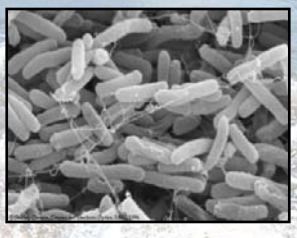
Impact of *Cladophora* mats on *E. coli* concentrations in beach water



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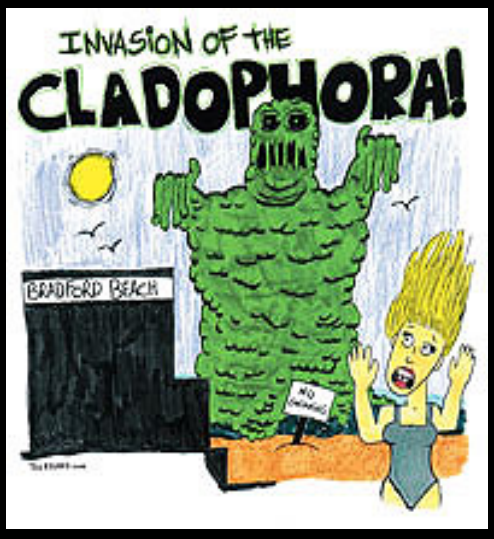
E-mail –mcdermot@uwosh.edu
http://bact.uwosh.edu/



Source: "Unknown"

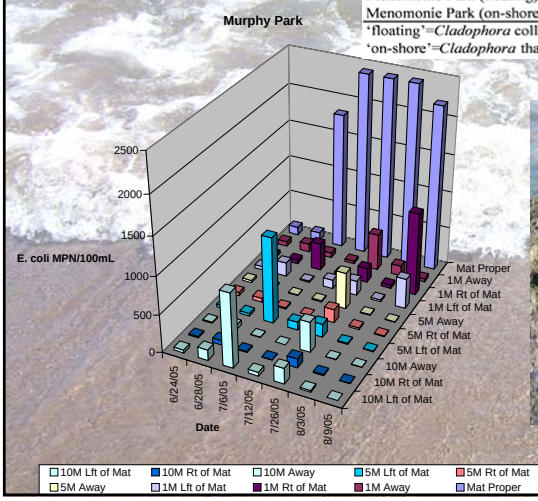
Overview

- Increased Beach Monitoring
- Increased *Cladophora*
- Previous research shows *Cladophora* can harbor large amounts of *E. coli*
- Impact of *Cladophora* on beach water quality?
- Impact of *Cladophora* on pathogens?
- What does it all mean?

Table 1. *E. coli* concentrations recovered from *Cladophora* mats on beaches in Wisconsin.

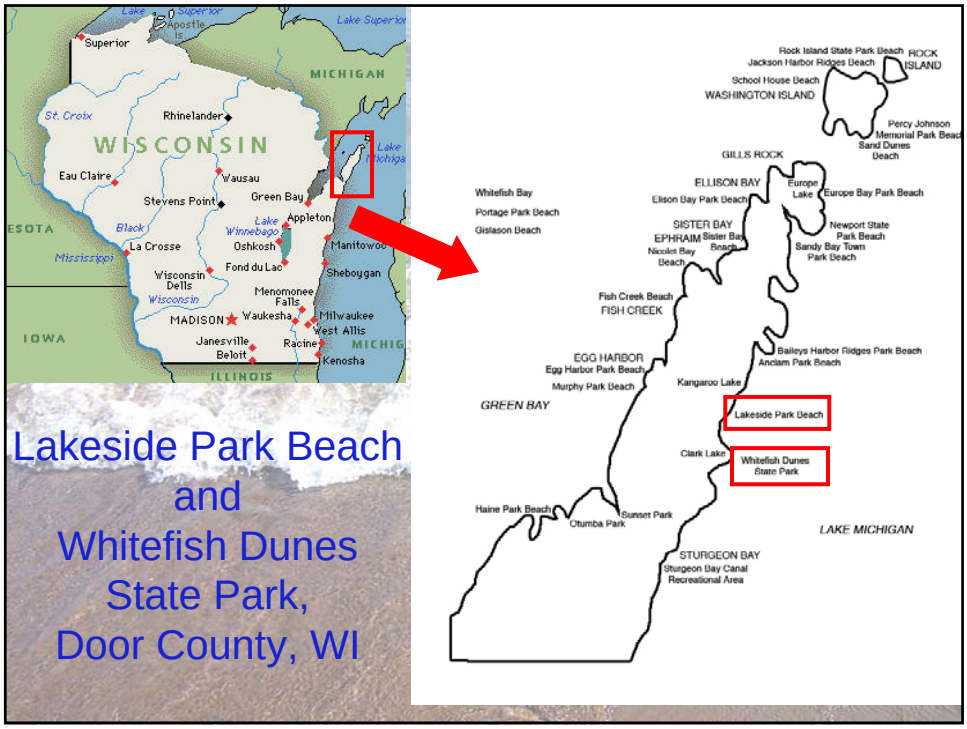
Beach	County	*E. coli* (CFU/g Dry Weight *Cladophora*)	*E. coli* seasonal average in water (MPN/100 ml water)
Murphy (floating)	Door	22,515	244.1
Murphy (on shore)	Door	17,670	244.1
Whitefish Dunes (on shore)	Door	39,501	50.1
Sister Bay (floating)	Door	10,874	89.8
Baileys Harbor (on shore)	Door	1,729	93.8
Sunset (floating)	Door	1,254	165.4
Menomonie Park (floating)	Winnebago	16,790	121.4
Menomonie Park (on-shore)	Winnebago	31,288	121.4

'floating'=*Cladophora* collected from beach water
'on-shore'=*Cladophora* that was on the beach



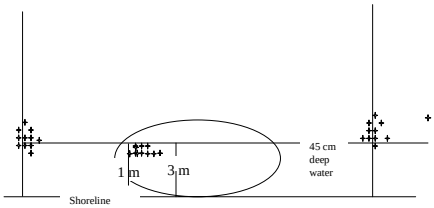
UW Sea Grant Project: *Cladophora* and Beaches 2007,2008

Sea Grant
University of Wisconsin

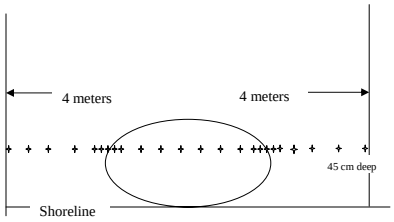

Lake Superior
Superior
Apostle Is.
MICHIGAN
St. Croix
Rhinelander
WISCONSIN
Eau Claire
Wausau
Green Bay
Stevens Point
Appleton
Lake Winnebago
Black
La Crosse
Oshkosh
Manitowoc
Mississippi
Fond du Lac
Sheboygan
Wisconsin Dells
Menomonee Falls
Wisconsin
MADISON
Waukesha
Milwaukee
West Allis
Janesville
Beloit
Racine
Kenosha
IOWA
ILLINOIS
Lakeside Park Beach
and
Whitefish Dunes
State Park,
Door County, WI
Rock Island State Park Beach
Jackson Harbor Ridges Beach
ROCK ISLAND
School House Beach
WASHINGTON ISLAND
Percy Johnson Memorial Park Beach
Sand Dunes Beach
GILLS ROCK
ELLISON BAY
Europe Lake
Europe Bay Park Beach
Whitefish Bay
Portage Park Beach
Gislason Beach
Ellison Bay Park Beach
SISTER BAY
EPHRAIM
Sister Bay Beach
Newport State Park Beach
Sandy Bay Town Park Beach
Nicolet Bay Beach
Fish Creek Beach
FISH CREEK
Baileys Harbor Ridges Park Beach
Anclam Park Beach
EGG HARBOR
Egg Harbor Park Beach
Murphy Park Beach
Kangaroo Lake
GREEN BAY
Lakeside Park Beach
Clark Lake
Whitefish Dunes State Park
Haine Park Beach
Otumba Park
Sunset Park
LAKE MICHIGAN
STURGEON BAY
Sturgeon Bay Canal Recreational Area

Sea Grant Methods

- **Collaborative project – UW-Oshkosh, U.-MN, USGS, and Racine Health Department**
- **Investigate spatial distribution of *E. coli* around *Cladophora* mats and the spatial distribution within mats.**
- **Investigate the occurrence of pathogens associated with mats.**
- **Other questions related to genetic make-up of *E. coli* recovered within mats.**

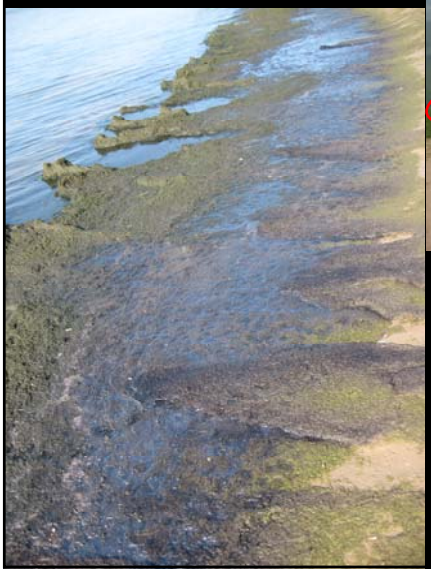


Sampling protocol for Objective 1, Question 1 (Is there a difference in *E. coli* concentrations inside and outside the mat?). Oval = *Cladophora* mat, Star = random sample site location (approximate).



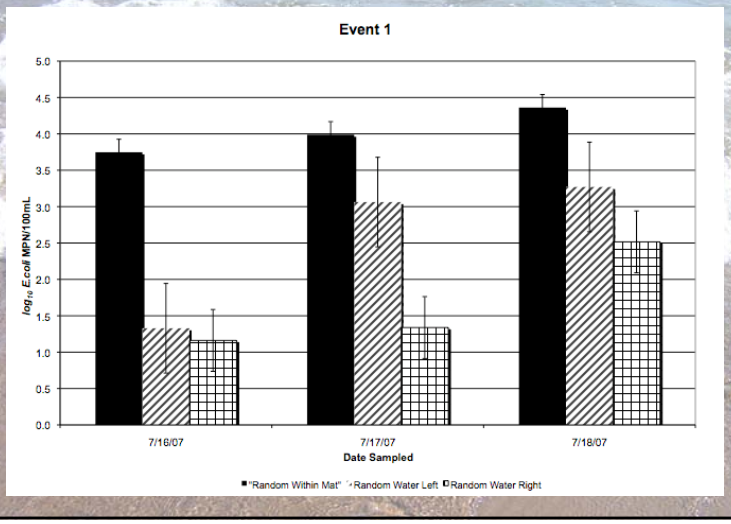
Sampling scheme for Objective 1, Question 2 (Is there an *E. coli* gradient within and outside the *Cladophora* mat?). Oval = mat, Stars = sampling locations.

Whitefish Dunes

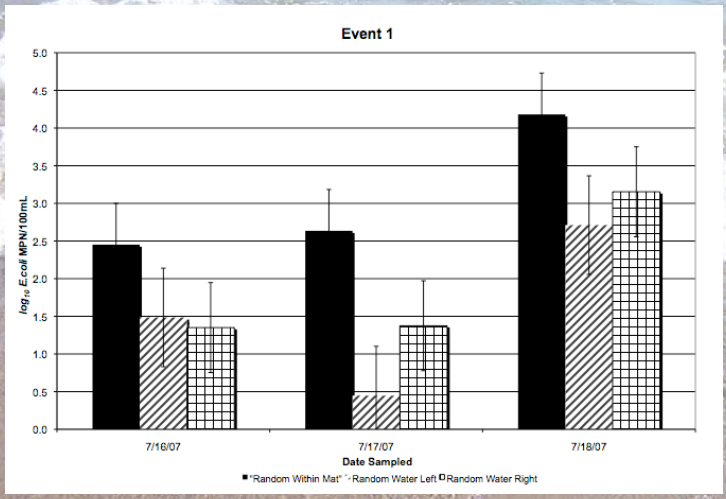


2007

Water samples underlying & adjacent to mat- WFD 2007

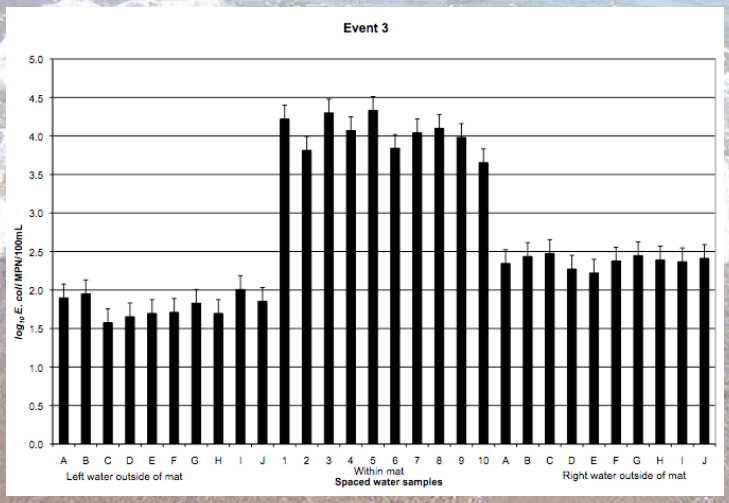


Water samples underlying and adjacent to mat- Lakeside 2007

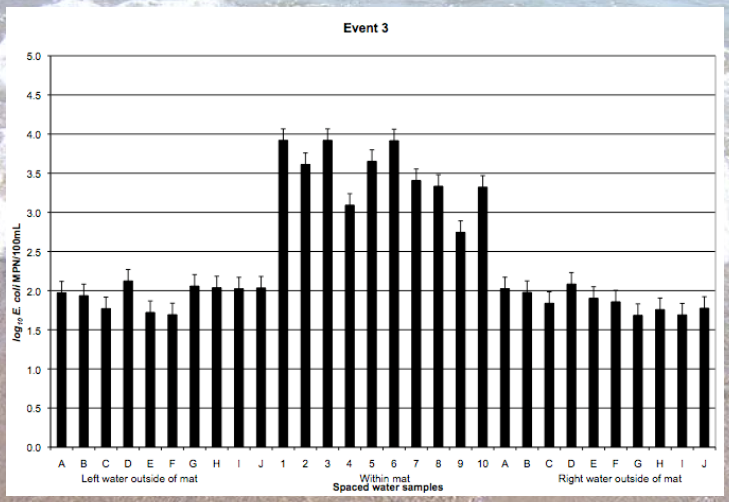


- Elevated concentrations of *E. coli* found in water within mats
- Elevated concentrations of *E. coli* found in water adjacent to mats
- *E. coli* concentrations in water underlying mats increases with time mat is stranded in nearshore water

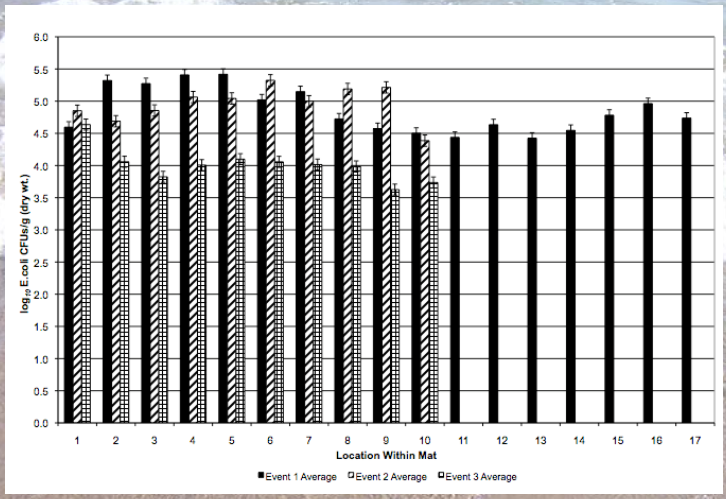
Water samples underlying and adjacent to mat- WFD 2007

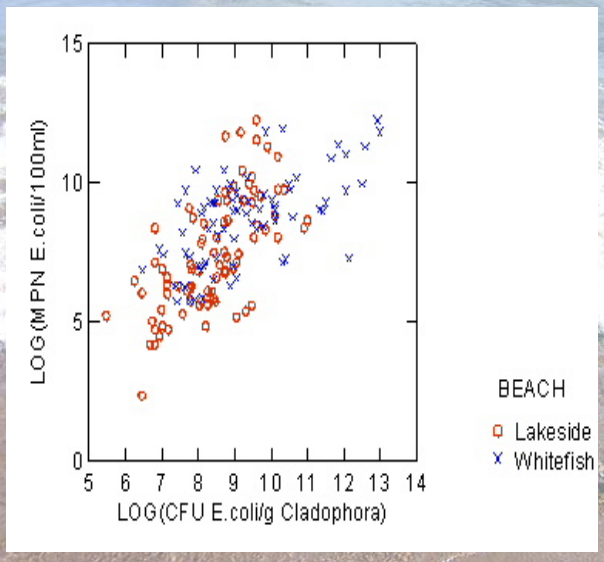


Water samples underlying and adjacent to mat- Lakeside 2007



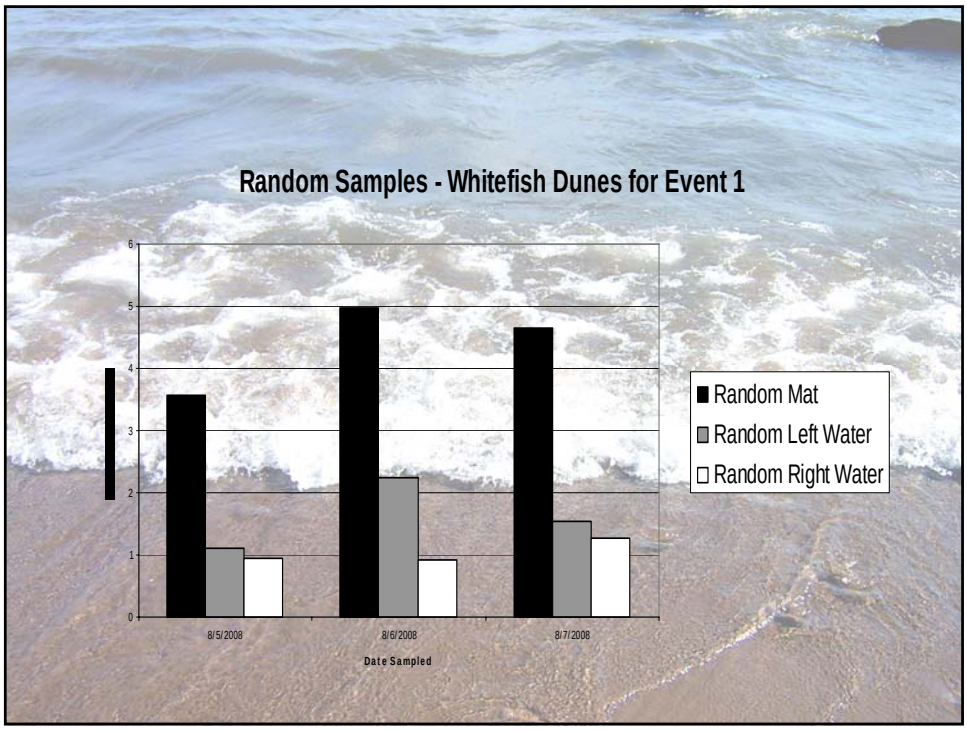
E. coli attached to mat biomass- WFD 2007

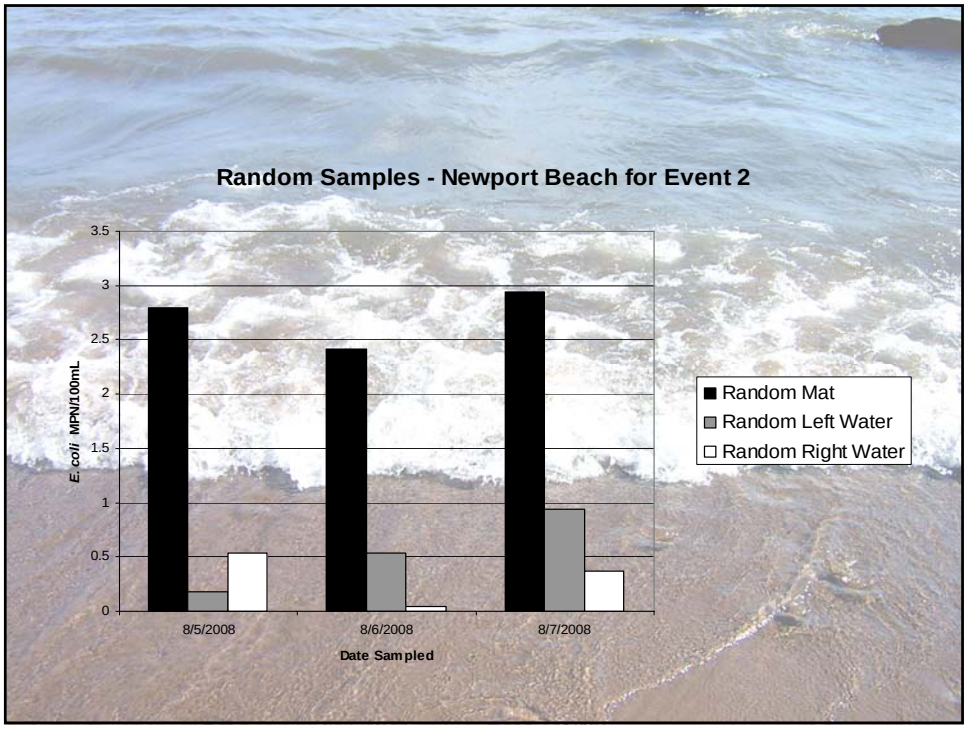


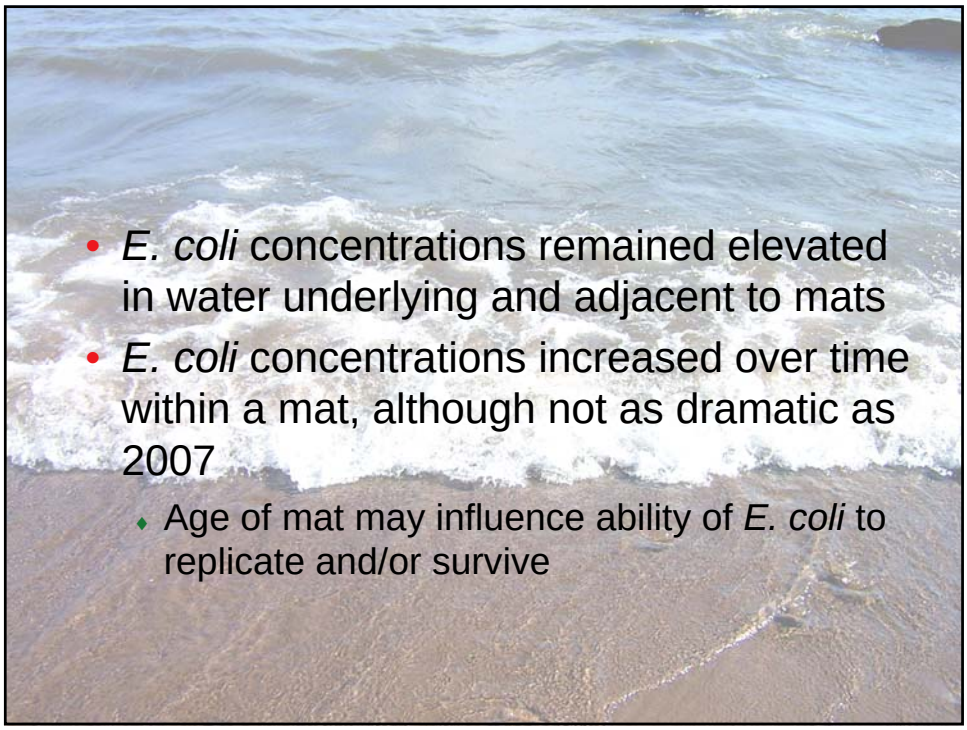


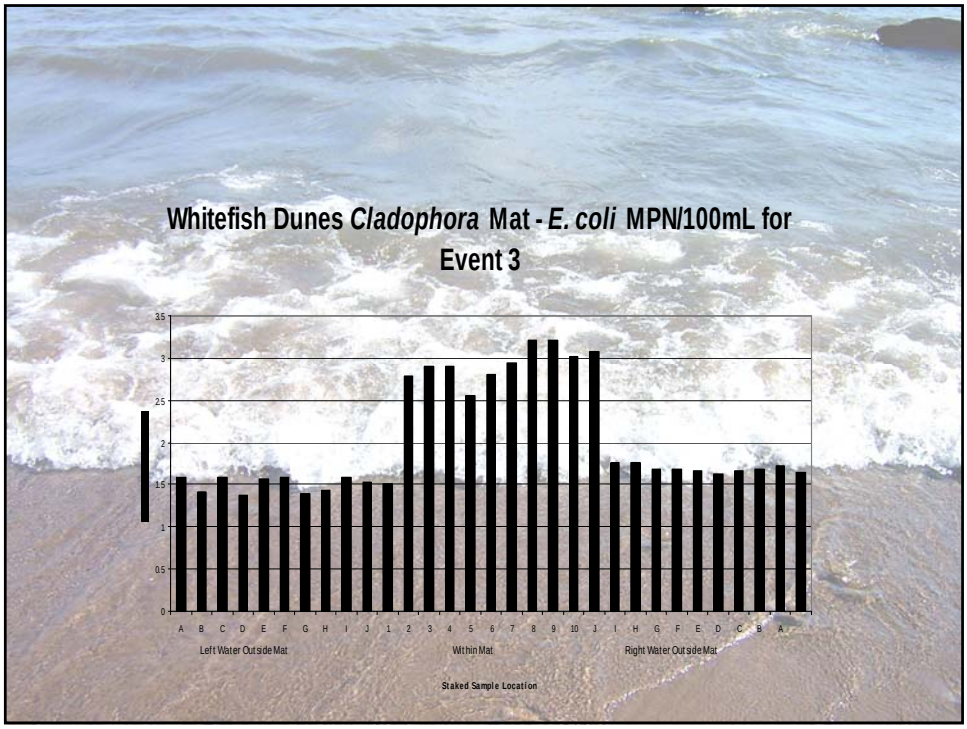
2008

- Wanted to return to same beaches
- Paucity of *Cladophora* for most of summer
- WFD= okay (3 Events)
- Lakeside = little *Cladophora* accumulation
- Added Newport Bay State Park Beach
  - Mat was stationary for most of summer
  - Did not have movement of mat in and out
  - Mat material was “aged”




Random Samples - Newport Beach for Event 2
3.5
3
2.5
2
1.5
1
0.5
0
E.coli MPN/100mL
8/5/2008
8/6/2008
8/7/2008
Date Sampled
Random Mat
Random Left Water
Random Right Water

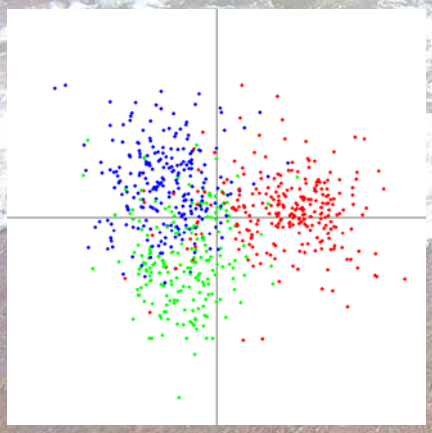

• *E. coli* concentrations remained elevated in water underlying and adjacent to mats
• *E. coli* concentrations increased over time within a mat, although not as dramatic as 2007
◆ Age of mat may influence ability of *E. coli* to replicate and/or survive



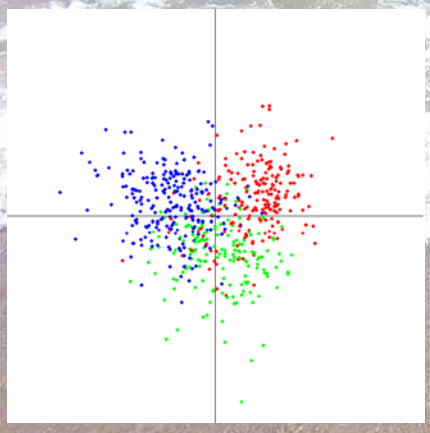
E. coli Genetic Fingerprinting

- To determine if *E. coli* in mat is replicating or if new inputs of *E. coli* are occurring.
- Used the HFERP technique (fluorophore-enhanced, repetitive extragenic palindromic-PCR)
- Jack knife and Manova analysis of fingerprints

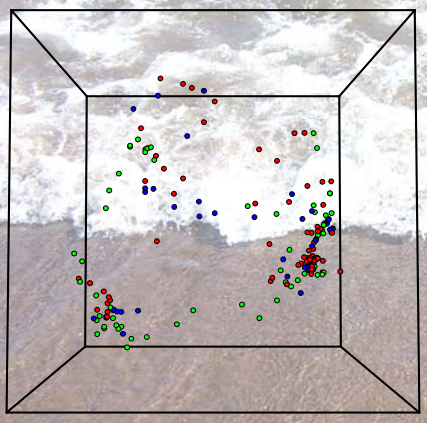
WFD 2007 *E. coli* isolates
All Mat Events



Lakeside 2007 *E. coli* isolates
All Mat Events



WFD 2007 *E. coli* isolates Mat Event #1: MDS



Fingerprinting Summary

- *E. coli* isolates from the same mat are similar from one day to next= replication
- "New" *E. coli* input also occurs day to day
- *E. coli* isolates (from same beach) are not similar from one mat to another


Cladophora impact on pathogens


Pathogens found in *Cladophora* mats
•Measured *Salmonella*, *Shigella* and *Campylobacter* (standard culture techniques) in water from mats and attached to *Cladophora* biomass
•Two of three sampling events in 2007 & 2008 in Door County

Pathogen Results

- Found **no** *Salmonella*, *Shigella*, or *Campylobacter* in water or associated with *Cladophora* in 2007
- Found *Salmonella* and *Campylobacter* in water underlying mat at Whitefish Dunes during **1** event in 2008

What we know

- Have <u>never</u> isolated *Salmonella* nor *Shigella* from water in Door County in previous years
- <u>Have</u> been able to isolate *Campylobacter* from water at beaches in previous years, not associated with *Cladophora* accumulations
- Will use more sensitive molecular techniques to try to identify pathogens in Summer 2009

- Other have consistently found pathogens in association with *Cladophora* mats in southern Lake Michigan (not in Door County) using molecular techniques
- Occurrence of pathogens in mats is likely a matter of input, rather than suitable habit

Conclusions

- ***Cladophora*** **harbors** ***E. coli*** **in high concentrations, allowing for survival and growth within mats. Our data (2007 & 2008) support results published by earlier researchers.**
  - **High** ***E. coli*** **concentrations attached to mat biomass**
  - **High** ***E. coli*** **concentrations in water underlying mat**
  - ***E. coli*** **concentrations within mats increases as mats linger on the beach**
- ***E. coli*** **within mats appears to be "clonal" (i.e. it replicates within mats). More work is needed on the genetic make-up of** ***E. coli*** **found in the** ***Cladophora*** **mats.**
- ***E. coli*** **within mats negatively impacts water quality adjacent to mats.**
- **Additional field data is needed on weather impacts and how mat movement may impact beach health.**
- **Pathogens may not survive to the extent that** ***E. coli*** **does in** ***Cladophora*** **mats and we see no evidence of possible pathogen growth.**
- ***E. coli*** **may not be the best indicator of recent fecal contamination of beach water**
- **Removal of** ***Cladophora*** **mats from beaches should improve water quality by reducing the number of** ***E. coli*** **in adjacent water**

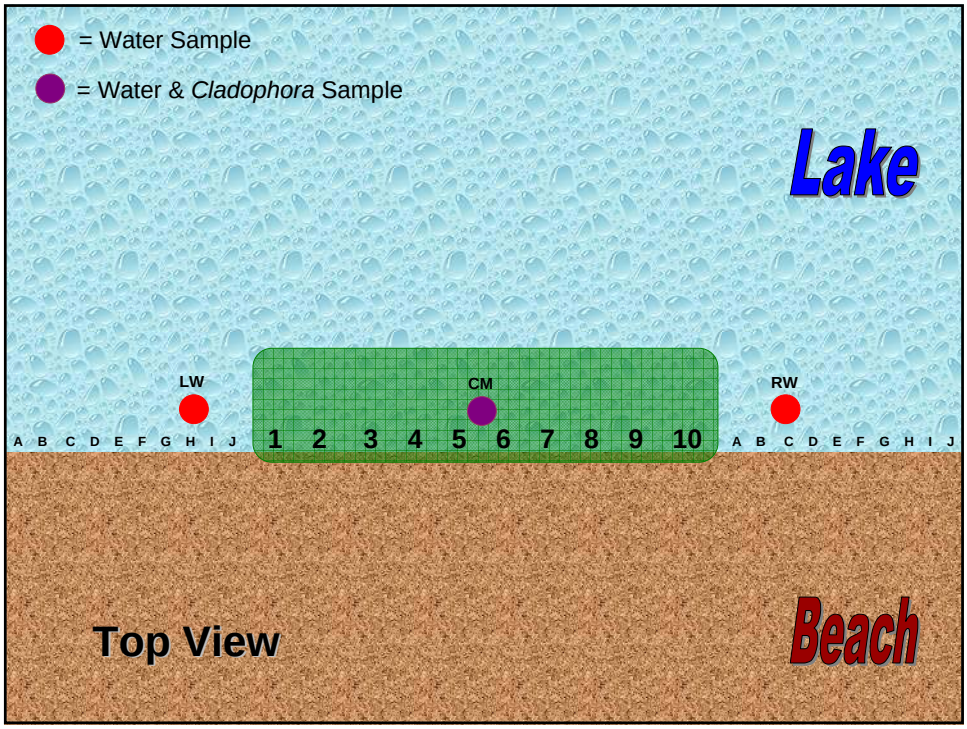
Acknowledgements

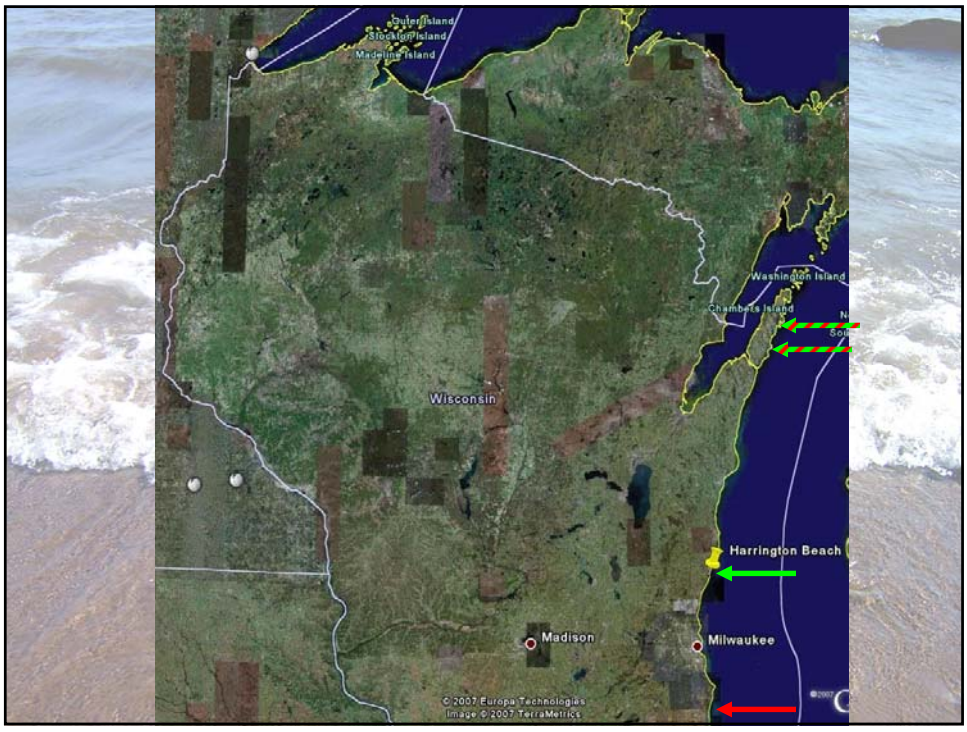
- UW-Oshkosh "Beach Group" – Drs. Todd Sandrin and Bob Pillsbury, Ms. Amy Vanden Heuvel, Mr. Ben Murphy, many other student researchers
- Collaborators:
  - Dr. Mike Sadowsky
  - Dr. Richard Whitman
  - Dr. Julie Kinzelman
- Partners:
  - Door County Soil and Water Conservation Dept.
  - Door County Health Department (Rhonda Kohlberg)
- Funding from:
  - UW Sea Grant
  - US EPA BEACH Act
  - UW-Oshkosh

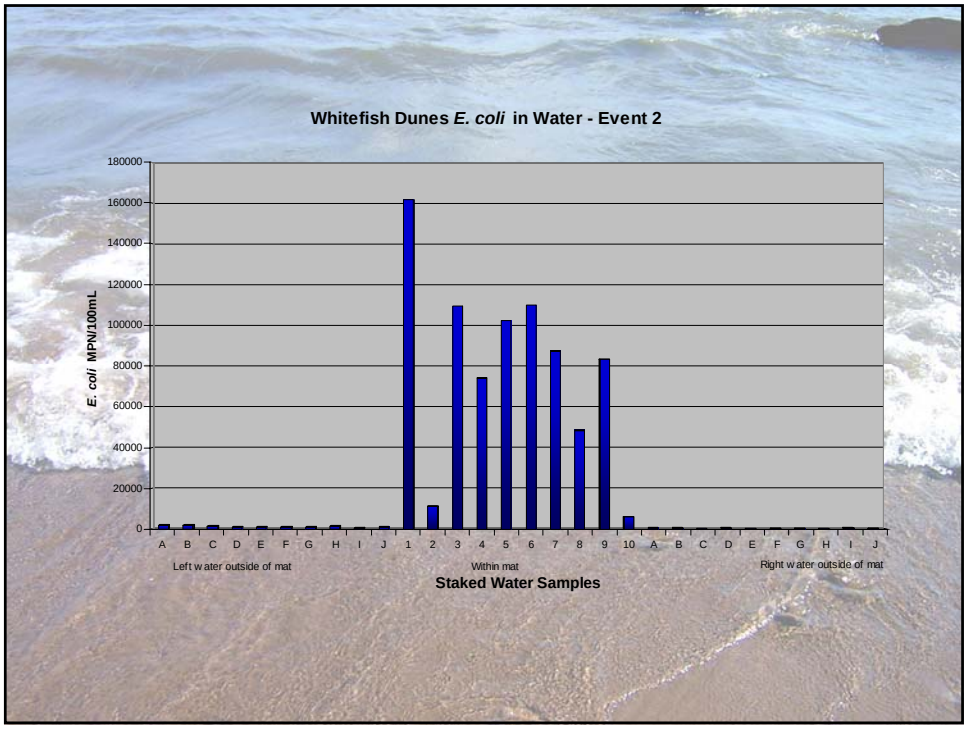
WISCONSIN COASTAL MANAGEMENT PROGRAM

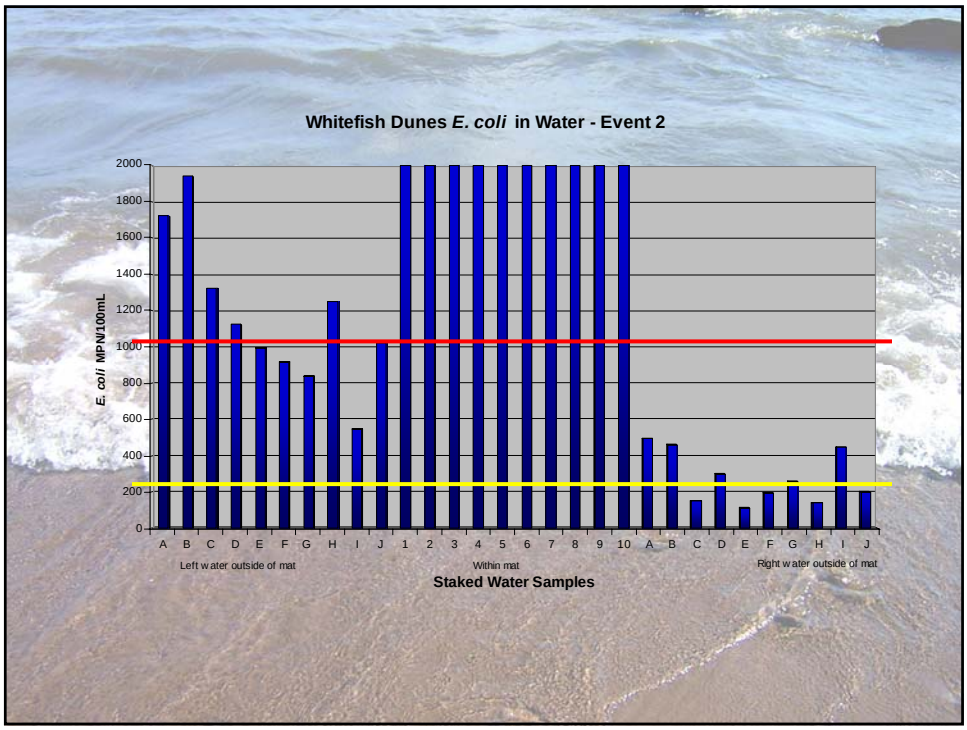
Sea Grant University of Wisconsin

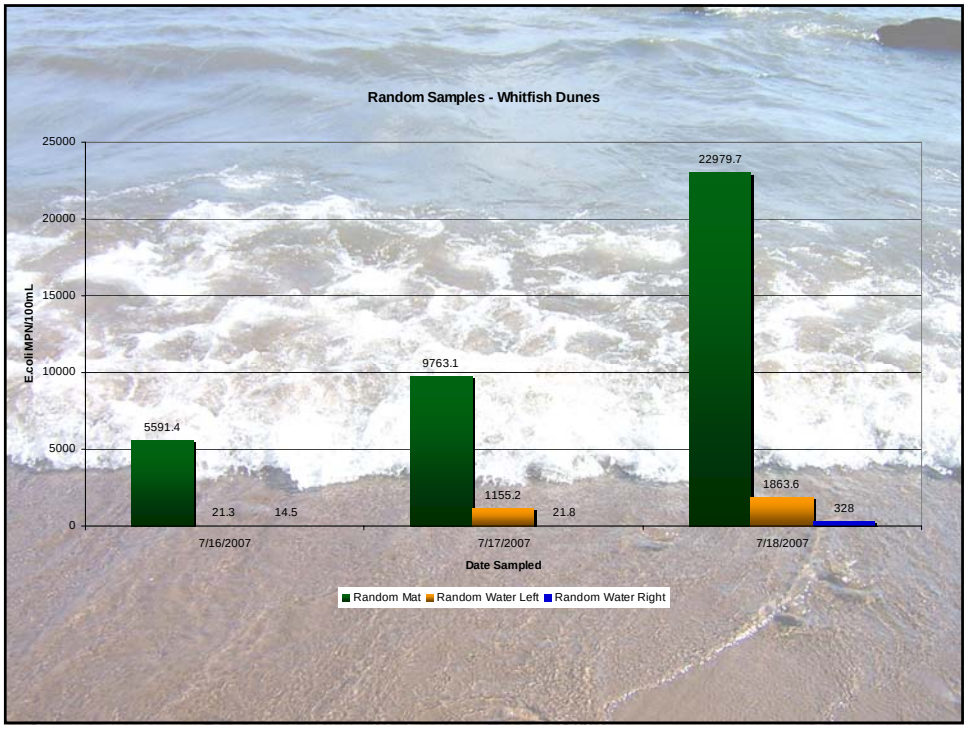
Thank you!

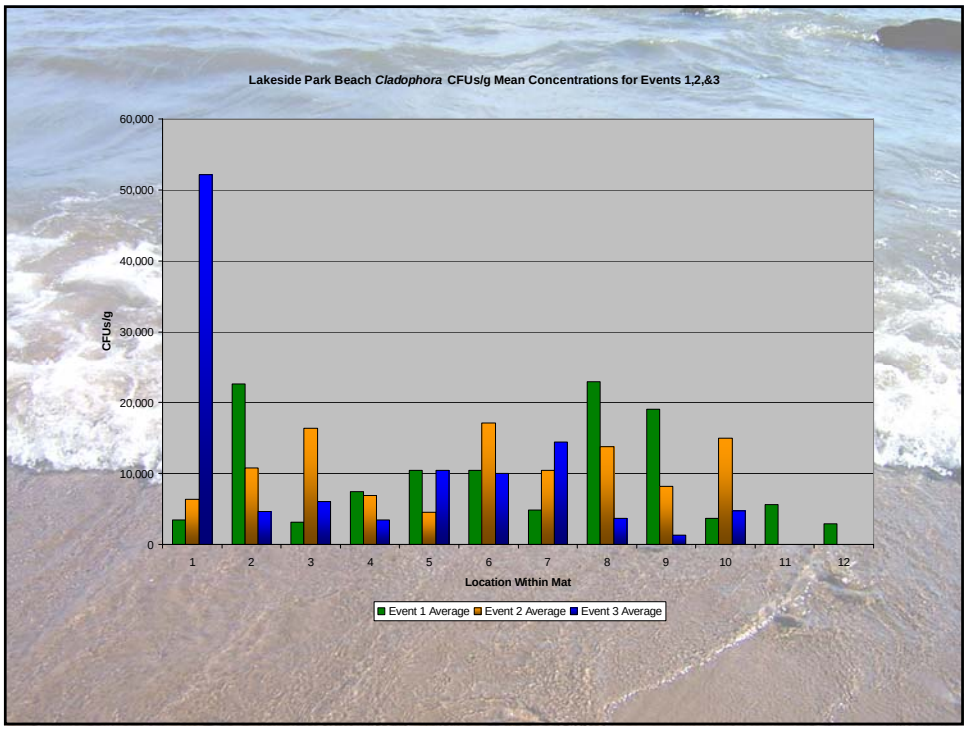

= Water Sample
= Water & *Cladophora* Sample
Lake
LW
CM
RW
A B C D E F G H I J
1 2 3 4 5 6 7 8 9 10
A B C D E F G H I J
Top View
Beach

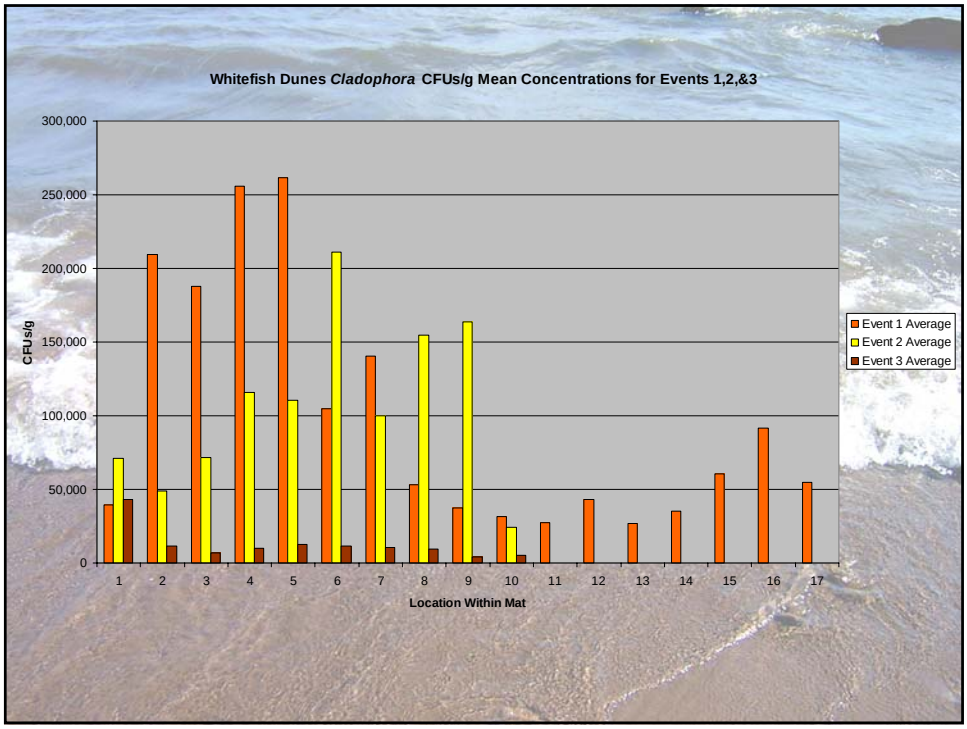

Outer Island
Stockton Island
Madeline Island
Washington Island
Chambers Island
Wisconsin
Harrington Beach
Madison
Milwaukee
© 2007 Europa Technologies
Image © 2007 TerraMetrics


Whitefish Dunes E. coli in Water - Event 2
E. coli MPN/100mL
180000
160000
140000
120000
100000
80000
60000
40000
20000
0
A B C D E F G H I J 1 2 3 4 5 6 7 8 9 10 A B C D E F G H I J
Left water outside of mat
Within mat
Right water outside of mat
Staked Water Samples


Whitefish Dunes E. coli in Water - Event 2
E. coli MPN/100mL
2000
1800
1600
1400
1200
1000
800
600
400
200
0
A B C D E F G H I J 1 2 3 4 5 6 7 8 9 10 A B C D E F G H I J
Left water outside of mat
Within mat
Right water outside of mat
Staked Water Samples


Random Samples - Whitfish Dunes
25000
20000
15000
10000
5000
0
E.coli MPN/100mL
5591.4
21.3
14.5
9763.1
1155.2
21.8
22979.7
1863.6
328
7/16/2007
7/17/2007
7/18/2007
Date Sampled
Random Mat
Random Water Left
Random Water Right


Lakeside Park Beach Cladophora CFUs/g Mean Concentrations for Events 1,2,&3
60,000
50,000
40,000
30,000
20,000
10,000
0
CFUs/g
1
2
3
4
5
6
7
8
9
10
11
12
Location Within Mat
Event 1 Average
Event 2 Average
Event 3 Average



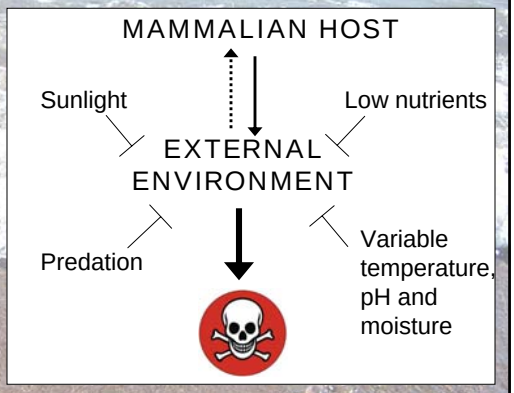
Factors Affecting Growth

- Phosphorous is an essential nutrient
  - Fertilizers, manure, & urban storm water
- Zebra mussels
- Light
- Temperature

Harris, 2005.

E.coli Outside the Host — Secondary Habitat

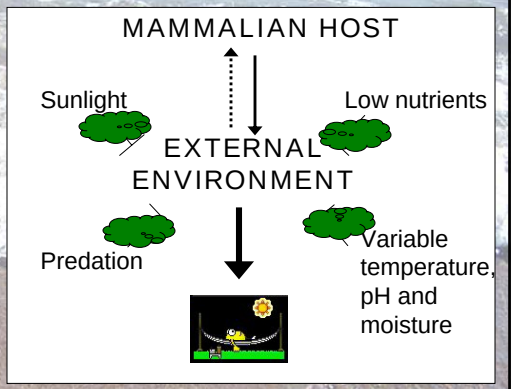
- A battle for survival
- Low rate of survival
- Low probability of colonizing a new host



Adapted from Winfield and Groisman. 2003.

Potential *Cladophora* impact on *E.coli* Survival

- Block UV light
- Increased nutrients
- Moisture retention
- More stable microbial community
- Favors growth and survival of microbes!

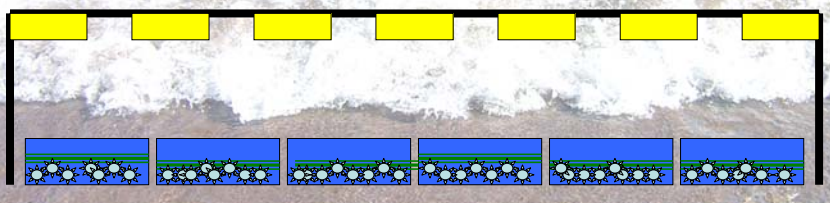


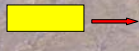
Adapted from Winfield and Groisman. 2003.

Lab Microcosm Study

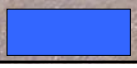
Not easy to assess pathogen survival in environment.

- Used lake water & *Cladophora* from local lake
- Looks at specific influence on *E. coli* and pathogens in a controlled setting.
  - Microbe + Water
  - Microbe + Water and *Cladophora*
- Measured:
  - microbes free in water
  - Microbes attached to *Cladophora*

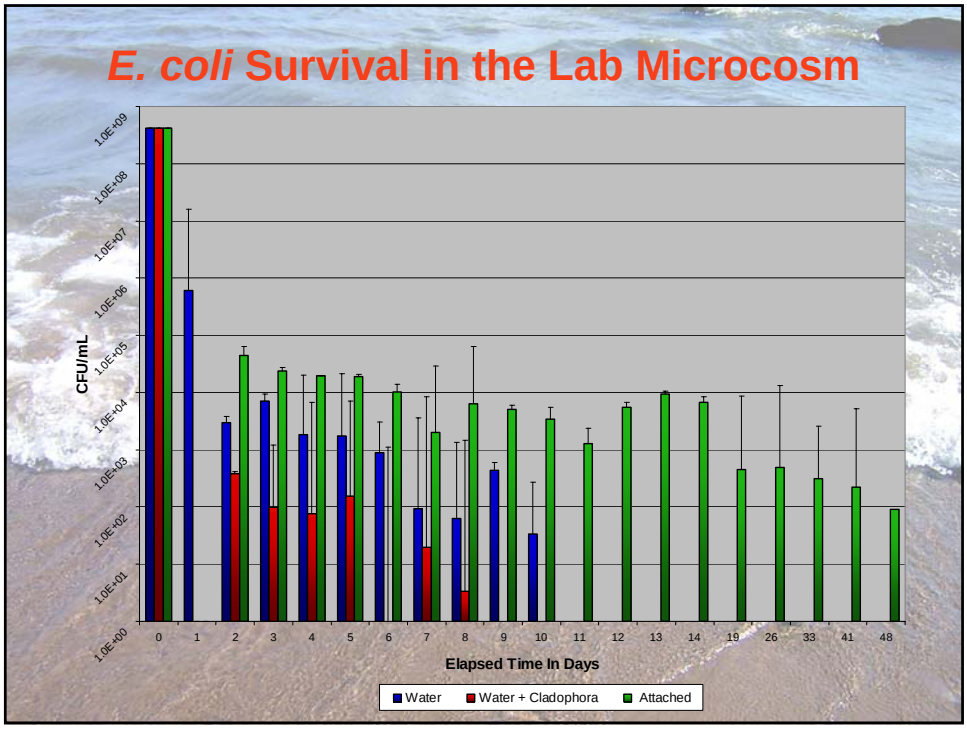


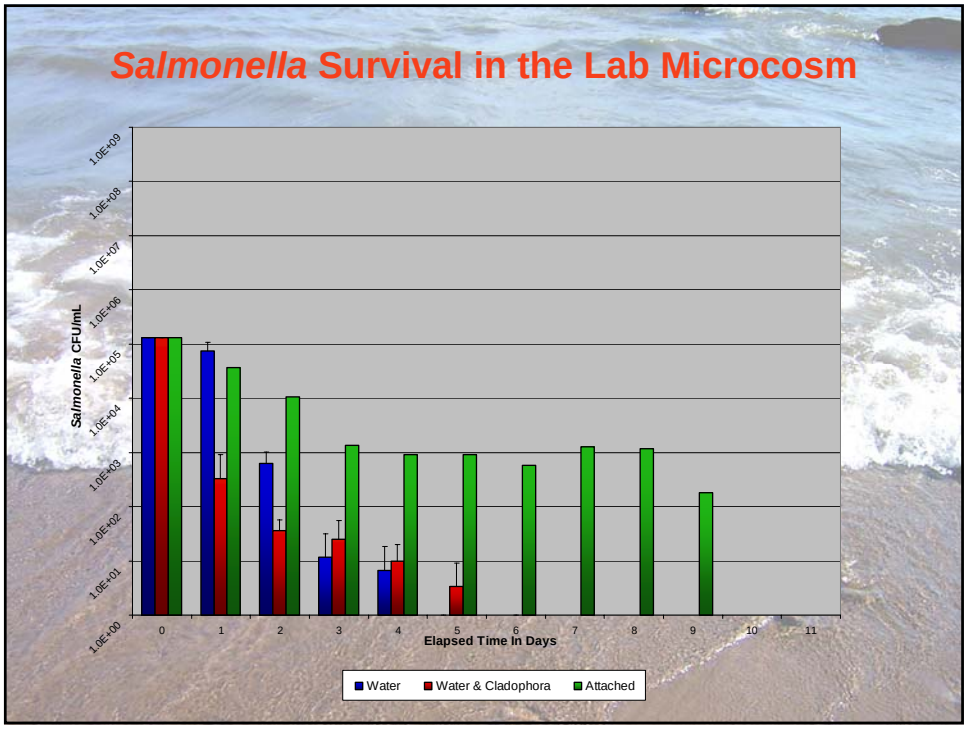


Grow Light (24" 20W)



Open Container w/H_2O and/or *Cladophora*


E. coli Survival in the Lab Microcosm
CFU/mL
1.0E+09
1.0E+08
1.0E+07
1.0E+06
1.0E+05
1.0E+04
1.0E+03
1.0E+02
1.0E+01
1.0E+00
0 1 2 3 4 5 6 7 8 9 10 11 12 13 14 19 26 33 41 48
Elapsed Time In Days
Water
Water + Cladophora
Attached


Salmonella Survival in the Lab Microcosm
Salmonella CFU/mL
1.0E+09
1.0E+08
1.0E+07
1.0E+06
1.0E+05
1.0E+04
1.0E+03
1.0E+02
1.0E+01
1.0E+00
0 1 2 3 4 5 6 7 8 9 10 11
Elapsed Time In Days
Water
Water & Cladophora
Attached

Time Required for 50% Decline of Microbes in Lab Microcosms

Microoganisms	Sample	Trial #	Hours to 50% Decline
E.coli	Water Only	1	82.6
		2	77.8
	Water + Cladophora	1	37.7
		2	57.8
	Attached	1	92.6
		2	90.5
Salmonella	Water Only	1	72.2
		2	*
	Water + Cladophora	1	57.8
		2	65.0
	Attached	1	*
		2	80.9
Shigella	Water Only	1	40.1
		2	40.3
	Water + Cladophora	1	37.7
		2	47.5
	Attached	1	42.5
		2	54.2

*Not enough data to conduct calculation.

DNR Observations vs Beach Monitoring

- Measure *Cladophora* using a WI DNR scale and data obtained from routine beach monitoring in Door County, WI
- Preliminary 'look' at what the observable *Cladophora* meant, if anything, to the data being obtained as part of a comprehensive monitoring program.
- Regression analysis of observational data versus *E. coli* data.

Picture from WI DNR 2004.

Cladophora Distribution Study

Ranked the presence of *Cladophora* on the beach on a scale of:

- None (0)
- Low (1)
- Moderate (2)
- High (3)

Slide adopted from WI DNR 2004.

Scale

Low *Cladophora* minimally present in patches on the beach.

Moderate *Cladophora* noticeably present on the beach and a nuisance.

High *Cladophora* covers beach in wind rows.

Relationship Between Observation and Beach Monitoring

Beach	F value	P value
Anclam	0.156	0.695
Bailey's Harbor	0.681	0.413
Egg Harbor	6.621	0.013
Ellison	0.114	0.737
Ephraim	1.572	0.215
Europe 1	0.002	0.969
Europe 2	0.002	0.969
Europe 3	0.491	0.487
Fish Creek	0.071	0.791
Gislason	0.397	0.541
Haines	1.367	0.247
Jackson Harbor Ridges	0.016	0.900
Lakeside	0.022	0.884
Murphy	2.192	0.144
Newport	0.036	0.850
Nicolet	3.281	0.076
Otumba	0.002	0.962
Percy Johnson	3.041	0.109
Portage Park	0.156	0.696
Rock Island	0.234	0.637
Sand Dune	0.042	0.840
Sandy Bay	1.382	0.252
Schoolhouse	0.009	0.926
Sister Bay	1.855	0.179
Sturgeon Bay Rec. Canal	0.037	0.848
Sunset	0.016	0.901
Whitefish Bay	1.869	0.177
Whitefish Dunes	0.319	0.575

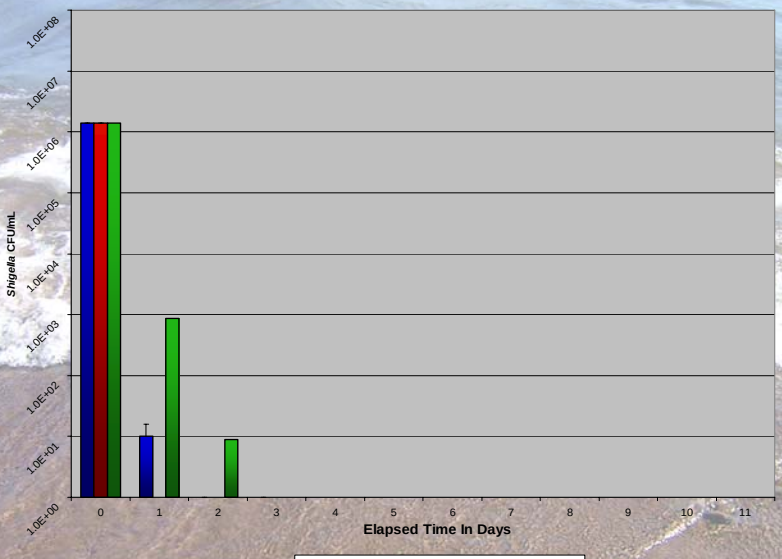
2005

2006

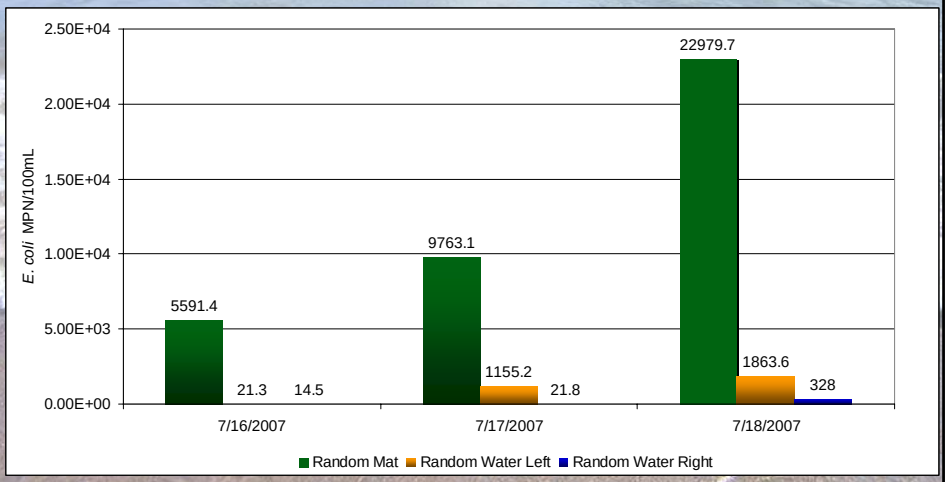
Beach	F value	P value
Anclam	1.047	0.315
Bailey's Harbor	0.173	0.679
Egg Harbor	0.766	0.385
Ellison	0.117	0.734
Ephraim	0.691	0.410
Europe 1	0.033	0.858
Europe 2	0.92	0.346
Europe 3	0.246	0.624
Fish Creek	0.464	0.499
Gislason	0.390	0.544
Haines	1.311	0.273
Jackson Harbor Ridges	4.957	0.043
Lakeside	0.678	0.417
Murphy	2.121	0.151
Newport	0.609	0.438
Nicolet	6.486	0.014
Otumba	1.257	0.267
Percy Johnson	0.384	0.545
Portage Park	0.611	0.441
Rock Island	0.092	0.767
Sand Dune*	n/a	n/a
Sandy Bay	4.659	0.043
Sturgeon Bay Rec. Canal	3.387	0.079
Sunset	0.112	0.740
Whitefish Bay	0.073	0.791
Whitefish Dunes	0.413	0.523

**Not possible to analyze due to lack of data.*

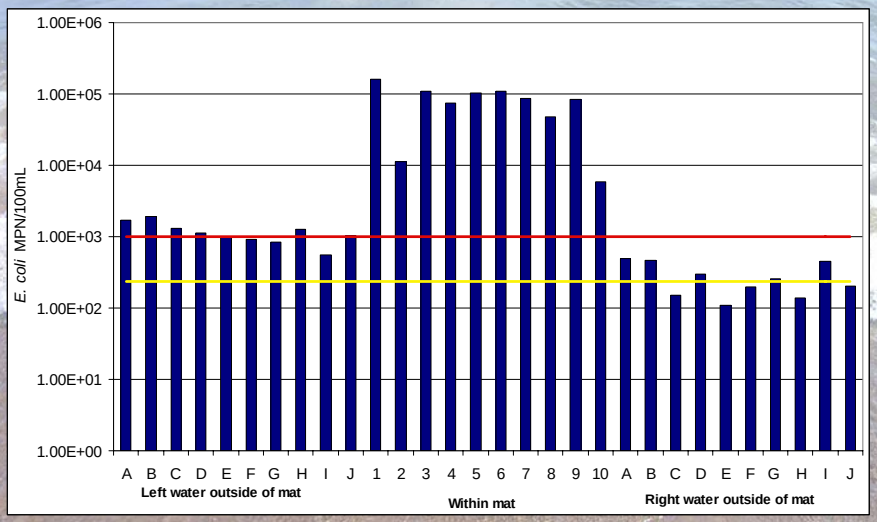
Shigella Survival in the Lab Microcosm



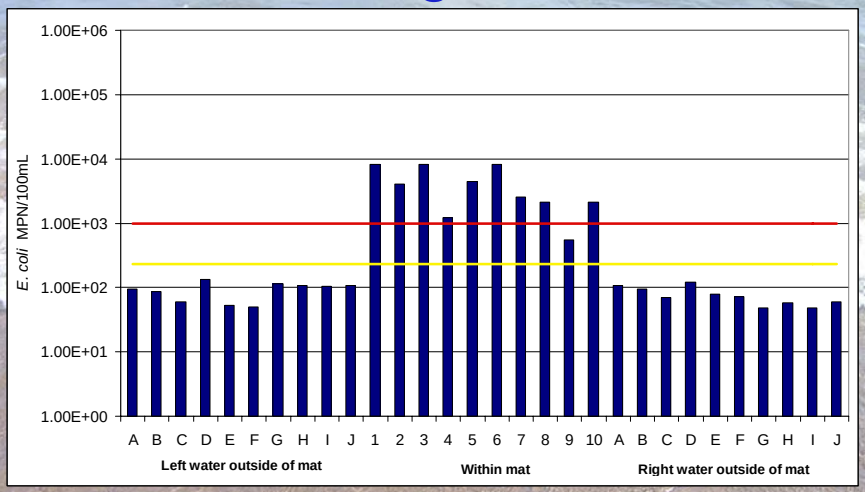
Whitefish Dunes-Event 1



Whitefish Dunes – Event 2

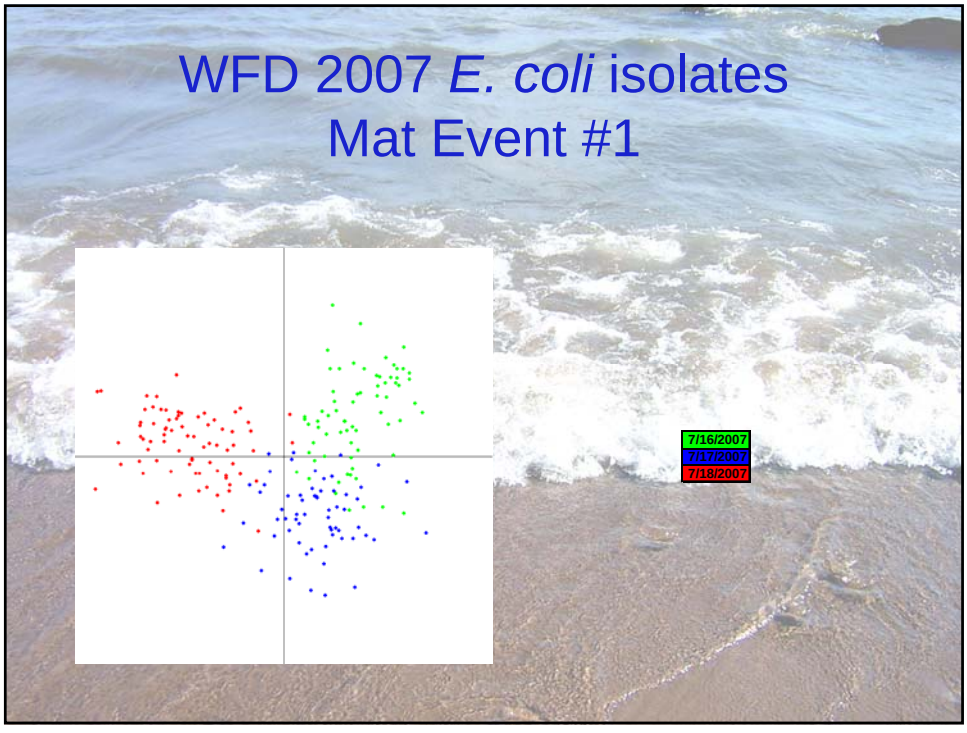


Lakeside Park Beach – Event 3




Uses for stranded
Cladophora?
What can be done with it, once it strands on my beach?
-Composting
-Use as pelleted heating product
-Ethanol production
-Methane production
-Biofuel
-And the list goes on…..


What we know now…
Presence of contaminating heavy metals or organics in *Cladophora*= very low
-BTU potential= similar to White Pine
-Composting project in progress in Ephraim
-Use in methane digestor in progress in Manitowoc County


WFD 2007 *E. coli* isolates
Mat Event #1
7/16/2007
7/17/2007
7/18/2007