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Microbial Pathogens and Biotoxins: State of the Gulf of Maine Report

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MICROBIAL PATHOGENS AND BIOTOXINS

STATE OF THE GULF OF MAINE REPORT

Gulf of Maine Council on the Marine Environment

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MICROBIAL PATHOGENS AND BIOTOXINS

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The *State of the Gulf of Maine Report*, of which this document is a part, is available at: www.gulfofmaine.org/stateofthegulf.

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1. Issue in Brief

Recreational activities, such as swimming and boating, and seafood consumption can expose humans to health hazards from water-borne microbial pathogens and biotoxins. Pathogens are microorganisms that cause disease, the most common being bacteria, viruses and protozoa (Table 1). Although there are numerous bacterial pathogens, those that cause the greatest public health concern for seafood consumption in the US and Canada are species from the genus *Vibrio*, which are naturally occurring bacteria in marine and estuarine habitats. Sewage-borne bacteria such as *Escherichia coli* and *Enterococcus* species have decreased in significance with improvements in sewage treatment, effective disinfection and diminished discharge of untreated sewage over the past four decades. Their role in public health assessments remains pivotal as the indicators used for detecting the presence and extent of fecal and sewage contamination, and as such, indicators of bacterial pathogens and viruses, in marine waters. Viruses are also significant public health threats, although their presence is indirectly inferred by coliform and enterococci data, and the incidence of illness is significantly under-reported. Equally important in the Gulf of Maine is the release of toxins from organisms such as algae, a condition also know as harmful algal blooms. Paralytic shellfish poisoning (PSP), caused by *Alexandrium fundyense*, is the most significant harmful algal threat in the Gulf of Maine. The presence and levels of all of these organisms is influenced by both human-induced and natural factors (Figure 1). In particular, increased population and coastal development have placed pressure on the water treatment facilities, resulting in an increase in sewage effluent and urban runoff. Non-point source pollution (e.g. agricultural and urban-stormwater runoff) is a significant source of increased contaminant loading into the Gulf of Maine (see also paper on Eutrophication). Climate change may cause changes to microbial community structures, due mainly to changes in water temperature and salinity. Legislation and policy within the Gulf of Maine focus on pollution control to assist in controlling the source of the problem. However, reactive responses, such as shellfish closures, beach closures and good medical assistance are also important. Long-term monitoring and research programs in the Gulf of Maine provide information for managing seafood harvesting and recreational activities to help prevent disease and harm to humans and ecosystems.

LINKAGES

This theme paper also links to the following theme papers:

- Land Use and Coastal Development
- Toxic Contaminants

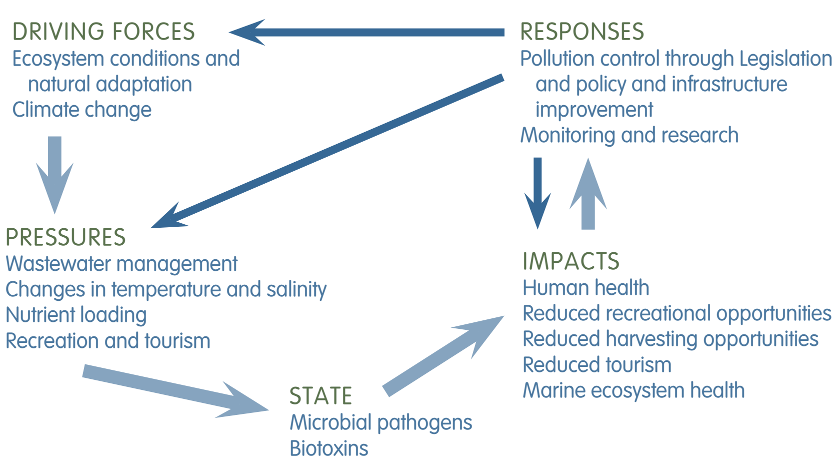

Figure 1: Driving forces, pressures, state, impacts and responses (DPSIR) to microbial pathogens and biotoxins in the Gulf of Maine. The DPSIR framework provides an overview of the relation between the environment and humans. According to this reporting framework, social and economic developments and natural conditions (driving forces) exert pressures on the environment and, as a consequence, the state of the environment changes. This leads to impacts on human health, ecosystems and materials, which may elicit a societal or government response that feeds back on all the other elements.

2. Driving Forces and Pressures

2.1 ECOSYSTEM CONDITIONS

Natural conditions and the adaptation of organisms to those conditions largely influence extent, distribution and structure of microbial and algal communities. Two different types of public health threats come from microorganisms found in the Gulf of Maine. Some microorganisms, called pathogens, cause infectious diseases, while others produce toxins that can poison humans who eat contaminated shellfish. There are some similarities between the two, like in mode of exposure (contaminated shellfish), but also some important differences, like the fact that cooking shellfish can kill disease-causing microorganisms and render the shellfish safe for consumption, while cooking does not neutralize biotoxins, so contaminated shellfish cannot be safely consumed even if they are fully cooked.

2.1.1 Microbial Pathogens

The microbial pathogens include different bacterial species, viruses and protozoa, with some that occur naturally in the marine environment and others that are sewage-borne pollutants. Different bacterial species respond to environmental conditions in different ways, partially because they associate with different environmental matrices (sediment, shellfish, plankton, infaunal burrows, etc.). For *Vibrio* spp.(see box), temperature is the major driving force. All three of the major pathogenic species thrive best in warm waters, and actually appear to vanish from colder waters (temperatures below 15°C (FAO/WHO 2005)), as they 'go dormant' during fall through spring in the Gulf of Maine. During the summer when their populations are at their peaks, nutrients indirectly affect Vibrio concentrations

Table 1: Types, species, sources, indicators for, potential health effects of and safety standards for microbial pathogens and marine biotoxins in the Gulf of Maine.

MICROBIAL ISSUE	SPECIES	SOURCES	INDICATORS	POTENTIAL HEALTH EFFECTS	SAFETY STANDARD
Fecal-borne microbial pathogens	Numerous bacteria, viruses and protozoan pathogens	Human sewage, feces from warm blooded animals; shellfish and recreational waters	Fecal coliform (FC), enterococci (ENT), *Escherichia coli* (Ec)	Mostly gastroenteritis, hepatitis	Shellfish: 14 FC/100 ml Marine recreation: 35 ENT/100 ml
Naturally-occurring bacterial pathogens	*Vibrio vulnificus* (Vv), *Vibrio arahaemolyticus, Vibrio cholerae* (Vc), *Vibrio alginoyticus*	Naturally occurring in estuarine and marine ecosystems; shellfish and recreational waters	No indicators, only direct detection of the species or suspected virulence marker genes	Gastroenteritis (Vp,Vc,Vv), wound infections (Vp,Vv), cholera (Vc), severe septicemia and death (Vv)	Shellfish (Gulf of Maine): monitoring of water temperature for favorable conditions
Harmful algal blooms	*Alexandrium fundyense*	Naturally occurring in marine ecosystems; shellfish	Mouse bioassay	Tingling, numbness, paralysis, death	80 µg PSP toxin/ 100 g shellfish meat or scallop roe

through stimulation of phytoplankton growth then death, a process that produces organic carbon, the energy source for growth of vibrios (Watkins and Cabelli 1985, Jones and Summer-Brason 1999). One significant knowledge gap is in understanding ecosystem conditions that favor strains of disease- causing vibrios and how those may be different from conditions that favor un-harmful strains. This is important information for shellfish harvest management. In the Gulf of Maine, the pathogenic vibrio species are most prevalent and persistent in warm estuaries, like the Great Bay Estuary of Maine (ME) and New Hampshire (NH), where they co-exist with oysters, yet studies to date suggest that strains that can cause disease are present at extremely low levels, if at all.

Estuarine and marine ecosystems are not the preferred environment for *E. coli* and other fecal-borne bacteria. They rapidly die under unfavorable high salinity conditions, are grazed by protozoa, or are inactivated by ultraviolet (UV) radiation and elevated water temperatures. Recent studies, however, have shown how these same organisms can persist and even grow in estuarine ecosystems under more favorable, lower salinity and higher nutrient level conditions (Bolster et al. 2005, Henroth et al. 2010) The key requirements for their survival are nutrients and low salinity. Intestinal viruses, including norovirus, astrovirus, adenovirus, rotavirus and enterovirus, generally mimic bacterial indicator responses to ecosystem conditions in the sense that they also persist during cold weather, then diminish in numbers during summertime in both the marine environment and within wastewater treatment systems (Burkhardt et al. 2000).

2.1.2 Harmful Algae

The growth of the harmful algae *A. fundyense* in the Gulf of Maine is complicated (Anderson et al. 2008). There is evidence for localized

VIBRIOS

Vibrios are a genus of curved, rod shaped bacteria that are natural inhabitants of estuarine and coastal ecosystems. They play key roles in carbon and nutrient cycling, closely associate with shellfish and plankton, are key partners in numerous host-microbe symbiotic relationships and can produce light via bioluminescence.

Three species are noteworthy, *Vibrio cholerae, Vibrio vulnificus* and *Vibrio parahaemolyticus,* because they are significant human pathogens. Human exposure is most commonly through consumption of raw or undercooked shellfish (most often oysters) or via exposure to water or surfaces in coastal areas. *V. cholerae* is perhaps the most well known vibrio species because it causes an intestinal infection and cholera. *V. parahaemolyticus* causes gastrointestinal illness (gastroenteritis) and, more rarely, wound infections. Disease symptoms may be less severe, but the incidence of disease in the US is the highest amongst the vibrios. *V. vulnificus* is a dangerous threat to people with compromised immune systems, i.e., liver disease, and has a 25% fatality rate with wound infections and a 50% fatality rate for shellfish-borne illnesses. It can cause severe septicemia or less severe gastroenteritis. Other vibrio species can also cause disease but are less common, although *Vibrio alginolyticus* causes numerous wound infections each year in the US and occasionally in the Gulf of Maine.

Until recently, vibrio diseases were rare and considered to be of minor significance in areas like the Gulf of Maine, where the incidence of vibrio diseases is still low. Recent outbreaks of *V. parahaemolyticus* disease in New York, Alaska and the Pacific Northwest, however, have caused a re-evaluation by public health officials of their potential significance in northern temperate ecosystems.

HARMFUL ALGAL BLOOMS (HABs)

The most common and important biotoxin-producing microorganisms are all marine algae and are called harmful algal blooms (HABs). These include the organism, *Alexandrium fundyense*, responsible for 'red tide', a condition that quite commonly causes shellfish harvesting to cease in the Gulf of Maine. These organisms are natural inhabitants of the Gulf of Maine where they pass through various life stages, some associated with toxin production. When there are high enough populations of these organisms that are producing toxins, the toxin-containing cells are taken up by filter-feeding shellfish, and/or accumulated in lobster internal organs to the point that ingestion of the shellfish by humans can cause severe toxin poisoning. The red tide HAB causes paralytic shellfish poisoning (PSP), a condition that affects the nervous system and paralyzes muscles. Other types of HABs include those that cause diarrhetic shellfish poisoning (DSP), a condition that includes rather mild and generic symptoms like diarrhea, cramping, and vomiting, and amnesic shellfish poisoning (ASP), a condition that affects the brain and causes memory loss and brain damage. These symptoms are all caused by different types of heat (cooking) insensitive toxins, including neurotoxins for PSP and ASP.

increases in PSP occurrence linked to elevated, near-shore nutrient (nitrogen) concentrations, especially in Casco Bay. For the larger regional open-water blooms, eutrophication effects are not well quantified, although a recent report on long-term monitoring in the Bay of Fundy suggests there is no link between nutrient (nitrogen, phosphorus, silica) concentrations and cell densities of *A. fundyense* and other harmful algal species (Martin et al. 2009).

2.2 HUMAN POPULATION AND COASTAL DEVELOPMENT

The continuing increase in human population in coastal areas (see Land Use and Coastal Development) and accompanying development, with its impacts to the natural ecosystem, are factors that have significant impacts on the concentrations and spatial extent of microbial pathogens and harmful algal blooms. Increases in population place greater demand on wastewater treatment facilities and associated infrastructure.   There are limitations on existing infrastructure for conveying waste, and the need for increased resource allocation to maintain and upgrade facilities. Wastewater pipes in sewered areas can convey too much volume to treatment facilities due to from improper connections, stormwater and infiltration of groundwater into leaky pipes. Leaky pipes can also release untreated sewage to surface and ground waters. Pressure on these facilities is worsened by the increasing number of large rainfall events that may cause water treatment facilities to be bypassed and result in discharges of untreated, pathogen-laden waste matter. Land areas not served by centralized sewage treatment systems are also potential problems, with septic systems causing contamination both to ground water and surface waters.

The discharge of nutrients have no direct public health impacts, but can have

SPATIAL INFLUENCE OF RAW SEWAGE DISCHARGE

The discharge of plastic discs from the Hooksett NH Wastewater Treatment Facility (WWTF) in March 2011 when heavy rains caused the discharge of 300,000 gallons of raw sewage discharge to the Merrimack River illustrates the potential spatial influence (southern Maine to Martha's Vineyard) and long-term potential impact of effluent discharge from one facility located well up into the Gulf of Maine watershed. Catastrophic events that cause bypass of treatment and discharges of untreated, pathogen-laden effluent can thus have major effects on water quality over time and space. An animated model of the spread of the discs with support from NERACOOS (Northeast Regional Association of Coastal Ocean Observing Systems):

The modeled distribution of discs reflects the eventual fate of the discs (Figure 2).

Figure 2: Distribution of sewage treatment discs in the Gulf of Maine on June 13, 2011 that were discharged from the Hooksett, NH WWTF on March 6, 2011. Map provided by NH DES Department of Environmental Services.

ecosystem impacts that stimulate the growth and persistence of harmful algae (Anderson et al. 2008), pathogenic vibrio species (Jones and Summer-Brason 1999, Watkins and Cabelli 1985) and even *E. coli* (Bolster et al. 2005). Long-term discharge of sanitary waste to groundwater can build up nutrient concentrations that leach to coastal surface waters and may have long-term influences on estuarine and marine water quality.

Development is accompanied by increases in surfaces, such as pavements and roofs. As a result, stormwater cannot as readily penetrate into the ground and runoff from these surfaces typically ends up as untreated discharges to surface waters. Rainfall is a well-documented cause of increased bacterial concentrations in coastal waters and is used as a criteron for closing shellfish harvesting in all Gulf of Maine jurisdictions. Bacteria are transported from roofs and pavement to surface waters. Increased runoff can also wash riparian areas of feces from wild animals, livestock and pets. Urbanized and suburbanized areas have characteristic wild animal (raccoon, skunk, deer) populations that adapt to land-use changes. These areas are also often favorable areas for Canada geese, ducks, and other water birds. All of these are sources of fecal-borne bacteria and can contain pathogenic microorganisms that can cause disease in humans. Although point source control has diminished the significance of human-borne sources of microbial pathogens, non-point sources continue to pose public health risks. The importance of non-human sources, however, has increased dramatically, and although the public health risk is not as severe as with human sources, animals do harbor microbial pathogens. Recent studies provide evidence suggesting increased public health risk from climate change related influences on microbial pathogens and harmful algal blooms is likely (Epstein 2005, Hunter 2003).

Recreation and tourism, mostly through boating and shipping also have an impact on the extent of microbial pathogens and biotoxins in the Gulf of Maine. Discharges of untreated sewage from cruise ships, small boats, and other vessels are a direct and potentially significant source of nutrients and of microbial pathogens; witness the high incidence of norovirus outbreaks on cruise ships (CDC 2011). Sewage waste also contains nutrients. Ballast water can contain microbial pathogens and biotoxin-producing algae, both native and exotic, which are potentially invasive species.

2.3 CLIMATE CHANGE AND EXTREME WEATHER EVENTS

The two main climate change factors important for microbial pathogens and marine biotoxins are increased water temperature and increased frequency of extreme weather events and the accompanying storm runoff. The continued increase in warm weather expected with current climate trend analyses, predict warmer summers and milder winters. Higher temperatures increases growth and metabolic rates for microbial pathogens under favorable conditions, resulting in greater persistence and higher concentrations of fecal-borne bacteria, *Vibrio* species and possibly harmful algae. In contrast enteric viruses tend to be less prevalent as environmental temperatures increase. A concern with *Vibrio* species is that increased levels and persistence may lead to evolution of virulent strains in existing populations that do not already include virulent strains (Mahoney et al. 2010).

The occurrence and dynamics of harmful algal blooms in coastal areas of the Gulf of Maine are driven by physical aspects of climate and weather conditions, including ocean currents, photosynthetically available radiation, the timing of freshening events, the nature of the North Atlantic Oscillation and hurricanes. Baker-Austin et al. (2010) presented recent evidence of the importance of changing ocean currents as a means for transport of pathogenic strains of Vibrio species to areas where they do not normally exist.

Runoff is a significant source of microbial pathogens, nutrients and toxic chemicals to coastal waters. The reduced salinity from increased freshwater volumes from more severe rain events can help to increase the persistence of enterococci, *E. coli* and sewage-borne pathogens in estuarine and coastal marine waters. Lower salinity also tends to favor the growth of *V. cholerae* and *V. vulnificus*. Runoff also delivers nutrients to coastal waters. Increased nutrients from runoff indirectly stimulates all three *Vibrio* species and the growth and persistence of nuisance harmful algae (eg. *A. fundyense*; Anderson et al. 2008).

3. Status and Trends

Based on microbial source tracking studies in the Gulf of Maine, sources of fecal-borne pollution vary widely by location, land use and season (Jones 2009, 2008). The trends for harmful algal blooms, fecal-borne microbial pollution and pathogenic vibrios are all increasing in the Gulf of Maine.

3.1 FECAL-BORNE MICROBIAL PATHOGENS

Fecal-borne microbial pathogens are not routinely measured across the Gulf of Maine as part of any monitoring effort. Instead, bacteria that are indicators of fecal pollution are used as surrogates that indicate the risk of exposure to viral and bacterial pathogens in fecal material and sewage.

Coastal beach monitoring programs in all five Gulf of Maine provinces and states assess risks due to fecal-borne microbial pathogens during the summer months. These provide a proxy indicator for water quality. The US Environmental Protection Agency (EPA) Beaches Program reports on a variety of statistics related to bacterial postings at beaches in Maine, New Hampshire and Massachusetts (MA). Information is not available on the frequency of beach postings in the Bay of Fundy. The EPA Beaches Program reports on how many beaches had advisories warning people to avoid contact with the ocean water, and on what percentage of days beaches were under these advisories. The trends in these statistics for each of the three states show a general increase from 2004 to 2009, with similar ranges of for each state (Figure 3). These increasing trends may in part be due to increased monitoring over the past decade.

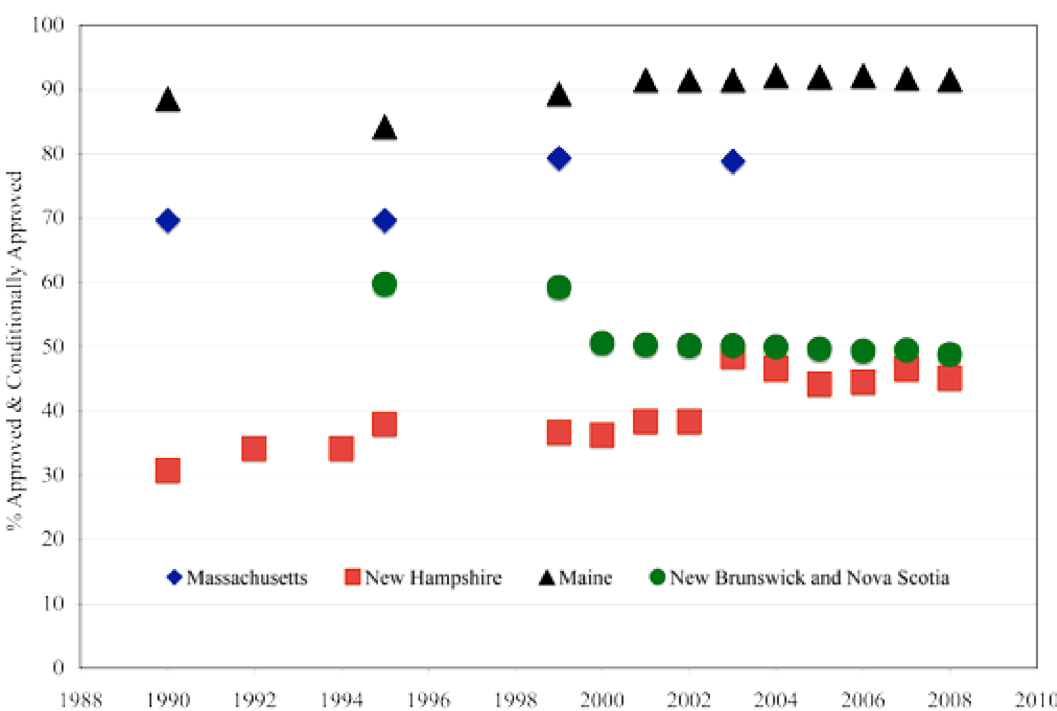

Figure 3: Trends in the percentage of shellfish harvest areas classified as approved or conditionally approved in the three states and the Bay of Fundy: 1990-2009.

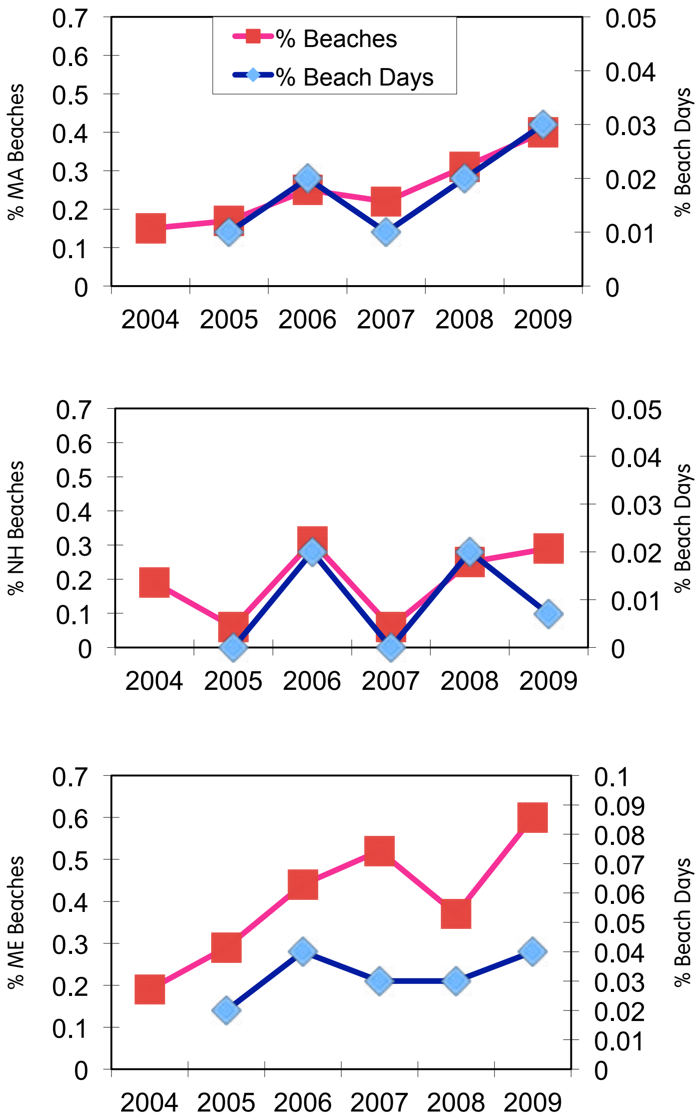

Figure 4: The annual percentage of days and the number of coastal beaches that were under advisories warning against contact with ocean water. Data from US EPA Beaches website: http://water.epa.gov/type/oceb/beaches/index.cfm

Closure of shellfish harvesting areas is an indicator for presence of microbial pathogens and toxin-producing algae. The Gulf of Maine Council's Ecosystem Indicator Partnership (ESIP) program has recently compiled shellfish harvest closure information for all five jurisdictions in the Gulf of Maine, building on previous efforts to track trends for pollution impacts on shellfish harvesting (Jones 2004).

The chosen indicator for levels of bacterial pollution is the percentage of harvest area classified as approved or conditionally approved, as opposed to prohibited (closed) or restricted (requiring depuration). The approved and conditionally approved areas in Massachusetts and New Hampshire have increased since the early 1990's, whereas little change has occurred in Maine and an overall decrease is apparent in the Bay of Fundy (Figure 4). These trends reflect a variety of influences, most notably efforts to eliminate pollution sources and changes to the way jurisdictions classify coastal waters. Based on the most recent available information, 80-90% of the harvest areas in Massachusetts and Maine are approved/conditionally approved, in contrast to <50% of the areas in the Bay of Fundy and New Hampshire.

3.2 NATURALLY OCCURRING BACTERIAL PATHOGENS

Vibrios tend to be most prevalent in the Gulf of Maine during warm summer months, and marine invertebrates may serve as reservoirs during colder months (Preheim et al. 2011).Vibrio studies in the Gulf of Maine have shown *V. parahaemolyticus* (Bartley and Slanetz 1971 Shiaris et al. 1987), *V. vulnificus* (O'Neill et al. 1990) and *V. cholerae* (Jones et al. 2010) to be present in coastal waters. Since the 1990s, studies in the Great Bay Estuary show increased occurrence and persistence of vibrios during 2007-2011, even during winter months when they were previously not detected (Jones et al. 2010). This is a concern, given the increasing occurrence of significant vibrio disease outbreaks in more northern US waters over the past 15 years. At present, no strains of *V. parahaemolyticus, V. vulnificus* and *V. cholerae* collected from

NH and ME waters have contained genetic markers indicating them as virulent strains (Jones et al. 2010). The incidence of disease in the Gulf of Maine has been relatively steady since 2000, though the reason for an increase in Massachusetts during 2007 and 2008 (see below) is unknown, and may be a change in the presence of virulent strains in local waters and shellfish.

3.3 TOXIN-PRODUCING ALGAE

A. fundyense is a naturally-occurring alga found throughout Gulf of Maine waters that blooms to elevated concentrations during summer months. Cells found during red tide blooms in the Gulf of Maine are in the reproductive stage of their life cycle. During cold winter months, *A. fundyense* exists as cysts lying on the ocean floor that will germinate once warmer water and increased light stimulate this transformation. Cells can then reproduce exponentially by simple division if nutrients are abundant and produce biotoxins until conditions are no longer favorable for growth. The cells then form gametes that join in pairs to form single cells, which develop into zygotes and then cysts that which remain dormant on the ocean floor until favorable conditions reoccur.

Whereas levels of fecal indicator bacteria are generally considered to correlate to pathogen levels, *A. fundyense* cell abundance in the Gulf of Maine is not consistently related to shellfish toxicity (McGillicuddy et al. 2005). *A. fundyense* cell abundance varies from year to year and by geographical area in the Gulf of Maine. Toxicity onset and disappearance tends to occur earlier and last longer in eastern Gulf of Maine than in the western areas. In addition, it has been hypothesized by scientists at Woods Hole Oceanographic Institute (WHOI) that the intensity of PSP bloom events is directly related to the number of *A. fundyense* cysts in bottom sediments (Figure 5), but the 2010 season did not bear this out. Though the number of cysts in sediments in the fall of 2009 was much higher than observed prior to the historic 2005 bloom, there were few PSP closures during 2010 in the western Gulf of Maine.

There is a general trend of increasing intensity in PSP blooms in the Gulf of Maine. A good illustration of this can be shown by the maximum toxicity measured in shellfish in Massachusetts Bay from 1972 to 2007 (Figure 6; WHOI, http://www.whoi.edu/northeastpsp/). The data start at 1972 because no PSP toxicity had been detected prior to that year in

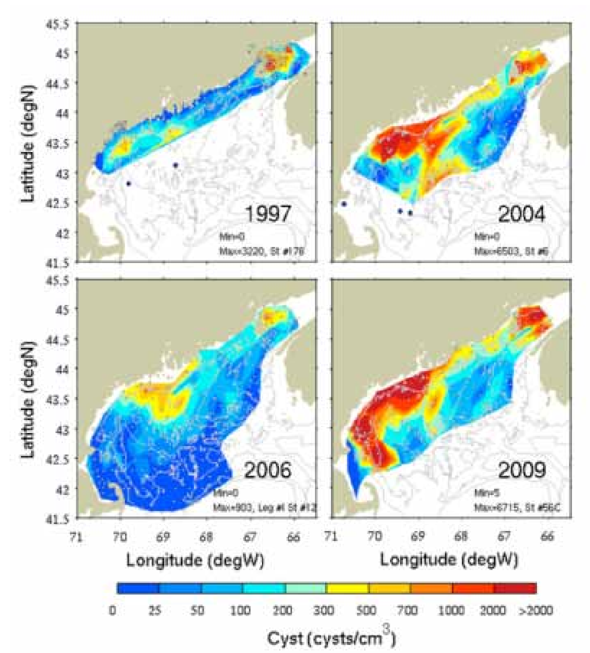

Figure 5: Maps of the Gulf of Maine reveal the concentration of *Alexandrium* cysts buried in seafloor sediments, as detected by WHOI-led surveys in the fall of 1997, 2004, 2005, 2006, 2007, 2008 and 2009. Four of those surveys are shown. The cyst abundance in 2009 is higher than ever observed in these past surveys. Note also that the *Alexandrium* cyst "seedbed" extends further to the south than was ever observed before. (NOAA, 2010)

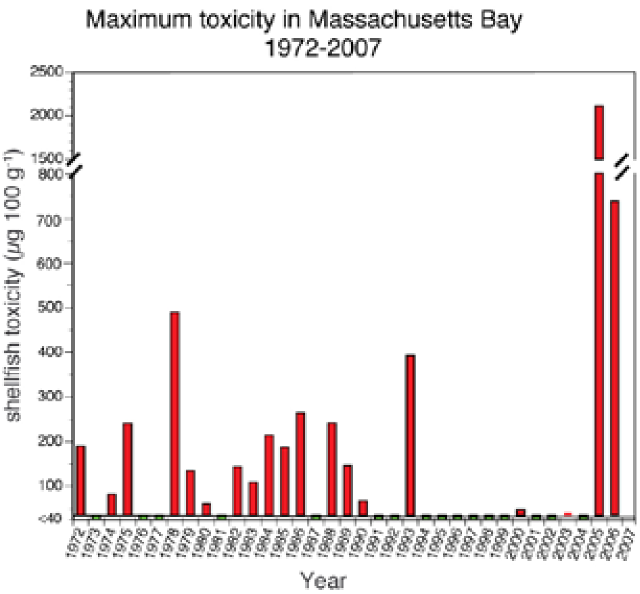

Figure 6: Maximum shellfish toxicity in Massachusetts Bay, 1972-2007 (WHOI, Don Anderson et al. 2011).

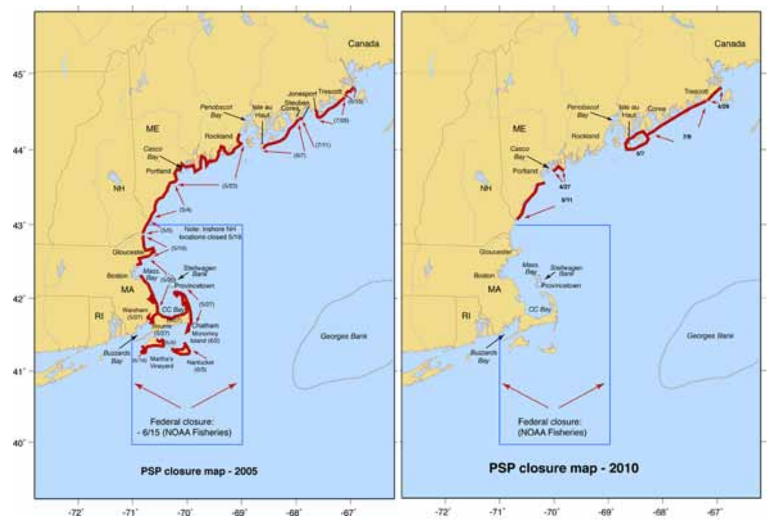

Figure 7: Interannual comparison of shellfish harvest closure maps caused by PSP toxicity: 2005 and 2010. Figures from WHOI/Don Anderson laboratory

Massachusetts waters, though it had been documented in Maine since 1958. From 1972 to 2004, PSP blooms occurred almost yearly in Maine but less frequently in Massachusetts and New Hampshire, with practically no toxicity detected in Massachusetts from 1994 to 2004 (Figure 6). In 2005, a bloom occurred that extended further south, and had both higher shellfish toxicities and cell densities of *A. fundyense*. A significant regional bloom occurred again in 2008, and variable degrees of severity in the blooms has occurred in other years since 2004, a factor due in part to different hydrographic and nutrient conditions in the Gulf of Maine. The spatial extent of shellfish harvesting closures due to PSP can differ greatly between years (Figure 7).

Blooms of *Psuedo-nitzschia* species, producers of domoic acid and the cause of amnesic shellfish poisoning, have been observed infrequently in the Bay of Fundy (Martin et al. 2009), with peaks in 1988, 1995 and 2004. Eight different *Psuedo-nitzschia* species have been observed in the Bay of Fundy, but they differ in domoic acid production and not all are detected each year. They also tend to bloom from May to October and the different species differ in ecosystems conditions favorable to blooms.

4. Impacts

4.1 HUMAN HEALTH

Fecal-borne pathogens cause mainly gastro-intestinal symptoms that are most often self-limiting, but also include hepatitis and wound infections, though some can cause much more serious infections. People can be exposed to pathogens mainly through consumption of raw or undercooked shellfish or through exposure to contaminated water and surfaces in coastal areas. The occurrence of diseases is probably under-reported to a significant level, mainly because the nature of most of the diseases is relatively mild and often difficult to attribute to a single activity. Large outbreaks, like norovirus infections associated with contaminated shellfish or more serious infections, like severe wound infections from exposure to vibrio pathogens in coastal areas, are perhaps the pathogens that people know best. This issue is the main force for both chronic shellfish harvest limitations and beach safety issues in the Gulf of Maine.

Studies have shown there to be a positive relationship between detected levels of bacterial indicators in water and the number of water-borne and shellfish-borne cases of human illness (EPA 1986). Routine water monitoring for human health protection often involves use of indicator bacteria and not the actual viral and bacterial pathogens. Based on risk analyses, beaches are posted as being polluted and shellfish harvesting is closed when the different indicators reach levels either as single samples or average concentrations over time that exceed acceptable risk levels.

Infectious enteric disease incidence is rarely reported according to source, except in larger outbreak cases where details are provided that could discern shellfish or swimming as sources or conditions responsible for infections. Vibrio infections are reported. The most critical vibrios relative to shellfish consumption are *V. vulnificus* and *V. parahaemolyticus*, and to a lesser extent *V. cholerae*, although there was a recent (March-May, 2011) non-cholera outbreak of gastroenteritis caused by consumption of raw oysters from Florida that contained *V. cholerae* (Onifade et al. 2011). Incidences of vibrio infections in Maine have been caused by *V. vulnificus, V. parahaemolyticus* and *V. alginolyticus*, and are reported each year by the ME Center for Disease Control and Prevention (CDC). Over the past 10 years, the incidence of vibrio infections has remained low (Robbins 2009), averaging 2.6 cases per year and ranging from 0 to 5 cases (Figure 8). Similar numbers (~2 cases/ year) of annual cases have been reported by the US CDC for New Hampshire from 2000 to 2008 through their national

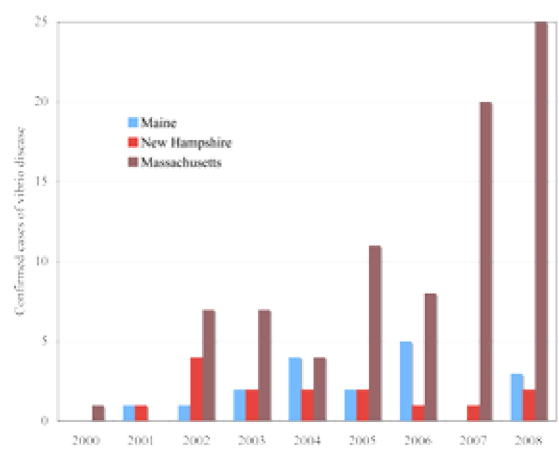

Figure 8: Annual confirmed cases of non-cholerae vibrio infections in Maine, New Hampshire and Massachusetts: 2000-2008. Data collected from ME CDC, MA Department of Public Health and the US CDC website: http://www.cdc.gov/nationalsurveillance/cholera_vibrio_surveillance.html

Vibrio surveillance system. It is important to reiterate that most, though not all, incidences of shellfish-borne vibrio infections have been attributed to shellfish harvested from areas outside of the Gulf of Maine. Marketed shellfish are tagged to identify harvest location, and many shellfish consumed around the Gulf of Maine are imported from outside the region. Massachusetts publishes the annual incidence of vibrio infections and have reported a higher rate of infections than in New Hampshire and Maine over the past 10 years (MADPH 2009).

Only *V. cholerae* is a reportable disease in the Gulf of Maine, as it is considered to be of great public health importance and thus is required to be reported by healthcare providers. For the three states in the Gulf of Maine, the National Shellfish Sanitation Program only requires assessment of shellfish growing waters for conditions that may be conducive to vibrio growth; if favorable conditions persist and/or there is incidence of disease, then active monitoring would be required. The Canadian Shellfish Sanitation Program requires the same for the two Gulf of Maine provinces. *Vibrio alginolyticus* is also a concern for recreational activities, as are *V. vulnificus* and *V. parahaemolyticus*, as wound infections can occur in contaminated waters. Recreational activities where people can be exposed to pathogens include swimming, surfing, diving, wading, boating, skiing and walking of falling on shore or rocks (Yoder et al. 2008).

For PSP toxins, the concern is poisoning of humans who consume shellfish with toxin levels that cause health-threatening symptoms. However, biotoxins and pathogenic vibrio species can also cause diseases in marine biota. The rare Gulf of Maine cases of poisoning from marine biotoxins are reported based on source location and shellfish species. PSP poisoning has recently become a reportable disease in Canada. A critical issue is educating the public about risks associated with recreationally harvested shellfish. A recent consumer awareness survey in Canada showed little public awareness and understanding about the risks of consuming bivalve shellfish, especially for recreationally harvested oysters (CREATEC 2006). The survey also reported Canadians on the East coast are more at risk than West coast residents.

PSP illnesses occurred in Maine in 2007, 2008 and 2009, all from consumption of shellfish taken from closed areas. The most notable set of cases was the July 31, 2007 incident where a family of four from Washington County consumed contaminated mussels and two people were hospitalized in critical condition, though they recovered. Recent investigations have shown lobster ‘tomalley’ should also be avoided during the summer as toxins can accumulate there and cause poisoning. Consumption warnings for lobster tomalley due in part to the potential of elevated levels of PSP toxin have been recently released in both Canada and the US.

The spatial and temporal trends for PSP closures Gulf-wide are complex, as closure of areas to shellfish harvesting changes annually, seasonally and spatially, with no obvious, discernable trends from year to year. Following a near absence of detectable shellfish toxicity during most of the 1990s, toxicity detection became more frequent in the 2000's (Figure 6). The worst years for the whole Gulf of Maine region were 2005 and 2008, based on the general increase in the incidence and intensity of PSP toxicity in the Gulf of Maine.

4.2 ECONOMIC AND SOCIAL IMPACTS

The perceived health benefits in seafood consumption and an increase in population have fueled increased worldwide consumption of shellfish and seafood. Recent estimates on the annual monetary losses associated with medical expenses, lost wages and lower productivity resulting from foodborne diseases in the US range in the billions of dollars (Scharf, 2010). Based on medical cost, quality of life losses and lost life expectancy, *V. vulnificus* had the highest cost per case of any pathogen. In addition, the public perception of shellfish safety can be negatively affected to a significant degree by reports of poisoning or infections. Perception of increased risk tends to decrease demand and have a negative impact on the seafood industry. Beyond foodborne illnesses, water-borne illness from marine pathogens and toxins also has significant costs (Ralston et al. 2011), including $300 million annually in the US for gastrointestinal illnesses associated with beach recreation. The same amount ($300 million) is the annual cost in Canada for treating health related problems related to water pollution, but the overall cost is much higher when considering beach and shellfish harvesting closures (Environment Canada 2001). Losses to the local Atlantic Canada (excluding Quebec) economy from shellfish harvesting closures are $10-12 million per year.

Significant economic impacts to the region have resulted from shellfish harvest closures due to elevated PSP levels in shellfish, especially in 2005 (Jin et al. 2008) and in 2008. Estimates of costs from the 2005 event ranged from a low estimate of $2.8 million for Maine to $18.4 million in Massachusetts. Federal relief was provided following the 2005 and 2008 bloom seasons through Red Tide Technical Assistance and Disaster Relief programs to each of the three states, but losses that occurred in other years were not supported with relief. Because marine biotoxins cannot be destroyed with cooking, closures are absolute and last until toxins diminish to non-threatening conditions.

The increase in potential pollution loading threatens shellfish harvesting areas and the livelihood of shellfish harvesters and aquaculturists. Many shellfish harvesting areas are routinely classified in ways that prevent the direct harvest and marketing of contaminated shellfish. Ongoing efforts throughout the Gulf are underway to identify and eliminate sources of fecal pollution to help open more shellfish harvesting area. Significant rainfall events, which are likely to increase in frequency with predicted climate changes, most likely will result in widespread closures in all areas of the Gulf for periods of time related to the severity of the event. This increases desirability and adds pressure on harvest areas that are free from mooring areas, wastewater effluent discharge and other pollution sources and are clean enough to support shellfishing. For restricted areas, harvesting is allowed if shellfish are then depurated. In Massachusetts, the Newburyport Shellfish Purification Plant is managed by the Department of Fish and Game, Division of Marine Fisheries and processes an average of 560 bushels of soft shell clams every week that are harvested from conditionally restricted areas of Boston Harbor. In Maine, many areas have become available for harvest because the shellfish can be depurated at a commercial facility (Jones et al. 1991). This helps to maintain a greater degree of employment for local clam diggers. Unfortunately, the demand for soft shell clams in the Gulf of Maine is highest during summer months when red tides occur.

4.3 BIOLOGICAL IMPACTS OF INCREASED FREQUENCY, DURATION AND INTENSITY OF MICROBIAL PATHOGEN AND BIOTOXIN OCCURRENCE

Significant mortalities of caged Atlantic salmon occurred in New Brunswick during a bloom of *Alexandrium fundyense* in 2003 (Sephton et al. 2007). PSP toxins in right whales have also been detected during a bloom of *Alexandrium fundyense* (Doucette et al. 2006). These studies suggest that elevated concentrations of PSP toxin can be lethal to salmon and compromise the health of whales. Both studies investigated links through the marine food chain and found elevated levels of PSP toxin in zooplankton, blue and horse mussels and lobsters, even well after the blooms.

Vibrio species, including species not yet mentioned as human pathogens, are significant and well documented pathogens of finfish, eels and other marine organisms. In the Gulf of Maine, limp lobster disease has been a significant loss to the lobster fishery, and there is evidence that vibrios are the causative agent (Tall et al. 2003). Fecal-borne microbial pathogens are generally thought to have little impact on the marine ecosystem, as they are allochthonous organisms. *Salmonella* spp. have been implicated in fledgling tern die off on Cape Cod, and there are some suggestions that enteric bacteria may be involved in seal diseases. Fecal-borne bacteria can be transported from wastewater facilities and landfills to marine ecosystems via sea gulls (Nelson et al. 2008), thus representing a potential zoonotic (i.e. non-human animal) reservoir and vector for human diseases. Their build up in favorable niches within coastal ecosystems suggests they may have some influence on microbial communities and their potential for ecosystem impacts.

5. Actions and Responses

The Gulf of Maine faces significant economic issues related to microbial pathogens and harmful algal blooms, despite having a relatively low incidence of seafood borne and recreational diseases and toxin poisoning cases. The threat of economic loss has helped to keep these issues in the forefront of management and legislative agendas. A variety of policy, legislative, program and research actions and efforts directly address the impacts of microbial pathogens and harmful algal blooms. The regulations and policies addressing these issues have been established and modified over the long term by several mechanisms, including detailed state and provincial legislative actions and ongoing considerations through organizations like the Interstate Shellfish Sanitation Conference (ISSC).

5.1 POLICY AND LEGISLATION

Both the National (US) and Canadian Shellfish Sanitation Programs (NSSP & CSSP) control and regulate the sanitary quality of shellfish harvested for human consumption (Table 2). In Canada, the Canadian Food Inspection Agency, Environment Canada and Fisheries and Oceans Canada (DFO) work collaboratively to meet CSSP requirements and to minimize health risks from biotoxins and microbial pathogens. The NSSP is a cooperative federal/state program for sanitary control of shellfish used for human consumption in the US involving the US Food and Drug Administration and the ISSC.

Ongoing federal programs in the US and Canada continue to support upgrades to wastewater treatment facilities and infrastructure. The US EPA is currently in the process of permitting stormwater discharges from small Gulf of Maine municipalities through their Stormwater Discharges from Municipal Separate Storm Sewer Systems (MS4) program, following the permitting of larger cities over the past few years under preceding stormwater management programs. Impaired waters throughout the US Gulf of Maine watershed are managed through the EPA Total Maximum Daily Load (TMDL) program. TMDLs designed to address microbial pathogen pollution problems have been developed for a number of impaired waters in the three states, and Maine and New Hampshire have developed statewide bacterial TMDLs.

The percentage of municipalities with wastewater treatment in Canada increased from ~70% in 1983 to 97% in 1999 (Environment Canada 2001). The degree to which the effluent is treated has also improved in Canada, yet there remained a significant problem in Atlantic Canada with the discharge of untreated or poorly treated effluent. Environment Canada has recently been pursuing studies to assess municipal sewage pollution sources that include hydrodynamic modeling to estimate impacts to shellfish beds and appropriate re-classification for harvesting (Klaamas et al. 2011). Part one of a new set of regulations for wastewater

Table 2: Legislative actions related to microbial pathogens and harmful algal blooms in the Gulf of Maine.

JURISDICTION	LEGISLATIVE ACTION	AGENCY	DESCRIPTION
Canada	Canadian Shellfish Sanitation Program (CSSP)	Canadian Food Inspection Agency, Environment Canada, Fisheries and Oceans Canada	Interagency effort to inspect shellfish, monitoring growing areas and enforcement in accordance with CSSP requirements, including issues related to microbial pathogens and biotoxins
US	National Shellfish Sanitation Program (NSSP)	Food & Drug Administration (FDA), Interstate Shellfish Sanitation Conference (ISSC)	Cooperative federal/state program for sanitary control of shellfish used for human consumption
		MICROBIAL PATHOGENS	
Canada	Wastewater System Effluent Regulations	Environment Canada	A new federal set of regulations and standards
US	Beaches Environmental Assessment and Coastal Health Act	US Environmental Protection Agency	Improve the quality of coastal recreational waters in the US
US	Stormwater Discharges from Municipal Separate Storm Sewer Systems (MS4)	US Environmental Protection Agency	Under the NPDES program of the Clean Water Act (CWA), to require NPDES permit coverage for stormwater discharges
US	Impaired Waters and Total Maximum Daily Loads (TMDL)	US Environmental Protection Agency	Under section 303(d) of the CWA, list and develop TMDLs for impaired waters
		HARMFUL ALGAL BLOOMS	
US & Canada	Consumption advisory for lobster tomalley	US FDA, Health Canada, MA & ME state shellfish programs	Advisories issued following the severity of the 2008 red tide bloom in the eastern seaboard
US	Red Tide Relief Program	NOAA	Following the 2005 and 2008 red tides, shellfish industry stakeholders in ME, MA & NH were compensated and monitoring, research and outreach were supported.

systems and national standards for wastewater effluent quality has been published (Canada Gazette 2010). Part two, which will include responses to part one, will be published in 2011 in the Canada Gazette under the title "Wastewater System Effluent Regulations."

The recent focus in the northeast US on reducing nutrient loading to coastal waters where harmful algal blooms and vibrios problems often occur will in part be addressed through the National Pollutant Discharge Elimination System (NPDES) permitting and the Total Maximum Daily Load (TMDL) processes. Municipal wastewater treatment facilities (WWTF) may well face expensive upgrades to reduce nitrogen. It is critical with these significant expenditures for nutrient removal that reducing the discharge of microbial pathogen should also be considered.

US federal support was made available through the National Oceanic and Atmospheric Administration (NOAA) following the 2005 and 2008 red tides in the

Gulf of Maine to help affected stakeholders in Maine, Massachusetts and New Hampshire. The support included funding to help improve monitoring in the region through support of red tide research and biotoxin detection method development.

The US Food and Drug Admistration (FDA) has used male specific coliphage as a viral indicator to determine conditions under which depuration of shellfish impacted by treated wastewater effluent could be used as a potential mitigation strategy for limited harvesting (Burkhardt et al. 2011). The US FDA has also initiated requirements for tighter controls on conditions that can increase the risk of *Vibrio* disease incidence. Even tighter controls for reducing both *V. vulnificus* and *V. parahaemolyticus* in harvested shellfish throughout the US and Canada are currently being considered.

5.2 MONITORING AND RESEARCH

There are monitoring programs that track microbial pathogens and toxin-producing algae in the Gulf of Maine to protect human health from risks associated with seafood consumption and swimming in contaminated water. The US National Shellfish Sanitation Program (NSSP) and Canadian Shellfish Sanitation Program (CSSP) manage shellfish harvesting for the most part based on risks from fecal-borne microbial pathogens and toxin-producing algae. Indirect management for pathogenic vibrios through water temperature monitoring is now a part of the US Gulf of Maine state (ME, NH, MA) shellfish programs. The NSSP and CSSP guidelines have recently included management guidelines for harvesting shellfish to reduce the incidence of shellfish-borne vibrio infections (NSSP 2009). Fecal-borne microbial pathogens are also monitored by provincial and state agencies to protect swimmers at marine beaches. Efforts in the Gulf of Maine states and provinces are also underway to identify sources of bacterial contamination in shellfish and bathing beaches using microbial source tracking and chemical tracers, and to improve wastewater disinfection strategies.

Harmful algal blooms and PSP biotoxin levels are tracked by shellfish programs in each of the three states and by Fisheries and Oceans Canada in the Bay of Fundy. Monitoring efforts and shellfish harvest management take into account location, timing, species and the use of some early warning efforts to limit public exposure to toxic shellfish. The latest closure information is posted on websites for all Gulf of Maine areas, delineated by monitoring areas and shellfish species. Warnings are also communicated for recreational shellfish harvesting through web postings and press releases. Biotoxin monitoring in the Bay of Fundy occurs mostly in Southwest New Brunswick. Monitoring for PSP and domoic acid, along with less frequent diarrheic shellfish poisoning (DSP) monitoring, occurs at least monthly (Martin et al. 2006, 2009, personal communications Canadian Food Inspection Agency, New Brunswick Operations). In the US, PSP monitoring is conducted at ~40 sites in Massachusetts from the North Shore into Cape Cod Bay, at two sites in New Hampshire and at many sites in Maine on a weekly basis from March through October. Volunteer phytoplankton monitoring programs in the three states, and the DFO monitoring program in southwest New Brunswick serve as useful early warning systems for the presence of HABs and other phytoplankton. Though most monitoring effort is for PSP, there is evidence that domoic acid, amnesic shellfish poisoning and DSP may become future problem areas that would require enhanced effort and monitoring.

Non-governmental organizations are putting pressure on the US EPA to improve water quality at beaches by conducting new swimmer health studies, develop tests that give same-day results, and addressing a wider array of potential water-borne illnesses. The Natural Resources Defense Council (NRDC) rates each state's water quality conditions. In 2010, Maine ranked 25th nationally, Massachusetts ranked 15th and New Hampshire 1st in the nation in beach water quality. This is an effective way for communicating to the public the potential risks and needs for addressing ongoing pathogen issues.

5.3 MODELING AND FORECASTING NEEDS

The most critical need for addressing impacts from microbial pathogens and harmful algal blooms is improved risk prediction. Development of predictive models for forecasting the potential for red tide incidence and intensity is well underway through partnered research involving academic and government researchers across the Gulf of Maine. These models are focusing on the relationships between numbers of *A. fundyense* cysts in sediments, nutrients (particularly nitrogen), climatic conditions and physical forcing factors. For all categories of pathogens and harmful algae, improved understanding of climate and ecosystem factors is also a critical research need. The potential emergence of new microbial pathogens, and the potential for continued increases in the extent and intensity of HABs and pathogenic vibrio problems are real issues. Given the nature of our collective response to the soaring increase in Lyme disease incidence in the Gulf of Maine states, it would be prudent to anticipate these problems and be better prepared to educate the public of risks and to improve our detection methods and disease diagnostic capabilities.

One of the key limitations of monitoring programs for both types of microbial issues is detection methods. The traditional use of a mouse bioassay has worked well, but it has a number of limitations and is undesirable because of the necessity to kill mice in the process. Efforts worldwide and within the Gulf of Maine are underway to replace this method with methods involving chemical detection of toxins. Detection methods that can detect virulent strains of all three vibrio species in a timely and cost-effective fashion are needed, especially for colder water areas like the Gulf of Maine.

For fecal-borne pathogens, the traditional bacterial indicators have generally been effective, but their limitations are numerous and well known for a long time. There are new efforts to use assays to target enteric viruses, or indicators thereof like male-specific coliphage. This has recently been approved for use in a Maine shellfish harvesting 'prohibited' area to allow for seasonal (summertime) harvest of soft shell clams. A critical need for same-day detection of waterborne pathogens for use at coastal beaches is a focus of research efforts at EPA and other labs. EPA has developed a rapid, molecular method for detecting enterococci, and this is being tested in coastal waters nationwide.

INDICATOR SUMMARY

INDICATOR	POLICY ISSUE	DPSIR	TREND*	ASSESSMENT
Percent harvest area classified as approved	Pollution impacts on shellfish harvesting	State	/	Good
Annual percent of days beaches were under notification actions	Pollution impacts on beach safety	State	-	Fair
Frequency of shellfish harvest closures due to PSP toxin levels	Safety of shellfish consumption	State	-	Poor
Occurrence of storm events	Worsens impacts from rainfall runoff	State	-	Poor
Annual incidence of PSP poisoning, vibriosis and gastroenteritis	Human health effects of exposure to contaminated shellfish	Impact	-	Poor
Costs of disease incidence and toxin poisoning	Increasing costs of impacts	Impact	-	Poor

* KEY:
– Negative trend
/ Unclear or neutral trend
\+ Positive trend
? No assessment due to lack of data

Data Confidence

- The classification of shellfish harvesting areas is updated each year in each jurisdiction based on test results of the 30 most recent water samples for bacterial indicators of pollution.
- The assessment of beach water quality is summarized each year in each state based on weekly water sample testing for bacterial indicators of pollution.
- PSP toxin levels are directly measured in shellfish collected at least weekly in many locations.
- The frequency of PSP poisoning is based on actual reported incidences, and is probably underreported, especially mild cases, and it has only been nationally notifiable in Canada since 2008 and is not a reportable illness in the US. The incidences of vibriosis and gastroenteritis are underreported to a large degree because of the relative mild symptom in many cases, and the wide array of potential causes.

Data Gaps

- The information on incidence of infectious diseases associated with marine water contact and seafood, and of biotoxin poisoning from seafood consumption, is not readily available and should be tracked in an organized way.
- The incidence and timeframe for persistence of pathogenic vibrio species and sewage-borne viruses should be determined across the Gulf of Maine.

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