

Lake Granbury and Lake Whitney Assessment Initiative

**Final Scientific/Technical Report
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For full Final Report & Appendices see <http://twri.tamu.edu/publications/reports> TR-392

3. Executive Summary:

A team of Texas AgriLife Research, Baylor University and University of Texas at Arlington researchers studied the biology and ecology of *Prymnesium parvum* (golden algae) in Texas lakes using a three-fold approach that involved system-wide monitoring, experimentation at the microcosm and mesocosm scales, and mathematical modeling. The following are conclusions, to date, regarding this organism's ecology and potential strategies for mitigation of blooms by this organism.

In-lake monitoring revealed that golden algae are present throughout the year, even in lakes where blooms do not occur. Compilation of field monitoring data with data collected by Texas Parks and Wildlife and Brazos River Authority (a period spanning a decade) revealed that inflow and salinity variables affect bloom formations. Thresholds for algae populations vary per lake, likely due to adaptations to local conditions, and also to variations in lake-basin morphometry, especially the presence of coves that may serve as hydraulic storage zones for *P. parvum* populations.

More specifically, the in-lake monitoring showed that the highly toxic bloom that occurred in Lake Granbury in the winter of 2006/2007 was eliminated by increased river inflow events. The bloom was flushed from the system. The lower salinities that resulted contributed to golden algae not blooming in the following years.

However, flushing is not an absolute requirement for bloom termination. In Lake Whitney, the highly toxic bloom that occurred that same winter was also stopped by high river inflow events. Flushing, however, did not terminate this bloom as the lake rose 10 feet, but no water was released from the dam at this time. It was the influx of nutrients that stopped toxin production. This, coupled with the high rates of photodegradation for *P. parvum* toxins (which we determined in laboratory experiments), allowed other phytoplankton to out-compete golden algae.

Laboratory experiments have shown that growth of golden algae can occur at salinities ~1-2 psu but only when temperatures are also low. This helps to explain why blooms are possible during winter months in Texas lakes.

In-lake experiments in Lake Whitney and Lake Waco, as well as our laboratory experiments, revealed that cyanobacteria, or some other bacteria capable of producing algicides, were able to prevent golden algae from blooming. Identification of this organism is a high priority as it may be a key to managing golden algae blooms.

Our numerical modeling results support the idea that cyanobacteria, through allelopathy, control the timing of golden algae blooms in Lake Granbury.

In-lake experiments in Lake Whitney and Lake Waco also revealed that as golden algae blooms develop, there are natural enemies (a species of rotifer, and a virus) that help slow the population growth. Again, better characterization of these organisms is a high priority as it may be key to managing golden algae blooms.

Laboratory and in-lake experiments and field monitoring have shown that nutrient additions will remove toxicity and prevent golden algae from blooming. In fact, other algae displace the golden algae after nutrient additions. Additions of ammonia are particularly effective, even at low doses (much lower than what is employed in fish hatchery ponds). Application of ammonia in limited areas of lakes, such as in coves, should be explored as a management option.

In addition, laboratory experiments and field monitoring also show that the potency of toxins produced by *P. parvum* is greatly reduced when water pH is lower, closer to neutral levels. Application of mild acid to limited areas of lakes (but not to a level where acidic conditions are created), such as in coves, should be explored as a management option.

Field monitoring and mathematical modeling revealed that flushing/dilution at high enough levels could prevent *P. parvum* from forming blooms and/or terminate existing blooms. This technique could work using deeper waters within a lake to flush the surface waters of limited areas of the same lakes, such as in coves and should be explored as a management option. In this way, water releases from upstream reservoirs would not be necessary and there would be no addition of nutrients in the lake.

Biomanipulation of *P. parvum*, i.e., through taxon-specific algal pathogens and/or grazing by toxin-resistant zooplankton, has promise and should be explored in future research.

Finally, our laboratory and in-lake experiments have also shown that additions of barely straw extract (useful for controlling some nuisance algae) have no effect on golden algae blooms and should not be employed as a management technique.

Figures, tables and a thorough description of project activities are detailed in the full text version of the Lake Granbury and Lake Whitney Assessment Initiative Technical/Final Report, TR-392. The full project Final Report and related Appendices can be found on the Texas Water Resources Institute (TWRI) technical report website at <http://twri.tamu.edu/publications/reports>.

4. Project Goals and Accomplishments:

Objectives:

This research expanded the scope and extended the duration of previous research supported by a FY06 federal earmark and Texas Parks and Wildlife Department. In the expanded project our overarching objectives were two-fold:

- Addressed the role of interactions between *P. parvum* and phytoplankton competitors through the mechanism of allelopathy as it relates to bloom initiation, persistence and termination.
- Focus on the continued development of a predictive numerical model where competitive interactions, life history, the physicochemical environment, and lake dimensions were more accurately depicted.

Accomplishments:

We accomplished these objectives by:

Interactions between P. parvum and plankton as it relates to bloom initiation, persistence and termination

1. Conducting system-wide, year-round sampling at monthly intervals in Lakes Granbury and Whitney where parameters measured included multiple characterizations of water quality and plankton community composition. This effort included high-resolution spatial mapping of these lakes.
2. Performing multivariate statistical analysis of field data.
3. Performing controlled laboratory experiments using cultured competitors and grazers common to Lakes Granbury and Whitney, as well as cultured *P. parvum*, and investigating the potential roles of allelopathy from cyanobacteria in *P. parvum* bloom formation, persistence and termination.

Continued development of a predictive numerical model

4. Building phytoplankton competitors into our existing numerical model where interactions between species through allelopathy were represented.
5. Performing controlled laboratory experiments focused on the role of mixotrophy in *P. parvum* bloom formation and termination.
6. Building *P. parvum* mixotrophy effects into the numerical model.
7. Expanding a *P. parvum* population dynamics “box” model into a 1-dimensional, spatially explicit model more representative of a reservoir environment, thereby enabling the continued study of the factors influencing *P. parvum* bloom demographics.

5. Summary of Project Activities:

5.1 System-wide, year-round sampling in Lakes Granbury and Whitney (1) and statistical analyses (2)

The field-monitoring component of this project was developed with the objective of characterizing *P. parvum* bloom dynamics. Lakes Granbury and Whitney, reservoirs constructed on the main stem of the Brazos River, have experienced recent toxic blooms of *P. parvum* that have resulted in massive fish kills and concerns about general water quality. Both lakes are critical to this region as being primary water supplies, sources of revenue and recreational areas.

This project addressed these water quality issues by providing critical information about the relationships between *P. parvum*, salinity, nutrients and other water quality and food web parameters. In Lakes Granbury and Whitney, plankton, nutrient and water quality samples were collected at fixed-location stations, and high-resolution spatial maps were generated, using an on-board dataflow technology, of various plankton and water quality parameters. Linkages between the toxic *P. parvum* blooms and environmental conditions were examined. All the data from monitoring of Lakes Granbury and Whitney are included in the Appendices.

Our data record for Lake Granbury spans September 2006 through May 2010. For Lake Whitney, our data record spans September 2008 through August 2009. The September 2008 through August 2009 dates correspond to the work performed as a part of this contract. For our analysis, we added information collected from other projects focused on these lake systems, as well as monitoring data made available to us from Texas Parks and Wildlife and Brazos River Authority. The larger data set spans 10 years, i.e., 2000 to 2009. We also added a third lake to this analysis, Lake Possum Kingdom, which is also a reservoir constructed on the main stem of the Brazos River.

Sampling of fixed stations

During each monitoring trip we sampled fixed-location stations, which enabled us to characterize the seasonal succession pattern of plankton communities (phytoplankton and zooplankton) and determine seasonal changes in various water quality parameters (inorganic nutrients, toxicity, pH, temperature, dissolved oxygen, Secchi depth). We sampled 20 stations in Lake Granbury and 10 stations in Lake Whitney.

While examining the phytoplankton samples for *P. parvum* cells, we observed other phytoplankton species present and the condition of cells. Specifically, we looked for the presence of other harmful algal blooms (HAB) species common to Texas lakes (e.g., multiple cyanobacteria that will include *Microcystis*, *Anabaena* and *Cylindrospermopsis*), for signs of algal pathogens (e.g., remains of lysed cells and presence of parasitic fungi), and took note of the dominant taxonomic groups present in each sample. Our sampling also included enumeration of zooplankton and bacteria. Since toxins produced by *P. parvum* under various physiological states are not fully understood, standards for measuring concentrations of toxins are not available at this time; toxicity can be estimated using other methods.

Samples were collected from Lakes Granbury and Whitney and transported to the laboratory where toxicity tests were initiated within 24 hrs. For each *P. promelas* toxicity test sample, three replicate chambers with 7 organisms per chamber were used to assess toxicity at each dilution level.

High-resolution spatial mapping

We measured spatial patterns of water quality in Lakes Granbury and Whitney at monthly intervals with Dataflow, a high-speed, flow-through measurement apparatus developed for mapping physicochemical parameters in shallow aquatic systems (Madden and Day 1992). We used this integrated instrument system to concurrently measure chlorophyll *a*, dissolved organic matter, transparency, salinity, and temperature from a boat running closely spaced transects. Measurements were taken at 4-second intervals from ~20 cm below the surface. An integrated GPS was used to simultaneously plot sample positions, allowing geo-referencing of all measurements for each variable.

GPS and dataflow information were used to create highly detailed contour maps of water quality parameters in relation to physiographic features. We also collected discrete water samples from the flow-through system during our continuous sampling for laboratory analysis of multiple parameters that were used to calibrate the dataflow unit. In addition to these samples, we measured profiles of water quality parameters that include dissolved oxygen and temperature to determine the degree of stratification of the water column.

High-resolution spatial maps are provided in Appendix E for Lake Granbury.

Other data collection

Daily discharges from the Brazos River were measured at the following upstream locations: South Bend, USGS Station Number 08088000 (Lake Possum Kingdom); Dennis, USGS Station Number 08090800 (Lake Granbury); and Glen Rose, USGS Station Number 08091000 (Lake Whitney). Salinities were measured during sampling using water quality multiprobes (Quanta, Hydrolab) and refractometers.

To better relate inflow magnitudes to the specific growth rate of *P. parvum*, we estimated daily system flushing rates during the time of peak flows. For this purpose, flushing rates of the system were estimated by dividing the daily inflow from the Brazos River by the lake volume.

Statistical analysis and simple model application

Correlations using linear-, exponential- and power-fit functions (Kaleidagraph, v.4.03), and multiple regression analysis (Matlab, v.7.5.0.338) were performed between *P. parvum* population density, inflow and salinity using the 10-year compiled data record of multiple lakes. We used correlation analyses to estimate the percent variability in *P. parvum* population density explained by either inflow or salinity (based on R^2), and the multiple regression analysis enabled us to simultaneously compare the relative roles of inflow and salinity as they affect *P. parvum* population density (based on the weighting coefficients). For inflows, we used 7-day, 10-day, 30-day and 365-day cumulative inflow

prior to each sampling date. The 7-day cumulative inflows showed the best relationships. So, only those results are reported here.

[Findings](#) (Figures can be found in the Appendices)

Results from multiple lake comparison over 10-year period (2000-2009)

Our data suggest that invading *P. parvum* established quickly in lakes downstream from early bloom events. For example, fish-killing blooms appeared sequentially down the watershed after they were first noticed in late 2000-early 2001. However, between 2004 and 2007, fish-killing blooms were concurrent, suggesting that immigration of *P. parvum* from upstream sources was no longer necessary for bloom initiation. The apparent rapid spread of *P. parvum* across the southern USA, and now with fish-killing blooms in northern areas (e.g., West Virginia and Pennsylvania, USA), also suggests that this species is an effective invader.

Our synopsis of the multiple lake comparison over a 10-year period, then, is that *P. parvum* blooms (and annual population maxima for the period after early-spring 2007) were recurrent winter phenomena in this area of the south-central USA. Bloom initiation and development only occurred at a time of year when inflows were low, and large fish-killing blooms occurred only when salinity was higher. Bloom termination followed high inflow events, likely through direct flushing of cells and indirect physiological effects. This linkage between incidence of *P. parvum* blooms, inflows and salinity raises concern because sequestration of water continues to increase in this area with rising human population. Combined with variations in precipitation and evaporation predicted from climate change, flows in this area may decrease by 60% (Cai and McCarl, 2009). Though not the focus of climate change models, it is likely that increased evaporation rates associated with regional warming will also result in higher salinity. Consequently, both human population increase and climate change may lead to an increased incidence of *P. parvum* blooms.

Statistical analysis and simple model application

Based on our observations of the system-wide, fish killing bloom, hydraulic flushing played an important role as a mechanism influencing *P. parvum* population dynamics. To better evaluate its relative impact, we estimated in-lake specific growth rates for *P. parvum* using a mathematical model. The model (eq. 1) was based on laboratory experiments using a strain of *P. parvum* isolated from Texas waters where salinity, temperature and light were varied (Baker et al. 2007, 2009), and was used previously to investigate *P. parvum* population dynamics (Grover et al. 2010). These equations were formulated under experimental conditions where inorganic nutrients did not limit *P. parvum* specific growth rate.

The calculated specific growth rate was used along with flushing losses to estimate the change in population density attributable to flushing during the period when the bloom was terminated using a simplified differential equation employed in previous studies (Roelke et al. 2003, Roelke and Eldridge 2008, 2009). Based on the dataflow maps for chlorophyll *a*, the length over which lake waters were well-mixed was determined to range between 1 and 6 km for the March and April samplings, respectively. By

employing average lake depth and width dimensions, the volume of well-mixed segments was estimated.

5.2 Microcystin ecological hazard assessment and laboratory experiments of microcystin-LR allelopathy to *P. parvum*

Recent studies on *P. parvum* in Texas reservoirs by our research team identified a possible allelopathic response of *P. parvum* to cyanobacterial allelochemicals (Grover et al., 2010; Roelke et al., 2010). Such observations provided the impetus for the present study in which we examined whether *P. parvum* growth can be inhibited by the cyanobacterial toxin microcystin-LR (MC-LR).

In the present study, we specifically examined the influence of MC-LR on *P. parvum* growth. To support selection of experimental treatment levels of MC-LR, a probabilistic distribution of environmental exposure was developed and compared to adverse response thresholds of other aquatic organisms. An environmental exposure distribution (EED), a probability distribution of reported concentrations of a compound measured in the environment, was developed for MC-LR following an extensive literature review. EEDs, a technique commonly employed in probabilistic ecological hazard and risk assessment (PEHA; Solomon et al., 2000; Dobbins et al., 2008, 2009), provides an approach to estimate the probability of observing a concentration of an environmental contaminant at or below a selected value, which can then be interpolated from the log-normal probability of aquatic exposure concentrations.

Probabilistic exposure and effects distributions

An extensive literature review was conducted to find reported environmental levels in surface waters for microcystins (Table 5.2.1) in order to develop a probabilistic EED. Measurements reported in the literature came from water bodies with microcystin-producers present or likely to be present; thus, there were few non-detect values. This EED represents a distribution of microcystin levels when microcystin producers are likely to occur.

Prymnesium parvum stock culture

A stock culture of *P. parvum* was initiated from a strain obtained from The University of Texas at Austin Culture Collection of Algae (strain UTEX LB 2797) that was originally isolated from the Colorado River in Texas. The culture was maintained in a 20-L glass carboy filled with 10 L of 2.4 psu Artificial Seawater (ASW) (Berges et al., 2001) enriched with f/2 levels of vitamins, trace metals, and nutrients (Guillard, 1975). The carboy was maintained in an incubator (VWR Model 2015, West Chester, PA, USA) at 20°C on a 12:12 light:dark cycle. The culture was swirled daily and allowed to grow until stationary phase, at which time these organisms were used to inoculate the experimental units at each treatment level. Cell density of the stock culture at the time of inoculation was approximately 2×10^5 cells mL⁻¹.

Experimental design

Experimental units consisted of five replicate culture tubes (25x200 mm) each containing 50 mL of test solution at one of six different nominal concentrations of MC-LR (0,

0.0623, 0.807, 13.9, 239, and 3090 $\mu\text{g L}^{-1}$, corresponding to the 10, 25, 50, 75, and 90th centiles, respectively). After all test solutions were made, each culture tube (except for blanks) was inoculated with 10,000 cells mL^{-1} of *P. parvum*.

After inserting foam plugs, all flasks were placed on slanted test tube racks in an incubator (VWR model 2015, West Chester, PA, USA) maintained at 20°C on a 12:12 L:D cycle for 27 days. Culture tubes were swirled once in the morning and gently vortexed in the afternoon before taking a fluorescence reading, after which their positions were rotated within the incubator.

Statistical analyses

Cell density of each treatment level was compared to the controls during each day of the study by a one way analysis of variance (ANOVA) and a Dunnett's test (JMP v6.1, SAS Institute, Cary, NC, USA) when a significant difference ($\alpha = 0.05$) was detected. In addition to determining cell densities, we also examined specific growth rates (μ , day^{-1}), which were calculated for days 2 through 7 for each replicate at each treatment level by regressing the natural log of the cell density against time.

Findings

Lake Waco, Texas has a high density of cyanobacteria, including species that produce microcystin-LR. In a recent microcosm study by Roelke et al. (2010), a dose-dependent decline of *P. parvum* cell density was observed when organisms were exposed to varying proportions of filtered Lake Waco and Lake Whitney water: the higher the proportion of Lake Waco water, the lower the growth of *P. parvum*. Based on these experimental observations, Roelke et al. (2010) concluded that grazers, salinity, nutrients, and anthropogenic contaminants were not responsible for such responses. However, Roelke et al. (2010) could not rule out toxins or other allelochemicals produced by cyanobacteria or bacteria. Though MC-LR was not quantified during their study, because MC-LR had been measured in Lake Waco in the past (Brooks, unpublished data) and species that produce MC-LR were abundant at the time of the Roelke et al. (2010) study, they proposed that allelochemicals produced by cyanobacteria may have inhibited growth of *P. parvum*.

Microcystin exposure distribution

The EED (Figure 5.2.1) included 211 concentrations of microcystins reported in the peer-reviewed literature (Table 5.2.1). Most values were available from water bodies in the United States, but data also came from Canada, Finland, Australia, and Japan. The highest reported value used to develop the distribution was 12,176 $\mu\text{g L}^{-1}$ microcystin from the Copco Reservoir in California, USA (Jacoby and Kann, 2007); 121 values were under 1 $\mu\text{g L}^{-1}$. A wide range of adverse acute invertebrate mortality values were identified from the literature (Table 5.2.2).

Table 5.2.1. Reported microcystin concentrations used to develop an environmental exposure distribution.

Reference	[MC] $\mu\text{g L}^{-1}$	Location	Reference	[MC] $\mu\text{g L}^{-1}$	Location	
McDermott, 1995	200	Wisconsin, US Suamico River	Graham and Jones, 2007	0.8	Missouri/ Iowa, US	
	180	Green Bay		7	Missouri/ Iowa, US	
	1.7	Green Bay	Fristachi <i>et al.</i> , 2007; Carmichael, 2001	0.007	US/ Canada	
	1.2	Quarry Park Lake		0.053	US/ Canada	
	0.7	Long Lake #1		0.013	US/ Canada	
	19	Long Lake #2		0.007	US/ Canada	
	0.3	Becker Lake		0.04	US/ Canada	
	3.8	Round Lake		147.1	US/ Canada	
	2.4	Boot Lake		4.7	US/ Canada	
	22	Beaver Dam Lake #2		0.03	US/ Canada	
	8	Beaver Dam access stream				
	2	Long Lake				
	6.8	Kettle Moraine Lake	Kotak and Zurawell, 2007	2.48	Canada Little Beaver Lake	
	0.5	Green Lake		11.2	Little Beaver Lake	
	17	Green Lake Dam		1.5	Nakamun Lake	
	50	Lake Weyauwega		1	Driedmeat Lake	
	12.4	Mirror Lake				
	7.4	Shadow Lake				
		11.2	White Lake	Murphy <i>et al.</i> , 2003	0.17	Canada Hamilton Harbor, Lake Ontario
		5	Hartman Lake		0.19	Hamilton Harbor, Lake Ontario
	1	Fox River-Omro Park	0.01		Hamilton Harbor, Lake Ontario	
	52	Lake Butte de Mortes	0.01		Hamilton Harbor, Lake Ontario	
	3	Wolf River	0.02		Hamilton Harbor, Lake Ontario	
	51	Lake Winnebago #1	0.06		Hamilton Harbor, Lake Ontario	
	56	Lake Winnebago #2	33.1		Hamilton Harbor, Lake Ontario	
			0.25		Hamilton Harbor, Lake Ontario	
			5.03		Hamilton Harbor, Lake Ontario	
			0.96		Hamilton Harbor, Lake Ontario	
Jacoby and Kann, 2007	12176	Pacific Northwest, US Copco Reservoir, CA		0.47	Hamilton Harbor, Lake Ontario	
	2032	Iron Gate Reservoir, CA		238.8	Hamilton Harbor, Lake Ontario	
	46.73	Klamath River, CA		202.2	Hamilton Harbor, Lake Ontario	
	0.7	Tenmile Lakes, OR		0.98	Hamilton Harbor, Lake Ontario	
	1.61	Tenmile Lakes, OR		0.24	Hamilton Harbor, Lake Ontario	
	0.19	Crane Prairie, OR	Murphy <i>et al.</i> , 2003	0.024	Lake Erie	
	4.92	Crane Prairie, OR		0.035	Lake Erie	
0.68	Lava Lake, OR	0.142		Lake Erie		
0.009	Paulina Lake, OR	0.302		Lake Erie		
84	Paulina Lake, OR	0.016		Lake Erie		
2.9	Wickiup Reservoir, OR	0.009		Lake Erie		
2.54	Diamond Lake, OR	0.008		Lake Erie		
0.36	Suttle Lake, OR	0.001		Lake Erie		
0.51	Suttle Lake, OR	0.006		Lake Erie		
0.19	Lake Selmac, OR	0.002		Lake Erie		
13.5	Lake Selmac, OR	0.371	Lake Erie			
0.03	Odell Lake, OR	0.407	Lake Erie			
5.01	Odell Lake, OR	0.2	Lake Erie			
10	Odell Lake, OR	0.028	Lake Erie			
600	Upper Klamath (Agency) Lake, OR	0.031	Lake Erie			
14.6	Owyhee Reservoir, OR	0.009	Lake Erie			
18.5	Vancouver Lake, WA	0.008	Lake Erie			
0.91	Vancouver Lake, WA	0.009	Lake Erie			
7	Vancouver Lake, WA					
9.5	Vancouver Lake, WA					
0.25	Lake Garrett (Hicks), WA	Young <i>et al.</i> , 2006			Aland, Finland	
0.85	Lake Garrett (Hicks), WA		0.070		Hogbolstad	
0.26	Lake Garrett (Hicks), WA		1.400		Godby trask	
0.75	Lake Garrett (Hicks), WA		12.100		Prasttrasket	
0.74	Lake Garrett (Hicks), WA		0.130		Basttjarnan	
0.68	Lake Garrett (Hicks), WA		0.260		Brantsbole trask	
0.86	Lake Garrett (Hicks), WA		6.400		Nato Hemviken	
1	Green Lake, WA		0.200		Gloskars trask	
32	Green Lake, WA		0.250		Gloskars trask	
3	Green Lake, WA		0.760		Brantsbole trask	
100	Green Lake, WA	0.620		Brantsbole trask		
0.17	Green Lake, WA	0.670		Brantsbole trask		
2.2	Green Lake, WA	1.000		Stromma trask		
1	Lake Sammamish, WA	0.700		Kaldersfjarden		
43	Lake Sammamish, WA	2.300		Lenbote Bytrask		
0.13	Lake Sammamish, WA	6.000		Lenbote Bytrask		
0.17	Lake Sammamish, WA	5.400		Vargata trask		

Table 5.2.1. Continued

Jacoby and Kann, 2007	0.16	Lake Sammamish, WA	Young <i>et al.</i> , 2006	4.600	Vargata trask
	0.1	Lake Sammamish, WA		5.100	Godby trask
	0.14	Lake Union, WA		0.100	Norra Langsjon, Saltvik
	0.15	Lake Union, WA		0.940	Overby insjo
	0.12	Lake Union, WA		0.240	Sodra Slemmern
	0.14	Lake Union, WA		0.610	Hogskar
	0.18	Lake Washington, WA		0.160	Lillfjarden
	0.62	Lake Washington, WA		1.640	Kathavet
	0.68	Lake Washington, WA		3.100	Prasttrasket
	1.15	Lake Washington, WA		0.440	Hagn trask
	4	Kitsap Lake, WA		42.300	Vargata trask
	13	Steilacoom Lake, WA		36.800	Vargata trask
	0.8	Waughop Lake, WA		7.540	Prasttrasket
	0.13	Moran State Park Lake, WA		3.100	Brantsbole trask
	0.5	Lake Cassidy, WA		0.210	Brantsbole trask
	3	Lake Cassidy, WA			
	0.5	Ketchum Lake, WA	Jones and Orr, 1994		Temora, Australia
	3	Ketchum Lake, WA		990	Lake Centenary
	2.73	Lake Lawrence, WA		1830	Lake Centenary
	0.99	McIntosh Lake, WA			
	0.54	McIntosh Lake, WA	Harada and Tsuji, 1998		Japan
	0.51	McIntosh Lake, WA		0.02	Lake Sagami
	1.52	McIntosh Lake, WA		0.04	Lake Sagami
	0.54	McIntosh Lake, WA		0.05	Lake Sagami
	0.48	McIntosh Lake, WA		0.6	Lake Sagami
				0.04	Lake Tsukui
				0.08	Lake Tsukui
				3.18	Lake Tsukui
				0.14	Lake Tsukui
				52	Lake Tsukui
				3.9	Lake Tsukui
				2.5	Lake Tsukui
				0.05	Lake Tsukui
				1.78	Lake Tsukui
				0.24	Lake Tsukui
				0.04	Lake Tsukui
				1.07	Lake Tsukui
	0.14	High Point Lake	Harada and Tsuji, 1998	93.8	Lake Tsukui
				0.08	Lake Tsukui
				0.02	Lake Tsukui
Williams <i>et al.</i> , 2007		Florida, US			
	0.05	Harris Chain of Lakes			
	0.5	Harris Chain of Lakes			
	3.6	Harris Chain of Lakes			
	0.01	St. Johns River			
	0.6	St. Johns River			
	31	St. Johns River			
	0.1	Lake Okeechobee			
	0.1	Lake Okeechobee			
	95	Lake Okeechobee			

Prymnesium parvum growth

From day 17 until the end of the study on day 27, *P. parvum* density in the lowest MC-LR treatment level ($0.09 \mu\text{g L}^{-1}$) significantly increased *P. parvum* cell density ($p < 0.05$) relative to controls (with day 24 as an exception; Figure 5.2.2). However, on day 6-8, 13-14, and 22-23 (approximately one quarter of the duration of the study), the highest MC-LR treatment level ($4,392.8 \mu\text{g L}^{-1}$) significantly decreased *P. parvum* cell density relative to controls ($p < 0.05$; Figure 5.2.2). There was a significant ($p < 0.05$) decrease in cell density by the $16.6 \mu\text{g L}^{-1}$ treatment on day 13 only. Cell densities in all other treatments were not significantly different from the controls throughout the study period ($p > 0.05$). Specific growth rate was similar in all treatments for most of the test duration. Specific growth rate was the highest in the first five days, decreased until around 15 days, then leveled off through day 27 (Figure 5.2.3). Analysis of exponential growth rate (Table 5.2.5; Figure 5.2.3) from days two to seven showed that there was no significant difference of exponential growth rate among the treatment levels ($p > 0.05$).

6. Product Descriptions:

a. Publications/presentations:

Grover, J.P., Crane, K.W., Baker, J.W., Brooks, B.W., and Roelke, D.L. (2010). Spatial variation of harmful algae and their toxins in flowing-water habitats: a theoretical exploration. *Journal of Plankton Research*. In Press.

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- b. **Websites:** Information from this project, as well as other projects in the watershed, is available online at: <http://lakegranbury.tamu.edu>. A general project description, a list of collaborators, funding agencies, news stories, technical reports and quarterly progress reports are all available at this site.
- c. **Networks or collaborations fostered:** Thru this and previous projects conducted in the Lake Granbury Watershed, continued collaboration and support has been received from the following groups:
 - Texas Water Resources Institute
 - Texas AgriLife Research
 - Texas AgriLife Extension Service
 - Texas Commission on Environmental Quality
 - Brazos River Authority
 - USDA Natural Resources Conservation Service
 - Texas Parks and Wildlife Department
 - Baylor University
 - University of Texas at Arlington
 - Hood County, Texas
- d. **Technologies/Techniques:** N/A
- e. **Inventions/Patent Applications:** N/A
- f. **Other Products:** N/A

7. Computer Modeling:

Building phytoplankton competitors into numerical model

- a. Model description, key assumptions, version, source, and intended use
A prior grant from DOE supported the construction and initial parameterization and calibration of a suite of models simulating *P. parvum* dynamics. These models were designed to be forced with limnological observations from reservoirs in Texas where *P. parvum* blooms occur, and appropriate data from Lake Granbury were used as input data in model development and testing. Although development of these models was largely completed under a prior grant, the current grant supported preparation of a manuscript reporting on these models (Grover et al., 2010).

During the current grant some additional modifications were made to the one-competitor model with allelopathy. A dynamically coupled zooplankton population was added, grazing on both *P. parvum* and the cyanobacteria. The grazing of this zooplankton population on *P. parvum* was inhibited by a toxin excreted by *P. parvum*. This section of the full final report details the development and calibration of this extended model of *P. parvum* and competing algae. The calibration uses the first year of data that our team obtained from Lake Granbury, which included a large bloom. We also report an initial test of validity done by attempting to predict the dynamics of *P. parvum* subsequent to our first year of data, using the necessary input data on limnological conditions over that time period.

- b. Performance criteria for the model related to the intended use
Model PP1 from Grover et al. (2010) was modified to construct a basic model with *P. parvum*, a competitor population (cyanobacteria) and a grazer population.

In general, all parameters carried over from the suite of models in Grover et al. (2010) were assigned the default (uncalibrated) values from that paper. The modified models are formulated in terms of quotas rather than yield coefficients, and the initial quota values were taken as reciprocals of the previous yields. In the JAWRA paper, models with cyanotoxin assumed a value of 1.0 for the exponent ξ governing inhibition of *P. parvum* growth, so that is the initial value here.

- c. Test results to demonstrate the model performance criteria were met (e.g. code verification/validation, sensitivity analyses, history matching with lab or field data, as appropriate)
With the initially assigned parameters, the model was run using one year of limnological observations from Lake Granbury as input data (Grover et al. 2010) until the predicted dynamics stabilized to an annual cycle. In this model, both cyanobacteria and *P. parvum* produce toxins. The cyanotoxins are allelopathic and reduce the growth rate of *P. parvum*; the *P. parvum* toxins inhibit the ingestion and growth rates of zooplankton. Dominance of cyanobacteria is predicted, with no blooms of *P. parvum* (Fig. 5.3.1). Cyanotoxin concentrations

are predicted to be much higher than those of *P. parvum* toxins. Accordingly, allelopathic effects on *P. parvum* are predicted to be strong, but the inhibition of grazers by *P. parvum* toxins is predicted to be weak.

d. **Theoretical framework of the model**

Population dynamics of *P. parvum* and other, competing algae are modeled as functions of physical factors such as temperature, irradiance, and salinity, and as functions of dissolved nitrate and phosphate concentration. Dynamics of these nutrients are coupled to population growth, so that large populations deplete nutrients leading to competition within and between populations. All algal populations suffer losses due to flow (washout from the habitat), grazing by zooplankton, and sinking in the case of diatoms. Some models also assume that a cyanobacterial population, competing with *P. parvum*, produces a toxin that reduces the growth rate of *P. parvum*.

e. **Mathematics to be used, including formulas and calculation methods**

These are fully summarized and listed in the full final report and appendices. Using laboratory cultures, Baker et al. (2007, 2009) determined that *P. parvum* displays rapid growth at relatively warm temperatures. This property was incorporated into mathematical models for *P. parvum* growth in Texas (Grover et al 2010). These models were based on conventional approaches for modeling in water quality studies (Chapra 1997), and designed to predict algal dynamics given input data on commonly measured limnological conditions. The models included up to five algal populations with two limiting resources, and allelopathic effects. The model consists of three ordinary differential equations. The model tracks the densities of *P. parvum* and their edible bacterial prey along with the concentration of a limiting inorganic resource here taken to be phosphorus.

Though this model was written with the idea that values from the input array would change every day for time-variable simulations, the first analyses addressed steady states using average values from the input array over one year as constant parameters. Solutions for the steady state were found both numerically and analytically. The analytic solutions were calculated using Mathematica's Solve function while the numeric solutions were found using MATLAB's numerical solvers for ordinary differential equations. Results from MATLAB's ode45 solver indicated convergence to a steady state to within 4 or 5 decimal places. Analytical solutions and numerical integration to achieve steady state agree well (Table 5.4.3).

Table 4.3. Analytical and numerical steady state solutions.

Variable	Analytical	Numerical
<i>N</i> (cells / liter)	1.67617×10^{10}	1.676174×10^{10}
<i>B</i> (cells / liter)	1.17543×10^6	1.175×10^6
<i>R</i> (μmol P / liter)	0.00312755	0.00312

f. **Strengths and weaknesses of the mathematical algorithms**

Possibly, a more sophisticated description of mixotrophy would prove more helpful. Alternatively, other processes may be more influential on bloom formation than the growth physiology of *P. parvum*. There has been some success with mathematical modeling that stresses spatial dynamics and hydrology as factors in bloom formation (Grover et al. 2010), and field observations support a strong role for hydrology (Roelke et al. 2010a, b). There has also been some success with models that include zooplankton grazing on *P. parvum* that is inhibited by toxin production (unpublished work included later in this report).

g. **Hardware requirements**

All models were written in Fortran 77 and compiled with the Digital Visual Fortran compiler, version 6. Numerical work was done on several IBM PC type computers with Pentium 4 or higher processors. However, the program code is portable to a wide range of hardware for which Fortran compilers are available. The programs use only simple ASCII text files for input and output and thus can be used on a wide range of hardware enabling use of this file format.

h. **Documentation (e.g. users guide, model code)**

These can be found in the full project Final Report and related Appendices on the TWRI technical report website at <http://twri.tamu.edu/publications/reports> (TR-392).

Expanding “box” model into a 1-dimensional model

a. **Model description, key assumptions, version, source, and intended use**

A one-dimensional model was constructed to study spatial variations of harmful algae such as *P. parvum* and their toxins, in riverine reservoirs. Inland waters present a spectrum of flowing-water habitats that are inhabited to varying degrees by planktonic algae. In small, low-order streams with rapid currents, no true plankton develops and suspended algae arise transiently from the benthos. As order increases, and especially in broad rivers and riverine reservoirs, development of plankton becomes more likely. Nevertheless, the ecology of planktonic algae in flow-dominated systems has been somewhat neglected, and raises many paradoxes (Reynolds, 1990).

This study addressed two basic questions: What is the longitudinal distribution of algal abundance and toxicity under various flows, in a riverine reservoir? And what differences arise between a fringing cove and a main lake?

In this study, steady state analyses of the models constructed were complemented by examining event-driven dynamics using documented, extreme flow events from a riverine reservoir to force model output. Although the model structures are idealized, they support elaboration with details necessary for modeling specific systems, and although parameters are based on riverine reservoirs in Texas, USA, where harmful blooms have occurred, they could readily be changed to represent conditions found in other riverine systems worldwide.

b. Performance criteria for the model related to the intended use

Extensive sensitivity analyses of the riverine reservoir model examined how parameter variations influenced the spatial patterns predicted at steady state.

c. Test results to demonstrate the model performance criteria were met (e.g. code verification/validation, sensitivity analyses, history matching with lab or field data, as appropriate)

In all variable flow simulations, algal abundance and toxin concentrations were predicted to be similar in the main channel and storage zone at most times (e.g. Figs. 5.5.7 – 5.5.10). However, during high flows approaching or exceeding those producing washout, large transient differences were predicted in upstream reaches, with the storage zone having several-fold larger algal abundance and toxin concentration than the main channel (e.g. Fig. 5.5.11).

Under most conditions explored, the cove-main lake model also predicted that a fringing cove would have similar algal abundance and toxin concentration to the main lake. Exceptions occurred on a transient basis in response to high flow, but only for coves that were relatively isolated from the main lake (with exchange rates \leq about 0.01 d^{-1}), receiving much higher nutrient supply (e.g. Fig. 5.5.12). The cove-main lake model was also less susceptible than the riverine reservoir model to washout and prolonged low algal populations during high flow events (e.g. compare Fig. 5.5.12 to Fig. 5.5.7).

d. Theoretical framework of the model

The occurrence of harmful algal blooms in riverine ecosystems demonstrates a viable phytoplankton community. In addition to a basic question of persistence in the presence of flow, other questions arise concerning the spatial variation of algal abundance and toxicity during bloom and flow events. Manipulation of flow is possible in some river systems, and has been suggested as a potential technique for managing and mitigating harmful algal blooms (Maier *et al.*, 2001; Roelke *et al.*, 2010a; Mitrovic *et al.*, 2010). This possibility motivates the theoretical exploration of harmful algal dynamics in flowing conditions undertaken here. Another motivation lies in the fact that coves along the shoreline of riverine reservoirs might represent habitats where dynamics of harmful algae differ from the main reservoir. Such differences could perhaps also be exploited to mitigate harmful blooms and their effects.

e. Mathematics to be used, including formulas and calculation methods

All equations can be found in the full project Final Report and related Appendices on the TWRI technical report website at <http://twri.tamu.edu/publications/reports> (TR-392).. For both modeling approaches, the same sets of assumptions were applied for algal population dynamics, and toxin production and decay. Three modes of production for dissolved toxins were explored.

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