other databases (e.g., sediment toxicity), facilitate summarization, and link to Geographic Information System applications.

Routine water quality monitoring conducted at a site that could support Pfiesteria.

CONCLUSIONS

These Workshops filled a need expressed by both federal and state agency managers for consistent protocols to monitor suspected toxic *Pfiesteria* outbreaks. More than 60 managers and scientists who participated in the two NOAA workshops reached consensus on the need for consistency in the parameters measured, the analysis of samples collected by those States that have been monitoring over the past few years for *Pfiesteria* and PLOs, and quality control/quality assurance involving cross-corroboration of results by *Pfiesteria* specialists with demonstrated expertise in research on toxic strains of TPC species. All recommendations put forth by this group were agreed upon, including consensus on sharing agency monitoring data with other state and federal agencies. This body of experts proposed a three-tier monitoring program: (1) rapid response to fish kill events, assisted by appropriate experts; (2) comprehensive surveys and assessments in areas that have experienced or are at risk for toxic *Pfiesteria* outbreaks (as well as other harmful algal blooms); and (3) routine monitoring at sites that might support toxic strains of TPC species. Attendees called for concurrent collection of phytoplankton, fish health, and water quality samples for each tier of their program.

These recommendations will be valuable for developing and improving rapid response and environmental assessment at sites suspected of suspected toxic *Pfiesteria* outbreaks, for state and federal monitoring of PLOs, and for comprehensive surveys to determine if and where potentially toxic strains of TPC species may be present.

About NOS/NCCOS/CCMA

The National Ocean Service (NOS), a part of the National Oceanic and Atmospheric Administration (NOAA), approaches the 21st century with a clear vision — that our coasts and oceans enjoy robust health, provide a rich bounty of resources, and are wisely managed to balance competing uses.

NOS's National Centers for Coastal Ocean Science (NCCOS) works to understand the ocean and coastal environment, and the influences of human activities, by conducting research, monitoring, and assessments of these delicate and important areas.

NCCOS's Center for Coastal Monitoring and Assessment (CCMA), monitors, surveys, and assesses coastal environmental quality, habitats, and resource distribution. The Center's National Status and Trends Program conducts long-term monitoring and bioeffects assessments of toxic contaminants at more than 350 coastal sites. The Center's Biogeography Program develops regional and national assessments on living marine resource distributions and ecology.

Credits:
The Cover, and pages 5, 6, and 10 - the North Carolina Department of Environment and Natural Resources; Opposite page, page 4 diagram, page 8 right photo, page 18, the first photo - J. Burkholder; page 1 - J.A. Purcell, The Washington Post; page 2 - The Washington Post; page 7 - North Carolina State University; page 8, left photo - Florida Marine Research Institute; page 9 - P. Gilbert; page 17 - A. Kane; and page 18, the second, third, and fourth photos - K. Steidinger.

Proper Citation of this document is as follows:
Turgeon, D.D, J.L. Higgins, D. Luttenberg, H. White, M. Orencia, N. Craig, K. Bushaw-Newton. 2001. Protocols for Monitoring Pfiesteria and Related Fish Health and Environmental Conditions in US Coastal Waters. NOAA, NOS,

With contributions from:

Mark Alling
Donald Anderson
Bill Andrews
Larry Ausley
Kay Austin
Lowell Bahner
Joan Bernstein
Vicki Blazer
Scott Brown
JoAnn Burkholder
Chris Caudle
Cindy Contreras
Cindy Driscoll
Joyce Evans
Bill Fisher
Howard Glasgow
Pat Glibert
Dave Goshorn
Sherwood Hall
Wick Harlan
John Heisler
Ken Hemphill
Edythe Humphries
Andrew Kane
Jan Landsberg
Mac Law
Mary Ley
Michael Mac
Joe Macknis
Harold Marshall
Pete Maye
Ron Michaels
Cynthia Moncreiff
Bill Muir
Larry Needham
Amanada Niskar
Jimmie Overton
Shelly Paraso
John Pennock
John Plumb
Renate Reimschussel
John Schachte
Helen Schurz Rogers
Kevin Sellner
Harley Speir
Karen Steidinger
Tracy Villareal
Barbara Viskup
Wolfgang Vogelbein
Butch Younginer

U.S. Secretary of Commerce
Donald L. Evans

Acting Under Secretary for Oceans and Atmosphere and NOAA Administrator/ Deputy Under Secretary for Oceans and Atmosphere
Scott B. Gudes

Acting Assistant Administrator for Ocean Services and Coastal Zone Management, National Ocean Service
Margaret A. Davidson, Ph.D.

U.S. Department of Commerce
National Oceanic and Atmospheric Administration
National Ocean Service
April 2001