ENVIRONMENT AND CLIMATE CHANGE CANADA

Great Lakes Studies

CRUISE REPORT

Microbial Ecology of the Lake Erie Ecosystem (MELEE) Cruise

CRUISE NUMBER:	2016046 - 001 – 008
DATES:	February 15th – 19th, 2016
REGION:	Lake Erie
VESSEL:	CCGS GRIFFON
COMMENDING OFFICER:	Capt. M. Head
STUDY LEADER:	Dr. S. Watson
OPERATIONS OFFICER:	B. Lalonde

Date of issue: March 4, 2016

CCGS GRIFFON

Winter MELEE

20160215 – 01 - 008

February 15th	Departed – Amherstburg 1530 hours
February 19th	Arrived – Port Colborne 1215 hours

Personnel:

Mr. B. Lalonde	A/Operations Officer
Mr. J. McVea	Technical Operations Services Research Support Branch
Ms. C. Watson	Aquatic Ecosystem Management Research
Mr. J. Guo	Division, WS&T
Mr. D. Depew	
Mr. A. Zastepa	
Ms. J. Fernandez	University of Cincinnati
Mr. R. M. McKay	Bowling Green State University
Mr. M. Twiss	Clarkson University
Mr. D. Smith	Potsdam, NY
Mr. A. B. Scott	Trent University, ON

The project is a collaboration between Canadian and American researchers representing Government and academic interests**.**

PURPOSE:

Characterize the spatial and vertical distribution in Lake Erie of
i) benthic seed populations
ii) physico-chemical parameters (temperature, conductivity etc.) and major nutrients in water column
iii) primary productivity, carbon processing and bacterial activity
iv) algal nutrient and physiological status; toxins
v) phytoplankton, picoplankton and algal taxa; samples for DNA barcoding and metagenomic analyses

RELEVANCE:

Key component of WS&T GLNI workplan to evaluate harmful algal and plankton communities across nutrient gradients, along inshore-offshore transects and areas of high and low urban/industrial impact in the lower Great Lakes, and evaluate the use of remote imaging, fluorescence, molecular and chemotype biomarkers as tools to detect differences in community composition, biological activity, toxigenicity and stress.

CRUISE OUTLINE:

The outline is as per the attached station positions and schedule of observations and analyses.

SCHEDULE OF OBSEVATIONS AND ANALYSES:

At all stations the following tasks performed:

1. **Temperature** - At all stations, a surface temperature observation was taken.

2. **Secchi Disc** - At all stations during daylight hours, a 30 cm Secchi disc observation was taken from the shaded side of the ship.

3. **pH, Dissolved Oxygen and Conductivity** - At all stations, determination of pH, dissolved oxygen, and conductivity of water collected from 1 m, bottom minus 1m. pH was calibrated once a day, D.O. was calibrated before every station.

4. **Hand-Deployed Profiler** – At all stations a YSI, Fluoroprobe, Phytoflash and PAR cast was performed.

5. **Hand-deployed small net samples** – At selected stations, nets were hauled to obtain sufficient material for inspection/ID and imaging using an EVOS

microscope for community composition. Samples were placed in plastic bottles and stored at 4°C for later isolation.

6. **Hand-deployed large phytoplankton net samples** – At selected stations, nets were hauled to obtain sufficient material for inspection/ID and imaging using an EVOS microscope for community composition. Subsamples preserved with Lugols (20mL vials) and live isolation (plastic bottles in fridge); remaining sample placed in plastic bottles and stored at -20 °C for metabolite analysis (-80 °C in lab). Two additional net hauls for isotopes (Depew); Two additional hauls for BGSU/Clarkson.

7. **Hand-deployed zooplankton net hauls 150µm mesh** – At selected stations, nets were hauled to obtain sufficient material for: DNA (Watson) plus 1 additional haul for Depew, two additional hauls for BGSU/Clarkson and at sites 970, 880 and 879- one additional tow for Trent.

8. **APMM (Autonomous Phytoplankton Metabolic Monitor)** – At selected stations, water was sampled to measure community photosynthesis and respiration.

Water Sampling

A. Water samples were taken for the following WQ parameters at 1 metre using a 30 L Niskin.

1. Unfiltered phosphorus; NLET vial filled exactly with 10 mL of raw water, stored at 4°C.

2. Filtered phosphorus (TP-Dissolved) cellulose acetate 0.45µm filtrate, NLET vial filled exactly with 10 mL, stored at 4°C.

3. Dissolved nutrients (SRP, NO2/3, NH4, TKN-F) cellulose acetate 0.45µm filtrate, stored in Boston round 100mL bottles, stored at 4°C.

4. Major Ions (Cl, Ca, SO4) and SiO2; cellulose acetate 0.45µm filtrate in 250mL bottle, stored at 4°C.

5. DIC/DOC (Carbon) cellulose acetate 0.45um filtrate stored at 4°C.

6. Chla filtered on GF/F (2) and GF/C (2) filters, frozen at -20C.

7. Seston/TSS filtered through pre-ashed pre-weighed Ready to USE (934-AH RTU) GF filters, stored in petri plates at -20C; volume recorded on petri plate and in seston sample record.

8. POC/PON (Carbon and Nitrogen); collect 250 mL on pre-ashed 25mm GF/C filters, add 2-3 drops of 0.3% H_2SO_4 when 25 mL remains to be filtered. Stored in petri plates in -20C freezer.

9. Particulate phosphorus (TP_Particulate GF/C) 100mL (or more in oligotrophic water) collected on pre-ashed 25mm GF/C filters preserved with 1mL 30% H_2SO_4 in 100mL glass bottle with volume filtered recorded on bottle, stored at 4°C. 100mL reagent grade distilled water added prior to submission WQMS to provide.

10. Phycocyanin filtered onto GF/F and frozen at -80C in cryovials.

11. Suspended Si - from 250mL or more volume; collected on 1.2µm polycarbonate membrane filters, folded particulate surface to particulate surface to avoid loss of material and stored in petri plates at -20C.

12. Phytoplankton in 125 mL boston round glass bottle, preserved with 2% Lugol's.

13. Picoplankton (x 2) in 1mL cryovial pre-spiked with 50 µL of 10% paraformaldehyde (0.5% final conc). Allow fixation for 1 – 8 h in refrigerator (4°C). Store at -80C.

14. Photosynthetic efficiency: ~15mL each of 0.45 filtrate and whole water immediately dark adapted in pre-rinsed glass tubes (note start time on the tube for dark incubation); analyzed on board by PhytoPAM.

15. Cyanotoxins: >1000 mL filtered onto duplicate GF/C (or more in oligotrophic water), frozen in 50 mL falcon tubes at -20°C.

16. Toxins-GFC-150mm: Filter several liters onto 150 mm GF/C (1.2 um) using peristaltic pump. Frozen in 50 mL falcon tube at -20°C.

17. Metabolites: >1000 mL filtered onto duplicate GF/C (or more in oligotrophic water), frozen in 50 mL falcon tubes at -80°C

18. DNA: use 6% bleach to presoak plankton collection containers followed by several rinses in sterile distilled water before use; these sample bottles are rinsed several times with water from the niskin before taking the final samples. If more than one container is available the soaking time in bleach can be extended.
    i. Plankton (x3): 200-2500mL filtered onto 0.2 um, polyethersulfone hydrophilic filters frozen in 15mL falcon tubes (or 1.5 mL Eppendorf tubes) at -80°C; ii) >1000 mL filtered onto a 0.22 um Sterivex cartridge with peristaltic pump, frozen -80°C.
    ii. Sediment triplicate surface sediment collected from box core and frozen at -80C in sterile 50mL Falcon tubes (fill tube, but leave headspace).

B. Deep chlorophyll layers sampling did not occur

C. Ice-water (2L worth of melted water)

1) Photochemical efficiency: ~15mL each of 0.45 filtrate and whole water immediately dark adapted in pre-rinsed glass tubes (note start time on the tube for dark incubation); analyzed on board by PhytoPAM.

2) Phytoplankton in 125 mL boston round glass bottle, preserved with 2% Lugol's. It is imperative that specific black lids with cone-shaped inserts are used to cap bottle

3) Picoplankton (x 2) in 1mL cryovial pre-spiked with 50 µL of 10% paraformaldehyde (0.5% final conc). Allow fixation for 1 – 8 h in refrigerator (4°C). Store at -80C.

4) DNA as above

D) Water samples were taken for the following WQ parameters at bottom -1metre, unless otherwise indicated using a 30 L Niskin

1) Unfiltered phosphorus; NLET vial filled exactly with 10 mL of raw water, stored at 4°C.

2) Filtered phosphorus (TP-Dissolved) cellulose acetate 0.45µm filtrate, NLET vial filled exactly with 10 mL, stored at 4°C.

3) Dissolved nutrients (SRP, NO2/3, NH4, TKN-F) cellulose acetate 0.45µm filtrate, stored in Boston round 100mL bottles at 4°C.

4) Major Ions (Cl, Ca, SO4) and SiO2; cellulose acetate 0.45µm filtrate in 250mL bottle, stored at 4°C.

5) DIC/DOC (Carbon) cellulose acetate 0.45µm filtrate stored at 4°C.

6) Chla filtered on GF/F (2) and GF/C (2) filters, frozen at -20C.

7) Seston/TSS filtered through pre-ashed pre-weighed Ready to USE (934-AH RTU) GF filters, stored in petri plates at -20C; volume recorded on petri plate and in seston sample record WQMS to provide TSS filters, petri dishes and clean water for blanks and rinsing.

8) Phytoplankton in 125 mL boston round glass bottle, preserved with 2% Lugol's. It is imperative that specific black lids with cone-shaped inserts are used to cap bottle.

9) Photosynthetic efficiency: ~15mL each of 0.45 filtrate and whole water immediately dark adapted in pre-rinsed glass tubes (note start time on the tube for dark incubation); analyzed on board by PhytoPAM.

10) At stations where sediment is obtained the following additional parameters should be preserved according to above instructions.
    - Phycocyanin, cyanotoxins, toxins, metabolites, DNA

Sediment Sampling

At stations, 879, 880, 970, 1057 sediment was collected using a mini box corer.
Attempts at stations 218 and 937 were unsuccessful.
3 core tubes were inserted in the undisturbed substrate.

1) Section each core (top 3 cm)

2) DNA/RNA: Freeze 10 mL of wet sediment in 50 mL falcon tube at -80°C.

3) Chemotyping: Dilute 36 mL of sediment 10X with distilled water and mix 70:30 Ludox solution. Distribute resulting volume into 6x85 mL centrifuge tubes and centrifuge at 400 g at 4°C for 20 mins. Pool and filter supernatants onto GFC filter. Store at -20C.

4) Phytoplankton; 0.5 g into 20 mL glass scintillation vials. Add 20 mL of distilled water and 2 drops of 30% formaldehyde, store at 4°C.

DATA QUALITY ASSURANCE:

Duplicate samples were collected at the 1m depth at two stations for parameters described in items A; Total Phosphorus filtered and unfiltered, Major Ions, Nutrients and DIC/DOC, and filtered samples POC/PON, TSS. Chlorophyl*a* were conducted in duplicates throughout the cruise.

Bottle blanks and method blanks were performed three times during the cruise for parametres described in the duplicate section, with the exception of unfiltered parameters, for which only method blanks were performed. The supplied carboys of MilliQ water was used for the TSS method blanks, to match the volume filtered from the same station.

Method blanks are treated in the same fashion as samples, with the exception that distilled deionized water is used rather than sample water. Method blanks are identified as depth from = 998 on STAR submission forms.

Bottle blanks are sample bottles filled with distilled deionized water, without rinsing, and preservatives are added as required. Bottle blanks are identified as depth from = 999 on STAR submission forms.

Additional work:

Depew, Environment and Climate Change Canada:
^{18}O phosphate isotopes – at stations 1156, 970, 880, 879 and 931 – collect ~ 200L using a marsh pump into collapsible carbuoys, and rigid carbuoys for settling /concentrating.
Water/zooplankton – At selected stations, collect 1 extra zooplankton net haul (150mm mesh) and ~ 2-4 L of water for POC/PON, P and lipids.
Phyto net - At all stations, Phyto net haul samples were collected for isotopes.

Smith, University of Michigan:
CDOM/H2O2 - At all stations, 10 L of water from 1m and bottom – 2m was sampled for CDOM/H2O2 filtration.

Scott, Trent University:
At all stations 10 L of water from surface (1m) and bottom – 2m - was filtered for nutrient a carbon fractions, dissolved organic phosphorus speciation (phospholipids, monophosphoesters and diphosphoesters), DOM age and isotopic composition and phytoplankton pigments
Zooplankton - At stations 970, 880 and 879 an additional zooplankton tow

Mackay/BGSU - Twiss/Clarkson
Use TWISS to grab 1.3 L of surface water; record lat/long/time and sea state. Transfer water to rinsed 2-L PC bottle for transport to lab.

1. **TP**: 2 x 35 mL into 50 mL borosilicate tubes for analysis back in lab at Clarkson University .
2. **Si, NO3**: syringe filtered (<0.2 micron) water into 50 mL PP conical tube for analysis back in lab at Clarkson University.
3. **%T**: used unfiltered water to measure % transmittance using spectrophotometer (5 cm cell).
4. **Phytoplankton community**: syringe filtered 25 mL into FluoroProbe (FP) cuvette to blank instrument for %T and yellow substances; measured community (100 repeated measurements) on FP Workstation 25. Also collected sample (100-200 mL filtered through Sterivex cartridge) for high throughput sequencing.
5. **CDOM**: Measured CDOM on filtered (<0.2 micron) lake water using TD-700.
6. **Size fractionated filtration for chl-a**: 2 x 20 micron, 2 x 2 micron, 2 x 0.2 micron. Froze filters in tubes for analysis back in lab at Clarkson University.
7. **COD**: Chemical Oxygen Demand measured in 2 mL samples in duplicate following digestion; used HACH spectrophotometer and digestion block.
8. Repeated (a-j) as many times as possible.

9. **Net Hauls**: When possible during station visits, conducted duplicate water column phytoplankton (filamentous diatoms)/zooplankton net hauls.
10. **Zooplankton counts**: Contents of one haul was fixed with sugar formalin and used to quantify zooplankton counts
11. Contents of duplicate haul was delivered to separatory funnel and incubated in darkness for 60 min. Diatoms sedimented whereas zooplankton was expected to swim. Diatom fraction was removed and processed for nucleic acid extraction for metatranscriptome analysis. Zooplankton were isolated to determine zooplankton gut contents – thus an approach to address zooplankton grazing on filamentous diatoms.

F. Longstaff, University of Western Ontario

Isotopic Composition - At all stations, 15 mL of water from a depth of 1 m and bottom – 2m when depth was greater than 50 m was collected for isotopic composition.

NARRATIVE

Thursday February 11th, 2016
B. Lalonde, T. Breedon, J. McVea, J. Paynter departed CCIW to Amherstburg, ON with a lab trailer and a cargo trailer transporting scientific equipment to be loaded onto the CCGS Griffon docked at the CG Base. The lab trailer was lifted with the ship's crane and secured to the deck as well as a hydrographic winch. The rest of the equipment was stowed in the tween deck and cargo hold. The TechOps crew returned to CCIW that evening.

Monday February 15th, 2016
TechOps, WHERD and visiting scientists departed CCIW to Amherstburg, ON. The crew embarked the CCGS Griffon and completed administrative issues. The Griffon departed the CG base at 1530 and initiated the Microbial Ecology of the Lake Erie Ecosystem (MELEE) Cruise. The ship arrived at station 1156 at 1630. Considering that this was the first station and that setting up of scientific equipment still needed to be completed a time on station of 3 hours was unusually long but expected.
Upon completion of station 1156, the Griffon headed S-S-E and "parked" in ice approximately a nautical mile off station 970 in order not to contaminate the study area with grey water discharge.
Note (The majority of the sampling effort occurred between the hours of 0800 and 2000 with the assistance of deckhands to operate winches or crane). Some scientists elected to sample that evening given that the parameters and conditions of their studies did not require ship personnel. Scientists from Clarkson and BGSU sampled periodically as the vessel was underway. This procedure required the vessel to slow down to a near halt, so the scientist could toss a specially designed sampler over the side. The Griffon would resume its course upon notification by the study team. This sampling exercise occurred throughout the duration of the cruise.

Tuesday February 16th, 2016
The Griffon repositioned on station 970 at 0800 and sampling started. The cold temperatures attained during the previous four days, ranging from -7°C to -12°C, consolidated the ice cover in the western basin. The Griffon headed to station 966 on the west side of Point Pelee. Upon completion the Griffon went around the point and reached station 218 at 1835, off of Weatley. Station 218 was completed shortly before 2000. In as much as sampling activities ceased at 2000, the decision was made to detour by station 1365 located 13 nautical miles to the east and especially were gas pipelines are located. A minimal volume of water was sampled for some visiting scientists. No water was sampled for ECCC staff. The Griffon headed toward station 880 in the central basin at slow speed.

Wednesday February 17th, 2016
The Griffon anchored at station 880 and sampling started 30 minutes later. The station was completed within 1.5 hours. The Griffon proceeded N-N-E toward station 1057.

This station was sampled in lieu of station 341 (originally in the cruise plan). The following two stations sampled were 1208 and 1210. Once completed the Griffon continued on a variable course around Long Point

Thursday February 18th, 2016
The Griffon arrived at station 937 in Long Point Bay and the station was completed within 1.5 hours. The Griffon headed toward station 879 and deployed the anchor despite the depth of 62 m. Upon completion of the station, the Griffon headed toward station 67 near Port Maitland. The Griffon anchored at station 931 overnight

Friday February 19th, 2016
A layer of ice (3 to 5 cm) kept being pushed by a S-W wind and made it challenging to keep an ice free area near the ship to lower the various sampling equipment at station 931. The station was completed at 0900 and the Griffon headed toward Port Colborne. The ship was secured by 1215. Scientific equipment, hydrographic winch and lab trailer were all unloaded in the afternoon. The scientific crew returned to CCIW by 1600.

CHRONOLOGY OF EVENTS

Monday February 15th
1530 CCGS Griffon departed CG Base Amherstburg, completed fire and boat drill
1635 Arrived station 1156
1910 Departed station 1156

Tuesday February 16th
0900 Arrived station 970
1000 Departed station 970
1200 Arrived station 966
1415 Departed station 966
1835 Arrived station 218
1950 Departed station 218
2120 Arrived station 1365
2145 Departed station 1365

Wednesday February 17th
0830 Arrived station 880
1005 Departed station 880
1335 Arrived station 1057
1450 Departed station 1057
1715 Arrived station 1208
1830 Departed station 1208
1945 Arrived station 1210

Thursday February 18th
0810 Arrived station 937
0940 Departed station 937
1140 Arrived station 970
1410 Departed station 970
1640 Arrived station 931
1845 Departed station 931
2225 Arrived at anchor1 mile off of station 931

Friday February 19th
0730 Raised anchor and arrived station 931
0900 Departed station 931
1215 Arrived Port Colborne

END OF CRUISE

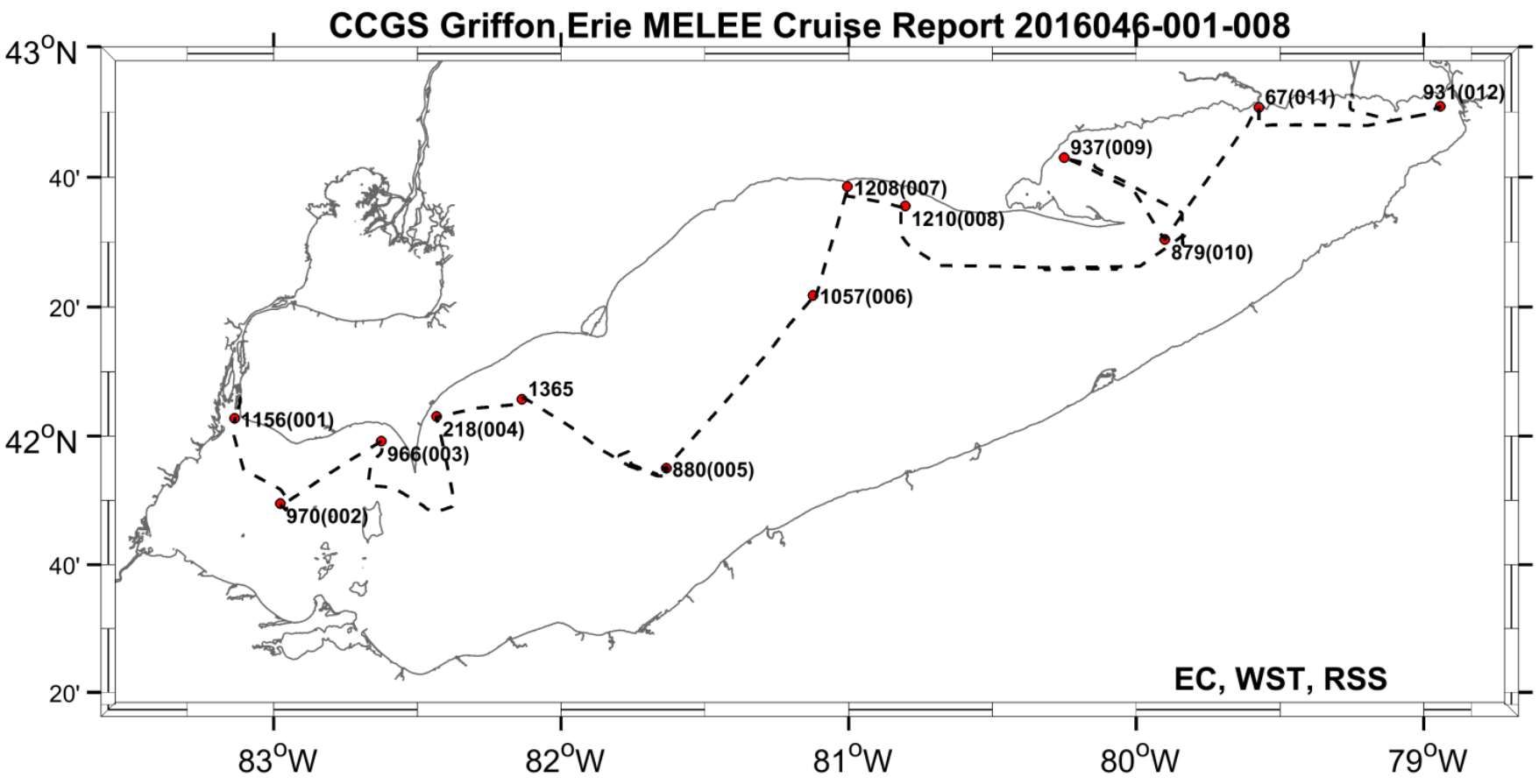
STATISTICS SUMMARY

CRUISE NO.	2016046-001-008	**SHIP**	GRIFFON
DATE:	2016 February 15	**REGION**	Lake ERIE
CRUISE TYPE	MELEE	**N.MI. STEAMED**	346

DESCRIPTION	TOTAL	DESCRIPTION	TOTAL
Stations Occupied	13	Zooplankton Hauls 20 µm	2
YSI casts	12	Zooplankton Hauls 64 µm	16
Fluoroprobe	10	Zooplankton Hauls 80 µm	17
Phytoflash	10	Zooplankton Hauls 153 µm	9
Secchi Disk Observations	9		
Water Samples Collected (D.O.)	27		
Water Samples Collected (Cond/pH)	28		
Water Samples Collected (TP uf)	33		
Water Samples Collected (Isotopic Composition)	12		
Water Samples Collected (Green House gases)	78		
Water Samples Collected (Picoplankton)	24	Cores Taken, Mini Box	9
Water Samples Collected (Phytoplankton)	24		
Water Samples Filtered (TSS)	29		
Water Samples Filtered (Chlorophyll a Watson method)	38		
Water Samples Filtered (Chlorophyll a NLET method)	38		
Water Samples Filtered (Photosynthetic efficiency)	24		
Water Samples Filtered (Cyanotoxins)	24		
Water Samples Filtered (Toxins)	12	Observations, Weather	
Water Samples Filtered (POC/PON)	30		
Water Samples Filtered (Phycocyanin)	12		
Water Samples Filtered (TP f)	33		
Water Samples Filtered (Nutrients)	33		
Water Samples Filtered (Major Ions)	33		
Water Samples Filtered (DIC / DOC)	33	**ONBOARD ANALYSIS**	
Water Samples Filtered (Particulate phosphorus)	10	Manual Chemistry, Tech. Ops.	
Water Samples Filtered (Suspended Si)	12		
Water Samples Filtered (Metabolites)	24		

STATION POSITIONS
LAKE ERIE

STATION NUMBER	LATITUDE N.	LONGITUDE W.
67	42° 50’ 41”	79° 34’ 23”
218	42° 03’ 02”	82° 26’ 01”
879	42° 30’ 24”	79° 54’ 00”
880	41° 55’ 00”	81° 38’ 04”
931	42° 50’ 51”	78° 56’ 33”
937	42° 42’ 59”	80° 14’ 59”
966	41° 59’ 12”	82° 37’ 32”
970	41° 49’ 32”	82° 58’ 35”
1057	42° 21’ 48"	81° 07’ 24"
1156	42° 02’ 45"	83° 08’ 10"
1208	42° 38’ 32”	81° 00’ 14”
1210	42° 35’ 35”	80° 48’ 12”
1365	42° 05’ 42”	82° 08’ 12”


CCGS Griffon Erie MELEE Cruise Report 2016046-001-008
43°N
40'
20'
42°N
40'
20'
83°W
82°W
81°W
80°W
79°W
1156(001)
966(003)
218(004)
1365
970(002)
880(005)
1057(006)
1208(007)
1210(008)
937(009)
879(010)
67(011)
931(012)
EC, WST, RSS