# Investigation of Fish Kills Occurring in the Peconic River - Riverhead, N.Y. Spring 2015



Suffolk County Department of Health Services James L. Tomarken, M.D., M.P.H., M.B.A., M.S.W. Commissioner

New York State Dept. of Environmental Conservation Marc Gerstman, Acting Commissioner

Stony Brook University School of Marine and Atmospheric Sciences Christopher J. Gobler, Ph.D., Associate Dean for Research, Professor

January, 2016

#### Acknowledgements

This investigation was conducted by personnel of the Suffolk County Department of Health Services, New York State Department of Environmental Conservation, Cornell University, and Stony Brook University, School of Marine and Atmospheric Sciences. This is in acknowledgement of the many contributions to the project by the following individuals:

## SUFFOLK COUNTY DEPARTMENT OF HEALTH SERVICES

Division of Environmental Quality: Walter Dawydiak, Director

**Office of Water Ecology:** Chris Lubicich, Robert Waters, Mike Jensen, Nancy Pierson, Gary Chmurzynski, Philip DeBlasi, Andrew Seal, Lorian Peterson, Kathy Governale, Mark Long, Shawn Droskoski

**Public and Environmental Health Laboratory:** Joette Pavelka, Vito Albanese, Paul Ames, Craig Baier, Cassandra Boulukos, Jeanette Calicchio, Robin Carpenter, Christopher Conte, Alice Curtis, Preciossa David, Anthony Emanuele, Catherine Krupp Thompson Lu, Scott Manuel, Theresa Marrone, Natalia Medvedeva, Scott Mirabella Eileen Paley, Lance Rosenberg, William Sarovec, April Wolf

### NYS DEPARTMENT OF ENVIRONMENTAL CONSERVATION

**Bureau of Marine Resources:** Stephen Heins, John Maniscalco, Sandra Dumais, Leonora Porter, Joe Orlando

Bureau of Fisheries: Andrew Noyes

### **CORNELL UNIVERSITY**

Aquatic Animal Health Program: Helene Marquis, DVM, PhD, Rod Getchell, PhD, Kelly Sams

### STONY BROOK UNIVERSITY

School of Marine and Atmospheric Sciences: Dr. Christopher Gobler, Dr. Theresa Hattenrath-Lehmann

\*Cover photo by Terry Hulse

## Table of Contents

1
2
4
5
10
20
21
24
26

# List of Tables

Table 1.	The historic occurrence of depressed DO levels at the CR105 Bridge water quality sonde site (June-August)
Table 2.	Densities of the most abundant phytoplankton species (groups) found in the water column in the Peconic River region during May - June of 2015
	List of Figures
Figure 1.	Peconic River sampling station locations
Figure 2.	Daily fluctuations in dissolved oxygen concentrations during an algal bloom in the Peconic River
Figure 3.	Western Peconic Estuary Dissolved Oxygen Concentrations (Daily Means, June-August, 2000-2015)7
Figure 4.	Dissolved oxygen and temperature levels at the Peconic River CR105 Bridge - SCDHS sonde data, May-August, 20089
Figure 5.	Dissolved oxygen, temperature and chlorophyll concentrations at the Peconic River CR105 Bridge - USGS water quality sonde data, April-August, 2013
Figure 6.	Dissolved oxygen, temperature and chlorophyll concentrations at the Peconic River CR105 Bridge - USGS water quality sonde data, April-August, 2014
Figure 7.	Dissolved oxygen, temperature and chlorophyll concentrations at the Peconic River CR105 Bridge - USGS water quality sonde data, April-July, 2015
Figure 8.	Maximal densities (cells/mL) of the most abundant dinoflagellates in the lower Peconic River region during May - June of 201516
Figure 9.	Minimum dissolved oxygen and mean chlorophyll concentrations at the Peconic River CR105 Bridge - USGS water quality sonde data, May-July, 2013-2015
Figure 10	. Salinity, nitrate, dissolved oxygen and chlorophyll concentrations at the Peconic River CR105 Bridge - USGS water quality sonde data, May-June, 2015. Yellow bars indicate time of fish kills

# **Appendices**

Appendix I.	SC Dep't. of Health Services fish kill sampling results	.28
Appendix II.	SC Dep't. of Health Services recreation advisory	.31
Appendix III.	Cornell University College of Veterinary Medicine	
	pathology report	.32

#### Abstract

This investigation was undertaken to identify factors that may have contributed to three fish kill events involving adult Atlantic Menhaden (*Brevoortia tyrannus*), that occurred in the tidal portion of the Peconic River during the spring of 2015. In a collaborative multiagency effort, extensive monitoring was conducted to document existing physical, chemical and biological conditions in the river that may have been associated with the events. Data on historical water quality conditions as well as past fish kills was also examined to provide possible clues to the occurrences.

Staff from the Suffolk County Department of Health Services (SCDHS) collected samples for the analysis of standard water quality parameters as well as constituents that may be toxic if present in sufficient quantities (organic solvents, pesticides and radiological residues) from a number of sites in and adjacent to the river. Additionally, sanitary conditions at the closest bathing beach (South Jamesport Beach) were periodically evaluated to insure that public health impacts from dead and decaying fish were not occurring, and an advisory issued to warn those recreating throughout the impacted area of potential conditions. Researchers from the School of Marine and Atmospheric Science (SoMAS) at Stony Brook University analyzed water samples to characterize the phytoplankton (micro-algae) community in the river and to determine if any harmful algal blooms (HABs) were impacting the fish. Staff from the New York State Department of Environmental Conservation (NYSDEC) recorded observations and collected moribund menhaden from several locations for pathological examination, and communicated with local commercial fisherman regarding conditions in and around the Peconic River.

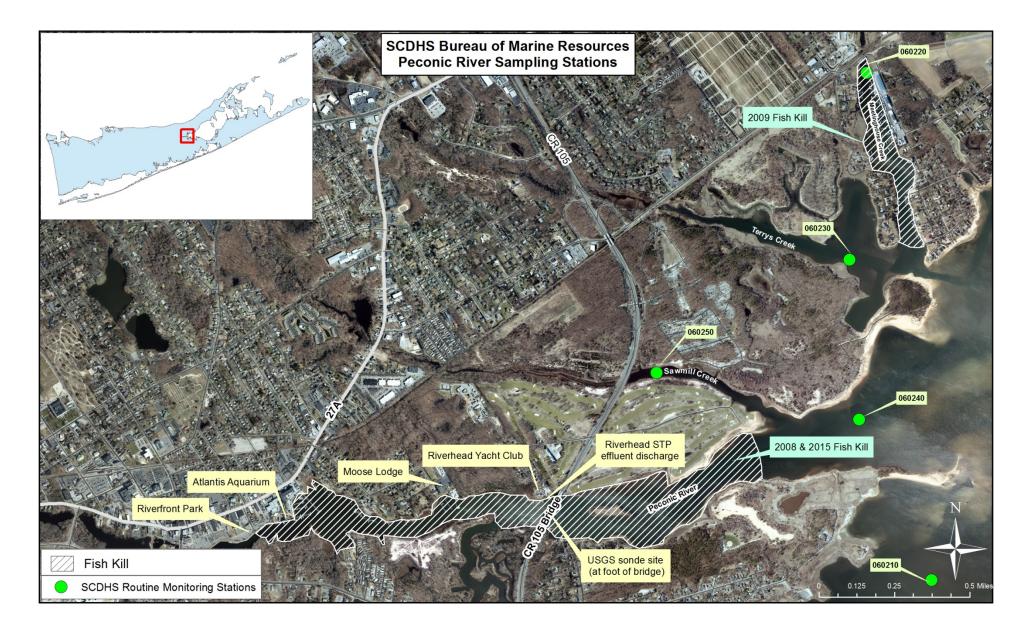
The weight of findings suggests that rapidly rising water temperature, the timing and magnitude of algal blooms and an unusually large biomass of adult menhaden confined in the river, were all contributing factors that resulted in prolonged periods of extremely low dissolved oxygen (DO) levels and ultimately caused large numbers of the menhaden to expire. Results for other water quality measures (nutrients, bacteria, VOCs, pesticides and radiological constituents), showed results within normal ranges,

discounting the possibility that illegal discharges, spills and/or the presence of toxic substances were contributing factors. On a local scale, the presence of HAB species known to be ichthyotoxic may have been an important factor acting in combination with declining DO levels. The HAB *Gymnodinium instriatum* was also present during all three fish kill events and during the initial 16-May fish kill, two HABs (*Prorocentrum minimum* and *Karlodinium veneficum*) were noted in the river immediately preceding the event. The effect of the HABs was also evidenced by the pathology report for the fish specimens collected, which indicated the fish died of asphyxiation but also suggested that gill damage due to exposure to harmful algae was a likely contributing factor.

The Peconic River has a long history of degraded water quality, particularly with respect to nitrogen inputs, algal blooms and diminished DO concentrations. What seems to have made 2015 different from other years, is that spring algal blooms were more intense and the oxygen decline occurred much earlier than usual. Only two other years since 2003 showed similar early oxygen declines, 2008 and 2009, both years when major menhaden fish kills also occurred. Explanations for the enhanced blooms in 2015 are not apparent in the monitoring data collected, but what can be certain is that given the current state of eutrophication in the river, algal blooms and diminished oxygen levels will continue to be the norm. If the waters are warm enough for anoxia to develop and a body of fish are present, another fish kill is likely to occur.

### Introduction

A series of fish kills involving Atlantic Menhaden (*Brevoortia tyrannus*) occurred in the tidal portion of the Peconic River (Figure 1) during May and June of this year. Masses of dead and dying fish were reported in various locations along the river, with areas of highest concentrations varying from the mouth of the river off of Indian Island County Park, to the waters between the Suffolk County Route 105 (CR105) Bridge and the Riverhead Yacht Club. The initial kill occurred in the area of the Atlantis Aquarium on 16-May, and was reported to be comparatively minor in terms of numbers. This event was followed on 27-May by a major fish kill involving what was estimated to be 100,000s (hundreds of





thousands) of menhaden. A third, also relatively minor menhaden kill (estimated at 10,000s fish), followed on 14-June. Causes of the kills were quickly identified by researchers and involved agencies as low oxygen levels in the river. An ongoing algal bloom (*Prorocentrum*), increasing water temperatures and limited tidal flushing were also noted to be associated factors.

#### Causes of Fish Kills

Fish kills can occur for a variety of reasons, including insufficient DO, extreme water temperatures, sudden changes in water temperature and/or salinity, the discharge or spill of a toxic substance, the presence of diseases, parasites or harmful algal blooms, or from bodily injury. In the literature, past studies involving menhaden fish kills have noted predation, parasites, disease and low DO as potential causes (Smith, 1999).In laboratory experiments, menhaden showed 100% mortality in 2-6 hours at 0.6 mg/L DO, with large fish found to be less tolerant to hypoxia than small fish (Shimps, 2003). Other studies have shown that many fish, including menhaden, will actively avoid hypoxic waters if an escape route is available (Wannamaker and Rice, 2000). Of the menhaden kills that have occurred in the Peconic River over the past two decades, including major kills in September of 1999 (estimated at 1-3 million fish), August of 2000 (~750,000 fish), May of 2008 (100,000s) and May of 2009 (100,000s), low DO was identified as the main contributing factor. For the 2009 event, results of pathological examinations done on fish samples submitted to the Aquatic Diseases and Immunology Lab at SUNY Stony Brook, suggested that reduced fitness from parasitism and damage from either predation or avoidance behavior that reduced the animals' ability to escape the event, were possible secondary factors (M. Fast, email communication).

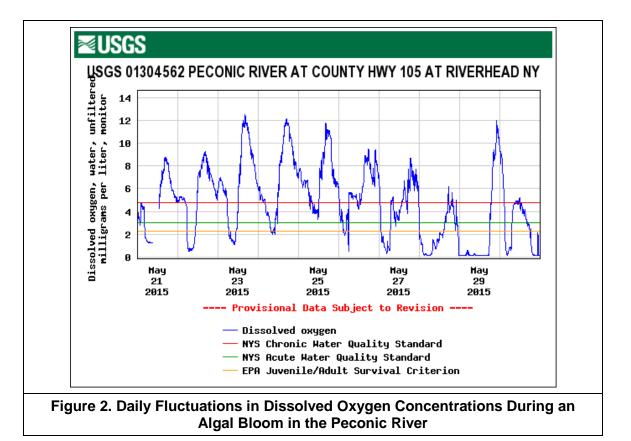
Low oxygen levels in surface waters may be associated with a number of factors, several of which periodically exist in portions of the lower Peconic River:

 Rising water temperatures – because the solubility of gasses in water (including oxygen) decreases with increasing temperature, DO levels tend to decline as waters warm up in the spring and summer.

- Algal blooms although algae produce an excess of oxygen during the day through the process of photosynthesis, they consume oxygen during the night through respiration. This results in a day-night cycle of fluctuating DO, as is shown in the USGS graphic below (Figure 2).
- Excess nitrogen although not a direct cause of low DO, anthropogenic nitrogen loading from point and non-point sources, including septic systems, residential and agricultural fertilizers, sewage treatment plant effluents and atmospheric deposition, stimulates the algal production that subsequently acts to diminish oxygen levels. A spike in nitrate was noted prior the late May and mid-June fish kills.
- Excessive biochemical oxygen demand (BOD) the organic compounds contained in wastewater discharges, decaying algal blooms, and sediments washed into surface waters during heavy rainfall events, all exert an oxygen demand as they are decomposed by aerobic bacteria. In a study of the Neuse River Estuary in North Carolina, Paerl et al. (1998) noted that hypoxic events can be triggered in a matter of days by increased organic matter contained in stormwater runoff.
- Sediment oxygen demand (SOD) oxygen consumed by sediment microorganisms can affect water column DO levels in areas subject to significant organic deposition. Previous studies of benthic fluxes in the Peconic Estuary noted high levels of SOD in areas in the western portion of the system, including the lower Peconic River, Reeves Bay and Meetinghouse Creek (Howes et al., 1998). The highly organic nature of the upper sediments in these areas is the legacy of the many duck farms that once operated there, as well as the current wastewater discharges from the Riverhead STP and Atlantis Aquarium, and residues contained in stormwater runoff from roads and parking lots in the adjacent downtown Riverhead area.

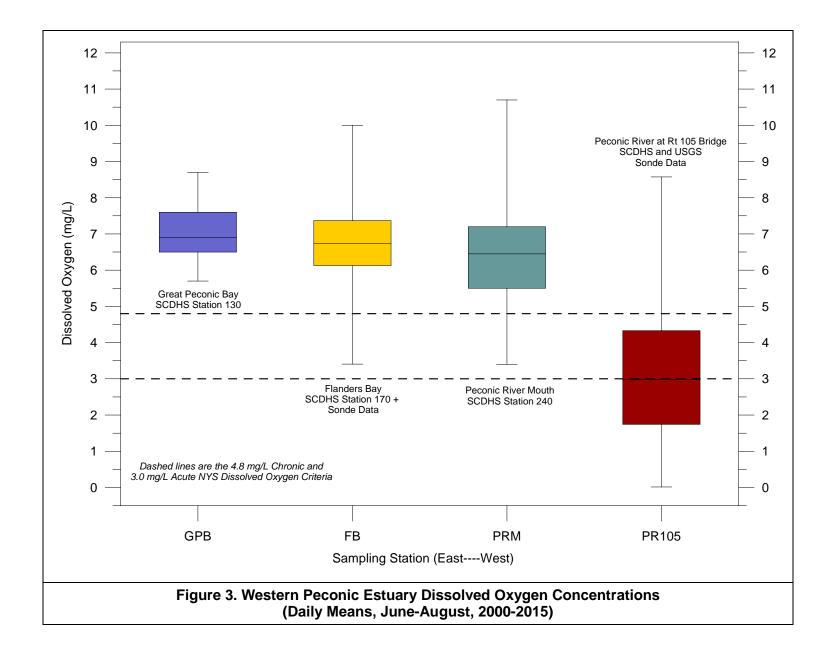
### Historic Water Quality

Although DO levels throughout much of the Peconic Estuary have historically been very



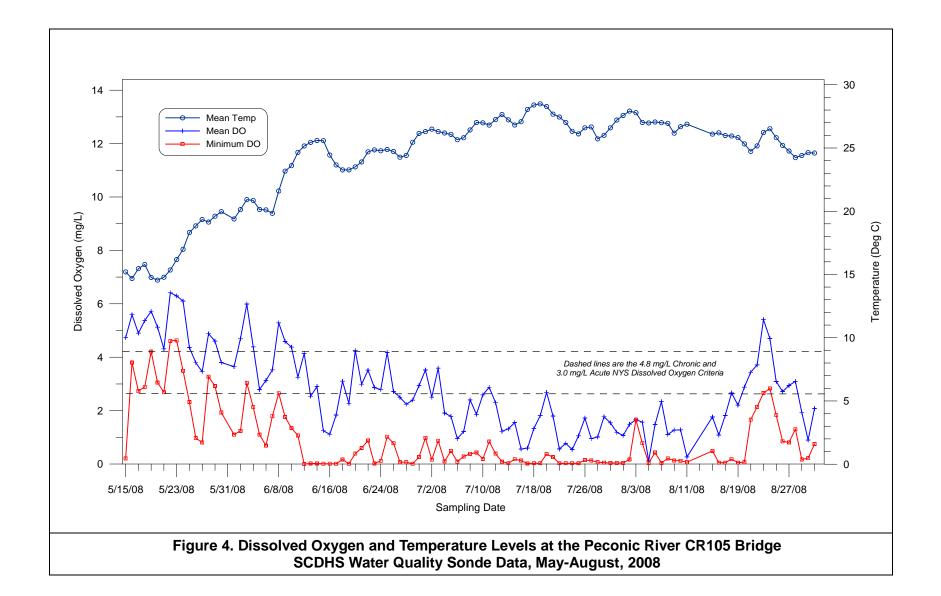
good, a number of sites in the western portion of the estuary, including the tidal portion of the Peconic River, have experienced periodic excursions below acceptable DO criteria (CCMP, 2001). These DO declines have been attributed to excessive algal growth that is associated with increased nitrogen loading. The box-plot graphic in Figure 3, depicting results of past DO monitoring done in the Peconic Estuary, illustrates an east to west pattern of declining DO concentrations as well as the significant degree of oxygen stresses that is characteristic of the CR105 Bridge location.

An examination of historic results from continuous monitoring water quality sondes deployed at the bridge by the SCDHS (2003 through 2011), similarly illustrates the magnitude of depressed oxygen levels in the river. The data consistently show oxygen levels declining rapidly through the spring as water temperatures increase, with mean levels during summer months rarely above the 4.8 mg/L NYS chronic DO criteria and



frequently below the 3.0 mg/L acute criteria (Table 1). As an example of a year when oxygen levels were particularly stressed in the river, Figure 4 shows the DO and temperature time series for 2008 (May-August), a year when a major fish kill also occurred in the river. As the graphic illustrates, from June through much of August, daily DO means were often in the hypoxic range (<2.0 mg/L) with episodes of anoxia (no oxygen) a frequent occurrence. An extended period during which daily oxygen minima were at or near anoxic levels started in mid-June, a typical occurrence for the site although unusually early compared to other years. A similar pattern was evident in 2009, another year when a major menhaden fish kill occurred. Conditions showed signs of improvement in 2011 (Table 1), when the number of days DO means were below the 3.0 mg/l NYS criteria and the 2.0 mg/l hypoxic benchmark, as well as the number of samples near anoxic levels, declined considerably. More recent data collected by the USGS at the bridge (site #01304562, data provisional), showed the level of summer hypoxia continuing to improve during 2013 and 2014.

	The Percen W	% Results — Near Anoxi		
Year	< 4.8 mg/L	< 3.0 mg/L	< 2.0 mg/L	(< 0.5 mg/L
2003	75	42	24	14
2004	76	61	38	13
2005	80	55	37	19
2006	98	63	35	10
2007	87	40	24	16
2008	97	73	46	16
2009	99	75	42	19
2010	97	74	52	20
2011	71	28	16	4
2013	70	30	8	0.2
2014	67	16	9	0.4
2015	79	28	17	8



### The 2015 Peconic River Fish Kills

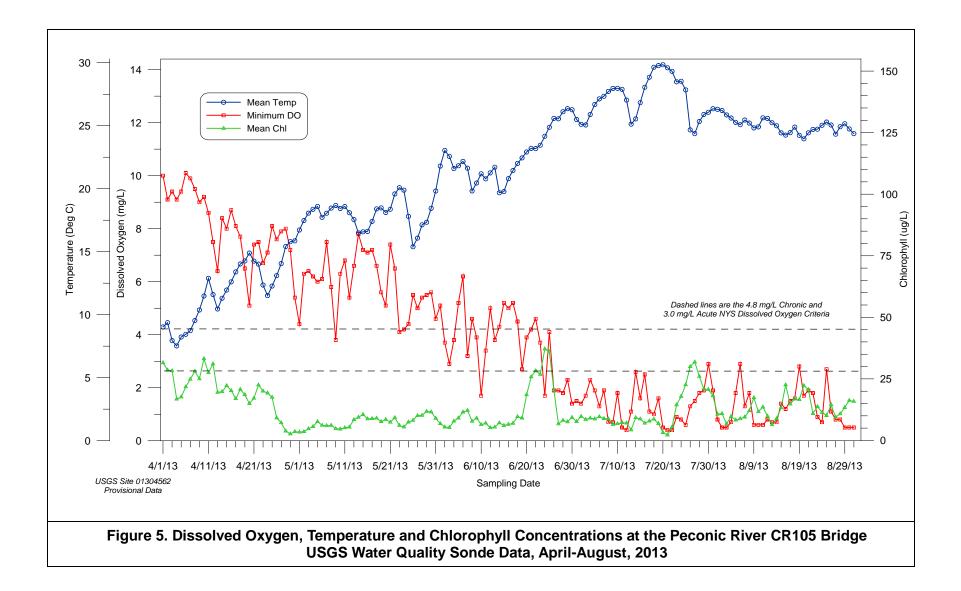
In an effort to identify factors that may have contributed to the 2015 Peconic River fish kills, and in response to public health concerns regarding the possible involvement of toxic substances, the SCDHS Office of Ecology initiated water quality monitoring at a number of sites in the Peconic River and in neighboring creeks and embayments in the days following the 27-May event. In addition to five locations in the river, sites monitored included Meetinghouse Creek, Terrys Creek, Sawmill Creek, Reeves Bay, Flanders Bay, East Creek and the Riverhead STP discharge (Figure 1). Sampling parameters included standard analytes such as salinity, temperature, DO, coliform bacteria, nitrogen and phosphorus nutrients and chlorophyll-a, as well as constituents that may be toxic if present in sufficient quantities, such as organic solvents (VOCs), pesticides and radiological residues. At the South Jamesport bathing beach, located approximately four miles to the east of the river, sanitary surveys and bacteriological sampling to evaluate beach water quality were also conducted immediately following the 27-May fish kill. Results from this sampling, as well as routine monitoring since the fish kill, has displayed very good water quality at the beach. In addition, the beach operator was instructed to keep the beach free of dead fish carcasses if any washed ashore. All available sampling are included in Appendix I. A copy of a recreational advisory issued by the Suffolk County Department of Health Services is included in Appendix II.

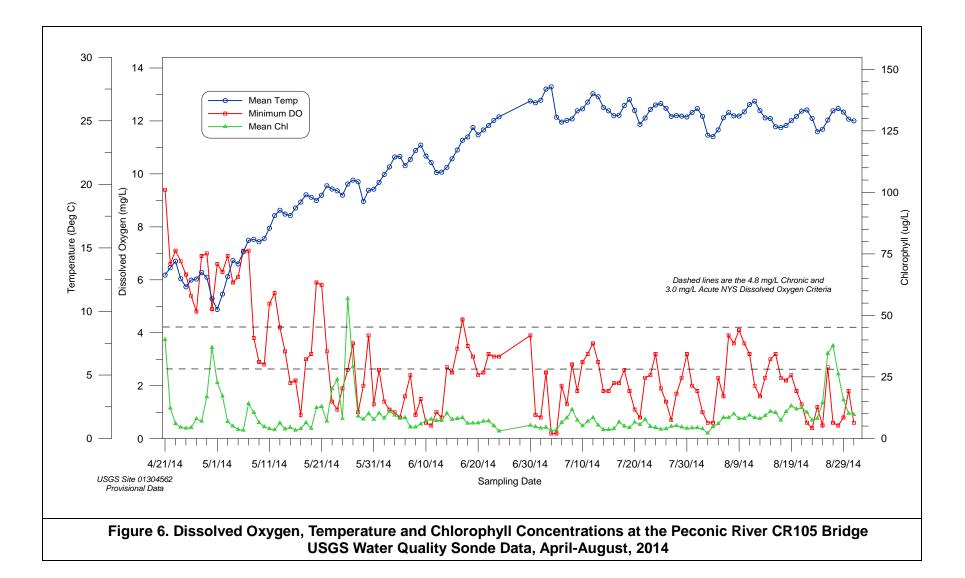
Supporting initial theories that low DO was the main contributing cause of the fish kills, results of samples collected in early June showed near-anoxic DO levels at mid-depth and bottom waters of the river extending from the Moose Lodge to Riverfront Park (Figure 1), with those at the CR105 bridge and nearby Sawmill and Meetinghouse Creeks at or marginally above hypoxic levels. Concentrations in better flushed waters to the east (the Peconic River mouth, Reeves Bay and Flanders Bay) were much improved (although less than ideal) in the 6.0-6.5 mg/L range. Subsequent monitoring done following the 14-June fish kill, similarly pointed to oxygen stress as the main contributing factor. Samples collected between 15-June and 17-June showed hypoxic to near anoxic DO levels present from the CR105 Bridge to the Atlantis Aquarium. Results for other water quality measures (nutrients, bacteria, volatile organic compounds, pesticides and

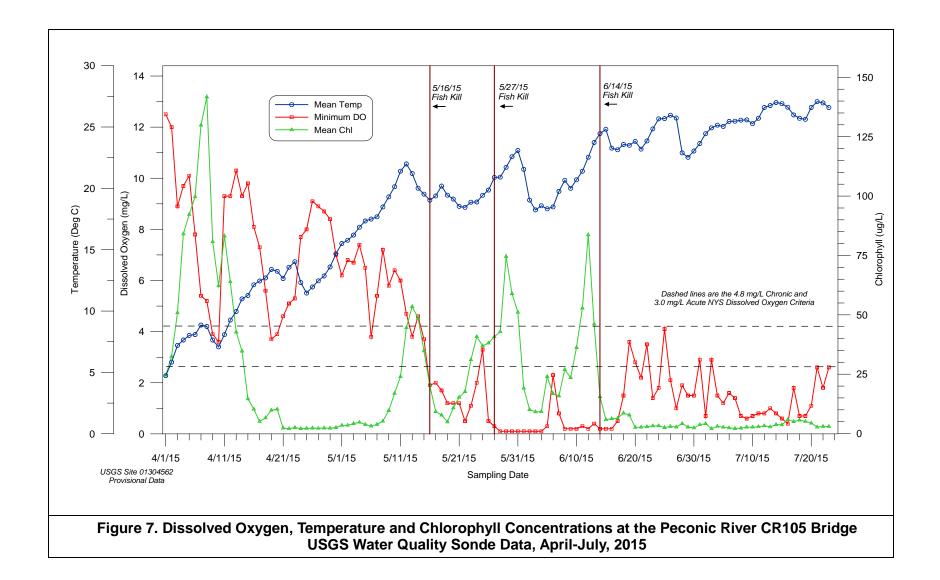
radiological constituents), showed results within normal ranges, discounting the possibility that illegal discharges, spills and/or the presence of toxic substances were contributing factors.

A closer look at recent sonde data (2013-2015) collected by the USGS at the CR105 Bridge site, similarly implicates low DO as a main contributing factor in the fish kills and suggests that water temperature in addition to the timing and magnitude of algal blooms (as indicated by chlorophyll levels) are key factors in oxygen dynamics in the river. As is shown in the time-series plots in Figures 5 to 7, a series of spring algal blooms occurred during each of the three years represented. During 2013, blooms with daily mean chlorophyll levels peaking in the 25-35 ug/L range, occurred in early April when water temperatures were relatively cool (10-15°C). Hypoxia didn't occur until 10-June, when temperatures had risen to ~23°C. After a bloom in late June, when water temperatures were in the 25-30°C range, daily minimum DO levels rarely rose above 3.0 mg/L, were frequently hypoxic (<2 mg/L) and occasionally approached anoxia. Had fish been present, conditions from June through August of 2013 were ideal for a fish kill.

During 2014, algal blooms occurred periodically from late April through May, with mean chlorophyll levels peaking in the 35-60 ug/L range (Figure 6). Daily oxygen minima fluctuated above and below hypoxic levels through August, but sustained periods of near-anoxia did not develop and no fish kills were reported. During the following year (2015) however, conditions were much different. An algal bloom with daily mean chlorophyll levels peaking at >125 ug/L, persisted for two weeks in early April (Figure 7). Oxygen minima subsequently showed periods of precipitous decline but quickly rebounded as water temperatures were still cool (5-10°C). In the following weeks, a rapid increase in temperature coincided with an algal bloom (>50 ug/L chlorophyll) that lasted approximately ten days (9-May to 19-May) and resulted in oxygen minima falling into the hypoxic range for the first time that year (on 16-May), the same day the initial 2015 menhaden fish kill was reported. Over the next three week period (late May through mid-June), a series of intense algal blooms occurred (with daily mean chlorophyll levels peaking at >75 ug/L) that resulted in daily periods of extended anoxia and the second and third fish kills.

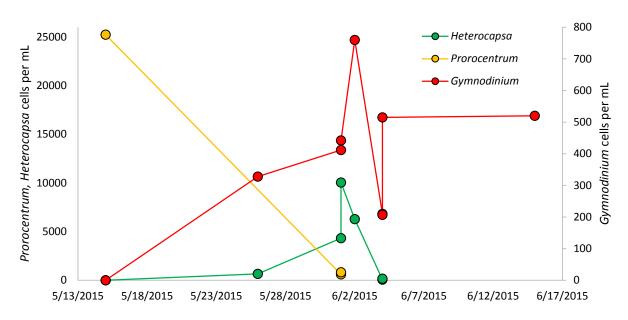






While phytoplankton are a pivotal component of the marine food web, algal blooms can be both harmful to humans through the production of shellfish poisoning toxins and/or considered ecologically destructive by disrupting ecosystem function (Sunda et al., 2006). During the fish kills that occurred in the Peconic River estuary a series of what can be considered ecologically destructive blooms were present in the water column, namely blooms of Prorocentrum spp., Gymnodinium instriatum, Heterocapsa spp., and Karlodinium veneficum, with maximal densities reaching 25,000, 760, 8,200, and 1,290 cells mL<sup>-1</sup>, respectively, during May through June of 2015 (Fig. 8, Table 2). Heterosigma akashiwo, an ichthyotoxic raphidophyte known to cause death of fish from asphyxiation via an undefined mechanism (Cochlan et al., 2013), was a smaller component of the phytoplankton community reaching >2,000 cells mL<sup>-1</sup> on 2-June and was not associated, in appreciable abundances, with any of these fish kills. Prorocentrum *minimum*, or mahogany tide, has been associated with fish kills in the Chesapeake Bay estuary at concentrations >10<sup>4</sup> cells mL<sup>-1</sup> with the Maryland Department of Natural Resources defining >3000 cells mL<sup>-1</sup> as a "threshold above which living resource" are impacted (Tango et al., 2005). While these blooms are typically considered ecologically destructive due to the association with hypoxic events and fish kills, there is also recent evidence for potential toxin production by *P. minimum*, specifically neurotoxins (Vlamis et al., 2015). The dinoflagellate Karlodinium veneficum and its associated toxins, karlotoxins, which have hemolytic and cytotoxic properties, and whose mechanism of action is disrupting gas transport across the gills of fish, has long been implicated in fish kills at densities >10<sup>4</sup> cells mL<sup>-1</sup> (Place et al., 2012). To our knowledge, however, Gymnodinium instriatum and Heterocapsa rotundata do not produce toxins but can contribute to biological oxygen demand at night and upon decay of high biomass blooms and have been associated with prior fish kills (Heil et al., 2001; Wang et al., 2005).

The first fish kill which occurred on 16-May was associated with a dense (>25,000 cells  $mL^{-1}$ ) *Prorocentrum minimum* and *Karlodinium veneficum* (>1000 cells  $mL^{-1}$ ) bloom (Fig. 8, Table 2). During this time mean DO levels dropped to <5mg L<sup>-1</sup> from >7 mg L<sup>-1</sup> just



Densities of dinoflagellates in the Peconic River, May, June 2015

Figure 8. Maximal densities (cells mL<sup>-1</sup>) of the most abundant dinoflagellates in the lower Peconic River region during May - June of 2015.

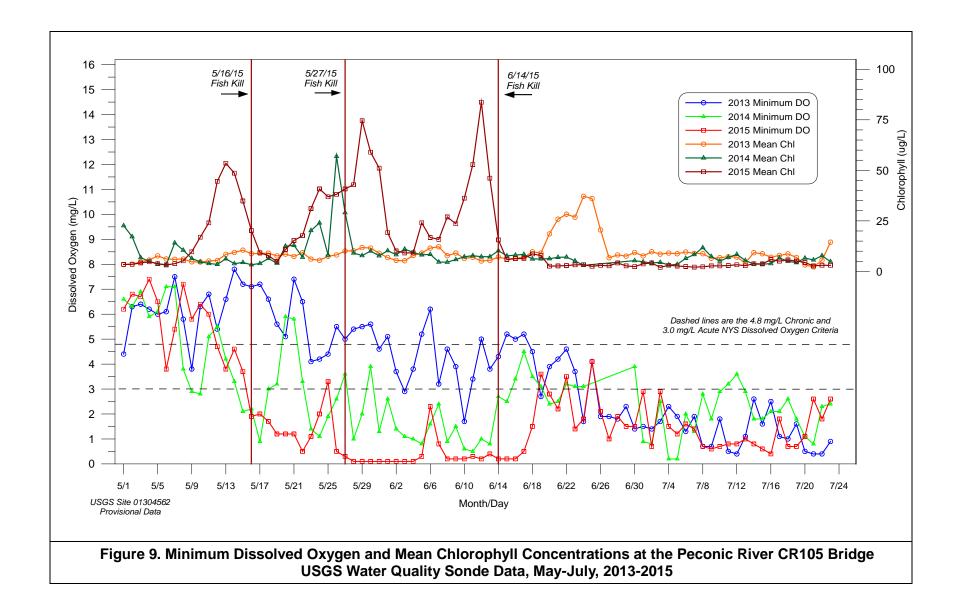
Table 2. Densities of the most abundant phytoplankton species (groups) found in the water
column in the Peconic River region during May - June of 2015.

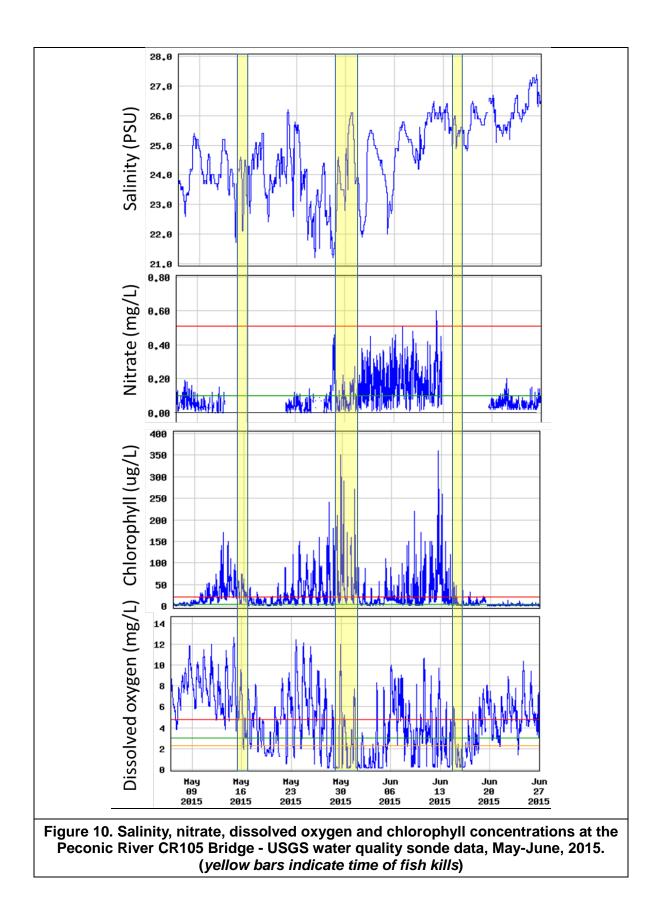
		Diatoms	Oxyrrhis	Prorocentrum	Gymnodinium	Mesodinium	Heterocapsa	Heterocapsa	Heterosigma	Karlodinium	Prorocentrum	Other
Date	Site		marina	minimum	instriatum		rotundata	arctica	akashiwo	veneficum	gracile	Dinoflagellates
		(cells mL <sup>-1</sup> )										
15-May-15	Peconic River			25240						1290		
15-May-15	Meetinghouse Creek	1520		745				1120				1680
18-May-15	Meetinghouse Creek	2920	9720	10360								1240
1-Jun-15	Moosehead Lodge	840	5520	616	412	880	2680	1650				2640
1-Jun-15	105 Bridge	1760	1760	820	442	1270	8200	1840				1240
1-Jun-15	Aquarium	1080	1120	522		246	642	488				760
2-Jun-15	Moosehead Lodge Surface	202	560		158		1620					381
2-Jun-15	Moosehead Lodge Mid	228	1205		67	212	480					515
2-Jun-15	105 Bridge Surface	1120			760		6280					520
2-Jun-15	105 Bridge Mid	524	316		242		1525					585
2-Jun-15	Aquarium Surface	143										7
2-Jun-15	Aquarium Mid	160	428				482		2840			19
4-Jun-15	Moosehead Lodge Surface	167	141		210	72	68					133
4-Jun-15	Moosehead Lodge Mid	282	1020		207	86	163					264
4-Jun-15	105 Bridge Surface	1720	76		49	94						83
4-Jun-15	105 Bridge Mid	4280	85		422	153						127
4-Jun-15	Aquarium Surface	193										13
4-Jun-15	Aquarium Mid	186	436		515	94						224
15-Jun-15	105 Bridge		1620		520							
26-Jun-15	105 Bridge									6680	2120	1360

prior to the fish kill while minimum DO levels dropped from  $<4mg L^{-1}$  to  $<2 mg L^{-1}$  (Figure 7). *P. minimum* densities were within the range of those known to be associated with fish kills. Two weeks later a larger and more extended fish kill occurred from 27-May through 4-June in the same region. By this time the phytoplankton community

shifted (samples from 1-June) and was comprised mostly of Prorocentrum minimum (>500 cells mL<sup>-1</sup>), Gymnodinium instriatum (>400 cells mL<sup>-1</sup>), Heterocapsa rotundata (<10,000 cells mL<sup>-1</sup>), Heterocapsa arctica (>500 cells mL<sup>-1</sup>), and Oxyrrhis marina (>1000 cells mL<sup>-1</sup>), while lower densities of *Heterosigma akashiwo* and *Chattonella* spp. were also present (Fig. 8, Table 2). Data collected from a USGS probe deployed in the Peconic River showed that while chlorophyll a levels were already elevated (>50µgL<sup>-1</sup>; 27-May), they increased dramatically (>350  $\mu$ gL<sup>-1</sup>) following a spike in nitrate concentrations (Fig. 10). Just prior to the fish kill, mean DO levels were 6 mg L<sup>-1</sup> while the DO minimum approached 0 mg  $L^{-1}$  (Fig. 7) with both of these values declining even further post 27-May likely due to a combination factors including increasing temperatures and increased water column oxygen demand due to decaying fish, and the dense algal blooms present. While these algal blooms occasionally pushed daytime DO concentrations above 5mg L<sup>-1</sup> in most cases daytime concentrations were <5mg L<sup>-1</sup> and nighttime DO concentrations were  $<2mg L^{-1}$  and often times close to 0 mg L<sup>-1</sup>, demonstrating that high phytoplankton biomass also contributed (respired) towards a larger nighttime oxygen demand (Fig. 10). Phytoplankton samples taken in response to the fish kill that occurred in the Peconic River on 14-June demonstrated that the phytoplankton community was dominated by Gymnodinium instriatum and the heterotrophic dinoflagellate, Oxyrrhis marina (Fig. 8, Table 2). Again, fish were exposed to a combination of factors, including increasing temperatures, minimum DO levels that were <1 mg L<sup>-1</sup> for extended periods, increased oxygen demand (both water column and sediment) due to a decaying algal bloom (peaked on 12-June; Fig. 7) and the presence of heterotrophic dinoflagellates that likely acted to exacerbate the oxygen demand, especially at night.

To better illustrate water quality conditions in the Peconic River that were likely associated with the 2015 fish kills, and to accentuate the impact that algal blooms can have of DO levels in the river, Figure 9 presents an overlay of the daily mean chlorophyll concentrations and daily minimum DO levels at the CR105 Bridge site for May-July of the last three years (2013-2015). As can be seen from the graphic, 2015 was unusual in that a series of algal blooms (as represented by chlorophyll levels) occurred during





the spring, resulting in an extended period where daily DO minima were near-anoxic. By the time the system began to recover (around 20-June) three fish kills had occurred.

Conditions that are conducive to fish and other kills have existed in the river for some time: an ample nutrient supply leading to a succession of algal blooms, subsequent stresses on oxygen levels as the blooms respire and die, and organics in the sediments and water column exerting an additional oxygen demand. As temperatures increased in the spring, all that was needed was a large body of panic-driven menhaden trapped by predators to finish off whatever oxygen was left.

Indeed, an unusually large body of adult menhaden was reported by commercial fishermen to enter the Peconic Estuary at the end of April, 2015, followed almost immediately by large schools of bluefish ranging in size from 5 to 15 pounds. On 7-May, NYSDEC observations in the Peconic River revealed surface activity indicative of the presence of large numbers of menhaden from Riverfront Park in downtown Riverhead to just east of Colonels Island (S. Heins, personal observation). Based on the sheer volume of large adult menhaden within the confines of the River, there was speculation among NYSDEC staff that a mass kill could occur later in the year if the fish did not leave the area. Bluefish feeding activity from the mouth of the River and Flanders Bay was also reported by commercial fishermen during that time period. On 29-May, two days into the second, largest kill, NYSDEC observed bluefish feeding on live menhaden at the edge of a floating mass of dead fish just west of the mouth of Sawmill Creek. This bluefish activity was clearly preventing menhaden from escaping the River. Dead and dying menhaden were estimated to number over 200,000 fish.

#### Fish Pathology

At the time of the fish kills in the Peconic River, other kills of adult menhaden were being reported throughout the marine district of New York. Kills were also being reported from the Hudson River, New Jersey, Connecticut and Rhode Island. It was commonly reported by witnesses that some of the dying fish exhibited a "spinning" behavior at the water's surface. Connecticut DEEP released a statement saying they believed the

deaths in their area were caused by a virus and referred to it as "whirling disease." It was later learned that DEEP had not sampled the dying fish, but were relying on descriptions of fish behavior in published literature (Stephens et. al., 1980). In response, NYSDEC collected moribund menhaden for pathological examination from several locations, including the Peconic River and Meetinghouse Creek. At the time of the collections (6/10), the fish in the Peconic River were not exhibiting the "spinning" behavior that had been previously noted and reported from other kill sites. They were swimming lethargically, but managed to evade capture when approached. Only a single specimen was obtained from the River, so the remaining samples were obtained from Meetinghouse Creek where dying fish were exhibiting the "spinning" behavior (J. Maniscalco, personal observation) and easily collected. Collected specimens were shipped on ice overnight to the Aquatic Animal Health Program at Cornell University's College of Veterinary Medicine. A preliminary pathology report is attached (Appendix III).

The pathology report stated that the fish died of asphyxiation, also showing excessive mucus in the gills and damage to the gill lamellae (see Appendix III), likely from exposure to harmful algae described previously. The evidence supports the diagnosis provided earlier in this document. In addition, viral pathology yielded positive results, though the virus has yet to be identified. Similar results were obtained in samples from the Hudson River and Manhasset Bay. It is unlikely the virus had any major role in the Peconic River fish kills, as the hypoxia/harmful algal blooms combination would explain 100% of the mortalities in this case. NYSDEC will continue to investigate the nature of the virus infection

### <u>Summary</u>

- A series of fish kills involving Atlantic Menhaden (*Brevoortia tyrannus*) occurred in the tidal portion of the Peconic River (Figure 1) during May and June of 2015.
- Masses of dead and dying fish were reported in various locations along the river, with areas of highest concentrations varying from the mouth of the river near In-

dian Island County Park to the waters between the CR105 Bridge and the Riverhead Yacht Club.

- At least three separate fish kills were reported to have occurred, with comparatively minor events noted on 16-May and 14-Jun (10,000s of fish) and a major event on 27-May that involved an estimated 200,000 fish.
- Based on real-time data recorded by a water quality probe deployed at the CR105 Bridge by the USGS, low levels of DO in the river was identified as the primary cause of the kills. Factors that were likely associated with the events included rising water temperatures, limited tidal flushing and the coincident occurrence of a succession of algal blooms.
- Dips in salinity coincident with a spike in nitrate in late May and a steady rise in nitrate in early June suggest that the delivery of nitrate via a pulse of riverine or run-off may have intensified algal blooms that thus contributed to low DO (Figure 10).
- To provided data on other potential contributing factors, and in response to public health concerns regarding the possible involvement of toxic substances, staff from the SCDHS Office of Ecology initiated water quality monitoring at a number of sites in the river as well as in neighboring creeks and embayments. In addition to standard water quality parameters, samples were collected for constituents that may be toxic if present in sufficient quantities, including organic solvents (VOCs), pesticides and radiological residues.
- As a precaution, bacteriological monitoring of the only nearby bathing beach (South Jamesport Beach) was conducted as well, and an advisory issued to warn those recreating throughout the impacted area of potential conditions.
- Additional sampling done by researchers from the School of Marine and Atmospheric Science (SoMAS) at Stony Brook University provided key data on the identification and temporal variations of phytoplankton (micro-algae) at a number of locations in the river, including species that are potentially toxic to fish.
- To evaluate the potential role of diseases as a contributing factor in the fish kills, staff from the New York State Department of Environmental Conservation

(NYSDEC) collected moribund menhaden from several locations for pathological examination by staff from Cornell University's College of Veterinary Medicine.

- Results of SCDHS sampling done proximate to the 27-May and 14-Jun fish kills found hypoxic to near anoxic conditions existing throughout much of the lower river, echoing the initial conditions noted at the CR105 deployment site.
- Samples for other water quality measures (nutrients, bacteria, volatile organic compounds, pesticides and radiological constituents), showed results within normal ranges, discounting the possibility that illegal discharges, spills and/or the presence of toxic substances were contributing factors.
- Analysis of phytoplankton samples by the SoMAS lab revealed that a series of "ecologically destructive" algal blooms, some involving species that are potentially ichthiotoxic, were present in the water column during the fish kills.
- Algal blooms act to diminish dissolved oxygen levels through nighttime respiration and via the oxygen demand exerted by decaying cells as the bloom declines. The association of algal blooms with excess nitrogen inputs, and subsequently depressed DO levels, has been well documented for areas of the western estuary, including portions of the lower Peconic River and adjacent creeks.
- The initial fish kill on 16-May was associated with a dense bloom of two algal species, *Prorocentrum minimum* and *Karlodinium veneficum*, both known to cause fish kills when present in sufficient quantities. As such, it is plausible that this fish kill occurred due to the combined effects of bloom induced hypoxia and the toxicity of the involved species.
- Subsequent blooms, involving multiple algal species, increased in intensity through mid-June and periodically rose to dramatic densities (as indicated by chlorophyll-a levels >350 ug/L at the USGS probe).
- As a result, the set of environmental conditions that developed in the river, including elevated water temperature and an increasing level of oxygen demand from nighttime algal respiration as well as water column and sediment decay processes, quickly led to frequent periods of hypoxia and extended periods where minimum DO levels were near-anoxic.

- At the same time, large numbers of adult menhaden were present in the river, their escape blocked by aggressively feeding bluefish. The activity of the panicdriven fish eventually depleted the already low DO levels, resulting in massive menhaden mortality on 27-May.
- Pathological analyses of fish specimens confirmed the cause of death as asphyxiation, and noted that effects from harmful algal toxicity and possibly an unidentified virus, were possible secondary contributing factors.

## Conclusions

The primary cause of the fish kills in the Peconic River was asphyxiation, as a large school of menhaden, trapped in the river by predator bluefish, consumed what was left of an already diminished oxygen supply. The presence of toxic algae may have been a contributing factor, particularly in the initial 16-May fish kill, with the role of an unidentified virus still uncertain.

The low DO levels found in the river resulted from multiple factors acting in combination, including rising temperatures, elevated nitrogen inputs, a succession of algal blooms, the oxygen demand exerted by increased biological activity in the water column and sediments, and a limited degree of tidal flushing. The Peconic River has a long history of degraded water quality, particularly with respect to nitrogen inputs. The enriched nature of the river has enabled various algal species to flourish, has promoted the growth of a number of opportunistic harmful algae and has been responsible for periodic excursions of DO concentrations below standard criteria. What made 2015 different from previous years, was the timing of the oxygen decline. As historical data (2003-2011) collected by the SCDHS at the CR105 Bridge indicates, prolonged periods where DO minima approach anoxic levels typically doesn't occur in the river until sometime in July. In 2015, it happened during the last week in May when large numbers of adult menhaden were present. The only other years since 2003 when the onset of extended anoxia occurred prior to July (in 2008 and 2009 it occurred in mid-June), were also the only years when major menhaden fish kills occurred.

Although chlorophyll data for the 2003-2011 period isn't available, data collected by the USGS during the last three years (2013-2015) illustrate that the magnitude and timing of algal blooms in relation to water temperature, are key factors in the DO dynamics in the river. Unlike 2013 and 2014, a succession of major algal blooms occurred during the spring of 2015 that were undoubtedly associated with the early onset of hypoxia in the river. What was different about the water quality in 2015 that enabled the blooms to proliferate is uncertain. Data for the Riverhead STP discharge, located directly adjacent to the CR105 Bridge, showed nitrogen levels for April through June that were well within permit specifications, and routine monitoring data collected at nearby creeks and embayments by the SCDHS (under the Peconic Estuary Program) similarly did not suggest any anomalies.

What can be certain however, is that given the current state of eutrophication in the river, algal blooms and diminished oxygen levels will continue to be the norm. If the waters are warm enough for anoxia to develop and a body of fish are present, another fish kill is likely to occur.

## <u>References</u>

CCMP, 2001. Peconic Estuary Program Comprehensive Conservation and Management Plan.

Cochlan, W.P., Trainer, V.L., Trick, C.G., Wells, M.L., Eberhart, B.-T., Bill, B.D., 2013. *Heterosigma akashiwo* in the Salish Sea: defining growth and toxicity leading to fish kills., Proceedings of the 15th International Conference on Harmful Algae.

Fast, M. 2008. Aquatic Diseases and Immunology, School of Marine and Atmospheric Sciences, SUNY Stony Brook, N.Y. Email communication.

Heil, C. A., Glibert, P. M., Al-Sarawl, M. A., Faraj, M., Behbehani, M., & Husain, M. (2001). First record of a fish-killing Gymnodinium sp bloom in Kuwait Bay, Arabian Sea: chronology and potential causes. *Marine Ecology-Progress Series*, *214*, 15.

Howes, B.L., D.R. Schlezinger, N.P. Millham, G. Hampson, D.D. Goehringer and S. Aubrey. 1998. Oxygen uptake and nutrient regeneration in the Peconic Estuary. Final report to the Suffolk County Dep't. of Health Services. 21 pp.

Paerl, H.W., J.L. Pinckney, J.M. Fear and B.L. Peierls. 1998. Ecosystem responses to internal and watershed organic matter loading: consequences for hypoxia in the eutrophying Neuse River Estuary, North Carolina, USA. Marine Ecology Progress Series 166: 17-25.

Paerl, H.W., J.L. Pinckney, J.M. Fear, and B.L. Peierls. 1999. Fish kills and bottomwater hypoxia in the Neuse River and Estuary: reply to Burkholder et al. Marine Ecology Progress Series 186: 307-309.

Place, A.R., Bowers, H.A., Bachvaroff, T.R., Adolf, J.E., Deeds, J.R., Sheng, J., 2012. *Karlodinium veneficum*- The little dinoflagellate with a big bite. Harmful Algae 14, 179-195.

Shimps, E.L. 2003. Hypoxia tolerance in two juvenile estuary dependent fishes. Graduate thesis, North Carolina State University.

Smith, J.W. 1999. A large fish kill of Atlantic Menhaden, Brevoortia tyrannus on the North Carolina coast. Journal of the Elisha Mitchell Scientific Society 115(3): 157-163.

Stephens, E.B., M.W. Newman, A.L. Zachary & F.M. Hetrick, 1980. A viral aetiology for the annual spring epizootics of Atlantic menhaden Brevoortia tyrannus (Latrobe) in Chesapeake Bay. Journal of Fish Diseases 1980 (3) 387-398.

Sunda, W.G., Graneli, E., Gobler, C.J., 2006. Positive feedback and the development and persistence of ecosystem disruptive algal blooms. J. Phycol. 42(5), 963-974.

Tango, P., Magnien, R., Butler, W., Luckett, C., Luckenbach, M., Lacouture, R., Poukish, C., 2005. Impacts and potential effects due to *Prorocentrum minimum* blooms in Chesapeake Bay. Harmful Algae 4, 525-531.

Wang, Q., Deeds, J. R., Place, A.R., & Belas, R. (2005). Dinoflagellate community analysis of a fish kill using denaturing gradient gel electrophoresis. *Harmful Algae*, *4*(1), 151-162.

Wannamaker, C.M. and J.A. Rice. 2000. Effects of hypoxia on movements and behavior of estuarine species. Journal of Experimental Marine Biology and Ecology 249(2): 145-163.

Vlamis, A., Katikou, P., Rodriguez, I., Rey, V., Alfonso, A., Papazachariou, A., Zacharaki, T., Botana, A., Botana, L., 2015. First Detection of Tetrodotoxin in Greek Shellfish by UPLC-MS/MS Potentially Linked to the Presence of the Dinoflagellate Prorocentrum minimum. Toxins 7(5), 1779.

Location	Date Col- lected	S/M/B <sup>1</sup>	Station No.	Temp (C)	DO (mg/L)	Total Coli- form (mpn/100 ml)	Fecal Coli- form (mpn/100 ml)	TN (mg/L)	NH3-N (mg/L)	NOx-N (mg/L)	Organics <sup>2</sup>	Gross Alpha	Gross Beta	Tritium
	6/2/2015	S	PR-105	17.9	3.5									
Peconic River at	6/2/2015	М	PR-105	17.8	3.1									
Rt. 105 Bridge	6/16/2015	S	PR-105	23.6	2.2									
	6/16/2015	В	PR-105	23.1	2.8									
	6/2/2015	S	PR-AQ	16.7	4.8									
Peconic River at	6/2/2015	М	PR-AQ	21.3	< 0.05									
Atlantis Aquarium	6/16/2015	S	PR-AQ	23.2	1.2									
	6/16/2015	В	PR-AQ	23.9	0.2									
	6/2/2015	S	PR-ML	17.5	3.5									
Peconic River at	6/2/2015	М	PR-ML	20.1	< 0.05									
Moose Lodge	6/16/2015	S	PR-ML	23.5	0.9									
	6/16/2015	В	PR-ML	23.4	0.1									
	6/1/2015	S	PR-RFP	19.5	5.5	1,100	800	0.68			ND	ND	12.2 <u>+</u> 0.6	ND
Peconic River at	6/1/2015	В	PR-RFP	22.2	0.2									
Riverfront Park	6/15/2015	S	PR-RFP	22.3	5.6									
	6/15/2015	В	PR-RFP	23.2	7.0									
	6/15/2015	S	PR-RYC	22.5	1.1									
Peconic River at Riverhead YC	6/15/2015	В	PR-RYC	22.5	1.0									
	6/16/2015	S	PR-RYC	23.4	1.1									
	6/16/2015	В	PR-RYC	23.2	0.8									
Riverhead STP Discharge	6/17/2015		200-009			800	< 20	15.8	9.95					

<sup>1</sup> Sample location: Surface (S), Mid-depth (M) or Bottom (B) <sup>2</sup> Organic parameters included numerous volatile organic compounds (VOCs) and pesticides ND = "Not Detected"

Location	Date Col- lected	S/M/B <sup>1</sup>	Station No.	Temp (C)	DO (mg/L)	Total Coli- form (mpn/100 ml)	Fecal Coli- form (mpn/100 ml)	TN (mg/L)	NH3-N (mg/L)	NOx-N (mg/L)	Organics <sup>2</sup>	Gross Alpha	Gross Beta	Tritium
	5/27/2015	S	060-240	20.5	7.2	< 20	< 20	0.51	< 0.02	< 0.005				
	5/27/2015	В	060-240	20.2	7.6									
	5/31/2015	S	060-240	24.4	12.5	80	40	1.23			ND			
	6/2/2015	S	060-240	17.8	5.9	40	40	0.74	0.10	0.012	ND	ND	65.0 <u>+</u> 9.2	ND
<b>D</b> . D.	6/2/2015	В	060-240	17.6	6.0									
Peconic River Mouth	6/4/2015	S	060-240	18.1	10.5	< 20	< 20	0.85	0.05	0.020	ND			
	6/4/2015	В	060-240	18.3	8.4									
	6/16/2015	S	060-240	23.3	8.0									
	6/16/2015	В	060-240	22.7	5.2									
	6/17/2015	S	060-240	23.1	7.2	40	< 20	0.42	< 0.02	< 0.005				
	6/17/2015	В	060-240	23.1	7.1									
	5/27/2015	S	060-220	21.0	2.6	40	40	1.60	0.46	0.723				
	5/27/2015	В	060-220	20.7	2.1									
	5/31/2015	S	060-220	23.0	9.2	210	110	3.64			ND			
	5/31/2015	В	060-220	22.7	3.6									
	6/1/2015	S	060-220	23.0	5.1	220	220	0.93			ND			
Meetinghouse	6/1/2015	В	060-220	23.0	2.3									
Creek	6/2/2015	S	060-220	20.0	1.1	20	20	1.02	0.30	0.190	ND			
	6/2/2015	В	060-220	20.1	1.2									
	6/4/2015	S	060-220	19.0	6.6	300	170	3.56	0.83	3.42	ND			
	6/4/2015	В	060-220	19.0	5.2									
	6/17/2015	S	060-220	23.3	3.1	20	< 20	0.80	0.24	0.177				
	6/17/2015	В	060-220	23.0	3.9									

Location	Date Col- lected	S/M/B <sup>1</sup>	Station No.	Temp (C)	DO (mg/L)	Total Coli- form (mpn/100 ml)	Fecal Coli- form (mpn/100 ml)	TN (mg/L)	NH3-N (mg/L)	NOx-N (mg/L)	Organics <sup>2</sup>	Gross Alpha	Gross Beta	Tritium
	6/2/2015	S	060-230	18.1	6.7	90	90	0.73	< 0.02	0.007				
Tarmia Croak	6/2/2015	В	060-230	18.2	5.0									
Terry's Creek	6/16/2015	S	060-230	24.0	9.0									
	6/16/2015	В	060-230	22.7	4.7									
Sawmill Creek	6/2/2015	S	060-250	17.8	2.0	2,400	1,300	1.25	0.39	0.145				
Sawmin Creek	6/2/2015	В	060-250	19.0	0.7									
Flanders Bay	6/2/2015	S	060-170	18.0	6.3	< 20	< 20	0.37	0.02	< 0.005				
Fianuers bay	6/2/2015	В	060-170	18.1	6.2									
	6/1/2015	S	060-210	19.6	6.6	< 20	< 20	0.59			ND			
	6/1/2015	В	060-210	19.8	6.4									
	6/2/2015	S	060-210	17.3	6.6	< 20	< 20	0.50	< 0.02	< 0.005	ND			
Reeves Bay	6/2/2015	В	060-210	17.3	6.4									
	6/4/2015	S	060-210	17.0	12.8	< 20	< 20	0.53	< 0.02	< 0.005	ND			
	6/4/2015	В	060-210	17.0	12.6									
	5/27/2015	S	060-101	20.1	5.0	140	80	0.84	0.29	0.231				
	5/27/2015	В	060-101	19.6	4.6									
	6/2/2015	S	060-101	17.6	4.5	80	80	0.60	0.19	0.061				
	6/2/2015	В	060-101	17.6	4.6									
East Creek (So. Jamesport)	6/15/2015	S	060-101	22.6	6.0									
	6/15/2015	В	060-101	22.7	4.5									
	6/17/2015	S	060-101	21.8	3.2	20	20	0.62	0.18	0.123				
	6/17/2015	В	060-101	21.8	3.3									
	6/19/2015	S	060-101			40	40	0.98	0.10	0.250				
South Jamesport Beach	6/19/2015	S	R14			< 20	< 20							

# Health Officials Issue Peconic River Recreation Advisory

#### Department: Health Services | Posted: 6/18/2015 |

Following the amassing of thousands of dead bunker fish on the shores of the Peconic River on May 29, 2015 and June 14, 2015, health officials are warning residents and visitors who choose to recreate near these waters to follow some common-sense recommendations.

#### Swimming/Bathing

Swim only at regulated bathing beaches. Regulated beaches are monitored and usually safe for swimming. When the waters at any regulated beach reveal the presence of bacteria at levels that exceed New York State standards, the department closes that beach. Beaches that are not permitted for swimming are not monitored by the department and the waters may be unsafe for swimming. The status of regulated bathing beaches can be found at the beach program web page http://gis2.suffolkcountyny.gov/bathingbeaches/

#### Wading, Fishing, Boating, Kayaking, Canoeing

Recreating in water, even on a raft or boat, poses some potential for the skin and face to come into contact with water that may contain bacteria, parasites and other microorganisms. If you are exposed to water that may be unsafe, you can help protect yourself by following the advice below:

- · Avoid water with accumulations of dead fish.
- Avoid recreating in cloudy or discolored water, as it may contain more microorganisms that might make people sick and affect a person's ability to see underwater hazards.
- Don't swallow water and keep your face and head out of the water.-This reduces exposure to bacteria, parasites, and other microorganisms that might make people sick by entering the body by swallowing, and through eyes, ears and nose.
- Wash your hands when you leave the water and before eating. Do not touch your eyes, nose or mouth before washing your hands.
- Shower as soon as you are finished with your activities for the day.

#### **Contact with Dead Fish**

Do not handle or eat fish that are found dead, dying, acting abnormal or seem sick. If you must handle dead or decaying matter, make sure your hands are covered with disposable nitrile, rubber or plastic protective gloves or a plastic bag before touching the fish. If your skin is exposed to the dead fish, wash your hands thoroughly with soap and water. If you accidentally ingest any decaying matter, seek medical attention immediately.

#### Eating Fish Caught from Waters Where the Dead Fish were Found

Fish can be contaminated with bacteria, viruses or parasites that can cause illness. It is difficult to determine the risks from eating live fish caught from areas where there are large masses of dead fish. If you have caught a live fish and choose to eat it, be sure to cook the fish thoroughly to kill bacteria, parasites and other microorganisms, as is always good practice.

suffolkcountyny.gov

Facebook.com/SuffolkCountyHealthServices

Twitter.com/SuffolkCoHealth

## Appendix III.



# Cornell University

# Aquatic Animal Health Program

Dept. of Microbiology and Immunology College of Veterinary Medicine Cornell University Ithaca, NY 14853-6401 Tel: (607) 253-4028 Fax: (607) 253-3384

Case number:	FPL2015-011	<b>Report Date:</b>	7/2/2015
Date received:	6/11/2015	<b>Diagnosticians:</b>	Marquis, Sams, Getchell
<b>Client Name:</b>	Steve Heins	Type of sample:	3 whole fish
		Species:	Menhaden (Brevoortia tyrannus)

**History:** An unusual number of fish kills involving primarily Atlantic menhaden ("bunker") have been reported from around Long Island, NY; CT and RI. Following detection of a virus in bunker from the Hudson River, we attempted to collect distressed fish from Peconic River/Flanders Bay. Two large kills had occurred and it is possible that conditions were right for another. Initial belief has been that these large kills are due to low DO. Bottom water DO was very low (1-2 mg/l) on morning of collection but surface water was significantly higher. Note: the marine dinoflagellate Akashiwo sanguinea was present in large numbers from plankton tows conducted in the Peconic River concurrent w/ fish collection on 6/10/2015. Submitted samples were collected on 6/10/15 from Peconic River/Flanders Bay by John Maniscalco NYSDEC BMR. Water temperature at time of collection (12:15 PM) was 20.6 C with a DO of 7.21 ppm at the surface of Meetinghouse Creek (23.21 ppt salinity). Bottom water data from Meetinghouse Creek was 19.98 C, 25.69 ppt salinity, and 6.3 ppm DO). Specimen number 1 was dropped by an osprey and recovered by DEC staff still alive near the 105 bridge on Peconic Bay. Other fish were seen acting strangely (swimming alone, slowly, at surface) but still capable of avoiding capture by dipnet from the boat. Specimens 2&3 were dipnetted from Meetinghouse Creek by DEC staff in a tight space against the bulkhead. Fish were at surface with gills flared. No "whirling" was seen. Large schools of fish were seen behaving normally, large numbers of large bluefish were also present.

**Presentation:** Three fish were delivered on ice to Cornell AAHP on 11 June 2015.

**Gross examination:** The menhaden ranged in size from 295 mm to 333 mm in length and weighed from 270 grams to 321 grams (fish #1-#3). The condition of the fish were good. The gills all had a thick mucus layer and detritus adhering to this mucus (see photo).

External and internal gross pathological lesions of fish #1 were as follows:

Fish #1 had ecchymosis around the vent, and left operculum and a puncture wound on right side presumably from osprey (see photo). Internally fish #1 had hemorrhagic ascites, erythema of pyloric caeca, and hemorrhagic brain. No obvious parasites were observed in any of the fish. Fish #2 had a slight hemorrhage in the left eye and caudal fin, as well as a puncture wound on left operculum, (see photo). Internally there was hemorrhagic ascites, darkened, inflamed, possibly necrotic pyloric ceca and intestines and dark green/black liquid for stomach contents. The brain, stomach, gonads were also hemorrhagic. Fish #3 had similar internal appearance with less hemorrhagic gonads.

**<u>Histological examination:</u>** Two of the sets of gill filaments appeared normal, while the third set had significant necrosis present (see photos). Severe congestion of red blood cells was observed in the liver, brain, and heart (see photos) suggesting possible anoxia. Hemorrhaging was noted in the posterior kidney (see photo). The inner walls of the pyloric cecae appeared necrotic (see photo).

#### Laboratory results:

**Bacteriology:** Kidney loop samples were inoculated onto TSA/5%SB and marine agar. No significant growth after 14 days incubation was observed.

Toxicology: None performed.

**Virology:** Viral isolation was performed with CHSE, EPC, KF1, FHM and BF-2 cell lines. Filtered homogenates were prepared from pooled tissues (kidney, spleen, heart) or from the brain, and tissues collected on 6/10/15 were prepared separately. These tissue homogenates were used to inoculate cells. Cytopathic effects were observed in CHSE (see photo), KF1, and BF-2 cells inoculated with the filtered homogenate from pooled tissues, but not from the brain. These results are suggestive of viral replication. Further work is underway to identify these isolates including EM and genome sequencing.

**Diagnosis:** Viral infection may have contributed to these die-offs, but clearly the predator induced anoxia that was measured during these events and the algal blooms that were documented played a bigger role in these die-offs.

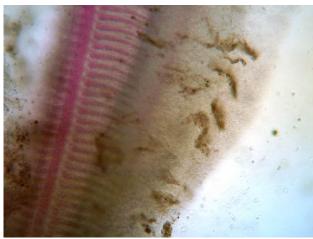
**Comments:** The histology slides were similar to those from Manhasset Bay (Case FPL2015-010). Pathological changes were again evident in multiple tissues including the pyloric ceca, and the gills from one of these specimens. The severity of the fish kills may be due to the cumulative effects of all three of these stressors on these menhaden schools, anoxia, algal blooms, and viral infection.

### Images:

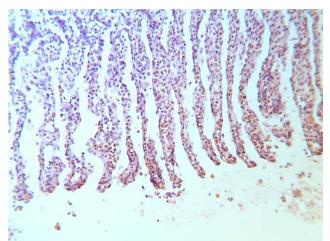


Fish #1 with some hemorrhaging of the fins.

Fish #2 with hemorrhages in eye.



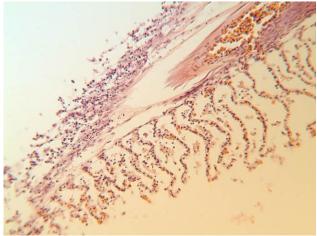
Gills with mucus and detritus



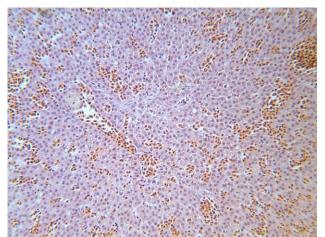
Menhaden gills (H&E 25X).



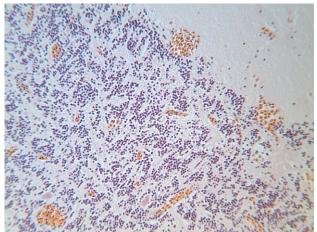
Menhaden gills (H&E 40X).



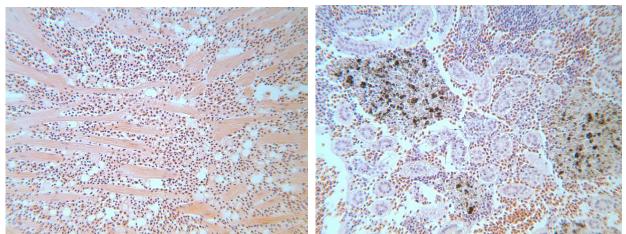
Menhaden gills with necrosis (H&E 25X).



Red blood cell congestion in liver (H&E 25X)

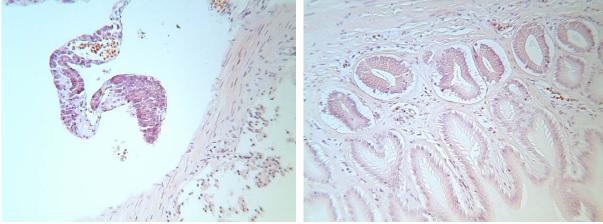


Red blood cell congestion in brain congestion

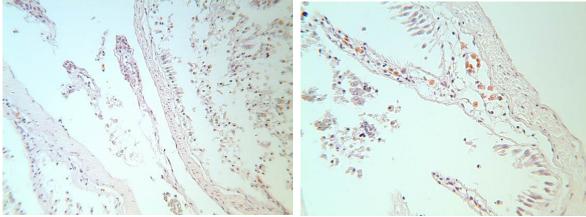


Red blood cell congestion in heart (H&E 25X)

Posterior kidney hemorrhages (H&E 25X)



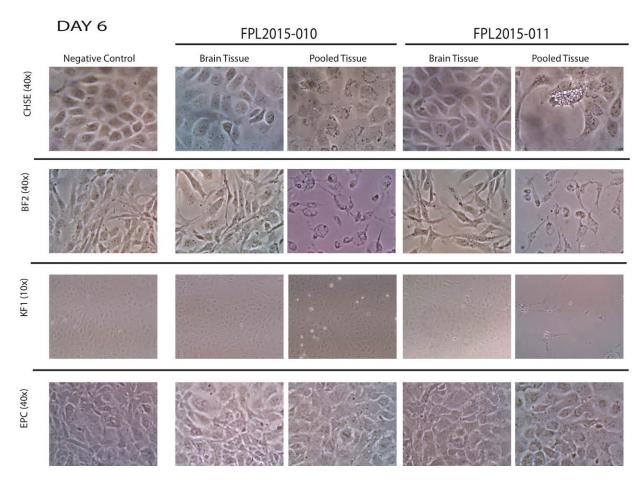
Pancreas with zymogen granule depletion (25X) Menhaden intestine (H&E 25X)



Pyloric ceca necrosis (H&E 25X)

Pyloric ceca necrosis (H&E 40X)

Panel of Viral CPE images from FPL2015 010 and 011



Helene Marquis, DVM PhD Professor

Rod Getchell, PhD Research Scientist

Kelly Sams, Technician