Chapter 6.2

Assessment of harmful algae bloom species in the Maryland Coastal Bays

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Abstract
Thirteen potentially harmful algae taxa have been identified in the Maryland Coastal Bays: *Aureococcus anophagefferens* (brown tide), *Pfiesteria piscicida* and *P. shumwayae*, *Chloromorum/ Chattonella* spp., *Heterosigma akashiwo*, *Fibrocapsa japonica*, *Prorocentrum minimum*, *Dinophysis* spp., *Amphidinium* spp., *Pseudo-nitzchia* spp., *Karlodinium micrum* and two macroalgae genera (*Gracilaria, Chaetomorpha*). Presence of potentially toxic species is richest in the polluted tributaries of St. Martin River and Newport Bay. Approximately 5% of the phytoplankton species identified for Maryland's Coastal Bays represent potentially harmful algal bloom (HAB) species. The HABs are recognized for their potentially toxic properties and, in some cases, their ability to produce large blooms negatively affecting light and dissolved oxygen resources. Brown tide (*Aureococcus anophagefferens*) has been the most widespread and prolific HAB species in the area in recent years, producing growth impacts to juvenile clams in test studies and potential impacts to sea grass distribution and growth (see Chapter 7.1). Macroalgal fluctuations may be evidence of a system balancing on the edge of a eutrophic (nutrient-enriched) state (see chapter 4). No evidence of toxic activity has been detected among the Coastal Bays phytoplankton. However, species such as *Pseudo-nitzschia seriata*, *Prorocentrum minimum*, *Pfiesteria piscicida, Dinophysis acuminata* and *Karlodinium micrum* have produced positive toxic bioassays or generated detectable toxins in Chesapeake Bay. *Pfiesteria piscicida* was retrospectively considered as the likely causative organism in a large historical fish kill on the Indian River, Delaware. Similarly *Chloromorum toxicum* (aka *Chattonella* cf. *verruculosa*) was implicated in a large fish kill and persistent brevetoxins detected in Delaware's Rehoboth Bay during 2000. Tracking potential HAB species diversity, abundance, distribution and toxic activity through time provides important indicators of environmental change for the Coastal Bays.

Introduction
Algae are important components of aquatic ecosystems, forming the base of the food chain by converting sunlight to energy (photosynthesis). Certain types of algae may become harmful if they occur in an unnaturally large abundance (termed a harmful algal bloom or HAB) or if they produce a toxin that can harm aquatic life or humans. HABs are increasing worldwide. Many have been related to increases of nutrients from human activities. Blooms of harmful algae cause the potential for economic loss related to decreased recreational and commercial fishing, and tourism.

Monitoring
Biomonitoring programs identify species and estimate abundance of algae through microscope counts and genetic probe technologies. There are recognized thresholds for some HABs from

regions in the world where particular organisms have presented chronic problems to human health and the environment. Such threshold levels have been used by managers or industries to initiate shellfish closures, beach closures and intensify monitoring which can include toxin testing. Toxin testing may proceed if human or living resource impacts are observed (Table 6.2.1). While no algae has shown toxicity from Maryland's Coastal Bays, some of the same organisms have proven toxic along eastern seaboard and in particular in the Chesapeake and Delaware bays. The list of HABs and published thresholds of management interest are being used here as a means of producing an environmental indicator for tracking by site, watershed and the bays overall: Threshold Level Exceedances of Abundance measured in samples for the list of recognized HABs in the region based on routine phytoplankton monitoring program results. For some species, no density threshold exists. The indicator may require evolving into toxin detections and exceedances of regulatory limits for toxin exposure as monitoring programs evolve with new technologies being brought online. A second indicator of relative condition may be the frequency of encounters for HAB species during routine monitoring. This information has been provided in the report.

***Draft* HAB Indicator:** threshold exceedances

Table 6.2.1 Summary of harmful algae species present in the coastal bays and associated threshold levels.

Species	Abundance Threshold	Comments
Akashiwo seanguienum	None	
Alexandrium sp.	500 cells/ml	
Amphidinium sp.	None available. Test for ciguatera toxin*.	**Amphidinium* has been found toxic in subtropical and tropical waters, not yet at temperate latitudes.
Aureococcus anophagefferens	Category 1 < 35,000 cells*ml^{-1} Category 2 $\geq$ 35,000 and $\leq$ 200,000 Category 3 > 200,000	Gastrich and Wazniak 2000
Chloromorum toxicum (Chattonella cf. verrculosa)	10,000 cells*ml^{-1} (Test for brevetoxin)	Estimated based on the 2000 Rehobeth Bay fish kill that included brevetoxin detection. Bourdelais et al. 2002.
Cyanobacteria	*Microcystis* 10,000 cells*ml^{-1} *Anabaena* *none* *Amphizomenon* *none* *Lyngbya* 10,000 cells*ml^{-1} *Oscillatoria* *none* *Synechococcus* 400,000 cells*ml^{-1}	
Dinophysis sp.	5 cells*ml^{-1} Test for okadaic acid.	Levels that can initiate further testing for toxins around the world.
Fibrocapsa japonica	None available, (Test for fibrocapsin or bioassay).	
Gonyaulax sp.	none	
Heterocapsa sp.	>100,000 cells*ml^{-1}	
Heterosigma akashiwo	1,000 cells*ml^{-1}	Average of 500-1,000 cells*ml^{-1} from fish kill events that require

		mitigation. Anderson et al.
Karlodinium micrum	10,000 cells*ml^{-1} Test for karlotoxin activity: hemolytic, cytotoxic and ichthyotoxic testing may occur.	Kempton et al. 2002 lower threshold for fish kill effects.
Pfiesteria piscicida, P. shumwayae	Low, Toxic bioassay tests required.	300 cells*ml^{-1} of *Pfiesteria* Complex Organisms has been considered but toxicity bioassays required.
Prorocentrum minimum	3,000 cells*ml^{-1} Bioassay toxicity tests – toxin is not yet characterized.	Initial effects thresholds on living resources, EPA 2003
Pseudo-nitzschia sp.	200-1000 cells*ml^{-1} Test for domoic acid (Some international standards available)	In Canada, Domoic acid only detected with > 1,000 cells*ml-1; New Zealand increases shellfish testing > 200 cells*ml^{-1} and closes shellfisheries > 500 cells*ml-1
Macroalgae	No threshold	

Status of potentially harmful algal bloom species

I. *Aureococcus anaphagefferens* (Brown Tide)
Brown Tides are not thought to be toxic in Maryland but are poor food for shellfish and produce such dense blooms that block light for underwater grasses. Below are results from the routine phytoplankton monitoring program for Brown tide using light microscopy; however, this small species generally requires a more specific technique to properly identify it. Since brown tide (*A. anophagefferens*) has been the most widespread and prolific HAB species in the area in recent years, producing growth impacts to juvenile clams in test studies and potential impacts to sea grass distribution and growth**,** DNR oversees a separate monitoring program for Brown Tide in cooperation with the National Park Service at Assateague Island (ASIS) and the Maryland Coastal Bays Program (MCBP) (see Chapter 6.1).

Routine light microscopy counts of *Aureococcus* in the Coastal Bays show peak blooms during the summers of 2006 and 2007. Blooms were also observed in fall of 2008 and summers of 2009, 2010 and lesser in 2011. Blooms have been found primarily in Chincoteague Bay and the Newport watershed and to a less degree in Turville and Manklin Creeks, the St Martin River and Isle of Wight Bay.

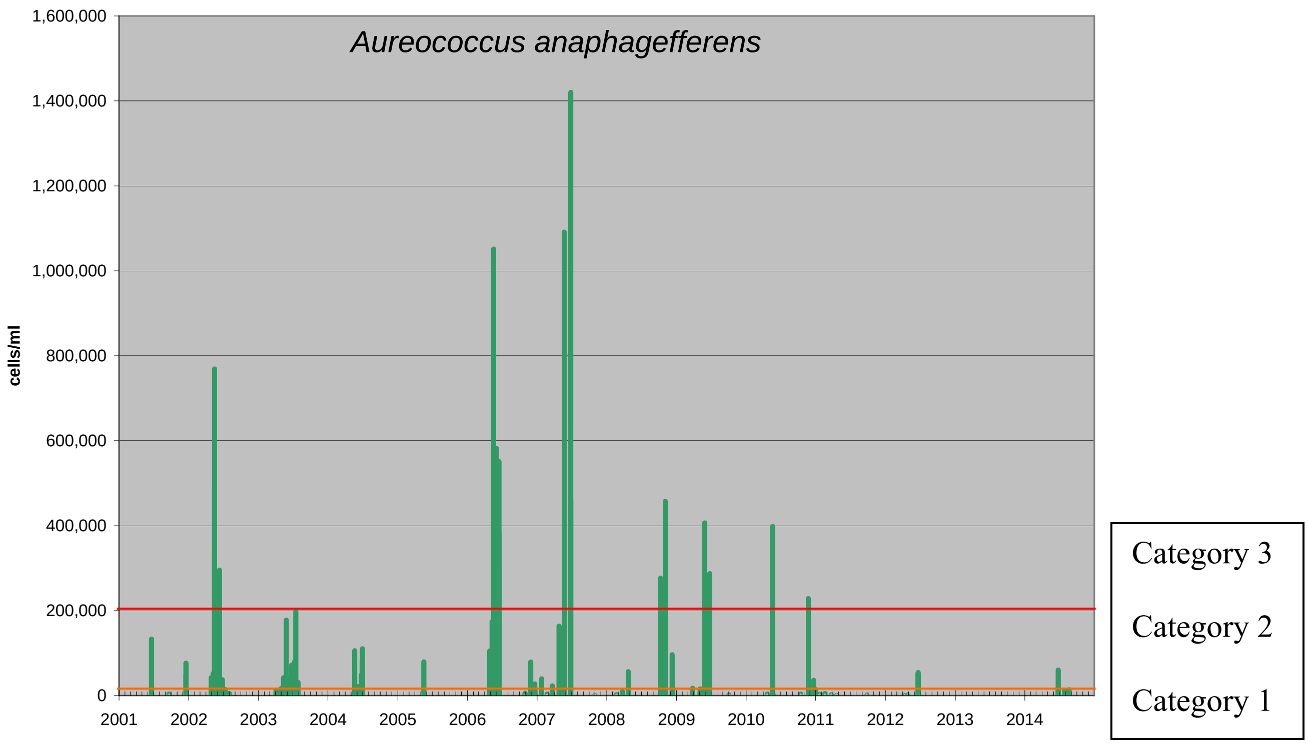

Figure 6.2.1 Occurrence of Brown Tide (*Aureococcus anaphagefferens* in the Maryland Coastal Bays between 2001-2014.

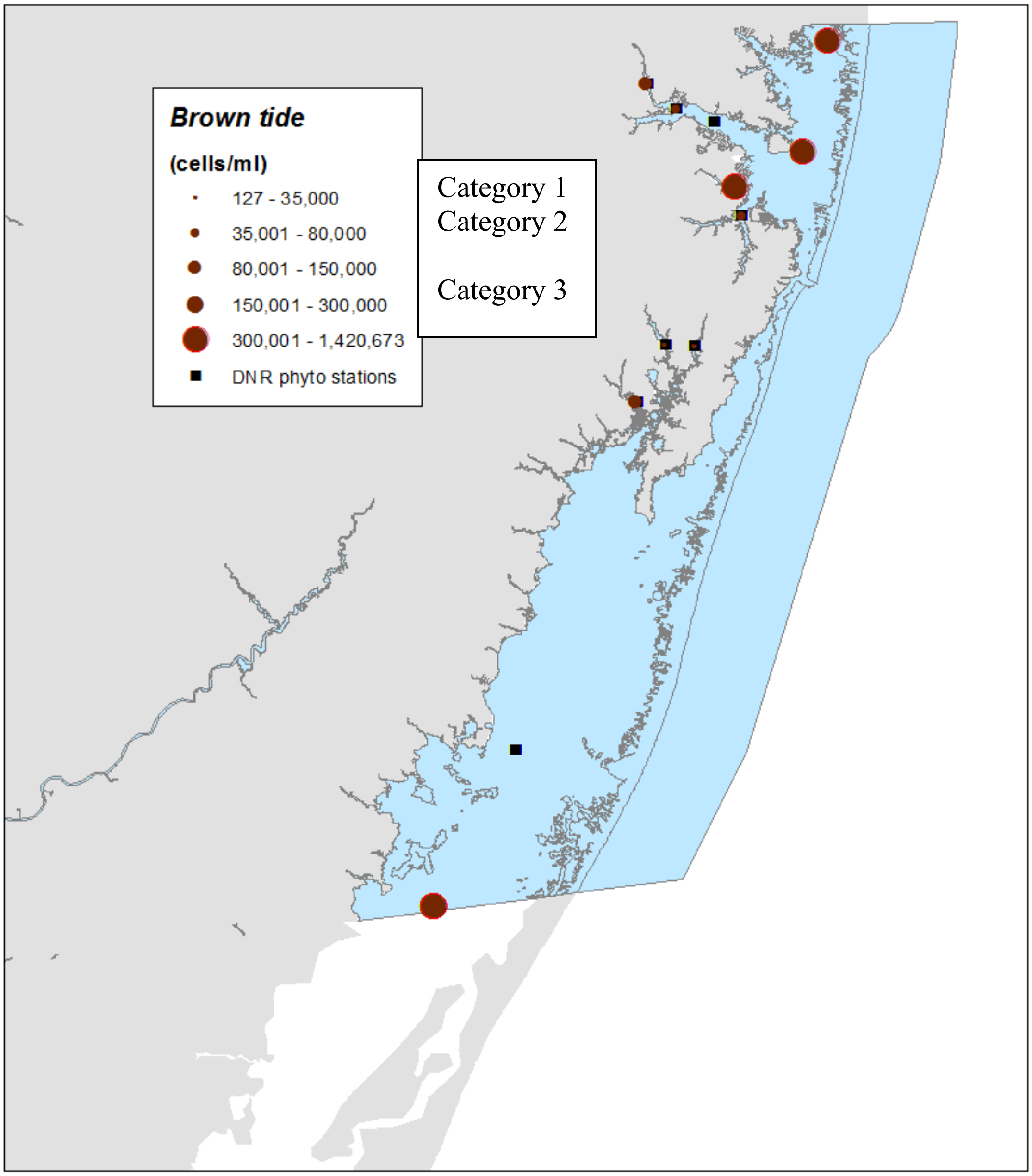

Figure 6.2.2 Distribution of Brown Tide (*Aureococcus anaphagefferens*) in the Maryland Coastal Bays between 2007-2014.

II. Raphidophytes: Chloromorum Chattonella, Heterosigma, and Fibrocapsa

The raphidophytes contain 12 known species, four such species have been identified from the Coastal Bays: *Chloromorum toxicum* (formerly *Chattonella* cf. *verruculosa*), *C. subsalsa, Heterosigma akashiwo* and *Fibrocapsa japonica*. Strains of *Chloromorum toxicum, H. akashiwo* and *F. japonica* have demonstrated toxic activity elsewhere in the world, however, there has been no evidence of toxins from any Raphidophytes in Maryland waters.

a. Chattonella

There are two species of *Chattonella* known in the Coastal Bays, *Chattonella* cf. *verruculosa aka Chloromorum toxicum* (may produce toxin), and *C. subsalsa* (not known to produce toxin). It includes the species *Chattonella subsalsa*, a bloom forming alga responsible for large scale fish deaths due to the synthesis of toxic compounds related to brevetoxin. *Chloromorum toxicum* is a potentially toxic species that has been associated with fish kills as near as in the Delaware Bays and can be potentially harmful to humans when producing brevetoxins. Brevetoxin is the same class of toxins as those produced by *Karenia brevis* (previously *Gymnodinium breve)*, associated with red tides, fish kills and sea mammal deaths in the Gulf of Mexico, and fish kills in Japan and Norway. Human exposure to brevetoxins can cause itchy skin, runny nose, watery eyes, wheezing and in some cases serious asthma attacks. Continued monitoring has not found the toxin in Maryland. Densities above 10,000 cells*ml^{-1} have been associated with toxin production and impacts on fish health (Bordelais et al. 2002). *Chloromorum toxicum* has been mainly found in Marshall Creek, Ayer Creek and St. Martin River.

Analysis of historic state phytoplankton data from intensive surveys of the St. Martin River in 1983 and 1992 suggested that *Chloromorum toxicum*, *Chatonella subsalsa* and *Fibrocapsa japonica* were present in what appears to be lower concentrations ten to twenty years ago than what has been observed in recent survey years. Historical identifications of Raphidophytes are based on journal drawings of cells identified in the Maryland Department of Environment monitoring program (MDE) (Walt Butler, Maryland Department of Natural Resources, Personal communication).

2007 Ten occurrences of *Chatonella* (8 *Chattonella subsalsa* and two *Chatonella cf)* were documented but all remained below 3,000 cells*ml^{-1}. Presence was limited to Ayres and Trappe Creeks.

2008 A *Chattonella subsalsa* bloom was documented in Ayres Creek on August 21 (12,954 cells*ml^{-1}). It was also present in Turville and Trappe Creeks (<300 cells*ml^{-1}) in July and August respectively. *Chloromorum toxicum* was detected cells*$ml^{\mathbf{-1}}$.

2009 A bloom was detected in July in Ayres Creek (56,515 cells*ml^{-1}) and just below bloom threshold levels were also detected in Trappe Creek (9,906 cells*ml^{-1}). No toxicity testing was performed. By August both blooms had dissipated to less that 2,600 cells*ml^{-1}.

2010 *Chattonella subsalsa* was detected three times in both Ayres and Trappe Creeks in July and August below bloom levels (254-6,350 cells*ml^{-1}). *Chloromorum toxicum* was detected in Turville, Bishopville and Marshal Creek.

2011 *Chattonella subsalsa* was again detected three times in both Ayres and Trappe Creeks in July and August below bloom levels (1-3,810 cells*ml^{-1}). *Chloromorum toxicum* was detected Ayres and Trappe Creeks in July and in Bishopville during the fall.

2012 *Chattonella subsalsa* was detected in July in Ayres and Trappe Creeks at low concentrations (633 and 1,899 cells*ml^{-1}respectively).

2013 *Chloromorum toxicum* (*Chattonella sp.)* was detected two times in 2013 at background levels (317 cells*ml^{-1}) in Ayres and Marshall Creeks.

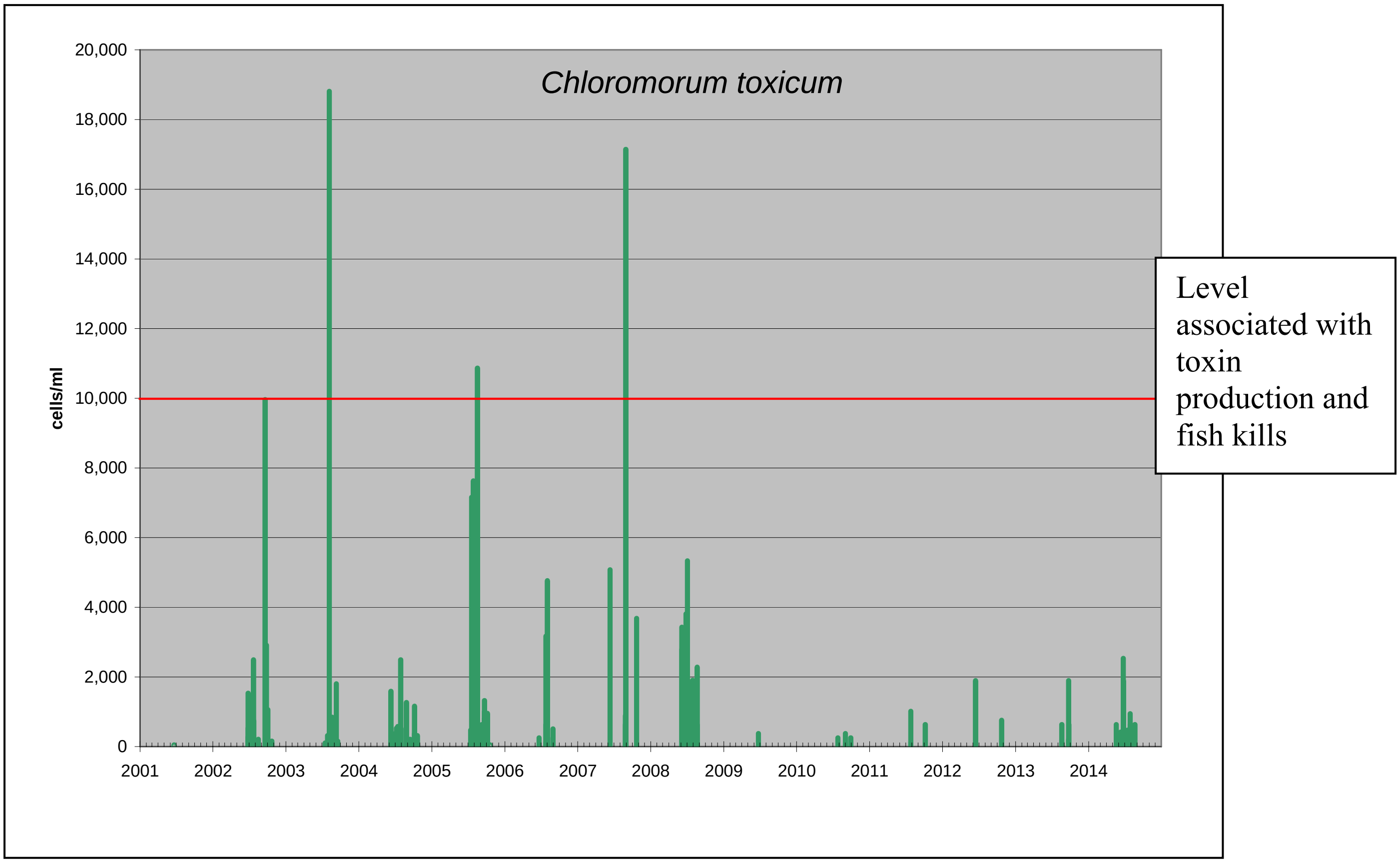

Figure 6.2.3 Occurrence of *Chloromorum toxicum* in the Maryland Coastal Bays between 2001-2014.

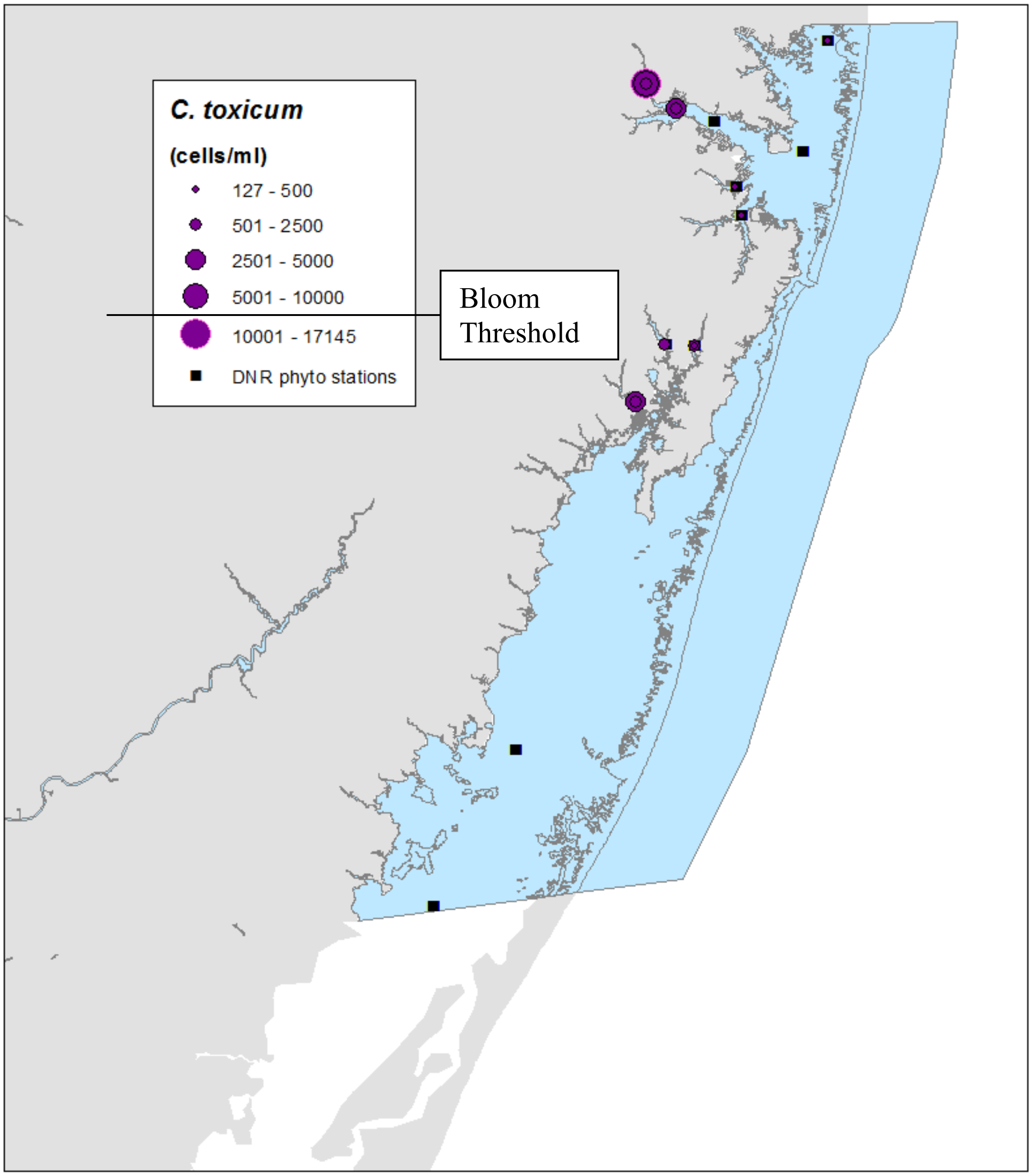

Figure 6.2.4 Distribution of *Chloromorum toxicum* in the Maryland Coastal Bays between 2007-2014.

b. <u>*Heterosigma*</u>

Heterosigma akashiwo has been found on both coasts of the United States (Hargraves and Maranda 2002) and is considered the causative organism involved in offshore fish farm kills in Washington State. Net-penned fish deaths related to *Heterosigma* have been particularly prominent in the northeast Pacific Ocean, notably around Japan. Predictability of blooms has been most related to temperature (warmer season waters >15 degrees C) and moderate salinity (approximately 15 ppt) in the coastal zone (Li and Smayda 2000, Connell and Jacobs 1997). Blooms have been observed to persist as long as stable water stratification persists in the warmer months. An unidentified ichthyotoxin (i.e., fish killing toxin) has been suggested as the causative agent in the mariculture fish kills. No documented effects to humans are evident from such blooms.

2007 *H. akashiwo* was detected in Trappe Creek (TRC0043) and Manklin Creek (MKL0010) in September and October respectively at background levels (1-127 cells*ml^{-1}).

2008 A bloom of *H. akashiwo* was detected in Marshall (MSL0011) in May (41,910 cells*ml^{-1}). Another bloom was detected in Ayres Creek (AYR0017) in June (1270 cells*ml^{-1}). Neither showed evidence of toxic activity.

2009 A bloom of *H. akashiwo* was found in June in Ayres Creek (AYR0017) and Trappe Creek (TRC0043) with concentrations of 25,718 and 1,143 cells*ml^{-1}respectively. *H. akashiwo* was also noted in Marshall Creek at 508 cells*ml^{-1}.

2010 *H. akashiwo* was observed in Trappe Creek (508 cells/ml) and Ayres Creek (635 cells*ml^{-1}) in June and September respectively.

2011 A bloom of *H. akashiwo* was documented in Turville Creek in September of 2011 (8,890 cells*ml^{-1}).

2012 *Heterosigma* was not detected in 2012.

2013 Both Ayres and Marshall Creeks had occurrences of *H. akashiwo* in May (1,400 and 1,600 cells*ml^{-1} respectively). It was also noted in Trappe, Manklin and Turville creeks at levels well below bloom thresholds.

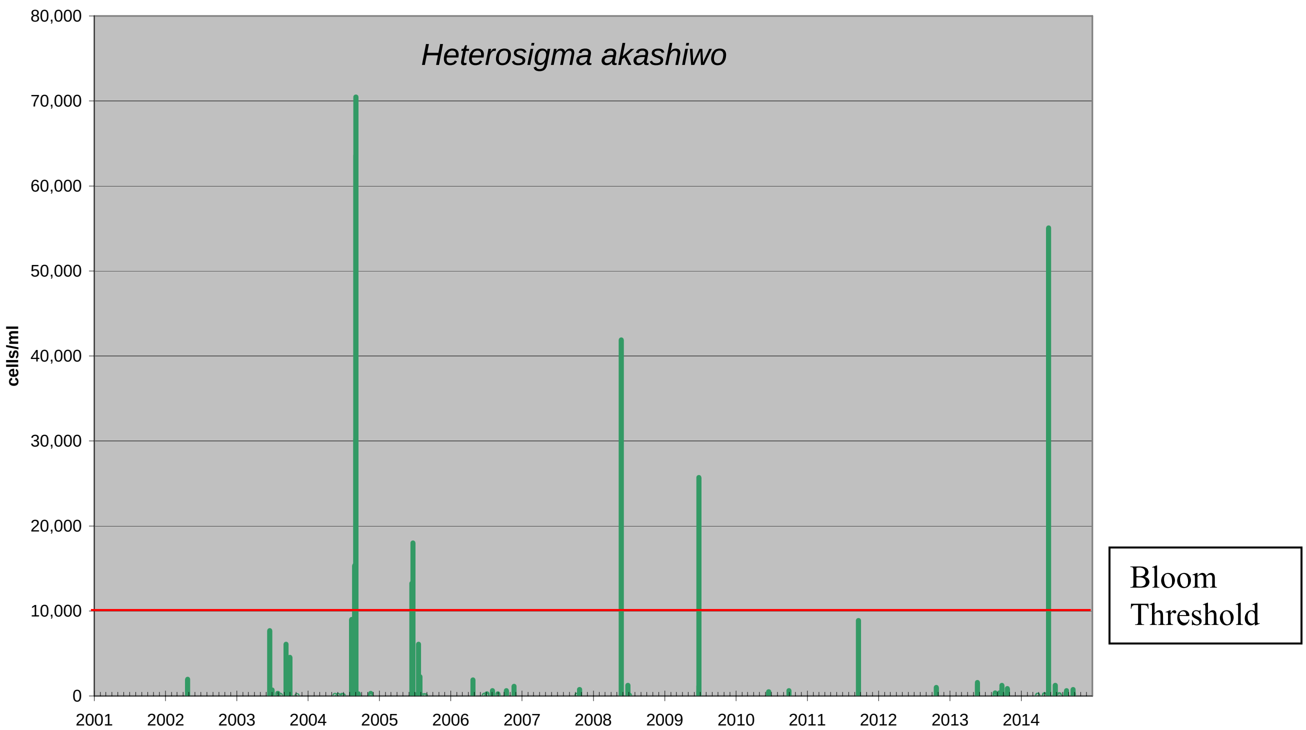

Figure 6.2.5 Occurrence of *Heterosigma akashiwo* in the Maryland Coastal Bays between 2001-2014.

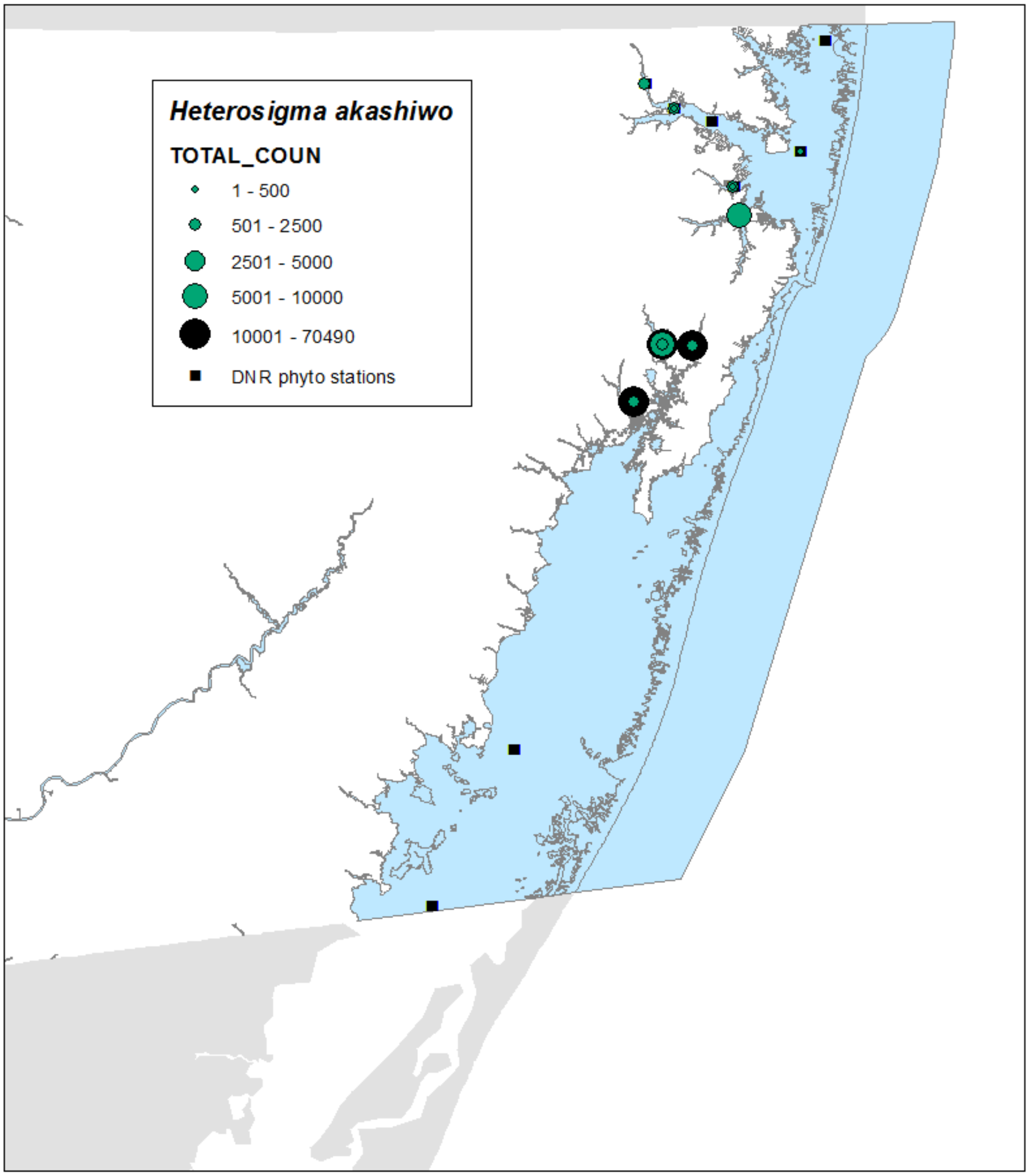

Figure 6.2.6 Distribution of *Heterosigma akashiwo* in the Maryland Coastal Bays 2007-2013.

c. <u>*Fibrocapsa*</u>

Fibrocapsa has had devastating impacts on mariculture operations in Japan. Strains of *F. japonica* collected from the North Sea in Europe have been capable of producing toxin that killed fish in laboratory tank studies. The body tissue of two seals that died in the Wadden Sea of Germany were found to have high levels of the toxin Fibrocapsin. North Sea strains of *F. japonica* grow well under laboratory conditions of 11-25^{o}C, 20-30 ppt salinity, and N/P ratio of 24. No samples were sent for toxin analyses.

2007 Low concentration detected in Ayres Creek in August (254 cells*ml^{-1}). A September bloom >1,000 cells*ml^{-1} was detected in Bishopville Prong.

2008 One bloom level cell count (>1,000 cells*ml^{-1}) was detected in the upper St Martin River during August (1,143 cells*ml^{-1}).

2009 No blooms detected.

2010 Blooms were observed in Bishopville Prong in early August (2,540 cells*ml^{-1}) and below bloom level counts detected in Ayres Creek in September (635 cells*ml^{-1}).

2011 No blooms detected

2012 No blooms detected

2013 Highest bloom recorded was detected in Marshall Creek in January (5,000 cells*ml^{-1}).

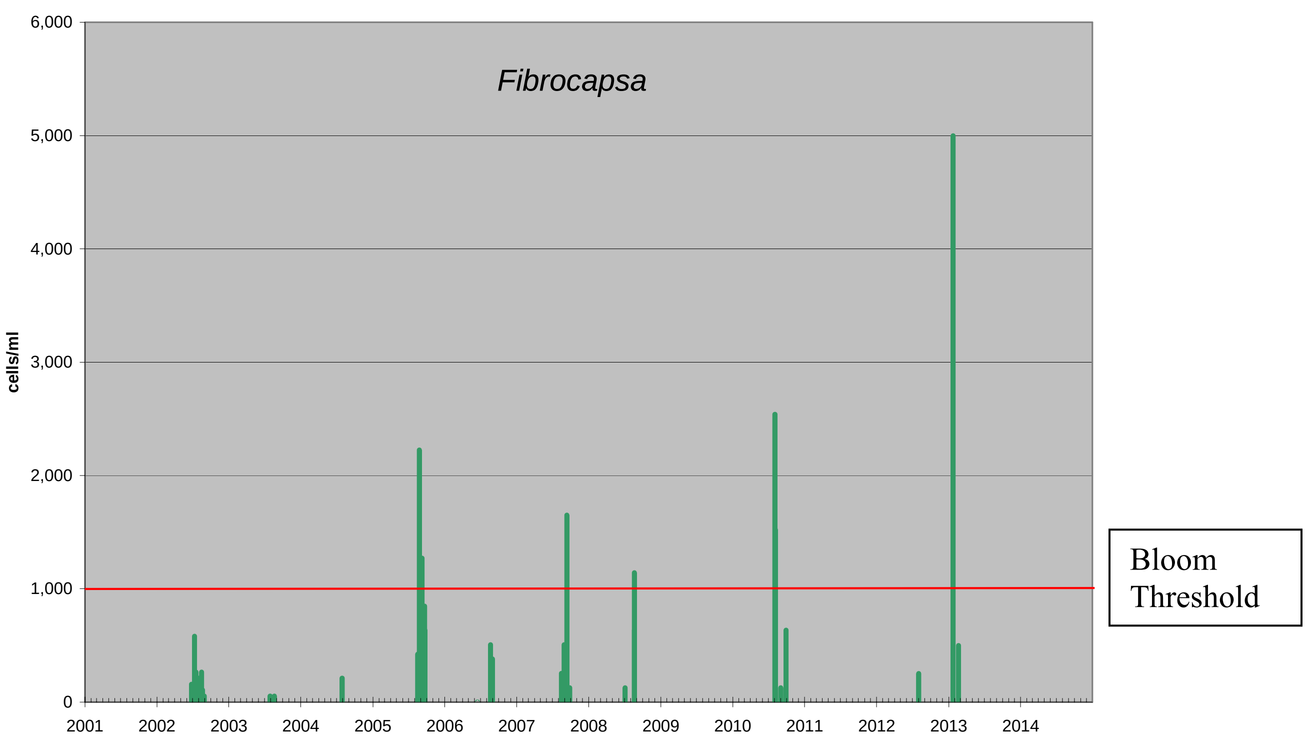

Figure 6.2.7 Occurrence of *Fibrocapsa* in the Maryland Coastal Bays between 2001-2014.

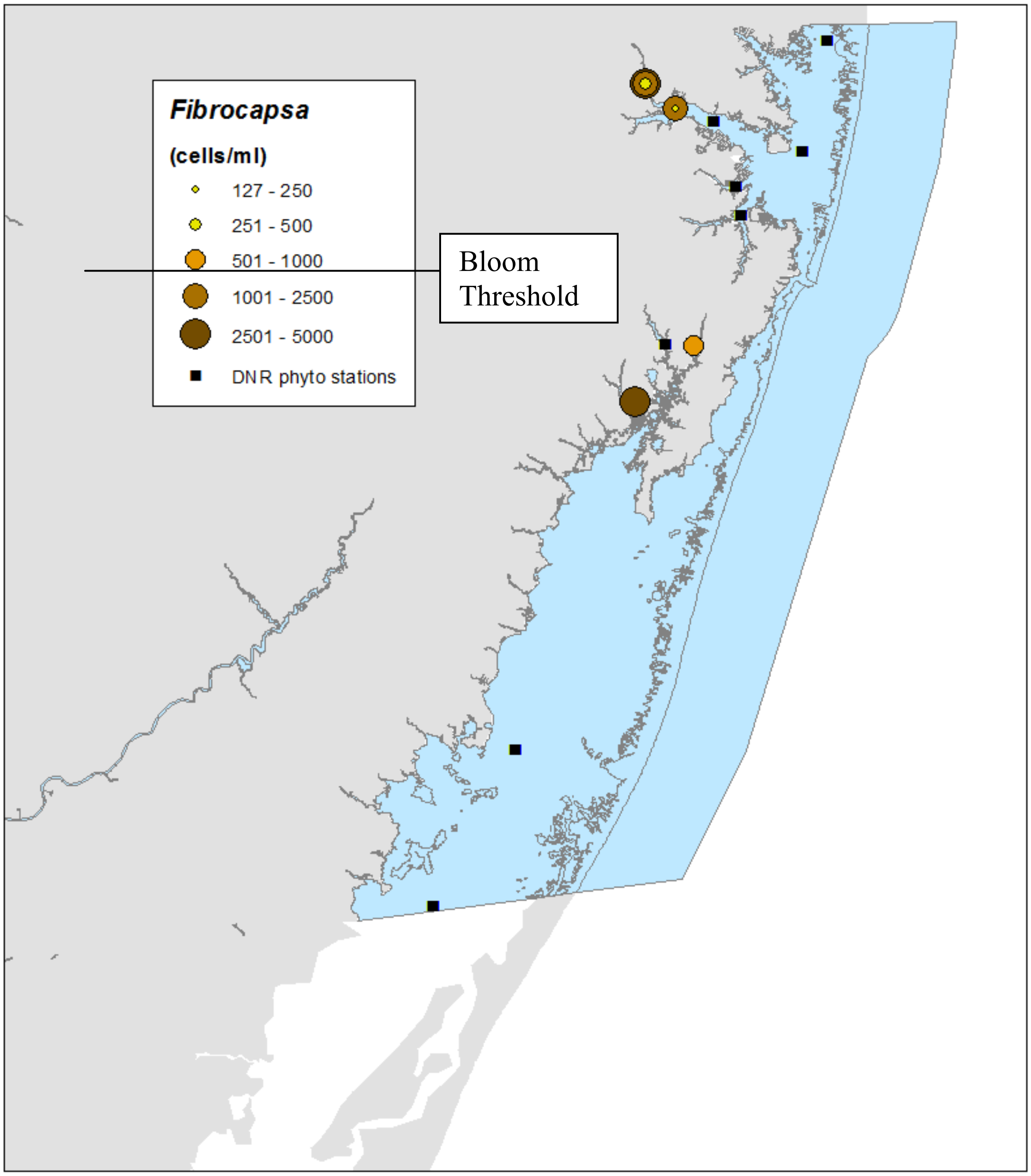

Figure 6.2.8 Distribution of *Fibrocapsa* in the Maryland Coastal Bays (2007-2014).

III. <u>*Pfiesteria: P. piscidia and P. shumwayae*</u>

There are two species of *Pfiesteria*, *Pfiesteria piscicida* and *Pfiesteria shumwayae*, both of which are potentially toxic to fish and people. *Pfiesteria* has been shown to have a highly complex life-cycle with more than 24 reported forms that live in either the bay sediment or water.

Pfiesteria was first detected with targeted sampling in the Coastal Bays of Maryland beginning in 1998. Water and sediment surveys have been conducted in the Coastal Bays using Polymerase Chain Reaction (PCR) techniques to detect these potentially harmful species. Rapid response efforts by MDE and DNR have examined fish kills annually since 2000 occasionally detecting *Pfiesteria* species at the events. Bioassays, however, have all been negative for signs of toxicity. No toxic Pfiesteria has ever been detected in Maryland's Coastal Bays. The presence of *Pfiesteria* has historically been in the Newport Bay system (Ayres, Trappe, Marshall and Newport Creeks).

2007-2012 No *Pfiesteria* cells were observed.

2013 Pi*esteria*-like species were detected during December in the upper St Martin River (low concentration of 506 cells*ml^{-1}). .

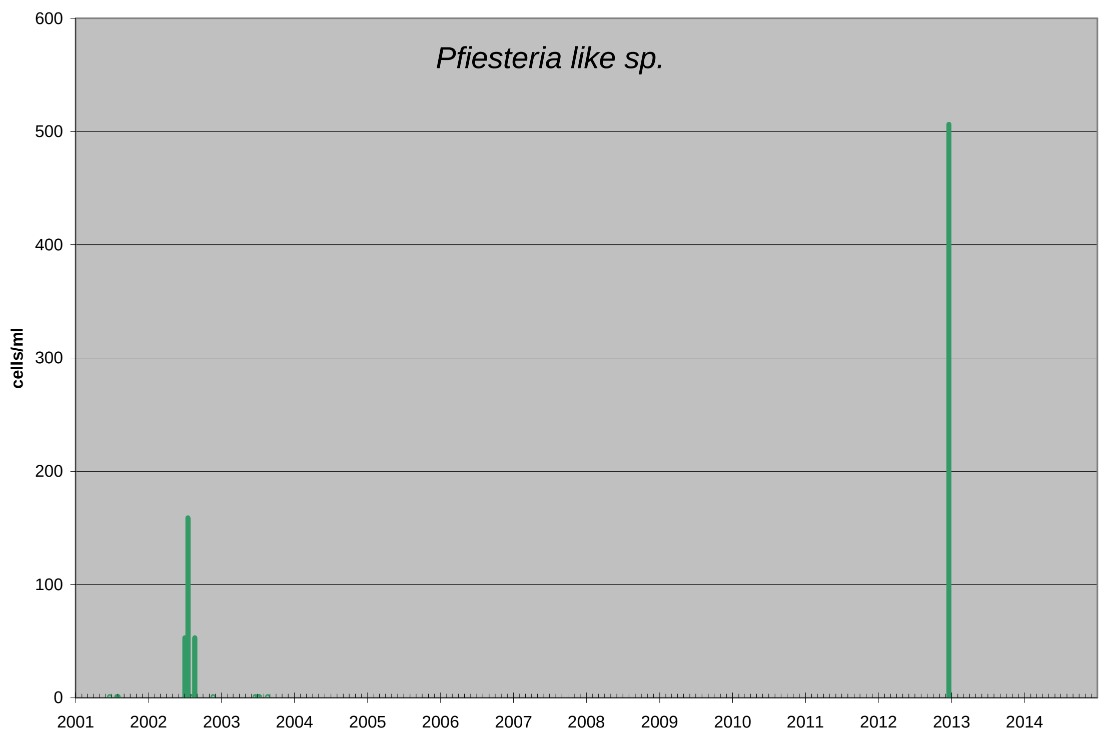

Figure 6.2.9 Occurrence of *Pfiesteria*-like species in the Maryland Coastal Bays between 2001-2014.

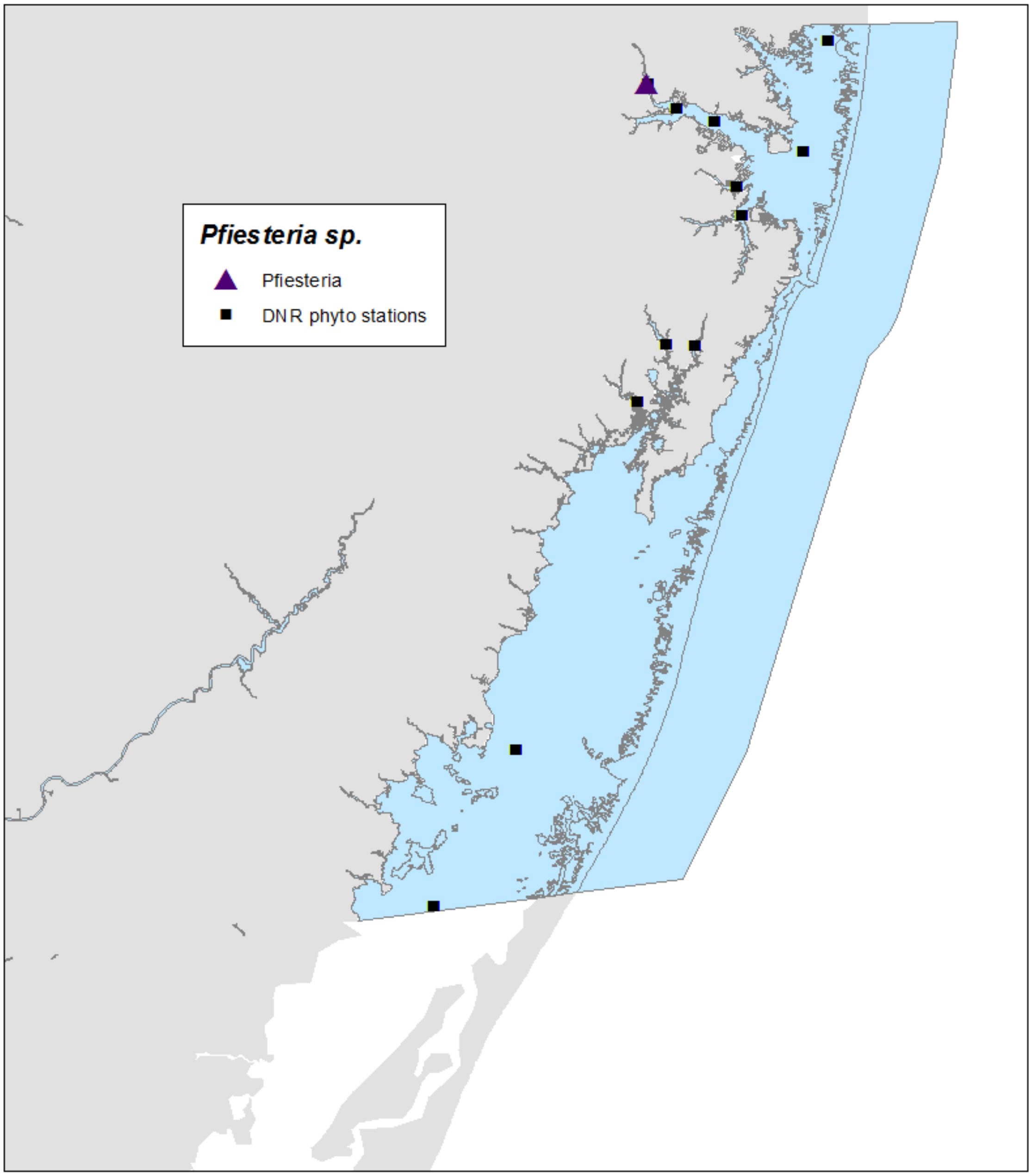

Figure 6.2.10 Distribution of *Pfiestera* in the Maryland Coastal Bays between 2007-2014.

IV. *Prorocentrum*

Prorocentrum blooms have been linked to widespread harmful ecosystem impacts including: anoxic and hypoxic events, finfish kills, aquaculture shellfish kills, submerged aquatic vegetation losses, and toxicity bioassays. Such events in this region are typically related to the planktonic species *Prorocentrum minimum*. In the Coastal Bays, blooms have occurred in April and May in mid-salinity waters (upper parts of creeks and rivers). This species is considered potentially toxic to humans with rare cases of associated shellfish poisoning worldwide. No such cases related to *P. minimum* have been reported from Maryland waters although isolates from the Choptank River (Chesapeake Bay watershed) indicated toxicity to shellfish larvae in laboratory testing. High biomass blooms have also been responsible for low dissolved oxygen events leading to fish kills in Chesapeake Bay embayments and an extended bloom in 2000 is suspected in declines of SAV in the mid-Chesapeake Bay region for 2001.

Impacts on bay organisms have been identified at concentrations as low as 3,000 cells*ml^{-1} (EPA 2003) providing a threshold for the tracking and assessment of blooms. Threshold exceedances were recorded only once during 2001 and 2002 in the St. Martin River. Impacts of high density blooms of *Prorocentrum* are most likely when blooms exceed 10,000 cells*ml^{-1}.

2007 Significant blooms were observed January through March in the Upper St Martins, Bishopville Prong, Marshall and Turville Creeks. Blooms were observed again in May in Bishopville Prong.

2008 No significant blooms observed.

2009 Bloom were detected in April and May in the upper St Martin River (19,685 cells*ml^{-1}), Bishopville Prong (~65,000 cells*ml^{-1}) and Turville Creek (>17,000 cells*ml^{-1}).

2010 No significant blooms observed. Cells counts above 3,000 cells*ml^{-1} (threshold which will discolor water) were seen in Marshall, Trappe and Newport Creeks.

2011 No blooms detected.

2012 A bloom was observed in January in the St Martin River, Bishopville Prong, due to a warm spell (19,177 cells*ml^{-1}). The rest of the year no blooms were detected.

2013 Significant blooms (>10,000 cells*ml^{-1}) were observed in Marshall Creek, Newport Creek and Trappe Creek in March and April.

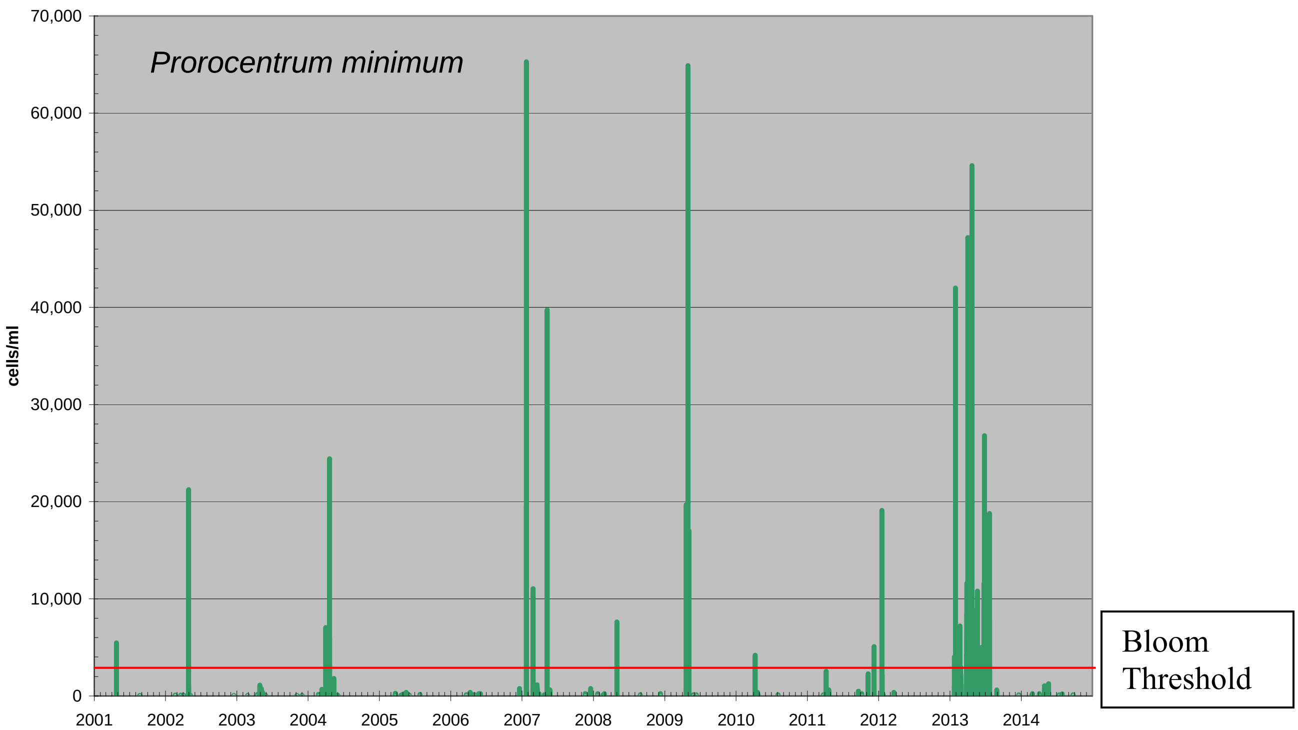

Figure 6.2.11 Occurrence of *Prorocentrum minimum* in the Maryland Coastal Bays 2001-2014.

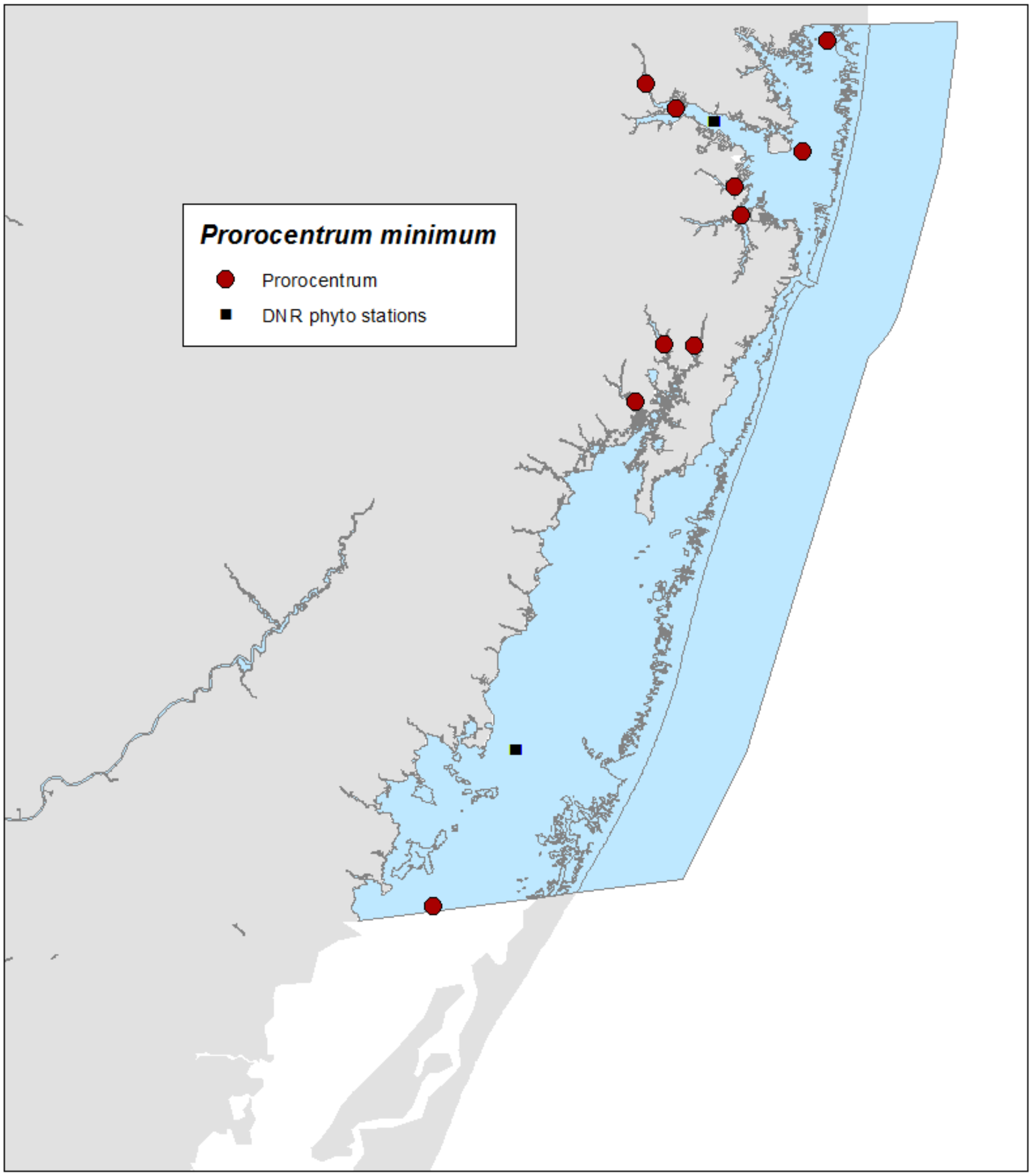

Figure 6.2.12 Distrbution of *Prorocoentrum minimum* in the Maryland Coastal Bays, 2007-2014.

V. *Dinophysis*

Dinophysis acuminata has been the most commonly encountered representative of this genus in Maryland's Coastal Bays. The genus *Dinophysis* is represented in Chesapeake Bay by five species (*D. acuminata, D. acuta, D. fortii, D. caudata* and *D. norvegica*) and are all known to produce okadaic acid or other toxins causing Diarrhetic Shellfish Poisoning (DSP) (Marshall 1996). DSP has occurred in humans consuming the contaminated shellfish resulting in symptoms that include intestinal discomfort, abdominal pain, nausea, headache, chills and vomiting. No cases of DSP have been reported in Maryland.

Management actions in the countries of Italy, Norway and Denmark to protect human health against DSP includes intensified monitoring of shellfish harvest waters, toxin testing of the shellfish and application of restrictions or closures of the fisheries. Thresholds of 500-1,200 cells*L^{-1} are used by managers in these countries to initiate temporary closures or intensified monitoring; toxin test results ultimately determine the extent of actions necessary (Anderson et al. 2001). Europe and Japan appear to be the most highly affected areas for cases of DSP, however, outbreaks in North America have been confirmed in Eastern Canada during 1990 and 1992. Okadaic acid was found in association with a *D. acuminata* bloom in 2002 on the Potomac River, however, levels were well below FDA levels for seafood safety. Despite thousands of documented cases of DSP worldwide since 1960, there are no reported fatalities associated with the illness.

A threshold 20x the minimum used in Europe (i.e., 0.5 x 20 = 10 cells*ml^{-1} threshold) has been implemented as a tracking indicator for this species. *Dinophysis* has been observed above threshold concentrations in Assawoman Bay, Isle of Wight and St. Martin River. Recently toxicity has been shown in the Coastal Bays and accumulation in shellfish. No shellfish in harvestable waters have shown levels above FDA thresholds and no closures have been implemented.

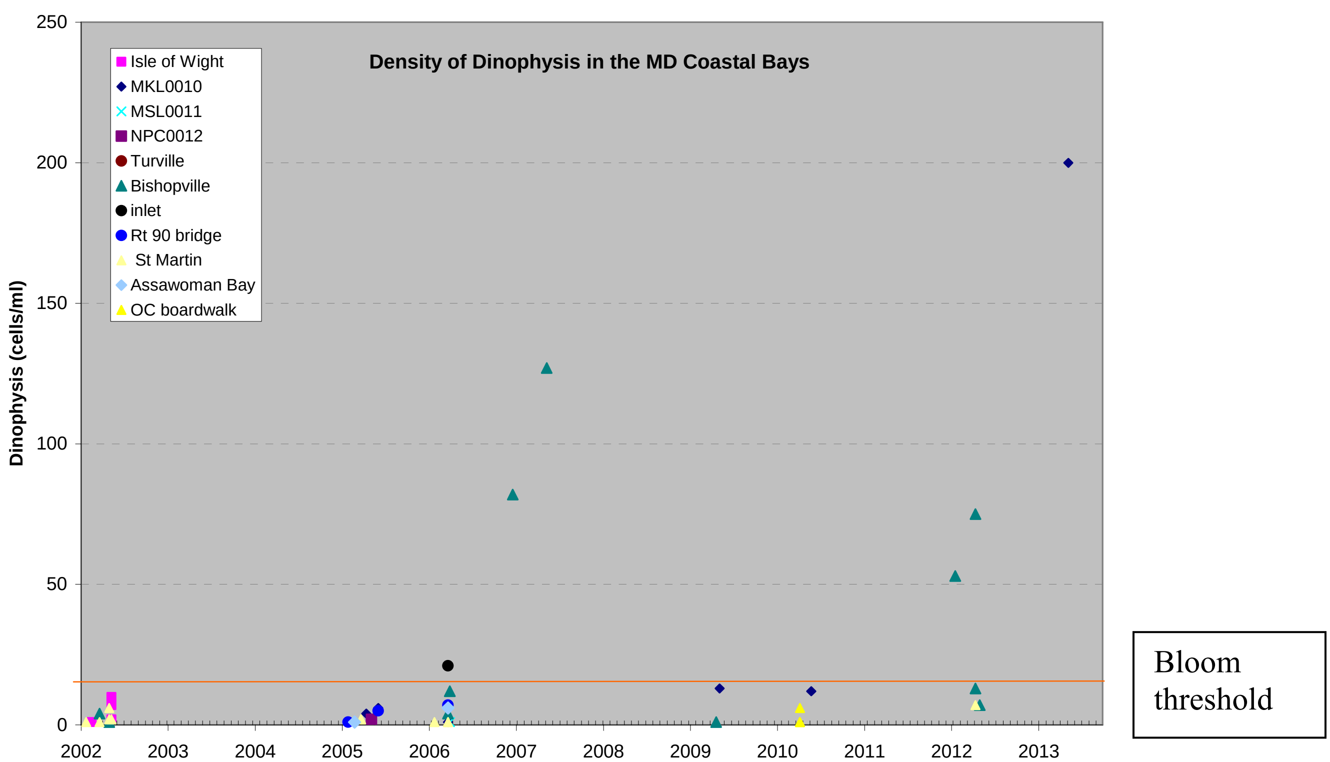

Figure 6.2.13 Occurrence of *Dinophysis sp.* at specific sites in the Maryland Coastal Bay from 2002-2014. Bloom threshold is 10 cells*ml^{-1}.

2007 Blooms of *Dinophysis* detected in May at Bishopville site (127 cells*ml^{-1}).

2008 No *Dinophysis* detected

2009 A bloom of *Dinophysis* was detected in Manklin and cells were present in Turville Creeks during May at 13 and 1 cells*ml^{-1} respectively.

2010 A bloom sample from Manklin creek in May (12 cells*ml^{-1}). A water sample was sent to FDA and confirmed DSP toxin presence in MD for the first time.

2011 No bloom level counts detected in routine samples; however, bloom levels were found in Turville Creek, Isle of Wight and Assawoman Bays during special study. Bloom in Bishopville, collected oyster for toxin analyses by FDA revealed toxin in shellfish. This is a no shellfish area.

2012 A bloom was observed in Bishopville Prong (53 and 75 cells*ml^{-1}). Mussels were collected for toxin analyses by U.S. Food and Drug Administration that revealed toxin above guidance levels. This is a no shellfish area. SPATT, solid phase adsorption toxin tracker, samplers indicated even higher toxin possible in Turville Creek (highest count 3 cells*ml^{-1}). Toxin also present in Manklin (count only 1 cells*ml^{-1}).

2013 Routine monitoring saw one bloom of *Dinophysis* in Manklin Creek during May (200 cells*ml^{-1}). An intensive cage study was also conducted in 2013 to compare the uptake of 'dinotoxins' in 4 bivalve species (scallops, mussels, oysters and clams) and the SPAT passive samplers. The study concluded that scallops take up the toxin most, followed by Clams and then oyster/mussels.

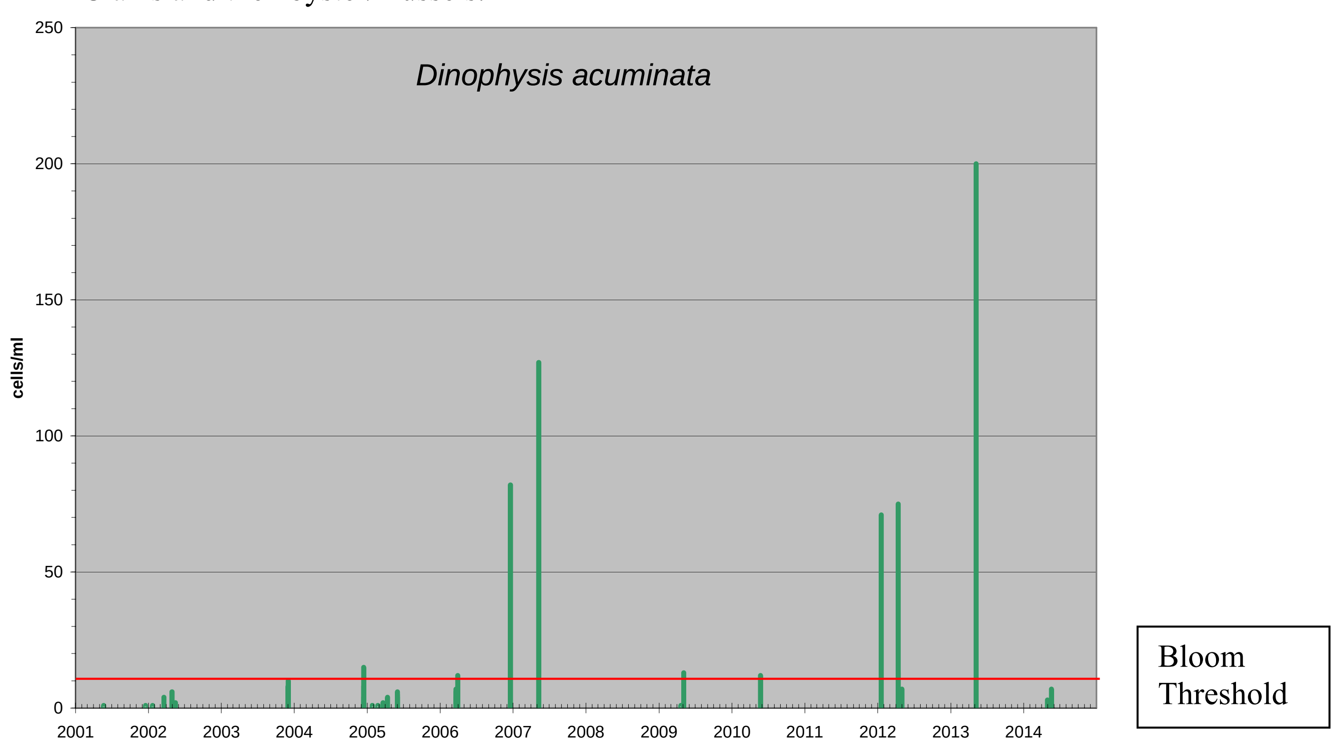

Figure 6.2.13b Occurrence of *Dinophysis sp.* in the Maryland Coastal Bays, 2001-2014.

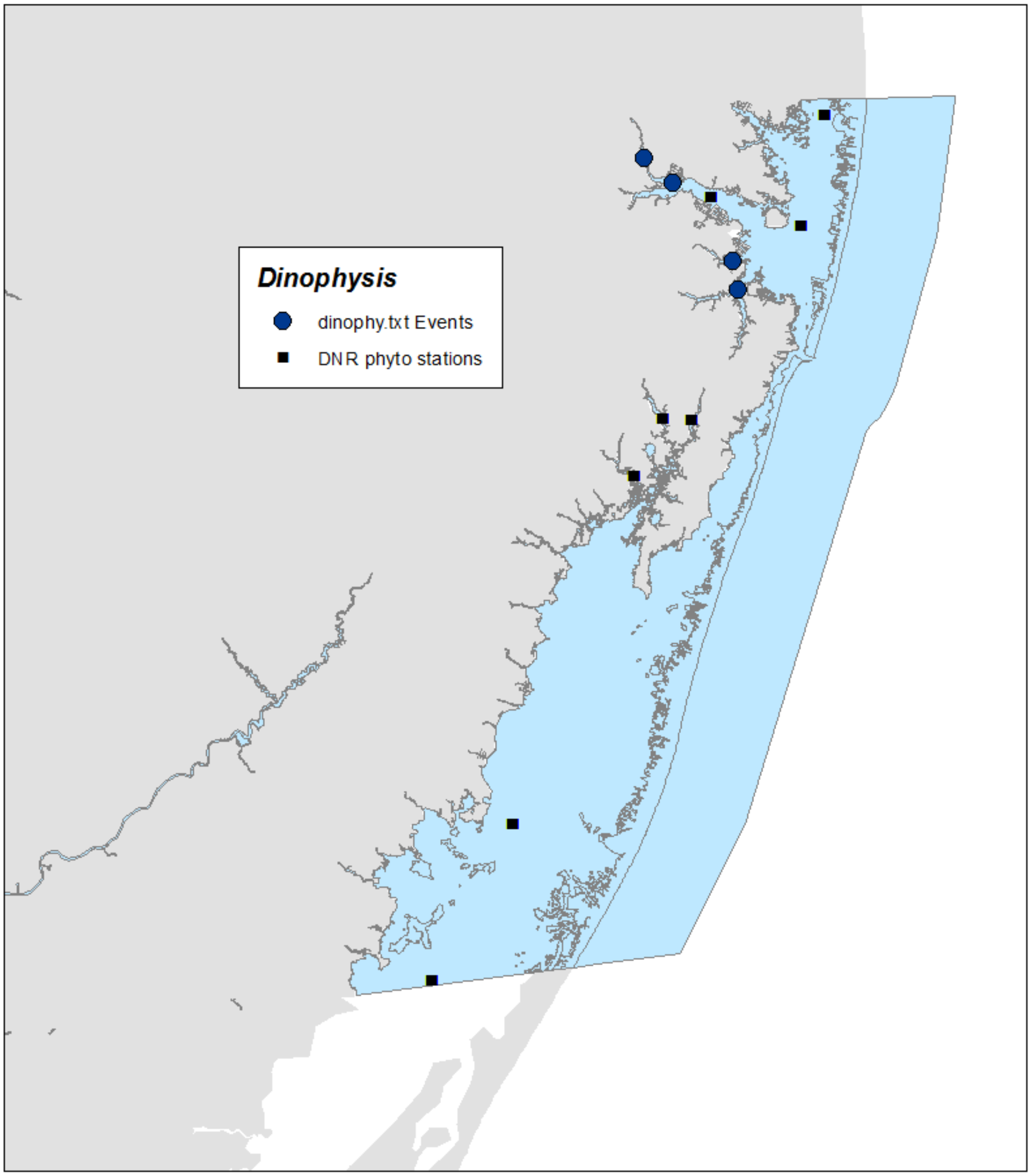

Figure 6.2.14 Distribution of *Dinophysis sp.* in the Maryland Coastal Bays (2007-2014).

VI. *Pseudo-nitzschia*

Diatoms in the genus *Pseudo-nitzschia* are recognized worldwide as potential producers of the toxin domoic acid (DA). Shellfish feeding on toxic *Pseudo-nitzschia* can accumulate domoic acid. Humans consuming the contaminated shellfish may subsequently experience Amnesic Shellfish Poisoning (ASP). Symptoms of ASP include vomiting, confusion, memory loss, coma or death. ASP was first identified on the east coast of North America at Prince Edward Island, Canada, in 1987. Despite a recall of all bivalve products from the Prince Edward Island region, the outbreak resulted in 107 illnesses that included 13 fatalities. In 1995, a shellfish closure occurred due to elevated levels of DA. Recent illnesses have only occurred from recreational harvests that have disregarded the shellfish closures.

In other countries: *Pseudo-nitzschia* cell densities of 200 cells*ml^{-1} of *P. seriata* are used in Denmark and 5-10 cells*ml^{-1} in New Zealand to trigger toxin testing of shellfish meats (Anderson et al. 2001). In New Zealand, the shellfish industry conducts voluntary closures of a fishery where cell densities measure > 5 x 10^5 cells*L^{-1} (Anderson et al. 2001). Canada has indicated detectable levels of DA in the shellfish at levels of at least 1,000 cells*ml^{-1} (Anderson et al. 2001).

In Maryland, low levels of domoic acid have been detected by Thessen and Stoeker 2008. However, blooms do not typically have high concentrations nor do they last

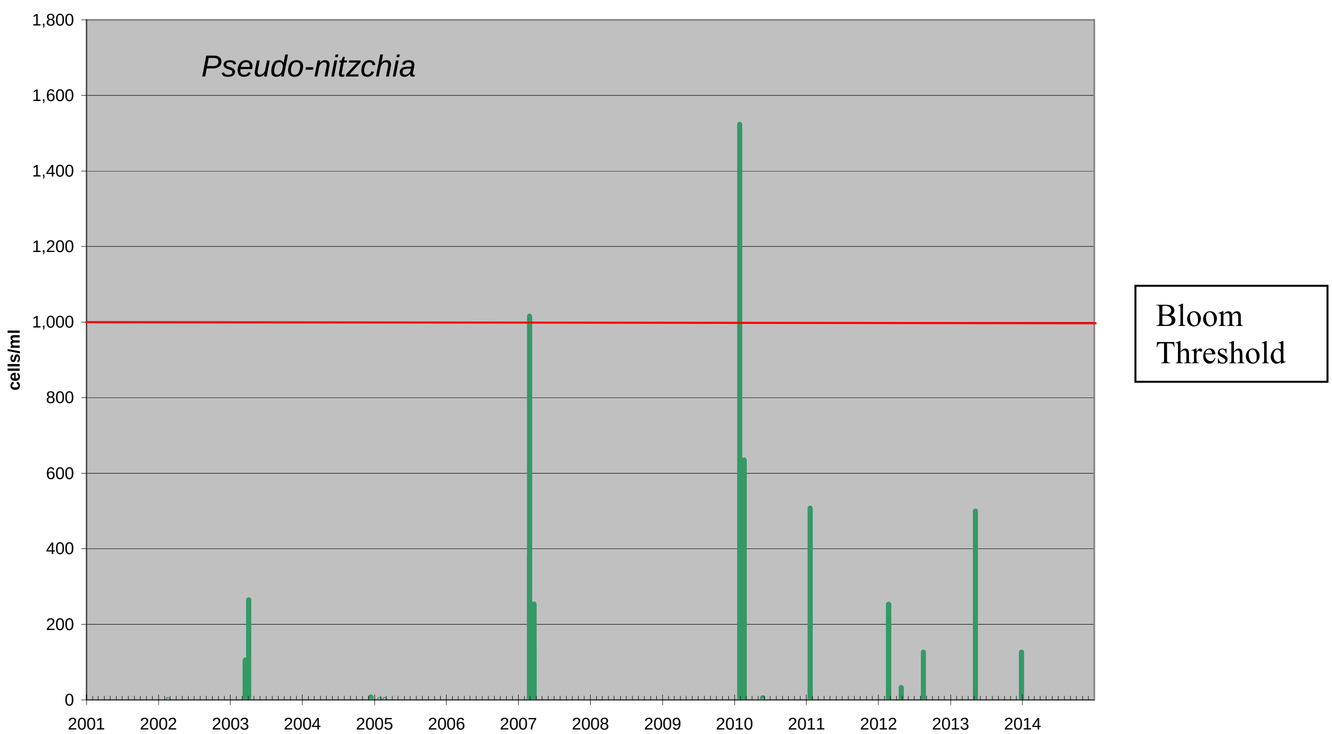

Figure 6.2.15 Occurrence of *Pseudo-nitzschia sp.* in the Maryland Coastal Bays (2001-2014). Bloom threshold is 1,000 cells*ml^{-1}.

2007 Bloom observed in Manklin Creek in February (1,016 cells*ml^{-1}) and also observed in Turville Creek and Isle of Wight Bay near the Rt. 90 bridge.

2008 No significant blooms observed.

2009 No significant blooms observed.

2010 Bloom observed (1,524 cells*ml^{-1}) during January in Isle of Wight Bay near the Rt. 90 bridge and lower counts observed in Turville and Manklin Creeks.

2011 No significant blooms observed.

2012 No significant blooms observed.

2013 No significant blooms observed. Present below bloom levels in Manklin Creek.

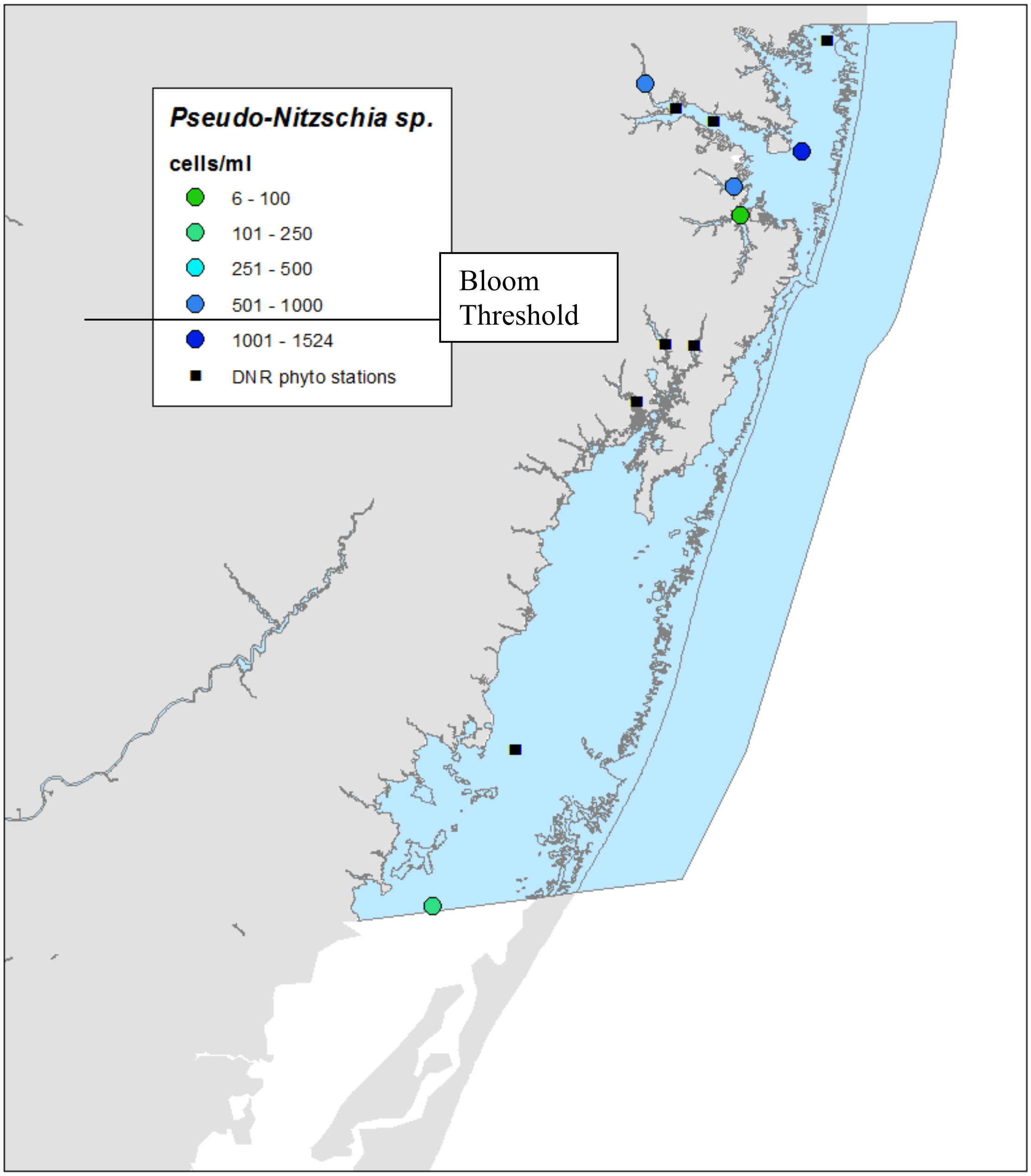

Figure 6.2.16 Distribution of *Pseudo-nitzschia sp.* in the Maryland Coastal Bays between 2007-2014.

<u>VII. *Amphidinium*</u>

The algae *Amphidinium operculatum* is an epi-benthic dinoflagellate was first found in Newport

Creek in October 1999 in very small numbers. This unusual organism was detected in a water sample through centrifuging 15 ml of the sample to look at another species. *Amphidinium* has been linked with ciguatera toxins in subtropical and tropical habitats. They are also known to produce several polyketides known as amphidinins. There is no evidence of toxicity for this species in the Coastal Bays.

Four occurrences of *Amphidinium spp*. were detected between 2007 and 2013 in water samples from the Coastal Bays. In March of 2007 in Turville Creek (TUV0011) and in November of 2009 in Ayres Creek (AYR0017) and last the species *A. glaucum* was observed in December of 2012 in Trappe Creek (TRC0043). Counts ranged from 1 cell/ml in Ayres Creek to 253 cells*ml^{-1} in Trappe Creek. No cell threshold. Better analyses of the benthic microphytobenthos community may reveal more of this genus.

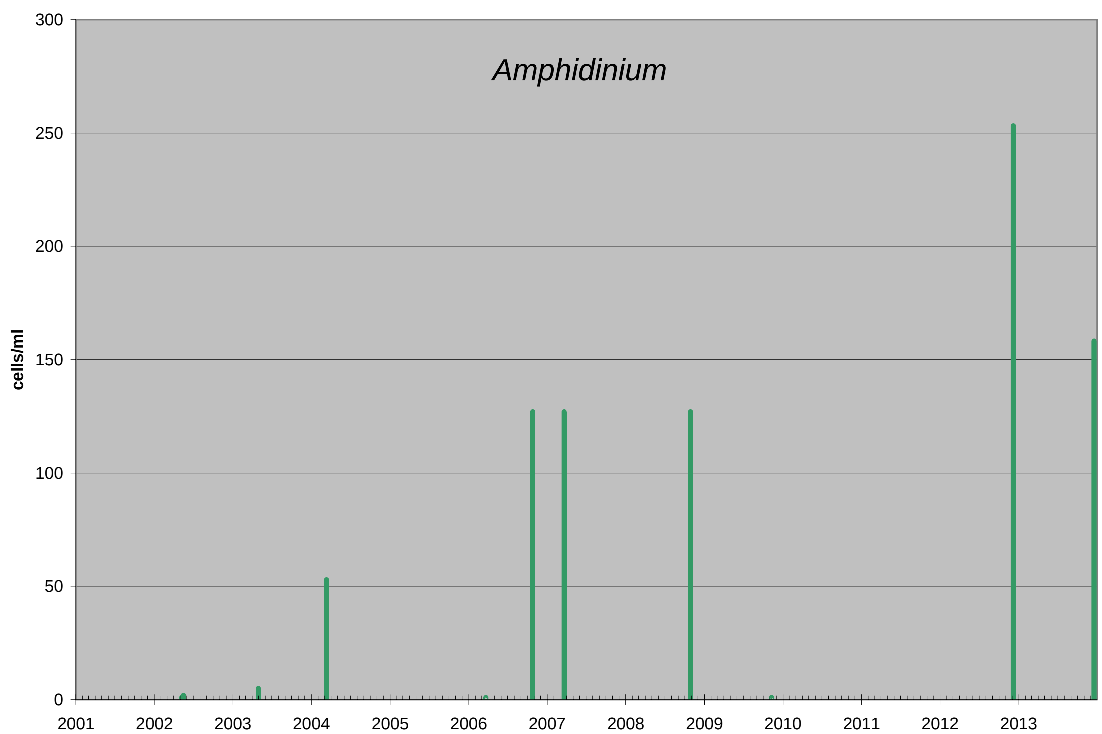

Figure 6.2.17 Occurrence of *Amphidinium sp.*in the Maryland Coastal Bays, 2001-2014.

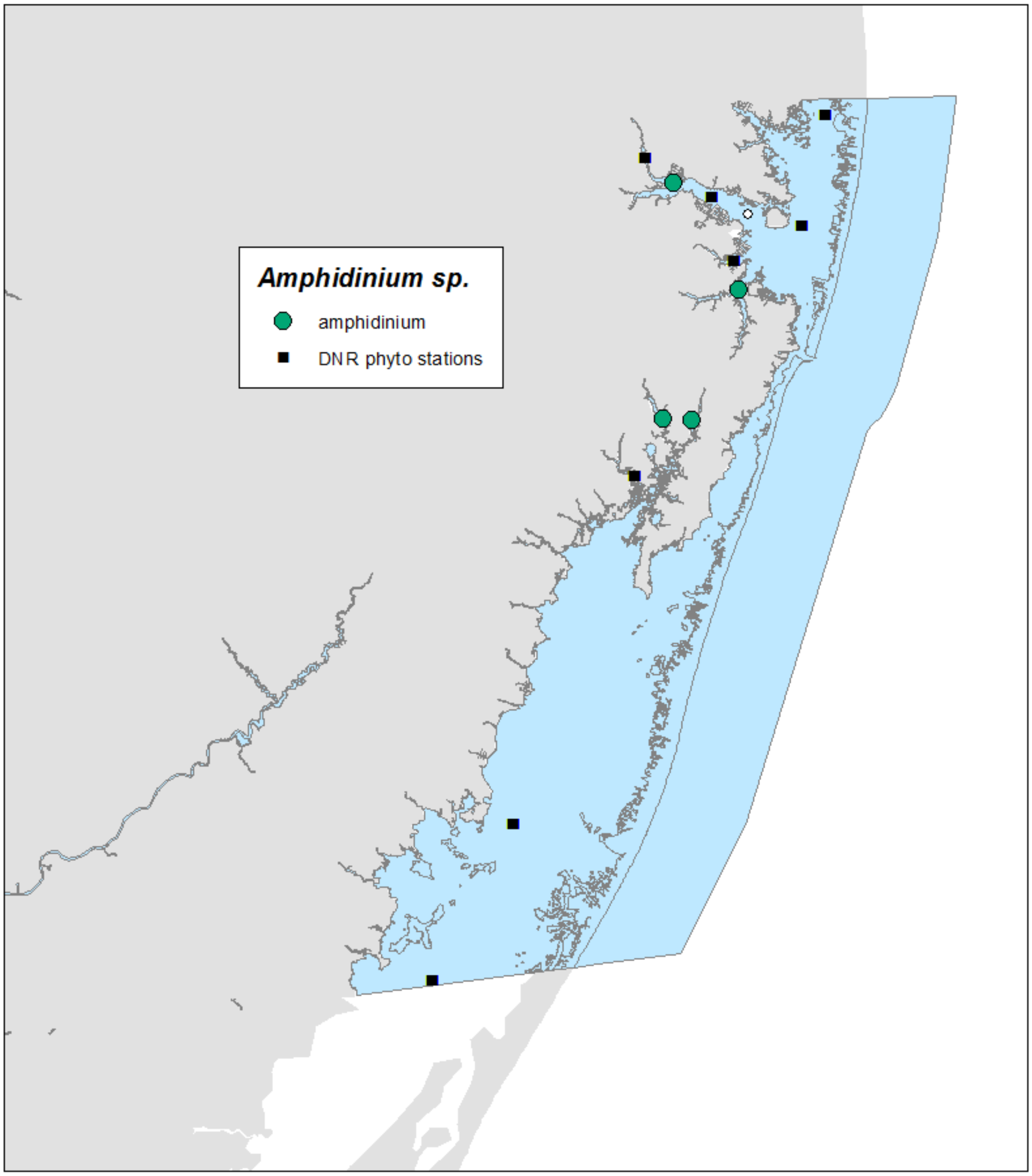

Figure 6.2.18 Distribution of *Amphidinium sp.* in the Maryland Coastal Bays, 2007-2014.

<u>VIII. *Karlodinium micrum*</u>
Karlodinium micrum may cause water to become discolored a reddish-brown and form Mahogany Tides. Mahogany tides may also severely reduce the amount of oxygen available to living resources at localized bloom sites. In large numbers, *Karlodinium micrum* will give the water a coffee color. *Prorocentrum minimum* tends to bloom earlier in the spring than *K. micrum* (late spring and early summer) although both species may occasionally be found blooming throughout the year on a local scale.

Karlodinium micrum is increasingly recognized for its ichthyotoxic effects in estuarine waters. Threshold levels for impacts on fish are considered 10,000 to 30,000 cells*ml^{-1}. *Karlodinium* *micrum*, is synonymous with *Gyrodinium galatheanum* Braarud and *Gymnodinium micrum*, and historically reported as *Gyrodinium estuariale* in Maryland. Recent work by Deeds et al. (2002) has demonstrated that Maryland isolates of the dinoflagellate from Chesapeake Bay produced toxins with hemolytic, cytotoxic and ichthyotoxic properties. Testing has not yet been conducted on samples from the Coastal Bays. Initial studies indicate *K. micrum* may produce sufficient toxin to result in fish mortality in the field at cell densities of 10,000 to 30,000 cells*ml^{-1} and above (Deeds et al. 2002, Goshorn et al. 2003). No human health effects have been associated with blooms of *K. micrum*.

2007 No bloom levels occurred.

2008 No bloom levels were detected.

2009 Blooms were found in Bishopville Prong in May (15,367 cells*ml^{-1}) and in the upper St Martin River in June (27,432 cells*ml^{-1}).

2010 No bloom levels were detected.

2011 Significant bloom during October in Bishopville Prong (58,801 cells*ml^{-1}) followed by lower abundance in November.

2012 No bloom levels were detected.

2013 No bloom levels were detected.

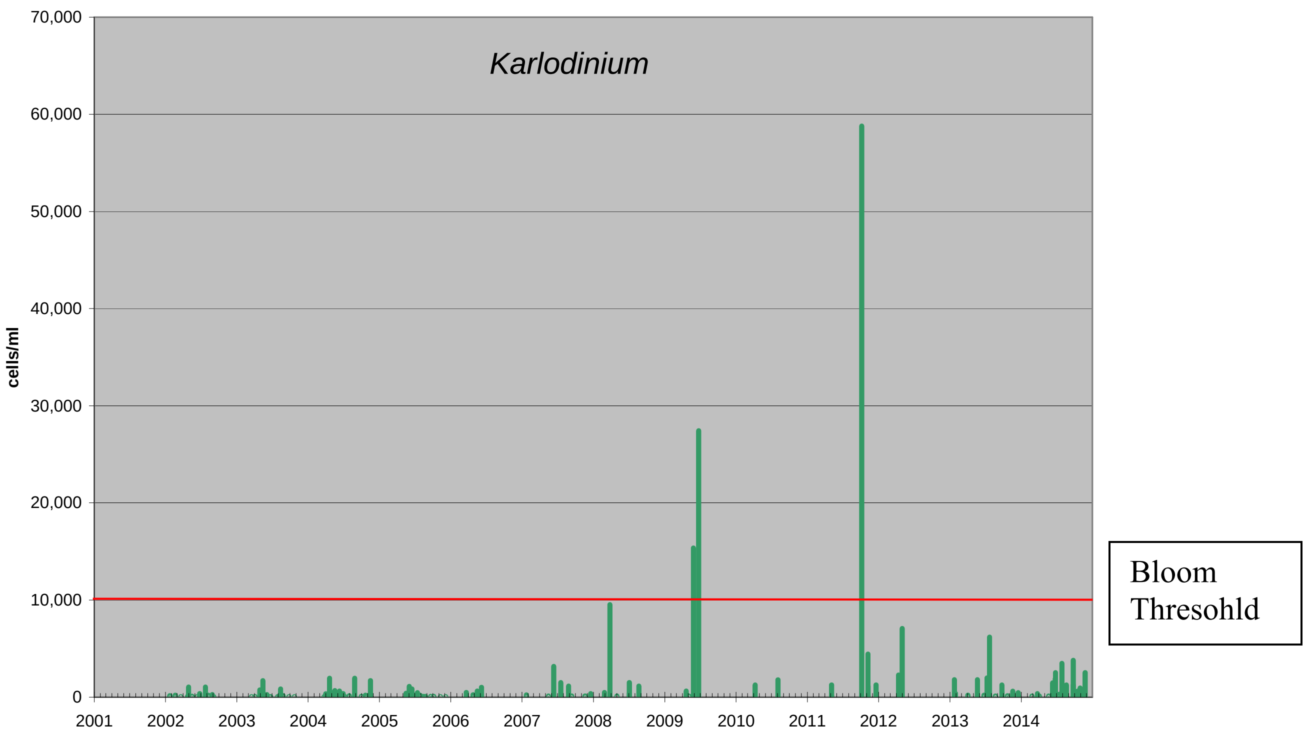

Figure 6.2.19 Occurrence of *Karlodinium veneficum* in the Maryland Coastal Bays between 2001-2014. Bloom threshold is 10,000 cells*ml^{-1}.

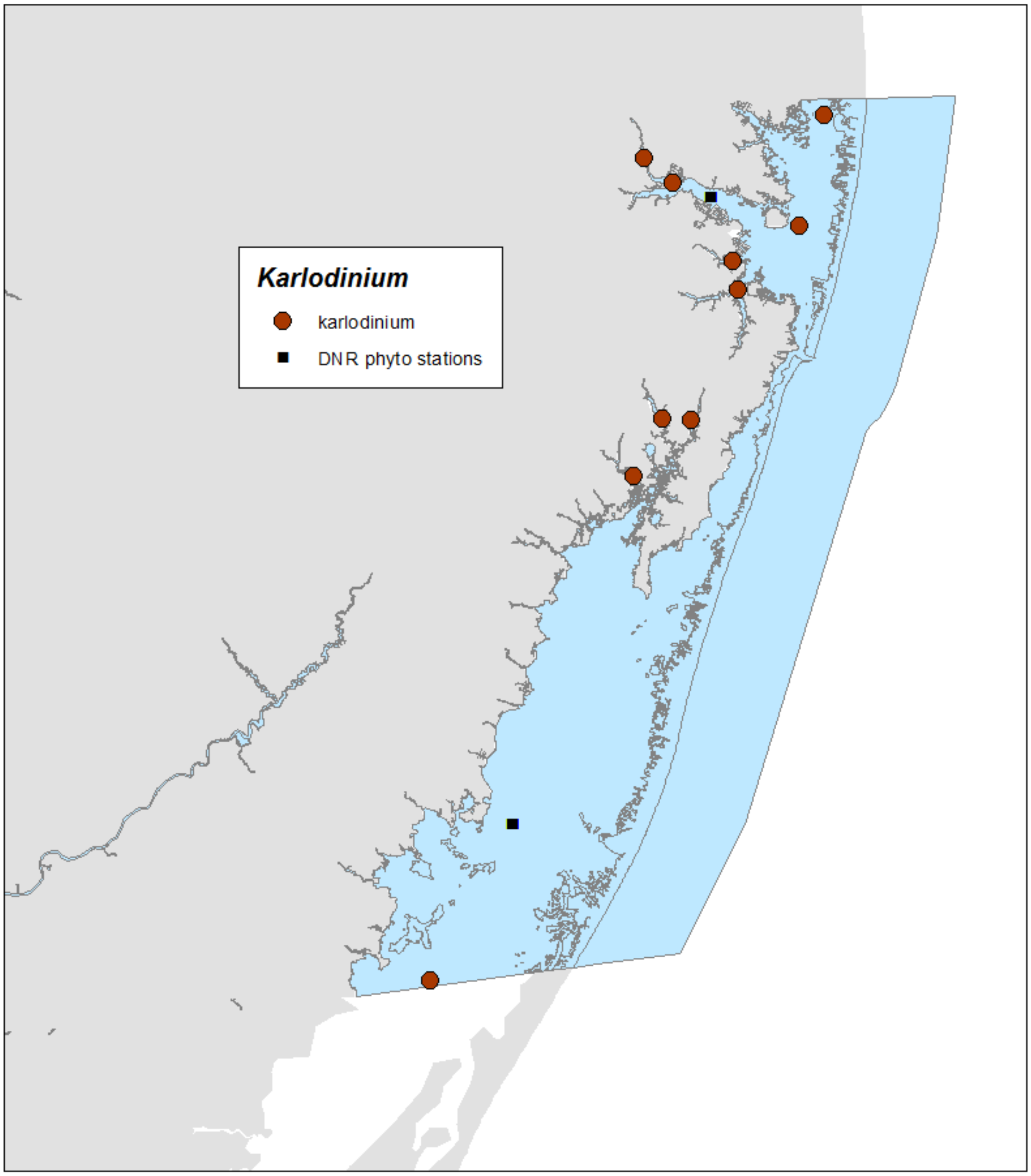

Figure 6.2.20 Distribution of *Karlodinium veneficum* in the Maryland Coastal Bays (2007-2014).

IX. Cyanobacteria

The winter of 2009 had blooms of cyanobacteria species in Trappe Creek including *Microcystis aeruginosa*, *Anabaena*, *Aphanizomenon* and *Oscillatoria*. Peaks in cyanobacteria abundance in 2009 is likely due to lower salinities resulting from higher rainfall that year (Figure 6.2.21). High levels of pico-cyanobacteria have been observed since a new microscope was implemented that allowed a greater magnification for small phytoplankton. Pico-cyanobacteria were very abundant in 2007 and 2008 (moderate in 2009-2012).

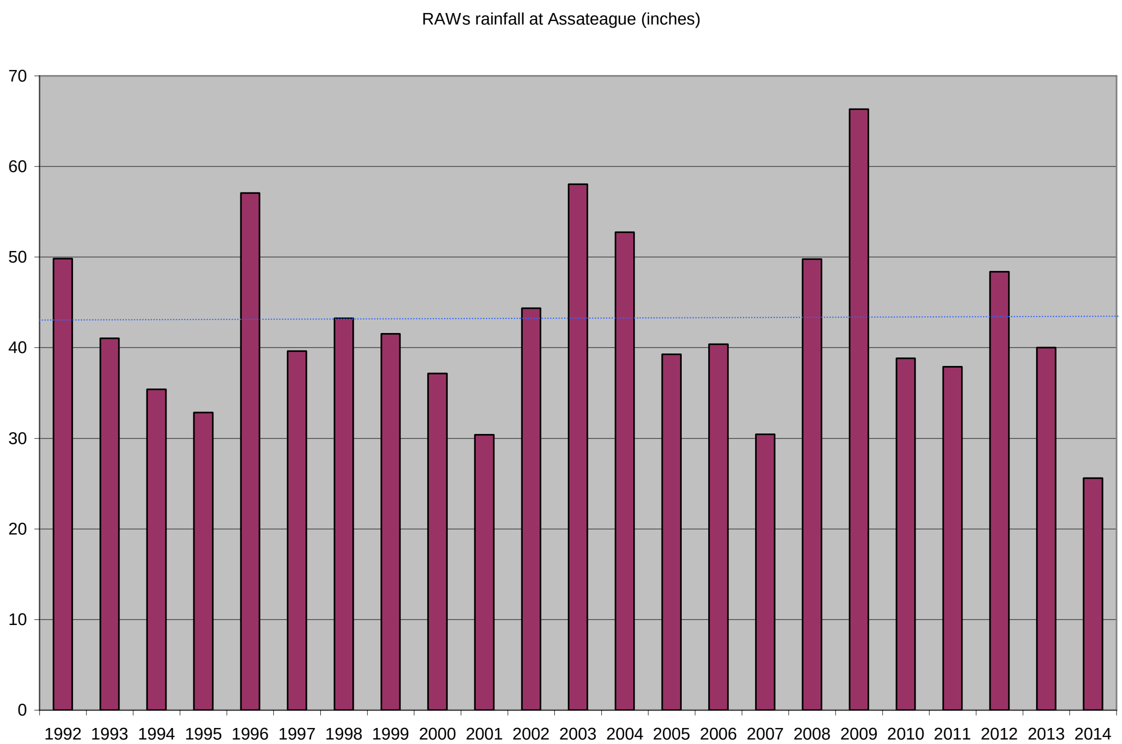

Figure 6.2.21 Rainfall record at Asssateague Island Remote Weather Station (RAWS) 1992-2014.

a. *Microcystis aeruginosa*

Microcystis is rarely seen in the Coastal Bays but was observed at significant bloom levels in 2009 in Trappe Creek. Toxic cyanophytes have been shown to affect a broad range of living resources. *Microcystis aeruginosa* is not unlike other possibly toxic phytoplankton species in that there may be a gradient of strain-related toxicity. Studies have shown negative effects on feeding to zooplankton by toxic and non-toxic *M. aeruginosa*. Fish kills have been attributed to cyanobacterial blooms and sub-lethal effects on fish can include reduced filtering rates, liver damage, modified ionic regulation and changes in behavior (Erickson et al. 1986, Rabergh et al. 1991).

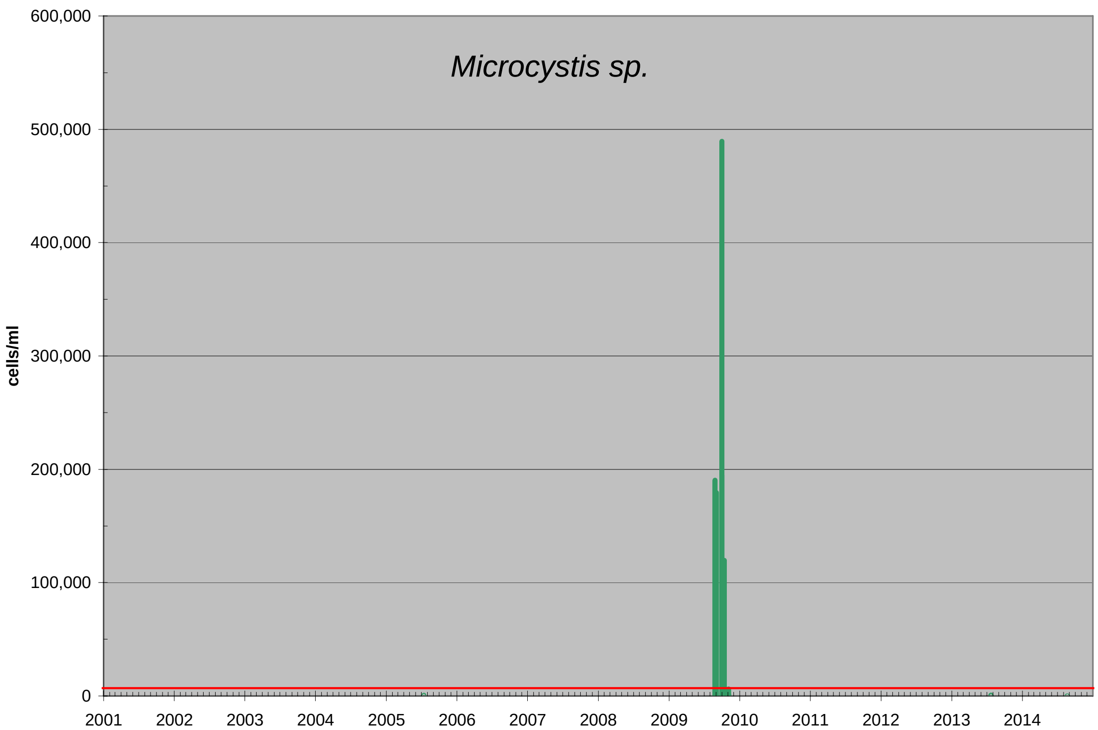

Figure 6.2.22 Occurrence of *Microcystis sp.* in the Maryland Coastal Bays, 2001-2014. Bloom threshold is 10,000 cells*ml^{-1} and potential toxin threshold exceedance may occur at 40,000 cells*ml^{-1}.

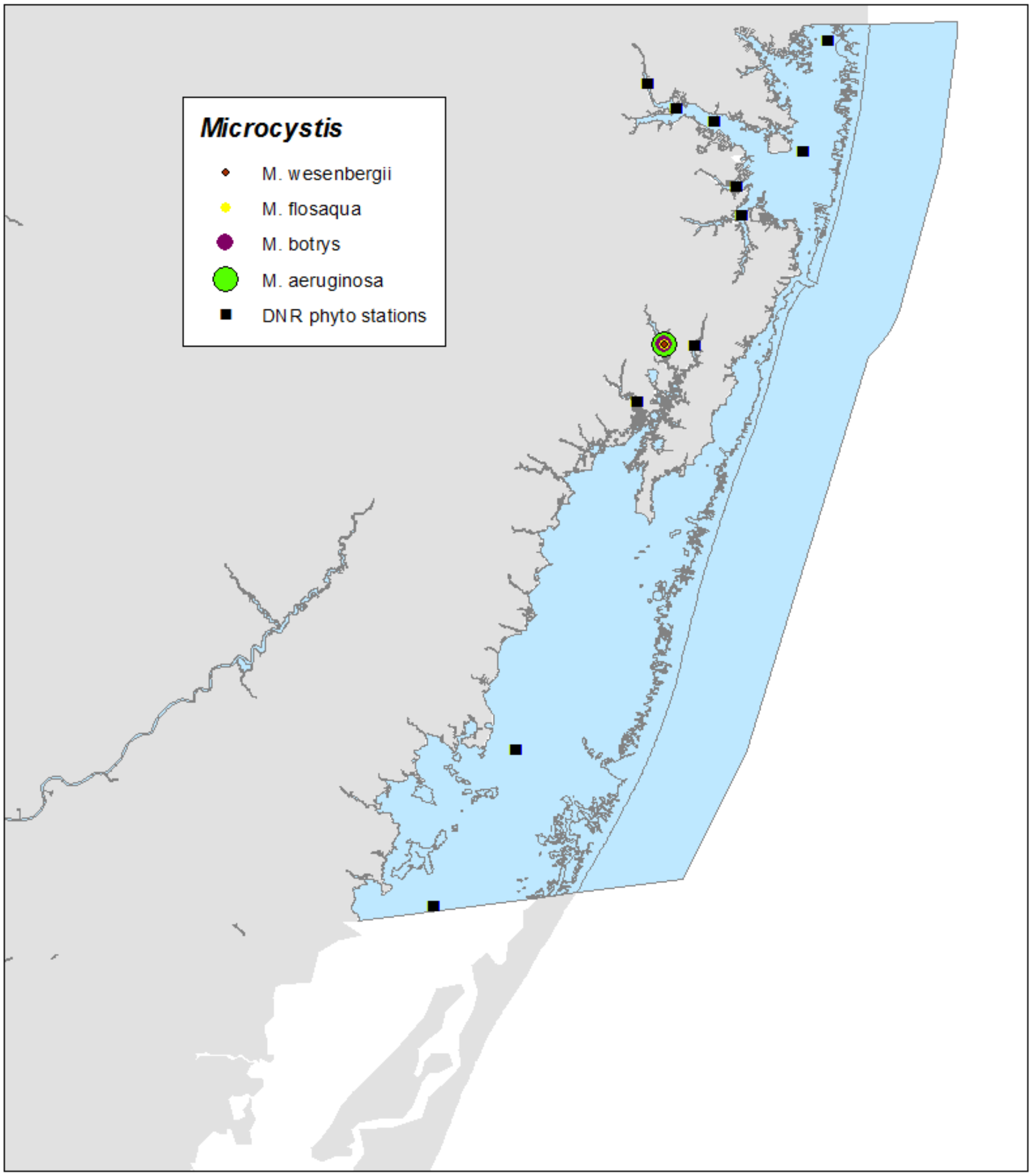

Figure 6.2.23 Distribution of *Microcystis species* in the Maryland Coastal Bays, 2007-2014.

2007, 2008 No blooms

2009 Bloom of *Microcystis aeruginosa* and *M. flosaguae* in Trappe Creek from late August to early November. Cell counts ranged from 101,346 to 489,500 cells* ml^{-1}.

2010-2013 No blooms.

b. <u>other Cyanophytes of Note</u>
Cyanophyte (bluegreen algae) concentrations in Bishopville Prong, Trappe Creek and Ayer's Creek have all shown declines from any pre-2000 phytoplankton sampling (Friedmen Chapter 3.3). Most species are not typically observed at the more saline stations in the coastal bays but in 2009 several species were detected at bloom levels in Trappe Creek (wet year and salinities were likely down).

Anabaena sp. were present in Trappe Creek in 2007 and bloom levels were observed in April 2009 and September/October (Figures 6.2.24 and 6.2.25). *Aphanizomenon sp.* also bloomed in Trappe Creek in 2009 (Figures 6.2.26 and 6.2.27). No toxin testing done. *Lyngbya sp* was not observed during the index time (2007-2013) but has been observed in 2005 and 2014 in the upper St. Martin River and Trappe Creek below bloom levels (Figures 6.2.28 and 6.2.29). *Oscillatoria sp.* was observed at blooms levels during September 2009 in Trappe Creek and St. Martin River (Figures 6.2.30 and 6.2.31).

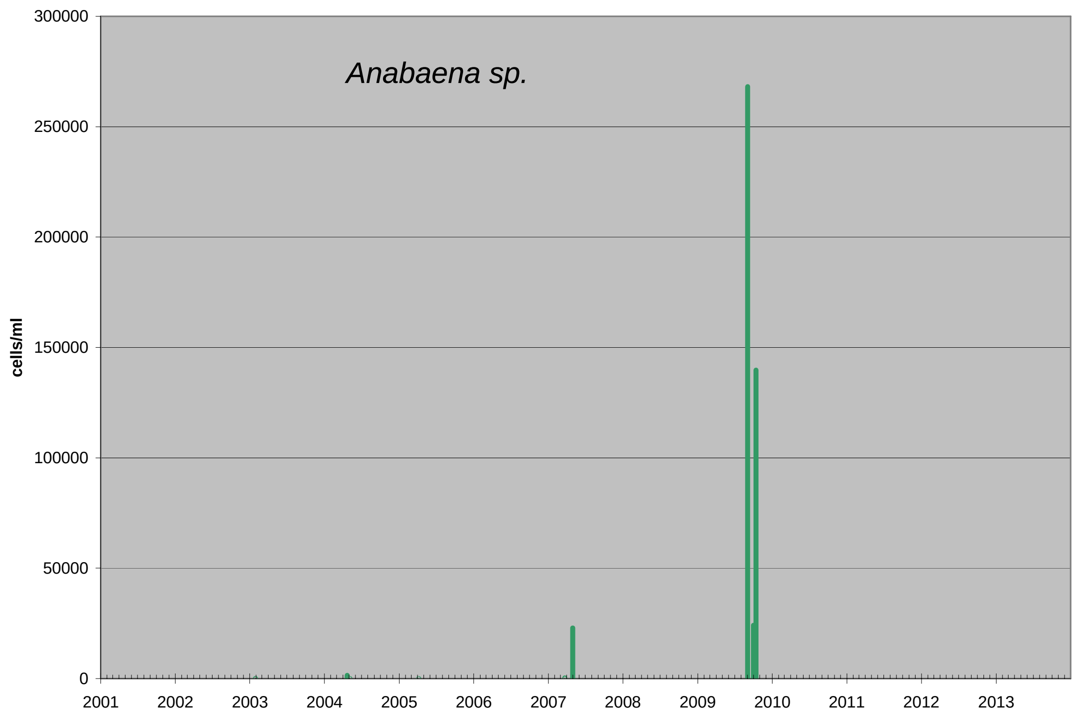

Figure 6.2.24 Occurrence of *Anabaena sp.* in the Maryland Coastal Bays, 2001-2014.

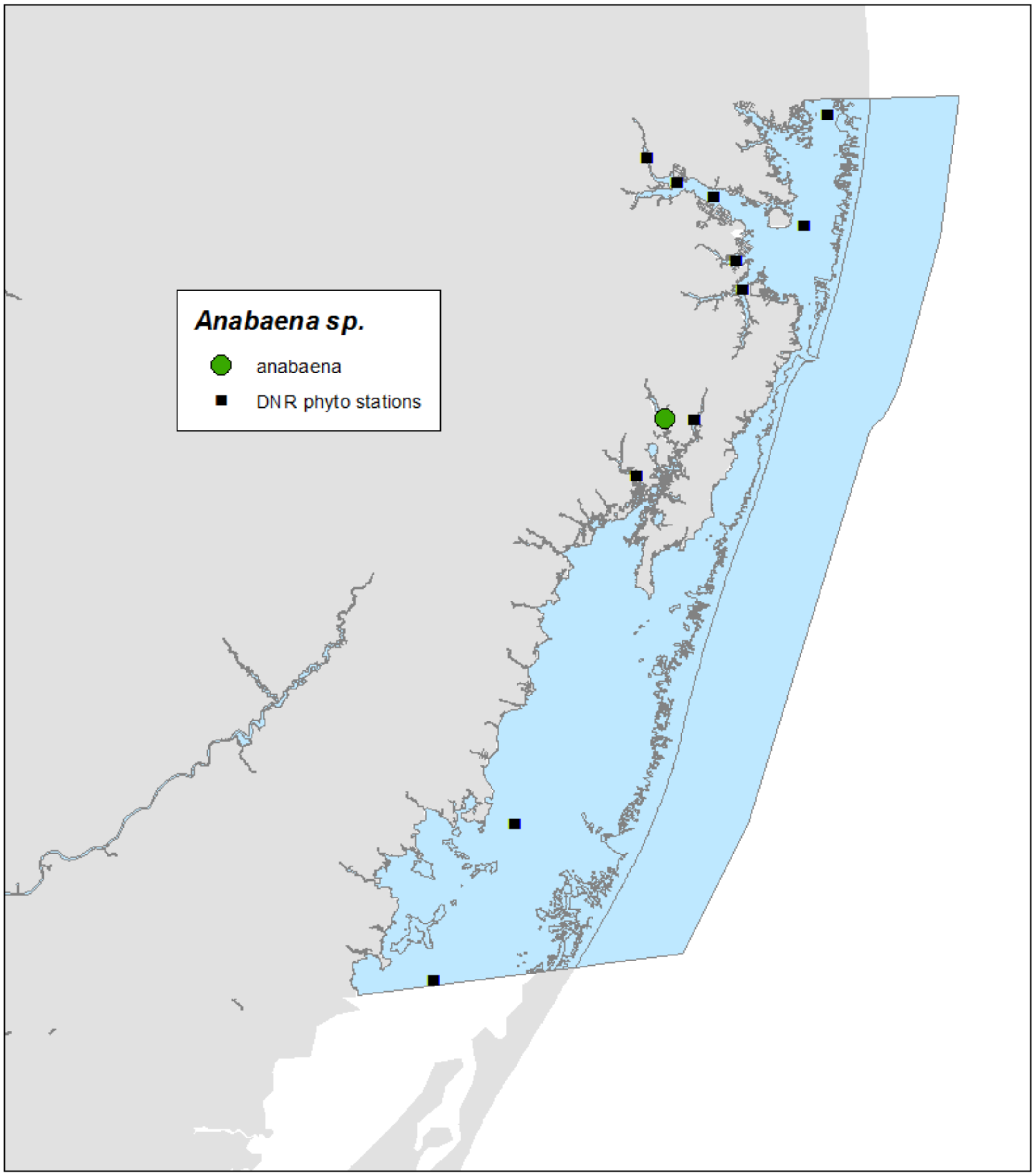

Figure 6.2.25 Distribution of *Anabaena sp.* in the Maryland Coastal Bays, 2007-2014.

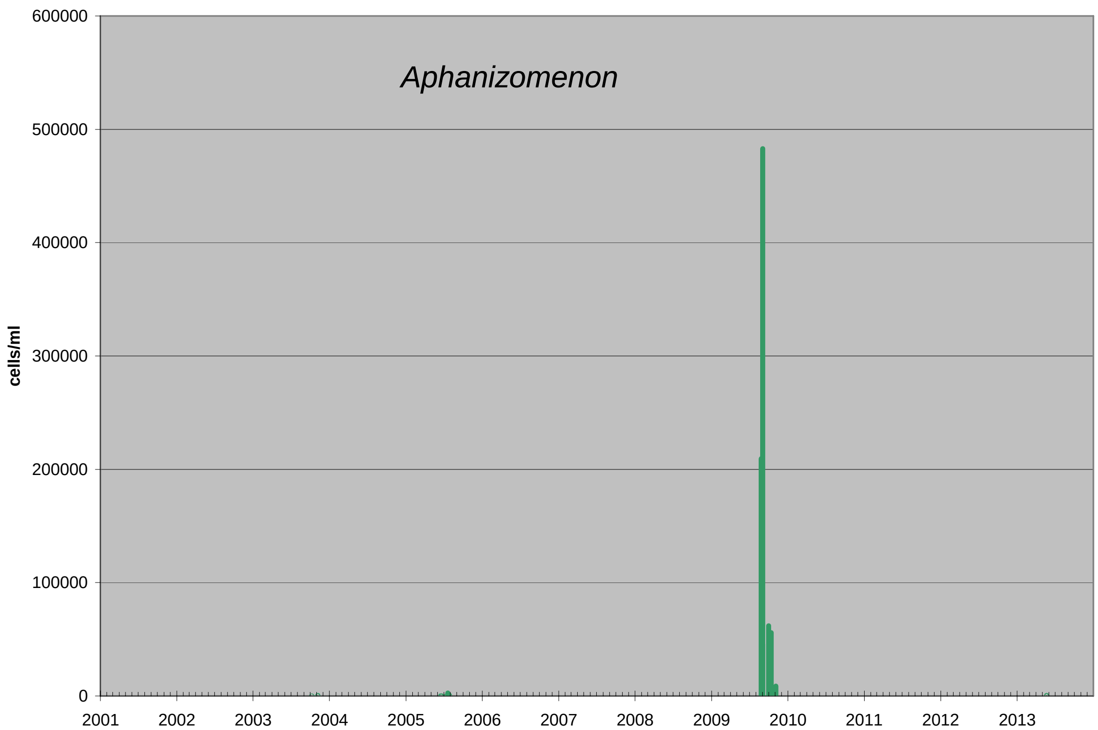

Figure 6.2.26 Occurrence of *Aphanizomenon* in the Maryland Coastal Bays from 2001-2014.

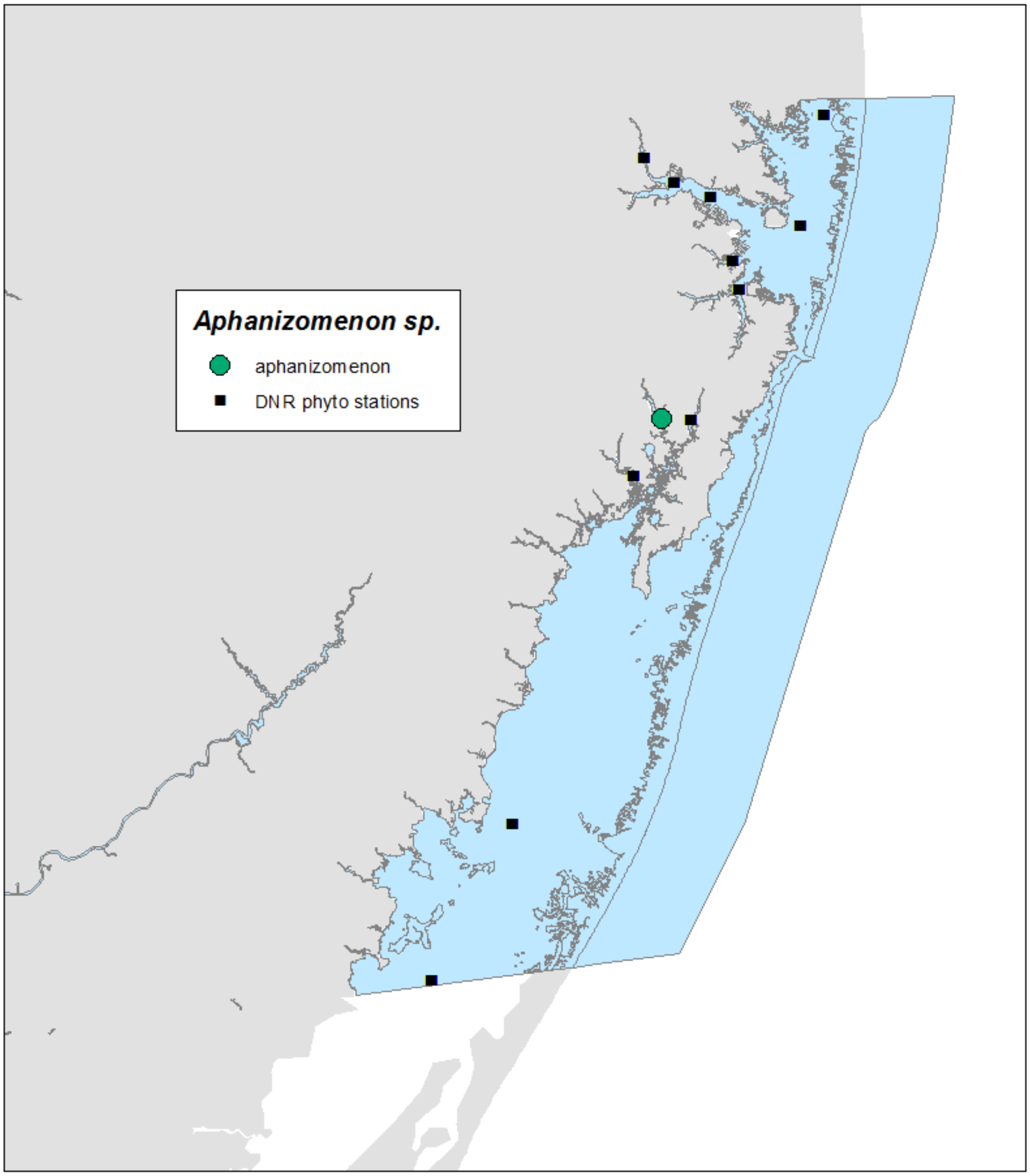

Figure 6.2.27 Distribution of *Aphanizomenon* in the Maryland Coastal Bays (2007-2014).

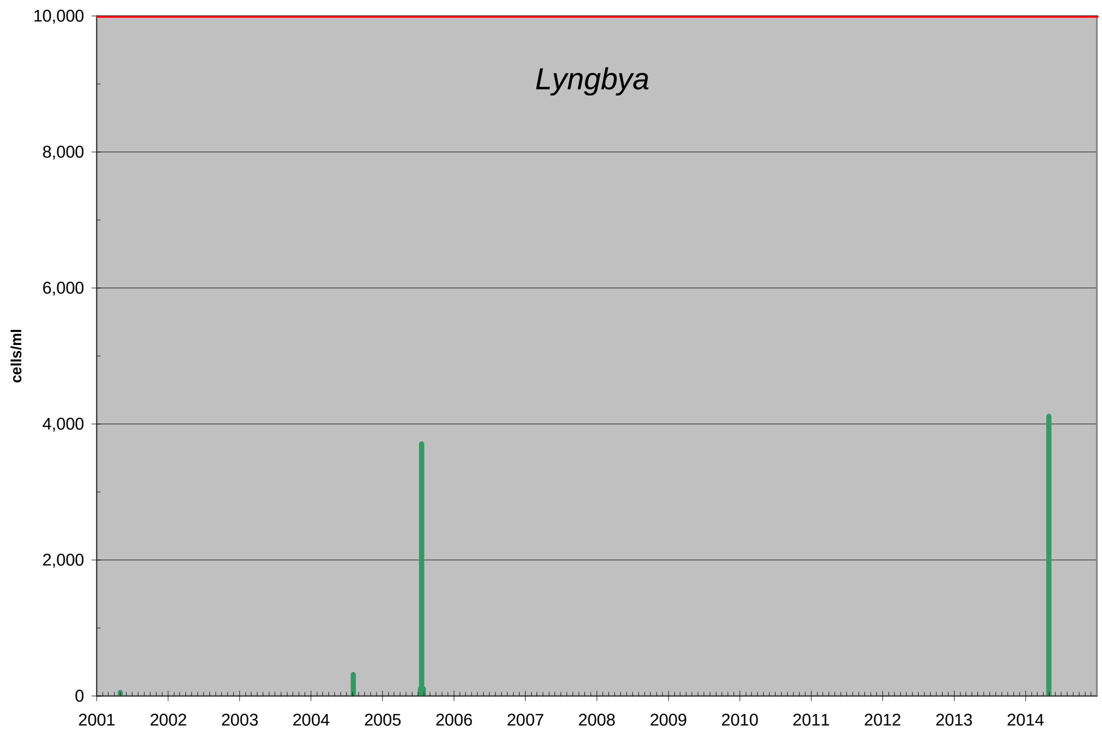

Figure 6.2.28 Occurrence of *Lygbya* in the Maryland Coastal Bays, 2001-2014.

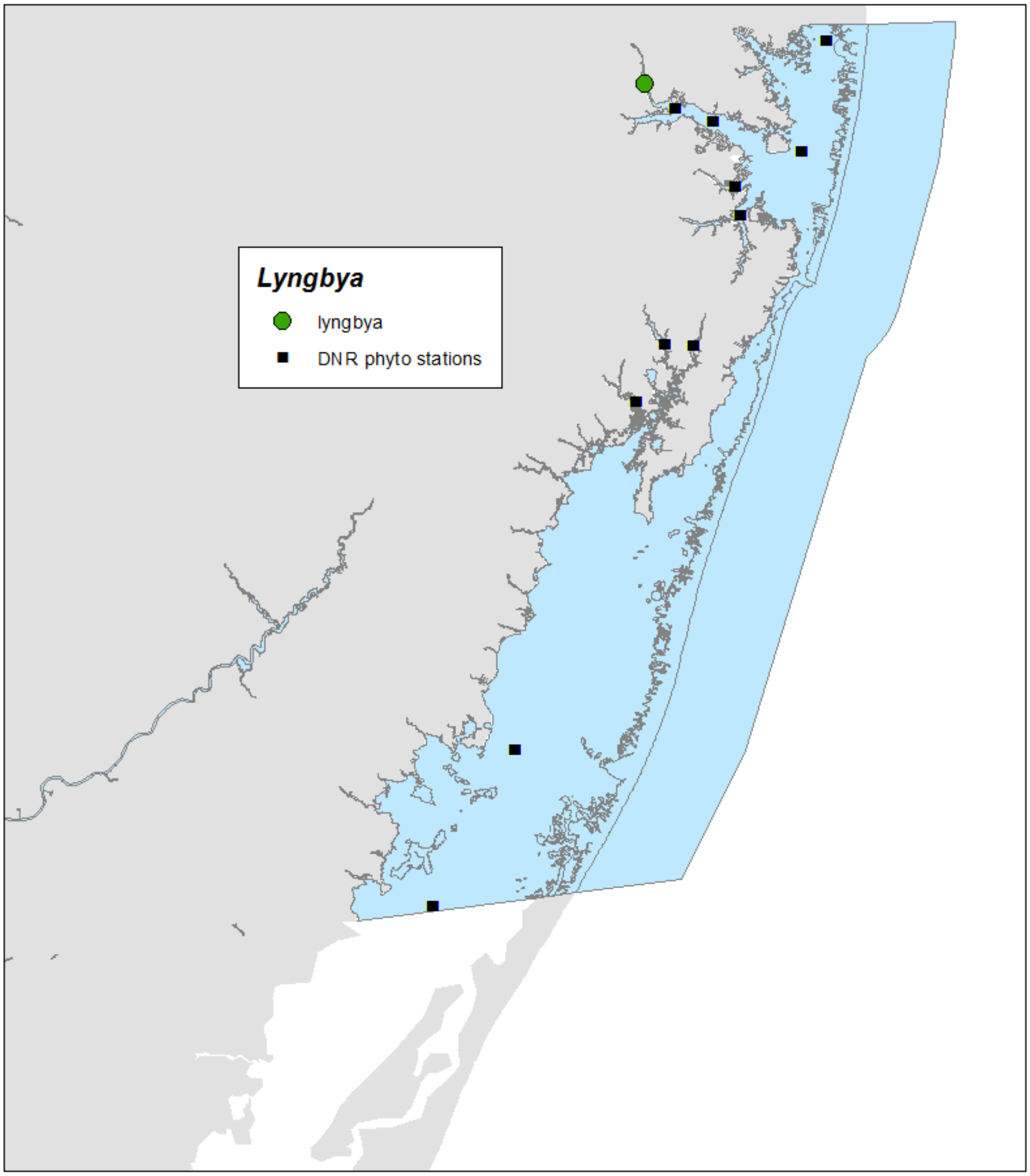

Figure 6.2.29 Distribution of *Lyngbya* in the Maryland Coastal Bays (2007-2014).

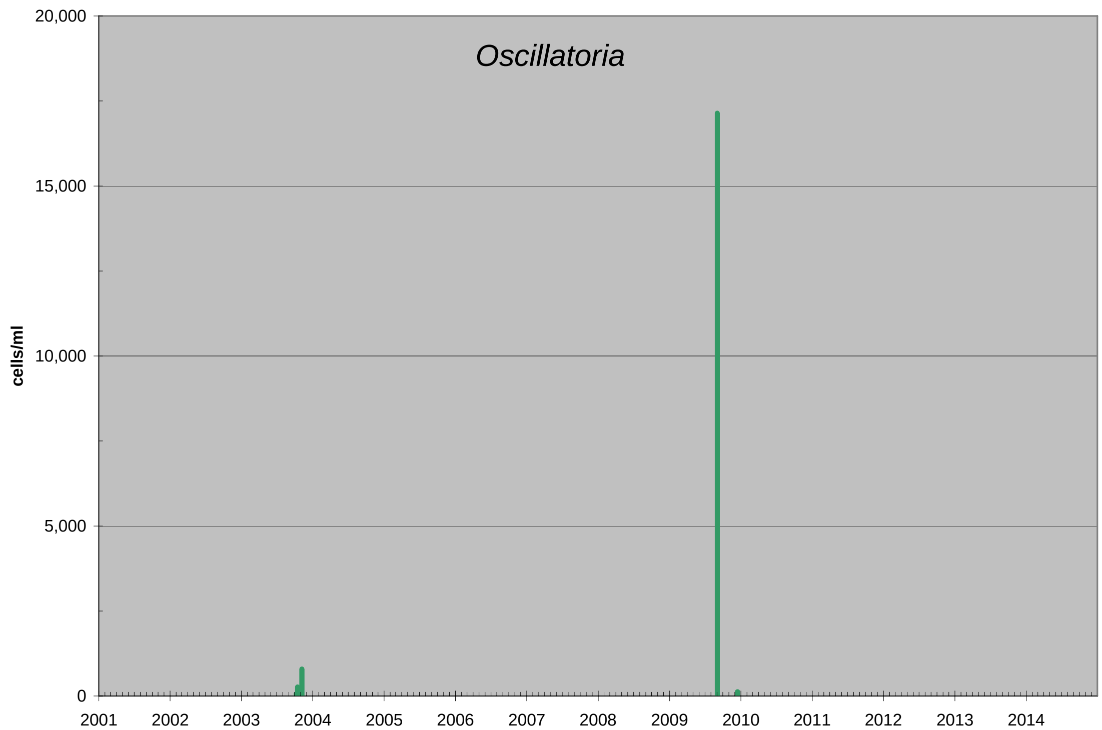

Figure 6.2.30 Occurrence of *Oscillatoria* in the Maryland Coastal Bays, 2001-2014.

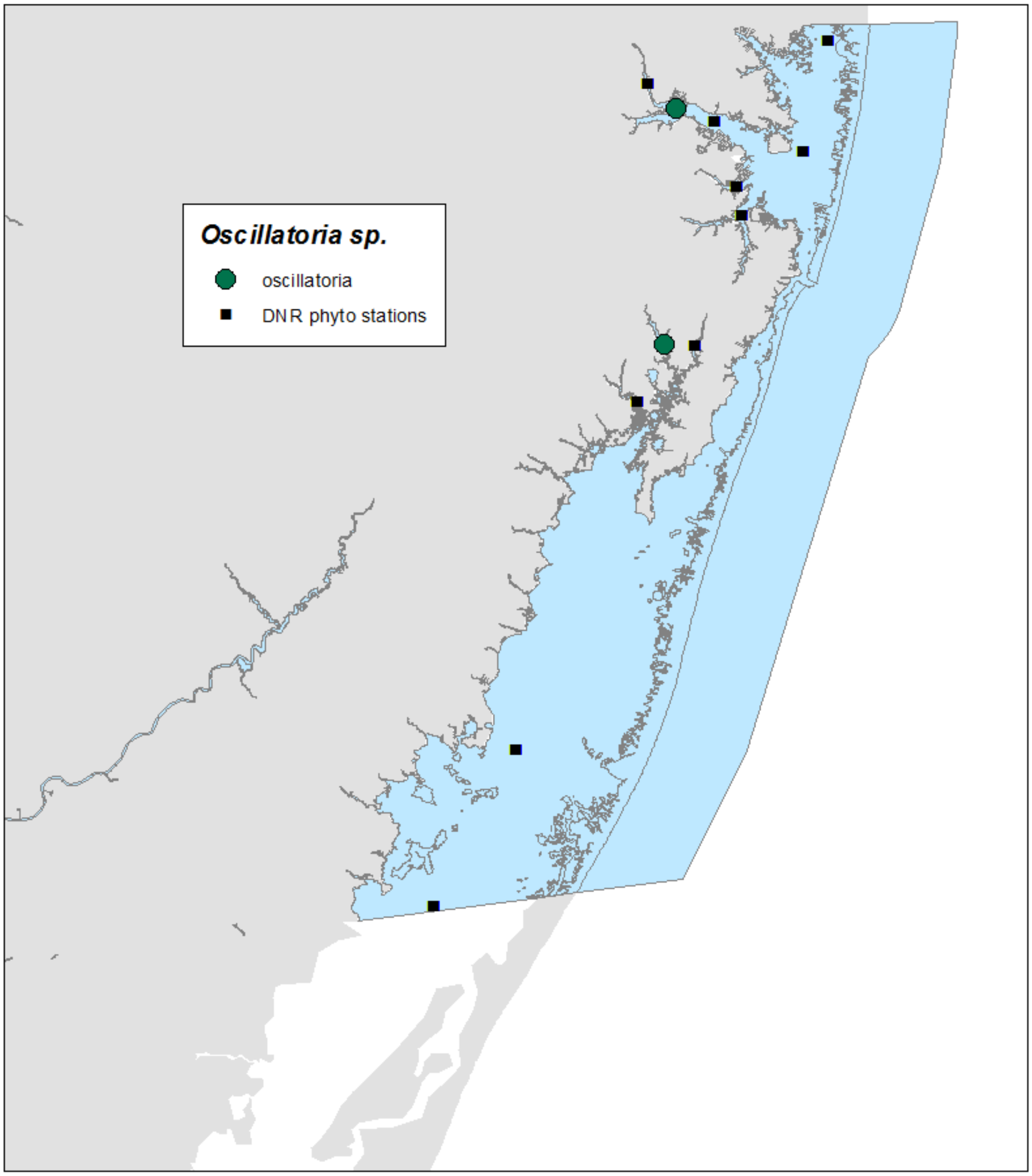

Figure 6.2.31 Distribution of *Oscillatoria* in the Maryland Coastal Bays (2007-2014).

b. <u>other Cyanophytes: Pico-cyanobacteria</u>

High levels of picoplankton have been observed since a new microscope was implemented in 2006 that allowed a greater magnification for small phytoplankton. Pico-cyanobacteria were abundant in 2007 and 2008 with highest concentrations (> one million cells* ml^{-1}) were observed in St Martin River, Trappe Creek, Ayers Creek and Marshal Creek. Concentrations of pico-cyanobacteria were moderate during 2009-2012. Positive identification of such small organisms is not possible using light microscopy alone. Further study is needed to identify the organism(s).

2007 Bloom levels exceeded two million cells*ml^{-1}in Trappe during September (bloomed from May to Sept). Blooms were also observed in Ayres Creek (786,587 cells*ml^{-1} in May), Marshal Creek and the St Martin River (July-October).

2008 Bloom levels exceeded one million cells*ml^{-1}.in Ayres Creek in June. Blooms also observed in Trappe Creek and St Martin River as well as Chincoteague (July 29 421,640 cells*ml^{-1}) and Assawoman (July 28- 589,280 cells*ml^{-1}) Bays.

2009 Blooms were found in Marshal Creek (447,040 cells*ml^{-1}) during June and in the upper St Martin River in September (497,840 cells*ml^{-1}).

2010 No bloom levels were detected.

2011 Significant bloom during April (923,036 cells*ml^{-1}) and June in Ayers Creek (409,346 cells*ml^{-1}) and in elevated in Trappe Creek, Marshall Creek and Bishopville Prong.

2012 No bloom levels were detected.

2013 No blooms were detected.

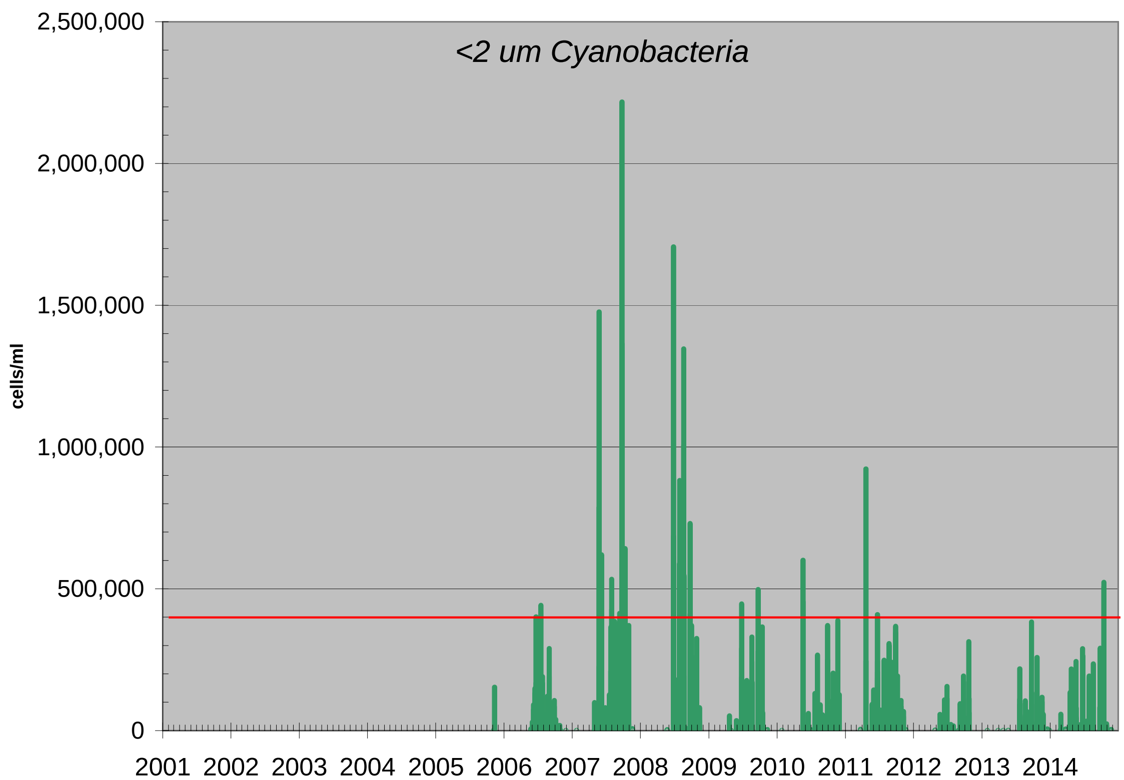

Figure 6.2.32 Occurrence of pico-cyanobacteria *(*<2μM) in the Maryland Coastal Bays, 2007-2014.

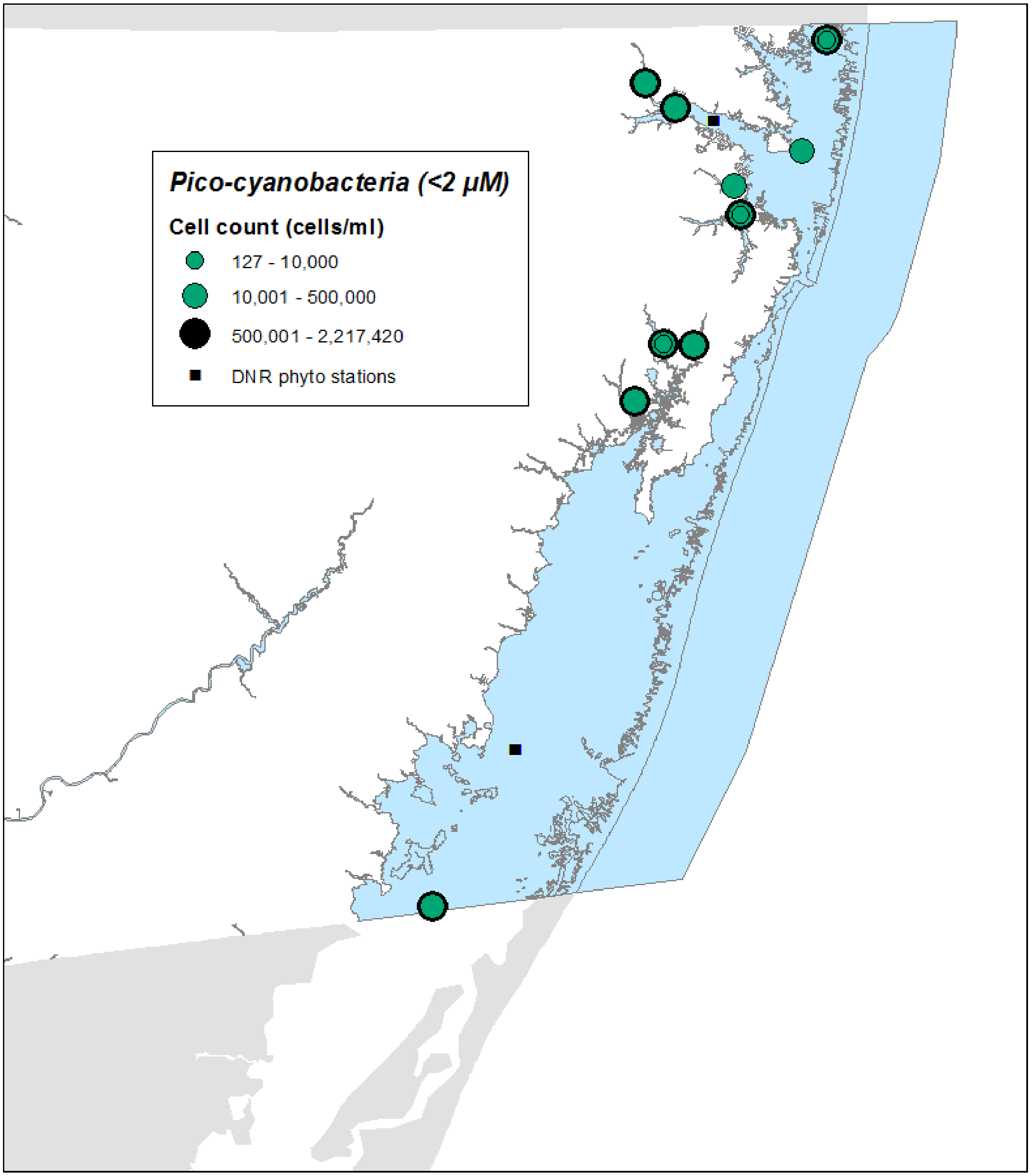

Figure 6.2.33 Distribution of pico-*cyanobacteria* (<2µM) in the Maryland Coastal Bays (2007-2014).

X. <u>*Gonyaulax*</u>
Gonyaulax is a genus of Dinoflagellates. *Gonyaulax spinifera* has been related to production of Yessotoxins (YTXs), a group of structurally related polyether toxins, which can accumulate in shellfish and can produce symptoms similar to those produced by Paralytic Shellfish Poisoning (PSP) toxins. All species are marine, except for one freshwater species *Gonyaulax apiculata.* It

previously included several species, which are now considered to belong to a separate genus, e.g. *Gonyaulax tamarensis* (now: *Alexandrium tamarense*).

Gonyaulax is observed infrequently in the Maryland Coastal Bays. Highest concentration was found in Bishopville Prong on the upper St Martin River during June 2004 (2,120 cells*ml^{-1}) and Trappe Creek in 2003 (864 cells*ml^{-1}). During 2007 to 2013 it was seldom recorded and only in the Bishopville Prong.

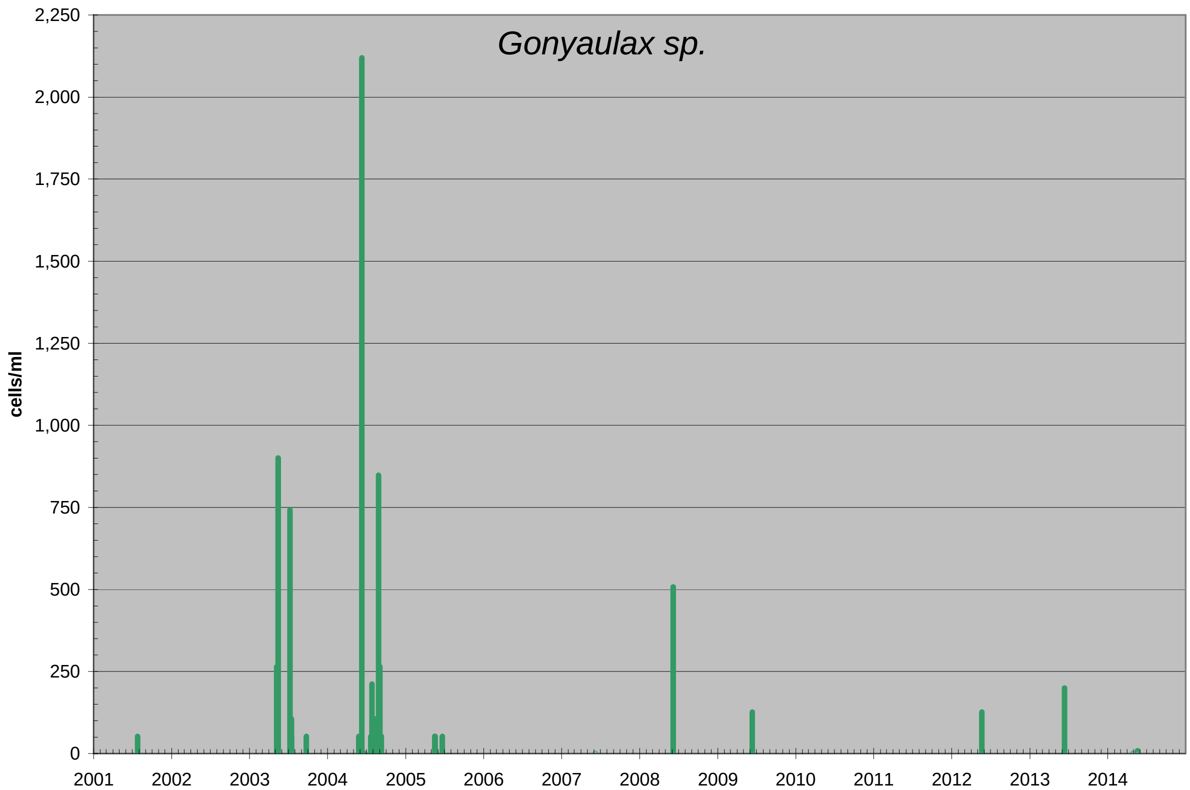

Figure 6.2.34 Occurrence of *Gonyaulax* in the Maryland Coastal Bays, 2001-2014.

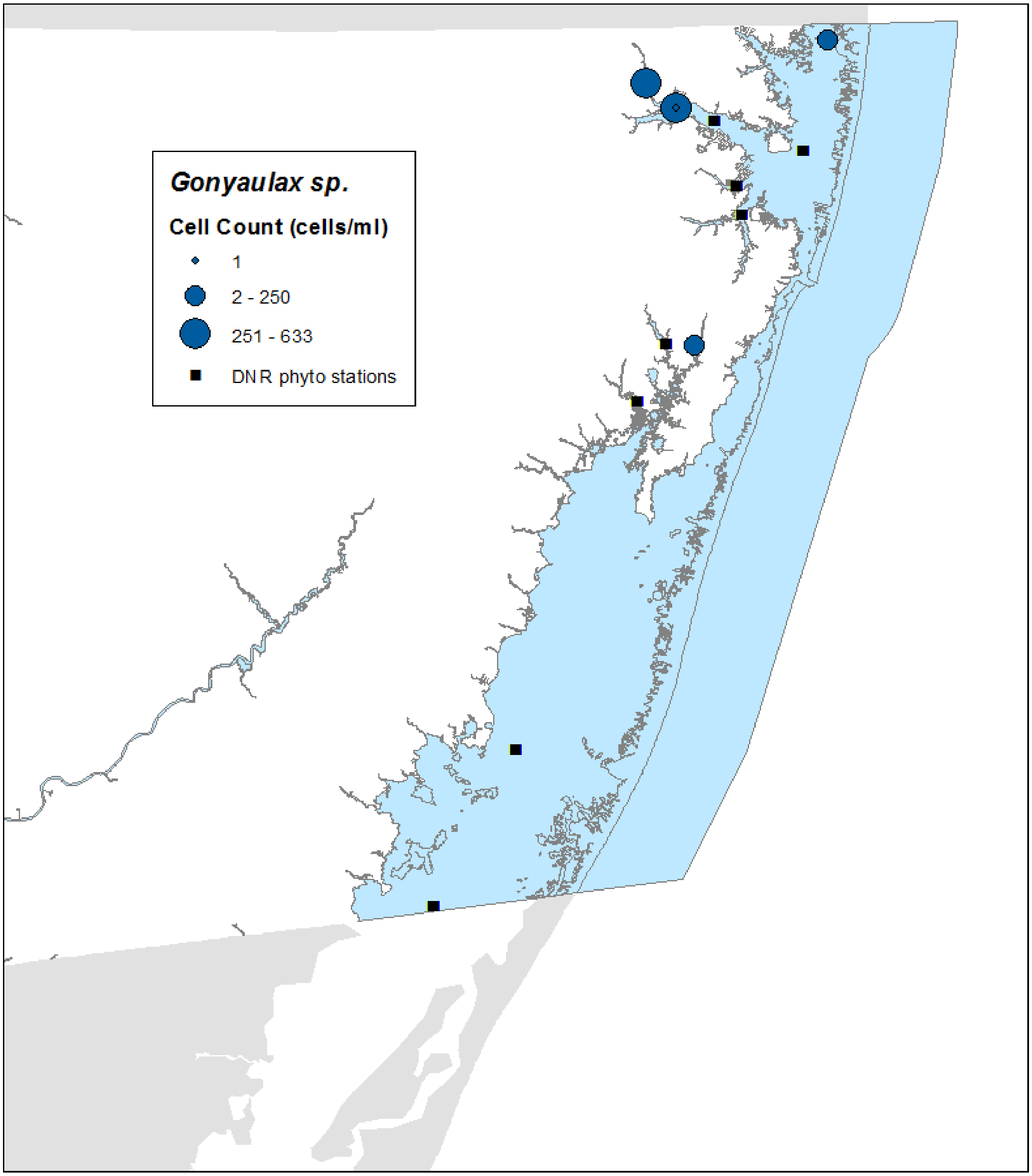

Figure 6.2.35 Distribution of *Gonyaulax* in the Maryland Coastal Bays (2007-2013).

XI. Ecosystem disruptive HAB: macroalgae

Macroalgae are considered harmful by the National Oceanographic and Atmospheric Administration (NOAA) when they produce dense overgrowth in localized areas, such as coastal embayments, receiving excessive nutrient loads. These accumulations can be so high as to cover the bottom, excluding other life. Also, when such large masses of macroalgae begin to die, excessive oxygen consumption associated with the decomposition process can rob the water of oxygen (Bushaw-Newton and Sellner 1999). Two genera of macroalgae are believed to qualify as HABs under NOAA's definition in specific areas of the Coastal Bays. *Gracilaria* in Turville Creek was so dense in 1999-2001 that it caused the fishery monitoring program to relocate a 25-plus year monitoring station. This system is prone to low dissolved oxygen levels that are probably influenced by these blooms. Furthermore, Total Maximum Daily Load models of this system were insufficient in predicting the low dissolved oxygen, likely because they failed to incorporate primary producers other that phytoplankton. *Chaetomorpha* levels in Chincoteague Bay were so dense during 1998-2001 that it is believed to have impacted scallop restoration efforts and seagrass density in some areas (R.Orth and M. Tarnowski, personal communication).

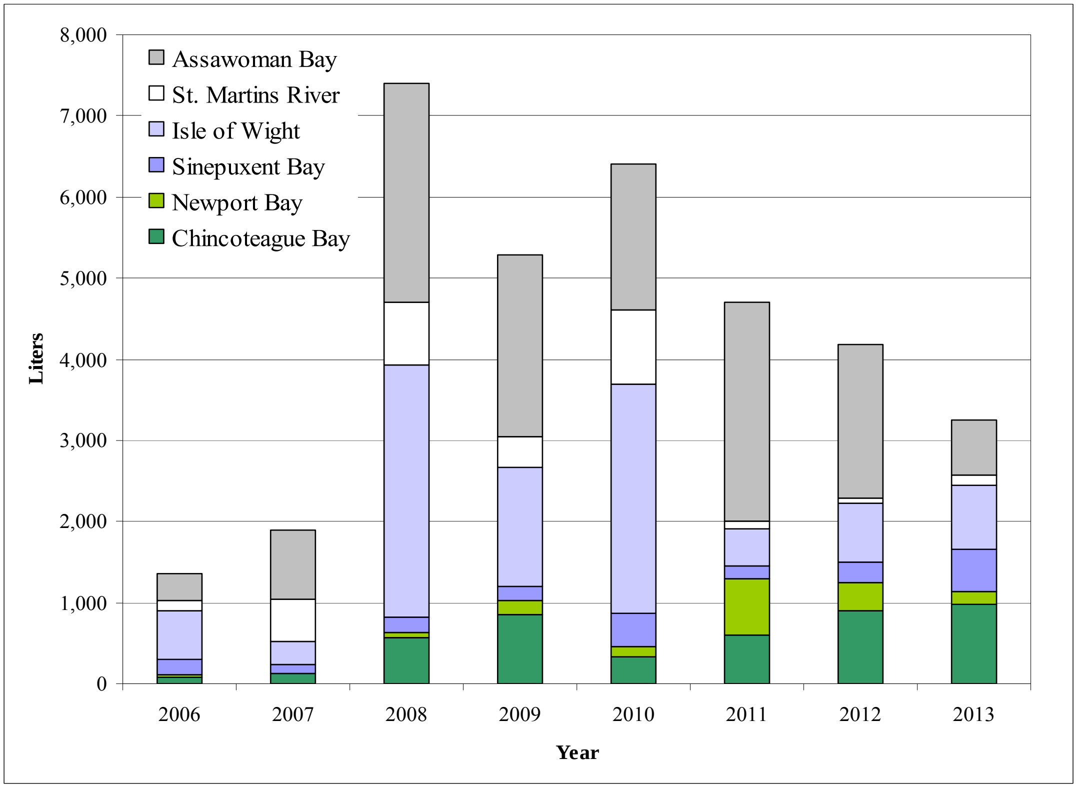

Figure 6.2.36 Macroalgae abundance by area and year from the Coastal Bays Fisheries Investigation Trawl and Beach Seine Survey (trawl n=140/year and beach seine n = 38/year). For more information please refer to Chapter 6.3 of this assessment.

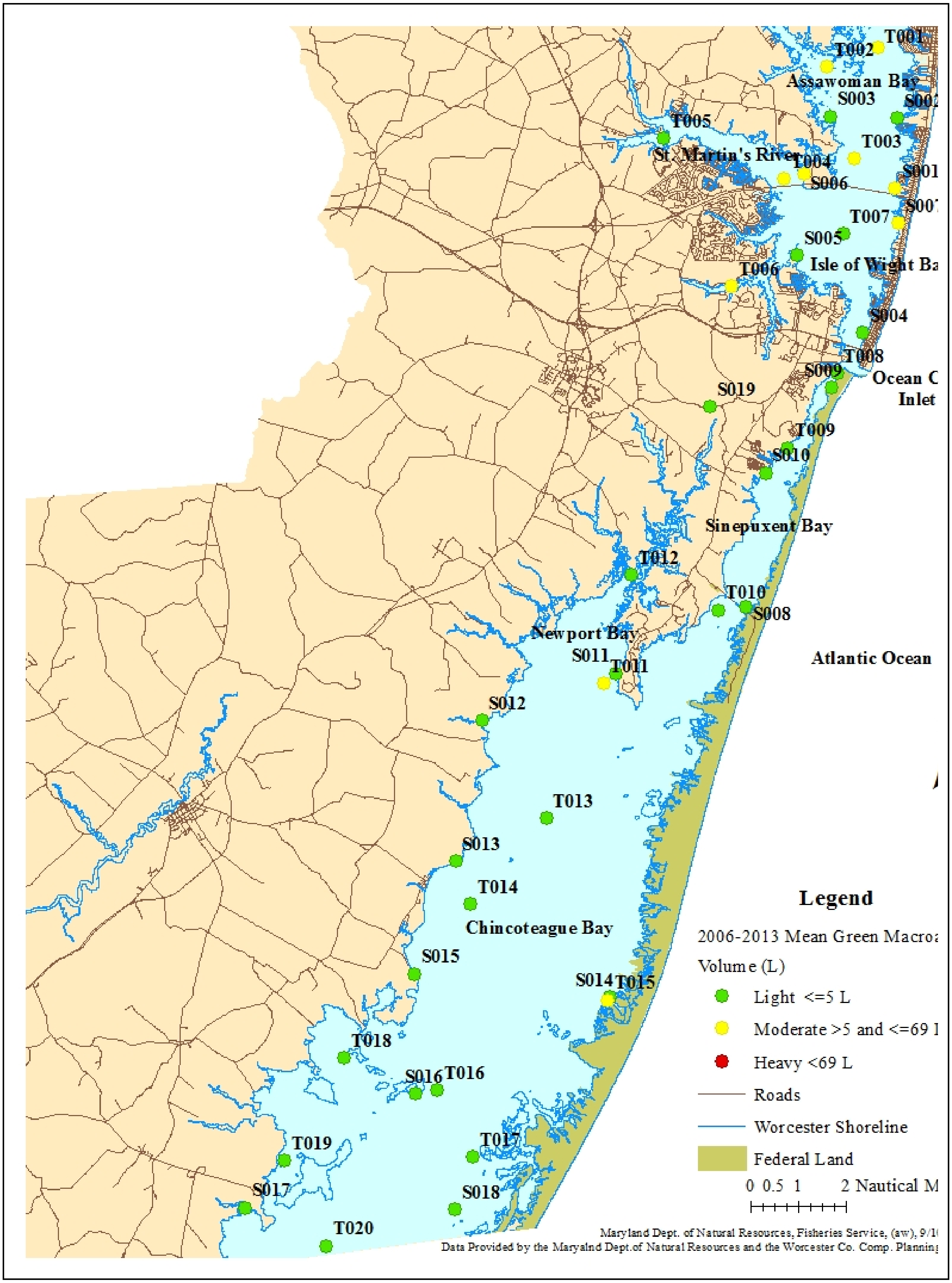

Figure 6.2.37a Distribution of green macroalgae in the Maryland Coastal Bays 2006-2013.

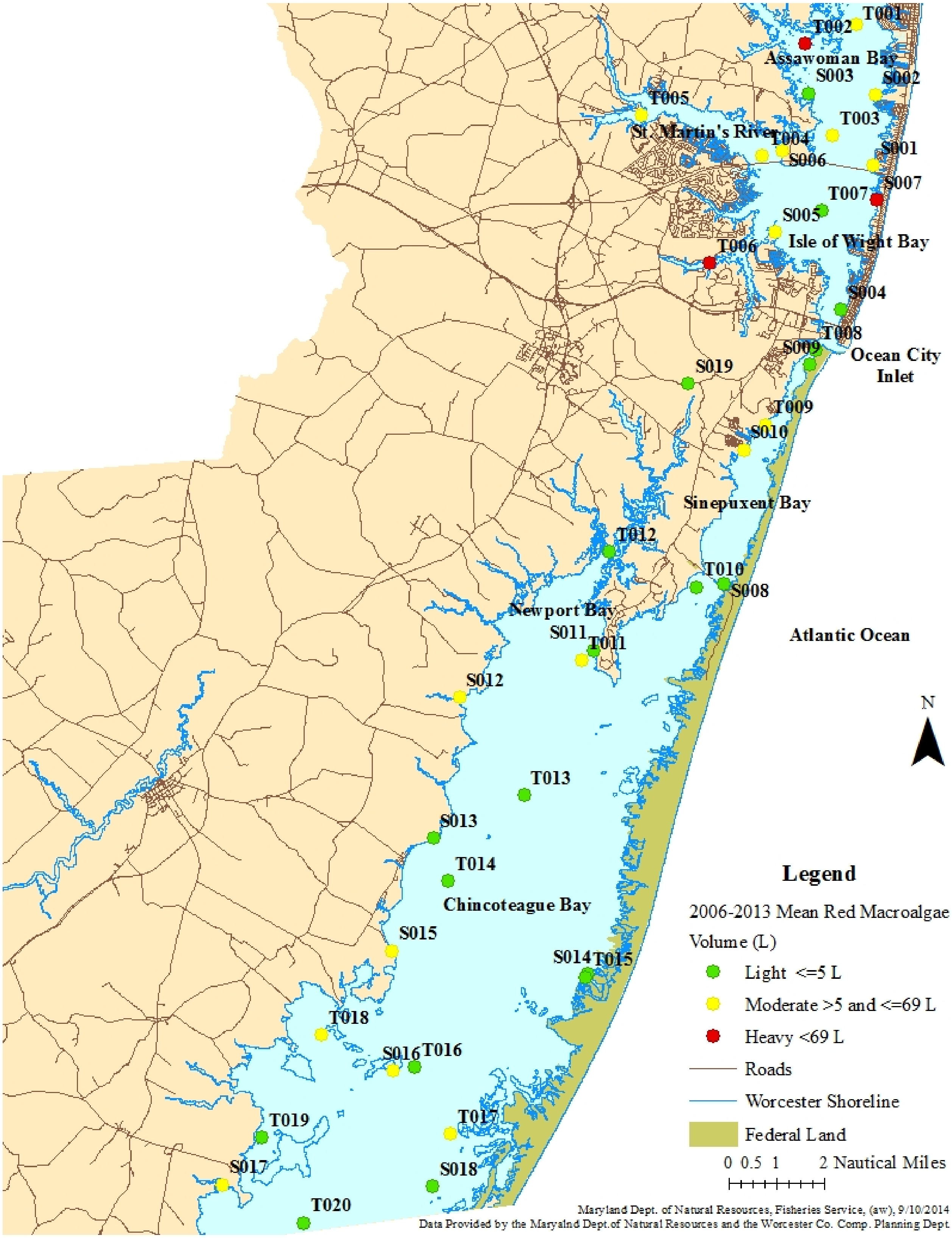

Figure 6.2.37b Distribution of red macroalgae in the Maryland Coastal Bays 2006-2013.

XII. Ecosystem Disruptive HAB: *Heterocapsa spp*.

The high biomass blooms of *Heterocapsa spp.* allows new nutrients delivered to the bays in the winter (generally a time of limited primary production) to be maintained in the bays and recycled for dpring blooms. *Heterocapsa rotunda* is more prevelant in the Coastal Bays than *H. triquetra*. *H. rotunda* showed a significant bloom in 2008 (Figure 6.2.38) and *H. triquetra* in 2010 (Figure 6.2.39 and Figure 6.2.40).

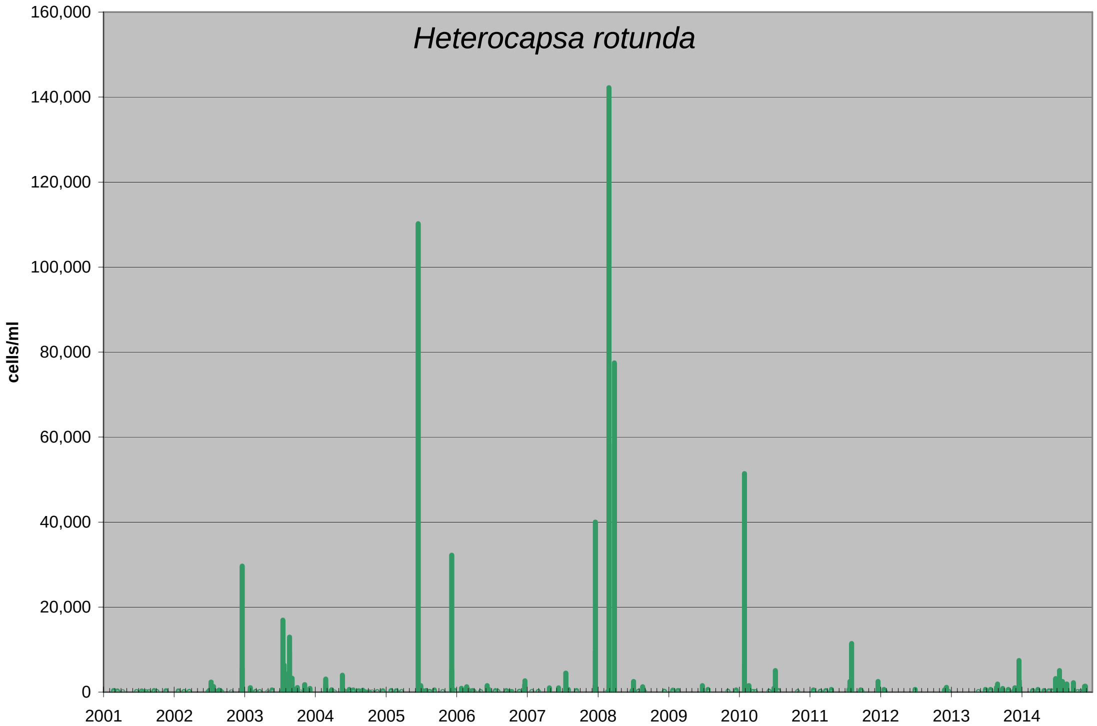

Figure 6.2.38 Occurrence of *Heterocapsa rotunda* in the Maryland Coastal Bays, 2001-2014.

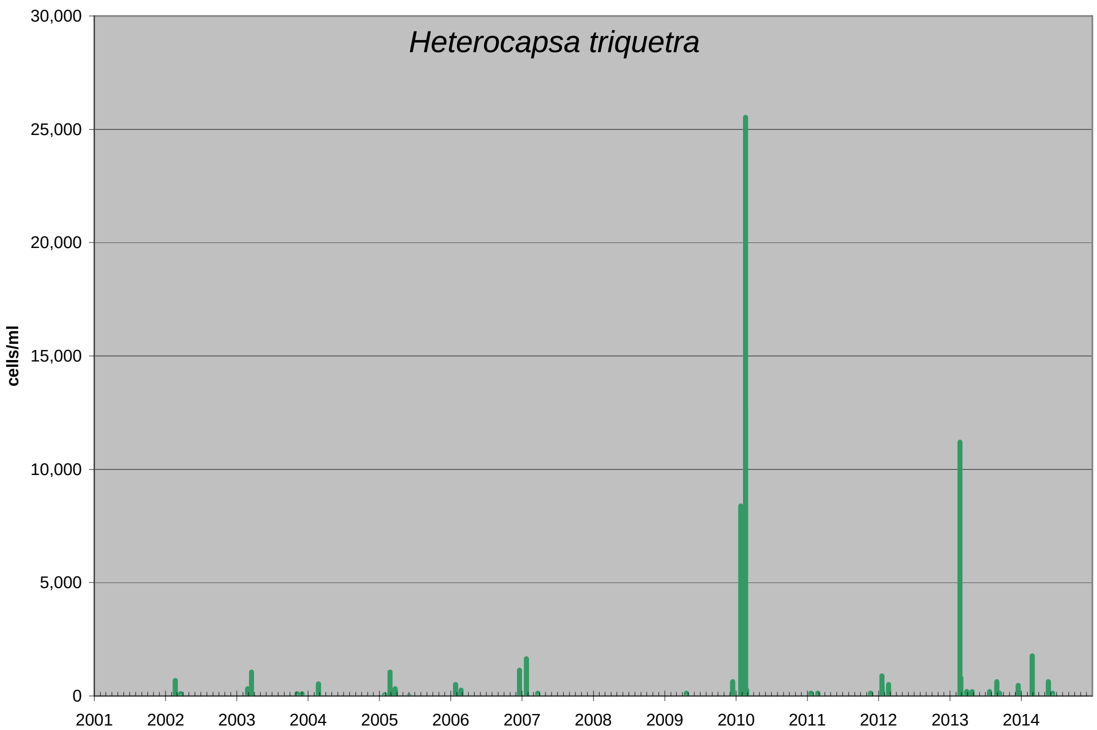

Figure 6.2.39 Occurrence of *Heterocapsa triquetra* in the Maryland Coastal Bays, 2001-2014.

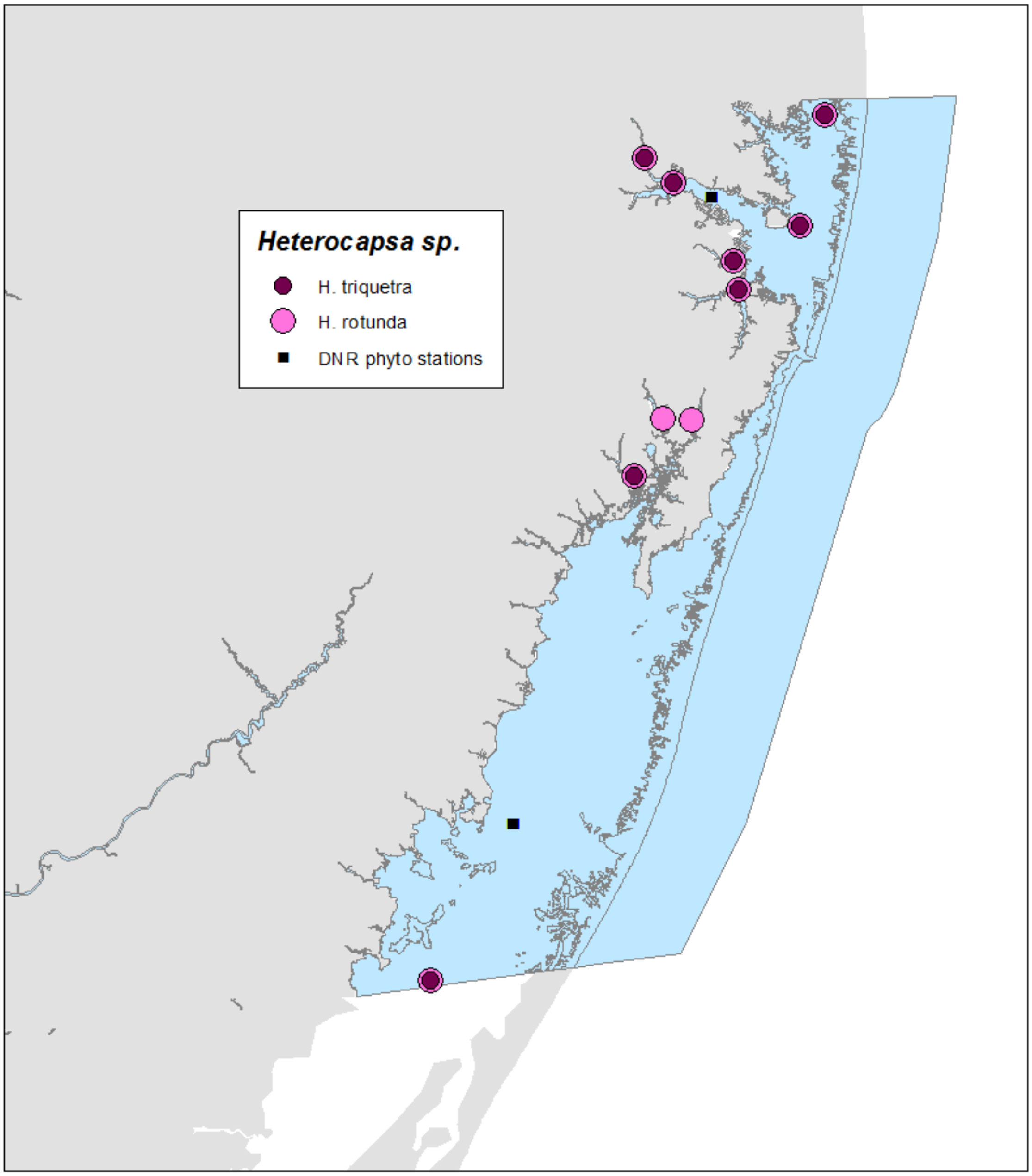

Figure 6.2.40 Distritubtion of *Heterocapsa sp* in the Maryland Coastal Bays (2007-2013).

XIII. Ecosystem Disruptive HAB: *Akashiwo sanguienum*

Harmful affects from this non-toxic dinoflagellate were first reported during a massive red tide (chlorophyll levels between 50-200 µg/L) that occurred November 2007 in Monterey Bay, California. Although this red tide bloom was ostensibly nontoxic, it was very harmful, causing unprecedented beach stranding of live and dead seabirds. Affected birds had a slimy yellow-green material on their feathers, which were saturated with water, and they were severely hypothermic. It was determined that foam containing surfactant-like proteins, derived from organic matter of the red tide, coated their feathers and neutralized natural water repellency and insulation This is the first documented case of its kind, but previous similar events worldwide may have gone undetected.

Akashiwo has been detected in the Coastal Bays with the highest concentrations found in the winter of 2013 in Manklin and Turville creeks (Figure 6.2.41s and 6.2.42). More offshore phytoplankton data would be useful to determine if blooms occur in the coastal Atlantic.

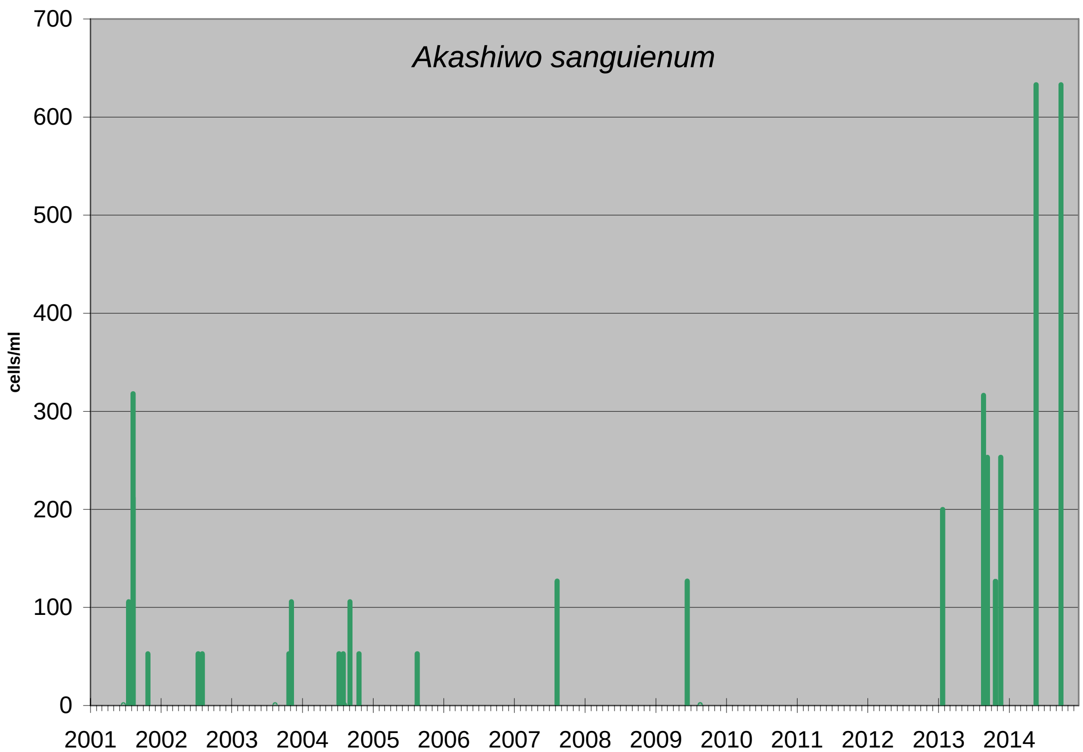

Figure 6.2.41 Occurrence of *Akashiwo seanguienum* in the Maryland Coastal Bays, 2001-2014.

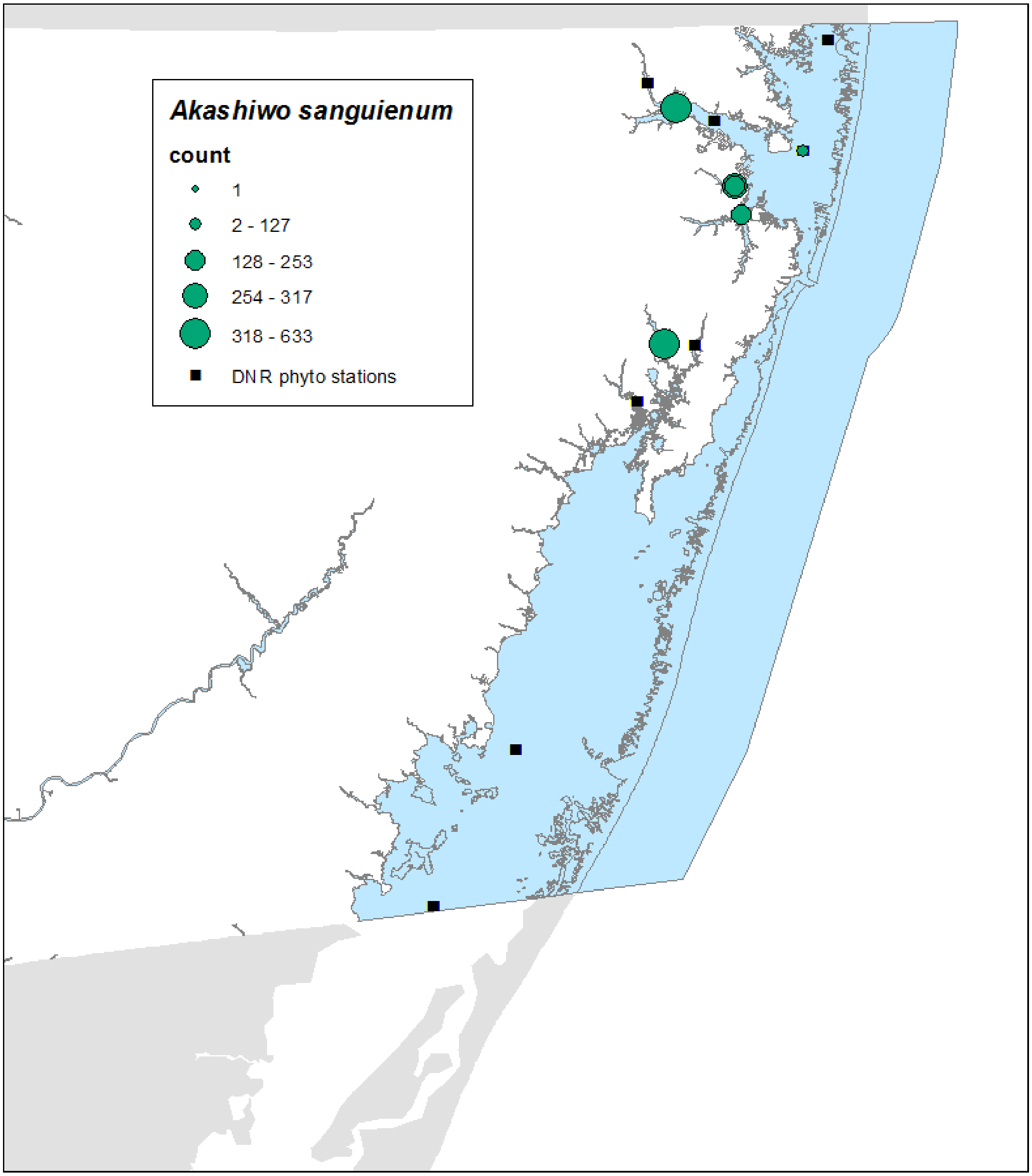

Figure 6.2.42 Distritubtion of *Akashiwo seanguienum* in the MD Coastal Bays (2007-2014).

Summary
Approximately 5% of the phytoplankton community identified in Maryland's Coastal Bays, represent HAB species. HABs are recognized for their potentially toxic properties as well as their ability to produce large blooms negatively affecting light and dissolved oxygen resources. Brown tide (*A. anophagefferens*) has been the most widespread and prolific species in the area in recent years producing growth impacts to juvenile clams in test studies and potential impacts to seagrass distribution and growth (see Chapter 6.1). Little toxic activity has been detected among the Coastal Bays phytoplankton such as *Dinophysis acuminate*, Raphidophyte species and recently some cyanobacteria species. However, other species such as *Pseudo-nitzschia seriata*, *Prorocentrum minimum*, *Pfiesteria piscicida,* and *Karlodinium micrum* have produced positive toxic bioassays or generated detectable toxins in the Chesapeake Bay. *Pfiesteria piscicida* was retrospectively considered as the likely causative organism in a large historical fish kill on the Indian River, Delaware. Similarly *Chloromorum toxicum* (formerly *Chattonella* cf. *verruculosa*) was implicated in a large fish kill and persistent brevetoxins detected in Delaware's Rehobeth Bay during 2000. Large increases in macroalgal may be evidence of a seagrass dominant system balancing on the edge of a eutrophic state. Tracking HAB species diversity, abundance, distribution and toxic activity through time provides important indicators of environmental change for the Coastal Bays.

Thirteen potentially harmful algae species have been identified as threats in the Maryland Coastal Bays including *Aureococcus anophagefferens* (Brown Tide), *Pfiesteria piscicida and P. shumwayae*, *Chattonella*, *Heterosigma akashiwo*, *Fibrocapsa japonica*, *Prorocentrum minimum, Dinophysis sp.*, *Amphidinium sp*., *Pseudo-nitzchia sp.*, *Karlodinium* and two macroalgae genera (*Gracilaria*, *Chaetomorpha*). Several other HAB species are also present from time to time including cyanobacteria species (*Microcystis*, *Anabaena*, and *Aphanizomenon*), *Gonyaulax*, and *Heterocapsa spp*, Presence of HAB species has been most diverse (i.e. greaterst richness of HAB species) in the polluted tributaries of St. Martin River and Newport Bay (figure 32).

Brown tide is the predominant species that exceeds published threshold levels (see Chapter 6.1). The years 1999 and 2002 had category two blooms in the northern and southern bays, while 2003 had the most extensive bloom (temporally and spatially) in the southern bays when no other area in the northeastern United States reported blooms.

Other threshold exceedances include *Chloromorum toxicum* (formerly *Chatonella cf. verruculosa*) in September 2002 on St. Martin River. A bloom of *C. cf. verruculosa* bloom during 1999 in Delaware Coastal Bays was related to a fish kill event, no evidence of toxicity by any of these species has been associated with similar events in Maryland waters. Threshold exceedances (3,000 cells*ml-1) of *P. minimum* were recorded once each year during April 2001 and 2002 on Bishopville Prong/St. Martin River. *Heterosigma akashiwo* 750-1,000 cells*ml^{-1} have been known to affect mariculture operations, however, *H. akashiwo* has thus far shown no evidence of toxic activity in the coastal bays when recorded above this threshold. *Fibrocapsa japonica* is present in the Coastal Bays but no known cell density thresholds are available to estimate possible effects or warrant intensified surveys for this species.

Dinophysis has been observed above threshold concentrations in Assawoman Bay (2001 once, 2003 once), Isle of Wight (2002 once) and St. Martin Creek (2001 once, 2002 seven times and 2003 twice). However, there is no evidence for toxicity to date in the Coastal Bays systems. All samples could potentially generate intensified monitoring for toxins but > 5 cells*ml^{-1} is probably a more appropriate threshold. The greatest concentrations of *Dinophysis* (up to 10 cells*ml-1, Canada action threshold is considered 5/ml) were found in areas closed to shellfishing (St. Martin, Turville and Herring Creeks), low concentrations (up to 2 cells*ml^{-1}) were observed in the Isle of Wight

Between 2007-2013, samples from the Maryland Coastal Bays have exceeded suggested living resource effects levels of $\geq$ 10,000 cells*ml-1 for *K. micrum* (2009 and 2011) or 200 cells*ml^{-1} for *Pseudo-nitzschia sp.* Cyanobacteria are encountered but have, in general, declined compared with pre-2000 data; however, pico-cyanobacteria may be increasing. Rare encounters of *Microcystis aeruginosa and other potentially toxic cyanobacteria (Anabaena, Aphanizomenon, Lyngbya, Oscillatoria)* are likely due to limited freshwater and low salinity habitat for this species.

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Table 6.2.3 Potential harmful algae bloom, HAB, species found at each sampling station from 1988 through 2013 in A) above the Ocean City Inet (northern bays) and B) below the Ocean City Inlet (sourthern bays). For a discussion of brown tide (*Aureococcus anafagefferens*) see Ch 6.1.

A. Northern Bays	XDN6454	XDN3445	XDM4486	XDN4797	XDN4312	TUV0011	TUV0019
Aureococcus anafagefferens	√		√	√	√	√	√
Chattonella cf. verruculosa			√	√	√	√	
Chattonella subsalsa			√	√			
Dinophysis sp.		√	√	√	√		√
Fribrocapsa japonica			√	√			
Heterosigma akashiwo			√	√	√		√
Karlodinium sp	√	√	√	√	√	√	
Microcystis sp							
Pfiesteria sp.			√				
Prorocentrum minimum			√	√	√	√	√
Pseudo-nitzschia		√				√	

B. Southern Bays	AYR0017	TRC0043	NPC0012	MSL0011	XDN3724	XBM1301	XDN3527
Brown tide	√	√	√	√	√	√	
Chattonella cf. verruculosa	√	√	√	√			√
Chattonella subsalsa	√	√	√	√			
Dinophysis sp.							
Fribrocapsa japonica							√
Heterosigma akashiwo	√	√	√	√			√
Karlodinium sp	√	√	√	√			√
Microcystis sp	√	√	√				
Pfiesteria sp.							
Prorocentrum minimum	√	√	√	√			
Pseudo-nitzschia							