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RUI: The Role of Dissolved Organic Material in Regulating Primary Production in Prairie Saline Lakes

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Final Report for Period: 09/2007 - 08/2008**Submitted on:** 11/04/2008**Principal Investigator:** Saros, Jasmine .**Award ID:** 0751273**Organization:** University of Maine**Submitted By:**

Saros, Jasmine - Principal Investigator

Title:

RUI: The Role of Dissolved Organic Material in Regulating Primary Production in Prairie Saline Lakes

Project Participants

Senior Personnel

Name: Saros, Jasmine**Worked for more than 160 Hours:** Yes**Contribution to Project:**

Saros is the lead investigator on the project. She coordinates all components of the project and oversees the work in the lakes of North & South Dakota. She has supervised the M.S. project as well as 6 of the undergraduate projects.

Name: Fritz, Sherilyn**Worked for more than 160 Hours:** Yes**Contribution to Project:**

Fritz oversees the sampling of the Nebraska lakes and supervises 3 additional personnel on this project.

Name: Osburn, Christopher**Worked for more than 160 Hours:** Yes**Contribution to Project:**

Osburn oversees all of the research related to the measurement and characterization of dissolved organic material in all of the lakes.

Post-doc

Graduate Student

Name: Salm, Courtney**Worked for more than 160 Hours:** Yes**Contribution to Project:**

Courtney is conducting her Master's research as part of this grant. The mesocosm experiments form the basis of her MS thesis. She is also assisting with the mentoring of undergraduate research.

Undergraduate Student

Name: Wigdahl, Courtney**Worked for more than 160 Hours:** Yes**Contribution to Project:**

Undergraduate research project paid with a stipend from the grant

Name: Erickson, Jarvis**Worked for more than 160 Hours:** Yes**Contribution to Project:**

Undergraduate research project paid with a stipend from the grant

Name: Henke, Margaret

Worked for more than 160 Hours: Yes

Contribution to Project:

Undergraduate research project paid with a stipend from the grant

Name: Martin, Callie

Worked for more than 160 Hours: Yes

Contribution to Project:

Undergraduate research project paid with a stipend from the grant

Name: McMullen, Brian

Worked for more than 160 Hours: Yes

Contribution to Project:

Name: Toban, Robert

Worked for more than 160 Hours: No

Contribution to Project:

Name: Czubakowski, Jessica

Worked for more than 160 Hours: Yes

Contribution to Project:

Jessica is conducting an undergraduate research project as part of this grant. She is performing a series of nutrient enrichment experiments.

Name: Wilcox, Erin

Worked for more than 160 Hours: Yes

Contribution to Project:

Erin is conducting an undergraduate research project as part of this grant. She is assessing the effects of cations on alkaline phosphatase activity.

Name: Daggett, Carmen

Worked for more than 160 Hours: Yes

Contribution to Project:

Carmen has conducted a series of nutrient enrichment experiments in three of the study lakes for her undergraduate research project.

Technician, Programmer

Name: Bennett, Danuta

Worked for more than 160 Hours: Yes

Contribution to Project:

Other Participant

Research Experience for Undergraduates

Organizational Partners

Bowling Green State University

Dr. Michael McKay has collaborated with Saros on the iron limitation components of this research. Dr. McKay has hosted an undergraduate from UW-L in his laboratory to analyze the ferredoxin ratios of our phytoplankton samples. He will also be joining us on our August 2006 trip to North Dakota to assist with collection of additional phytoplankton samples for this analysis.

Other Collaborators or Contacts

Nebraska sampling is coordinated with water quality sampling funded by the Nebraska Department of Environmental Quality and carried out by the University of Nebraska - Lincoln School of Natural Resources.

Activities and Findings

Research and Education Activities: (See PDF version submitted by PI at the end of the report)

Findings: (See PDF version submitted by PI at the end of the report)

Training and Development:

Seven undergraduates and one M.S. student from UW-La Crosse and two undergraduates from UN-Lincoln have participated in the field surveys & experiments. Most of these students had little or no research experience in aquatic ecology prior to this project. While each student has specialized in one or two techniques as part of their research project, they have learned all of the collection methods and observed all of the techniques involved. Saros also worked closely with the undergraduates during method development, which enhanced the students' understanding of the techniques as well as Saros' teaching skills in this area. As mentioned above, two of these students won poster awards for their presentations at national and international meetings.

With the collaborative nature of this project, students have access to investigators and equipment from other institutions. These students are also participating in the synthesis of data across the project.

Outreach Activities:

We have prepared reports for landowners in the vicinity of our sample lakes. These reports consist of basic water chemistry parameters as well as phytoplankton standing crop, and any specialized information pertinent to particular lakes. Where possible, we have contacted and informally discussed with landowners and interested parties our activities. We note a decided interest in these lakes from local communities, especially lakes developed (e.g, houses) and used for recreation. There is concern about the 'health' of these lakes and most people were receptive and supportive of the project.

We have also provided our data to the Fish & Game Departments in these states. This has been particularly helpful to the North Dakota Fish & Game Department, which has implemented a phosphorus reduction project in one of our study lakes. Our water chemistry and algal physiology data were particularly useful to them.

Journal Publications

Books or Other One-time Publications

Web/Internet Site

Other Specific Products

Contributions

Contributions within Discipline:

Saline lake ecosystems are model extreme environments that are ubiquitous in arid climates yet remain relatively unstudied. We have gained insight into the cycling of DOM in these systems and the coupling (or un-coupling) between DOM and primary production-- which is often quite high in saline lakes.

By using physiological metrics and multivariate statistics, we have begun to refine patterns in primary production across saline lakes. Our results indicate that numerous factors play a role in regulating primary production across these lakes, and challenge the paradigm of phosphorus control of primary production in all lake ecosystems.

Osburn has made advances in the analytical methodology of stable carbon isotopes of DOM in saline waters. The prairie saline lakes have been quite challenging for geochemical analyses and work on them has advanced methods for DOM stable isotope measurement that has extended to pore waters and ocean water.

Contributions to Other Disciplines:

One tangential aspect of this research is that these lakes are potential sources of feedstock for biofuels. Osburn is involved in research of novel feedstocks for synfuel development and has begun investigating the use of algal and microbial biomass from saline lakes and wetlands.

Contributions to Human Resource Development:

This grant funded summer research stipends for seven undergraduates at UW-La Crosse and two at University of Nebraska - Lincoln. These students typically fund their own education, and hence must be employed in some fashion during the summer. Many of our students do not engage in research due to financial difficulties. This grant is allowing these students to gain valuable professional experience while still earning a salary.

This grant also funded the education of a Master's student at UW-La Crosse. The majority of graduate students at UW-La Crosse are not financially supported by the program, hence the funding from this grant provides important support for the graduate student and also promotes the graduate program overall.

Contributions to Resources for Research and Education:

Funds from this grant have been used to acquire small pieces of equipment, such as a benchtop centrifuge, as well as to upgrade existing equipment, such as additional kits for a field fluorometer.

Contributions Beyond Science and Engineering:

Categories for which nothing is reported:

Any Journal

Any Book

Any Web/Internet Site

Any Product

Contributions: To Any Beyond Science and Engineering

Findings

The 30 lakes in the data set varied along gradients of primary productivity, salinity, ion composition and nutrient conditions, and thus serve as an excellent data set to investigate patterns of primary production. Rates of primary production (as measured by ^{14}C incorporation) generally followed patterns in chlorophyll concentration ($R^2=0.72$), but deviated enough to warrant the use of productivity measurements as the metric for comparison with other variables. Using Akaike's Index Criterion (AIC) as well as Classification and Regression Trees (CART), we have discerned patterns of nutrient limitation across these lakes (Table 1). Despite the fact that these lakes all have total nitrogen concentrations exceeding 1 mg/L, they are primarily nitrogen-limited, with a sub-set of lakes secondarily limited by phosphorus. The 2006 experiments confirmed the model results (Table 2).

Table 1. Summary of models developed with Akaike's Information Criterion (AIC) for 2004 comparative lake sampling.

Model	Model Predictors	D AICc	w	K	R ²
SPRING - All lakes (44)*	LogNO ₃ (+), C:N(+), LogDFE(-), LogA250:A365(-)	0.00	0.086	4	0.795
	LogNO ₃ (+), C:N(+), LogDFE(-),	0.72	0.060	3	0.756
	LogCA(-), LogSO ₄ (-), LogTN(+), LogDFE(-)	1.01	0.052	4	0.786
	pH(+), LogNA(-), LogNO ₃ (+), LogDFE(-)	1.29	0.045	4	0.783
	LogCA(-), LogSO ₄ (-), LogTP(+), LogDFE(-)	1.54	0.040	4	0.781
SPRING – NGP lakes only (3)	LogCOND(-), LogCA(-), LogTP(+), LogDFE(-)	0.00	0.652	4	0.753
	LogALK(-), LogCA(-), LogTP(+), LogDFE(-)	2.40	0.196	4	0.717
	LogCA(-), LogSO ₄ (-), LogTP(+), LogDFE(-)	2.91	0.152	4	0.709
SUMMER - All lakes (58)	LogNA(-), LogCL(+), C:P(+), LogDFE(+)	0.00	0.060	4	0.715
	TEMP(-), LogNA(-), LogCL(+), C:P(+)	0.03	0.059	4	0.714
	LogK(-), LogTP(+), C:P(+), LogDFE(+)	0.17	0.055	4	0.713
	LogK(-), LogCL(+), LogSI(+), C:P(+)	0.49	0.047	4	0.710
	LogK(-), LogNO ₃ (+), C:P(+), LogA350(+)	0.99	0.036	4	0.705
SUMMER - NGP lakes only (17)	LogTP(+), LogSRP(-), NP(+), LogDFE(+)	0.00	0.202	4	0.730
	LogTP(+), LogSRP(-), NP(+), LogTFE(+)	0.65	0.147	4	0.720
	LogTP(+), LogSRP(-), NP(+)	1.45	0.098	3	0.636
	LogFAPA(-), LogTP(+), LogSRP(-), NP(+)	1.84	0.081	4	0.701
	LogNA(-), LogCL(+), LogTDP(-), DO ¹³ C(-)	2.02	0.074	4	0.698

* indicates the number of models with DAICc < 4, which were used to compute the Akaike weights

Table 2. Summary of Tukey's post-hoc analyses for spring and summer 2006 experiments. Numbers in each column indicate groups of treatments with responses that are not significantly different from each other ($\alpha = 0.05$). Nutrient treatments: C=control, Fe=Fe addition, P=P addition, N=N addition, N+P=combined N and P additions. Lake abbreviations: Alk=Alkaline Lake, CW=Coldwater Lake, FP=Free Peoples Lake, ED=East Devil's Lake, Stk=Stink Lake, Clear=Clear Lake, Grg=George Lake.

Treatment	SPRING					SUMMER					
	Alk ^a	CW ^a	FP ^a	ED ^a	Stk ^b	Alk ^a	ED ^a	Stk ^c	Clear ^a	CW ^a	Grg ^a
<i>Prod. rate</i>											
C	1	1	1	1	-	1	1	-	1	1	1
Fe	1	1	1	1	-	12	1	-	1	1	1
P	12	1	1	1	-	2	1	-	2	1	1
N	2	2	2	2	-	2	2	-	1	2	2
N+P	3	3	2	2	-	3	2	-	2	3	3
<i>Chl a</i>											
C	1	1	1	1	1	1	1	1	2	12	1
Fe	1	1	1	1	2	1	1	1	1	2	1
P	1	1	1	1	12	2	1	1	3	1	2
N	2	2	2	2	3	2	2	2	12	3	2
N+P	3	3	3	2	3	3	3	2	4	4	3

^a $p < 0.001$ for analysis of variance for productivity and chl a data.

^b $p = 0.044$ for analysis of variance for productivity data, but post-hoc analysis did not detect different subsets of lakes. $p < 0.001$ for analysis of variance for chl a data.

^c $p = 0.325$ for analysis of variance for productivity data. Post-hoc analysis was not performed on these data. $p < 0.001$ for analysis of variance for chl a data.

The late spring phytoplankton assemblages in 2004 were dominated by diatoms, with coccoid cyanobacteria abundant in a sub-set of lakes. Despite the prevalent N limitation, cyanobacteria that are capable of N fixation were not abundant. Canonical correspondence analysis of 2004 data showed salinity (logCl), nitrogen, and phosphorus (N:P ratios) to be the main drivers of phytoplankton distribution in the spring, and phosphorus (C:P ratios), iron (logTFe), and nitrogen (logTN) as important factors in the summer (Fig. 1). Despite major differences in nutrient limitation patterns (P-limitation in freshwater systems, N-limitation in saline systems), seasonal patterns of phytoplankton phyla changes in these saline lakes were similar to those of freshwater systems. Dominance shifted from diatoms in the spring to cyanobacteria in the summer. Nutrient enrichment assays (control, +Fe, +N,+P, +N+P) in 2006 indicated that nutrient limitation is generally more consistent within lakes than for individual taxa across systems.

Bulk alkaline phosphatase (APA) activity measurements were essential to discerning nutrient limitation patterns in these lakes because of the high total nutrient pools. In addition, APA appeared low in high calcium lakes, suggesting a possible inhibitory effect of calcium on this enzyme. In our experiments to test the effects of cations on APA, we found that both calcium and potassium lowered APA. While potassium stimulated P uptake, calcium did not, hence the mechanism involved here is still not understood. In the field samples, enzyme-labelled fluorescence of alkaline phosphatase activity revealed interspecific differences in the expression of this activity, thereby allowing us to resolve

nutrient limitation patterns more fully. This technique also allowed us to separate bacterial from phytoplankton activity.

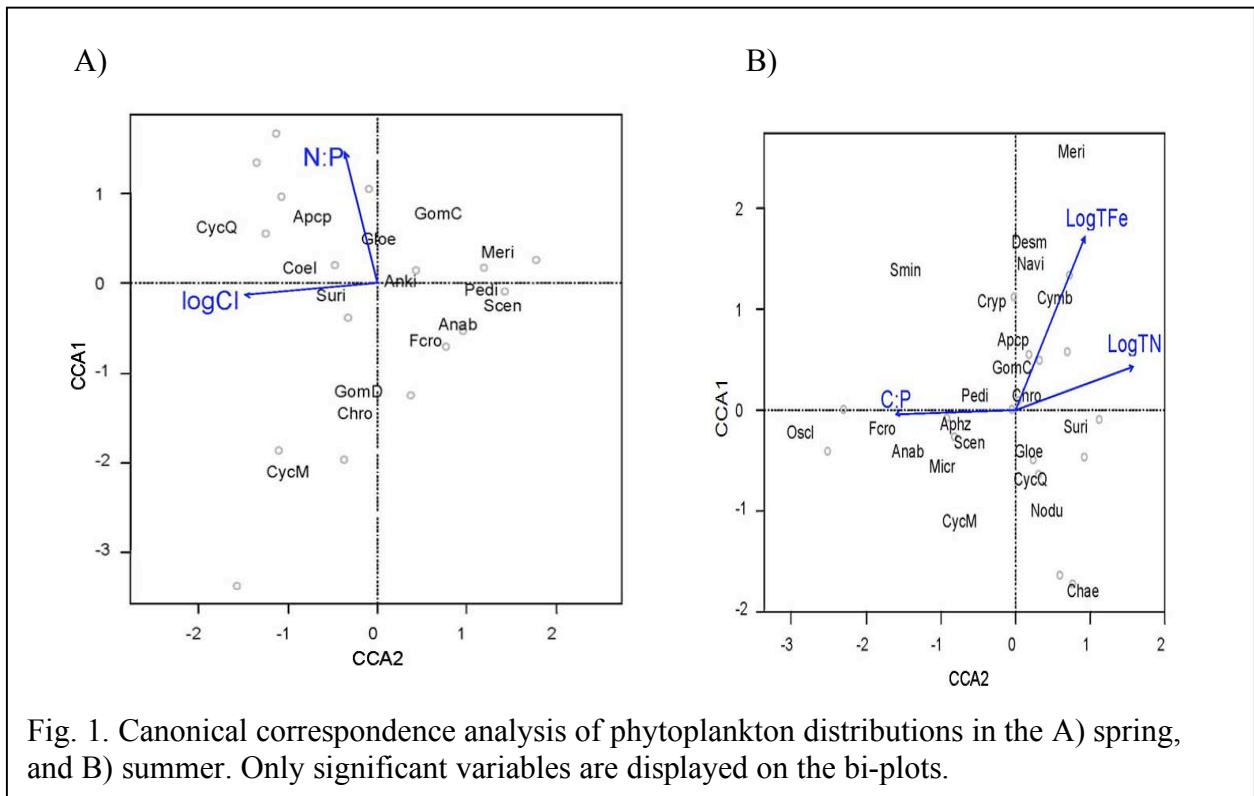


Fig. 1. Canonical correspondence analysis of phytoplankton distributions in the A) spring, and B) summer. Only significant variables are displayed on the bi-plots.

The photolysis experiments did not indicate that higher levels of UV exposure release nutrients for phytoplankton use. Bacterial productivity was also assessed in these experiments, and showed no change with photolysis.

DOC quantities varied considerably among the 30 lakes. In general, the much shallower NE lakes exhibited the highest DOC concentrations in Osburn's data set (> 2 mM DOC). Most lakes exhibited high UV attenuation, but when normalized to DOC, these lakes are quite transparent. Amino acid and carbohydrate results were further illuminating and surprising. Most carbohydrates were in the form of monosaccharides as opposed to polysaccharides in the Dakota lakes. Amino acid concentrations were less than 1 micromolar in most lakes, with the exception of Goose Lake, MT which had serine and glycine concentrations of several micromolar. The stable carbon isotope data for DIC and DOC revealed a complex geochemistry for DOM in these lakes. In general DOC stable isotopes were 3 to 9 per mil *heavier* than DOM measured from freshwater lake ecosystems. While this may be reflective of the prairie grassland vegetation, several lakes in our study (notably Goose in MT and Alkali in ND) have substantial autochthonous C input.

Activities

The goal of this research was to investigate patterns of primary production in prairie saline lakes of the Northern Great Plains of the U.S. These lakes do not behave similarly to freshwater temperate lakes, in that linear regression models of chlorophyll versus nutrients or salinity do not reveal a pattern in primary productivity trends. We hypothesized that the high dissolved organic material (DOM) concentrations in these lakes influence algal productivity by binding nutrients and reducing their availability.

A set of field surveys was conducted in May/June and August 2004 in North and South Dakota as well as Nebraska for a total of 30 lakes. During this field work, Saros and four undergraduates collected data on phytoplankton productivity patterns, including ^{14}C primary production, alkaline phosphatase activity, and seston nutrient ratios. A novel technique involving fluorescent labeling of phosphorus-limited cells was successfully conducted as an undergraduate project, and provided greater resolution of nutrient limitation patterns in these lakes. A second undergraduate assessed iron limitation by measuring the ratio of ferredoxin to flavodoxin in phytoplankton cells; we collaborated with Dr. Michael McKay of Bowling Green State University to conduct these analyses. Two additional undergraduates worked on ^{14}C primary productivity and phytoplankton species composition. Fritz and her research group, including two undergraduates assisting in the field, collected the same type of data for the Nebraska lakes. An additional undergraduate assisted with laboratory analyses of the samples.

Osburn collected samples from all lakes for DOM quantification and characterization. Lake water samples were filtered through 0.2 micron filtration and subsequently analyzed for UV-vis absorption, synchronous fluorescence, excitation-emission fluorescence (EEM), and dissolved organic carbon (DOC) concentration. (2) 500 mL aliquots of filtered lake water were individually extracted onto ca. 10 mg C18 sorbent. One column was reserved for dissolved lignin measurement, while the other column was reserved for humic-bound phosphorus analysis. Osburn also directed light attenuation profiles of each lake (weather permitting). During the deployment of the light meter and subsequent data acquisition, Osburn had the assistance of 1 to 2 undergraduates and was able to instruct them in rudimentary field measurements of light (UV and visible) attenuation.

In Spring 2005, Osburn measured stable isotopes of DIC and DOC on the 2004 lake data set. This information built upon lignin and optical information about DOM in the prairie lakes. In August 2005, Osburn returned to the majority of the 2004 lakes and again sampled for DIC, DOC, stable isotopes, and optical measurements. While DOM samples were not collected for lignin analysis in 2005, Osburn did sample vegetation in several of the lake's watersheds to constrain allochthonous C input to the lakes.

In August 2005, a series of nutrient enrichment experiments were conducted in 6 of the Dakota lakes. We also conducted an experiment with four types of dissolved organic material to assess the effects of this material on primary production. A third experiment was conducted to assess the effects of cations on the rate of alkaline phosphatase activity.

In May 2006, 12 of the lakes in the data set were re-sampled to assess inter-annual variability in the measured parameters. Two undergraduates and Saros conducted this field sampling. Saros'

lab also conducted three experiments. Two of these experiments were conducted by Courtney Salm, the Master's student on the project. She conducted a nutrient enrichment experiment with 5 lakes to test the primary productivity models that she has developed (see below), and she also conducted a DOM photolysis experiment. In the latter experiment, she tested the effects of UV photolysis on the release of nutrients from DOM, and the influence of this on primary production. The third experiment was conducted by an undergraduate, Carmen Daggett; she investigated iron limitation in three lakes through a nutrient enrichment experiment. This was coupled with additional ferredoxin measurements by McKay's group.

Eighteen presentations of this research were given at national and international meetings. Twelve of these presentations were given by undergraduates, including four presentations at the International Society for Salt Lake Research meeting in Perth, Australia. The University of Wisconsin-La Crosse primarily funded the travel to this meeting for Saros and all four students. Saros also presented at this meeting; her participation resulted in her election to the board of the society. Saros was also invited to present results from this project at a shallow lakes workshop held at the St. Croix Watershed Station in Minnesota. Saros & Osburn co-chaired a session on dissolved organic material in prairie saline lakes at the annual meeting of the American Society of Limnology & Oceanography in 2005.

Two of the student poster presentations received awards. Courtney Salm received a Best Student Poster Award at the International Society for Salt Lake Research in Perth, Australia, in September of 2005. In February 2007, Jessica Czubakowski received an honorable mention for her poster presentation at the annual meeting of the American Society of Limnology & Oceanography.

The following publications are resulting from this project:

Graduate advisees indicated with *; undergraduate advisees with a ^

- 1) Salm, C.R.*, J.E. Saros, C. Martin^ & J.M. Erickson^. Patterns of seasonal phytoplankton distribution in prairie saline lakes of the Northern Great Plains (USA). Submitted to *Saline Systems*, July 2008; currently undergoing minor revision.
- 2) Salm, C.R.*, J.E. Saros, S.C. Fritz, C.L. Osburn & D.M. Reineke. Lake classification models of primary production by phytoplankton in prairie saline lakes of the Great Plains (USA). Submitted to *Canadian Journal of Fisheries & Aquatic Sciences*, July 2008; currently undergoing revision.
- 3) Saros, J.E., C.R. Salm*, R.M. McKay, C.L. Osburn & S.C. Fritz. Biochemical tools and nutrient enrichment assays reveal phytoplankton nutrient limitation by nitrogen, phosphorus, and iron in lakes with high concentrations of non-chromophoric dissolved organic material. In preparation for *Journal of Plankton Research*.
- 4) Osburn, C.L., J.E. Saros, C.R. Salm*, L.J. Hamdan & S.C. Fritz. Optics and geochemistry of dissolved organic matter in prairie saline lakes. In preparation for *Biogeochemistry*.