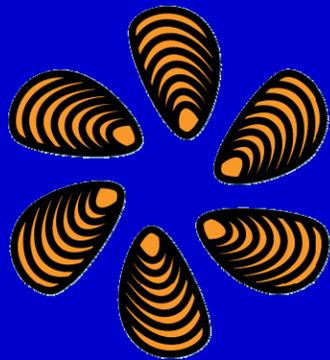


Network approaches to studying aquatic invasive species in Canada

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University of Windsor



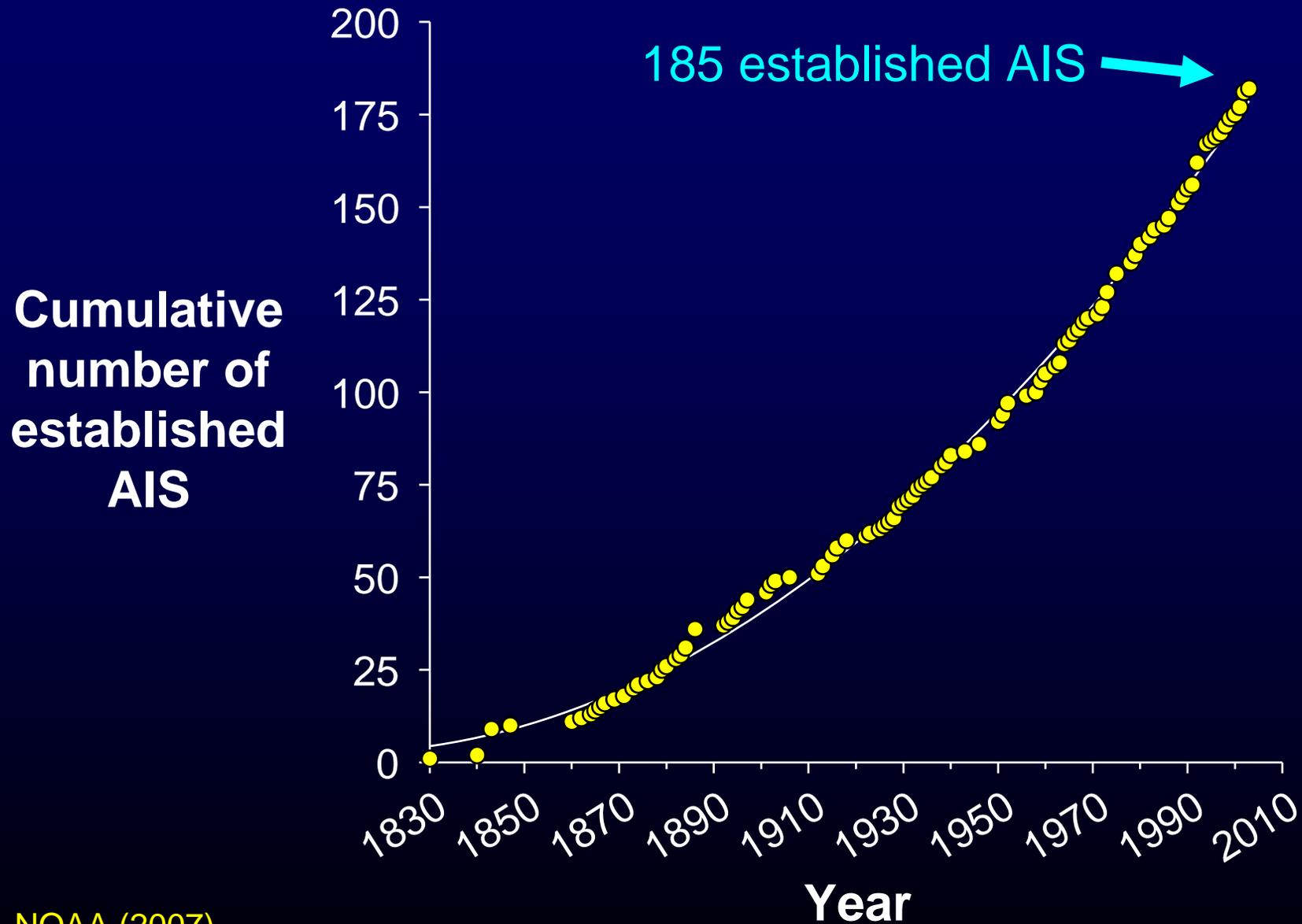
CAISN

CANADIAN
AQUATIC
INVASIVE
SPECIES
NETWORK

What is an Aquatic Invasive Species (AIS) and why should we care?

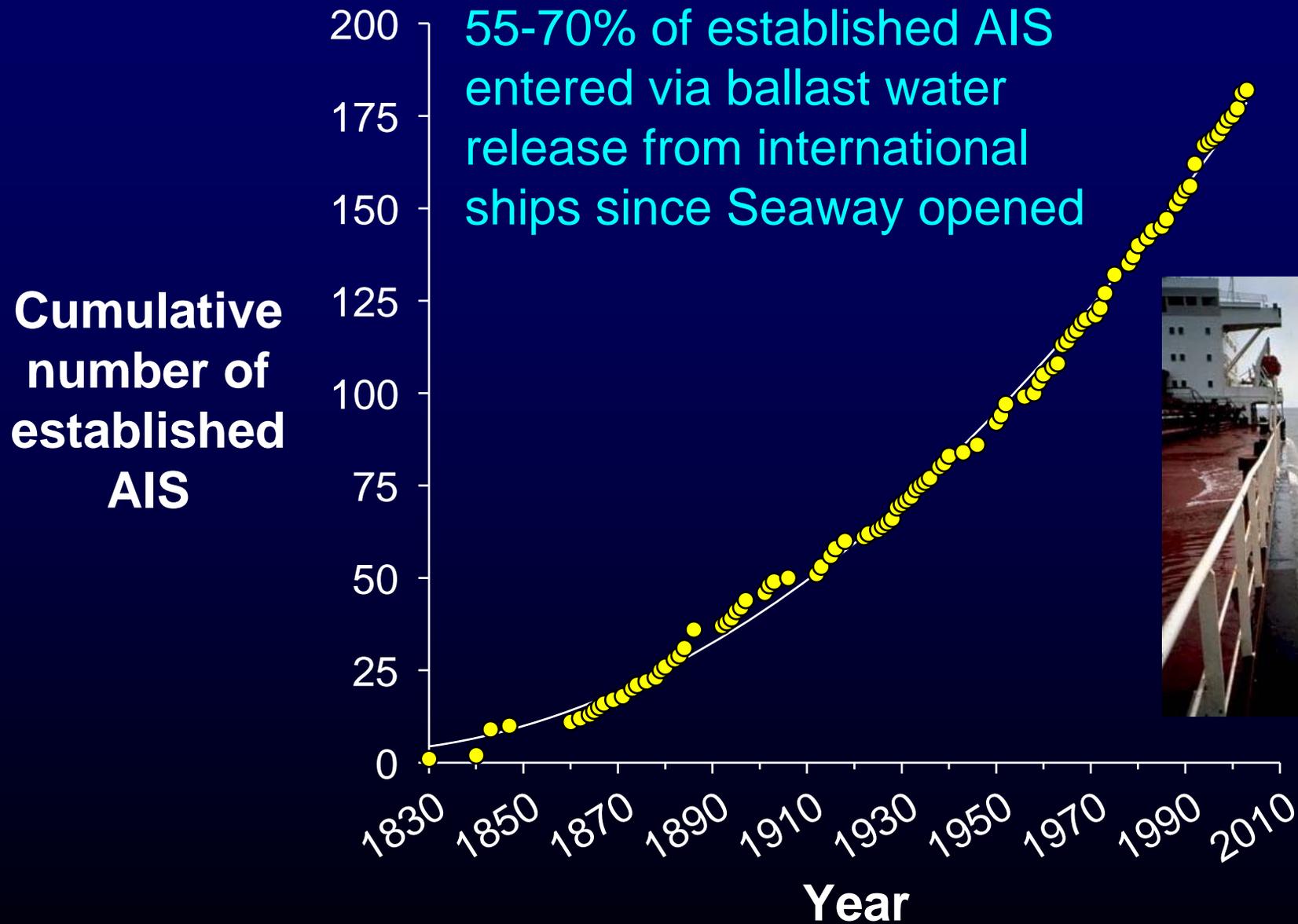
- Species introduced to regions outside of their historic native range;
- Spread at 10,000 times the natural dispersal rate;
- Cause up to \$132 billion in USA and \$34 billion in Canada per year in damage and control costs;
- 2nd leading cause of biodiversity loss globally;
- Biggest problem today in the Great Lakes and a major problem in marine coastal ecosystems;
- Auditor General (~General Accounting Office in USA) has addressed the issue 3 times since 2003.

Invasive Species in the Great Lakes



NOAA (2007)

Invasive Species in the Great Lakes



U.S. National Research Council (2008)

Marine Ecosystems are under siege by AIS



vase and clubbed tunicates



violet & golden star tunicate

threaten aquaculture industry in PEI, which provides 5% of jobs

CAISN Research Priorities and Areas

1) 3 approved theme areas:

- i) Vectors & Pathways
- ii) Factors Affecting Establishment Success
- iii) Risk Assessment
- iv) Trophic Disruption

2) 3 regions (nodes) studied:

- i) west coast Canada
- ii) Atlantic Canada
- iii) Great Lakes - St. Lawrence River, inland lakes

- Network operated from 2006-2011



CAISN

Participants:

- 25 faculty in 8 provinces (later expanded to 34 faculty with new projects)
 - 18 universities
 - 5 federal labs

Funding: NSERC (\$4.7 million)

- Fisheries and Oceans (\$1 million)
- Transport Canada (\$0.5 million)
- Ontario Ministry of Environment (\$360K)
- Ontario Ministry of Natural Resources (\$200K)
- University of Windsor (\$448K)

Partners:

- Aquaculture Industry (PEI)
- Shipping Industry (Shipping Federation of Canada, ports of Halifax, Vancouver)
- Outreach and education: Ontario Federation of Anglers and Hunters and its sister organizations

CAISN Projects

Theme	Project	Region
Vectors and Pathways	I-I <i>Ballast water discharge database</i>	WC, EC
	I-II <i>Plankton survivorship analyses</i>	WC, EC
	I-III <i>Propagule pressure in relation to shipping mode & route</i>	WC, GL, EC
	I-IV <i>Baseline coastal port surveys</i>	WC, EC
	I-V <i>500-lake survey</i>	GL
	I-VI <i>Dispersal modes of Bythotrephes</i>	GL
	I-VII <i>Vectors of invasive fishes</i>	GL
Factors Affecting Establishment Success	II-I <i>Analysis of source and destination port similarity</i>	EC, GL
	II-II <i>Effect of human disturbances on establishment success</i>	WC, EC
	II-III <i>Physiological tolerance of Bythotrephes</i>	GL
	II-IV <i>"Invasional meltdown" field studies and computer simulation</i>	GL, EC
Risk Assessment Modeling	III-I <i>Modeling risk of international ships</i>	WC, GL, EC
	III-II <i>Modeling of invasive fish vectors</i>	GL
	III-IV <i>HACCP model of tunicate AIS</i>	EC, WC
	III-V <i>Modeling establishment success of AIS in inland lakes</i>	GL

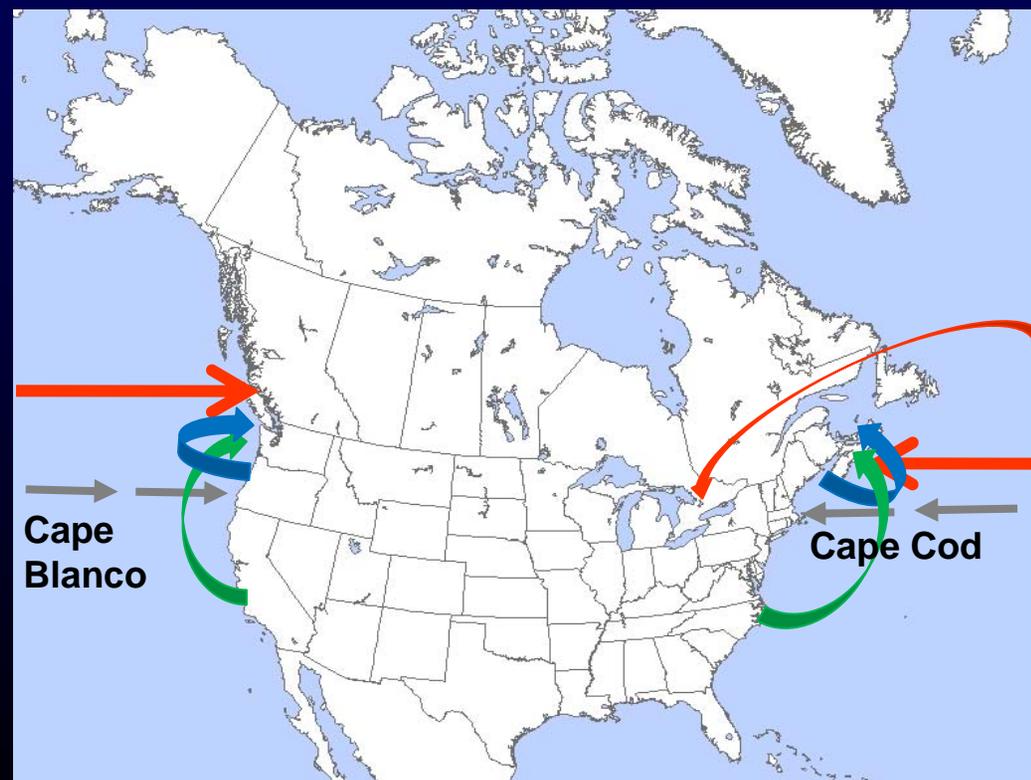
•Additional 5 projects funded via RFP process in 2007, 2008

Examples of Key Findings

1) Ballast water studies

3 different types of ships (20 each, per coast)

- Transoceanic ships with ballast exchange (red)
- Coastal ship (green) with exchange
- Coastal ship (blue) no exchange



- Vancouver gets the most ship traffic, followed by Halifax, and then Great Lakes ports
 - hull fouling risk -> Vancouver?
 - ballast water risk -> Halifax?

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Invertebrates

- Halifax has the highest diversity and abundance of living invertebrates of all ships examined
 - mid-ocean exchange did not reduce abundances relative to coastal vessels
 - All invertebrate eggs in ballast sediment are nonviable in transoceanic ships arriving to Vancouver but not elsewhere

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Dinoflagellates

- Dinoflagellates were present in almost all ships
 - Potentially toxic species found in 94% and 63% for the east and west coasts, respectively
 - more species were found in east coast transoceanic and west coast coastal non-exchanged vessels

Diatoms

- coastal unexchanged vessels had highest densities of diatoms
- 70% of tanks had at least one member of *Pseudo-nitzschia*, including many potentially toxigenic species

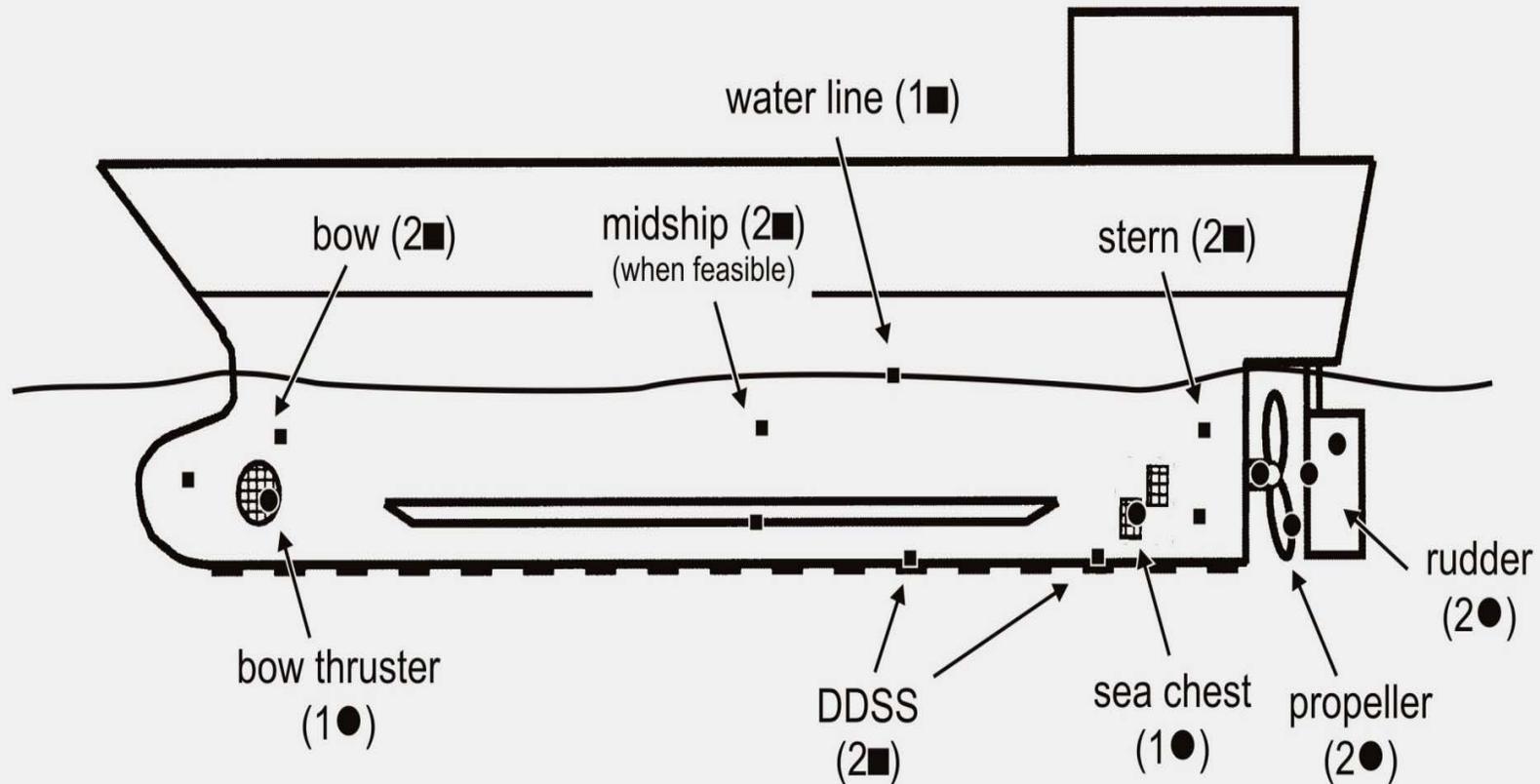
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Bacteria

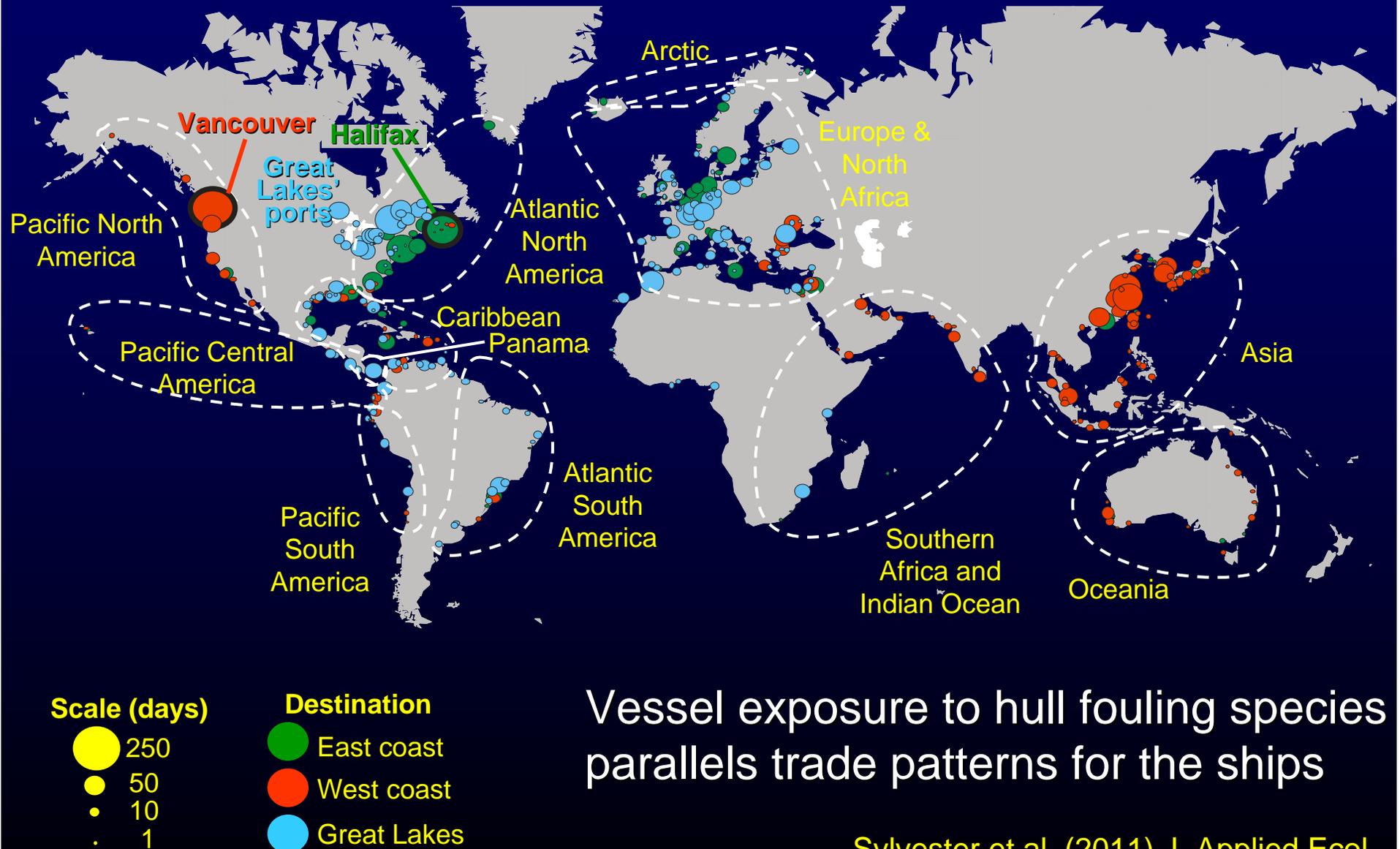
- On average, bacterial abundance in receiving port water was 3-4x higher than that in ballast water.
- Ballast water not exchanged at sea had higher bacterial abundance than exchanged ballast water
- Bacterial communities did not differ among ballast waters types
- *Vibrio* spp. was detected in 15% of ballast water samples, and comprised 5 to 20% of total bacterial abundance.

2) Hull Fouling



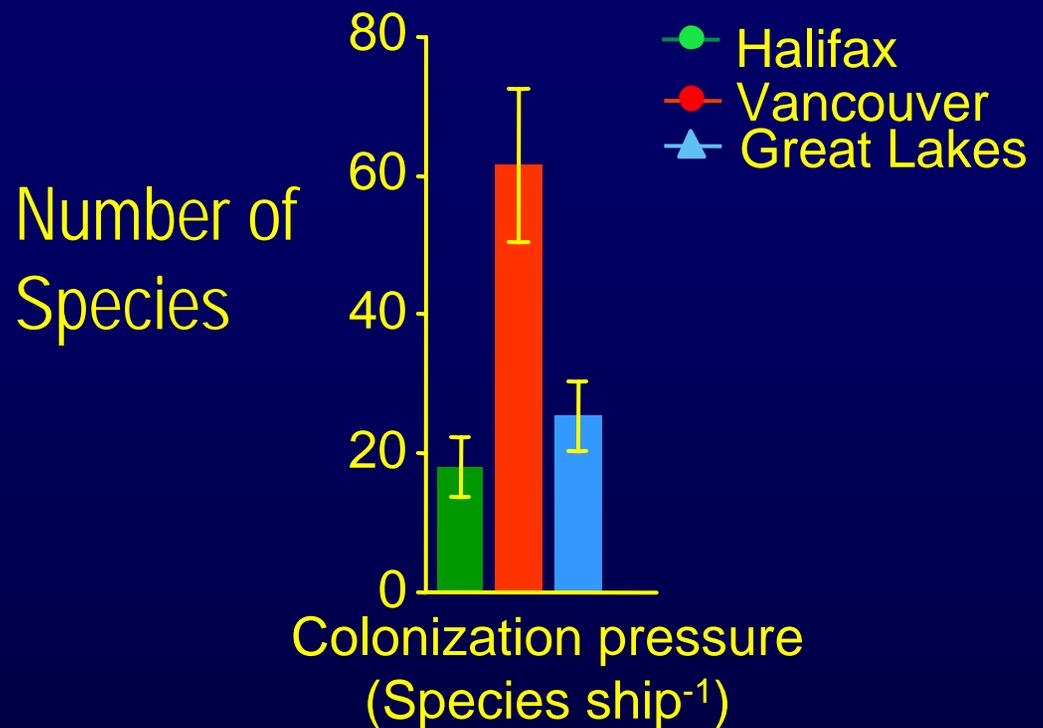
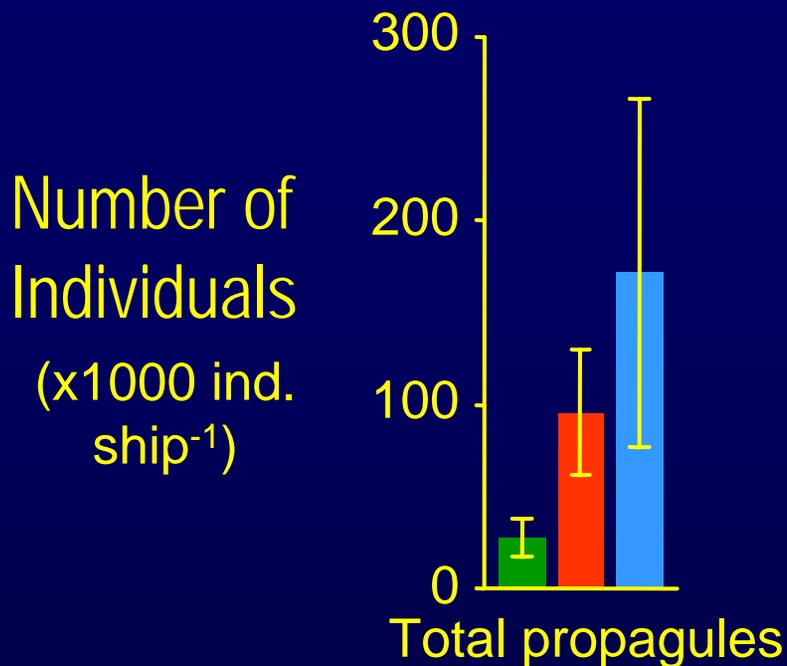
Ship sampling protocol based on previous experiences by workers in USA, New Zealand and Australia

Days Spent in Port in Different Regions Prior to Arrival in Canada (60 ships)



Vessel exposure to hull fouling species parallels trade patterns for the ships

Sylvester et al. (2011) J. Applied Ecol.



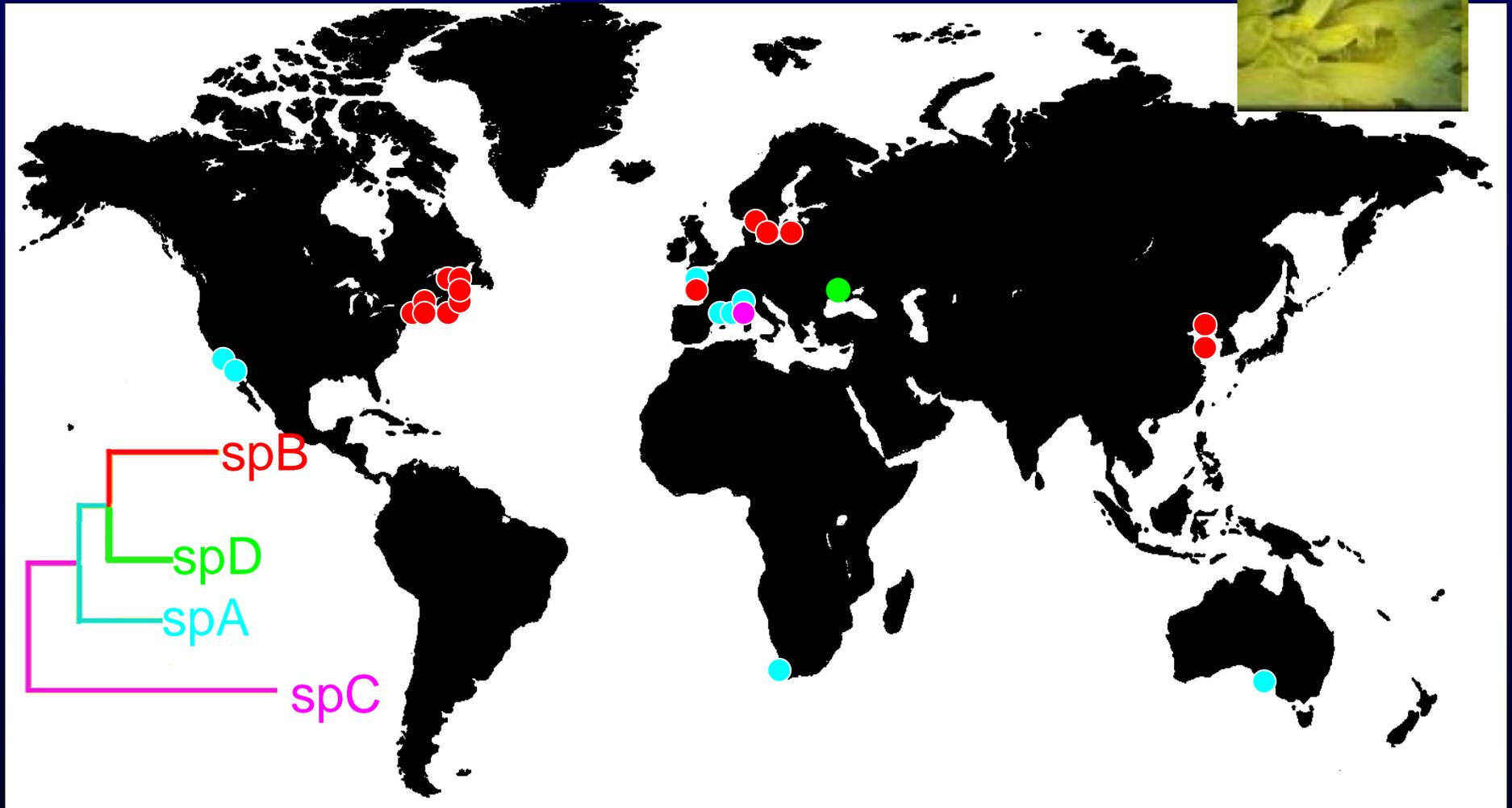
- Propagule and colonization pressure higher in Vancouver than Halifax
- Great Lakes appears at moderate risk but only 1 new species found there that could possibly survive, while 10 and 34 such species were found in Halifax and Vancouver, respectively
- Species on ships are almost completely different than those present in Halifax and Vancouver

3) Genetics of Invasive Tunicates



AVC - UPEI

i) *Ciona intestinalis* species

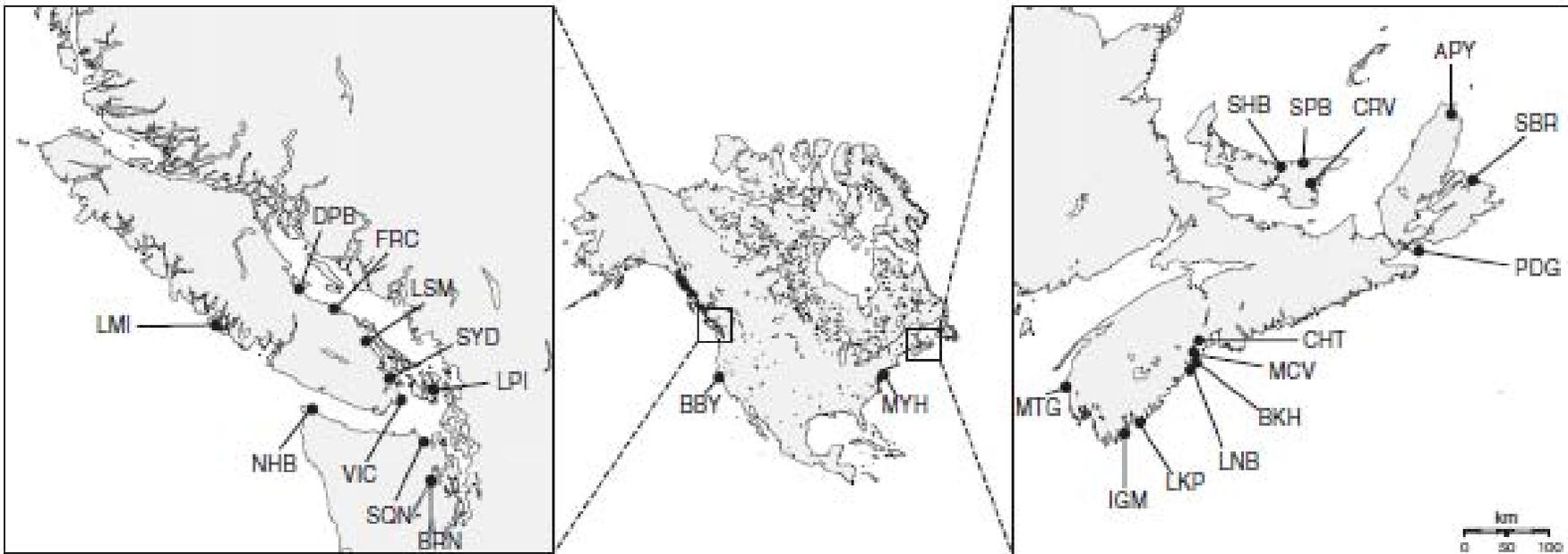


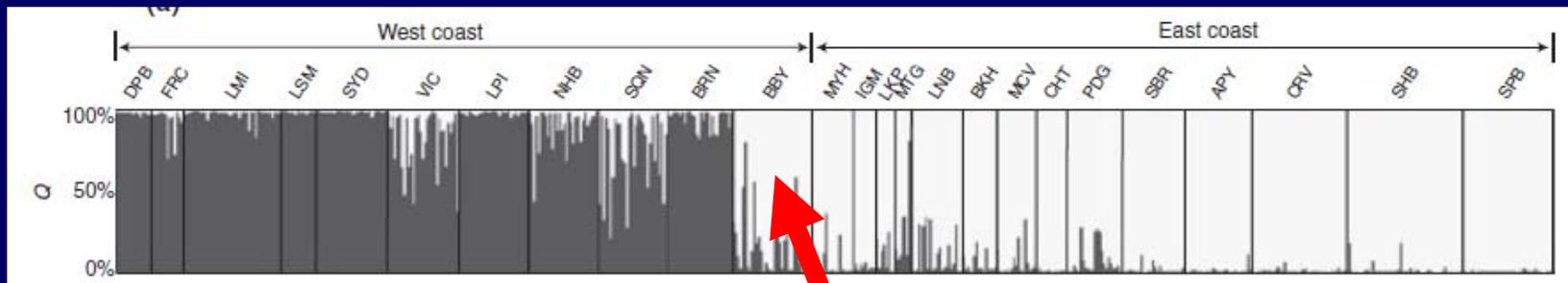
- 4 species detected in complex, two are invasive, only one is present on east coast
- high population connectivity in species A is likely a consequence of human and natural dispersal

ii) *Botrylloides violaceus*



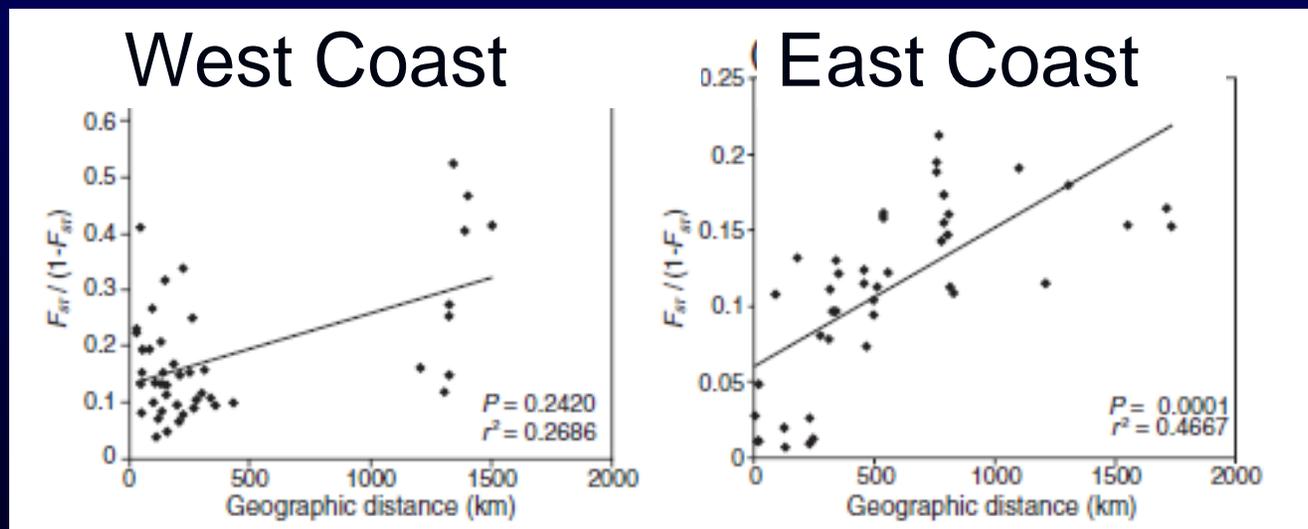
- Surveyed 25 populations
- Used mitochondrial and microsatellite markers





genetic assignment test

Bodega Bay population clusters with east coast stocks and likely was the source



Isolation by distance only on East coast

- West coast sites invaded via long-distance dispersal, east coast seeded from west
- East coast spread may be facilitated by aquaculture or boating practices

Key products from CAISN

- Advice to government and industry on current risk associated with ballast water (generally, non-flushed coastal vessels riskiest)
- Advice to industry and government on hull fouling
- Advice to government on risks associated with aquarium and baitfish trades
- Advice to industry to prevent regional spread of tunicates
- comprehensive conceptual and empirical models on spread of spiny waterfleas (and other species) to inland lakes

CAISN Gaps:

1. Dearth of knowledge about detecting new AIS
2. No consideration of management of AIS once detected
3. Poor understanding of how stressors interact
4. No consideration of uncertainty in development or implementation of management plans
5. No involvement in the Arctic

NSERC CAISN II

- Scheduled activities from 2011-2016
- Active in all same regions as CAISN plus Arctic
- Will train a further:
 - 14 MSc, 20 PhD, 6 PDF, 61 BSc students
- Outputs will fulfill Auditor General's 2008 call to establish a surveillance program across the country and create reference databases against which to compare future changes

Geographic Distribution of PIs and Projects



Research Themes

1. Early detection and identification of AIS – 4 projects
2. Rapid response to new AIS – 2 projects
3. AIS as part of multiple stressors – 3 projects
4. Reducing uncertainty in management of AIS – 4 projects

Theme 1: Early detection and identification of AIS

Projects

1. Surveillance for AIS throughout Canada's coastal waters.
2. Early detection of AIS using state-of-the-art technologies.
3. Reconciling large-scale model predictions with small-scale observations of AIS distributions to inform early detection.
4. Optimal methods for early detection based on life history traits.

Theme 2: Rapid Response to Newly Detected AIS

Projects

1. Evaluating Canada's preparedness to deal with AIS
 - education, training, media
2. Rapid response and decision support models for AIS
 - Andrea' Locke's (Nick Mandrak) talk

Theme 3: Multiple Stressors including AIS

Projects

1. Synergistic impacts of AIS and environmental stressors on native biodiversity and structure in coastal marine ecosystems. Chris McKindsey's talk.
2. Environmental modulation of multiple stressors in riverine ecosystems.
3. Stressor-dependent effects of AIS on native communities.

Theme 4: Reducing Uncertainty in Managing AIS

Projects

1. Explaining variation in AIS impact using invasion history and organismal traits.
2. Reducing uncertainty for risk based management through assessments of phenotypic plasticity and genetic diversity in AIS.
3. Embracing uncertainty in the management of AIS.

Examples of Research Projects

1. Early detection and identification of AIS

- Optical methods – FlowCam + image library
- barcoding
- 454 pyrosequencing
- microarray

2. Reducing uncertainty in management of AIS

- can combining technologies reduce the risk of ballast water introductions to the Great Lakes?

1) CAISN's Molecular Detection Methods

Identification of unknown species in samples

- DNA barcoding
- Pyrosequencing

Screening for presence of target species

- PCR based on species-specific primers
- Quantitative PCR (qPCR)
- Microarray

Why molecular methods?

Morphology-based methods (classical taxonomy)

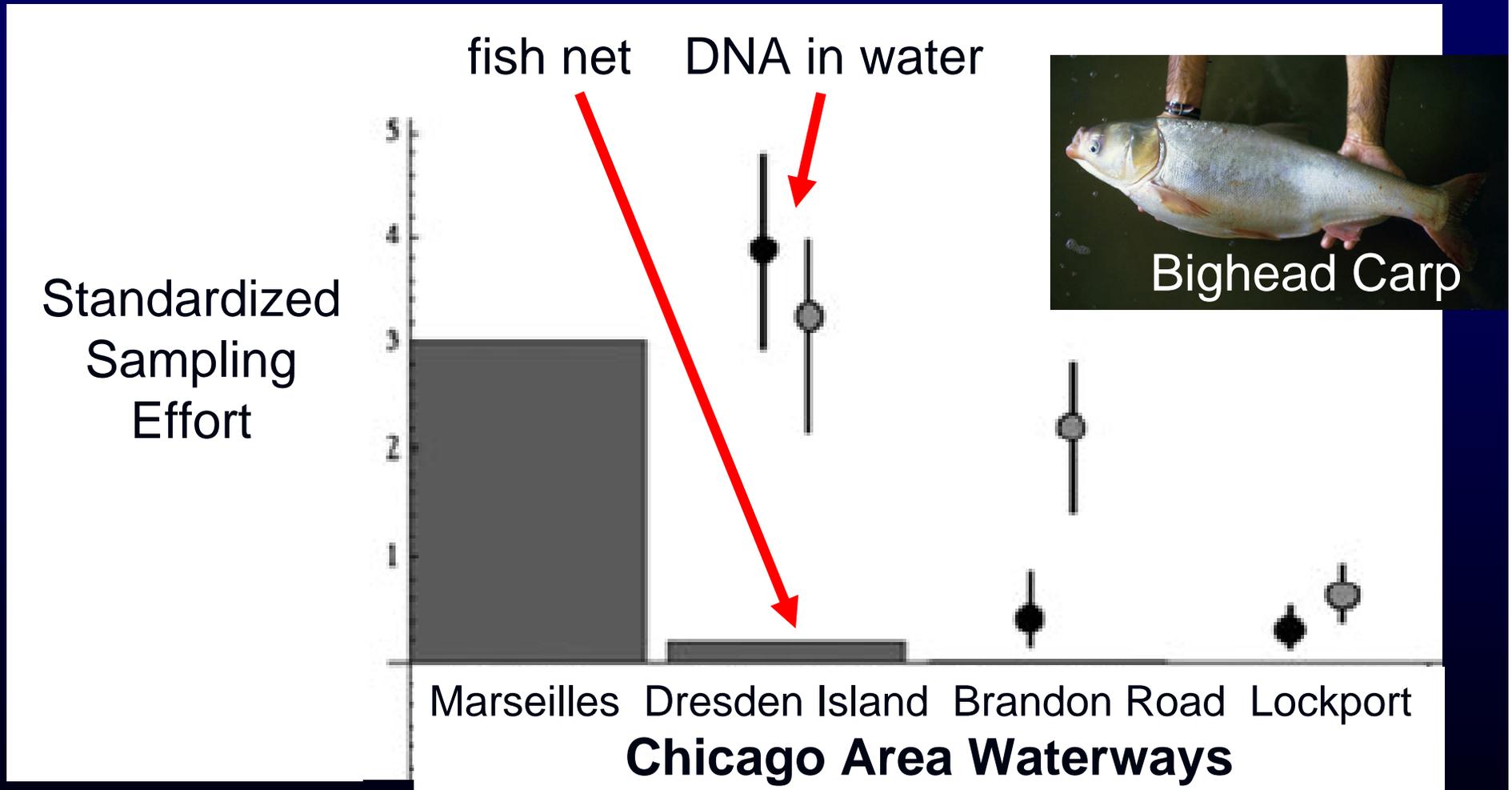
- Skills in taxonomy (less common every day)
- Unrealistic for some stages, e.g. eggs, larvae

Molecular methods

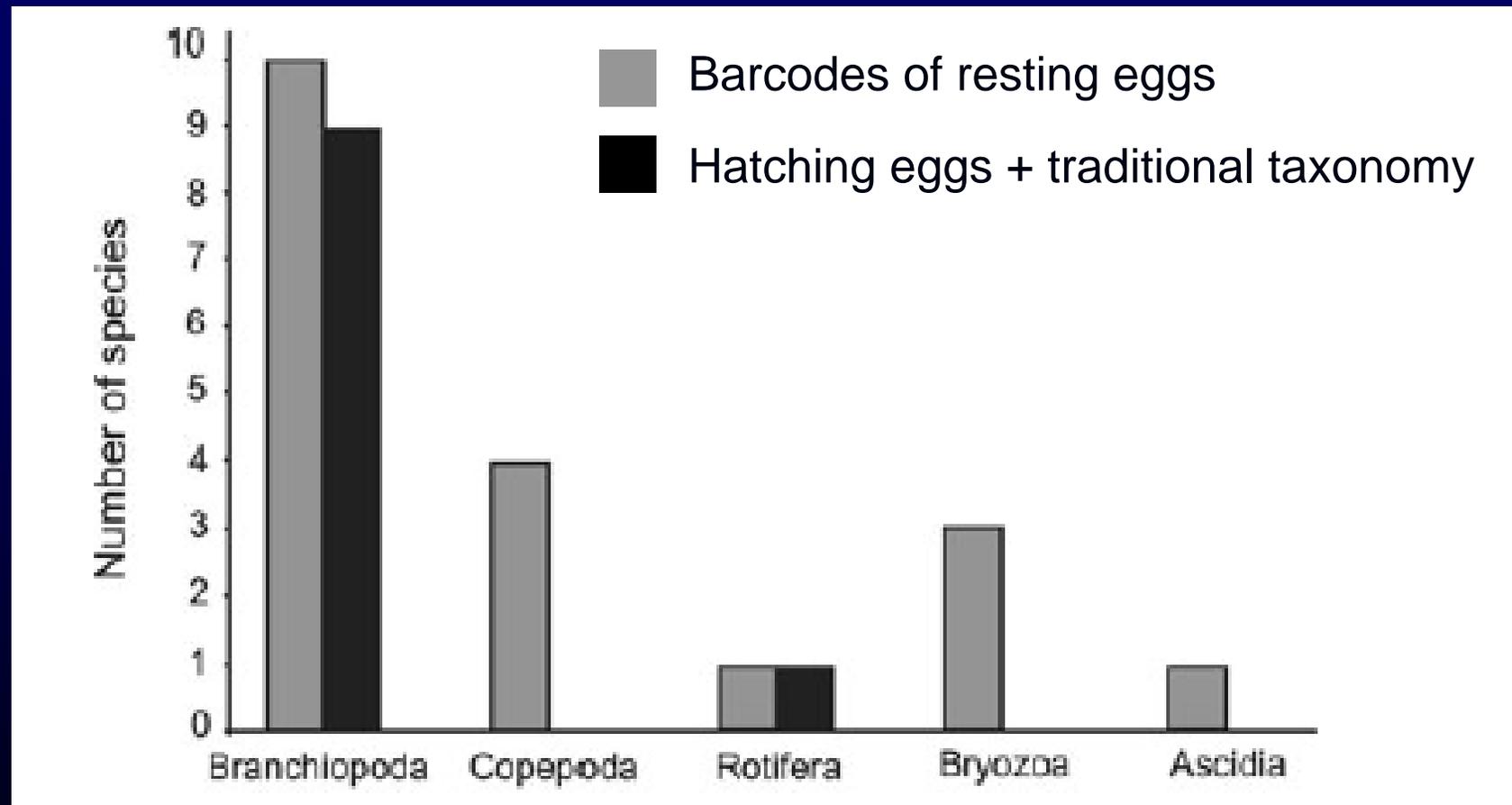
- High sensitivity, especially for low-density species
- High accuracy
- Powerful in species with limited taxonomic information
- Powerful in cryptic species

Why molecular detection?

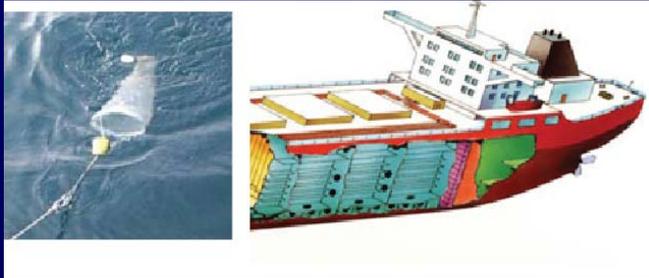
Detecting an invader when it is rare



Barcoding can resolve identity for some groups where hatching studies are a complete failure



Technique: DNA barcoding



↓ Sample sorting



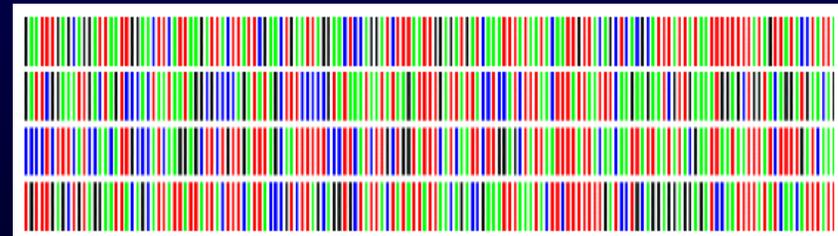
↓ DNA extraction



PCR & Sequencing



BOLDSYSTEMS



Technology: 454 Pyrosequencing



Sampling

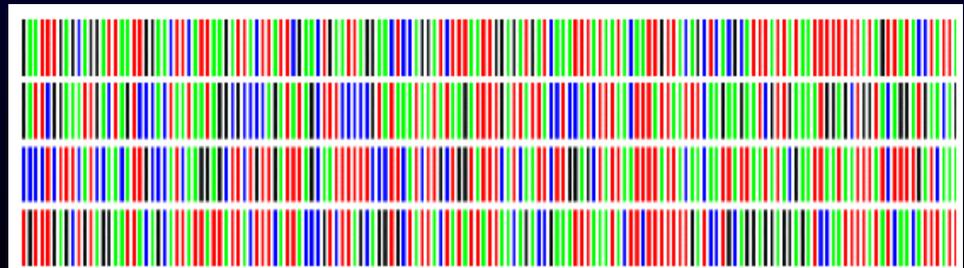
Targets:

- molluscs
- crustaceans
- ascidians

↓ Mass DNA extraction



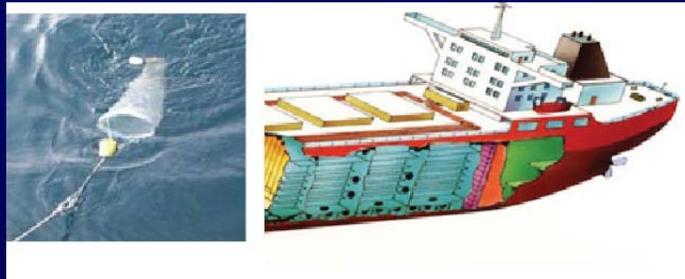
↓ Pyrosequencing



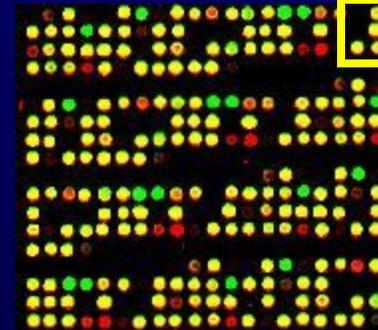
BOLDSYSTEMS



Identification of 40 target AIS Technology: Microarray



Sampling



Microarray read

DNA extraction, amplification



Hybridization



Key step

Microarray development

- Genechip will be offered globally for routine monitoring of these 40 AIS

40 targeted AIS for microarray

	Marine Species		Freshwater Species	
	Scientific name	Common name	Scientific name	Common name
1	<i>Carcinus maenas</i>	European green crab	<i>Dreissena polymorpha</i>	zebra mussel
2	<i>Eriocheir sinensis</i>	Chinese mitten crab	<i>D. rostriformis bugensis</i>	quagga mussel
3	<i>Styela clava</i>	clubbed tunicate	<i>Cercopagis pengoi</i>	fishhook waterflea
4	<i>Botrylloides violaceus</i>	violet tunicate	<i>Bythotrephes longimanus</i>	spiny waterflea
5	<i>Botryllus schlosseri</i>	golden star tunicate	<i>Chelicorophium curvispinum</i>	Black Sea amphipod
6	<i>Ciona intestinalis</i>	vase tunicate	<i>Dikerogammarus villosus</i>	killer shrimp
7	<i>Didemnum vexillum</i>	carpet sea squirt	<i>Cornigerius maeoticus</i>	Caspian cladoceran
8	<i>Diplosoma listerianum</i>	compound sea squirt	<i>Potamopyrgus antipodarum</i>	New Zealand mud snail
9	<i>Clavelina lepadiformis</i>	light-bulb sea squirt	<i>Orconectes rusticus</i>	rusty crayfish
10	<i>Littorina littorea</i>	common periwinkle	<i>Hemimysis anomala</i>	bloody red shrimp
11	<i>Mytilus galloprovincialis</i>	Mediterranean blue mussel	<i>Corbicula fluminea</i>	Asian clam
12	<i>Rapana venosa</i>	veined rapa whelk	<i>Channa argus</i>	northern snakehead
13	<i>Hemigrapsus sanguineus</i>	Asian shore crab	<i>Hypophthalmichthys molitrix</i>	silver carp
14	<i>Caprella mutica</i>	Japanese skeleton shrimp	<i>H. nobilis</i>	bighead carp
15	<i>Crepidula fornicata</i>	common Atlantic slipper snail	<i>Neogobius melanostomus</i>	round goby
AIS of global concern to be added to microarray chip				
1	<i>Mnemiopsis leidyi</i>	comb jelly	<i>Limnoperna fortunei</i>	golden mussel
2	<i>Perna perna</i>	brown mussel	<i>Evadne anonyx</i>	waterflea
3	<i>Xenostrobus securis</i>	black-pygmy mussel	<i>Daphnia lumholtzi</i>	waterflea
4	<i>Crassostrea gigas</i>	Pacific oyster	<i>Cornigerius maeoticus</i>	waterflea
5	<i>Mytilopsis sallei</i>	black-striped mussel	<i>Pacifastacus leniusculus</i>	signal crayfish

2) Combining technologies reduce the risk of ballast water introductions of AIS



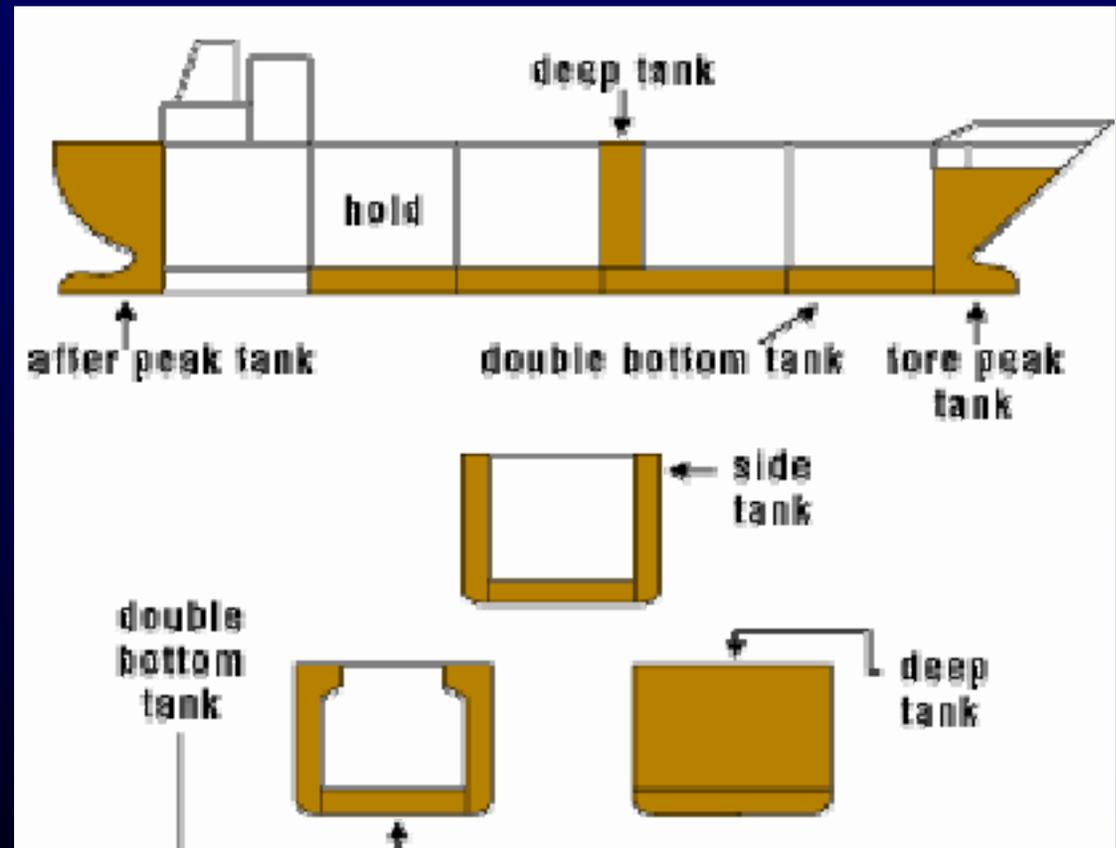
Massena, NY

- All ships entering Great Lakes must exchange ballast water on the open ocean, which has been proven effective
- A proposed global standard places limits on the density of live organisms in discharged ballast water
- In 2013, NY will implement a standard 100x more robust

IMO standard (if accepted): < 10 viable ind. m^{-3} $> 50\mu m$

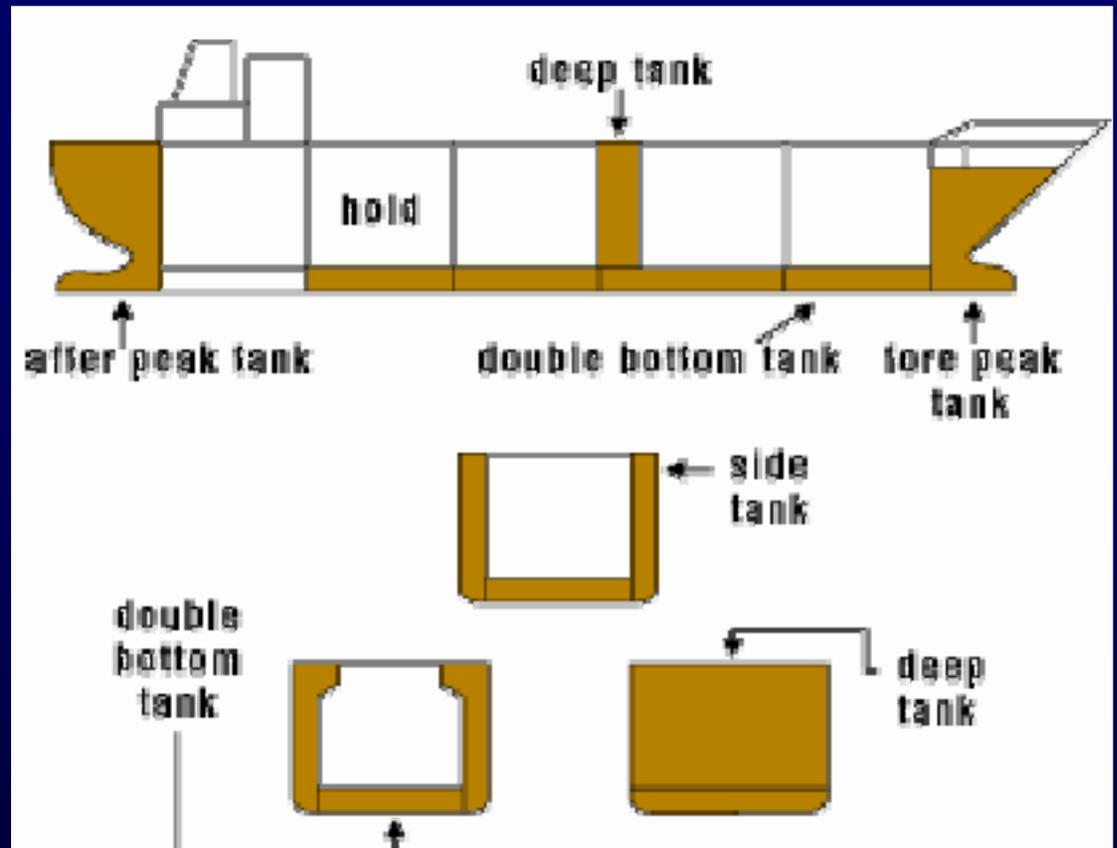
New York standard: < 0.10 viable ind. m^{-3}

CAISN will conduct on-board, trans-Atlantic experiments to achieve reduced densities of organisms using ballast water exchange in combination with chlorination



Tests:

- no exchange
- ballast exchange only
- chlorination only
- exchange & chlorination



- Experiments conducted on operational ships moving between Europe and Great Lakes
- Individual tanks can serve as separate experiments
- Demonstrated additive or synergistic benefits may be implemented globally by shippers

Advantages of a Network

1. Coordinated and efficient assembly of expertise
 - common sample teams spread across the country collecting samples the exact same way in each location
 - reduces cost, increases efficiency
2. Hierarchical studies from landscape to laboratory
3. National scope to address the most pressing questions
4. Multiple disciplines involved in each study
 - molecular, evolutionary, community and population ecology, statistics, modeling, hydrodynamics
5. Study large range of biota simultaneously
 - viruses, bacteria, dinoflagellates, diatoms, benthic and planktonic invertebrates
6. Allows experiments that are too big/expensive for one PI
7. International Collaborations (travel grants for students)
8. Large contingent of HQP trained (40-54 MSc, PhD, PDF in each network)

Thank You