

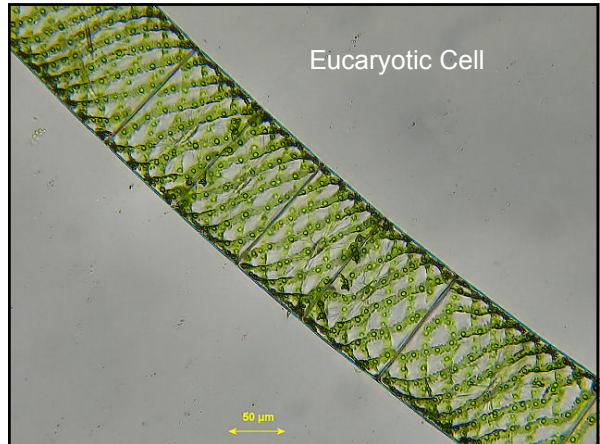


Taxonomy, Ecology and Control of Algae

2006 Aquatic Weed Control Short Course

John H. Rodgers, Jr.

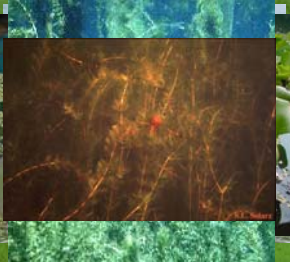
Environmental Toxicology Program
Department of Forestry and Natural Resources
Clemson University

Cyanobacterial Cell

- No membrane-bound organelles.
- Cellular processes:
 - Photosynthesis
 - Respiration
 - Electron transport systems

Why do we have problems?




- Invasive and exotic species move at unprecedented rates.
- We have changed the landscape – e.g. canals, reservoirs, stormwater detention basins, etc.
- Human population increase – algae / plant - people interface.
- Changing climate – globally
- Pressure on water resources.

Problem Definition

Do you have a problem?

Problems Caused by Vascular and Nonvascular (Algae) Plants

- Aesthetics
- Devalue property
- Disrupt transportation
- Taste and odor problems
- Impact fisheries and endangered species
- Impede irrigation
- Human health
- Interfere with water resource usages!



Problem or Not?

- **Aesthetics**
(property value, tourism)
- **Alter Water Characteristics**
(increase pH, decrease DO)
- **Taste and Odor Problems**
(MIB, geosmin)
- **Hinder Recreational Activities**
(swimming, fishing)
- **Toxin Production**
(neurotoxins, hepatotoxins)



Solutions for the problem

- **Risk assessment – problem or not?**
- No decision / action vs. decision / action
- Consider all available options
- Implement viable option(s)
- Monitor results
- Modify approach if indicated

Options for Remediation

- **Physical**
-dyes, aeration, precipitation
- **Mechanical**
-rakes, filters
- **Biological**
-grass carp, filter feeding bivalves
- **Chemical**
-Endothall, Diquat dibromide, Peroxide algaecides, Acrolein, Cu algaecides



IPM Approach Integrated Pest Management

- 1) **Environmentally sound**
 - Cure should not be worse than the disease
- 2) **Economically viable**
 - Solution should not cost more than the problem

Consider all available options

- Biological
 - Physical
 - Mechanical
 - Chemical
 - Combinations
- Algal Problem*





The Villains - Planktonic

- *Premnesium*
- *Microcystis*
- *Anabaena*
- *Cylindrospermopsis*
- *Hydrodictyon*
- *Anacystis*
- *Asterionella*, *Tabellaria*, *Synedra*

The Benthic Villains

- *Lyngbya*
- *Oscillatoria*
- *Oedogonium*
- *Spirogyra*
- *Pithophora*
- *Nitellopsis*

What is the Golden Alga?

- *Prymnesium parvum*
- Microscopic yellow-green alga
- Mixotrophic
- Produces toxins

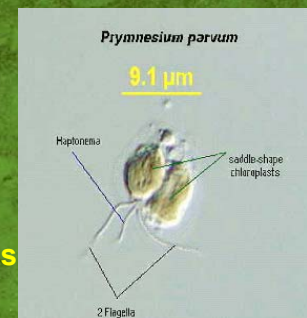
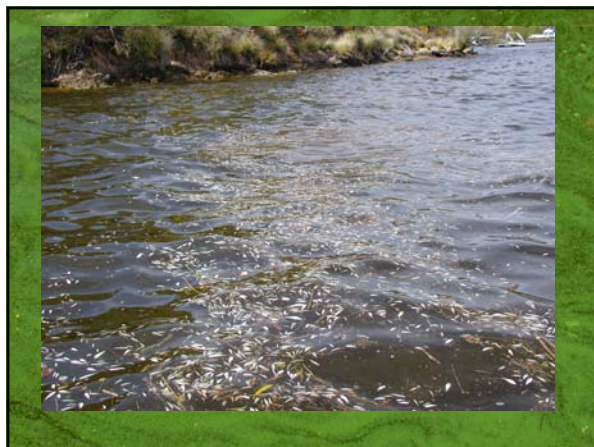


Photo by Greg Southard 2004



Prymnesium parvum

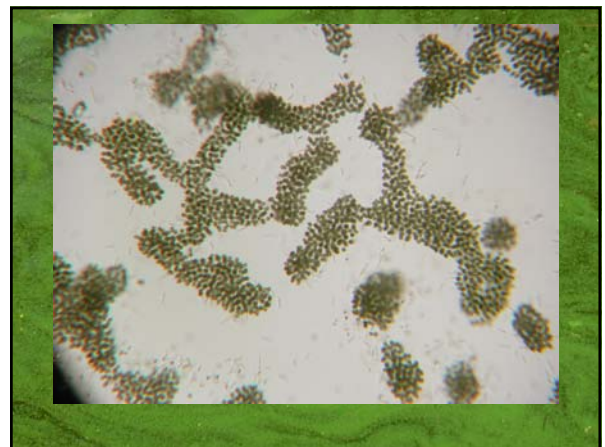
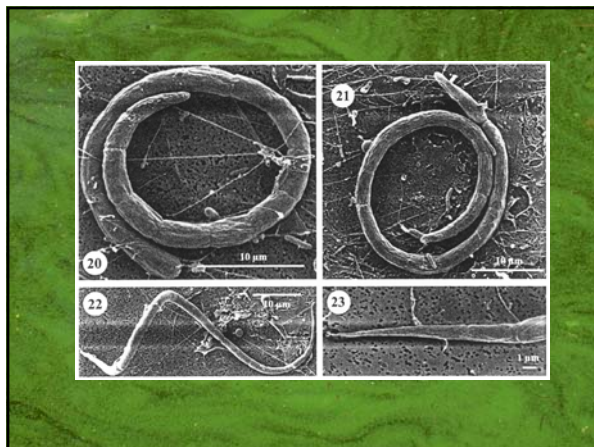
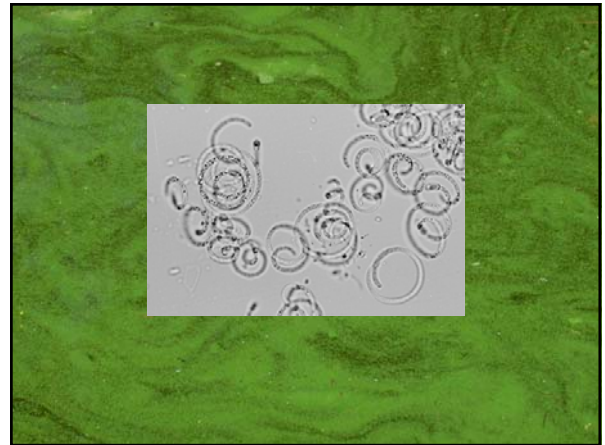
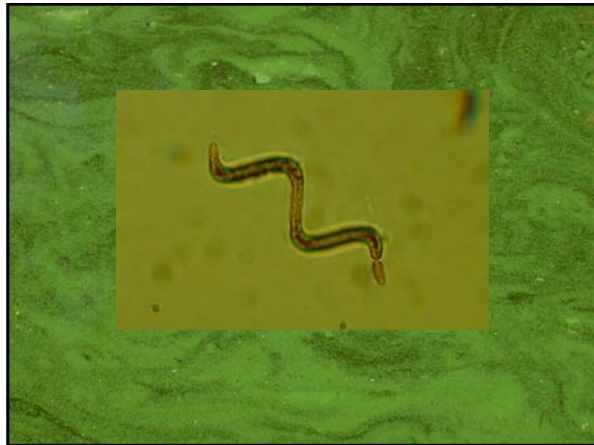
- Golden Brown Algae
- May produce a toxin
- Hemorrhaging gill filaments
- Causes mucus and blood to be present in fins of fish

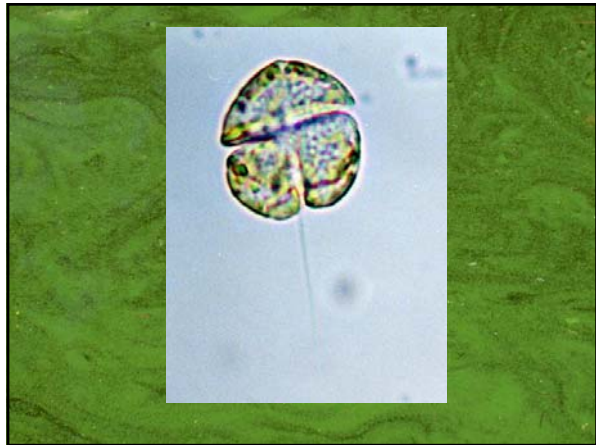
Photo: Dr. Carmelo Tomas UNC Wilmington

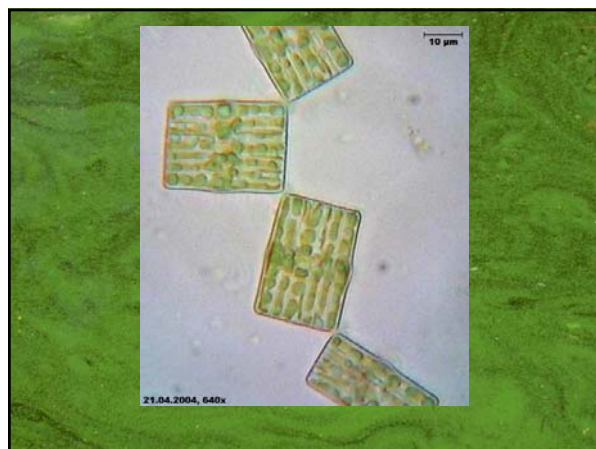
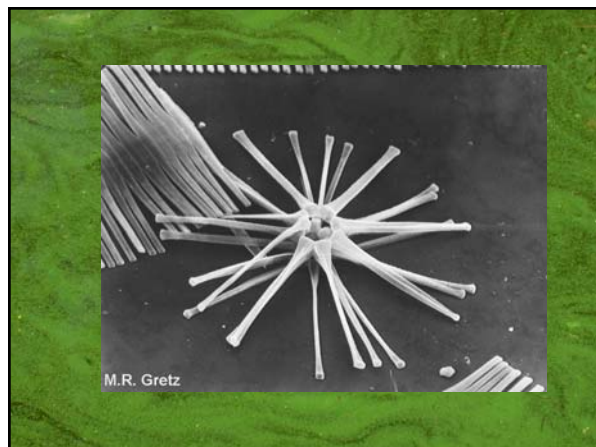
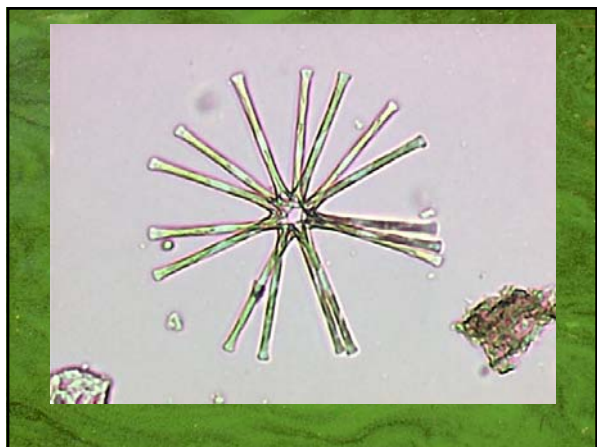


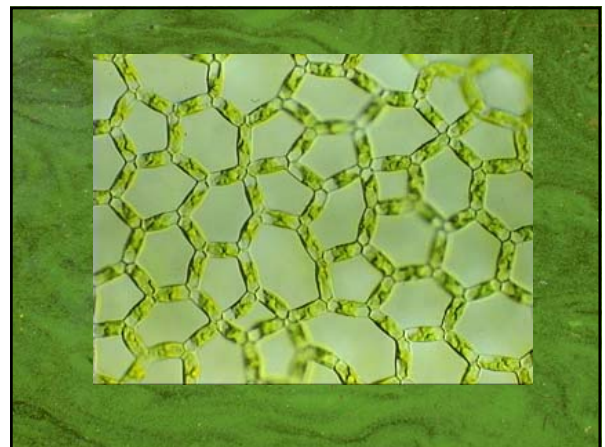
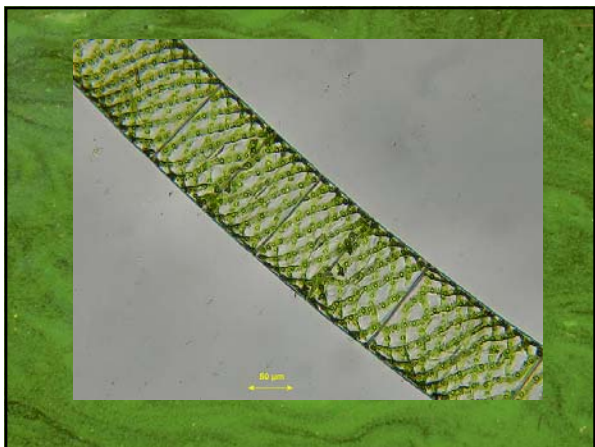
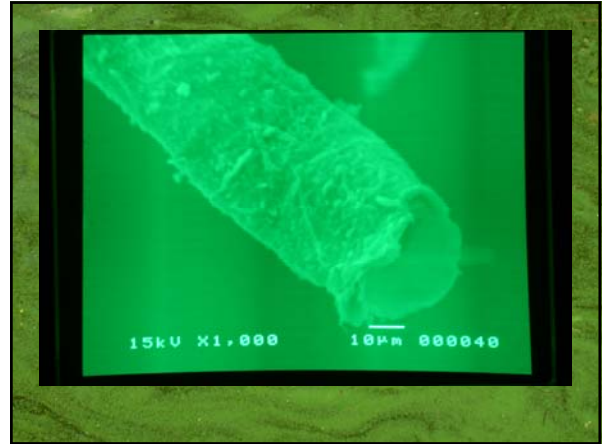
Photo: Dave Buzan, TPWD Possum Kingdom Reservoir

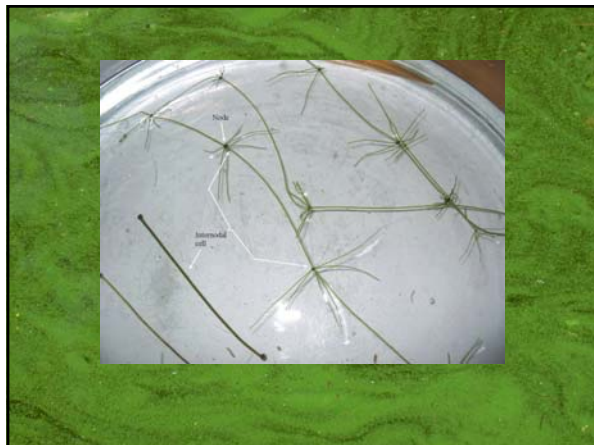
Does inventorying dead fish "fix" the problem?












Starry Stonewort

May have a whitish bulb at the base of each cluster

Green gelatinous stems

Listed as an invasive aquatic species in New York

Algae looks like an Aquatic vascular Plant



Rock Snot "Didymo"
Didymosphenia geminate

- Invasive diatom species
- Thrives in nutrient poor water
- Found on the bottoms of streams and rivers where it attaches itself by stalks
- Can cover 90% of stream bottom-South Dakota
- Affects the food chain and fish populations
- Tennessee anglers are encouraged to report sightings and properly clean all boats and equipment
- The alga has been found in Arkansas, Montana, South Dakota, Tennessee and British Columbia in North America




Lyngbya wollei






Prymnesium parvum

Lake Whitney (TX)




January to April 2003
 Fish Killed: 1,475,212
 Direct Cost: \$518,135

Photo: Dave Buzan, TPWD

Prymnesium parvum




March- April 2005
 Fish Killed: >1,000

Photo: Eric Swanson, AZ Game and Fish Department

MONITORING PLAN

- **Location:** Various- to cover the entire system.
- **Frequency:** Observe conditions and collect samples weekly.
- **Preservation:** Keep samples cool and analyze quickly. Do not preserve.
- **Results:** Report immediately.
- **Go to Action plan** if *P. parvum* is present.

Site Monitoring

hemorrhaging in and around gills
 necropsy to eliminate other causes







Options for Remediation

- **Physical**
-dyes, aeration, precipitation
- **Mechanical**
-rakes and filters
- **Biological**
-grass carp, filter feeding bivalves
- **Chemical**
-Endothall, Diquat dibromide, Peroxide algaecides, Acrolein, **Cu algaecides**

Experimental Objectives

- To determine the most efficient and effective [Cu] using Cutrine®-Plus to control *Prymnesium parvum* in two site-waters: Lake Whitney, Texas and Water Ranch Lake, Arizona.
- To compare responses of *Prymnesium parvum* from two locations to exposures of a copper-containing algaecide (Cutrine®-Plus).
- To measure the response of *Pimephales promelas* to concentrations of Cutrine®-Plus that control growths of *Prymnesium parvum*.

Prymnesium Toxicity Experiment

1. Test type:	Static, non-renewal
2. Temperature (°C):	23 ± 2 °C
3. Light quality:	"Cool white" fluorescent lighting
4. Light intensity:	86 ± 8.6 µE/m ² /s, or 400 ± 40 (ft-c)
5. Photoperiod:	16:8, light:dark
6. Size of test vessel:	250 ml Erlenmeyer flask
7. Genus and Species of Algae:	<i>Prymnesium parvum</i>
8. Treatments	0.2, 0.4, 0.6, 0.8, 1.0 mg / L of Cu as Cutrine®- Plus

Prymnesium Toxicity Experiment (Continued)

9. No. of replicate test vessels per concentration or dilution:	4 replicates
10. Algal Mixing:	Swirling once daily by hand
11. Aeration:	None, unless DO concentration falls below 40% of saturation, at which time start gentle, single-bubble, aeration
12. Test duration:	96 hours Observations daily
13. Endpoints measured:	Cell counts: microscope counting using a hemacytometer (APHA, 1995) and chlorophyll content: measuring chlorophyll a using a spectrophotometer (APHA 1995)

Cutrine®-Plus Algaecide

- **Copper – based algaecide, Aquatic herbicide (chelated)**
- **9 % elemental copper**
- **Manufactured by Applied Biochemists**
- **Maximum label limit 1 mg Cu / L**

Applied Biochemists
<http://www.appliedbiochemists.com>

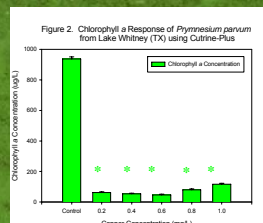
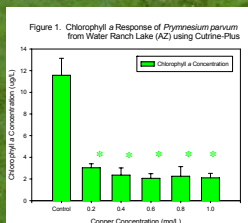
Analysis of Data

- $H_0 = 0.2-0.4-0.6-0.8-1.0$ mg Cu / L
- [Cu]s required to control *Prymnesium parvum* (EC_{100}^b) were determined.
- ANOVA and Regression Analysis were used to determine concentrations which were statistically significant.
- Water chemistry parameters such as DO, temperature, conductivity, pH, hardness, and alkalinity were measured and recorded throughout experiments according to Standard Methods (APHA 1995).

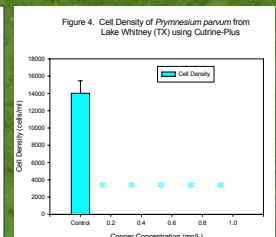
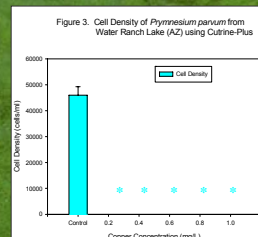
Site Water Characteristics

Problem algal species	Water Source	pH (avg.)	Hardness (avg.)	Alkalinity (avg.)	Conductivity (avg.)
Scientific name	Site	SU	mg / L as CaCO ₃	mg / L as CaCO ₃	µS / cm ²
<i>Prymnesium parvum</i>	Lake Whitney (Clifton, TX)	8.27	312	130	1604
<i>Prymnesium parvum</i>	Water Ranch Lake (Gilbert, AZ)	7.75	411	104	2700

Chlorophyll a Response of *Prymnesium parvum* from Arizona and Texas after Cutrine®-Plus Exposures



Cell Density of *Prymnesium parvum* from Arizona and Texas after Cutrine®-Plus Exposures



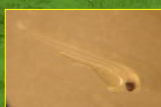
Margin of Safety (MOS)

Scientific name	Algaecide	96 h LOEC ^a <i>Pimephales promelas</i> (mg / L)	EC_{100}^b <i>Prymnesium parvum</i> (mg / L)	MOS ^c = LOEC of <i>Pimephales promelas</i> / [Cu] required to control <i>Prymnesium parvum</i> (mg / L)
<i>Pimephales promelas</i>	Cutrine®-Plus	0.750	0.2	3.75

^a Lowest Observed Effect Concentration (Murray-Gulde et. al, 2002)

^b [Cu] used to control *Prymnesium parvum* (EC_{100})

^c Margin of Safety (MOS)



Observations and Conclusions

- *Prymnesium parvum* produces a neurotoxin that has killed millions of fish in Arizona and Texas waters.
- *P. parvum* was controlled by a short-term (2 - 4 d) exposure of 0.2 mg Cu / L as Cutrine®-Plus in waters from both Arizona and Texas. Arizona site algaecide treatment recommendations were based on data presented in this presentation. Field applications have been successful in Arizona with no recurrence of *P. parvum*, and no fish mortality after several months.

Observations and Conclusions (Continued)

- A Margin of Safety (3.75) was calculated using EC_{100} for *P. parvum* at each site and the LOEC for *Pimephales promelas* using Cutrine®- Plus. Since the MOS is greater than 1, there is a margin of safety associated with using Cutrine®-Plus to control *P. parvum*.

Do Algae Spill Their Guts After Algaecide Treatment?: A Test of the Leaky Cell Hypothesis

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"Leaky Cell" Hypothesis

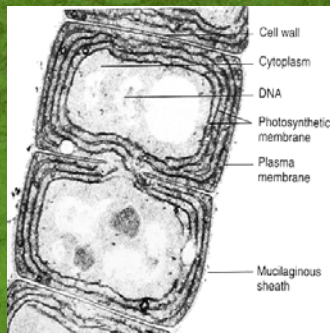
- Following exposure to algaecides or water treatment chemicals, algae (cyanobacteria) release intracellular contents.
 - Coptrol®.
 - Microcystin(s) (Jones and Orr 1994).
 - Chlorine, $KMnO_4$, $AlSO_4$, H_2O_2 .
 - MIB, GSM, DOC (Peterson et al. 1995).
- Appropriate consideration for action / no action?
- Appropriate consideration for no algaecide decision?

"Leaky Cell" History

- Algae release intracellular contents following exposure.
 - Algaecide (Coptrol®); water treatment chemicals (i.e. Chlorine, $KMnO_4$, $AlSO_4$, H_2O_2).
- Release following exposure not universal(?); however universally applied.
 - Lyngbya, *Cylindrospermopsis*,
***Microcystis*.

Sites of Potential Toxicity: Cyanobacterial Cell

- Sites other than cell membrane.
- Cellular processes:
 - Photosynthesis
 - Respiration
 - Electron transport systems



Specific Problem: Pawnee Reservoir, NE



- Re-occurring *Microcystis aeruginosa* "blooms".
 - Toxin (microcystin) producer / releaser
 - Toxic to some mammals / some fish low concentrations ($\leq 1 \mu g / L$).
 - Aqueous concentrations $\geq 10 \mu g / L$.
 - Risks.
 - Affects recreation, fishing, aesthetics.

Pawnee Reservoir Site Characteristics

- Approx. 300 ha. (~750 acres)
- >200 campgrounds / recreation areas.
- Primary uses:
 - Contact recreation:
 - boating, skiing, fishing.
 - Agricultural water supply.



Problems Caused by *Microcystis*

- Alter water characteristics.
- Alter aesthetic properties.
- Affect recreational activities.



Problems Caused by *Microcystis*

- Displace native / beneficial aquatic organisms.
- Toxin production / release.
 - *Microcystis aeruginosa*.
 - Microcystin(s).
 - Hepatotoxin(s).
 - Toxic to mammals, fish in low aqueous concentrations ($\leq 1 \mu\text{g} / \text{L}$).

Potential Solution: Pawnee Reservoir, NE

- Risk assessment:
 - Consider all available options.
 - Chemical control tactic.
 - Cu algacide.
 - Cost.
 - Application time.
 - Response time.



Experimental Objectives

- Determine effective [Cu] to control *M. aeruginosa*.
 - Site water; Pawnee Reservoir, NE.
- Measure responses in terms of chl *a* and cell density.
- Measure responses in terms of microcystin concentrations.
 - Pre- and post- exposure.

Exposure(s) / Response(s)

• Exposure(s):



• Response(s):



Algaecides

	Citrine®- Ultra	Cleargate®	Algimycin® PWF	Copper Sulfate Pentahydrate
% Cu as elemental	9.0	3.8	5.0	25.4
Formulation	Copper-Triethanolamine Complexes and D-limonene	Copper-Ethanolamine and D-limonene	Chelates of Copper Citrate and Copper Gluconate	CuSO ₄ ·5H ₂ O
Chemical class	Chelated Elemental Copper	Chelated Elemental Copper (Cu ₂ CO ₃)	Weakly Chelated Copper	Copper salt
Appearance	Blue Viscous Liquid	Blue Viscous Liquid	Blue Viscous Liquid	Blue Crystalline
Odor	Orange	Orange	Slight	Odorless
Water Solubility (mg/L)	Miscible	Miscible	Complete	316,000
pH	9.5-10.0	9.5-10.0	1.5-2.0	NA

Exposure Concentrations

Exposure Concentration (ppb Cu)	Algaecide			
	Citrine®-Ultra	Cleargate®	Algimycin® PWF	CuSO ₄ x 5H ₂ O
Initial (Background)	23	49	49	49
200	212	210	289	243
400	440	485	472	456
600	673	736	674	700
800	901	981	962	873
1000	1090	1160	1137	1148

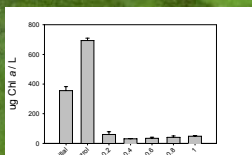
Water Characteristics

Water Characteristic Parameter	Range for Algaecide Exposures
pH (SU)	7.7-9.7
D.O. (mg O ₂ / L)	4.6-11.7
Alkalinity (mg CaCO ₃ / L)	136-200
Hardness (mg CaCO ₃ / L)	96-200
Conductivity (µS / cm ²)	418-513

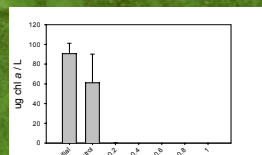
Statistical Analyses

- One-way ANOVA procedure
 - Chlorophyll a concentrations
 - Algal cell densities
 - Aqueous microcystin concentrations
- Differences identified were discerned using a multiple range test.

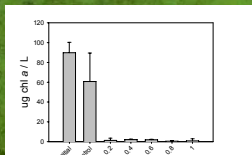
Results: Chl a Concentrations



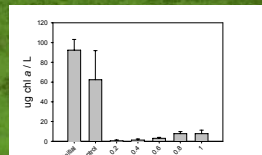
Treatment Concentration (mg Cu / L as Citrine-Ultra)



Treatment Concentration (mg Cu / L as Cleargate)

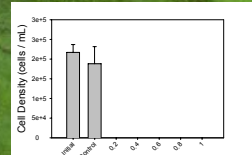


Treatment Concentration (mg Cu / L as Algimycin PWF)

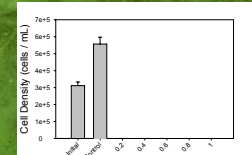


Treatment Concentration (mg Cu / L as CuSO₄ x 5H₂O)

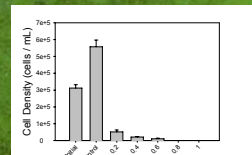
Results: Algal Cell Density



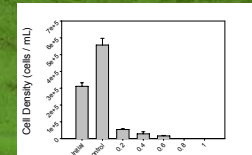
Treatment Concentration (mg Cu / L as Citrine-Ultra)



Treatment Concentration (mg Cu / L as Cleargate)

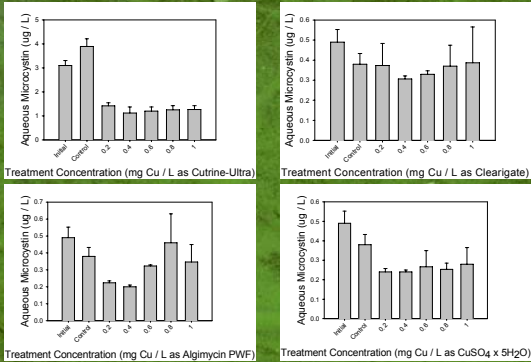


Treatment Concentration (mg Cu / L as Algimycin PWF)



Treatment Concentration (mg Cu / L as CuSO₄ x 5H₂O)

Results: Aqueous Microcystin Concentrations



Conclusions

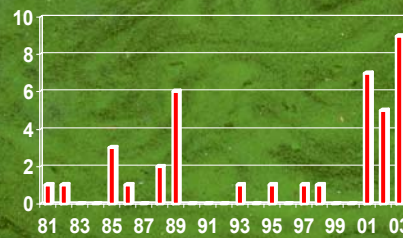
- Control of *Microcystis aeruginosa* was achieved:
 - Specific formulation, concentration, and duration of exposure.
 - Statistically significant decrease in aqueous microcystin concentrations (Cutrine®-Ultra).
 - Site-specific algaecide treatment strategy.
 - [Cu] as algaecide needed to achieve the desired level of control << 1 mg Cu / L.
 - Cutrine®-Ultra, Clearigate® = 0.2 mg Cu / L.
- Different forms of Cu exposure ≠ same responses; same algae.

References

- Peterson, H.G.; Hrudehy, S.E.; Cantin, I.A.; Perley, T.R.; Kenefick, S.L. 1995. Physiological toxicity, cell membrane damage and the release of dissolved organic carbon and geosmin by *Aphanizomenon flos-aquae* after exposure to water treatment chemicals. *Water Research* 29(6), 1515-1523.
- Jones, G.J., Orr, P.T. 1994. Release and degradation of microcystin following algicide treatment of a *Microcystis aeruginosa* bloom in a recreational lake, as determined by HPLC and protein phosphatase inhibition assay. *Water Research* 28(4), 871-876.

History in Texas

Fish Kills 1981 to 2003 (millions)



Treatments – Chemical

- **Copper algaecides**
 - **Copper sulfate – CuSO₄**
 - some species and life stages of fish are very sensitive to copper ion in water
 - not a preferred method (harmful to primary and secondary production)
 - **Cutrine® Plus - chelated form of CuCO₃**
 - effective treatment = 0.2 mg/L total copper
 - treatments ≥ 0.4 mg/L caused significant rainbow trout mortality



Lyngbya Growth And Control in Southern Reservoirs

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Scott Lankford and Wes Anderson
Alabama Power
Pelham, AL



The Villain = *Lyngbya wollei*

- Prokaryote – Cyanobacteria
- Filamentous
- Mucilagenous sheath
- Cell Wall
- Productivity – low light, rapid growth
- Production- standing crop
- Dispersal – fragments
- Toxin production

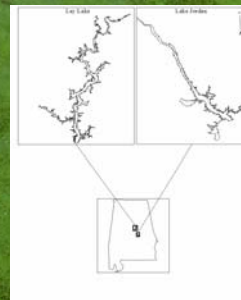
Lyngbya wollei in Jordon Lake



Study Sites – Alabama, Louisiana

Alabama

Louisiana



Issues with Lyngbya Infestations

Alabama

Louisiana

- | | |
|---|---|
| <ul style="list-style-type: none"> • Home owners • Boat owners • Taste and odor • Fish production | <ul style="list-style-type: none"> • Fish toxicity • Invertebrate toxicity • Fish production |
|---|---|

Approach to the problem

- Risk assessment – problem or not?
- No decision / action vs. decision / action
- Consider all available options
- Implement viable option(s)
- Monitor results
- Modify approach if indicated

Consider all available options

- Biological
- Physical
- Mechanical
- Chemical
- Combinations

Lyngbya wollei

Consider All Available Options

Laboratory Studies

- Copper formulations
 - Algimycin®-PWF, Earth Tec, Cutrine®-Plus, Cutrine®-Ultra, Clearigate
- Peroxide formulations
 - Green-Clean®, Green-Clean® PRO, PAK™ 27, Phycomycin
- Endothall Formulations
 - Hydrothol®191, Aquathol®
- Diquat dibromide
 - Reward®

Consider All Available Options

Laboratory Studies



Bluewell Bridge Nersons Slough Kelleys Slough

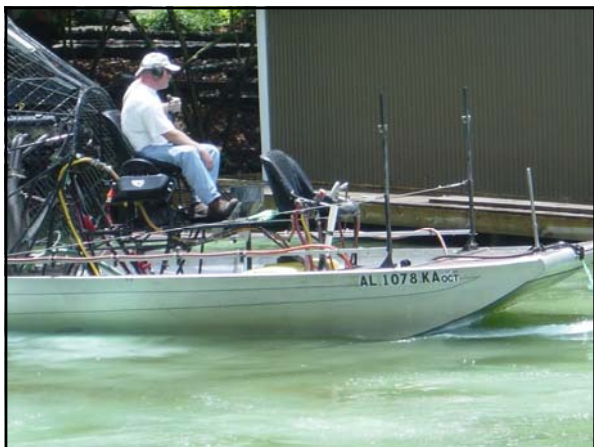
Implement Viable Options

Alabama Louisiana

- Pak-27
- Algimycin®-PWF
- Cidekick II
- Clearigate®

Implement Viable Options



Monitor Results

Measure:

- Chlorophyll *a*
- Mass (biomass)
- Density
- Oxygen production
- Respiration
- Responses of non-target species
- Residues



Modify approach if indicated



Conclusions and Observations to Date

- ~ 70% decrease in Chlorophyll a
- Absence of *Lyngbya* surface mats
- Light brown to yellow coloration observed - *Lyngbya* surface mats and benthic mats
- Treated benthic mats are less cohesive than untreated mats



Observations

- No decision is a decision. However, no decision does not = zero risk.
- Multiple treatments/ monitoring are often required (management).
- Success of treatments often depends on applicator skill and equipment.
- Timing of treatments can be important (less density, higher dissolved oxygen, etc.)
- Risk assessment can help to focus decisions.

Is this in your future?

HEALTH ADVISORY
 TOXIC ALGAE PRESENT IN THIS WATER.
NO Full Body Contact.
 Avoid Ingesting Lake Water.
 Monitor Small Children & Pets.
 Avoid Concentrations of Algae.

HAB Toxicology – Traditional vs. New

- | | |
|--|---|
| <ul style="list-style-type: none"> • TRADITIONAL • Who is it? • Where is it? • What is it doing? • What toxin is it producing? • Who is adversely affected? • Who is interested in a report? | <ul style="list-style-type: none"> • NEW • Who is it? • What are the probable risks (risk assessment)? • What are we going to do about it? What can we do? • Do it! • Monitor and learn. |
|--|---|

Factors influencing algaecide selection

- Target algal species (strain)
- Water resource usages
- Water body and water characteristics
- Efficacy
- Costs
- Margin of safety for non-target species

Animal Testing Species for Margins of Safety for Nontarget Species

Ceriodaphnia dubia

Pimephales promelas



Margin of Safety (MOS)

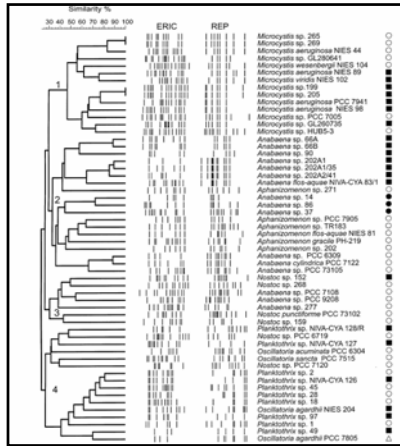
Scientific name	Algaecide	96 h LOEC ^a <i>Pimephales promelas</i> (mg / L)	EC ₁₀₀ ^b <i>Prymnesium parvum</i> (mg / L)	MOS ^c = LOEC of <i>Pimephales promelas</i> / [Cu] required to control <i>Prymnesium parvum</i> (mg / L)
<i>Pimephales promelas</i>	Citrine [®] -Plus	0.750	0.2	3.75

^a Lowest Observed Effect Concentration (Murray-Gulde et. al, 2002)

^b [Cu] used to control *Prymnesium parvum* (EC₁₀₀)

^c Margin of Safety (MOS)





Dendrogram of cyanobacterial genomic fingerprints (generated by REP- and ERIC-PCR). Scale = similarity coefficients. Numbers 1-4 = groups. Symbols: ● neurotoxic; ○ non-toxic; ■ hepatotoxic; □ toxicity not determined; △ type of toxicity unknown. *Lyra et al., 2001*

