2013 Project Abstract
For the Period Ending June 30, 2017

PROJECT TITLE: Evaluation of Lake Superior Water Quality Health
PROJECT MANAGER: Erik T. Brown
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WEBSITE: [If applicable] http://www.d.umn.edu/llo/
FUNDING SOURCE: (Environment and Natural Resources Trust Fund
LEGAL CITATION: M.L. 2013, Chp. 52, Sec. 2, Subd. 05f
M.L. 2016, Chapter 186, Section 2, Subdivision 18

APPROPRIATION AMOUNT: $600,000
AMOUNT SPENT: $600,000
AMOUNT REMAINING: $0

Overall Project Outcomes and Results

Although Lake Superior seems timeless, it has been changing dramatically—with shifting temperatures, ice cover, storminess, and biological activity. This project worked to build our knowledge of how the lake responds to external processes, including climate change and the introduction of invasive species. This will help us to protect and foster this resource during a time of unprecedented change.

We used state-of-the-art techniques to evaluate the lake's behavior from Fall 2013 through Fall 2016. This included an extreme range of winters—the "Polar Vortex" of 2013-14 and the mild conditions of 2015-16. Our field strategy included shipboard sampling (12 stations occupied four times each year) as well as use of autonomous gliders and moored instruments. These unmanned technologies provide cost-effective measurements at more places and times than possible with ship operations.

Major results include:

Lake circulation. Building on observations in the lake, we created a hydrodynamic numerical model of the St. Louis Estuary/Duluth Harbor/Lake Superior system that runs in real-time, providing estimates of currents and water levels across Lake Superior. Such information is useful to boaters and fishermen, and is being used in St. Louis River Estuary wastewater studies, and for studies of riverine nutrient dispersion and of nearshore wave action around the Apostle Islands

Lake acidity. We quantified seasonal shifts in lake pH due to river runoff, atmospheric inputs, and biological activity. Increased atmospheric CO_2 has acidified many lakes. In Lake Superior this appears to be mitigated by reductions in acid rain after clean air legislation of the 1990s.

Algae and plankton. We now have measurements of biological productivity from 2006 through 2016. Broadly, we see increased productivity in warmer years, with lower biomass of small algae that photosynthesize rapidly. These productive small algae might dominate a future, warmer Lake Superior. Such a shift could lead to significant changes for animals higher on the food chain.

Exotic species. Our work demonstrates that the invasive spiny water flea has damaged Lake Superior's lower food web. Our data provide a baseline for future evaluation of shifts in zooplankton.

Fish. We assessed historical patterns in growth of lake herring (cisco) using archived ear bones in combination with our current data. It appears that climate change and invasion by the spiny water flea have not greatly affected cisco growth rates to date. Nevertheless, spiny water fleas are a relatively poor prey item and could reduce growth rates of cisco that consume them.

Data obtained through this project have been utilized in proposals to the US National Science Foundation. Three successful proposals yielded ~$2.0M for Lake Superior research including 70 days of shiptime. A large proportion of this funding supports personnel and thus has a real impact on our local economy.

Project Results Use and Dissemination

We have worked to disseminate our results and information about Lake Superior science to the general public and the scientific community in several ways. These include: news reports on our work through print, television and radio; an ongoing social media presence; outreach events with public tours of UMD's research vessel; and publication of results in the scientific literature.

Environment and Natural Resources Trust Fund (ENRTF) M.L. 2013 Work Plan. Final Report

Date of Status Update Report: 11 August 2017

Final Report

Date of Work Plan Approval: June 11, 2013

Project Completion Date: June 30, 2017 **Is this an amendment request?** _No___

Project Title: Evaluation of Lake Superior Water Quality Health

Project Manager: Erik T. Brown

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Web Address: http://www.d.umn.edu/llo/

Location: St. Louis, Lake, and Cook Counties

Total ENRTF Project Budget:	**ENRTF Appropriation:**	**$600,000**
	Amount Spent:	**$600,000**
	Balance:	**$ 0**

Legal Citation: M.L. 2013, Chp. 52, Sec. 2, Subd. 05f
M.L. 2016, Chapter 186, Section 2, Subdivision 18

Appropriation Language:
$600,000 the first year is from the trust fund to the Board of Regents of the University of Minnesota to evaluate impacts to Lake Superior from a changing thermal structure and invasive species in order to implement lake water quality management strategies. This appropriation is available until June 30, 2016, by which time the project must be completed and final products delivered.
Carryforward: (a) The availability of the appropriations for the following projects are extended to June 30, 2017: (5) Laws 2013, chapter 52, section 2, subdivision 5, paragraph (f), Evaluation of Lake Superior Water Quality Health

I. PROJECT TITLE: Evaluating Lake Superior's Health in a Changing World

II. PROJECT STATEMENT: Lake Superior is among Minnesota's greatest natural resources. Shipping, recreation and tourism on the lake stimulate Minnesota's economy. It provides drinking water to thousands, and it symbolizes our natural heritage. The lake seems timeless, but recently it has been changing dramatically. For example:

- Surface summer water temperatures have increased by 5°F over the past 30 years, some of the most rapid change observed on the planet;
- Average ice cover on the lake over the past 15 years is 2-fold lower than the long-term average;
- Major storm events are becoming more frequent (the Duluth floods of June 2012 are an extreme example);
- The lake's nutrient content is changing markedly, with nitrate increasing 5-fold since 1900;
- The lake is increasingly affected by invasive species (spiny water flea, zebra mussels; etc);
- The lake's biological productivity is decreasing, providing less food for fish;
- While lake trout and herring have recovered from mid-20th Century decimation, lake herring populations rise and fall dramatically from year to year.

We do not fully understand the reasons for these changes, or complex interactions among them. Yet we need such an understanding to protect and foster this resource during a time of unprecedented change.

A team from the U of MN and the DNR will use state-of-the-art techniques to evaluate the lake's behavior in this rapidly changing temperature regime. Ship-based observations (using the research vessel *Blue Heron*) will focus on two transects across the western arm of Lake Superior. These results will be complemented with data obtained using an autonomous underwater glider and moored profiling instruments (purchased with National Science Foundation—NSF—grant support). These new unmanned technologies provide cost-effective measurements at more places and times than possible with labor-intensive ship operations. Our plan (see graphic) includes:

a. Shipboard sampling (from 5 to 10 depths at 12 locations along two transects occupied four times in 2014 and 2015) to measure nutrients, pH, carbon, oxygen, temperature, particle abundances and composition, activity of photosynthetic and other pigments. We will also measure primary productivity, carbon and nutrient cycling using sediment traps and species abundances of algae, zooplankton, and fish.
b. Use of an autonomous underwater glider for measurements of temperature, chlorophyll and other pigments, oxygen and water clarity. This unmanned device, which can be released and recovered from small boats, is programmed to "swim" repeatedly from surface to bottom as it navigates across the lake, and can provide results in near-real time via satellite telephone. This work during 2014 and 2015 field seasons will provide a detailed context for interpretation of the ship-based sampling efforts.
c. Moored profilers to be deployed from Fall 2013 to Spring 2016. These instrument packages are "parked" about ~100 feet below the lake's surface. Several times daily they unspool and float upward to record profiles of temperature, oxygen, nitrate, currents, chlorophyll and other biologically important pigments, and water clarity, providing real time data via satellite telephone. They can provide year-round observations, including under ice measurements never previously made in Lake Superior.
d. Biological and chemical analyses of archived samples collected over the past decades to identify historic trends—such as changes in fish feeding habits or timing of arrival of invasive species. We will use newly developed analytical techniques to examine samples collected in the past and stored at U of MN or the DNR.
e. Education and research opportunities for graduate and undergraduate students. Summer stipends will be provided for 5 graduate and 4 undergraduate students during the major field season, along with partial support for one graduate student during the academic year. Other graduate students involved with the project will receive academic year support through other funds such as U of MN teaching assistantships.
f. Public outreach through a partnership with existing programs and expertise at MN Sea Grant.

This work builds on projects that were initiated with support from MN SeaGrant, the US National Science Foundation (NSF) and from LCCMR. The U of MN's research ship, *Blue Heron*, was purchased with LCCMR support in 1998 and is directly tied to projects that have brought some $14 million in competitive external research funding to Minnesota. Research supported by MN Sea Grant has demonstrated the critical role that daily fish migration patterns play in the lake. With SeaGrant and NSF support, U of MN scientists have also made significant progress in understanding overall productivity and nutrient cycling. Recently, NSF provided funds for equipment purchases (glider and moored profiling system) that allow us to make cost-effective measurements to evaluate effects of lake circulation processes (currents/mixing) at far more places and more times than possible with ship-based operations. The work proposed here will merge these avenues of research, using measurements of lake circulation and mixing to improve our understanding of biological processes, including fish and plankton behavior. No prior large lake study has included the breadth of measurements, the geographic range, and the span of seasons we propose.

III. PROJECT STATUS UPDATES:

Project Status as of February 2014: We have been working to prepare for Lake Superior monitoring programs, and have made concrete plans for the first field season (May-October 2014). We had a limited deployment of moored systems in Fall 2013. Technical issues (this is a cutting edge technology) made us concerned about a full over-winter deployment so we pulled the gear in for the winter rather than risk losing it. Nevertheless, this deployment provided an unprecedented record of lake conditions during the fall seasonal transition.

We have made progress in outreach goals as well. We are in design and construction phases of a new interactive display at the Great Lakes Aquarium (supported by a number of entities including LCCMR) that enables the public to investigate how Lake Superior internal waves respond to changing wind conditions. Minnesota SeaGrant published a short newsletter article that included a description of the Fall 2013 moored instrument deployment (http://www.seagrant.umn.edu/newsletter/2014/02).

Project Status as of October 2014: We had a successful first field season with four sampling cruises in summer 2014 (total of 20 days on the lake) in addition to mooring deployments. The very cold conditions in Lake Superior during 2014 provide a key contrast to recent trends that can aid in understanding the lake's response to the generally warmer conditions projected for coming years. Cruises involved an intensive sampling program to determine abundances and species of plankton and fish, to study nutrient and carbon dynamics, and to measure rates of biological productivity. We have initiated analyses of archived plankton samples to provide a historic context for these results. The project provided research employment opportunities for undergraduate and graduate students.

We have expanded the scope of the project (at no cost to ENRTF) by partnering with other researchers and tapping other funding sources. A research team from Michigan Technological University joined the field program to undertake monitoring that will be used for ground-truthing satellite imagery of Lake Superior for evaluation of parameters including lake surface temperature, chlorophyll, and suspended sediment. We have coordinated with MN-DNR to take advantage of their scheduled fish trawling to expand our seasonal coverage. Thanks to funding from the Great Lakes Observing System, we expanded our proposed use of the autonomous glider to provide a total of 30 days of deployment in western Lake Superior.

A prototype of a display for the Great Lakes Aquarium has been completed, and we continue to have periodic meetings with Aquarium staff to discuss new initiatives to highlight Lake Superior research. UMD's External Affairs Office, released an article describing the summer 2014 field season (http://www.d.umn.edu/external-affairs/homepage/14/llo-lake-ecology.html). Minnesota Sea Grant produced a "Sea Grant Files" radio interview with Robert Sterner (to be broadcast on 4 November) that discusses research on Lake Superior productivity and carbon cycling. A second interview on the implications of observations from the 2014 field season is scheduled for broadcast in mid-December. "Sea Grant Files" programs are archived on line and are available as podcasts at (http://www.seagrant.umn.edu/radio/sgf/)

Project Status as of February 2015: A major results are coming from examination of the effects of the unusually cold conditions that persisted in the lake though much of the summer after the extremely cold winter temperatures and exceptional ice cover of the 2013-14 winter (2013-14 ice cover was far greater than any previous year on record). While the temperature structure of the lake was unusual for most of the summer, with colder than normal conditions persisting into August, biological activity was not far from the "normal" range observed in recent years. Overall biological productivity for the season was comparable to "normal" years, but was concentrated at the very end of the season. The cooler temperatures of early summer may have delayed the appearance of significant numbers of the invasive crustacean spiny waterflea, allowing native species of zooplankton to flourish in the early part of the summer.

We have made substantial progress in developing numerical modeling that will lead to real-time website displays of now-casts and forecasts of lake conditions. We anticipate that this modeling work will transition to becoming an "asset" of the Great Lakes Observing System (funded by NOAA) so that this website will be supported and maintained beyond the funding period of this project. We are working toward final installation of the Great Lake Aquarium display that enables the public to investigate how Lake Superior internal waves respond to changing wind conditions. MN Seagrant and UMD's External Affairs Office continue to expand our presence on online and in social media.

Project Status as of October 2015: The LCCMR project completed five cruises in 2015 for a total of 16 days on the lake (costs for 1.5 days covered by University funds). During the field season of 2015, samples were taken for the same package of chemical and biological parameters measured in 2014. These included carbon and nutrients in different forms as well as chlorophyll, the green pigment found in phytoplankton. These measurements continue to enhance our understanding of the lake's behavior and will allow us to assess the year-to-year variability in the base of the food web in the Lake Superior ecosystem. One of our findings so far was the algal abundance at times in 2014 and 2015 were higher than observed in earlier studies. This suggests that the cold winter conditions of the previous two years created conditions of high nutrients or changed mixing, which fostered the growth of algae.

We continue to monitor invasive species. In 2015, the invasive zooplankton, spiny waterflea or *Bythotrephes longimanus,* was observed in May in the relatively shallow waters near the Apostle Islands. This observation is two months earlier than the first observation of spiny waterflea in 2014 and is likely due to warmer water temperatures brought on by a much more mild spring. However, on subsequent 2015 cruises spiny waterflea densities were lower than those observed during corresponding months in 2014. These findings are surprising because the warmer surface temperatures on Lake Superior in 2015 compared to 2014 should have increased the growth and reproductive rates of spiny waterfleas, and suggest that other phenomena may be affecting spiny waterflea abundances.

As part of LCCMR field activities in 2015, we deployed a WetLABS Autonomous Underwater Profiler in the western arm of Lake Superior on 6 June, near a meteorological buoy operated by LLO. These profilers allow us to collect information on the distribution of various properties throughout the water column, such as temperature, chlorophyll content, nitrate and oxygen concentrations, and several other fields, and to do so repeatedly. The profiler achieved 160 profiles before being successfully recovered on 21 August. At the same time, a second profiler was deployed at the same location, and has made 105 more profiles. Our current plan is to recover this profiler mid-November, and redeploy another for an over-winter deployment. The data provided by these profiles represent the most highly-resolved long term time series of biogeochemical parameters *ever* collected in a large lake, and will help to put the shipboard sampling program in proper context.

Project Status as of February 2016: All samples from 2015 for zooplankton, particulate carbon, nitrogen and phosphorus as well as chlorophyll, ammonia and silica have now been processed and we are beginning evaluation of data and comparison with other years. In contrast to the cooler summer of 2014, during the warmer summer of 2015, the lake shifted to taking up carbon dioxide in the western arm, presumably due to higher biological productivity in warmer water. Looking at individual stations over time, there appears to be an inverse relationship between surface water chlorophyll concentrations and pCO2. Overall zooplankton biomass was higher in October of 2015 than October of 2014. The invasive zooplankton species, *Bythotrephes longimanus*, was uncommon in September and October of 2015, and *Bythotrephes* densities in October of 2015 were lower than the densities observed during October of 2014. We continue to evaluate the factors influencing year-to-year differences in *Bythotrephes* densities.

UMD's autonomous deployed several times in late 2015 and early 2016, leading to a 217-day long time series of key parameters, including temperature, chlorophyll content, dissolved organic matter content, dissolved oxygen concentration, nitrate concentration, and light field. This dataset is unprecedented for the breadth of measurements as well as the duration. In addition, as the profiler operation is not weather dependent, the data provide us with insights into lake conditions when it is not be safe to work on the deck of a boat. While our intention was to have a profiler deployed throughout the winter, power management issues on the platform necessitated an emergency recovery in early January. We are currently working with the company to determine the cause of this failure and hope to have a profiler deployed as soon as ice conditions in the harbor and on the lake allow.

We have completed a hydrodynamic numerical model of the coupled St. Louis Estuary/Duluth Harbor/Lake Superior system that runs in real-time, currently providing estimates of currents and water levels across Lake Superior. Every three hours, the model uses recent meteorological observations from around the lake and runs the model forward to estimate its current state- hence the term "nowcast." We are expanding the model in two areas. The first is adding additional meteorological parameters beyond wind and barometric pressure, including air temperature, relative humidity, and cloud cover; this will allow us to predict water temperature distribution and evolution. The second is developing a web platform for the results so that they can be widely disseminated. We will be working with colleagues at Minnesota Sea Grant to make the website useful to users, from researchers to managers to fishermen to curious citizens.

During this activity period, communication efforts supporting this project have relied on: Social media (about 100,000 potential impressions via 20 posts); two public service messages that televised in September in Duluth and archived on the web; a radio program that is available to the public via podcast and on the MN Sea Grant website.

Project status as of October 2016: We had another successful field season with 3 sampling cruises during the summer of 2016 (a total of 9 days on the lake) in addition to glider and moored profiler deployments and recoveries.

The suite of analyses undertaken through this LCCMR funding, allows detailed examination of the contrast between a very cold summer (2014) and a warm one (2016). Preliminary analyses suggest that with increasing temperature, phytoplankton abundance is lower, average cell size is higher, and primary production is higher. These trends may help us project what to expect if climate warming trends continue into the future.

We had an active season of glider and profiler deployments in 2016. The moored profiler was in the water from April to July, so that over the last year we have collected a wide variety of biogeochemically relevant data (including temperature, chlorophyll content, dissolved organic matter content, dissolved oxygen concentration, nitrate concentration, the light field, and several others) nearly around the calendar, the most complete and extensive dataset of its kind. In conjunction with the Great Lakes Observing System and the EPA's CSMI (Coordinated Science and Monitoring Initiative) program, the glider was deployed for over 100 days, covering the entire lake but continuing our emphasis on the western arm. We have developed a web application so people can follow the glider in real time when it is deployed, and explore a database of the last six years of glider deployments.

We are placing our results on the foodweb in historical contexts. Our comparisons of current measurements with measurements of archived samples indicate that plankton-eating fish diets are now composed of primarily native species. This contrasts the period 2005-2008 when the invasive spiny waterflea (*Bythotrephes longimanus*) composed upwards of 90% of lake herring diets. The current supply of calanoid copepods, the principle prey item of lake herring, exceeds planktivorous fish demand and suggests lake herring populations are not currently energy limited.

We continue our outreach efforts. Minnesota Sea Grant worked with the Great Lakes Aquarium to provide videos for their new Great Lakes Research display; several of the looping videos involve footage, commentary and researchers related to the ENRTF grant. A public service message that aired on local networks (western region of Lake Superior) in July is archived at: https://www.youtube.com/watch?v=jOJ2QS2CQ4E&feature=youtu.be

Project status as of February 2017: The full suite of chemical and biological analyses of samples from the 2016 field season is complete. Preliminary conclusions include: observation of a long term increase in lake pH, which is attributed to a combination of factors: warming surface waters (CO_2 is more soluble in cold than warm waters); an increase in alkalinity (perhaps due to more intense chemical weathering in the watershed) which can buffer the lake; and an increase in the time per year that the lake is stratified (which can lead to higher overall algal growth, which consumes acidic carbon dioxide).

Two MS theses evaluating effects of the invasive spiny waterflea based have been completed at UMD and will be archived at the UMN Library Digital Conservancy; we expect four publications to result from this work. These revealed that spiny water flea is a poor food source for cisco (lake herring). In addition, populations of a native zooplankton (Bosmina)—an important food source for fish—have declined since invasion by spiny water fleas in ways that are consistent with it being preyed upon by spiny water flea. These results indicate significant upper foodweb consequences if spiny water flea becomes more abundant in the lake.

Data collected by the Michigan Tech group that joined our cruises during the 2014 field season to make measurements needed for calibrating satellite imagery are available at NASA's data repository (https://seabass.gsfc.nasa.gov/archive/URI/Mouw/LakeSuperior/).

Our measurements of biological productivity now extend over nine years (some warm, some cold) from 2006 to 2016. In combination with with carbon and nutrient data we can explore the differences in structure and function in warm vs. cold years. Increasing temperature generally increased rates of primary production in the upper 20 m, but decreased the abundance and mean size of organisms in the mixed layer. It appears that bursts of primary production are associated with high wind events. We might surmise from the observed year-to-year variability that a future, warmer Lake Superior will have lower biomass of smaller primary producers that nevertheless fix carbon at a higher rate. Propagating effects in the rest of the food web can be expected.

Overall Project Outcomes and Results:

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Amendment Request (02/27/2015):
Modest reallocation of funds is requested within Activity 2. Costs of satellite telephone communication are lower than anticipated, while in-state travel expenses and support for undergraduate research assistants are projected to be higher than in our initial budget. We request reallocation of $7000 originally budgeted for

satellite telephone communication, transferring $1300 to in-state travel and $5700 to support of undergraduate research assistant salaries.
Amendment Approved (03/03/2015)

Amendment Request (01/27/2016):
We request reallocation of funds to take advantage of two circumstances that have arisen. The current winter (2015-16) is projected to be among the mildest on record in the Lake Superior region, a remarkable contrast to the "polar vortex" winter of 2013-14. In addition, our expenses for technician salaries and for satellite phone communication with moored instruments have been lower than originally budgeted as we have secured funding from other sources to cover those costs. A reallocation of funds will allow us to focus on the lake's response to the unusually warm winter, obtaining data for a full open-water field season in 2016. In combination with our data from the summer of 2014, when we saw the response to the extreme cold of the previous winter, the summer 2016 data will enhance our understanding of the lake's behavior under warming conditions, broadening the impact of the ENRTF-supported work we have undertaken over the past two and a half years. We also intend to request a legislative extension to the project (through March 2017) to allow for a full 2016 field season and for analyses of 2016 samples and data. No additional costs to the project will be associated with this legislative extension.

Our request primarily reallocates funds budgeted for technician and student salaries in order provide a total of 9 days of shiptime in 2016 and as well as laboratory supplies and analyses associated with the 2016 fieldwork. In addition, we request reallocation of funds originally budgeted for laboratory analyses within Activity 3 (Previous Ecosystem Conditions) to support work associated with Activity 2 (Current Ecosystem Health), allowing us to have a greater focus on the ongoing change in the lake, while maintaining interest in records of the past. If the 9-month legislative extension is not granted, we still would like to reallocate these funds, using them in support of an intensive sampling campaign in the earliest part of summer 2016.

Specific requested changes include:
Activity 1:
We request a reduction in technician support and support for undergraduate summer assistants totaling $15,359. We request that this be applied to an increase in funds for shiptime. In combination with requested reallocation in Activity 2, this provides a total of 39 days @$8850/day, 6 additional days beyond the shiptime included in the original project.

Activity 2:
We request a reduction in support for technician and undergraduate support totaling $35,060, a decrease in travel expenses of $727, a decrease in funds for satellite telephone communication of $6000, and a decrease of $1500 in funds requested for instrument calibration. We would like to apply $36,705 of this toward additional shiptime; this provides, in combination with requested reallocation in Activity 1, 6 additional days. We would like to reallocate the remainder of these funds toward increasing funds for field and laboratory supplies by $13,826 and for laboratory analyses by $11,831. This increased need for supplies and analyses will also require reallocation of $19,075 from Activity 3.

Activity 3:
To cover additional costs of field supplies and laboratory analyses supplies needed for the expansion of work in Activity 2, we request reallocation of $9575 originally budgeted for technician and undergraduate student support, $4000 that had been budgeted for laboratory supplies, and $5500 originally budgeted for Activity 3 laboratory analyses.
Amendment Approved (05/25/2016)

Amendment Request (10/31/2016):

We request reallocation of funds because our expenses for the final field season of the project differed somewhat from our earlier projections. Much of this is due to internal UMD billing, where some analytical work was billed as salary expense rather than per-sample analytical costs. We would thus like to transfer some of the funds allocated for analytical fees and for consumables to cover salary costs. There is no change in scope of the project.

Specific requested changes include:
Activity 1:
We request movement of $753 from shiptime to equipment/tools and supplies.

Activity 2:
We request an $8835 increase in personnel expense, accomplished by transfer of $6745 from equipment/supplies and $2090 from Other/Laboratory analyses.

Activity 3:
We request and increase of $4340 in personnel costs, accomplished by transfer of $1500 from equipment/supplies and $2840 from Other/Laboratory analyses.
Amendment Approved by LCCMR (11/01/2016)

IV. PROJECT ACTIVITIES AND OUTCOMES:

Activity 1: Abundance and distribution of native and invasive species
Biological sampling (phytoplankton, zooplankton, and fish) along our transects will provide the most detailed analysis yet of the distribution and abundance of both native and invasive plankton and fish and their relation to water quality. This work will be undertaken four times along the two transects during the 2014 and 2015 summer seasons. We will use acoustic techniques (more complex versions of “fish finders”) to evaluate fish populations; these will be calibrated as needed with fish trawls. Plankton will be sampled with net tows at approximately 20 stations on each cruise. Samples of both fish and plankton are processed and preserved on shipboard for subsequent laboratory evaluation. Resulting data will allow evaluation of invasive and native species response to changing lake conditions. Observed warming may affect growth of invasive and native species. For example, lake herring often reproduce more efficiently in warm years, but the mechanism for this is unknown. Undergraduate and Graduate students will participate in this research. As a side benefit, this work will contribute to training the next generation of scientists and research technicians.

Summary Budget Information for Activity 1: **ENRTF Budget: $186,860**
Amount Spent: $186,860
Balance: $ 0

Activity 1 Completion Date:

Outcome	Completion Date	Budget
1. *Reports on newly identified invasive species*	February 2015 & 2016	$115,000
2. *Distribution maps of known invasive species*	February 2016	$35,000
3. *Report on status of native species*	February 2016	$36,860

Activity Status as of February 2014: We have been working to prepare for Lake Superior monitoring programs, and have made concrete plans for the first field season (May-October 2014).

Activity Status as of October 2014: We had a successful first field season, with plankton tows on all cruises. In addition to the ENRTF-funded field program for fish trawls, we coordinated with MN-DNR for complementary work to expand our seasonal coverage. A Graduate Student in Integrated Biological Sciences at UMD joined the project.

Activity Status as of February 2015: Nine mid-water fish trawls were completed in August and six in October 2014. Native species captured in August were lake herring Coregonus artedi, bloater Coregonus hoyi, kiyi Coregonus kiyi, and siscowet Salvelinus namaycush siscowet. Rainbow smelt Osmerus mordax was the only non-native species captured in August. Native species captured in October were similar to August with the addition of nine-spine stickleback Pungitius pungitius, lean lake trout Salvelinus namaycush, and young of year (yoy) bloater, kiyi, and lake herring. Non-native species captured in October were similar to August with the addition of sea lamprey Petromyzon marinus. Catch compositions in August and October were predominantly native species (97% and 89%, respectively) and non-native species a lower portion of the catch (3% and 11%, respectively).

Summary of current invasive species findings: Zooplankton samples of the water column were collected from multiple sites on all 2014 cruises. Specimens of the invasive crustacean, Bythotrephes longimanus (spiny waterflea), were first observed in August samples. In previous studies, Bythotrephes have been observed earlier in the season and we suspect that the unusually cold water temperatures of 2014 may be responsible for their late first detection. The highest density of Bythotrephes observed in any single collection during 2014 was in August and was 233 individuals/m2. This density is similar to densities observed by others in recent surveys of the Western Arm of Lake Superior. By October, Bythotrephes densities had declined. Based on the samples analyzed thus far, the highest density observed in October was 28 individuals/m2. This decline is anticipated since Bythotrephes are seasonal and generally reach peak densities in relation to water temperature. Interestingly, a native predatory cladoceran, Leptodora kindtii, was observed in both our July and August samples. Typically Leptodora do not successfully co-exist with Bythotrephes, so their presence in these samples is consistent with a late buildup of Bythotrephes associated with the unusually cold spring and early summer conditions of 2014.

Activity Status as of October 2015: A total of 144 zooplankton samples were collected during the 2015 LCCMR cruises. The stations visited were the same as those sampled during the summer and fall of 2014. In 2015, the invasive cladoceran, *Bythotrephes longimanus,* was first observed in May in the relatively shallow waters near the Apostle Islands. This observation is two months earlier than the first observation of *Bythotrephes* in 2014 and is likely due to warmer water temperatures brought on by a much more mild spring. *Bythotrephes* were observed on each subsequent cruise in densities up to 30 individuals/m lower than the maximum densities observed during 2014. These findings are surprising because the warmer surface temperatures on Lake Superior in 2015 compared to 2014 should have increased the growth and reproductive rates of *Bythotrephes*. However, *Bythotrephes* populations are known to cycle in many systems, and zooplankton samples were not taken in August of 2015, which is when peak *Bythotrephes* abundance was observed in 2014. Therefore, it is possible that the *Bythotrephes* densities observed do not represent the actual peak densities for 2015.

To evaluate changes in the food web and its affect on fish populations, we are using bomb calorimetry to identify caloric content of abundant fish in the Lake Superior food web. We are also conducting extensive diet studies of all abundant fish species to identify food web connections and the contribution of invasives to the current food web.

Activity Status as of February 2016: Replicate zooplankton samples were taken at 5 stations during September and October 2015 LCCMR cruises. All samples from 2015 have now been processed. Overall zooplankton biomass was higher in October of 2015 than October of 2014. The invasive zooplankton species, *Bythotrephes longimanus*, was uncommon in September and October of 2015, and *Bythotrephes* densities in October of 2015 were lower than the densities observed during October of 2014. We continue to evaluate the factors influencing year-to-year differences in *Bythotrephes* densities.

Activity status as of October 2016: Zooplankton samples were collected during the May, July, and September 2016 cruises. These samples were taken using the same protocols as 2014-2015 zooplankton collections but have yet to be processed. Zooplankton data from 2014-2015 has been processed and analyzed in a variety of ways. Notably, there was some evidence that temperature has the potential to influence the timing of peak zooplankton biomass by several weeks between years.

Fish collections made in August and October of 2014 and 2015 have been processed. Additional fish collections were made in October 2016 but have not yet been analyzed. The offshore pelagic fish community is still dominated by native species and the invasive rainbow smelt Osmersus mordax is a minor component of total fish biomass. Planktivore diets were composed of primarily native species in contrast to diet surveys from 2005-2008 where Bythotrephes composed upwards of 90% of lake herring diets. Energetics modeling suggests that the utilization of Bythotrephes as a prey resource could reduce lake herring growth. However, more work is needed to understand how the inter-seasonal and inter-annual variation in Bythotrephes abundance affects total consumption of Bythotrephes by lake herring, and to examine its consequences for lake herring growth.

Lake herring growth and body condition (energy density) in current years is similar to growth and body condition since the late 1980's and early 1990's, respectively. However, juvenile lake herring growth from 1999-2000 was reduced which correlates with high adult lake herring density and suggests a possible density dependent interaction. The current supply of calanoid copepods, the principle prey item of lake herring, exceeds planktivorous fish demand and suggests lake herring populations are no currently energy limited.

Activity status as of February 2017: For Activity 1, all data analyses and sample processing are complete. Ian Harding completed his MS in UMD's Integrated Biosciences program, and his thesis "Retrospective analysis of growth and predatory demand by cisco (Coregonus artedi) in western Lake Superior" is likely to lead to publication of two journal articles, which we will provide to LCCMR. Harding reconstructed the growth history of cisco (Coregonus artedi) collected in western Lake Superior from 1984-2013. Cisco is a very important prey of lake trout. He used historical collections of cisco as well as cisco collected under this project for analysis. To age the cisco and determine their growth rates, he measured the inner ear bones called otoliths. He tested the hypotheses that changes in water temperature and changes in the lower food web wrought by spiny water flea (Bythotrephes longimanus) changed cisco growth rates. The evidence does not support either hypothesis at this time. However, his work revealed that spiny water flea is a poor food source for cisco and this could have consequences if spiny water flea becomes more abundant in the lake.

Final Report Summary:

Taken together our overall project results show that cisco in western Lake Superior are not currently prey limited during the growing season (May-October) but that they may exert top-down control on their winter prey resource. Future studies concerning the winter ecology of cisco and calanoid copepods may improve our understanding of resource use by this important planktivore in Lake Superior.

Two manuscripts have been produced by the zooplankton and fish data sets to date. The first entitled "Changes in the cladoceran community of Lake Superior and the role of Bythotrephes longimanus)" by M. Pawlowski, D. Branstrator and T. Hrabik has been submitted to the Journal of Great Lakes Research. Another manuscript which is currently in internal review entitled "Retrospective analyses of growth and predatory demand by cisco in western Lake Superior" by I. Harding, T. Hrabik, D. Branstrator, C. Goldsworthy and B. Ray is on track for submission to a peer reviewed journal this fall.

Despite Lake Superior's size, our work demonstrates that it is not immune to impact by the invasive spiny water flea. Samples collected and analyzed under this project show that Lake Superior's lower food web was damaged by spiny water flea, and hasp responded to invasion in much the same way as other Great Lakes, and numerous small inland lakes in North America. This information provides a new baseline of understanding of species composition and relative densities in Lake Superior's zooplankton against which future conditions can be compared.

Activity 2: Snapshots of current ecosystem health
Physical, chemical and biological results from shipboard sampling, the autonomous glider, and profiling moored instruments will be used to create a comprehensive assessment of the western Lake Superior ecosystem needed to understand responses to ongoing change. The proposed work will use measurements of lake circulation and mixing to improve our understanding of distributions of fish and plankton, building on knowledge acquired through previous work supported by MN Sea Grant and the National Science Foundation.

Shipboard sampling will occur on four cruises each year during the ice-free seasons of 2014 and 2015. The cruises will occupy 12 stations along two transects across the lake, providing some 400 water samples to be analyzed for a suite of chemical and biological parameters. We will undertake measurements of primary biological production at a subset of stations. These results will be complemented by autonomous glider deployments and will be evaluated in the contexts of the data provided by the moored profilers (which will be deployed from fall 2013 to spring 2016). Undergraduate and Graduate students will participate in this research. We also will make this data available to the public through a web-site that will show real-time predictions of lake circulation conditions (e.g. winds, currents, mixing) that are important to fisherman and boaters. The National Weather Office in Duluth already uses results from LLO-operated instruments in making decisions on issuing Small Craft Advisories and Surf Zone Forecasts (issued to alert swimmers to potential rip currents); we expect to build on this ongoing collaboration.

Summary Budget Information for Activity 2: **ENRTF Budget: $374,510**
Amount Spent: $374,510
Balance: $ 0

Activity 2 Completion Date: June 2017

Outcome	**Completion Date**	**Budget**
1. *Reports on seasonal changes in the lake ecosystem*	February 2015 & 2016	$184,000
2. *Maps and data cross sections displaying results—temperatures, chlorophyll, nutrients*	February 2015 & 2016	$175,000
3. *Website displaying lake circulation predictions for public use*	May 2016	$15,510

Activity Status as of February 2014: We have been working to prepare for Lake Superior monitoring programs, and have made concrete plans for the first full field season (May-October 2014). We had a limited deployment of moored systems in Fall 2013. Technical issues (this is a cutting edge technology) made us concerned about a full over-winter deployment so we pulled the gear in for the winter rather than risk losing it. Nevertheless, this deployment provided an unprecedented record of lake conditions during the fall seasonal transition.

Activity Status as of October 2014: We undertook four sampling cruises in summer 2014, in addition to mooring deployments. Cruises involved an intensive sampling program for studies of nutrient and carbon dynamics as well as measurements of biological productivity. Thanks to funding from the Great Lakes Observing System, we expanded our proposed use of the autonomous glider to provide a total of 30 days of deployment in western Lake Superior. The very cold conditions in Lake Superior during 2014 provide a key contrast that can aid in understanding the lake's response to the generally warming conditions projected for coming years. A Michigan Tech research team joined the field program (at no cost to ENRTF) to undertake monitoring that can be used for ground-truthing satellite imagery of Lake Superior to evaluate parameters such as lake surface temperature, chlorophyll, and suspended sediment. The real-time website displays are under development.

Activity Status as of February 2015: We have undertaken chemical and biological analyses of samples collected during the 2014 field season. A major result will be examination of the effects of the unusually cold conditions that persisted in the lake though much of the summer after the extremely cold winter temperatures and exceptional ice cover of the 2013-14 winter (2013-14 ice cover was far greater than any previous year on record). While the temperature structure of the lake was unusual for most of the summer, with colder than normal conditions persisting into August, biological activity was not far from the "normal" range observed in recent years. Overall biological productivity for the season was comparable to "normal" years, but was concentrated at the very end of the season. We have made substantial progress in developing numerical modeling that will lead to real-time website displays of now-casts and forecasts of lake conditions. We anticipate that this modeling work will transition to becoming an "asset" of the Great Lakes Observing System (funded by NOAA) so that this website will be supported and maintained beyond the funding period of this project.

Activity Status as of October 2015: During the field season of 2015, samples were taken for the same package of chemical and biological parameters measured in 2014. These included carbon and nutrients in different forms as well as chlorophyll, the green pigment found in phytoplankton. These measurements will allow us to assess the year-to-year variability in the base of the food web in the Lake Superior ecosystem. Laboratory analysis of 2015 samples is still underway. One of our findings so far was the algal abundance at times in 2014 and 2015 were higher than observed in earlier studies. This suggests that the cold winter conditions of the previous two years created conditions of high nutrients or changed mixing, fostering growth of algae.

Our results also tell us about the cycles of carbon in the lake. The partial pressure of carbon dioxide in surface waters relative to average atmospheric partial pressure of carbon dioxide indicates whether the lake is a source or a sink of CO2. In spring the lake outgasses carbon dioxide. In summer, several open-lake locations become sinks of atmospheric carbon dioxide, most likely due to increasing phytoplankton growth relative to grazing/remineralization at these locations. There were more sites acting as sinks in summer 2015 (the warmer year) than in summer 2014.

As part of LCCMR field activities in 2015, we deployed a WetLABS Autonomous Underwater Profiler in the western arm of Lake Superior on 6 June, near a meteorological buoy operated by LLO. These profilers allow us to collect information on the distribution of various properties throughout the water column, such as temperature, chlorophyll content, nitrate and oxygen concentrations, and several other fields, and to do so repeatedly. The profiler achieved 160 profiles before being successfully recovered on 21 August. At the same time, a second profiler was deployed at the same location, and has made 105 more profiles. Our current plan is to recover this profiler mid-November, and redeploy another for an over-winter deployment. The data provided by these profiles represent the most highly-resolved long term time series of biogeochemical parameters ever collected in a large lake, and will help to put the shipboard sampling program in proper context.

We also deployed a glider on a single cruise in October. The glider occupied a transect between the Wisconsin south shore and the north shore of Minnesota, collecting a range of physical, chemical, and biological parameters.

We continue to make progress on a numerical model of the Lake Superior/St. Louis Estuary system working toward the goal of an online presence for the model by early next year.

Activity Status as of February 2016: We have completed lab analysis of particulate carbon, nitrogen and phosphorus as well as chlorophyll, ammonia and silica from 2015 cruises. Data have all been entered into spreadsheets. We have been modeling the uptake vs release of carbon dioxide from lake surface waters using our field data from each station on each cruise. The western arm of the lake released carbon dioxide into the atmosphere in spring and in the cooler summer of 2014. In the warmer summer of 2015, the lake shifted to taking up carbon dioxide in the western arm, presumably due to higher biological productivity in warmer water. Looking at individual stations over time, there appears to be an inverse relationship between surface water chlorophyll concentrations and pCO_2.

We made three sequential deployments of an autonomous profiler in the western arm of Lake Superior, with deployments on 5 June, 22 August, and 11 November, and recovery on 8 January. The result is a 217-day long time series of a wide variety of biologically, chemically, and physically relevant parameters, including temperature, chlorophyll content, dissolved organic matter content, dissolved oxygen concentration, nitrate concentration, the light field, and several others. This time series is unprecedented in not only the breadth of measurements but in its duration. In addition, as the profiler operation is not weather dependent, the data provide us with insight into lake conditions when it would not normally be safe to work on the deck of a boat. While our intention was to have a profiler deployed throughout the winter, power management issues on the platform necessitated an emergency recovery in early January. We are currently working with the company to determine the cause of this failure and hope to have a profiler deployed as soon as ice conditions in the harbor and on the lake allow.

We have configured a hydrodynamic numerical model of the coupled St. Louis Estuary/Duluth Harbor/Lake Superior system that runs in real-time, currently providing estimates of currents and water levels across Lake Superior. Every three hours, the model ingests recent meteorological observations from around the lake and runs the model forward to an estimate of its current state- hence the term “nowcast”. We are currently working on two fronts to expand the capability of this model platform. The first is working to add additional meteorological parameters beyond wind and barometric pressure, including air temperature, relative humidity, and cloud cover, in order to estimate surface heat fluxes that will allow us to predict water temperature distribution and evolution. The second is to provide a useful web platform for the results so that they can be widely disseminated. We will be working with colleagues at Minnesota Sea Grant to develop web tools that will be relevant and useful to users, from researchers to managers to fishermen to curious citizens. Right now we have several of these web tools in testing, but have not released them publicly.

Activity status as of October 2016: For the May, July, and September 2016 cruises, we performed CTD profiles of temperature, salinity, chlorophyll fluorescence, colored dissolved organic matter fluorescence and transmissometry as before. We performed the same organic and inorganic carbon concentration measurements as previously as well as high precision pH determinations. For the May 2016 cruise we also collected surface water samples at 5 lake stations for caffeine analyses. Caffeine is not present in native plants in the watershed and is usually removed by waste water treatment, thus it acts as a tracer for incompletely treated wastewater inputs. Mass spectrometry analysis (positive ionization electrospray, multiple reaction monitoring via Agilent 6460 triple quadrupole LC-MS) showed that there was quantifiable caffeine in the open lake (5.7 to 26.4 ng/L). The highest concentration measured was at the station closest to Duluth and Superior; the lowest concentration at our most open-lake station, midway between Isle Royale and the Apostle Islands. The concentrations measured in Lake Superior are at the lower end of reported concentrations for rivers, streams, ponds, and lakes in the US and Canada (2 to 6000 ng/L).

In addition to maintaining the carbon, chlorophyll and nutrient measurements performed in the previous two field seasons, we performed 14C primary production studies at site WM. These were performed both in 2014 and 2016 under this LCCMR funding, allowing for a contrast between a very cold summer (2014) and a warm one (2016). Preliminary analyses suggest that with increasing temperature, phytoplankton abundance is lower, average cell size is higher, and primary production is higher. These trends may help us project what to expect if climate warming trends continue into the future.

In 2016, we continued with deployments of gliders and profilers. We had a profiler deployment that lasted from April to July, so that over the last year we collected a wide variety of biogeochemically relevant data (including temperature, chlorophyll content, dissolved organic matter content, dissolved oxygen concentration, nitrate concentration, the light field, and several others) nearly around the calendar, the most complete and extensive dataset of its kind. In conjunction with the Great Lakes Observing System and the EPA's CSMI (Coordinated Science and Monitoring Initiative) program, we had a very active glider deployment year, with well over 100 deployed days, covering the entire lake but continuing our emphasis on the western arm. In addition, we have developed a web application so that people can not only follow the glider in real time when it is deployed, but explore a database of the last six years of glider deployments.

The hydrodynamic modeling effort has moved forward, with an emphasis on improving our web interface, which now has many options for exploring the real-time model data. Significantly we have recently established two collaborations with external researchers to use the model to address applied problems. In one instance, we will be working with Nathan Johnson's lab at UMD to explore flushing rates of a small embayment within the St Louis estuary to help better understand the fate of mercury compounds in the water. In the second instance, we will be working with Mike Sadowski at UMTC to better understand the fate of the WLSSD outflow and how it might impact beach closures. We have also secured funding to continue the development of this model through the Great Lakes Observing System, and expect roughly $250,000 of support over the next five years to continue to improve and expand the model.

Activity status as of February 2017: Chemical analyses of samples from the 2016 field season are complete for the full suite of nutrient analytes as in previous years. Radiocarbon-based measurements of lake productivity are complete. We completed measurement of carbon cycle parameters (inorganic carbon, dissolved organic carbon, total organic carbon, pH and alkalinity) and have a good understanding of seasonal and spatial variations in pH within the lake. The balance between carbon dioxide inputs into the lake and out-gassing from the lake depends on a host of factors including concentrations of the gas (pCO_2) in the surface water and the overlying air and the rate of transfer between these reservoirs (a function of wind speed). Surface-water pCO_2 values were found to be higher on average (lower pH, more acidic) and more variable overall in the 2014 sampling season (relative to 2015) most likely due to the harsher preceding winter. After a colder winter (in which gas exchange is limited by ice cover and water stratification), the surface-lake releases more carbon dioxide to the atmosphere during spring and early summer. A higher overall average pH occurred after the milder 2014-15 winter as compared to the previous year.

Comparison of our data with historical data sets from spring and summer sampling also indicates that over recent decades the lake's surface-water pH has risen. This initially appears counterintuitive because as atmospheric CO_2 levels increase, equilibrium should drive more CO_2, which is acidic, into the lake waters, which should lower lake pH. A combination of factors—warming surface waters (CO_2 is more soluble in cold than warm waters), an increase in alkalinity (perhaps due to more intense chemical weathering in the watershed) which can buffer the lake, and an increase in the time per year that the lake is stratified (which can lead to higher overall algal growth, which consumes acidic carbon dioxide)—appears to be driving the decreased acidity of the lake's surface waters.

Caffeine is often used as a tracer of minimally-processed wastewater inputs, so we added a pilot study to our suite of analyses. Caffeine concentrations in western Lake Superior (5-26 ng/L) were higher nearshore and closer to Duluth, MN and Superior, WI, the human population centers in far-western Lake Superior. Comparison of caffeine concentrations from western Lake Superior with other surface water systems throughout the world shows that Lake Superior has minimal wastewater contamination. However, Lake Superior caffeine concentrations are similar to measurements seen in Lake Erie and Lake Ontario, which have far higher population densities in their watersheds. This suggests that western Lake Superior may experience more contamination per person compared to the other Laurentian Great Lakes. It should be pointed out that our study is preliminary and limited in scope to western Lake Superior and the month of May. Investigating the seasonal variations in delivery and processing of caffeine in the lake as well as expanding to greater spatial coverage within the lake (including multiple depths within the water column) would be logical next steps in studying caffeine distribution and fate and the possible fate of associated contaminants.

The Michigan Tech group that joined our cruises during the 2014 field season to make measurements needed for calibrating satellite imagery recently submitted all of their Lake Superior data to NASA's data repository (https://seabass.gsfc.nasa.gov/archive/URI/Mouw/LakeSuperior/). LCCMR cruises are noted in the archive making them easy to pull out from the rest. A dataset description paper is also available: http://www.earth-syst-sci-data-discuss.net/essd-2017-10/.

Final Report Summary:

Overall results in this activity fall into three areas: Lake circulation processes; variation of lake chemistry with particular emphasis on carbon cycles; and biological productivity.

Lake circulation processes
We have developed a hydrodynamic model of lake processes building on our observations of currents and physical properties of the lake. The model has been compiled on the Minnesota Supercomputing Institute (MSI) linux cluster, which is a University of Minnesota resource. Running the model on MSI facilitates faster model runtimes. The model grid was recreated and the cartesian model projection is now well-referenced to latitude/longitude, which was an issue in the previous model build. An overview of the model was presented at the Twin Ports Freshwater Folk March meeting to people from various agencies in the Duluth/Superior region. This has helped generate interest in the model, and helped identify a few collaborators and model applications:

1) A series of model runs has been conducted to assist Michael Sadowsky's lab group at the University of Minnesota Twin Cities campus. Their lab is investigating the source of microbes in the St. Louis River Estuary, and has identified wastewater effluent from the local treatment plants as a possible source of microbes. We have used the model to help that lab group quantify the amount of wastewater effluent from these treatment plants that may have been present at their sample locations when they sampled them.

2) The Apostle Islands National Lakeshore is interested in the potential of the model to address a variety of topics within the region, including concentrations of nutrients from rivers, current dynamics, and nearshore wave action in the park. A model grid focused on the Apostle Islands region was developed, and demonstration model runs are being conducted to model the fate of water from the Bad River (a major source of nutrients in the region) and the Nemadji River following a April 2016 storm event. This will be used as a demonstration of the types of useful data the model can produce, and allow us to discuss potential ongoing applications of the model with the National Lakeshore.

3) Carol Reschke at the National Resources Research Institute is interested in the potential of the model to characterize the response of currents in the shallow regions of the St Louis River Estuary to extreme storm events. She has vegetation data from before and after the 2012 flood event, and would like to characterize bottom currents generated by such events to better understand the extent to which currents may have impacted vegetation in the estuary. This problem requires shallower region of the model than are currently being modeled, which will test the limits of the model. We are assessing whether the model can appropriately address this question, which will help us understand the practical limitations of the model.

Lake chemistry
Our datasets (pH, total inorganic carbon (TIC), and alkalinity) are providing insights on how the lake is a source or a sink of CO_2 to the atmosphere and how the lake's acidity changes as a function of location and season. These measurements were used to calculate the CO_2 content of surface water ($pCO_{2(w)}$) using the model CO2SYS and to test model performance.

Western Lake Superior's $pCO_{2(w)}$ calculations show the lake is a source of carbon dioxide to the atmosphere in spring, that there was little net gas exchange in summer, and the lake was sometimes a source of carbon dioxide in the fall. On average, $pCO_{2(w)}$ values (and thus outgassing ability) were higher in 2014 as compared to 2015 and

2016, likely because of the harsher preceding winter. Seasonally, the lake was most acidic in the spring and acidity in surface waters decreased throughout the summer, more so after a milder winter than a harsh one. In deep waters acidity increased during the stratified summer season. As water temperature and organic carbon concentrations increased, the lake became less acidic. The carbon dioxide content in surface waters showed a weak positive correlation with water temperature.

To investigate longer term trends in lake acidity and alkalinity, we compared our pH and alkalinity data with the US EPA's long-term data set (GLENDA), which is collected from Lake Superior locations twice per year. Within the GLENDA data set we again see that acidity decreases as temperature increases. Over the past 20 years, alkalinity has been increasing, and acidity has been decreasing (i.e., pH has been increasing). Our results indicate that the small increase in pH (in other words, decrease in acidity) observed in Lake Superior over that time frame cannot be explained by the observed changes in atmospheric carbon dioxide content, temperature, and alkalinity. Instead pH must also be impacted by other processes, this may be associated with a concurrent shift in the onset of water column stratification and an increase in primary production as well as the mitigating effects of reductions of acid rain resulting from clean air regulations of the 1980s and 90s.

Biological production
Our measurements of biological productivity now extend from 2006 through 2016, including a wide range of warmer and colder years. In combination with carbon and nutrient data we can explore the differences in structure and function of the biological community in warm vs. cold years. Increasing temperature generally increased rates of primary production but decreased the abundance and mean size of organisms. It appears that bursts of primary production are associated with high wind events. We might surmise from the observed year-to-year variability that a future, warmer Lake Superior will have lower biomass of smaller primary producers that nevertheless fix carbon at a higher rate. The greater abundance of smaller algae would affect dietary options for planktivores, and we expect to see these effects propagate up the food chain.

Activity 3: Evaluation of previous ecosystem conditions
Identification of historic ecosystem trends using surface sediment cores and archived samples from sediment traps. These samples were collected in the past for other studies; we will examine them with sophisticated methods (e.g. stable isotope mass spectrometry) now available in U of MN laboratories. This is a cost-effective approach because archived samples require no field costs. Anticipated results: historic arrival of invasive species (spiny water flea); past changes in fish feeding habits; past changes in nutrient and carbon cycling; comparison with meteorological data. Undergraduate and Graduate students will participate in this research. As a side benefit, this work will contribute to training the next generation of scientists and research technicians.

Summary Budget Information for Activity 3: **ENRTF Budget: $19,173**
Amount Spent: $19,173
Balance: $ 0

Activity 3 Completion Date: June 2017

Outcome	Completion Date	Budget
1. *Reports on recent ecosystem trends relative to historic records*	*February 2015 & 2016*	$15,000
2. *Distribution map of arrival of invasive species*	February 2016	$4,173

Activity Status as of February 2014: We have identified sample archives and begun to assemble materials for analysis.

Activity Status as of October 2014: We have initiated analyses of archived plankton samples.

Activity Status as of February 2015: Analyses of archived sediment trap samples are underway.

Activity Status as of October 2015: We making measurements of the caloric content of abundant fish in the Lake Superior food web. These will be compared to historic samples to identify changes resulting from food web dynamics. We are also conducting diet studies of abundant fish species to evaluate food web connections and the contribution of invasive species to the current food web. Finally, we are using fine sections of otoliths ("fish ear bones") to identify sources of variability in growth in cisco from 1986 to present.

Activity Status as of February 2016: Analyses, as described above, continue.

Activity status as of October 2016: Historic comparisons lead to the following conclusions:

- Lake herring growth and body condition (energy density) in current years is similar to growth and body condition since the late 1980's and early 1990's, respectively. However, juvenile lake herring growth from 1999-2000 was reduced, which correlates with high adult lake herring density and suggests a possible density dependent interaction.
- Planktivore diets are composed of primarily native species in contrast to diet surveys from 2005-2008 where Bythotrephes longimanus composed upwards of 90% of lake herring diets. The current supply of calanoid copepods, the principle prey item of lake herring, exceeds planktivorous fish demand and suggests lake herring populations are not currently energy limited.
- We also found evidence that cladoceran communities have changed since the 1970s. The most conspicuous change was in the relative abundance of the small cladoceran species, Bosmina longirostris, which is less common now than during the 1970s. Several lines of correlative evidence point to a possible role of top-down effects of Bythotrephes in these cladoceran community changes.

Activity status as of February 2017: A master's student in UMD's Integrated Biosciences program, Matthew Pawlowski, completed his thesis "Changes in the zooplankton community of Lake Superior and the implications of climate change and Bythotrephes longimanus". We anticipate submission to peer-reviewed journals in the coming months and will provide these to LCCMR.

Pawlowski's work tested the hypothesis that the non-native species called spiny water flea (Bythotrephes longimanus), which invaded Lake Superior in 1987, had a measurable effect on the native zooplankton. To do this he compared the zooplankton community from periods before the invasion, based on the literature, to periods after the invasion based on the literature and the extensive data we collected under this project. He found that there is currently fewer of an important native species called Bosmina in the lake and that its decline is consistent with it being eaten by spiny water flea, supporting a hypothesis of predation. This same pattern has been found in numerous small lakes in North America but his study extends this known impact to the world's largest lake. Bosmina is a food source for many native species of zooplankton and fish so its decline is concerning.

Final Report Summary:

For this Activity we assessed historical patterns in growth of lake herring. We reconstructed variability in growth histories of cisco from western Lake Superior from 1984-2013 using archived otolith (inner ear bones) samples from agency collections and combined the results with bioenergetics modeling to explore how cisco have responded to changes in their density, the invasion of spiny water flea (Bythotrephes longimanus), and climate change. We also used bioenergetics modeling and concurrent estimates of calanoid copepod standing stock and production to estimate the current supply-demand relationship for this important prey resource of cisco in Lake Superior. Cisco growth rates have been relatively stable over the 25-year period analyzed in this study with the exception of the 1998 cohort, which showed reduced growth rates that may have been a consequence of density dependence. Climate change and invasion by spiny water flea do not appear to have affected cisco growth rates to date, however, bioenergetics modeling suggests that spiny water fleas are a poor prey item for cisco relative to native prey and could reduce growth rates of cisco that consume them.

Activity 4: Public outreach
We will maximize impact of our work by building upon ongoing Minnesota Sea Grant outreach efforts. To publicize the process of conducting science and the results of this project to Minnesotans, as well as to people within the Great Lakes region, the project will provide 4 weeks/year of salary support for a Minnesota Sea Grant outreach specialist. These efforts will include press releases, regular updates through social media outlets, and facilitation of newspaper, TV and radio interviews involving project investigators. Sea Grant will also produce short videos and other new media. Sea Grant will publish two newsletter stories about the work.

Summary Budget Information for Activity 4: **ENRTF Budget: $19,457**
Amount Spent: $19,457
Balance: $ 0

Activity 4 Completion Date: June 2017

Outcome	Completion Date	Budget
1. *Ongoing outreach and publicity in collaboration with Minnesota Sea Grant*	June 2016	$19,457

Activity Status as of February 2014: We are in design and construction phases of a new interactive display at the Great Lakes Aquarium (supported by a number of entities including LCCMR) that enables the public to investigate how Lake Superior internal waves respond to changing wind conditions. Minnesota SeaGrant published a short article that described the Fall 2013 moored instrument deployment (http://www.seagrant.umn.edu/newsletter/2014/02).

Activity Status as of October 2014: A prototype of Great Lakes Aquarium display has been completed, and we continue to have periodic meetings with Aquarium staff to discuss new initiatives to highlight Lake Superior research. Richard Ricketts made a shipboard presentation about our Lake Superior project as part of LLO's "Science Friday" public outreach campaign (http://www.d.umn.edu/llo/sciencefriday.html). UMD's External Affairs Office, released an article describing the summer 2014 field season (http://www.d.umn.edu/external-affairs/homepage/14/llo-lake-ecology.html). Minnesota Sea Grant produced a "Sea Grant Files" radio interview with Robert Sterner (to be broadcast on 4 November) that discusses research on Lake Superior productivity and carbon cycling. A second interview on the implications of observations from the 2014 field season is scheduled for broadcast in mid-December. "Sea Grant Files" programs are archived online and are available as podcasts at (http://www.seagrant.umn.edu/radio/sgf/)

Activity Status as of February 2015: The Great Lakes Aquarium display is moving forward, and MN SeaGrant outreach personnel are working with UMD faculty and GLA staff on logistics and planning issues needed for installation. We continue to have good interaction with Aquarium staff to find ways to publicize Lake Superior research. UMD's External Affairs Office released an article highlighting one of LLO's marine technicians and his work on Lake Superior, as well as other systems (http://duluth.umn.edu/external-affairs/homepage/15/agnich.html). A recent Minnesota Public Radio "Updraft Blog" focuses on a report from Jay Austin showing how small shifts in winter temperatures can lead to major changes in the amount of ice cover on Lake Superior (http://blogs.mprnews.org/updraft/2015/02/lake-superior-ice-sensitive-to-small-climate-shifts/). Minnesota Sea Grant produced a "Sea Grant Files" radio interview with Elizabeth Minor, broadcast on 16 December, presenting results from the LCCMR project. Another episode, focusing on aspects of Lake Superior biology, was broadcast on 24 February. "Sea Grant Files" programs are archived online and are available as podcasts at (http://www.seagrant.umn.edu/radio/sgf/). Social media posts related to this project have reached a potential audience of over 27,000 across the U.S. and, more importantly, engaged a minimum of 40 users:

https://twitter.com/MNSeaGrant/status/529672426594902016
https://twitter.com/MNSeaGrant/status/564816542123061249
https://www.facebook.com/mnseagrant/posts/797951886907313

https://twitter.com/MNSeaGrant/status/532635127952596992
https://www.facebook.com/mnseagrant/posts/753792304656605

Activity Status as of October 2015: MN Sea Grant produced a newsletter story about the project that went to 3000 subscribers. The article is available online at www.seagrant.umn.edu/newsletter/2015/06/from_hot_to_cold.html. Sea Grant also worked with a local news station (Northland Newscenter) to produce two "Lake Superior Basin Basics" public service messages that involved aspects of this research (fish and stratification). The 2-minute messages aired in September and are available through the MN Sea Grant website and YouTube channel (https://www.youtube.com/watch?v=TQeG4t3fvyU and https://www.youtube.com/watch?v=tf5tdb6Xr_w). Team members met to discuss the Great Lakes Aquarium installation. The artwork was finalized and the computer equipment was purchased through a public engagement grant from the University of Minnesota Duluth. Social media related to this work is ongoing.

Activity Status as of February 2016: During this activity period, communication efforts supporting this project have relied on:

- Social media (about 100,000 potential impressions via 20 posts),
- Two public service messages that played in September on a Duluth, MN, network are on YouTube https://www.youtube.com/watch?v=TQeG4t3fvyU;https://www.youtube.com/watch?v=tf5tdb6Xr_w
- An audio program that aired in December on a public radio station, via podcast and on the MN Sea Grant website (What Ice (or Lack Thereof) Means for Ecological Processes Underwater (http://www.seagrant.umn.edu/radio/sgf/)).

Activity status as of October 2016: Since Feb 2016, communication efforts supporting this project have relied on social media and a public service message that aired on local networks (western region of Lake Superior) in July and that is archived at: https://www.youtube.com/watch?v=jOJ2QS2CQ4E&feature=youtu.be. Minnesota Sea Grant worked with the Great Lakes Aquarium to provide videos for their new Great Lakes Research display; several of the looping videos involve footage, commentary and researchers related to the ENRTF grant. We continue to meet with Aquarium staff to discuss new initiatives to highlight Lake Superior research. Additionally, the team worked on developing a website that can deliver buoy data and related promotional material. A video summarizing this ENRTF project is being developed.

Activity status as of February 2017: During the October 2016-February 2017 period, Minnesota Sea Grant conducted video interviews with two project researchers, scripted and recorded a narrator voice-over, shot additional footage, and gathered drone footage and project images. These materials are being packaged into a 10-minute video about the research and its results. The video should be available online by May 2017.

Final Report Summary:

As noted throughout this section, we have used multiple forms of media to communicate with the public over the course of the project. In addition to those reported above for earlier periods we have recently (June 2017) released a crisp 8-minute video that features students, drone footage, with students and researcher interviews to showcase how LCCMR funding yielded new information about Lake Superior.

https://www.youtube.com/watch?v=iRmG8xIYtY8&feature=youtu.be

V. DISSEMINATION:

Description:
As described above, we will coordinate public outreach with ongoing Minnesota Sea Grant efforts. One highlight will be development of a real-time model of lake circulation processes (currents, winds, mixing) that will be available on the Internet. This will be a valuable resource for fishermen, boaters, and the interested public. We also intend to publish significant results in peer-reviewed journals to reach the broader scientific community.

Status as of February 2014: As noted above we are working on a new interactive display at the Great Lakes Aquarium and Minnesota SeaGrant published a short article that described the Fall 2013 moored instrument deployment (http://www.seagrant.umn.edu/newsletter/2014/02).

Status as of October 2014: As noted above a prototype interactive display has been completed in cooperation with the Great Lakes Aquarium. UMD's External Affairs Office released a description of the summer 2014 field season (http://www.d.umn.edu/external-affairs/homepage/14/llo-lake-ecology.html). Two MN Sea Grant radio shows (archived as podcasts) highlight results of the project. The real-time website displays are under development.

Status as of February 2015: The Great Lakes Aquarium display is moving forward, and MN SeaGrant outreach personnel are working with UMD faculty and GLA personnel on logistics and planning issues needed for its installation. An article on the effects of the 2012 Duluth Flood on western Lake Superior has been published: Minor, E.C., B. Forsman, and S.J. Guildford, 2014. The effect of a flood pulse on the water column of western Lake Superior, USA. Journal of Great Lakes Research, volume 40: 455-462. We have made substantial progress in developing numerical modeling that will lead to real-time website displays of now-casts and forecasts of lake conditions.

Status as of October 2015: LLO has continued and expanded its "Science on Deck" program that invite the public to visit the r/v Blue Heron and include presentations on scientific topics. A total of nearly 1000 visitors attended 2015 "Science on Deck" events that were held in Duluth (next to the Duluth Entertainment and Convention Center) on May 29th, June 26th, July 31st, August 21st and September 18th and in Two Harbors on July 11th. Minnesota Sea Grant's newsletter included results of LCCMR sponsored work (www.seagrant.umn.edu/newsletter/2015/06/from_hot_to_cold.html). Sea Grant also worked with a local television station (Northland Newscenter) to produce two short "Lake Superior Basin Basics" public service messages that were aired in September and are available on youtube: (https://www.youtube.com/watch?v=TQeG4t3fvyU and https://www.youtube.com/watch?v=tf5tdb6Xr_w).

Status as of February 2016: An audio program aired in December on a public radio station, via podcast and on the MN Sea Grant website (What Ice (or Lack Thereof) Means for Ecological Processes Underwater (http://www.seagrant.umn.edu/radio/sgf/)). Minneosota SeaGrant continues to provide a social media presence for the project with about 100,000 potential impressions via 20 posts.

Status as of October 2016: The popular "Science on Deck" program, which invites the public to visit the r/v Blue Heron and includes presentations on scientific topics, continued in summer 2016. A total of nearly 600 visitors attended events that in Duluth (next to the Duluth Entertainment and Convention Center) on May 27, July 22, and August 26. A fourth event, scheduled for September 23, was cancelled because heavy winds had the potential to lead to unsafe conditions. We continue to meet with Great Lakes Aquarium staff to discuss new initiatives to highlight Lake Superior research. In addition, we have developed a web application so that people can not only follow the autonomous glider in real time when it is deployed, but explore a database of the last six years of glider deployments.

Status as of February 2017: MS Theses based on research supported by this project are available at the UMN Library Digital Conservancy (https://conservancy.umn.edu/handle/11299/53656). These include:

“Analysis of Inorganic Carbon and pH in the Western Arm of Lake Superior” Tennant, Cody (2016);
"Retrospective analysis of growth and predatory demand by cisco (Coregonus artedi) in western Lake Superior" Harding, Ian (2016);
"Changes in the zooplankton community of Lake Superior and the implications of climate change and Bythotrephes longimanus" Pawlowski, Matthew (2016).

Final Report Summary:

We have disseminated results from this project through multiple forms of media, through public outreach events and through scientific literature. These efforts are summarized here:

Public outreach

1. Project summary video released by Minnesota Sea Grant in 2017
   https://www.youtube.com/watch?v=iRmG8xIYtY8&feature=youtu.be

2. Minnesota SeaGrant was a key player in the public outreach efforts of this project, producing media pieces available in multiple formats:
   a. Radio (available as podcast),
      http://www.seagrant.umn.edu/audio/2014.12.16_lizminor.mp3
      http://www.seagrant.umn.edu/audio/2014.11.04_bobsterner.mp3
      http://www.seagrant.umn.edu/audio/2015.02.24_creatures.mp3
      http://www.seagrant.umn.edu/audio/2015.12.22_winterIce.mp3
   b. Televised public service announcements (available on youtube),
      https://www.youtube.com/watch?v=TQeG4t3fvyU
      https://www.youtube.com/watch?v=tf5tdb6Xr_w
      https://www.youtube.com/watch?v=jOJ2QS2CQ4E&feature=youtu.be
   c. Newsletter articles in print and online
      http://www.seagrant.umn.edu/newsletter/2014/02/a_hard_days_and_nights_work_aboard_the_rv_blue_heron.html
      http://www.seagrant.umn.edu/newsletter/2015/06/from_hot_to_cold.html

3. UMD University Public Relations and Media pieces
   http://duluth.umn.edu/external-affairs/homepage/15/agnich.html
   http://www.d.umn.edu/external-affairs/homepage/14/llo-lake-ecology.html

4. Science On Deck. LLO continues its popular “Science on Deck” outreach events. In summers these monthly open house events attract hundreds of people to visit UMD’s research vessel Blue Heron and hear about ongoing projects, including those supported by LCCMR.

5. Great Lakes Aquarium exhibit. We developed an interactive exhibit in collaboration with the Great Lakes Aquarium in Duluth

6. Media reports on Lake Superior research efforts building on LCCMR support.
   http://www.duluthnewstribune.com/news/education/4142404-gliders-provide-depth-scientific-data-lake-superior
   http://www.fox21online.com/2017/05/26/umd-scientists-invite-area-students-visit-research-vessel/
   http://greatlakesecho.org/2016/11/02/peering-beneath-great-lakes-ice/
   https://www.outsideonline.com/2183826/superiority-complex
   http://www.wpr.org/lake-superior-researchers-studying-what-ice-bode-fish-commerce

http://www.twincities.com/2016/09/11/researchers-say-lake-superior-water-temperature-on-the-rise/
http://blogs.mprnews.org/updraft/2015/02/lake-superior-ice-sensitive-to-small-climate-shifts/

Scientific dissemination

1. *Journal articles to date*

   "The effect of a flood pulse on the water column of western Lake Superior, USA" E.C. Minor, E.C., Forsman, and S.J. Guildford. Journal of Great Lakes Research, volume 40: 455-462 (2014)

   "Changes in the cladoceran community of Lake Superior and the role of Bythotrephes longimanus)" M. Pawlowski, D. Branstrator and T. Hrabik in review at to the Journal of Great Lakes Research.

   "Retrospective analyses of growth and predatory demand by cisco in western Lake Superior" I. Harding, T. Hrabik, D. Branstrator, C. Goldsworthy and B. Ray to be submitted to the Journal of Great Lakes Research.

2. *Theses archived at the UMN Library Digital Conservancy (https://conservancy.umn.edu/handle/11299/53656)*

   "Analysis of Inorganic Carbon and pH in the Western Arm of Lake Superior" Cody Tennant (2016)

   "Quantification of Caffeine as an Anthropogenic Marker in Western Lake Superior" Kaila Hanson (2016)

   "Retrospective analysis of growth and predatory demand by cisco (Coregonus artedi) in western Lake Superior" Ian Harding (2016)

   "Changes in the zooplankton community of Lake Superior and the implications of climate change and Bythotrephes longimanus" Matthew Pawlowski (2016)

VI. PROJECT BUDGET SUMMARY:

A. ENRTF Budget:

Budget Category	$ Amount	Explanation
Personnel:		
Research technicians	$57,011	Skilled field and laboratory technicians are needed to undertake the research program, under the direction of project scientists.
Outreach specialist	$19,457	MN Sea Grant outreach specialists will aid in providing information on the results of this project to the general public.
Graduate students	$89,353	Support for 5 graduate students working on this project during summer months and to 1 graduate student for 1 academic year. Teaching assistantships or other research grants will provide academic year salary for students with summer support only from this project. Support from the LCCMR will be critical for their training to become scientists or research technicians.
Undergraduate students	$15,094	Support for 2 undergraduate students working on this project during summer months. This will provide hands-on educational experiences for the next generation of scientists and technicians.
Equipment/Tools/Supplies:		
Plankton nets and endcups	$3500	Needed for plankton surveys
Mooring hardware and expendable supplies	$3500	Supplies (shackles, batteries, anchors, cable, etc) necessary for deployment of moored profiler
Glider spare parts and supplies	$3500	Parts, supplies, and maintenance needed for autonomous glider deployment
Laboratory and field supplies	$22,240	Bottles, filters, plastic ware needed for collecting, processing and storing samples for the full range of proposed analyses.
Chemical reagents	$4000	Chemical reagents and standards needed for processing and analyzing samples.
Radiocarbon tracer	$2000	Needed for measurement of biological productivity in the lake
Travel Expenses in MN:	$1773	Mileage for UMTC employees to travel to Duluth for fieldwork
Other:		
Shiptime 39 days.	$344,397	Day rate of $8850 covers costs of fuel, crew salaries, insurance, basic maintenance, meals.
Laboratory analyses	$31,401	800 water samples analyzed for organic carbon, organic nitrogen, nutrients, pH, photosynthetic pigments, stable isotopes of C and N.
Satellite telephone	$1000	Monthly fees and data transfer charges for Iridium Satellite telephone service needed to control (and to transfer data from) unmanned

		instruments deployed in the open lake from fall 2013 through 2016.
TOTAL ENRTF BUDGET:	**$600,000**	

Explanation of Use of Classified Staff:
N/A
Explanation of Capital Expenditures Greater Than $3,500:
N/A

Number of Full-time Equivalent (FTE) funded with this ENRTF appropriation:

Technicians and the outreach specialist: 0.3 FTE per year.
Three-year total: 0.92FTE.

Graduate student support: 8 weeks of summer support for 5 students (0.77 FTE each year), plus one academic year of support (39 weeks) for 1 student in Year 2 (0.75 FTE).
Three-year total: 3.28 FTE

Undergraduate summer student support: part time support for 2 students (0.26 FTE each year).
Three-year total: 0.51 FTE

Ship's crew during LCCMR-paid cruise dates: 39 days x 5 crew x (12 hours/day)/(2080 hours/FTE)
Three-year total: 1.13 FTE

Overall three-year total: 5.84FTE

Number of Full-time Equivalent (FTE) estimated to be funded through contracts with this ENRTF appropriation:
N/A

B. Other Funds:

Source of Funds	$ Amount Proposed	$ Amount Spent	Use of Other Funds
Non-state			
UMD, Large Lakes Observatory	$106,200	$	*12 days of Blue Heron shiptime*
UMD, Large Lakes Observatory	$3000	$	Cost of small boat rentals for glider operations
NSF grant	$485,000	$485,000	*Purchase of moored profiling instruments*
TOTAL OTHER FUNDS:	**$594,200**	**$485,000**	

U of MN scientists associated with this project are *not* requesting salary support, and are providing their expertise—project design, supervising technicians, and advising students—*at no cost to LCCMR*.

VII. PROJECT STRATEGY:

A. Project Partners:

The following scientists and are providing time and expertise in the areas noted, but are are *not* requesting Trust Fund support for salaries:

U of M Duluth Large Lakes Observatory:

Erik Brown (project management, carbon & nutrient cycling)
Jay Austin (physical processes, moored and autonomous instruments)
Elizabeth Minor (biochemistry, carbon cycling)

Richard Ricketts (ship operations and logistics)

U of M Duluth Department of Biology:

Donn Branstrator (zooplankton ecology)

Tom Hrabik (fish ecology)

U of M Duluth MN SeaGrant:

Jeff Gunderson (public outreach)

U of M Twin Cities Department of Ecology, Evolution and Behavior:

Robert Sterner (biological productivity; nutrient distributions, data management)

Providing services at no cost:

DNR Duluth Office:

Don Schreiner (fish population dynamics); $0

B. Project Impact and Long-term Strategy:

The ecosystem of Lake Superior is unquestionably changing, due to human activities, invasive species, and long-term warming, but little is being done to monitor those changes. In particular, no baseline exists from which to measure future changes.

The proposed project will fill a gap left by a spectrum of scientific and regulatory agencies. At the Federal level, EPA participates in the bi-national Lakewide Management Plan (LaMP), but this effort involves little data collection. An interagency initiative called the "Coordinated Science Monitoring Initiative" (CSMI) is also underway, but again this does not involve the level of sampling (in space and time) that is truly needed to understand the lake. On the State side, the Department of Natural Resources (DNR) conducts a Coastal Program on Lake Superior, but this program focuses on the landward side of the coastline. DNR also conducts limited small-boat fish surveys; we continue to have good working relations with the DNR, particularly the Duluth Office, in this regard.

The researchers involved in this project have a strong history of support from the National Science Foundation and MN Sea Grant. These agencies fund work that involves specific scientific hypotheses and questions, along with constrained sampling programs to address those specific questions. NSF does not fund data collection or monitoring efforts that are not intimately tied to such topical scientific questions. Despite this, there are urgent applied and scientific reasons to extend the topical NSF studies to repeated sampling and transects studies. In particular, the spatial and seasonal variation of processes and properties in the lake need to be characterized. It is our expectation that results of this LCCMR sponsored work will be used as seed data for additional external funding.

C. Spending History:

Funding Source	**M.L. 2007 or FY08**	**M.L. 2008 or FY09**	**M.L. 2009 or FY10**	**M.L. 2010 or FY11**	**M.L. 2011 or FY12-13**
NSF grant. Equipment (Austin PI)*				485,000	
NSF grant. Radiocarbon (Minor PI)*	160,000	160,000	160,000		
NSF grant. Carbon and Ice (Austin PI)*	200,000	200,000	200,000		
SeaGrant. Fish migration (Hrabik PI)*	50,000	50,000			
NOAA Great Lakes Observing System (Austin PI)			120,000	120,000	120,000

GLPA (Colman PI)	86,000 subd. 4(i)				

*These projects also provide for shiptime, not part of these research budgets, totalling approximately 30 days per year.

VIII. ACQUISITION/RESTORATION LIST:
N/A

IX. MAP(S):
See attachment with schematic map of our field strategies.

X. RESEARCH ADDENDUM:
See attachment.

XI. REPORTING REQUIREMENTS:
Periodic work plan status update reports will be submitted not later than February 2014, October 2014, February 2015, October 2015, February 2016, October 2016 and February 2017. A final report and associated products will be submitted between June 30 and August 15, 2017 as requested by the LCCMR.

Attachment A: Budget Detail for M.L. 2013 Environment and Natural Resources Trust Fund Projects

Project Title: Evaluating Lake Superior's Health in a Changing World
Legal Citation: M.L. 2013, Chp. 52, Sec. 2, Subd. 05f; M.L. 2016, Chapter 186, Section 2, Subdivision 18
Project Manager: Erik Brown
M.L. 2013 ENRTF Appropriation: $ 600,000
Project Length and Completion Date: 1 July 2013 to 30 June 2017
Date of Update: 28 February 2017

ENVIRONMENT AND NATURAL RESOURCES TRUST FUND BUDGET	Activity 1 Budget	Amount Spent	Balance	Activity 2 Budget	Amount Spent	Balance	Activity 3 Budget	Amount Spent	Balance	Activity 4 Budget	Amount Spent	Balance	TOTAL BUDGET	TOTAL BALANCE
BUDGET ITEM	*Abundance and distribution of native and invasive species*			*Snapshots of current ecosystem health*			*Evaluation of previous ecosystem conditions*			*Public outreach*				
Personnel: Total annual compensation (%Wages and %Benefits)	49,181	49,181	0	95,264	95,264	0	17,013	17,013	0	19,457	19,457	0	180,915	0
S. Grossheusch, Chemistry technician: $60,360 (71.6% salary; 28.4% benefits); 5.2% FTE														
S. Brovold, Biology technician: $72,229 (71.6% salary; 28.4% benefits); 9.0% FTE														
M. James, Physics technician and programmer: $76,222 (71.6% salary; 28.4% benefits); 5.0% FTE														
S. Moen, Outreach specialist: $81,031 (71.6% salary; 28.4% benefits); 7.7% FTE														
Graduate Research Assistant Academic Year: $36,748 (49.2% salary; 50.8% benefits that include a tuition fellowship of $13,120); 1 student at 75% FTE in year 2														
Graduate Research Assistant Summer; $24,167 (80.5% salary, 19.5% benefits); 5 students at 15.4% FTE														
Undergraduate Assistant summer; $22,329 (93.2% salary, 6.8% benefits); 2 students at 8.5% FTE														
Equipment/Tools/Supplies	5,217	5,217	0	34,797	34,797	0	500	500	0				40,514	0
Puget Sound-style zooplankton net ($2,500)														
End buckets for plankton net ($1,000)														
Mooring hardware and expendables (batteries, anchors, cables, shackles, etc) ($3,500)														
Expendables and supplies for Autonomous Glider ($5,216)														
Radiocarbon tracer ($3,000)														
Consumable lab and field supplies and chemical reagents ($32,790)														
Travel expenses in Minnesota														
Mileage for U of M Twin Cities employees to join Duluth-based field programs				1,773	1,773	0							1,773	0
Other														
Shiptime: 39 days. Day rate=$8850. Covers cost of fuel, crew salaries, insurance, basic maintenance, meals.	132,462	132,462	0	211,935	211,935	0							344,397	0
Laboratory analyses: 800 water samples analyzed for organic carbon,organic nitrogen, nutrients, pH, photosynthetic pigments, stable isotopes of C and N.				29,741	29,741	0	1,660	1,660	0				31,401	0
Satellite telephone links for instrument control and data transfer of the autonomous glider and the moored profilers				1,000	1,000	0							1,000	0
COLUMN TOTAL	**$186,860**	**$186,460**	**$0**	**$374,510**	**$372,166**	**$0**	**$19,173**	**$19,173**	**$0**	**$19,457**	**$19,457**	**$0**	**$600,000**	**$0**

Changes in the cladoceran community of Lake Superior and the role of *Bythotrephes longimanus*

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Abstract

Introductions of *Bythotrephes longimanus* have resulted in reduced cladoceran species richness and biomass in the Laurentian Great Lakes and many inland lakes. *Bythotrephes* was first observed in Lake Superior in 1987 but its effect on the cladoceran community has been unknown. We compared the composition of the offshore cladoceran community of Western Lake Superior during 2014 and 2015 to zooplankton surveys from 1971-2001 to determine whether changes in the cladoceran community have occurred. Monthly comparisons show that the contribution of *Bosmina longirostris* to offshore cladoceran numbers was generally twice as much in the 1970s than during 2014-2015 while the relative contribution of *Daphnia mendotae* increased after the 1970s. These community changes are consistent with changes due to *Bythotrephes* observed in other lakes. To evaluate evidence for the role of *Bythotrephes* in these community changes, we used data from 2014-2015 to analyze patterns in spatial and vertical overlap between *Bythotrephes* and its cladoceran prey species (*Bosmina*, *Daphnia*, and *Holopedium*) and compared estimates of consumption by *Bythotrephes* to production of these potential prey. *Bosmina* was the species whose vertical position and rate of production made it most vulnerable to suppression by *Bythotrephes*. Of the potential cladoceran prey species, *Bosmina* densities were also the most negatively correlated with *Bythotrephes* densities. These findings support a hypothesis of top-down effects on *Bosmina* by *Bythotrephes* in Lake Superior. This work informs future zooplankton research in Lake Superior and furthers our understanding of the effects of *Bythotrephes* on the Lake Superior food web.

Keywords: *Bythotrephes*, Lake Superior, Zooplankton, Great Lakes, Invasive species, *Daphnia*

Introduction

Species introductions in aquatic ecosystems can alter the abundance and community composition of zooplankton (Brooks and Dodson, 1965; Carpenter et al., 1987). In North America, introductions of *Bythotrephes longimanus*, a predatory cladoceran native to northern Europe and Asia (Lehman, 1987; Burkhardt and Lehman, 1994), have resulted in reduced zooplankton species richness (particularly in cladocerans) and abundance in small inland lakes and in the Great Lakes (Barbiero and Tuchman, 2004; Strecker et al., 2006; Azan et al., 2015). *Bythotrephes* selects slow-moving cladocerans such as *Bosmina longirostris* and *Daphnia spp.* (Vanderploeg et al., 1993; Grigorovich et al., 1998; Schulz and Yurista 1999). *Bosmina* and *Daphnia* consistently become less abundant in lakes following *Bythotrephes* invasion and multiple studies suggest that this is a direct result of consumption by *Bythotrephes* (Yan and Pawson, 1997; Yan et al., 2002; Strecker et al., 2006; Kerfoot et al., 2016).

Rapid cladoceran community changes in the Great Lakes following *Bythotrephes* invasion occurred in Lake Michigan in the late 1980s, when two of three common *Daphnia* species nearly disappeared within a year of the first detection of *Bythotrephes* (Lehman, 1988; Lehman, 1991; Lehman and Cáceres, 1993). *Daphnia mendotae* has remained common in Lake Michigan since *Bythotrephes* establishment, but appears to have done so partly by reducing the extent of its vertical overlap with *Bythotrephes* (Pangle and Peacor, 2006; Pangle et al., 2007). Other small cladocerans such as *Bosmina* have also become less common in Lake Michigan since the establishment of *Bythotrephes* (Makarewicz et al., 1995; Schulz and Yurista, 1999). Similar changes have been described in the cladoceran communities of Lakes Huron and Erie (Barbiero and Tuchman, 2004; Bunnell et al., 2012). In Lake Ontario, *Bosmina longirostris* and *Eubosmina spp*. abundance has declined by more than half since 2003 (Barbiero et al., 2014; Rudstam et al., 2015). These changes coincided with an order of magnitude increase in *Bythotrephes* abundance after 2003 which suggests that *Bythotrephes* has exerted top-down control on bosminids in Lake Ontario.

Bythotrephes was first detected in Lake Superior, the largest lake on Earth by surface area, in 1987 (Cullis and Johnson, 1988), but its effect on the zooplankton community is largely unknown. The Lake Superior zooplankton community is calanoid-dominated in terms of density and biomass (Patalas, 1972; Barbiero et al., 2001). Historically, the cladoceran community in Lake Superior was dominated by three species including *Bosmina longirostris*, *Daphnia mendotae*, and *Holopedium gibberum* (hereafter referred to by genus, unless otherwise noted). Brown and Branstrator (2004) reported a lower abundance of *Bosmina* in August of 2001 compared to observations from the early 1970s. Though reductions in *Bosmina* abundance are consistent with planktivory by *Bythotrephes*, the authors could not clearly demonstrate such a relationship given the temporal limitations of their dataset. Zooplankton surveys spanning multiple sampling seasons are needed to determine whether these or other community changes have occurred in the decades since *Bythotrephes* establishment.

Previous studies in Lakes Michigan and Huron have suggested that consumption by *Bythotrephes* can exceed production of cladocerans (Lehman and Cáceres, 1993; Bunnell et al., 2011; Bunnell et al., 2012). Consumption by *Bythotrephes* has not yet been estimated in Lake Superior. *Bythotrephes* densities in Lake Superior are generally lower than in Lakes Michigan and Huron (Barbiero et al., 2001; Brown and Branstrator, 2004; Brown et al., 2012; Pothoven et al., 2012; Bunnell et al., 2014). However, Lake Superior is also colder and less productive than the other Great Lakes (Patalas 1972) and the consumptive demands of *Bythotrephes* in Lake Superior may still approach the rate of production of their preferred prey species. Cladoceran production is subject to a variety of losses other than by invertebrate predation. Thus, increased mortality due to *Bythotrephes* predation might be enough to reduce cladoceran abundance even if consumption by *Bythotrephes* does not exceed cladoceran production.

The addition of *Bythotrephes* to the Lake Superior food web is not the only important change that has occurred in this ecosystem in recent decades. Since the 1970s, population densities of planktivorous fish have fluctuated in Lake Superior and average summer surface temperatures have increased (Austin and Colman, 2007; Gorman, 2012; Pratt et al., 2016). One key change in the planktivorous fish community since the 1970s has been an overall increase in lake herring (*Coregonus artedi*) densities. Increasing vertebrate planktivory would be expected to cause a downward shift in the average body size of zooplankton (Brooks and Dodson, 1965). Temperature is a key factor driving zooplankton production and spatial aggregation in Lake Superior and increases in surface temperature might increase zooplankton production and density (Watson and Wilson, 1978; Zhou et al., 2001). In addition, the increase in summer surface temperatures in Lake Superior since the 1970s has the potential to favor warm water taxa such as small cladocerans (Lehman, 2002). Though zooplankton abundance and production can also be influenced by changes in primary production, changes in food quality and quantity for herbivorous zooplankton are unknown over the period of *Bythotrephes* invasion. However, changes in food availability would be expected to have similar effects on the densities of all herbivorous zooplankton rather than effects on specific taxa. Therefore, while changes in temperature, vertebrate planktivory, and primary production all can cause changes in zooplankton community structure, the effects of these ecosystem changes on the cladoceran community should be distinguishable from top-down effects by *Bythotrephes*.

The purpose of this study is to determine whether long-term changes in the cladoceran community of Lake Superior have occurred since the introduction of *Bythotrephes*. Based on changes observed in the cladoceran communities of the other Great Lakes and smaller, inland lakes following *Bythotrephes* invasion, we hypothesized that small cladocerans such as *Bosmina* would be less common in offshore areas of Lake Superior than before *Bythotrephes* invasion. To test this, we compared the offshore cladoceran community observed during the 2014 and 2015 growing seasons to past zooplankton surveys. Further, we used the data from 2014 and 2015 to evaluate three lines of evidence that *Bosmina*, *Daphnia*, and *Holopedium* populations are currently negatively impacted by the presence of *Bythotrephes*. These lines of evidence were: 1) the extent of synchronous spatial overlap among the prey taxa and *Bythotrephes*, 2) patterns in vertical position of the prey taxa relative to *Bythotrephes*, and 3) the difference in temperature-driven production of the prey taxa versus temperature-driven consumptive demands of *Bythotrephes*.

Methods

Sampling sites and dates

Zooplankton and water temperature data were collected in the western arm of Lake Superior (Fig. 1) from the R/V Blue Heron. In 2014, collections occurred on June 3-6, July 23-25, August 11-14, August 17-19, October 1-2, and October 16-19; in 2015 collections occurred on May 20-22, July 15-17, September 8-10, October 5-7, and October 16-19. The number of stations visited in each month is indicated in Table 2. Stations 5, 12, and 15 were sampled in every month and station 7 was sampled in every month except July and October of 2014 (Fig. 1). The remaining stations were sampled on only one or two occasions. Zooplankton samples were collected primarily during low light conditions (between dusk and dawn) although some samples were collected during daylight hours. Additional archived zooplankton samples collected at station 6 were available from the summer of 1996 (See Table 1 for details). All stations visited were greater than 70 m in depth and the depths and coordinates of all stations are described in Appendix A.

Figure 1 here

Zooplankton collection

Zooplankton were collected using a conical plankton net with a mouth opening of 1-meter diameter, 153 µm mesh, and a 4:1 aspect ratio (length to opening). A metered winch was used to collect vertical tows to depths of 60 m and 15 m at each site to study both total zooplankton and the shallow dwelling taxa. Recent studies have shown that more than 90% of the zooplankton biomass in Lake Superior exists at depths less than 50 m (Oliver et al., 2014; Pratt et al., 2016). At all stations visited after July of 2014, time allowed for triplicate samples to be taken at both depths to increase accuracy in zooplankton density estimates. Zooplankton samples were preserved in 70% ethanol (final concentration). In 2015, nets were equipped with a RBR TWR-2050 pressure gauge to verify that nets reached target depths. The gauge was secured to the steel bridle at the mouth opening of the net and was sensitive to changes of 0.0001 dbar. Pressures were converted to depth according to Sea-Bird Electronics, Inc. (2002) as:

$$depth\ (m) = pressure\ (dbar) \times 1.0197 \qquad \text{(Eq. 1)}$$

Zooplankton processing

Specimens were sorted, counted, and identified under a Nikon SMZ 1500 dissecting microscope. Identification was done to species level according to Balcer et al. (1984). All *Bythotrephes* and *Leptodora kindtii* were removed with forceps and counted in full. Replicate subsamples were taken until approximately 100 individuals of each taxon were counted. For less abundant taxa, larger subsample volumes were used and counting stopped when adequate replication of individuals per subsample was achieved or 10% of the sample volume was counted. The lengths of the first ten individuals in each taxon were measured using the ocular micrometer. The lengths of all *Bythotrephes* and *Leptodora* were measured using the method in Branstrator (2005). For *Bythotrephes*, the number of barbs on the caudal spine (indicator of developmental instar) of each individual was also recorded for later consumption estimates. The length of each taxon was averaged for each sample (June-July 2014) or set of triplicate samples (Aug 2014-Oct 2015).

Areal (individuals m^{-2}) and volumetric (individuals m^{-3}) densities of each taxon were calculated. Volumetric density calculations were based on the amount of cable paid out (15 m or 60 m) for all stations. Estimated densities in triplicate tows were averaged for analyses.

Zooplankton biomass and taxonomic composition

The average individual dry weight of *Bosmina*, *Daphnia*, and *Holopedium* in a sample was estimated using the average length of the species and the length-weight regressions described in Bottrell et al. (1976), Persson and Ekbohm (1980), and Dumont et al. (1975), respectively. Average individual dry weight was multiplied by the average density of the species to estimate total biomass for that species in each tow.

Large cladocerans shrink when placed in preservatives (Yan and Pawson, 1998). This is problematic because length-weight regressions for *Bythotrephes* and *Leptodora* were based on unpreserved animals (Branstrator, 2005). To estimate the original lengths of preserved *Bythotrephes* and *Leptodora* individuals for biomass estimates, 64 live *Bythotrephes* (17, 36, and 11 individuals of instar 1, 2, and 3, respectively), and 98 live *Leptodora* individuals were measured to the nearest tenth of a millimeter and placed in 70% ethanol for three weeks. Because of the difficulties associated with collecting and handling live zooplankton in Lake Superior, *Bythotrephes* were collected in Island Lake Reservoir (Duluth, MN) on July 1, 2015 and *Leptodora* were collected in Caribou Lake (Duluth, MN) on July 7, 2015. After three weeks of storage in ethanol, each individual was re-measured. To determine whether *Bythotrephes* shrinkage was instar-specific, the shrinkages of each instar were compared using one-way ANOVA ($\alpha = 0.05$).

Bythotrephes core body length (length of animal excluding caudal spine) and *Leptodora* total body length shrank an average of 19.45% and 16.67%, respectively, when preserved in 70% ethanol. One-way ANOVA indicated that shrinkage in *Bythotrephes* was not instar-specific ($p = 0.51$, $F_{2, 61} = 0.689$) and therefore all instars were combined for the *Bythotrephes* shrinkage regression. The fresh lengths of *Bythotrephes* (equation 2) and *Leptodora* (equation 3) preserved in 70% ethanol can be predicted as:

$$Fresh\ length\ (mm) = 0.8166\ l_p\ (mm) - 0.0366, \qquad n = 64, R^2 = 0.8083 \qquad \text{(Eq. 2)}$$

$$Fresh\ length\ (mm) = 0.8953\ l_p\ (mm) - 0.3038, \qquad n = 97, R^2 = 0.9193 \qquad \text{(Eq. 3)}$$

where l_p is the length of the preserved individual. The lengths of *Bythotrephes* and *Leptodora* captured in Lake Superior in this study were corrected for shrinkage before biomass estimates were made. Because all *Bythotrephes* and *Leptodora* present in the samples were measured, biomass estimates for these species were based on the sum of all individuals in a sample.

We compared the monthly composition of the offshore herbivorous cladoceran community (in terms of percent contribution to total numbers) observed in 2014-2015 to historical estimates of cladoceran composition starting as early as the 1970s to identify long-term changes in the cladoceran community. Changes in absolute densities of cladocerans over time could not be compared because densities were not consistently reported in the literature. To account for the relatively low number of stations visited in some months, we used a bootstrapping technique in R software to estimate the average density of each species in each month sampled. This was done by randomly selecting density estimates from the 60 m net tows from each month 1000 times with replacement and calculating average monthly densities and standard errors from the vectors of density estimates. The resulting average monthly densities of each species were used to calculate percent contribution of each species to total offshore herbivorous cladoceran numbers in each month sampled.

Data for historical comparisons came from a variety of sources and we subsetted observations from these sources that were from comparable depths (stations greater than 60 m) and regions of the lake

Table 1 here

to eliminate depth-related biases from long-term community comparisons. Sources of historical data are described in Table 1.

Determining the vertical distributions of cladocerans

We compared the estimated areal densities (individuals m^{-2}) of *Bythotrephes*, *Bosmina*, *Daphnia*, and *Holopedium* in the 15 m and 60 m tows taken at each of the stations using a separate paired, one-tailed *t*-test ($\alpha = 0.05$) for each species. These tests were done separately for 2014 and 2015 samples. Because areal densities are not adjusted for the depth of the net tow, it can be inferred that most individuals of a species were present in the upper 15 m of water when the average density of the species in 15 m and 60 m tows are not significantly different. A species whose average areal density is significantly higher in 60 m tows must be present at depths greater than 15 m in considerable numbers.

Spatial overlap of Bythotrephes *and cladoceran prey*

To determine whether increased densities of *Bythotrephes* are associated with decreased densities of their potential cladoceran prey species, we plotted the simultaneous densities of *Bythotrephes* with *Bosmina*, *Daphnia*, and *Holopedium*. The densities of each species were displayed as the percent of the maximum observed density for each respective species to control for differences in the ranges of observed densities between taxa. Densities derived from 15 m and 60 m tows were plotted separately to control for the potential effect of depth on estimated species density. Data from 2014 and 2015 were pooled for these comparisons.

Bythotrephes *consumptive demands and prey production*

Two published models were used to estimate average daily *Bythotrephes* consumptive demands and average daily production of the available cladoceran prey for each month sampled. Both models are based on temperature and estimated biomass. Temperature data were from CTD (Seabird Electronics) casts taken at zooplankton sampling sites. Past work suggests that in lakes with planktivorous fish such as lake herring, *Bythotrephes* generally remains in the upper 15 m of water during day and night (Young and Yan, 2008). For this reason, estimates of *Bythotrephes* consumptive demands and cladoceran production were based on biomass estimates from 15 m net tows only. The use of biomass data from 15 m tows also reduced uncertainty regarding vertical overlap between predator and prey compared to biomass data from 60 m tows.

Because of the relatively few stations sampled in some months and the potential biases that could result from differences in the frequency of sampling at certain stations, *Bythotrephes* consumption and prey production estimates were made by bootstrapping the temperature and biomass observations from stations 5, 7, 12, and 15 (Fig. 1). These four stations were sampled every month with the exception of July of 2014 and October of 2014, when station 7 was not sampled. The bootstrapping procedures used in consumption and production estimates are described below.

Estimating Bythotrephes *consumptive demands*

Bythotrephes has three developmental instars and the consumptive demand (*C*) of each instar was estimated separately for each month sampled using the instar-specific general linear models from Yurista et al. (2010). Model parameters include epilimnetic temperature, median individual dry weight of each instar, and the density of each instar. Because the number of *Bythotrephes* caught in some months was sometimes limited, we determined the median individual dry weight for each instar during each month using all of the individuals observed in that month. Thus, all monthly consumption estimates for an instar used the same month-specific individual dry weight for that instar.

The other model parameters, epilimnetic temperature and instar density, were bootstrapped by resampling the monthly observations 1000 times with replacement. This generated a single vector of 1000 temperature estimates for each month and a monthly density vector for each instar. Because temperature and instar densities were bootstrapped separately, the temperature and density parameters from each station were decoupled from one another. We did this to generate the largest possible range of monthly consumption estimates possible based on observed conditions. Model inputs were pulled component by component from the bootstrapped vectors of epilimnetic temperature and instar density into the instar-specific consumption equations. This generated 1000 population consumption estimates for each instar in each month. The three resulting instar consumption vectors for each month were summed to generate 1000 estimates of consumption for the entire *Bythotrephes* population which were then used in later surplus and deficit production estimates.

Estimating cladoceran production

Production (*P*) was estimated for each herbivorous cladoceran species individually based on Shuter and Ing (1997) as:

$$P = 10^{\alpha_{taxon} + \beta \times t(°C)} \times B \qquad \text{(Eq. 4)}$$

where α_{taxon} is a cladoceran-specific intercept (-1.725), β is the slope (0.044), *B* is the biomass (dry weight) of each cladoceran species, and *t* is the average daily temperature in the upper 15 m of water. We chose this model for estimating cladoceran production over other methods (i.e., the egg ratio method) because the time intervals between sampling events in this study were too long to accurately estimate the rate of population growth used in other methods. In addition, previous studies have shown that the *P*:*B* method of Shuter and Ing (1997) produces reliable estimates of production for the small herbivorous cladoceran taxa observed in Lake Superior (Stockwell and Johannsson, 1997; Carter and Schindler, 2012).

The bootstrapping protocol for cladoceran production estimates was similar to that used for *Bythotrephes* consumption. Prey biomasses in each month were bootstrapped together by resampling the biomass observation for each species at each station 1000 times with replacement. Componentwise multiplication was used to generate 1000 production estimates for each taxon in each month from the monthly biomass vectors and the same monthly temperature vectors from the *Bythotrephes* consumption estimates. The three monthly taxon-specific production estimates from each bootstrap iteration were also summed to produce 1000 estimates of total cladoceran production. These steps decoupled the prey biomass and temperature observations from each station but did not decouple biomass observations of the prey taxa at each station from each other. This was done to generate the largest range of potential production outcomes for each month and to ensure that temperature experienced was not a source of variation between taxa for monthly production estimates.

Comparing consumptive demands of Bythotrephes *to herbivorous prey production*

To determine the relative vulnerability of each cladoceran species to suppression by *Bythotrephes*, the vector of consumption estimates for the *Bythotrephes* population was subtracted, component by component, from the vectors of production estimates for *Daphnia*, *Bosmina*, and *Holopedium* for each month where *Bythotrephes* were observed. Because of the wide range of consumption and production estimates for different taxa and months, consumption and production estimates were log (x + 1) transformed before subtracting the two vectors. The resulting vectors were used to produce box and whisker plots from which we could visualize the deficit or surplus production rate for each species in each month. Taxa with median monthly production rates less than *Bythotrephes* consumption were interpreted to be more vulnerable to top-down effects of *Bythotrephes* than taxa with production estimates in excess of the consumptive needs of *Bythotrephes*. The log-transformed consumptive demands of the *Bythotrephes* population were also subtracted from the vector of log-transformed total cladoceran production estimates to compare *Bythotrephes* consumptive demands to total epilimnetic cladoceran production.

Results

Pressure gauges deployed during net tows in 2015 indicated that nets reached average depths of 13.9 m (SE = 0.62) and 55.6 m (SE = 0.10) when 15 m and 60 m of cable were paid out, respectively. While net tows did not generally reach target depths, the ratios of depths reached in deep and shallow tows indicate that the deep net tows consistently reached depths four times those of shallow tows in each month sampled.

Cladocerans became common in August in 2014 but in 2015 they were already common in July (Table 2). *Daphnia* was the most common cladoceran by density and biomass during both years sampled. The largest average monthly densities of *Bosmina* and *Holopedium* were observed in 2014 while densities of *Daphnia* were greater in 2015 than in 2014 (Table 2). *Bythotrephes* densities varied widely by month during 2014 and 2015 and were only detected during July-October (Table 2). *Bythotrephes* was observed in densities up to 267 individuals m^{-2} but was usually much less abundant and average monthly *Bythotrephes* densities never exceeded 23 individuals m^{-2} (Table 2). Small densities of *Leptodora* were detected in August of 2014 and July of 2015 but accounted for less than 0.1 percent of cladoceran numbers and biomass.

Table 2 here

The structure of the offshore herbivorous cladoceran community in Lake Superior, which includes primarily *Daphnia*, *Bosmina*, and *Holopedium*, appears to have shifted in the past four decades. Most notably, the monthly contribution of *Bosmina* to total offshore herbivorous cladoceran numbers appeared to be smaller during the 1970s than in observations after 1996 (Fig. 2). *Bosmina* contributed 50-90% of offshore cladoceran numbers in July and August during the 1970s but only accounted for 20-30% during the summers of 2014 and 2015 (Fig. 2a, b). *Bosmina* was also a less important member of the cladoceran community in fall of 2014 and 2015 than it was during fall observations from the 1970s (Fig. 2c, d). Relatively speaking, *Daphnia* was a more important contributor to total offshore cladoceran numbers in Lake Superior in 2014 and 2015 than it was in the past. It is unclear whether absolute densities of *Daphnia* have increased since the 1970s due to differences in reporting of densities in Watson and Wilson (1978) and Conway et al. (1973). There were no obvious changes in the contribution of *Holopedium* to cladoceran numbers in 2014 or 2015 compared to the 1970s.

Figure 2 here

Vertical distributions of Bythotrephes *and cladoceran prey*

Table 3 here

The average areal density of *Bythotrephes* was higher in 2014 than in 2015 but the differences in *Bythotrephes* densities between 15 m and 60 m tows were not significantly different in either year (Table 3). Average areal densities of *Bosmina* and *Holopedium* were also not significantly different between 15 m and 60 m tows in either year (Table 3). *Daphnia* areal densities were significantly higher in 60 m tows than in 15 m tows in 2014 ($p = 0.04$) but were not significantly different in 2015 (Table 3). Separating day and night samples before running t-tests produced similar results but the number of samples collected during the day were limited.

Simultaneous densities of Bythotrephes *and cladoceran prey*

Large densities of *Holopedium* often occurred when *Bythotrephes* were also abundant in both years sampled (Fig. 3). *Daphnia* was generally most common when *Bythotrephes* was absent or in low densities, however, large densities of *Bythotrephes* and *Daphnia* occasionally coincided in each year sampled (Fig. 3). High simultaneous densities of *Bythotrephes* and *Bosmina* were never observed in 2014 or 2015. The maximum *Bosmina* density occurred when *Bythotrephes* densities were about 5% of the *Bythotrephes* maximum (Fig. 3). Above 5% of the maximum *Bythotrephes* density, *Bosmina* densities never exceeded 10% of their maximum.

Figure 3 here

Comparisons of Bythotrephes *consumptive demands and epilimnetic prey production*

August of 2014 had the highest monthly *Bythotrephes* density with approximately 23 individuals m^{-2} (Table 2). This density was strongly influenced by a station where *Bythotrephes* densities were in excess of 150 individuals m^{-2}. While this density was greater than the density observed at any other stations in 2014 or 2015, it is not an unreasonable *Bythotrephes* density for Lake Superior (Brown and Branstrator, 2004; Yurista et al., 2009; Isaac et al., 2012). Estimated median *Bythotrephes* consumption was well in excess of total cladoceran production during August of 2014 (Fig. 4). Even when the large *Bythotrephes* density observed in August of 2014 was omitted from the model inputs, consumption by *Bythotrephes* still overwhelmed the production rate of each prey taxon as well as total cladoceran production although not as drastically.

Median *Bythotrephes* consumption did not exceed total cladoceran production in any month besides August of 2014. However, median *Bythotrephes* consumption in July of 2015 was greater than the median production rates of each of the three individual taxa due to relatively low densities of herbivorous cladocerans observed in this month (Table 2, Fig. 4). *Bythotrephes* consumption exceeded *Bosmina* and *Holopedium* production in every month where *Bythotrephes* was observed but did not exceed *Daphnia* production in September of 2015 or October of either year (Fig. 4).

Figure 4 here

Because of the limited dataset, there was a relatively large amount of variability in the bootstrapped surplus and deficit production estimates for some months (Fig. 4). Median to mean ratios of the surplus and deficit production estimates are summarized in Table 4. The median to mean ratio during

Table 4 here

July of 2015 was negative because mean *Bythotrephes* consumption was slightly negative while median consumption was slightly positive. The median to mean ratios did not become closer to 1 when production and consumption estimates were repeated with 10,000 bootstrapping iterations. This indicates that 1000 iterations adequately resamples the available data but that the model outputs for some months are non-normally distributed.

To evaluate the effect of decoupling biomass and temperature observations at individual stations, we estimated production and consumption for each station and bootstrapped the resulting station-specific estimates. Under this scenario, *Bosmina* production was still exceeded by *Bythotrephes* consumption in every month and *Daphnia* production was not exceeded in the September or October observations. However, assuming site dependence of biomass and temperature caused *Holopedium* production to be in excess of *Bythotrephes* production in September and October of 2015. We also evaluated the effect of using only the data from routinely sampled stations on model outputs by repeating the consumption and production estimates using data from all of the stations visited in each month. We found that the proportion of times that each prey taxon was overwhelmed by *Bythotrephes* consumptive requirements was the same as when only data from routinely sampled stations were used.

Discussion

In the decades since the establishment of *Bythotrephes* in Lake Superior, the composition of the offshore cladoceran community appears to have changed. Past studies suggest that *Bosmina* was often equally or more numerically common in Lake Superior than *Daphnia* from spring through fall (Swain et al., 1970; Patalas, 1972; Schelske and Roth, 1973; Conway et al., 1973; Selgeby 1975). *Bosmina* contributed less to cladoceran numbers in 2014 and 2015 than it did during the 1970s which is consistent with the findings of Brown and Branstrator (2004). Our results show that *Daphnia* has become a more important contributor to the cladoceran community in the offshore regions of Lake Superior and suggest that the importance of *Holopedium* has remained stable. Similar changes have occurred in the cladoceran communities of the other Great Lakes and inland lakes following *Bythotrephes* invasion (Yan and Pawson, 1997; Barbiero and Tuchman, 2004; Rudstam et al., 2015; Kerfoot et al., 2016). We analyzed patterns in simultaneous densities of predator and putative prey to evaluate evidence for consumptive effects of *Bythotrephes* on any of the cladoceran species. We also analyzed vertical distributions of *Bythotrephes*, *Bosmina*, *Daphnia*, and *Holopedium* and compared the consumptive demands of *Bythotrephes* to the production rates of each herbivorous cladoceran species to determine which were most vulnerable to top-down control.

Bosmina

Both *Bosmina* and *Bythotrephes* were caught primarily in the upper 15 m of water during both 2014 and 2015 (Table 3). This is consistent with the vertical distribution of *Bythotrephes* observed in other lakes with planktivores like lake herring (Young and Yan, 2008). Past studies of the vertical distributions of cladocerans in Lakes Michigan and Erie reported a downward shift in the average vertical position of *Bosmina* following *Bythotrephes* invasion (Pangle et al., 2007). In this way, *Bythotrephes* might indirectly reduce *Bosmina* production by causing *Bosmina* to inhabit colder water (Pangle et al., 2007). However, Lakes Michigan and Erie have greater summer surface temperatures than Lake Superior and have historically had deeper thermoclines (Reavie et al., 2017). These conditions allow cladocerans in Lakes Michigan and Erie to migrate to greater depths before experiencing temperatures that cause appreciable reductions in production.

Despite occupying similar depths, the simultaneous densities of *Bosmina* and *Bythotrephes* imply a general lack of co-presence between these species. This pattern is consistent with recent observations in Lake Ontario (Barbiero et al., 2014; Rudstam et al., 2015) and could be evidence for predation on *Bosmina* by *Bythotrephes*. Finally, the consumptive demands of *Bythotrephes* in this study always exceeded *Bosmina* production when *Bythotrephes* was present. This was the case regardless of whether we used data from all stations or from only the routine stations and regardless of whether or not we assumed site-dependence of temperature and biomass data. These results demonstrate that it takes very few *Bythotrephes* eating *Bosmina* to overwhelm average *Bosmina* production. In addition to being a preferred prey species for *Bythotrephes*, new evidence suggests that *Bosmina* may lack avoidance responses to *Bythotrephes* (Kerfoot et al., 2016). Therefore, it is possible that *Bosmina* remains an easy prey item for *Bythotrephes* in the offshore regions of Lake Superior despite being present at relatively low densities.

Daphnia

In 2014, larger *Daphnia* densities were observed in 60 m tows than 15m tows, suggesting that overlap between *Bythotrephes* and *Daphnia* may have been reduced. However, this difference was relatively small and the difference in *Daphnia* densities between deep and shallow tows was not significantly different in 2015. *Daphnia* has remained common in Lake Superior, as it has in several other lakes, since the establishment of *Bythotrephes* (Lehman, 1991; Azan et al., 2015). Like *Bosmina*, the average vertical position of *Daphnia* in Lake Michigan shifted downward after *Bythotrephes* invasion (Lehman and Cáceres, 1993; Pangle et al., 2007). This change in habitat use has been used to explain the persistence of *Daphnia mendotae* in Lake Michigan since the establishment of *Bythotrephes* despite reduced biomasses of other species of *Daphnia*. While occupying deeper depths may have allowed *Daphnia* to avoid predation, it has also resulted in reduced *Daphnia* production in Lake Michigan due to the lower water temperatures at greater depth. In Lake Superior, where surface temperatures are lower and the thermocline is generally shallower, the benefits of migrating to deeper waters may not outweigh the costs, especially at the relatively low *Bythotrephes* densities observed in 2014 and 2015.

Regardless of whether or not *Daphnia* actively reduce their vertical overlap with *Bythotrephes* in Lake Superior, monthly epilimnetic production rates of *Daphnia* exceeded the consumptive demands of *Bythotrephes* in three of the five months where *Bythotrephes* was observed. Similar trends were observed when we assumed site dependence of temperature and biomass data and when we used data from infrequently sampled locations. *Daphnia* also co-occurred with *Bythotrephes* more often than did *Bosmina* which is consistent with lower relative consumption rates of *Daphnia* by *Bythotrephes*. These findings suggest that *Daphnia* in Lake Superior is less susceptible to being overwhelmed by *Bythotrephes* and may explain why *Daphnia* has remained common in Lake Superior since the establishment of *Bythotrephes*.

Holopedium

Most *Holopedium* biomass occurred in the upper 15 m of water, implying vertical overlap with *Bythotrephes*. In addition, the monthly consumptive demands of *Bythotrephes* exceeded *Holopedium* production when *Bythotrephes* was present under most modeling scenarios. While these findings suggest that *Holopedium* could be suppressed by *Bythotrephes* in Lake Superior, *Holopedium* and *Bythotrephes*, like *Daphnia* and *Bythotrephes*, commonly occurred together. *Holopedium*, which has not become less common in Lake Superior since the arrival of *Bythotrephes*, may be less vulnerable to predation by *Bythotrephes* because of its gelatinous coating and have occasionally become more common in other lakes after *Bythotrephes* invasion (Yan and Pawson, 1997; Barbiero et al., 2014; Kerfoot et al., 2016). The relative frequency of vertical and spatial overlap between these species in the present study suggests that *Holopedium* is not a preferred prey source for *Bythotrephes* in Lake Superior. Taken together, these results support the hypothesis that *Holopedium* is less susceptible to predation by *Bythotrephes* and help to explain why *Holopedium* does not appear to have become less common since *Bythotrephes* establishment.

The role of Bythotrephes *in cladoceran community change*

Of the common herbivorous cladocerans in Lake Superior, *Bosmina* was the only species that met all three of the criteria used in this study to evaluate the possibility of suppression by *Bythotrephes*. Because *Bosmina* is also the only herbivorous cladoceran species that appears to have become less common in the offshore regions of Lake Superior since the establishment of *Bythotrephes*, these findings support the hypothesis that *Bosmina* has become a less important member of the cladoceran community in Lake Superior as a result of top-down control by *Bythotrephes.*

It is unlikely that increasing surface water temperatures or vertebrate planktivory were responsible for the decline in this formerly abundant species. Herbivorous cladocerans are less abundant in Lake Superior than in the other Great Lakes and this is partially due to the low surface water temperatures in Lake Superior (Patalas, 1972; Lehman, 2002). Therefore, increasing temperatures in Lake Superior should not lead to a reduction in *Bosmina* abundance. Populations of planktivorous fish like lake herring have increased in Lake Superior since the 1970s, but *Bosmina* is not effectively retained in the gill rakers of mature lake herring and *Bosmina* is not observed in the diet of this or other common pelagic planktivorous fish (e.g., rainbow smelt, *Coregonus spp.*) in Lake Superior (Link and Hoff, 1998; Isaac et al., 2012; Gamble et al., 2011). Juvenile fish may feed on *Bosmina*, but year classes of smelt and the various coregonines have been irregular in Lake Superior in recent decades (Bronte et al., 2003; Johnson et al., 2004; Gorman, 2012; Pratt et al., 2016) while the reduction in *Bosmina* abundance appears to be persistent.

It is important to note that the cladoceran community observations made in 2014 and 2015 differ from other recent zooplankton surveys in Lake Superior. For one, densities of *Bythotrephes* observed in the present study were considerably lower than those reported in other Lake Superior zooplankton surveys conducted since 2001 (Brown and Branstrator, 2004; Yurista et al., 2009; Isaac et al., 2012). It is unclear why the *Bythotrephes* densities observed in 2014-2015 were so much lower than in previous summers. Past work shows that peak *Bythotrephes* densities in an inland reservoir are short-lived (Brown et al., 2012) and it is therefore possible that the true peaks in *Bythotrephes* density did not coincide with sampling events. We were unable to collect zooplankton samples in September of 2014 and August of 2015 which is when *Bythotrephes* densities have peaked in Lake Superior in other years (Isaac et al., 2012) and because of this the estimates of *Bythotrephes* consumptive demands in this study are likely to be conservative.

Another difference between our observations and other recent surveys has to do with *Holopedium*. Brown and Branstrator (2004) reported a possible increase in *Holopedium* abundance between the 1970s and 2001. The densities of *Holopedium* we observed in 2014-2015 were considerably lower than those reported in Brown and Branstrator (2004). However, Yurista et al. (2009) and Pratt et al. (2016) also report *Holopedium* densities in 2006 and 2011 that were much higher than we observed. Therefore, while the present study does not indicate *Holopedium* is more common than it used to be, there is good evidence that *Holopedium* is often more abundant in Lake Superior than before the establishment of *Bythotrephes*. Because *Holopedium* is not thought to be a preferred prey species for *Bythotrephes*, the higher *Holopedium* densities reported in other recent studies may be evidence of a species replacement resulting from reduced competition with *Bosmina*.

Future food web implications of Bythotrephes

These findings suggest that *Bythotrephes* has had a measurable impact on the offshore cladoceran community in Lake Superior. Seasonally, cladocerans are important sources of prey for some species of planktivorous fish in Lake Superior (Gamble et al., 2011; Isaac et al., 2012). While *Bosmina* is not an important food source for planktivorous fish in Lake Superior, *Bythotrephes* necessarily consumes other species and may therefore reduce the amount of cladoceran biomass available for such planktivores, especially those that are unable to eat *Bythotrephes*. Furthermore, though the densities of *Bythotrephes* observed in this study were generally not high enough to overwhelm total cladoceran production, the densities needed to do so are within the range of *Bythotrephes* densities observed in Lake Superior in the past.

This study does not address the relationship between *Bythotrephes* and copepods in Lake Superior. While *Bythotrephes* is known to occasionally consume small copepods, cladocerans are their preferred prey (Schulz and Yurista, 1999; Dumitru et al., 2001). Because the standing stock of copepod biomass in Lake Superior greatly exceeds cladoceran biomass (Yurista et al., 2009; Barbiero et al., 2012), *Bythotrephes* is unlikely to influence the copepod community structure or biomass in Lake Superior at the densities observed in this study. In addition, zooplankton biomass has been relatively stable in Lake Superior in recent decades and the dominant copepod species in Lake Superior have not changed since the 1970s (Barbiero et al., 2001; Barbiero et al., 2012).

The consumptive demands of *Bythotrephes* relative to prey production in Lake Superior are likely to change with further climate warming because temperature is one of the factors that influences both zooplankton production and *Bythotrephes* consumption. We explored the consequences of further warming on predator consumption and prey production by projecting the production and consumption estimates made in this study over a higher range of temperatures. When both predator and prey biomass were held constant, prey production increases faster than *Bythotrephes* consumption. However, predicting the effects of continued warming in Lake Superior on this predator-prey interaction is difficult because the response of *Bythotrephes* and their prey species to further warming may not be equivalent. For example, past studies indicate that the optimal temperatures for *Bythotrephes* growth and reproduction are in the range of 18-22°C (Kim and Yan, 2010; Yurista et al., 2010), which are temperatures not consistently met in the offshore regions of Lake Superior (Austin and Colman, 2007).

This suggests that low surface temperatures might be one factor limiting *Bythotrephes* abundance in Lake Superior. Warming water temperature might also favor herbivorous cladocerans over copepods in Lake Superior as it has in other cold water systems (Carter and Schindler, 2012). However, herbivorous cladoceran abundance in Lake Superior is probably also limited by food quality and quantity (Patalas 1973) and the effects of continued warming on primary production in Lake Superior are difficult to predict (Reavie et al., 2017). As such, the future top-down effects of *Bythotrephes* on Lake Superior zooplankton will depend on the extent of warming, changes in primary production, and the many possible responses of *Bythotrephes* and its potential prey species to warming.

Factors other than temperature also influence *Bythotrephes* abundance in Lake Superior and increase the uncertainty regarding the future effects of *Bythotrephes* on the Lake Superior food web. Though increased surface temperatures could result in higher abundances of *Bythotrephes* in Lake Superior, *Bythotrephes* is also heavily consumed by planktivorous fish like lake herring (Isaac et al., 2012; Keeler et al., 2015). Keeler et al. (2015) showed that *Bythotrephes* production can be overwhelmed by vertebrate planktivory in the offshore regions of the Apostle Islands. If this occurs throughout the lake, a small positive interaction between increasing surface water temperatures and *Bythotrephes* density may be masked by vertebrate planktivory. Because the consumptive demands of the *Bythotrephes* population are highly dependent on *Bythotrephes* density, future populations of the fish that consume *Bythotrephes* will also influence the effects of *Bythotrephes* on the Lake Superior zooplankton community in the coming decades.

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Figure captions:

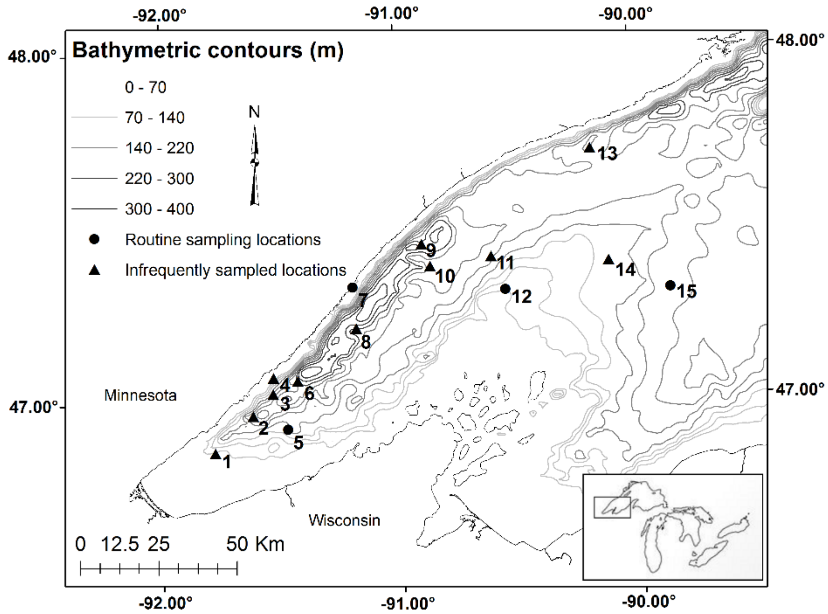

Fig. 1. Zooplankton sampling locations in 2014 and 2015. Routine sampling stations are those that were sampled during each month with the exception of station 7, which was not sampled in July or October of 2014. Infrequently sampled locations are those that were sampled on one or two occasions. Zooplankton samples were collected at station 6 once in 2015 and additional archived samples from 1996 were available for this station. Depths and coordinates for all sites are summarized in Appendix A.

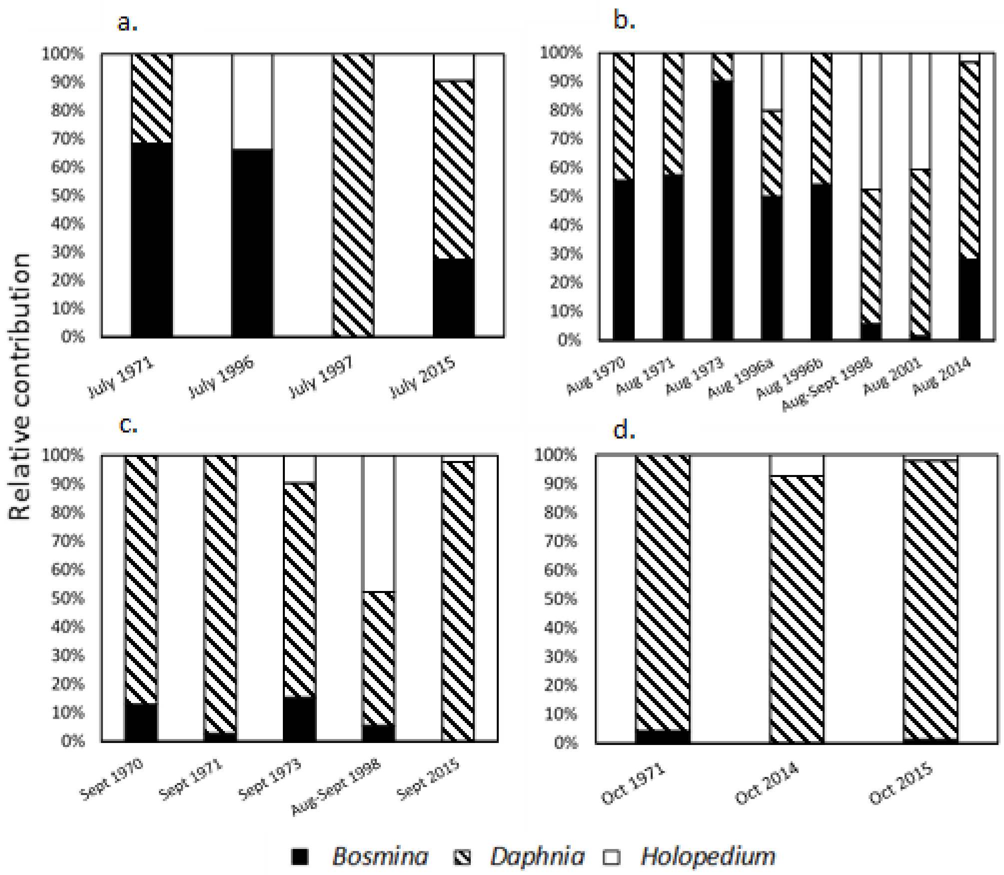

Fig 2. Comparisons of the relative contribution of *Bosmina*, *Daphnia*, and *Holopedium* to total offshore herbivorous cladoceran densities in Lake Superior during a) July; b) August; c) September; and d) October from 1971-2015. Observations 1996a and 1996b in panel b refer to different surveys (see Table 1). All 2014 and 2015 observations were from the present study and sources for previous observations are listed in Table 1.

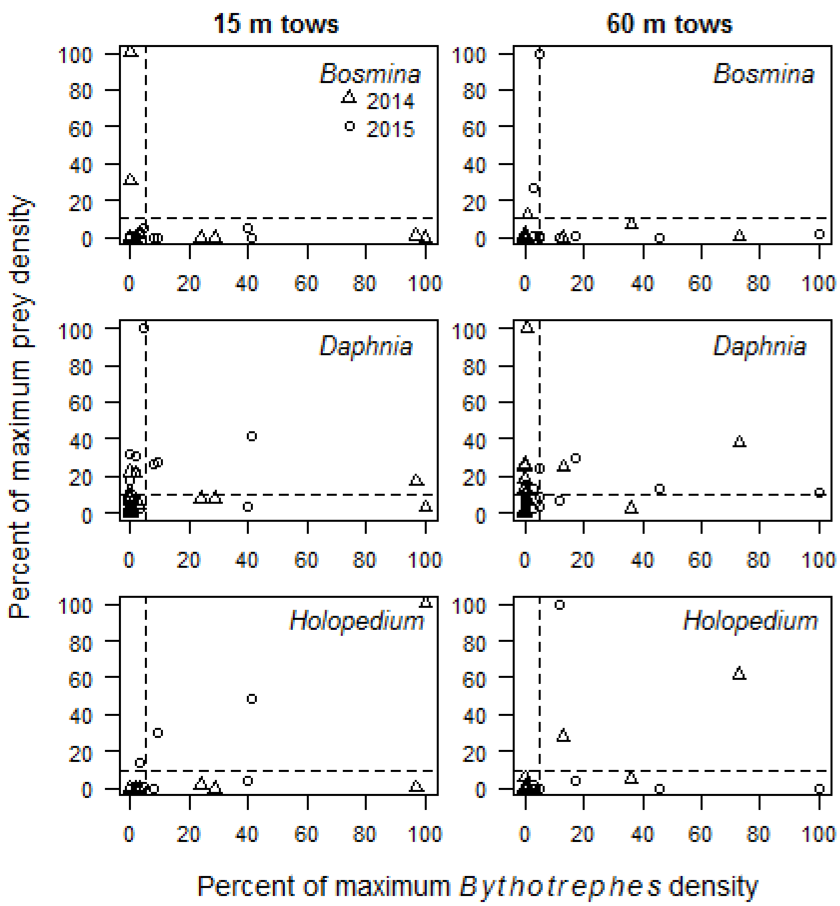

Fig. 3. Simultaneous densities of *Bythotrephes* with *Bosmina*, *Daphnia*, and *Holopedium* in 15 m net tows (left panels) and 60 m net tows (right panels) during 2014 and 2015. Densities for each species are relative to the maximum observed density for the species. The dashed horizontal and vertical lines at y = 10% and x = 5% provide a reference window to facilitate species-to-species comparisons of coexistence with *Bythotrephes*.

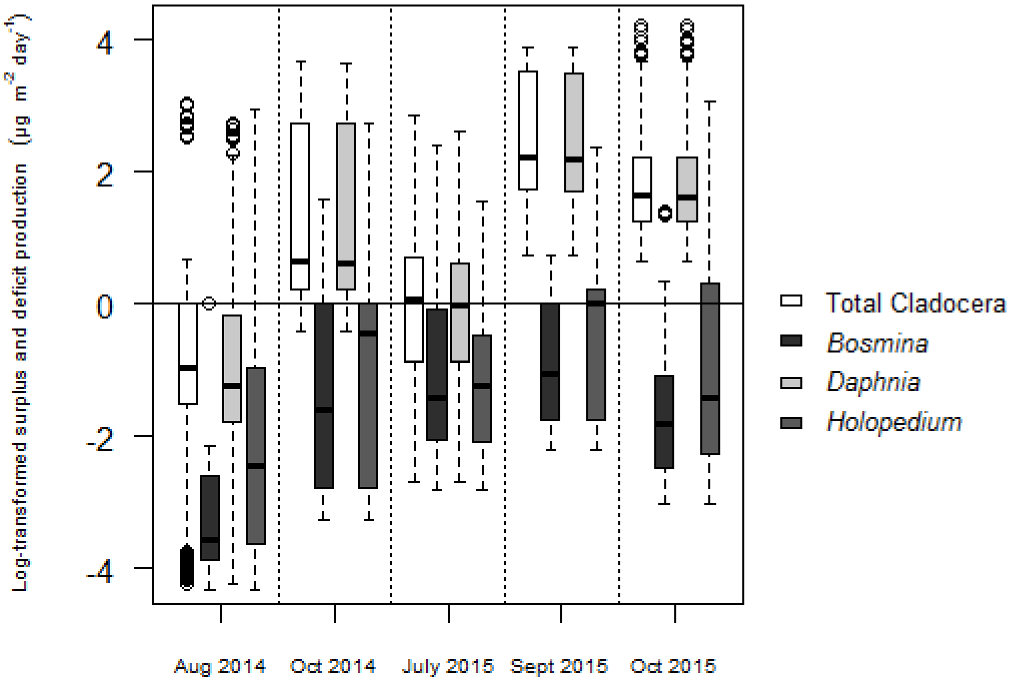

Fig. 4. Box and whisker plot of log-transformed surplus and deficit production rates (µg m^{-2} day^{-1}) for total Cladocera, *Bosmina*, *Daphnia*, and *Holopedium*. Median production values are indicated by the horizontal black lines within boxes. The lower and upper limits of the boxes represent the 25th and 75th percentiles, respectively. Whiskers represent 1.5 times the interquartile range below and above the 25th and 75th percentiles and points represent production estimates above or below the range covered by whiskers. Taxa with negative median production rates are those that would be overwhelmed by *Bythotrephes* if *Bythotrephes* fed exclusively on that taxon.

Table 1. Sources of monthly cladoceran community data for long-term comparisons. The “Data used” field describes how the data were subsetted to ensure compatibility with data from the present study. The letters a and b appended to the two August 1996 surveys differentiate columns in Fig. 2b.

Source	Months available	Data used
Conway et al., 1973	Aug-Sept 1970, July-Oct 1971	Larsmont and Stony Point stations at 2 miles from shore (155 m and 110 m deep respectively)
Watson and Wilson 1978	Aug-Sept 1973	Lake Regions 5 and 6 (Table 3 and Fig. 5 in original source)
Sterner (archived samples)	July 1996, Aug 1996a	Archived samples from R. Sterner. Collected at station 6 (Fig. 1) using 80 µm 0.5-meter diameter conical zooplankton net towed from bottom to surface. Samples processed Sept 2015.
Johnson et al., 2004	July 1997, Aug 1996b	“Open lake” region (See Fig. 2 and Table 2 in original source)
Barbiero et al., 2001	Aug-Sept 1998	All zooplankton collection done at stations of depth >90 m. Data used as is.
Brown and Branstrator 2004	Aug 2001	Stations deeper than 60 m (see Fig. 1 and Table 1 in original source).

Table 2. Average monthly density as individuals m^{-2} (standard error in parentheses) for common offshore cladocerans in Lake Superior during 2014-2015 in 60 m net tows. Averages and standard errors were estimated by bootstrapping density observations from each month. The number of stations visited during each month is indicated by n.

	Bythotrephes density (+/-SEM)	*Bosmina* density (+/-SEM)	*Daphnia* density (+/-SEM)	*Holopedium* density (+/-SEM)	n
June 2014	0	0	44.0 (2.35)	0	4
July 2014	0	0	0	0	3
Aug 2014	23.0 (1.1)	1388.8 (99.54)	3393.5 (137.71)	155.9 (12.77)	10
Oct 2014	3.6 (0.1)	6.3 (0.38)	4280.7 (85.43)	330.9 (25.43)	13
May 2015	0	0	0.5 (0.03)	0	5
July 2015	2.6 (0.07)	217.4 (9.75)	500.2 (13.49)	73.2 (2.36)	4
Sept 2015	0.4 (0.32)	14.2 (0.75)	9083.8 (124.47)	210.2 (10.65)	4
Oct 2015	1.3 (0.05)	225.0 (13.96)	16214.9 (492.0)	292.3 (19.83)	7

Table 3. Average areal densities of *Bythotrephes*, *Bosmina*, *Daphnia*, and *Holopedium* in 15 m and 60 m net tows. The 2014 comparisons were based on 30 paired observations and the 2015 comparisons were based on 20 paired observations. Differences in densities were identified with a paired, one-tailed *t*-test ($\alpha = 0.05$).

	2014			**2015**		
	# m^{-2} (15 m)	# m^{-2} (60 m)	*p*-value	# m^{-2} (15 m)	# m^{-2} (60 m)	*p*-value
Bythotrephes	11.6	9.2	0.22	1.3	2.4	0.13
Bosmina	457.4	484.5	0.19	67.6	114.7	0.12
Daphnia	2227.8	3142.6	0.04	6312.3	6709.6	0.25
Holopedium	106.3	87.1	0.12	189.2	127.2	0.12

Table 4. Monthly median to mean ratios for the surplus and deficit production estimates for total Cladocera, *Bosmina*, *Daphnia*, and *Holopedium* (see Fig. 4). Medians and means were calculated from the 1000 bootstrapped estimates of log-transformed surplus and deficit production for each month where *Bythotrephes* were observed.

	Total Cladocera	*Bosmina*	*Daphnia*	*Holopedium*
August 2014	1.04	1.21	1.13	1.28
October 2014	0.57	1.04	0.54	0.39
July 2015	-0.49	1.38	0.27	1.02
September 2015	0.88	1.34	0.88	0.00
October 2015	0.86	1.15	0.85	1.53

Appendix A. Location and depth of sampling stations shown in Fig. 1.

Site number	Latitude (degrees North)	Longitude (degrees West)	Site depth (meters)
1	46.8614	-91.7815	77
2	46.9666	-91.6200	208
3	47.0281	-91.5354	231
4	47.0753	-91.5330	102
5	46.9271	-91.4753	137
6	47.0650	-91.4317	255
7	47.3313	-91.1925	87
8	47.2129	-91.1810	276
9	47.4505	-90.8979	241
10	47.3868	-90.8633	200
11	47.4110	-90.6049	170
12	47.3155	-90.5476	77
13	47.7129	-90.1720	157
14	47.3911	-90.1089	140
15	47.3088	-89.8514	167