

FLORIDA'S HARMFUL ALGAL BLOOM TASK FORCE: HISTORY AND FOCUS

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INTRODUCTION

Harmful Algal Blooms or HABs occur worldwide in marine and freshwater environments from Norway to New Zealand (1,2). They can be defined as above background concentrations of aquatic nonvascular plants that have harmful effects on natural and human resources. Blooms can occur in the open ocean, bays and lagoons, and freshwater environments. Most of the harmful algae are microscopic and photosynthetic with plant pigments such as chlorophyll and accessory pigments. If the cell concentrations are high enough they can cause a visible discoloration of the water.

Dinoflagellates, a group characterized by two whip-like flagella, comprise >70 percent of the known harmful microalgae. At least one toxic dinoflagellate is documented in the fossil record back 50 million years. There are >100 toxic HAB species worldwide. Most of these species produce potent toxins, e.g., neurotoxins, ichthyotoxins, hemolysins, cytotoxins, or reactive biocompounds. HABs that cause known public health impacts in marine waters, e.g., shellfish toxicity, are routinely monitored as part of programs to reduce or prevent human exposure (IOC, website, 3).

In Florida, there are about 45 marine/estuarine and 15 freshwater toxic microalgae that can or could cause animal mortalities, animal disease, and even human illness (4). In the marine environment, the organism that causes almost annual coastal impacts is a dinoflagellate known as *Karenia brevis*, a dinoflagellate that produces a potent neurotoxin. These events have been referred to as Florida red tide.

Obviously, such living and human resource impacts result in economic impacts to local communities through reduced tourism and recreational activities, reduced work attendance, and the resultant loss of revenue. Marine and estuarine HABs affect coastal communities, but freshwater HABs can affect both inland and coastal communities because they can affect lakes, rivers, reservoirs and potable water supplies. In the case of cyanobacteria blooms, particularly in surface water supplies that are used for drinking water, toxins released into the water, if not removed, can cause mild to serious illness, even death. This has been documented for animals such as cows and dogs as well as humans (2,5). Human death from cyanotoxin exposure is rare but documented in unusual circumstances such as contaminated water for dialysis in Brazil or drinking directly from a contaminated ditch in China. The cost of these HAB events, whether marine or freshwater, is not well characterized but can be in the millions and tens of millions USD per episode depending on the extent and duration of the event. In the 1970s, two red tide

outbreaks caused by *K. brevis* affected several west coast counties for three to five months and caused an estimated 15 to 20 million-dollar USD impact to those counties (4). Today, an economic report estimates that from 1987 to 1992 the average annual cost for US marine HABs was 49 million USD (in 2000 dollars, 6).

In October of 1997, the then Secretary of the Florida Department of Environmental Protection, Virginia Wetherall, created an Ad Hoc Task Force on Harmful Algal Blooms with the then Secretary of the Florida Department of Health, Dr. James Howell, as Vice-Chairman. The Task Force was created to address harmful algal blooms in Florida waters, particularly red tides caused by *Karenia brevis* and potential fish kill/disease events caused by *Pfiesteria piscicida*. The concept for the Task Force followed a Mid-Atlantic States crisis with *Pfiesteria* fish kills and a major 1996 manatee mortality in Florida due to an uncommon winter-spring *K. brevis* red tide. These two events in 1996 and 1997 provided the impetus for the task force. The Task Force originally consisted of > 50 representatives from state agencies, e.g., DEP, DOH, Game and Fresh Water Fish, water management districts, and the governor's office, state and private universities, private laboratories, and interested parties. In 2000-2001, 35 members were reappointed and represented scientists, engineers, economists, citizens groups and government. The Task Force was originally charged with identifying existing information and data on HABs in Florida, identifying data gaps, and recommending how the data gaps could be filled by new research and monitoring programs. The Task Force created a 15 member Technical Advisory Group (TAG) to develop a review and make recommendations for the whole Task Force to consider and approve. The recommendations were made in March 1999 and presented to the HAB Task Force (4). These were approved by the Task Force and finalized in June 1999 and submitted to the Legislature by October 1, 1999, a requirement of Florida law.

Until July of 1999, the Task Force was an ad hoc body. After the 1999 legislative session it became a legislatively created Task Force with a mandate, Ch. 370.06092, F.S. The Task Force had four specific tasks and serves as an advisory body to the Director of the Florida Marine Research Institute (FMRI): 1) determine the status and adequacy of existing information, 2) develop research and monitoring priorities including detection, prediction, mitigation, and control, 3) develop recommendations for government whereby they can create response/contingency plans fitting their specific needs and resources, and 4) make recommendations to FMRI by October 1, 1999. Another statute (Ch. 370.06093, F.S.) authorized FMRI to implement a cooperative HAB program and spend authorized funds on specific marine and estuarine topics if State monies are appropriated or if federal and private grants are received. State funds were appropriated in fiscal years 1998-1999 to 2000-2001 for Task Force activities. Between July 1998 and July 2001, the Task Force received three million dollars through the Florida Fish and Wildlife Conservation Commission's Florida Marine Research Institute. In addition, Task Force projects received \$270,000 from FMRI's 2001-2002 appropriation. Each fiscal year, the appropriated monies were non-recurring which meant that each year the monies had to be requested in the agency's budget request and lobbied for by the agency and private groups interested in Task Force priorities. Other State HAB monies during this time period were appropriated for FMRI and Mote Marine Laboratory (MML), a private

laboratory in Sarasota, Florida, through FMRI. All monies were directed toward the data gaps identified in the 1999 Task Force report or toward supplementing a federal grant awarded to Florida scientists called ECOHAB: Florida with FMRI as the coordinating entity.

The March 1999 “white paper” on Harmful Algal Blooms in Florida was restricted to HABs that Task Force members identified as priorities, i.e., red tides caused by *Karenia brevis*, possible fish kills or disease events caused by *Pfiesteria piscicida*, ciguatera fish poisoning events due to *Gambierdiscus toxicus* and other ciguatera dinoflagellates, blue-green algae or cyanobacteria blooms in freshwater and estuaries, potential tumor causing dinoflagellates such as *Prorocentrum*, and macroalgal blooms caused by *Caulerpa*, *Codium*, *Gracilaria* and other marine macrophytes. Although Florida has microalgal species that cause diarrhetic shellfish poisoning and amnesic shellfish poisoning, no public health cases of these illnesses have been reported to date and therefore this is not a current problem although it still remains a potential problem. At the time of the 1999 report, the Task Force considered red tides caused by *K. brevis* to be the most significant HAB factor to impact Florida in terms of natural resources, public health and economics because of its long history of neurotoxic shellfish poisoning cases, aerosol irritation to beachgoers, almost annual occurrences with thousands of square kilometers affected, and duration of blooms from one to 18 months.

RED TIDE

Florida red tides, one type of HAB, result in mortalities of aquatic animals such as fish, birds or even marine mammals. Massive fish kills have been documented since the 1800s off southwest Florida and were even mentioned in Spanish explorer diaries. Offshore reef fish or bottom fish, on the bottom or floating, were often the first casualties to be noticed by fishermen or divers. Beached dead fish present esoteric problems in a tourist based economy and offer a challenge for removal, particularly once red tide is transported into bays and lagoons. In addition to living natural resources, Florida red tides can impact humans in several ways.

Some HABs can cause human illness or discomfort through consumption of toxic shellfish (1,2). Shellfish such as oysters, clams, mussels and other bivalves can filter out toxic microalgae as prey items and bioaccumulate the toxins. Humans in turn can become ill from toxic shellfish meats. In the case of Florida red tides, the human illness from consumption of bivalves that have accumulated the toxin from *K. brevis* is called Neurotoxic Shellfish Poisoning (NSP). With other toxic microalgae, the shellfish poisoning may have another name, like Paralytic Shellfish Poisoning. Each type of shellfish poisoning has different symptoms. Some cause death. Fortunately, NSP has not been associated with human fatalities. Florida red tides also produce a toxic aerosol that can irritate people’s eyes, nose, and throat if they are exposed to the aerosol at the beach or in an area where there is sea spray or a boat wake. Usually the respiratory effects disappear within several hours after leaving the beach or the exposure area. Respiratory irritation due to toxic particles airborne in sea spray is typical of some *Karenia* species

and has been reported for a *Trichodesmium* bloom in Brazil (2). Otherwise, respiratory irritation is not commonly associated with HABs.

The top eight Task Force priorities for red tide data and information (4) were 1) predict onset and movement of red tides in shelf waters, 2) develop rapid chemical assays to replace the mouse bioassay, 3) develop economic impact studies to evaluate losses by location or industry, 4) develop epidemiological studies to determine public health risks, 5) investigate existing technologies for dead fish cleanup, 6) investigate applicability and efficacy of control and mitigation methods, 7) determine the fate and effect of toxins in the marine environment, and 8) continue and enhance public information and outreach (4). Items 3) thru 6) and 8) were funded with State funds and items 1), 2) and 7) were funded with both federal and state funds. Since the initial funding sponsored by FMRI for Task Force initiatives, more federal funds have been made available for epidemiological and control studies through other sources.

About one-half of all Task Force dollars were devoted to red tide projects. Additional state dollars for HAB research were appropriated for FMRI and MML red tide studies. Task Force red tide projects funded included: supplemental funds for ECOHAB: Florida cruises and research; economic impacts caused by red tides, development of new, rapid immuno-chemical assay for brevetoxins, occupational exposure to red tide aerosol and its effects, a red tides and public health literature review, and removal of floating dead fish at sea. In addition to targeted Task Force funded projects, FMRI funded a business outreach and information project that relied on the Red Tide Alliance and business community for input. The product of this venture can be seen at www.redtideonline.com. Solutions to Avoid Red Tide (START), a grassroots citizen group, conducted the project, which has been extremely successful.

The legislative language that established the Task Force and its duties specifically mentioned the federal program called ECOHAB: Florida. This is a five-year federal, state, academic, and private laboratory partnership to study the development of Florida red tides and be able to predict their occurrence, movement, and landfall through coupled biophysical models. ECOHAB: Florida consists of 23 principal investigators from 13 institutions in Florida, Michigan, Mississippi, North Carolina, South Carolina and New Jersey. The program is in the fifth and final year. Task Force funds were used to supplement ship time and analysis of data for the coupled physical-biological 3D model being developed by Drs. John Walsh and Bob Weisberg of the University of South Florida based on their own studies and those of other PIs. The backbone of the ECOHAB: Florida program consists of three sampling strategies to gather data to develop the model, i.e., monthly cruises onto the shelf in a closed hourglass configuration so that boundary conditions of a control volume of seawater could be identified (USF, Dr. Gabriel Vargo), a weekly transect out to 30 miles during red tide season and at other times as well (MML, Dr. Gary Kirkpatrick), and a process cruise of several weeks during red tides in 1998, 1999, 2000, 2001 and 2002 (collaborating PIs).

Although the ECOHAB studies are not complete, it is obvious that physical processes of water movement and physical concentration at fronts as well as nutrients influences plant abundance both offshore and inshore. ECOHAB: Florida funded studies are evaluating physical processes and nutrients and their sources in relation to coastal red tides.

Nutrients can be organic or inorganic, major such as nitrogen and phosphorus, minor such as the trace element iron, and come from very diverse sources, e.g., offshore upwelling, atmospheric deposition, and terrestrial river runoff and estuarine flux.

Karenia brevis is light adapted, favored by colored dissolved organic matter near shore waters, and is able to utilize organic N which has many sources (Walsh and Vargo, pers. comm.). Different sources of nutrients coupled with phytoplankton growth strategies can influence what HABs occur when on the West Florida shelf. For example nitrate and silicate favor diatom blooms as would be expected.

One of the theories for why Florida has *K. brevis* blooms involves species adaptations and interactions in a physical and chemical matrix on the west Florida shelf. This theory can account for most of the large red tides off the west coast of Florida (7). The theory recognizes that Saharan dust contains iron and iron is deposited over the west coast during storm events when the dust is carried from South Africa across the Atlantic to the Gulf of Mexico (7,8). These dust events precede *Trichodesmium* blooms based on an analysis of the historical database. The *Trichodesmium* blooms precede *K. brevis* blooms, which lag behind the cyanophyte blooms. *Trichodesmium* is an atmospheric nitrogen fixer but requires among other elements, iron for its enzymes. This cyanophyte or blue-green alga can occupy thousands of square kilometers of the Gulf of Mexico and can be very dense. The regenerated nutrients from these blooms, particularly regenerated nitrogen, could fuel large red tides, as could decomposing dead fish from fish kills. Phosphorus and its importance in Florida red tides are also being evaluated (Vargo, pers. comm.).

In addition to providing ECOHAB support for cruises, analyses, and modeling, Task Force and other funds have supported the development of an ELISA technique for the detection of brevetoxins in seawater and shellfish (9). The end product of this test hopefully will be a rapid, accurate test kit that can replace the mouse bioassay which is the current public health standard used for NSP. Although there are many more red tide studies funded with state funds, particularly at FMRI and MML, one effort points out that gathering data is not enough. There has to be adequate storage, compatibility and retrieval of data. Accurate metadata are among the critical needs. The coastal oceans observing system for HABs in the Gulf of Mexico has the acronym HABSOS (www.ncdda.noaa.gov/habsos) and through a state-federal partnership it has the capacity to store, integrate, and layer red tide data from the Gulf states and provide information to the public and educators. The future of red tide and other HAB monitoring and forecasting in Florida and elsewhere is in automated instrumentation packages aboard various fixed and not fixed platforms such as buoys or underwater vehicles or even aboard satellites that transmit real time or near real time data to facilities that can package it for analyses. These monitoring networks will feed directly to data receiving and/or management portals such as HABSOS.

PFIESTERIA

In 1997, the Mid-Atlantic States were faced with an environmental scare from a dinoflagellate that was associated with fish kills and fish lesions (10). The organism was called the “cell from hell” because it was reported to attack and kill fish. Although put with other HAB species, these dinoflagellates do not have their own chlorophyll like most other HAB species but they are capable of taking in pigmented organisms as prey. At first the only described species was *P. piscicida* meaning “fish killer”, later another species named *P. shumwayae* was discovered (11). Most recently this second species has been described from New Zealand, Australia, Norway, Japan and other locations (12,13).

There are other *Pfiesteria* appearing dinoflagellates in estuarine and marine waters that have not been described yet, but may be in this complex. Today, it is recognized that *Pfiesteria*, particularly *P. shumwayae* is widely distributed and is being studied from the aspect of being ichthyotoxic to fish. Different studies yield different results, yet all indicate *P. shumwayae* can kill fish via one or another mechanisms (14, 15). In *Pfiesteria* there are varying degrees of toxicity that are defined by whether they can be induced to kill fish; there are also nontoxic strains. Finding such species in water or sediment samples does not automatically mean that there is a problem since they are widely distributed. If the area has blooms of *Pfiesteria* and the strain is toxic, there could be a problem that is visualized in fish kills. On the other hand, *Pfiesteria* is often thought of as causing lesions in fish, particularly characteristic lesions near the caudal fin and the anal vent. Recent evidence suggests that a fungus causes the lesions, which can be a primary pathogen (16, 17). Studies at FMRI are looking at this pathogenetic pathway for induced lesions using the fungus *Aphanomyces* (Sosa and Landsberg, pers. comm.).

The Task Force identified nine priorities for *Pfiesteria* and *Pfiesteria*-like species (4), five of which are: 1) determine occurrence and distribution of PLS (*Pfiesteria*-like species) in Florida, 2) develop molecular probes for identification and differentiation of PLS in Florida, 3) develop molecular probes for detection of PLS toxins or bioactive compounds in natural waters, 4) determine if PLS isolates are toxic or produce bioactive compounds that affect fish, and 5) determine the distribution of PLS in relation to fish disease or fish kill “hot spots” and relationship to other environmental factors such as nutrients. Targeted funds for Task Force priorities were directed to 2) and 4). Probes were developed for cryptoperidiniopsoids and *P. piscicida* and bioactive compounds were detected in cryptoperidiniopsoid and in a PLS species known as “Lucy”. The latter produced a lipid-soluble ichthyotoxic substance.

CIGUATERA

Ciguatera is known as tropical fish poisoning or TFP. It is associated with the consumption of tropical fish usually from reef areas. It can also be associated with invertebrates such as gastropods. The symptoms are similar to neurotoxic shellfish poisoning however, there is a very low human mortality rate. There are over 175 known human symptoms from rashes to aching teeth to disorientation and paralysis (2). It is thought that more than one toxin is involved and that is what accounts for the multitude

of symptomology. It is projected that there are 900 cases per year in Dade County alone (4).

The Task Force identified 10 priorities for ciguatera research (4), five of which were: 1) develop accurate and rapid tests to detect ciguatera toxins in fish, 2) expand the testing network to document occurrence and extent of ciguatoxic fish, 3) develop a better monitoring system for recording and documenting confirmed incidences of ciguatera poisoning, 4) increase awareness and training in the medical community for recognizing, documenting, and treating symptoms, and 5) determine the economic impacts, including loss of revenue and productivity due to illness and treatment. No ciguatera priorities were funded. Dr. Lora Fleming, a University of Miami epidemiologist/physician and a Task Force member, considers ciguatera a significant public health problem in Florida that deserves investigation and research support.

TUMOR PROMOTING MICROALGAE

Bioactive compounds in dinoflagellates and cyanobacteria have been associated with tumors in experimental animals (2). Since the causative organisms occur in Florida, the Task Force identified five priorities (4) for research on tumor promoting substances produced by HAB species: 1) explore potential role of biotoxins such as okadaic acid in tumor development in sea turtles, either for direct tumorigenic effects or as sublethal immunosuppressive factors, 2) determine fate and effects of toxins in food web, 3) conduct animal exposure studies to determine effects of specific tumor-promoting compounds, 4) isolate and maintain potentially toxic species for toxin or bioactive compound identification, and 5) include potential species of concern in any statewide survey. Number 5) is being addressed in event responses, but none of the priorities were funded for targeted Task Force funds.

MACROALGAE

Macroalgal blooms in Florida can be extensive whether they occur in bays and lagoons or on the shelf, east coast or west coast. These blooms have had significant impact by altering habitat and affecting trophic pathways. The Task Force identified eight priorities (4), four of which are listed here. 1) Survey *Codium* populations out to 300 ft. depth in area of known occurrence for spatial and temporal variability and determine sources of drift algae on the east coast. 2) Survey invertebrate herbivore populations and determine food sources and feeding rates. 3) Verify environmental regulators of growth, e.g., light, temperature, and nutrients. 4) Determine nutrient sources and if there is a lag in macroalgal growth influenced by nutrient source or storage. One project was funded to investigate the source of nitrogen for *Codium* and *Caulerpa* blooms on the southeast coast and map the distribution of bloom forming macroalgae.

CYANOBACTERIA (BLUE-GREEN ALGAE)

The Task Force identified cyanobacteria blooms in fresh and brackish water as an emerging HAB problem in Florida and elsewhere. Blooms occur in surface freshwaters

that either are used for drinking water or are being considered for drinking water (Mr. John Burns and Dr. Chris Williams, pers. comm.). The six Task Force priorities (4) were 1) determine the distribution of toxic and nontoxic strains, 2) develop epidemiological studies to determine public health risks, 3) develop economic impact studies to evaluate losses by location or industry, 4) determine the roles of nutrient enrichment and managed freshwater flow in blooms, 5) determine fate and effects of toxins in the food web, and 6) investigate control and mitigation methods.

Examples of cyanobacteria projects funded with targeted Task Force funds were “Assessment of Cyanobacteria in Florida Lakes, Reservoirs, and Rivers”, “Cyanobacteria Automated Detection Workshop”, “*Cylindrospermopsis* Culture for Production of Cylindrospermopsin”, and “Cyanobacteria Public Health Issues: Education and Epidemiologic Study. Many of these Task Force projects will be discussed in this meeting. Some of the results that can be highlighted are 1) *Microcystis*, *Cylindrospermopsis*, and *Anabaena* occur state-wide, 2) microcystin, cylindrospermopsin, and anatoxin-A have been found in surface waters, 3) microcystins have been found in finished drinking water, and 4) no human health problems from cyanotoxins in finished drinking water have been documented in Florida.

In June 2001, the Task Force met to review interim progress of Task Force funded projects. At that meeting summaries prepared by the principal investigators were discussed and questions were raised about the projects. The following were some of the questions about the cyanobacteria projects.

- “When will standards for toxins be available?”
- “Can blooms be prevented or reduced?”
- “What is the effectiveness of home filters on tap water for removing toxins if present?”
- “What is the lag time between the bloom and the spike at the water treatment plant?”
- “What are effective water treatments for toxins in drinking water?”
- “If toxins can be detected, can they be removed?”

For FY 2000-2001 cyanobacterial project recommendations, either specific projects were recommended to be continued, e.g., surveys, epidemiology studies, and educational materials or new time sensitive projects, e.g., culture of *Cylindrospermopsis* for toxin standards and a workshop to discuss probes or sensors to detect cyanobacteria and quantify their toxins. The latter would be appropriate for natural waters as well as water treatment plants. The workshop entitled “Cyanotoxin Detection and Quantification and Instrumentation” laid out a plan for initial collaboration and approaches. It looked at instrumentation and packaging of the instrumentation for deployment into natural water bodies or installation in treatment plants.

The three million dollars for Task Force projects to fill in data gaps was successful, but just as importantly, project results were used to leverage for additional funds and other phases of research. Additional funds were made available through other sources to 1) promote chemical or cell assays to replace mouse bioassays, 2) investigate effects of red tide aerosol on humans, 3) investigate the possibility for cyanotoxins in drinking water, 4) promote automation of detection and quantitation methods for natural waters, aquaculture, and water treatment plants, 5) promote state monitoring programs, and 6) promote new HAB studies not considered in the first three rounds of Task Force funding by pursuing new grants. Lastly, the Research Contracts Review Committee of the Task Force recommended that two workshops be held, one to address treatment methods for blooms and cyanotoxins in drinking water and the other to address public health and cyanobacteria. The Florida Department of Environmental Protection held the first meeting recently and the second is this meeting you are attending today that is sponsored by the Florida Department of Health. These workshops and other Task Force deliberations will lay the groundwork for a remaining Task Force activity, that of proposing recommendations for state and local governments to consider in the development of their response/contingency plans.

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Cyanobacteria and their toxins in Florida Surface Waters

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Introduction

Cyanobacteria have likely been an important ecological component of aquatic systems in Florida for thousands of years. However, increased eutrophication (e.g., nutrient enrichment) of lakes, rivers, and estuaries has led to a growing trend in the incidence of cyanobacterial blooms and the production of toxic metabolites. Florida's sub-tropical climate, prolonged drought, significant hydrologic modifications, and continued urbanization have also contributed to the increased occurrence of toxic cyanobacterial blooms.

Toxic cyanobacterial blooms in Florida were first recorded by Wayne Carmichael (1992) in Lake Okeechobee (1987, 1989) and Lake Istokpoga (1988). Dead cattle, signs of poisoning in laboratory mice, and contact irritation were found associated with *Anabaena* and *Microcystis* blooms. The toxin microcystin and an unidentified neurotoxin were attributed to the toxic effects found in Florida lake samples. Although toxic cyanobacterial blooms had become a major concern throughout the world, and the World Health Organization had set provisional guidelines for the consumption of microcystin-LR (WHO 1998), little information on toxic cyanobacteria in Florida waters had been published since those first toxic events identified in 1992. Following formation of the Florida Harmful Algal Bloom Task Force (FHABTF) in 1997 (Steidinger et al. 1999), cyanobacteria were recognized as a group of potentially toxic species in Florida that had increased in distribution and abundance and possibly represented major threats to water quality, ecosystems, drinking water supplies, and public health. Moreover, the FHABTF found that the growing dependence on cyanobacterial bloom effected lakes and rivers for drinking water supply was a primary impetus for the evaluation of the distribution and potential toxicity of cyanobacterial blooms throughout the state (SJRWMD 2000).

Methods

During 1998, the FHABTF, Florida Marine Research Institute, and St. Johns River Water Management District initiated a collaborative study with the Florida Department of Health and Wright State University to identify potential cyanobacterial toxins in Florida's lakes, rivers, reservoirs, and estuaries. Samples were collected and analyzed during 1999 and extended through 2000 to increase the understanding of cyanobacterial toxins in waters currently utilized for drinking water or identified as a potential future drinking water source.

Methods employed to identify alga taxa included microscopic examination and epifluorescence. Algal toxins were characterized and quantified by enzyme linked immunosorbant assay (ELISA), protein phosphatase inhibition assay (PPIA), HPLC-FI,

HPLC-UV, and LC/MS/MS. Mouse bioassays were used to characterize toxicity by intraperitoneal injection of freeze-dried sample extracts into ICR-Swiss male mice.

Results & Discussion

With the assistance of numerous state and local agencies in 1999, a total of 167 samples were collected throughout Florida, eighty-eight of these samples, representing 75 individual water bodies, were found to contain cyanotoxins. Approximately 80% of the samples containing cyanotoxins were found to be lethal to mice following intraperitoneal injection.

Seven genera of cyanobacteria were identified from water samples collected. *Microcystis* (43.1%), *Cylindrospermopsis* (39.5%), and *Anabaena* (28.7%) were observed most frequently and in greatest concentration. *Planktothrix* (13.8%), *Aphanizomenon* (7.2%), *Coelosphaerium* (3.6%) and *Lyngbya* (1.2%) were found less frequently, but at times accounted for a significant proportion of the planktonic and macroalgal species composition. *Aphanizomenon* and *Anabaenopsis* were also found consistently during the 2000 survey. Cyanobacterial blooms were common throughout the state, some of which formed continuous blooms in eutrophic and hypereutrophic systems. Many of the water bodies affected by cyanobacterial blooms were identified by water management agencies as areas of current concern or were being addressed by ongoing or proposed restoration efforts.

Algal toxins identified from bloom material during the study included hepatotoxic microcystins, neurotoxic anatoxin-A, and the cytotoxic alkaloid cylindrospermopsin. Subsequent identification of lyngbyatoxin-A and debromoaplysiatoxin were found associated with *Lyngbya wollei* blooms collected from Florida springs.

Microcystins were the most commonly found toxins in Florida waters, occurring in all 87 samples analyzed during 1999. During the 2000 survey, microcystins were detected in pre- and post-treated drinking water. Finished water concentrations ranged from below detection levels to 12.5 $\mu\text{g L}^{-1}$. Microcystins are considered the most frequently found cyanobacterial toxins around the world. Over 60 structural variants of this cyclic peptide have been reported, causing considerable concern due to their high chemical stability, high water solubility, environmental persistence and exposure to humans in surface water bodies. The World Health Organization has set a provisional consumption limit of 1 $\mu\text{g L}^{-1}$ for microcystin-LR (WHO 1998). The mammalian toxicity of microcystin occurs by active transport across membrane boundaries and is mediated through binding to protein phosphatases (Runnegar et al. 1991; Falconer et al. 1992). Analysis of protein phosphatase inhibition activity, an index of microcystin bioactivity, was found to be positive in 44 (69%) of 64 Florida samples tested. There is limited evidence of tumor promotion (Sato et al. 1984; Falconer 1991, Nishiwaki-Matsushima et al. 1992, Wang and Zhu 1996) and clastogenic dose-related increases in chromosomal breakage by microcystin (Repavich et al. 1990), but no mutagenic evidence has been reported (Runnegar and Falconer 1982, Repavich et al. 1990).

Anatoxin-a is a potent neurotoxic alkaloid that has been frequently implicated in animal and wildfowl poisonings (Ressom et al. 1994). It is considered a nicotinic agonist that binds to neuronal nicotinic acetylcholine receptors which leads to depolarization and a block of electrical transmission in the body (Soliakov et al. 1995). At sufficiently high doses (oral LD₅₀ = >5,000 µg kg⁻¹ body weight), it can lead to paralysis, asphyxiation, and death (Carmichael et al. 1975, Carmichael 1997). The authors have suggested that unexplained bird and alligator mortality events during cyanobacterial blooms in Florida lakes may be due to exposure to neurotoxic compounds produced by species of *Anabaena*, *Aphanizomenon*, and *Cylindrospermopsis*. Anatoxin-a was found in three finished water samples and in tissues from Blue Tilapia and one White Pelican during surveys in 2000. Anatoxin-A was found in the gut and liver of a White Pelican and in Blue Tilapia (0.51 to 43.3 µg g⁻¹) and in finished drinking water (below detectable limits to 8.46 µg L⁻¹). Preliminary evidence suggests that *Cylindrospermopsis raciborskii* isolates from Florida lakes and rivers can produce anatoxin-a (Mark Aubel, CyanoLab; Wayne Carmichael, Wright State University).

All 1999 samples containing the organism *Cylindrospermopsis* were positive for the toxin cylindrospermopsin. Nine (9) finished drinking water samples collected during the 2000 survey were positive for cylindrospermopsin and ranged in concentration from 8.07 to 97.12 µg L⁻¹. Identification of the algal toxin cylindrospermopsin during this study represented the first record of this hepatotoxic alkaloid in North America. This toxin primarily affects the liver, but extracts given orally or injected in mice also induce pathological damage to kidneys, spleen, thymus, and heart (Hawkins et al. 1985, 1997). Cylindrospermopsin is a potential important contaminant of drinking waters in Australia, Central Europe, South America, and the United States. The toxin was identified after 138 children and 10 adults were poisoned following a *Cylindrospermopsis* bloom and copper sulfate applications in a water supply reservoir on Palm Island, Australia (Hawkins et al. 1985). Over 69% of the affected individuals required intravenous therapy for electrolyte imbalance, and the more severe cases for hypovolemic and acidotic shock (Byth 1980). The oral toxicity or lethal dose of cylindrospermopsin has been reported between 4.4 and 6.9 mg kg⁻¹ mouse body weight with death occurring 2-3 days after treatment (Humpage and Falconer 2002). In experiments where cell free extract of *Cylindrospermopsis* was administered to mice in drinking water over 90 days, no pathological symptoms were recorded up to a maximum dose of 150 mg kg⁻¹ day⁻¹ (Shaw et al. 2001). Humpage and Falconer (2002) suggest a Tolerable Daily Intake and Guideline Value for cylindrospermopsin in drinking water of 1 µg L⁻¹ based on an oral No Observed Adverse Effect Level of 30-µg kg⁻¹ day⁻¹ and a Lowest Observed Adverse Effect Level of 60-µg kg⁻¹ day⁻¹.

Lyngbyatoxin-a and debromoaplysiatoxin have been identified from *Lyngbya wollei* and *L. majuscula* samples collected from Florida springs and marine embayments, respectively (J. Burns, N. Osborne, and G. Shaw, unpublished data). Concentrations have ranged from below detection to approximately 4.5 mg kg⁻¹. The aplysiatoxins and lyngbyatoxins are considered dermatotoxic alkaloids, causing severe dermatitis among swimmers and other recreational users of water bodies that come into direct contact with the organism (Mynderse et al. 1977, Fujiki et al. 1990). Aplysiatoxins are lethal to mice

at a minimum dose of 0.3 mg kg⁻¹ (Moore 1977). Aplysiatoxins and lyngbyatoxins are also considered potent tumor promoters and protein kinase C activators (Fujiki et al. 1990). Osborne et al. (2001) reviewed the human and ecological effects of *Lyngbya majuscula* blooms and reported acute contact dermatitis in Hawaii, Japan, and Australia. One potential death due to exposure via ingestion of turtle meat containing lyngbyatoxin-a was reported by Yasumoto (1998). Recent reports from Cambodia (December 2002) include 140 illnesses, 100 individuals hospitalized, and 3 deaths following consumption of marine turtle meat suspected to contain *Lyngbya* toxins (Helen Rogers, Centers for Disease Control & Prevention, Atlanta, GA). Severe dermatitis has also been reported in Florida following recreational activities in waters supporting *Lyngbya wollei* blooms in Florida's springs (John Burns, Ian Stewart and G. Shaw, unpublished data).

Summary

The occurrence of toxic cyanobacterial blooms in Florida have become more prominent following increased growth, declining groundwater supplies, and identification of impaired surface waters as future drinking water sources. Cyanobacterial toxins have been identified in source waters used for drinking water supply and in post-treated drinking water during algal bloom events. Algal toxin concentrations in post-treated drinking water have exceeded existing and proposed World Health Organization guidelines for the oral consumption of microcystin and cylindrospermopsin. Severe dermatitis has also been reported by swimmers in Florida springs where *Lyngbya wollei* mats have expanded following significant increases in groundwater nitrate levels.

This information should be useful in developing appropriate Total Maximum Daily Loads for impaired Florida waters that do not currently meet their designated use. It could also support further efforts to characterize potential ecological and human health risks due to toxic cyanobacterial blooms. Identification of algal toxins in finished drinking water and reports of severe skin irritation following contact with toxic cyanobacteria should be utilized for justification and implementation of increased monitoring of potentially toxic cyanobacterial blooms by surface water managers and water utilities. Epidemiological studies may also be required in Florida to assess potential human health risks due to algal toxin consumption at the tap and for those exposed to cyanotoxic blooms during recreational users of lakes, springs and rivers.

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The Distribution of Potentially Toxic Cyanobacteria in Florida

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The cyanobacteria, or blue-green algae, are the oldest group of algae on earth, dating back over 3 billion years, and are still one of the most widespread and abundant forms of photosynthetic organisms. The fact that a number of species in the group not only form blooms but also produce powerful toxins, places them into a position of particular human interest and concern. This concern is particularly acute in Florida, which is exceptionally blessed with an abundance of aquatic natural resources. There is perhaps no other state in the continental United States where the quality of life and economy is more closely tied to the integrity and sustainability of these resources. There is no doubt that the issue of potentially toxic cyanobacteria blooms deserves the attention of researchers and managers throughout the state. In this paper we examine this issue from the perspective of our research activities around the state over the past twenty years. During this time we have carried out long-term studies of planktonic algae populations in three major marine ecosystems (Florida Bay, Indian River Lagoon and the Suwannee Estuary) and five major freshwater ecosystems (Lake Okeechobee, the St. Johns River, Lake Griffin, the Rainbow River and the Suwannee River)(Figure 1). Despite the broad range of environmental conditions represented by these seven ecosystems, cyanobacteria play an important role in the structure and function of all seven. There are a number of factors that play a prominent role in the control of algal blooms in many of these ecosystems, including trophic state, water exchange rates, light availability and growth-limiting nutrients status.

Algal Blooms and Trophic Status

Many of Florida's lakes and rivers have an ample supply of nutrients to sustain high levels of phytoplankton biomass. In water management and limnological circles such lakes are called eutrophic, Greek for 'well-fed'. One of the common public misconceptions is that eutrophic conditions are a certain sign of the influence of human activity. Actually, eutrophic lakes and rivers can arise naturally due to accumulation of organic matter over time or edaphic factors, like surface or ground water input from naturally nutrient-rich sediments. These phenomena, in part, explain the high frequency of eutrophic lakes in certain regions of central Florida, where phosphorus-rich sediments are widespread, as indicated by the phosphate mining activities in the region. This does not imply that humans do not play a major role in promoting algal blooms. There is substantial evidence that the explosion of human development in Florida over the past century has elevated the trophic status of many lakes and rivers, in some cases dramatically. With this elevation in trophic status, the frequency and intensity of algal

blooms has increased. In Florida, cyanobacteria have been exceptionally successful in taking advantage of the increase in bloom potential (Canfield et al. 1989).

The Distribution of Potentially Toxic Algae in Florida

For most lakes and rivers in Florida, historical records of algal composition are absent or very sparse, making it almost impossible to define the distribution of potentially toxic algae species in past years. Even where significant data sets are present, information on the toxicity of algal species is largely absent. Therefore, in most cases it is only possible to discuss the distribution of ‘potentially’ toxic algae species, which currently includes the species listed in Table 1. The five cyanobacterial genera (groups) that contain the freshwater species of greatest concern are *Microcystis*, *Oscillatoria*, *Anabaena*, *Cylindrospermopsis* and *Aphanizomenon*. There are also groups of cyanobacteria that form blooms, but whose toxicity is not as well established, like *Lyngbya* and the widely occurring marine species *Synechococcus*.

Since the greatest risk associated with cyanobacterial toxins is found under bloom conditions, it is obvious that eutrophic and hypereutrophic lakes, rivers and coastal ecosystems are the focus of much attention. As expected, recent surveys of Florida indicate that cyanobacteria are common features of ecosystems that exhibit elevated trophic status (Canfield et al. 1989). Even in lakes, rivers and estuaries subject to high levels of nutrient loading, the expression of that nutrient availability in the form of algal bloom can be effected by other factors, like residence time, light limitation or biomass loss through grazing, senescence or disease. The influence of these factors is manifested in our observations of the seven aforementioned ecosystems.

Lakes

Lake Okeechobee, Florida’s biggest lake and the second largest lake in the continental United States, has been the site of some of the most widely publicized algal blooms in the country. Since intensive monitoring of the lake began in the 1970’s, blooms of the cyanobacteria *Microcystis*, *Anabaena*, *Oscillatoria* and *Cylindrospermopsis* have been observed on numerous occasions, in many cases covering over 100 square miles of the lake. Most of the largest blooms have been confined to a crescent-shaped region along the western side of the lake. The center and eastern part of the lake generally contain relatively low algal standing crops, due in large part to the low light availability caused by the re-suspension of muddy flocculent sediments (Phlips et al. 1995, 1997). These observations demonstrate that the distribution of potentially toxic algal species in structurally complex ecosystems, like Lake Okeechobee, may not be uniform (Phlips et al. 1993). Cyanobacteria blooms in Lake Okeechobee also exhibit significant temporal variation. The intensity and composition of blooms vary on both intra- and inter-annual time scales (Figure 2), reflecting changes in lake stage, nutrient limitation and nutrient loading (Phlips et al. 1997).

In contrast to the spatial and temporal heterogeneity of blooms in Lake Okeechobee, in some lakes, blooms of key species can persist for extended periods of time. For example,

bloom concentrations of *Cylindrospermopsis* in the hypereutrophic Lake Griffin have been observed to extend over the entire lake for periods in excess of a year (Figure 3). The ability of algae blooms to persist for long periods of time in Florida is in part attributable to the sub-tropical climate experienced over the peninsula, which diminishes the seasonality of blooms so often seen in temperate latitudes. This does not mean that temporal variations of climate are not important in Florida. Recent drought and flood periods have had a dramatic impact on the ecology of Florida's aquatic environments. In Lake Griffin, drought periods have been characterized by low water turnover rates and elevated algal standing crops. Conversely, flood periods, like the *El Nino* of 1997/98, can reduce phytoplankton concentrations for periods of time by increasing the rate water turnover and dilution of biomass. This is a feature shared by many lakes in Florida, which are often characterized by shallow depths and low water volume.

From a broader geographic perspective, there are few regions of Florida where blooms of the major species of algae outlined above cannot be found in a multitude of nutrient-rich lakes. While some regions of Florida have a higher proportion of eutrophic and hypereutrophic lakes and rivers than others, recent monitoring of over almost a 1000 Florida lakes indicates that about half are subject to at least periodic algal blooms, if defined as chlorophyll *a* concentrations exceeding 40 µg / liter (Lakewatch, 2003). There are also factors other than trophic status that encourage the predominance of cyanobacteria over other algal groups in Florida. For example, *Anabaena*, *Cylindrospermopsis* and *Aphanizomenon* all fall into a select category of photosynthetic organisms that can convert biologically unusable elemental nitrogen (which comprises 80% of the air) into the plant nutrient ammonia through a process known as nitrogen fixation. Since the growth of algae in many of Florida's lakes and rivers is periodically limited by the supply of nitrogen fertilizer (like ammonia) the ability of these organisms to carry out nitrogen fixation places them at a distinct selective advantage over all other algae and plants, which are incapable of fixing nitrogen.

Another example of an advantageous feature shared by *Microcystis*, *Oscillatoria*, *Anabaena*, *Cylindrospermopsis* and *Aphanizomenon* is buoyancy regulation. Unlike many species of algae that depend on mixing energy to stay afloat, these five cyanobacteria can adjust their position in the water column by inflating or deflating gas chambers in their cells. This ability is a great advantage in highly productive lakes and rivers where light available for photosynthesis can be restricted to only the top portion of the water column. This attribute also explains why blooms of the aforementioned cyanobacteria are often observed as the dreaded surface scum.

Rivers

Many of Florida's rivers are characterized by high nutrient levels, due to both natural and anthropogenic factors. However, these high nutrient levels do not always lead to planktonic algae blooms. Most often, this is due to the impact of short residence time. The Suwannee and Rainbow Rivers are good examples of the latter phenomenon. While both rivers contain sufficient concentrations of both nitrogen and phosphorus to sustain bloom concentrations of planktonic algae, neither does, due to the fact that water seldom

stays in either system for more than a few days, preventing the accumulation of planktonic biomass.

The Suwannee River is a blackwater river, which originates in the Okefenokee Swamp in southeastern Georgia before winding southward 394 km to the Gulf of Mexico. The Suwannee River, with its tributaries the Alapaha, Withlacoochee, and Santa Fe Rivers, drains 28,500 km² of southern Georgia and north-central Florida. Phosphorus-rich freshwater inflow from the upper portions of the watershed are augmented by substantial nitrogen-rich groundwater contributions from numerous springs along the riverbanks, giving the river the second highest mean annual discharge of any river in Florida. Despite these high nutrient levels, their impact is not generally realized as plankton production within the river (Bledsoe and Philips 2000), although accumulations of benthic cyanobacteria, like the potentially toxic *Lyngbya*, can occur, particularly when the normally highly colored river water becomes clearer due to drought conditions in the watershed. In the case of the Rainbow River, the shallow depths and crystal clear water provide the light necessary to support large populations of the potentially harmful benthic cyanobacterium, *Lyngbya* (Phlips 1999). Blooms of *Lyngbya* have become a management issue in a number of Florida's river systems, like Crystal River.

In contrast to the high flow environments of the Suwannee and Rainbow Rivers, there are rivers in Florida whose gentle slope results in very low flow rates and sufficient residence time to sustain planktonic algae blooms. Most prominent among these is the St. Johns River, which during drought years can actually flow backwards for significant periods of time due to tidal forces. Cyanobacteria blooms in the St. Johns River can be both extensive and intensive; reaching chlorophyll *a* concentrations in excess of 100 µg / liter (Phlips et al. 2000). Blooms of all five of the potentially toxic algae groups outlined above have been observed in the river (Cichra and Philips 2002). Sometimes these blooms follow a recurring pattern. For example, blooms of *Cylindrospermopsis* and *Anabaena* in the St. Johns River occur in the summer, although the intensity of blooms can vary from year to year (Figure 4). In contrast, blooms of *Microcystis* in the same river are less predictable, occurring at any time during the year.

Estuaries

The estuaries of Florida are often the recipients of the nutrient loading experienced in their respective watersheds. Therefore, the impact of human development on land is by no means restricted to freshwater ecosystems. By the same token, the impact of human development on coastal ecosystems extends beyond just the issue of nutrient loading. In restricted coastal ecosystems like Florida Bay and the Indian River Lagoon, changes in water flow patterns can have as dramatic an effect on the dynamics of algal blooms as changes in nutrient loading.

In Florida Bay, a 1600 km² inner-shelf lagoon located just south of the Everglades and west of the Florida Keys, a network of very shallow mud banks located throughout the Bay restrict water exchange between inner regions of the bay and the Atlantic Ocean and Gulf of Mexico. One of the dominant features of phytoplankton communities in Florida

Bay is the potentially harmful picoplanktonic cyanobacterium *Synechococcus*, which frequently reaches cell densities in excess of 10^6 cells / ml in parts of the bay (Phlips et al. 1999). The relative dominance of *Synechococcus* during bloom events in Florida Bay is manifested by temporal patterns of phytoplankton biovolume at four sampling sites (Figure 5) representative of ecologically distinct regions in the bay (Phlips et al. 1995b). The north central region of Florida Bay is the focal point for blooms of *Synechococcus*, which spread south during the fall and winter due to shifts in wind direction. The physical restriction of tidal water exchange between the interior of the bay and the surrounding oceanic environment contribute to the build up of cyanobacterial standing crop by lowering the rates of water turnover. Prior to human development, surface water in South Florida flowed from Lake Okeechobee down through the Everglades and into Florida Bay. Extensive development of South Florida has restricted this flow and exacerbated the conditions favorable for algal blooms in the bay.

In another one of Florida's largest flow restricted estuaries, the Indian River Lagoon, the picoplanktonic cyanobacterium *Synechococcus* is once again a prominent component of planktonic algae blooms (Phlips and Badylak 2002). Although dinoflagellates and diatoms are the dominant bloom-forming groups in the lagoon overall, numerous cyanobacterial blooms, dominated by *Synechococcus*, have been observed in the most flow-restricted portions of the lagoon, like the Banana River region (Figure 6). Bloom levels of another cyanobacteria genus known to contain toxic species, i.e. *Oscillatoria*, have also been observed in the lagoon. The latter events appear to be associated with periods of high rainfall and may be indicative of washout of freshwater algae into the lagoon, rather than *in situ*, production (Phlips, unpublished data).

On the west coast of Florida there are a number of major river discharges that contain high concentrations of nutrients. These include the two rivers with the highest freshwater discharge in Florida, the Apalachicola and the Suwannee Rivers, both of which flow into the area of Florida known as the Big Bend. The Big Bend is noted as one of the most productive areas of Florida's coastline and certainly the nutrient discharged by these rivers contributes to that productivity. There is, however, concern that continued escalation of nutrient levels due to anthropogenic activities may soon create serious environmental problems, similar to those experienced in the Mississippi delta, like hypoxia and seagrass die-off. In the estuarine region of the Suwannee River, blooms of planktonic algae are common and are most often dominated by diatoms and dinoflagellates (Bledsoe and Phlips 2000, 2003). There are, however, periods in which small spherical cyanobacteria dominate the plankton community and on occasion reach bloom proportions. Blooms of these species are most common during the summer, particularly during periods of low freshwater outflow (Figure 7). The potential toxicity of these species is not known.

Control of Potentially Toxic Algae Blooms

The ideal long-term strategy for dealing with toxic algae is to prevent or reduce the occurrence of blooms. The most fundamental way of addressing this challenge is to reduce the availability of nutrients that support the growth of algae. There are lakes,

rivers and estuaries in Florida where such water management efforts have or could be used to achieve this goal. The restoration of lakes, rivers and their watersheds is a major on going activity in many regions of Florida. The reduction of nutrient availability has been pursued in several different ways, including the control of external nutrient loading from the watershed and the binding or removal of nutrients within a system using chemical treatment (e.g. alum) or sediment removal (e.g. dredging) (Cooke et al. 1993). In terms of ecosystems subject to high levels of external nutrient loading, long-term improvements of conditions generally require a strategy for the reduction of external load.

There are of course ecosystems where it may be impractical, too expensive or fundamentally impossible to eliminate the occurrence of potentially toxic algae blooms. In such cases it may be necessary to carefully weigh the risk associated with certain activities in the system and make appropriate use recommendations or requirements. Unfortunately, there is currently insufficient information for most systems to make a meaningful risk determination at the present time. To make such a determination a number of important pieces of information about individual lakes and rivers need to be available to water managers, including: (1) the distribution of potentially toxic algal species in time and space (i.e. temporal and spatial patterns), (2) the potential impact of future changes in the character of the ecosystem and its watershed on algal communities, (3) the amount and relative strength of the specific toxins associated with blooms of the strains of potentially toxic algae species present in the ecosystem in question and (4) the pathways and mechanisms by which critical biological elements of the ecosystem, including humans, may be exposed to, accumulate or pass-on toxins to other organisms.

It is well known that different strains of potentially toxic algae species can vary in toxicity. Some of this variation is due to genetic differences between populations of algae, some of the variation is due to the impact of environmental variables (e.g. temperature, light, nutrient levels) and some of the variation is due to the life cycle of the algae (e.g. young versus old blooms). Therefore, determining the risk associated with algal toxins in individual lakes, rivers or estuaries require a well-designed and implemented monitoring program that can be used to gather essential information.

For short-term remediation of problems with algal blooms in smaller systems, including man-made ponds, traditional algaecide applications, like copper-based products, have been employed for control. Unfortunately, such applications can worsen the problem by accelerating the release of toxins stored inside algal cells. In dealing with toxic algae blooms it may be necessary to employ an alternative approach that removes algae from the water column without killing the cells. Research is underway to explore alternative methods for treating blooms, like flocculation. Overall, post-bloom treatment is an expensive alternative. In situations where surface water is used for human consumption, such expense may be acceptable and a number of water treatment technologies for eliminating toxicity are in the process of development (Chorus and Bartram 1999).

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Table 1. Major groups of toxins, their primary site of action and the algae groups that contain species and strains capable of producing the toxins. Underline indicates the group(s) it is most commonly associated with. Note that the potential for toxin production is restricted to certain strains within the algal groups and can vary depending on environmental conditions (After Chorus and Bartram 1999).

Toxin	Site of Action	Algal Group
Microcystin	Hepatotoxin – liver damage	<u>Microcystis</u> , <u>Anabaena</u> , <u>Planktothrix (Oscillatoria)</u> , <u>Hapalosiphon</u> , <u>Anabaenopsis</u> , <u>Nostoc</u>
Cylindrospermopsin	Hepatotoxin – liver and other associated organs	<u>Cylindrospermopsis</u> , <u>Aphanizomenon</u>
Nodularin	Hepatotoxin – liver	<u>Nodularia</u>
Lyngbyatoxin	Dermatotoxin – skin and GI-tract	<u>Lyngbya</u>
Aplysiatoxin	Dermatotoxin – skin	<u>Lyngbya</u>
Anatoxin	Neurotoxin – nerve synapse	<u>Anabaena</u> , <u>Aphanizomenon</u> , <u>Planktothrix (Oscillatoria)</u>
Saxitoxin	Neurotoxin – nerve axons	<u>Anabaena</u> , <u>Aphanizomenon</u> , <u>Lyngbya</u> , <u>Cylindrospermopsis</u>

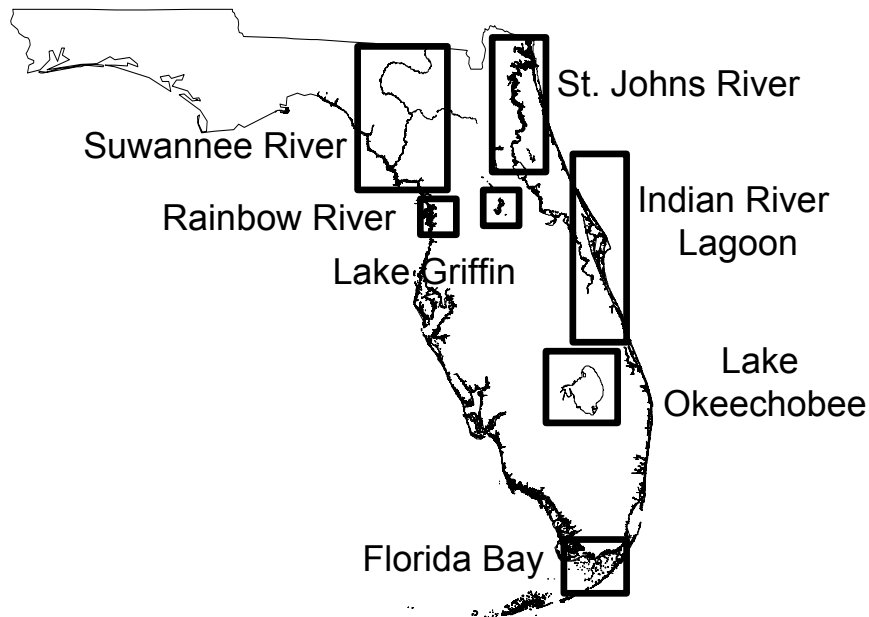


Figure 1. Long-term study of planktonic algae was conducted on three major marine systems (Florida Bay, Indian River Lagoon and Suwannee River Estuary) and five major

freshwater ecosystems (Lake Okeechobee, the St. Johns River, Lake Griffin, Rainbow River and the Suwannee River) in Florida.

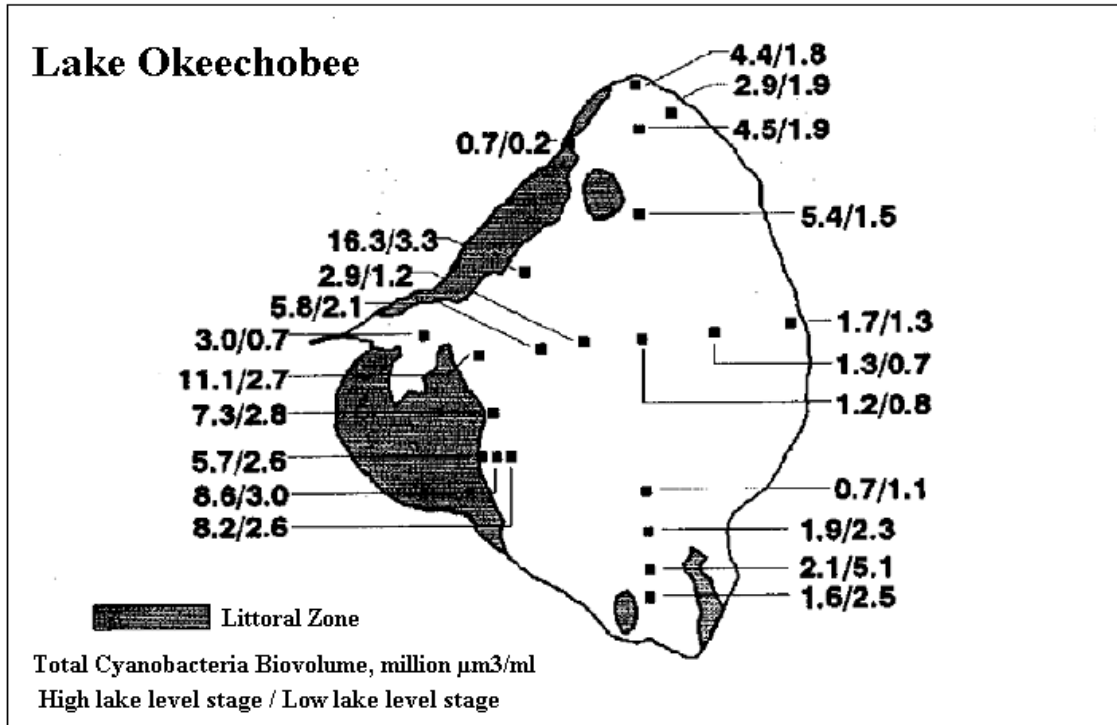


Figure2. The spatial and temporal distribution of cyanobacteria biomass (million $\mu\text{m}^3/\text{ml}$) during a high lake stage year (first number)(1988-1989) and a low lake stage year (second number)(1989-1990)(Cichra et al. 1995).

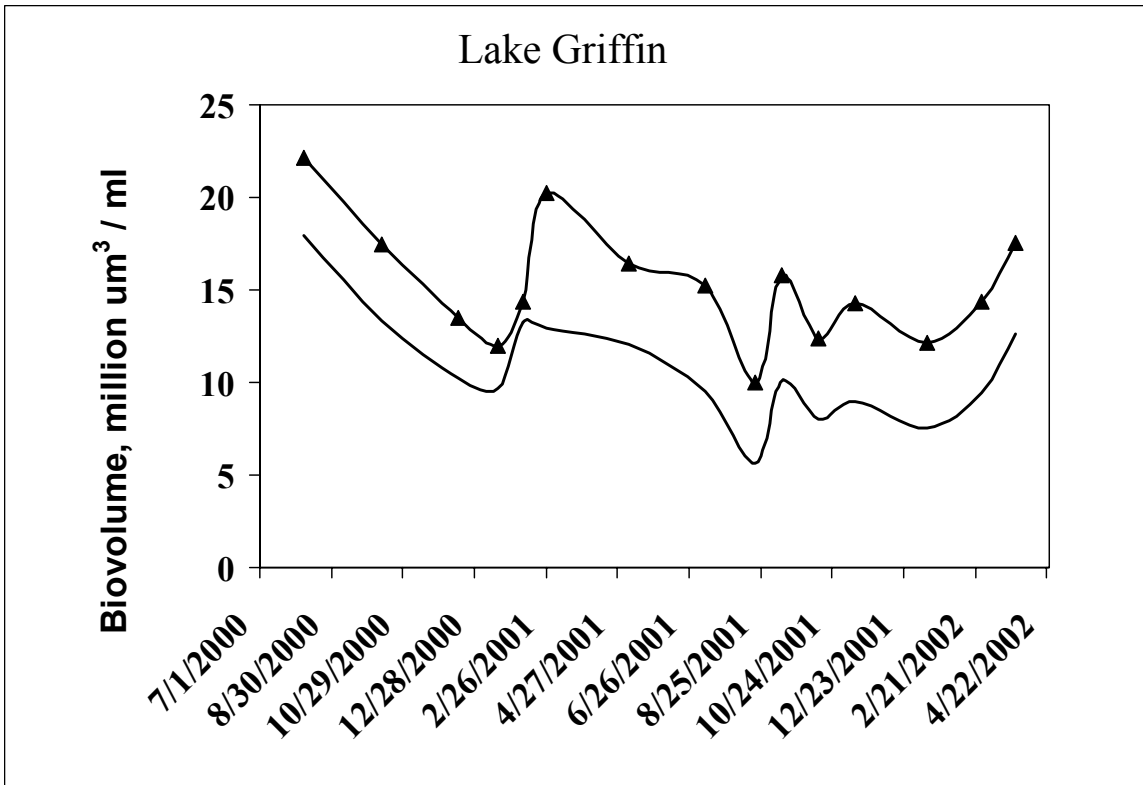


Figure 3. Two key taxa of cyanobacteria, *Cylindrospermopsis* and *Oscillatoria* (solid line) played an important role in the overall phytoplankton biomass (triangle)(million μm^3 / ml) in the Lake Griffin (Phlips, Cichra and Frost 2002).

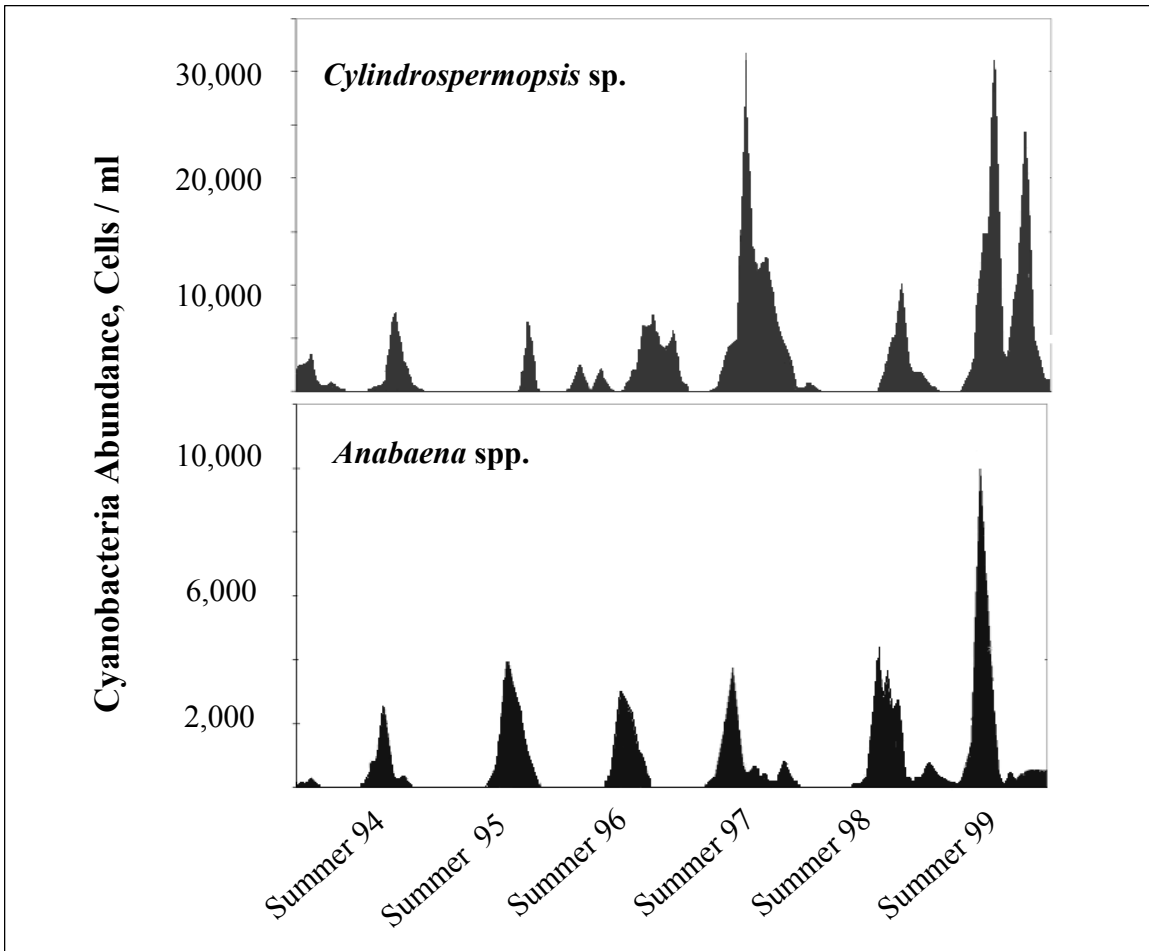


Figure 4. The temporal distribution of two potentially toxic forms of cyanobacteria in the St. Johns River (cells / ml) from 1994 through 1999 (Cichra and Philips 2002).

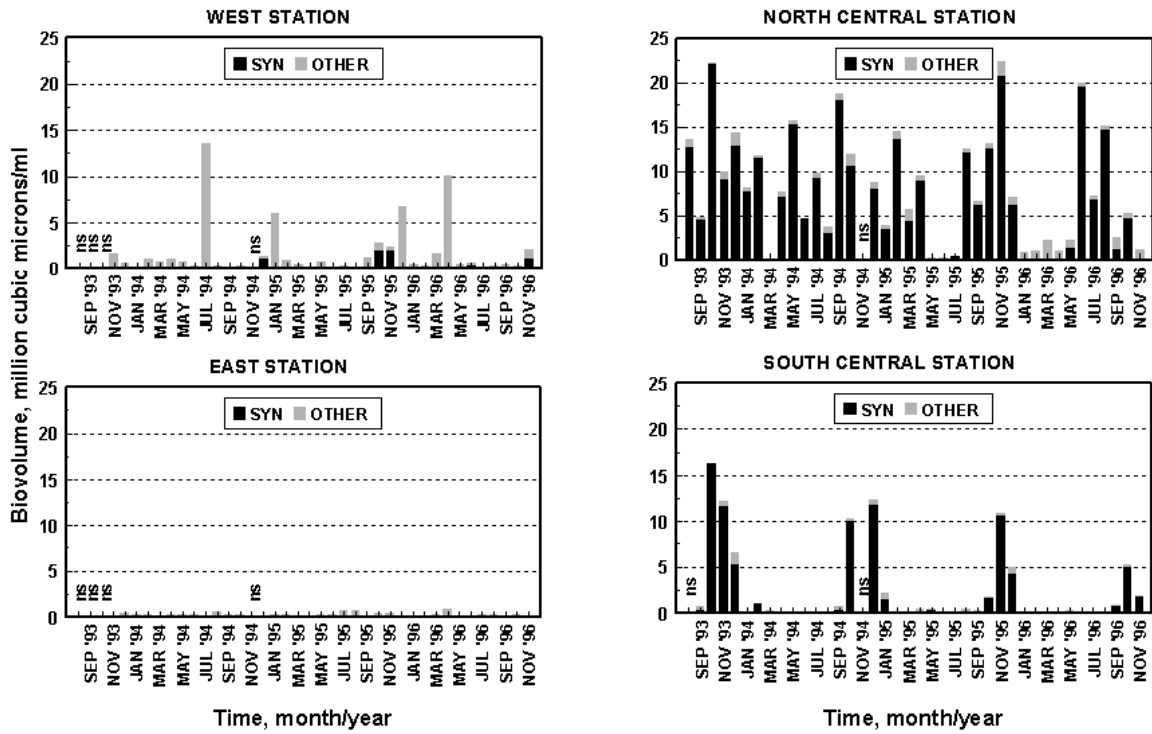


Figure 5. Phytoplankton biomass (million $\mu\text{m}^3 / \text{ml}$) in four distinct regions of Florida Bay. The dominant cyanobacteria, *Synechococcus* cf. *elongatus*, is indicated in black (Phlips et al.1999).

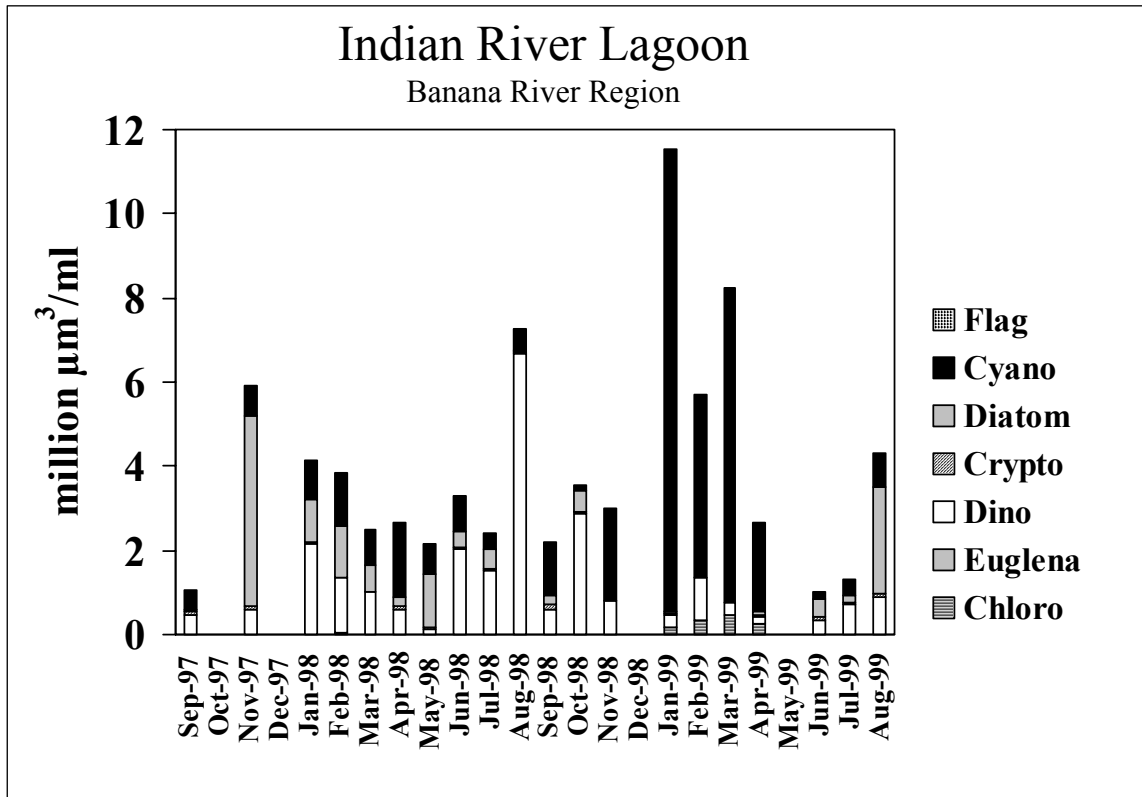


Figure 6. Phytoplankton biomass (million $\mu\text{m}^3 / \text{ml}$) in the Indian River Lagoon. Cyanobacteria (black) was dominated by *Synechococcus* sp. (Badylak and Philips 2002).

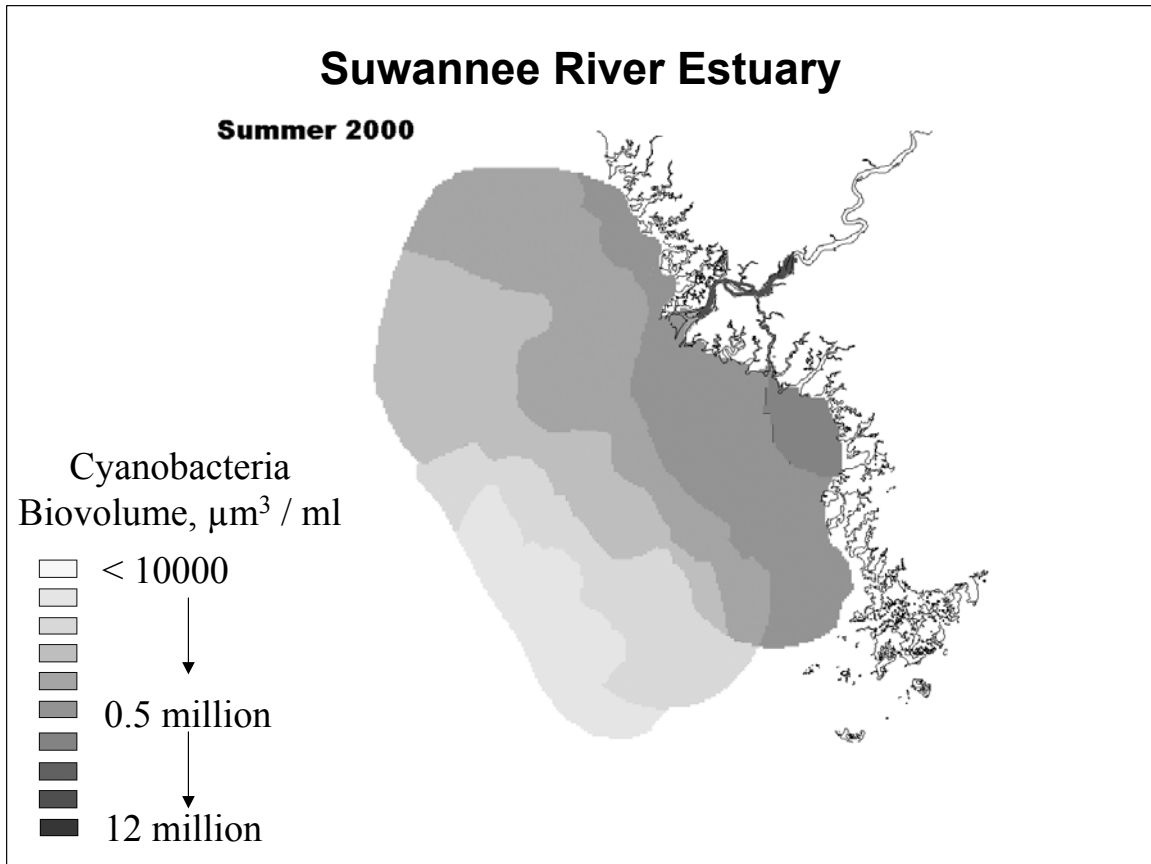


Figure 7. Picoplanktonic cyanobacteria biovolume ($\mu\text{m}^3 / \text{ml}$) distribution in the Suwannee River estuary during peak biomass observed during the summer of 2000. Darker shading indicates increased biomass (Bledsoe and Philips 2002).