https://greatlakescenter.buffalostate.edu/

LAKE SUPERIOR BENTHOS SURVEY COOPERATIVE SCIENCE AND MONITORING INITIATIVE 2022

Technical Report

Principal Investigators: Lyubov E. Burlakova
Alexander Y. Karatayev

Great Lakes Center
SUNY Buffalo State
1300 Elmwood Ave, Buffalo, New York USA 14222

September 2023

Suggested citation for the report:

Burlakova, L. E., Karatayev, A.Y., S. E. Daniel. 2023. Lake Superior Benthos Survey Cooperative Science and Monitoring Initiative 2022. Technical Report. USEPA-GLRI GL00E02259. Great Lakes Center, SUNY Buffalo State, Buffalo, NY. Available at: https://greatlakescenter.buffalostate.edu/sites/glc/files/documents/LakeSuperiorBenthosSurveyCSMI2022FinalReport_0.pdf

TECHNICAL REPORT

LAKE SUPERIOR BENTHOS SURVEY

COOPERATIVE SCIENCE AND MONITORING INITIATIVE 2022

Project Title: "Great Lakes Long-Term Biological Monitoring 2017-2023"

Grant/Award Number: Subaward # 82839-10916 from Cornell University

U.S. Environmental Protection Agency Award GLRI GL00E02259 (PI Lars Rudstam)

Principal Investigators: Lyubov E. Burlakova and Alexander Y. Karatayev

Great Lakes Center, SUNY Buffalo State

1300 Elmwood Ave., Buffalo, NY 14222

Phone: 716-878-4504, 716-878-5423

E-mail: burlakle@buffalostate.edu; karataay@buffalostate.edu

September 2023

CONTENTS

1.1. Project Overview

In this report, we present results of a benthic survey of Lake Superior conducted as part of the United States Environmental Protection Agency (U.S. EPA) Great Lakes National Program Office (GLNPO) Great Lakes Biology Monitoring Program (GLBMP) and Cooperative Science and Monitoring Initiative (CSMI) benthic surveys. Consistent with the sampling scheme of previous CSMI surveys, a lake-wide benthic survey was conducted in 2022 at 66 stations in Lake Superior to assess the status of the benthic macroinvertebrate community. This study advanced the Lake Superior CSMI priority of monitoring and understanding Lake Superior's lower food web. The primary focus of this survey was to assess the status and long-term trends in benthic community, including the native amphipod *Diporeia,* Oligochaeta, Chironomidae, and Sphaeriidae.

1.2. Study Highlights

- 57 species and higher taxa of benthic macroinvertebrates were found in Lake Superior in 2022. The most diverse taxa throughout the lake were Chironomidae (24 genera), and the most abundant taxa throughout the lake were native amphipods *Diporeia* sp., representing 57% of lake-wide density and biomass.
- *Diporeia*, one of indicator species to assess the state of the Great Lakes, was found at 95% of all stations, with an average lake-wide density of 655 per m^2, and a maximum average density at the 51-90 m depth zone (1,333 per m^2). *Diporeia* populations remain in good conditions (at densities above 220-320 m^{-2} in nearshore waters (<100 m) and 30-160 m^{-2} in offshore (>100 m) waters) at 82% of the sampled stations.
- *Diporeia* abundance declined significantly by 70% at the 19 historical nearshore stations compared to 1994 and was the lowest of all previous surveys (1994, 2000, 2003 and 2016). The proportion of the historical nearshore stations where densities fell below 220/m^2 increased from 0% in 1994, to 10-15% in 2000 and 2003, 20% in 2016 and to 37% in 2022, and *Diporeia* was not found at two of these stations in 2022. *Diporeia* densities declined significantly in 2022 compared to previous surveys and were almost 3 times lower than in 2016 at 17 deep (>100 m) stations as well. Nevertheless, the current *Diporeia* abundance is still almost twice as high than in the 1970s. No temporal trends in *Diporeia* densities were found at the 11 deep GLBMP long-term monitoring stations from 1998 to 2022.
- At the historical nearshore stations, mean Oligochaeta and Sphaeriidae densities declined significantly two-fold compared to 1994 and were the lowest of all five surveys. At mostly deep GLBMP stations, however, there were no significant changes in Oligochaeta and Sphaeriidae densities from 1998 to 2022. Densities of Chironomidae at the historical nearshore stations did not change from 1994 to 2022, nor at the GLBMP stations since 1998.

1.3. Major findings from the CSMI Benthic Macroinvertebrate Survey in Lake Superior in 2022 with an emphasis on long-term trends in benthic community

1.3.1. Overview

A lake-wide benthic survey of Lake Superior was conducted in 2022 as part of the U.S. EPA Great Lakes National Program Office (GLNPO) Great Lakes Biology Monitoring Program (GLBMP) and Cooperative Science and Monitoring Initiative (CSMI) survey efforts. Consistent with the sampling scheme of previous CSMI benthic surveys, benthic samples were collected at 66 stations to assess the status of the benthic macroinvertebrate community.

Benthic surveys in Lake Superior were conducted in 1973 (lake-wide survey, Cook, 1975), 1994, 2000, 2003, and 2016 (U.S. nearshore region, Scharold et al., 2004, 2009; Mehler et al., 2018). A dramatic increase in all major taxonomic groups was found in surveys conducted in 1994 - 2003 compared to the 1973 survey (Scharold et al., 2009). The abundance of *Diporeia* in 2003 was 5 – 8 fold higher, and the abundance of Oligochaeta, Sphaeriidae (now Pisidiidae), and Chironomidae was 2 – 3 fold higher (Scharold et al., 2009). The decline in benthivorous fish and the reduction of heavy metal loadings and siltation at the beginning of the 1980s were identified as possible reasons for the higher abundance of benthos found in later surveys (Bronte et al., 2003; Scharold et al., 2009).

As a key component in the food web of offshore regions, and an important pathway by which energy is cycled through the ecosystem, *Diporeia* is used as sub-indicator of ecological condition in the State of the Great Lakes reports to assess ecosystem health in Lake Superior and in the cold, open-water portions of other Great Lakes. Densities of *Diporeia* in Lake Superior, however, declined by 59% in 2016 compared to 2003 at the 25 resampled historical nearshore stations and was the lowest of all four previous surveys (837 ± 132 m^{-2}, Mehler et al., 2018). *Diporeia* populations are considered in good conditions at densities 220 to 320 m^{-2} at depths less than 100 m, and 30-160 m^{-2} at greater depths (EPA and ECCC, 2022). While 94% of the 59 sampling stations in 2016 still had *Diporeia* densities exceeding the indicator thresholds lake-wide, the significant decline in 2016 compared to 1994 was found at 95% of the 25 previously sampled stations, and *Diporeia* were not collected at three of these stations. The proportion of the 25 historical stations that fell below the nearshore indicator threshold for *Diporeia* density increased from 0% in 1994 to 10 – 15% in 2000 and 2003 (Scharold et al., 2009), and to 20% in 2016 (Mehler et al., 2018). In contrast to other Great Lakes, *Dreissena* spp. have not established in Lake Superior and therefore cannot account for the decline in *Diporeia* abundance, suggesting that benthivorous fish predation or other environmental factors might have caused the changes. The decline in nearshore *Diporeia* density in 2016 could be an indication of a long-term trend of decreasing *Diporeia* densities or could be a result of inter-annual variation, reinforcing the importance of monitoring historical stations to detect statistically significant temporal trends. Densities of other major taxonomic groups including Oligochaeta, Sphaeriidae and Chironomidae declined in 2016 compared to 1994 as well, but the changes were not significant.

The objective of this study was to advance the 2021 Lake Superior CSMI priority of monitoring and understanding Lake Superior's lower food web and describe the status and trends in Lake Superior benthos, with a special emphasis on *Diporeia*, which has experienced widespread declines in the other Great Lakes. This report contains detailed descriptions of benthic communities in Lake Superior in 2022, including information on sampling design (station locations, sampling, and laboratory procedures), the taxonomy and abundance of benthic invertebrates, and changes in major taxonomic groups since the 1990s.

1.3.2. Methods

1.3.2.1. Sampling protocol

A total of 197 benthic samples from 66 stations were analyzed for benthic macroinvertebrates in this study: 164 samples from 55 CSMI stations and 33 samples from 11 GLBMP long-term monitoring stations (Fig. 1.1, Table 1.1). The CSMI stations were sampled from September 7 through October 5, 2022, and GLBMP stations were sampled in September 2022. All CSMI stations were sampled aboard the U.S. EPA R/V *Lake Guardian* using a standard Ponar grab (sampling area 0.0523 m^2) (US EPA, 2021). The long-term monitoring stations were collected from the R/V *Blue Heron* using a regular Ponar grab at all stations except one (SU 16), where a larger Ponar was used (sampling area of 0.0929 m^2).

Nineteen of the 55 CSMI stations (referred as "historical stations", LS94) were previously sampled in 1994, 2000, 2003 and 2016. The other 36 stations had only been previously sampled in 2016. Sixteen of these 36 stations were nearshore stations (BE-01 through BE-56, 23-103 m), 18 were deep-water stations (SEQ 40 through SEQ 139, 92-257 m), and 2 stations (HN40 and HN50) were characterized by previously high *Diporeia* densities (Table 1.1). Only two replicate samples were retrieved at one of the historical stations LS94-82781. Samples from 18 planned stations were not collected due to bad weather, hard substrates, or survey cancellation.

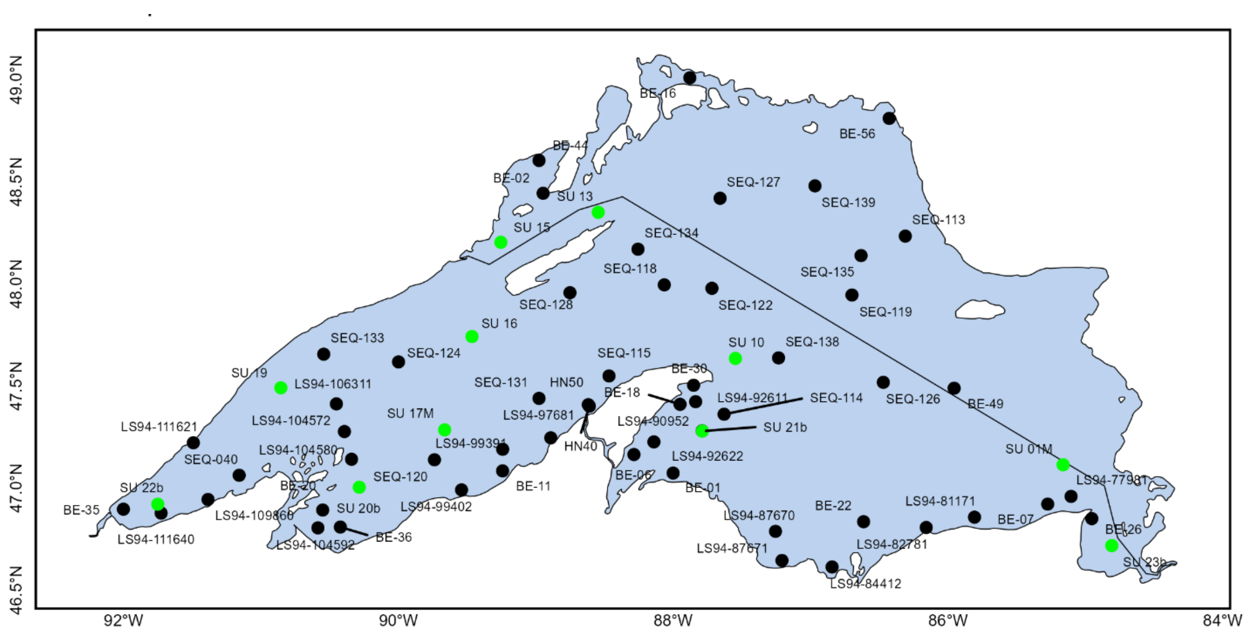

Figure 1.1. Location of 66 benthic stations surveyed in Lake Superior in 2022. The map indicates the locations of 55 CSMI benthic stations (black), and 11 GLBMP long-term monitoring benthic stations (in green; SU 01M, SU 10, SU 13, SU 15, SU 16, SU 17M, SU 19, SU 20b, SU 21b, SU 22b, SU 23b). LS94 stations are those sampled in 1994, 2000, 2003, and 2016, all other stations were previously sampled in 2016.

Table 1.1. A list of 66 stations, including 55 CSMI stations sampled on Lake Superior in September and October 2022 and 11 GLBMP long-term monitoring benthic stations sampled in September 2022 with information on location (decimal degrees), water depth (both planned and actual), and main substrates. NA – not available. BE stations: 16 nearshore stations previously sampled in 2016. HN stations: 2 stations characterized by previously high *Diporeia* densities. LS94 stations: 19 historical stations sampled in 1994, 2000, 2003, and 2016. SEQ stations: 18 deep-water stations previously sampled in 2016. SU stations: 11 GLBMP long-term monitoring stations.

Station	Latitude	Longitude	Actual depth, m	Substrate
BE-01	46.95267	-87.99928	58.0	sand and clay
BE-02	48.32294	-88.94550	33.4	clay and sand
BE-06	47.04352	-88.28437	23.0	sand and clay
BE-07	46.80078	-85.27342	26.0	sand and gravel
BE-11	46.96350	-89.24044	30.1	sand
BE-16	48.88835	-87.87760	46.0	silt and clay
BE-18	47.28941	-87.94888	55.0	clay and gravel
BE-20	46.77176	-90.54955	53.0	sand
BE-22	46.71458	-86.61311	48.5	gravel and sand
BE-26	46.72972	-84.95239	63.0	silt and sand
BE-30	47.38279	-87.85036	54.6	silt, clay, gravel, and detritus
BE-35	46.77579	-91.99999	30.0	silt, sand, and detritus
BE-36	46.68835	-90.42104	47.7	sand
BE-44	48.48355	-88.97523	28.4	clay
BE-49	47.36825	-85.95407	71.5	silt, clay, and sand
BE-56	48.68958	-86.42668	103.1	clay
HN40	47.28046	-88.60881	71.0	silt, sand, and detritus
HN50	47.28777	-88.61655	115.0	clay, gravel, and detritus
LS94-104572	47.15600	-90.39153	64.5	clay and sand
LS94-104580	47.02047	-90.33973	39.7	clay and sand
LS94-104592	46.68433	-90.58433	22.0	sand
LS94-106311	47.29153	-90.45000	91.0	clay and gravel
LS94-109860	46.82347	-91.38433	28.9	sand, clay, and gravel
LS94-111621	47.10120	-91.49093	104.3	clay, sand, gravel, and detritus
LS94-111640	46.75660	-91.72527	28.1	sand and clay
LS94-77981	46.83855	-85.10300	40.0	sand
LS94-81171	46.73673	-85.80600	25.0	sand and gravel
LS94-82781	46.68613	-86.15720	18.0	gravel and sand
LS94-84412	46.49295	-86.84247	41.2	sand
LS94-87670	46.66747	-87.25420	91.2	clay, sand, and gravel
LS94-87671	46.52467	-87.20720	34.2	sand

Station	Latitude	Longitude	Actual depth, m	Substrate
LS94-90952	47.15960	-87.78793	110.0	clay, gravel, and detritus
LS94-92611	47.30240	-87.83553	56.0	clay, gravel, and detritus
LS94-92622	47.10540	-88.13913	65.0	clay, sand, and gravel
LS94-97681	47.12527	-88.88973	27.0	gravel and sand
LS94-99391	47.06927	-89.23973	84.6	silt and sand
LS94-99402	46.87047	-89.53913	30.0	sand
SEQ-040	46.94188	-91.15741	92.6	silt and clay
SEQ-113	48.11308	-86.30946	167.4	clay
SEQ-114	47.24081	-87.62844	102.7	clay
SEQ-115	47.42837	-88.46593	223.0	silt, clay, sand, and detritus
SEQ-118	47.87444	-88.06359	238.5	silt and clay
SEQ-119	47.82477	-86.69773	257.0	silt and clay
SEQ-120	47.01792	-89.73626	199.0	silt and clay
SEQ-122	47.85755	-87.71684	235.0	silt and clay
SEQ-124	47.49716	-89.99779	147.4	clay
SEQ-126	47.39711	-86.46955	294.0	clay and silt
SEQ-127	48.29852	-87.65762	221.4	clay
SEQ-128	47.83599	-88.75010	231.5	clay
SEQ-131	47.31857	-88.97530	129.1	clay, silt, sand, and detritus
SEQ-133	47.53495	-90.54224	175.4	silt and clay
SEQ-134	48.04910	-88.25508	243.8	silt, clay, and detritus
SEQ-135	48.01783	-86.63151	163.2	clay
SEQ-138	47.51673	-87.23194	282.0	clay
SEQ-139	48.35920	-86.96694	196.0	clay
SU 01M	46.9933	-85.1611	94	NA
SU 10	47.5142	-87.5461	150	NA
SU 13	48.2297	-88.5444	150	NA
SU 15	48.0828	-89.2533	224	NA
SU 16	47.6214	-89.4631	178	NA
SU 17M	47.1644	-89.6619	198	NA
SU 19	47.3703	-90.8539	184	NA
SU 20b	46.8833	-90.2833	110	NA
SU 21b	47.1583	-87.7861	112	NA
SU 22b	46.8000	-91.7500	53	NA
SU 23b	46.5975	-84.8069	63	NA

Upon collection, each sample was placed separately into an elutriation device and then washed through a 500-µm mesh screen. All retained organisms and sediments were placed into a collection jar and preserved with neutral buffered formalin with Rose Bengal stain to a final formalin concentration of 5 – 10%. Detailed methods are described in the EPA GLNPO Standard Operating Procedure for Benthic Invertebrate Field Sampling (US EPA, 2021: SOP LG406, Revision 14, January 2021).

1.3.2.2. Laboratory Procedures

All organisms found in each replicate sample collected at the 66 benthos stations were sorted, identified, counted, and weighed (total wet weight). Organisms were separated under low magnification using a dissecting microscope. Oligochaetes and chironomids were mounted on slides and identified using a compound microscope; other organisms were identified using a dissecting microscope. Naididae and mature Tubificidae and Lumbriculidae were identified to species; Enchytraeidae, immature Tubificidae and Lumbriculidae were identified to the lowest taxonomic level possible, usually family, and included in density and biomass estimates. Counts of oligochaete fragments were excluded from density analyses but fragment weight was considered in the determination of biomass. Chironomids were identified to the lowest practical taxonomic level, usually genus. Other invertebrates were identified to species, when possible. Details are described in the EPA GLNPO Standard Operating Procedure for Benthic Invertebrate Laboratory Analysis (US EPA, 2021a: SOP LG407, Revision 10, March 2021).

1.3.2.3. Historical data

Historical data collections spanning between 1994 and 2016 (Scharold et al., 2004, 2009; Mehler et al., 2018) were used to examine long-term changes in major benthic taxonomic groups in Lake Superior. Four sets of previous data were analyzed to compare with 2022 data (Table 1.1):

1) 19 historical stations (LS94-77981 through LS94-111640) in the U.S. nearshore waters of southern Lake Superior from a set of 27 previously sampled in 1994, 2000, and 2003 (Scharold et al., 2009) and 2016 (Mehler et al., 2018). Initial selection of these stations in 1994 followed a randomized, probability-based sampling design used in the Great Lakes Environmental Monitoring and Assessment Program (EMAP-GL) (Scharold et al., 2009). Seven of these stations were located at ≤30 m depth, 4 at >30-50 m depth, 4 at >50-90 m depth, and 4 stations at >90 m (90-116 m).
2) 16 stations (BE-01 through BE-56, 23-103m) from a set selected in 2016 at 20-80 m depths zone using a Generalized Random Tessellation Stratified (GRTS) sampling design and sampled previously in 2016. Four of these stations were located at ≤30 m depth, 7 at >30-50 m depth, 4 at >50-90 m depth, and 1 station at >90 m (103 m).
3) 18 deep-water (SEQ 40 through SEQ 139, 92-257 m) stations previously sampled in 2016.
4) 2 stations (HN40, 65 m and HN50, 107 m depth) in the south-central region near Houghton, MI characterized by previously high *Diporeia* densities (Auer et al., 2013).

1.3.2.4. Data analysis

To compare 2022 benthos abundance among depth zones we used one-way ANOVA on log-transformed total density and biomass followed by pairwise Tukey HSD test. To test for significant differences in taxa densities collected at the same stations in two different years we used two-sided paired *t*-tests. To find significant changes in taxa densities at the same stations sampled over time we used repeated measures ANOVA on log-transformed densities from 1994, 2000, 2003, 2016 and 2022 followed by Tukey HSD test. All test effects were considered significant at $P < 0.05$.

1.3.3. Results and Discussion

1.3.3.1. Status of the Lake Superior benthic community in 2022

We found 57 species and higher taxa of benthic macroinvertebrates in Lake Superior in 2022, in addition to unidentified immature tubificids and unidentified Chironomidae. The most diverse were Chironomidae (24 species and higher taxa), Oligochaeta (21 species and higher taxa), Mollusca (3 species: 2 Gastropoda and 1 Bivalvia); and Malacostraca (2 species: 1 Amphipoda and 1 Mysida). Other taxa were represented by fewer than 3 species, or were not identified to species level (e.g., Trichoptera, Hydrozoa, Nemertea). Among Oligochaeta, the most diverse were Tubificidae (12 species and higher taxa), and Naididae (7).

The most widely occurring taxon throughout the lake was *Diporeia*, which was found at 63 stations (95% of stations), followed by Bivalvia recorded at 53 stations (*Pisidium:* 80%), Oligochaeta at 52 stations (79%), and chironomids at 45 stations (68%, mostly *Heterotrissocladius subpilosus* group). Among Oligochaeta, the most widespread were Lumbriculidae found at 62% of stations (*Stylodrilus heringianus* and immatures), Enchytraeidae (59%), and Tubificidae (25%).

Diporeia comprised the largest percentage (57%) of lake-wide benthos density, followed by Oligochaeta (23%), *Pisidium* (15%), and Chironomidae (4%) (Table 1.2). Contribution of other groups (Hirudinea, Trichoptera, Platyhelminthes, Nemertea, etc.) to total benthos density was less than 1% each. Among Oligochaeta, the most numerous were Lumbriculidae (67% of total Oligochaeta density), Enchytraeidae (17%), and Tubificidae (12%). *Diporeia* also comprised the largest share of lake-wide benthos by biomass (57% of total wet biomass). The remaining benthic biomass was represented by Oligochaeta (29%, dominated by Lumbriculidae), *Pisidium* bivalves (10%), and Chironomidae (2%).

Total benthos density and biomass were significantly lower at depths >90 m ($P < 0.0001$, one-way ANOVA on log-transformed total density and biomass, $P < 0.02$, Tukey HSD tests) compared to benthos abundance at ≤30 m, >30-50 m and >50-90 m depths.

Table 1.2. Average (± standard error) density (ind. m^{-2}) and wet biomass (g m^{-2}) of major taxonomic groups of benthic invertebrates collected from 66 benthic stations in Lake Superior in 2022 and averaged by depth zones and lake-wide (not weighted). Number of stations given in parentheses.

Taxa	≤30 m (11)	>30 – 50 m (11)	>50 – 90 m (11)	>90 m (33)	Lake-wide (66)
Diporeia sp. (ind. m^{-2})	564±193.3	940.4±211.7	1333.2±303.3	365.5±118.6	655.7±99.9
Diporeia sp. (g m^{-2})	1.14±0.44	1.95±0.47	2.28±0.51	0.5±0.16	1.15±0.18
Chironomidae (ind. m^{-2})	123.4±39.8	28.4±6.8	60.3±15.3	20.3±8	45.5±9.2
Chironomidae (g m^{-2})	0.08±0.04	0.03±0.01	0.08±0.02	0.02±0.01	0.04±0.01
Pisidium sp. (ind. m^{-2})	206.3±63.9	351.1±101.5	300.1±101.6	58±15.2	171.9±30.3
Pisidium sp. (g m^{-2})	0.21±0.08	0.4±0.13	0.36±0.17	0.06±0.02	0.19±0.04
Mysis relicta (ind. m^{-2})	0.6±0.6	5.8±2.5	5.2±1.9	7.1±2.2	5.5±1.2
Mysis relicta (g m^{-2})	<0.01	0.06±0.04	0.06±0.03	0.05±0.01	0.05±0.01
Oligochaeta (ind. m^{-2})	398.3±100.2	335.5±62.9	406.2±104.7	155.3±46	267.6±36.9

Taxa	≤30 m (11)	>30 – 50 m (11)	>50 – 90 m (11)	>90 m (33)	Lake-wide (66)
Oligochaeta (g m^{-2})	0.69±0.19	0.62±0.13	0.8±0.25	0.48±0.14	0.59±0.09
- Enchytraeidae (ind. m^{-2})	16.2±6.7	38.2±11.3	96.2±40.2	40±22.5	45.1±13.4
- Enchytraeidae (g m^{-2})	<0.01	0.02±0.01	0.04±0.02	0.02±0.01	0.02±0.01
- Lumbriculidae (ind. m^{-2})	244.8±62.9	255.5±60.8	243.9±85.5	110.9±31	179.5±26.3
- Lumbriculidae (g m^{-2})	0.39±0.11	0.4±0.11	0.46±0.18	0.33±0.1	0.38±0.06
- Naididae (ind. m^{-2})	48.1±26.6	0	0	0	8.0±4.8
- Naididae (g m^{-2})	0.02±0.01	0	0	0	<0.01
- Tubificida (ind. m^{-2})	84±62.8	40.6±18.4	59.1±54.7	4.2±1.6	32.7±14.2
- Tubificida (g m^{-2})	0.08±0.06	0.05±0.02	0.06±0.06	0.01±0	0.03±0.01
Total Benthos (ind. m^{-2})	1301.3±254.4	1665.2±288.7	2107.3±443.2	606.5±177.2	1148.9±148.1
Total Benthos (g m^{-2})	2.14±0.5	3.07±0.56	3.58±0.69	1.12±0.27	2.02±0.25

1.3.3.2. Long-term trends in the Lake Superior benthic community

We found declines in the major groups of nearshore benthic invertebrates in 2022 compared to the previous surveys in 2016 and in 1994. These declines were especially pronounced in *Diporeia,* Oligochaeta and Sphaeriidae, and are described below.

Diporeia

In 2022, *Diporeia* density declined significantly by 70% compared to 1994 (571 vs. 1934 m^{-2}, $P < 0.001$, paired *t*-test, Table 1.3) at the subset of 19 historical nearshore stations sampled in 1994, 2000, 2003 and 2016 (Scharold et al., 2004, 2009; Mehler et al., 2018) and was the lowest of all four previous surveys (Fig. 1.2). Negative temporal trends in *Diporeia* density were found at 89% (17 of the 19) historical stations, and 24% of them were significant ($P < 0.05$, Fig. 1.2). The decline in densities was significant throughout all sampled years ($P = 0.0002$, repeated measures ANOVA). *Diporeia* density, however, was not significantly different between 2016 and 2022 (744 vs. 571 m^{-2}, $P = 0.08$, paired *t*-test). Likewise, at other nearshore stations first sampled in 2016 and resampled in 2022, there was no significant difference in *Diporeia* densities between the two years (1140 vs. 1058 m^{-2}, $P = 0.56$).

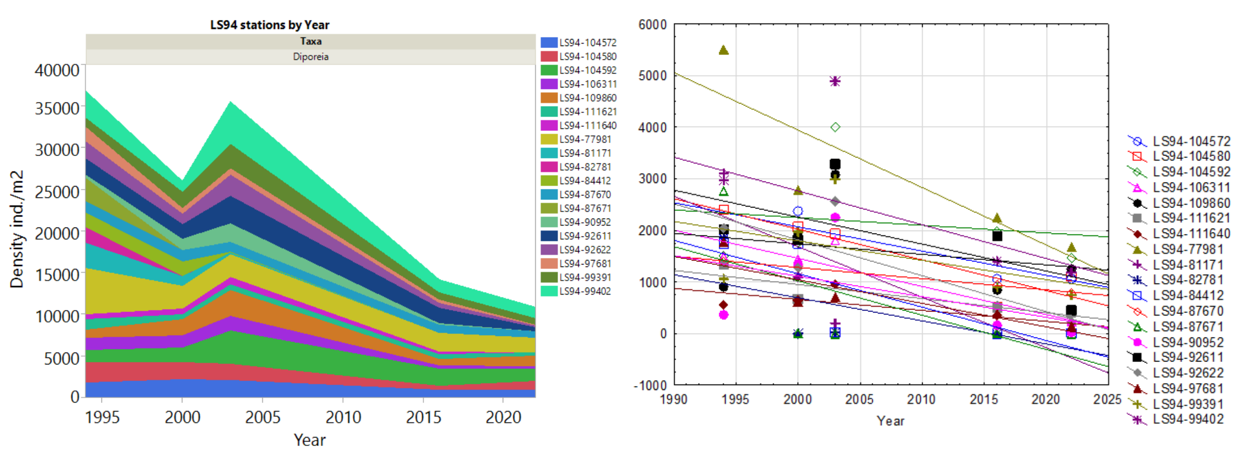

Figure 1.2. Stacked area plots of *Diporeia* densities at 19 stations sampled in 1994, 2000, 2003, 2016 and 2022 (left). Most of the trends in *Diporeia* densities at individual stations were negative (right).

Diporeia abundance declined at deeper stations as well. Pairwise comparison of 17 deep (>100 m) stations sampled previously in 2016 found that *Diporeia* densities in 2022 declined significantly and were 2.8-fold lower than in 2016 (282 vs. 101 m^{-2} in 2022, $P = 0.036$).

Table 1.3. Dynamics of mean (± standard error) densities of major benthic macroinvertebrate taxa in Lake Superior from 1994 to 2022. Density data for 1994 - 2003 are from Scharold et al. (2004, 2009), for 2016 are from Mehler et al. (2018), and 2022 - this report. n/a – data not available.

Depth zone	Stations	Taxa/Year	1994	2000	2003	2016	2022
0-114m	LS94-76401 - 111640	*Diporeia*	1934±261	1377±176	1937±347	744±160	571±130
	(n=19)	Oligochaeta	654±104	594±156	807±169	518±97	328±56
		Sphaeriidae	524±121	456±78	561±99	308±59	223±56
		Chironomidae	81±24	108±28	130±39	90±28	86±25
>90m	SEQ 113-139	*Diporeia*	n/a	n/a	n/a	282±81	101±20
	(n=18)	Oligochaeta	n/a	n/a	n/a	n/a	69±42
		Sphaeriidae	n/a	n/a	n/a	n/a	16±7
		Chironomidae	n/a	n/a	n/a	n/a	0.7±0.5
<90m	BE 01-56	*Diporeia*	n/a	n/a	n/a	1140±165	1058±187
	(n=16)	Oligochaeta	n/a	n/a	n/a	723±141	406±78
		Sphaeriidae	n/a	n/a	n/a	364±75	311±88
		Chironomidae	n/a	n/a	n/a	55±8	46±11
51-90	HN040 (n=1)	*Diporeia*	n/a	n/a	3870	4041±113	3582±637
>90	HN050 (n=1)	*Diporeia*	n/a	n/a	2976	2059±166	3748±236

In contrast, no temporal trends in *Diporeia* densities were found at the 11 GLBMP long-term monitoring stations from 1998 to 2022 (Fig. 1.3). Most of these stations are located >90 m deep except stations SU22 and SU23, which are 54 and 62 m deep, respectively.

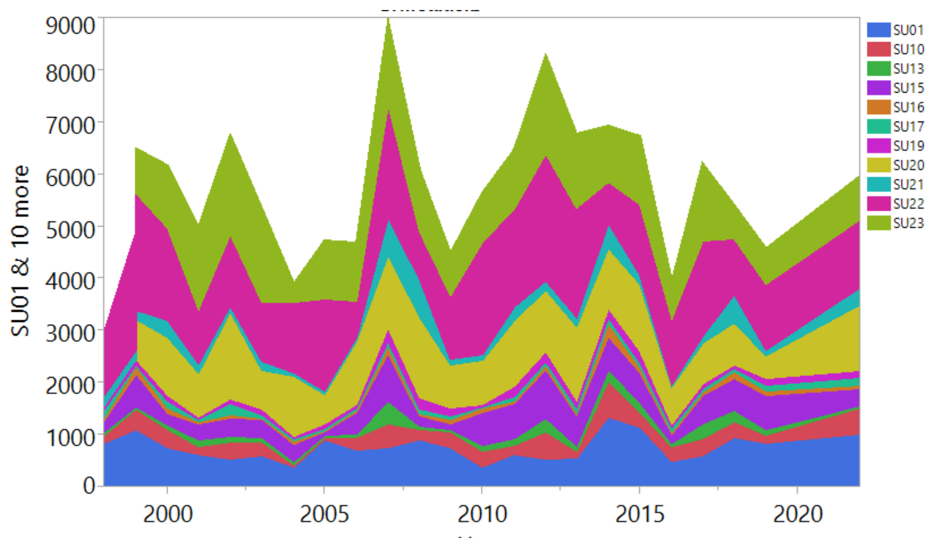

Figure 1.3. Stacked area plots of *Diporeia* densities at all 11 GLBMP long-term monitoring stations sampled from 1998 to 2022. No significant temporal trends in *Diporeia* densities were found at these stations.

The two additional stations in the south-central region near Houghton, MI were characterized by previously elevated *Diporeia* densities (simple mean average density (± standard deviation): 1143 ± 1311 m^{-2} between 2000 and 2009, Auer et al., 2013). At these two stations mean *Diporeia* densities were still high in 2003, 2016 and 2022 (Table 1.3).

Eighty-two percent (54 out of 66) of the 2022 CSMI and GLBMP sampling stations had *Diporeia* densities exceeding indicator threshold of 220 to 320 m^{-2} in nearshore areas of less than 100 m and 30-160 m^{-2} at greater depths (EPA and ECCC, 2022). In 2022, 79% of stations exceeded indicator threshold at depths <100 m, and 86% of stations > 100 m. The proportion of the 25 historical nearshore stations that fell below indicator threshold increased from 0% in 1994, to 10 – 15% in 2000 - 2003 (Scharold et al., 2009), and to 20% in 2016 (Mehler et al., 2018). In 2022, 37% (12) of 19 sampled historical stations fell below the indicator threshold and *Diporeia* was not found at 2 of these stations.

Diporeia was historically the dominant component of the Great Lakes benthic communities that played an important role in the ecosystem (Cook and Johnson, 1974) contributing from 22 to 80% to the total benthic density in Lake Superior in the 1970s (Cook, 1975). A substantial increase in *Diporeia* abundance in U.S. nearshore waters of Lake Superior was found in 1990s. The mean abundance of *Diporeia* in 1973 at the 26 Cook stations that were comparable by location and depths to the stations sampled in 1994 (243 ± 55 m^{-2}) increased eight-fold to 1,990 ± 208 m^{-2} (Scharold et al., 2004). This increase in *Diporeia* abundance did not appear to result from an increase in food supply but rather a decline in predation (Scharold et al., 2004). Bottom trawl surveys conducted by the United States Geological Survey revealed substantial declines in important bottom fish predators of *Diporeia* from 1970s and 1980s slimy, spoonhead and deepwater sculpins, nine-spined stickleback, burbot, rainbow smelt, and bloater (Gorman et al., 2002; Bronte et al., 2003). Another factor contributing to increases in *Diporeia* populations from the 1970s to the 1990s in the western part of Lake Superior could be reductions in harmful contaminants released into the lake (Scharold et al., 2004). Increases in *Diporeia* abundance in

1960-1980s were found in other Great Lakes as well. In Lake Michigan, *Diporeia*, Oligochaeta, and Sphaeriidae increased in density in nearshore waters (<50 m) during 1964–1980, when phosphorus loading was presumably increasing, and declined in the nearshore in the next decade when phosphorus loading was decreasing (Mehler et al., 2020). Increased *Diporeia* abundance in southern nearshore Lake Michigan from the 1960s to the 1980s was also attributed to reduced fish predation (Nalepa, 1987). The large increase in abundances of *Diporeia* in Lake Ontario in the 1980s could have been caused by predation release due to declines in sculpins (Barton and Anholt, 1997).

Currently, *Diporeia* densities are lower in the nearshore zone of Lake Superior compared to the 1990s and 2000s; nevertheless, the current *Diporeia* abundance is still almost twice as high than in the 1970s (570 vs. 260 m^{-2} at depths <90 m, Cook, 1975). The reasons for declines compared to the 1990s and 2000s are unknown, but they likely not due to fish predation given recent bottom trawl survey results indicating that total annual fish biomass in bottom trawls was almost three times lower in 2022 than in 1990s, with rainbow smelt, bloater and sculpins biomass being lower than at the end of 1970s (Vinson et al., 2022).

The decline in *Diporeia* abundance in 2016 and 2022 compared to previous studies calls for further investigations. The GLNPO Biology Monitoring Program has been effective at detecting negative trends in *Diporeia* abundance in the past (Barbiero et al., 2011). To increase the number of monitoring stations in the nearshore zone, GLBMP is currently monitoring *Diporeia* and other benthic groups annually at several of these historical stations since 2016. Further monitoring of nearshore historical stations is essential to determine whether the current change in *Diporeia* and other major groups in Lake Superior indicate a long-term trend in the decline in the nearshore benthos.

Other major groups

Oligochaeta accounted for 23% of total benthos densities and 29% of biomass in 2022, comparable to the previous survey (>30% of both density and biomass in 2016). At the 19 historical nearshore stations, mean Oligochaeta density declined two-fold compared to 1994 and by 37% compared to 2016 (Table 1.3, Fig. 1.4). In 2022 *Diporeia* density was the lowest of all five surveys. Although the decline in densities between 1994 and 2022 was significant ($P = 0.007$, paired *t*-test), the declining trend through the years was only marginally significant ($P = 0.066$, repeated measures ANOVA). A significant decline in Oligochaeta density was also found at nearshore stations sampled both in 2016 and 2022 ($P = 0.022$, paired *t*-test). In contrast, there were no significant temporal trends in Oligochaeta densities at mostly deep (>50 m) GLBMP stations from 1998 to 2022.

Sphaeriidae (mainly *Pisidium*) accounted for 15% of total density and 10% of total biomass in 2022, similar to 2016 (16% and 10%, respectively). The densities in 2022, however, were the lowest of all five surveys at the historical nearshore stations. We found a significant 27% decline in Sphaeriidae at historical nearshore stations compared to 2016 (223 vs. 308 m^{-2}, $P = 0.029$, paired *t*-test, Fig. 1.4), over two-fold decline compared to 1994 (223 vs. 524 m^{-2}, $P = 0.019$), and overall declining trend in densities measured from 1994 to 2022 (repeated-measures ANOVA, $P < 0.0001$, Table 1.3). There was no decline in Sphaeriidae at the rest of nearshore stations previously sampled in 2016 (364 vs. 311 m^{-2}, $P = 0.38$, paired *t*-test), and no significant temporal trends in Sphaeriidae densities at GLBMP stations from 1998 to 2022, indicating that the changes might have occurred mainly in the nearshore zone. As in case with *Diporeia*, and Oligochaeta, the reasons behind the declines in Sphaeriidae are unclear.

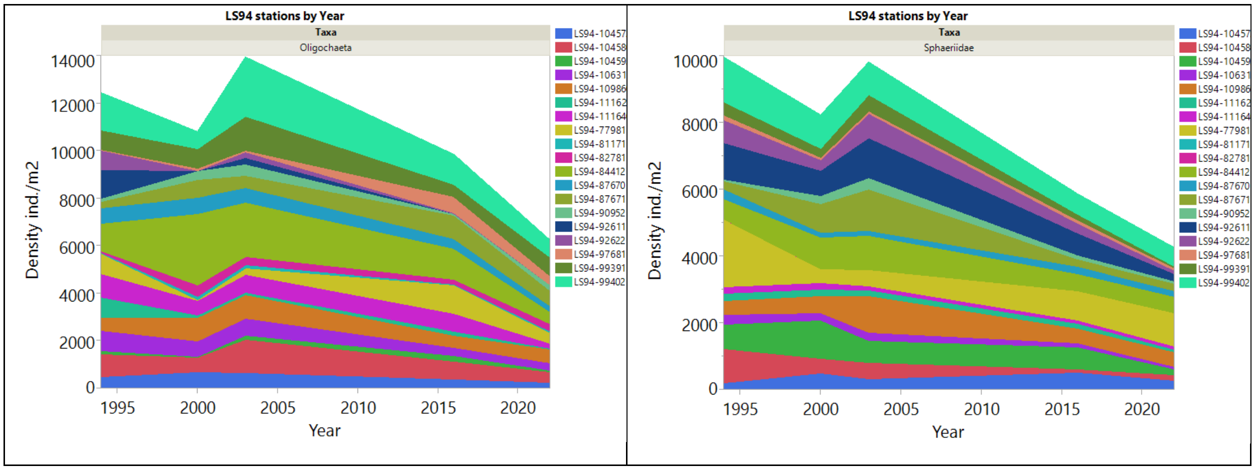

Figure 1.4. Stacked area plots of Oligochaeta (left) and Sphaeriidae (right) densities at 19 nearshore stations sampled in 1994, 2000, 2003, 2016 and 2022 (left).

Chironomidae represented a minor component (4% by density and 2% by biomass) of the Lake Superior benthic community in 2022, similar to the 2016 survey (5% by density and 2% by biomass). From 1994 to 2022, densities of Chironomidae at the 19 historical stations did not change (P = 0.64, repeated measures ANOVA, Table 1.3). Likewise, there were no significant changes in the densities of Chironomidae from 2016 at the rest of the nearshore stations (P = 0.52, paired t-test), and at the 11 GLBMP long-term monitoring stations since 1998. Average lake-wide densities of bottom-dwelling *Mysis* in 2022 (Table 1.2) were comparable to densities recorded in 2016 (6.2+1.5 m^{-2}).

1.4. Resources and Products

A manuscript is being prepared for a peer-reviewed publication on long-term trends in Lake Superior benthic community. Long-term monitoring data are available from the EPA's Great Lakes Environmental Database (US EPA 2023). Additional data are available by request.

1.5. Acknowledgements

This study was funded by US EPA through the Great Lakes Restoration Initiative via a Cooperative Agreement with Cornell University, Department of Natural Resources under Award GL00E03089 "Great Lakes Biology Monitoring Program: Zooplankton, Mysis, Benthos 2022-2027" (PI J. Watkins) and Subaward # 82839-10916 to SUNY Buffalo State. We appreciate the assistance of the captain and crew of the US EPA R/V *Lake Guardian* and SUNY Buffalo State research scientist Allison Hrycik in sample collection**.** We thank Great Lakes Center technicians K. Hastings, E. M. Hartnett, B. Tulumello, S. Geary, A. Tulumello, N. Mikulska, and student technicians M. Basista, K. Glenn, K. Albayed and Y. Mikulska for help with sample processing. We are grateful to L. Denecke for preparing the map, to J. Scharold (US EPA Mid-continent Ecology Division), A. Scofield and E. K. Hinchey (U.S. EPA GLNPO) for reviewing this report, and Great Lakes Center Administrative Assistant Susan Dickinson for proofreading the report. Any views expressed in this report are those of the authors and do not necessarily represent the views

or policies of the US EPA. Any use of trade, product or firm names is for descriptive purposes only and does not imply endorsement by the US EPA.

1.6. Literature Cited

Auer, M. T., Auer, N. A., Urban, N. R., and Auer, T. 2013. Distribution of the amphipod *Diporeia* in Lake Superior: The ring of fire. Journal of Great Lakes Research 39: 33-46.

Barbiero, R. B., Schmude, K., Lesht, B. M., Riseng, C. M., Warren, G. J., and Tuchman, M. L. 2011. Trends in Diporeia populations across the Laurentian Great Lakes, 1997-2009. Journal of Great Lakes Research 37: 9-17.

Barton, D. R., and Anholt, B. 1997. The macrobenthos of Lake Ontario during 1964 to 1966, and subsequent changes in the deepwater community. Aquatic Sciences 59: 158-175.

Bronte, C. R., Ebener, M. P., Schreiner, D. R., DeVault, D. S., Petzold, M. M., Jensen, D. A., Richards, C. and Lozano, S. J. 2003. Fish community change in Lake Superior, 1970-2000. Canadian Journal of Fisheries and Aquatic Sciences 60: 1552-1574.

Cook, D.G. 1975. A preliminary report on the benthic macroinvertebrates of Lake Superior. Environment Canada, Fisheries and Marine Service, Burlington, ON. Technical Report No. 572.

Cook, D. G., and Johnson, M.G. 1974. Benthic macroinvertebrates of the St. Lawrence Great Lakes. J. Fish. Res. Board Can. 31:763–782.

Gorman, O.T., Hoff, M.H., and Cullis, K. 2002. Status and trends in the Lake Superior fish community, 1978–2001. In Superior State of the Lake Conference Program and Abstracts, p. 20.

Mehler, K., Burlakova, L. E., Karatayev, A. Y., and Scharold, J. 2018. Major Findings from the CSMI Benthic Macroinvertebrate Survey in Lake Superior in 2016 with an Emphasis on Temporal Trends. Lake Superior Benthos: Cooperative Science and Monitoring Initiative, Final Report. USGS-GLRI G14AC00263. Great Lakes Center, SUNY Buffalo State, Buffalo, NY. Available at: https://greatlakescenter.buffalostate.edu/sites/glc/files/documents/LakeSuperiorBenthosSurveyCSMI2016FinalReport.pdf

Mehler, K., Burlakova, L. E., Karatayev, A. Y., Elgin, A. K., Nalepa, T. F., Madenjian, C. P., & Hinchey, E. 2020. Long-term trends of Lake Michigan benthos with emphasis on the southern basin. Journal of Great Lakes Research 46, 528-537.

Nalepa, T. F. 1987. Long term changes in the macrobenthos of southern Lake Michigan. Can. J. Fish. Aquat. Sci. 44, 515-524.

Scharold, J. V., Lozano, S. J., and Corry, T. D. 2004. Status of the amphipod *Diporeia* sp. in Lake Superior, 1994-2000. Journal of Great Lakes Research 30: 360-368.

Scharold, J., Lozano, S. J., and Corry, T. D. 2009. Status of benthic macroinvertebrates in southern nearshore Lake Superior, 1994-2003. In: Munawar, M. and I. F. Munawar (eds.) 2009. State of Lake Superior. Aquatic Ecosystem Health and Management Society. Ecovision World Monograph Series, p. 473-492.

EPA and ECCC, 2022. State of the Great Lakes 2022 Technical Report EPA 905-R-22-001. Great Lakes National Program Office, U.S. Environmental Protection Agency, Chicago, IL.

Vinson, M. R., Evrard, L. M., Gorman, O. T., Rosinski, C., and Yule, D. L. 2019. Status and Trends in the Lake Superior Fish Community. Report, U.S. Geological Survey Great Lakes Science Center Lake Superior Biological Station. Available at: http://www.glfc.org/pubs/lake_committees/common_docs/2019LSPreyfishReport_v9_FINAL_tbell-edits.pdf

Vinson, M. R., Yule, D. L., Evrard, L. M., and Phillips, S. B. 2022. Status and Trends in the Lake Superior Fish Community, 2022. Report, U.S. Geological Survey Great Lakes Science Center Lake Superior Biological Station. Available at: http://www.glfc.org/pubs/lake_committees/common_docs/LS_2022_FishSurveyReport.v4.FINAL.pdf

US EPA (2021). SOP LG406, Standard Operating Procedure for Benthic Invertebrate Field Sampling, Revision 14, January 2021. Great Lakes National Program Office, U.S. Environmental Protection Agency, Chicago, IL.

US EPA (2021a). SOP LG407, Standard Operating Procedure for Benthic Invertebrate Laboratory Analysis, Revision 10, March 2021. Great Lakes National Program Office, U.S. Environmental Protection Agency, Chicago, IL.

U.S. EPA (2023). Great Lakes National Program Office. EPA_GLBMP_Benthic Macroinvert Data 1998-2022_2023XXXX_V1 [Microsoft Excel file]. Retrieved from https://cdx.epa.gov/