Identification of *Clostridium botulinum* type E in Lake Erie Sediments and Benthic Invertebrates

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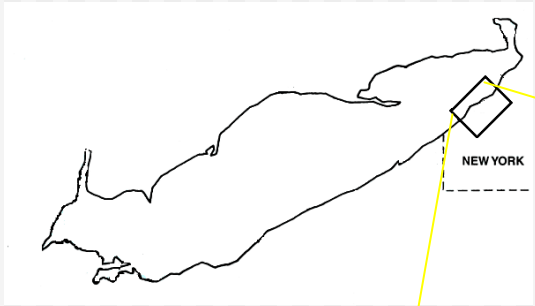
SUNY FREDONIA

Lake Erie Outbreak

- 1999-2002- Large Outbreaks
- Fish and Aquatic Salamanders
  - Freshwater Drum, Smallmouth bass, Lake Sturgeon (threatened species in NYS)
- Birds
  - Common Loons, Red-breasted mergansers, Ring-billed gulls, Long-tailed ducks

Lake Erie Outbreak

- Confined to Eastern Basin
- Smaller Outbreak in 2003
- Minimal reports of mortality in 2004


NEW YORK


GlobeXplorer™
Eastern Lake Erie
*
Offshore (20 m deep)
Dunkirk
Harbor
* Inshore (10 m deep)
van Buren Bay
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SUNY Fredonia Study

- To identify environmental conditions in Eastern Lake Erie associated with the presence of *Clostridium botulinum* type E.

- To determine presence of the botulism bacteria (type E) in sediments and in invertebrate organisms.

Data Collection

- Measure physical and chemical conditions at 0.5 m above sediments

  (D.O., Redox potential, pH, T, Conductivity, Nitrates, Phosphates, Chl *a*)

- Collect sediments and invertebrates to test for presence of the *C. botulinum* Type E bacterium

Isolation of DNA

- Sediments
  - Epicentre SoilMaster DNA Extraction Kit
- Invertebrates
  - Qiagen DNeasy Tissue Kit
- Purified DNA sent to Cornell U. for Q-PCR

Results

- Botulism outbreaks are associated with:
  - Elevated temperatures
  - Low levels of dissolved oxygen
  - Low redox potential
- *C. botulinum* bacteria present in:
  - Sediments
  - Mussels
  - Other aquatic invertebrates

2002 Sediment Data

Date	Site	# DNA Copies/mg Sediment
6-11	Offshore	456
6-28	Inshore	27.6 - 282
8-8	Inshore	215
8-21	Inshore	5520
9-18*	Inshore	16.8

*Pseudofeces

2003 Sediment Data

Date	Site	# DNA Copies/mg Sediment
5-30	Offshore	275
6-30*	Dive 46’	11.4
8-18	Offshore	6.6
8-26*	Dive 43’	15

*Pseudofeces

2003 Invertebrates: Diptera

Date	# DNA Copies/mg sample
6-11	820,000
6-17	<1,660
6-23	312-648 (4)
7-22	76-476 (2)
8-4	705-1550 (2)
9-1	50-1,000 (4)
9-10	13.3-600 (4)

2003 Invertebrates: Oligochaeta

Date	# DNA Copies/mg sample
6/17	<35,400 (2)
7-22	283
8-4	144-444 (3)
9-1	10-372 (5)
9-10	10

2003 Invertebrates: Dreissenids

Date	# DNA Copies/mg sample
5-30	1920
6-17	108
6-23	2280
6-30	23.2
8-26	224

2003 Invertebrates: Other

Date	Organisms	# DNA Copies/mg sample
5-30	Nematodes + Oligochaetes	4000
6-17	Nematodes	56
6-17	Nematodes + Oligochaetes	<1800
6-23	Nematodes	<3088
7-22	Ephemeroptera (Mayflies)	210

Vegetative Cells or Spores?

- *C. botulinum* spores are believed to be widespread
- DNA from bacterial endospores is not likely isolated using gentle conditions (such as those used in this study)
- Experiments are being conducted to determine if endospores are lysed during DNA extractions

Future Studies

- Continue collections
  - Sediments
  - Invertebrates
  - Physical/chemical data

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