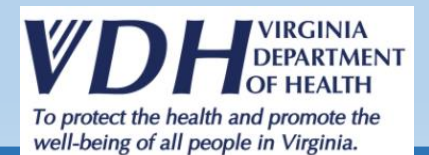


2022 Virginia estuarine HABs: marine biotoxins update

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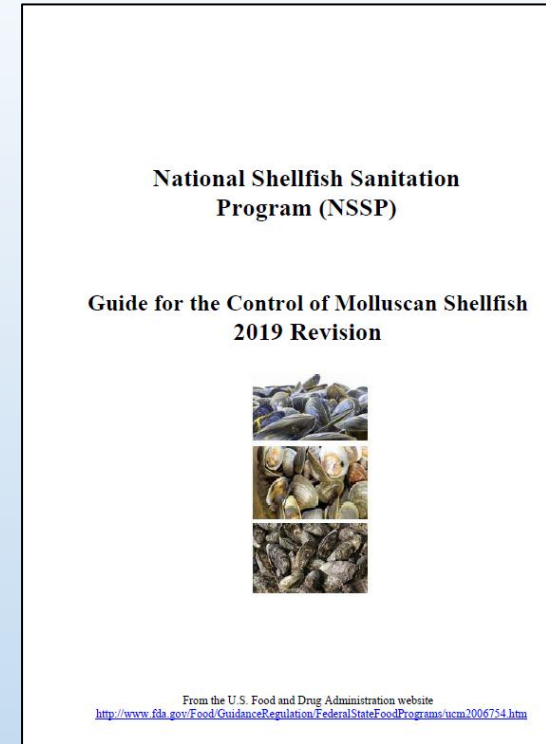
VA HAB Taskforce Meeting
Zoom-VIMS
2/24/2023



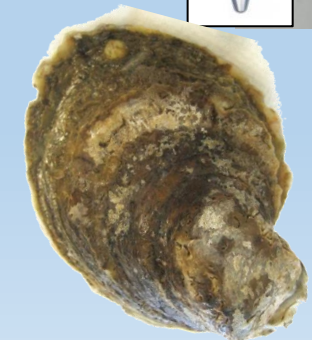
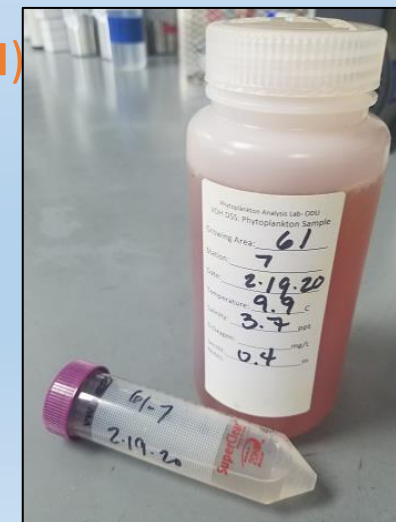
www.SwimHealthyVA.com www.vdh.virginia.gov/shellfish

Shellfish marine biotoxin control

- Biotoxin contingency plan for:
 - Paralytic shellfish poisoning (PSP)
 - Amnesic shellfish poisoning (ASP)
 - Neurotoxic shellfish poisoning (NSP)
 - Diarrhetic shellfish poisoning (DSP)
 - Azaspiracid shellfish poisoning (AZP)
- VDH:DSS Biotoxin plan and flow chart:
<http://www.vdh.virginia.gov/content/uploads/sites/20/2016/05/BiotoxinControlPlan.pdf>
- Monthly collections- routine fixed sites
 - Lugol's solution (250mL) – phytoplankton analyses (ODU)
 - Screened at VDH field offices
 - **Unpreserved frozen sample (50mL)- targeted ELISA screening (VDH)**
 - Unpreserved frozen filter sample (100mL, 3µm)- qPCR (VIMS) (all samples/stations)
- Bloom samples
 - Response to bloom reports or visual observation by field staff
- **Oyster Deployments:**
 - **Expanded since 2019- (VDH)**



Phyto Kit: Extra bottles, vials, lugol's, rubber gloves, marker



2022

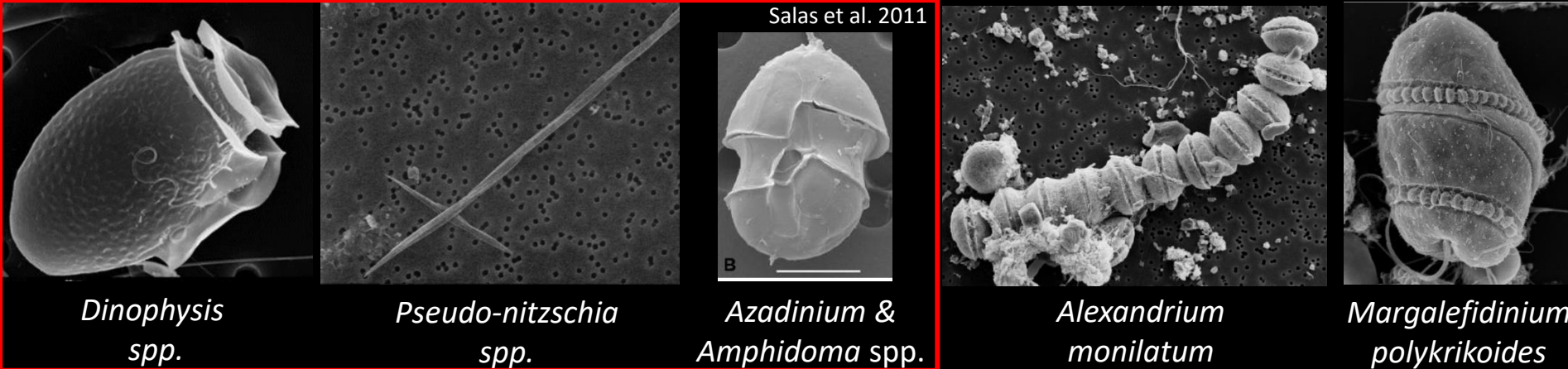
Updates

- Year-round sampling qPCR analyses of all DSSWH collections for marine Biotoxin producers DSP, ASP, AZP

*Differentiating thick and thin *Pseudo-nitzschia* +/- 5µm width

- Expanded routine seawater toxin ELISAs**

- Additional shellfish deployments and expanded sites**



Dinophysis
spp.

Pseudo-nitzschia
spp.

Azadinium &
Amphidoma spp.

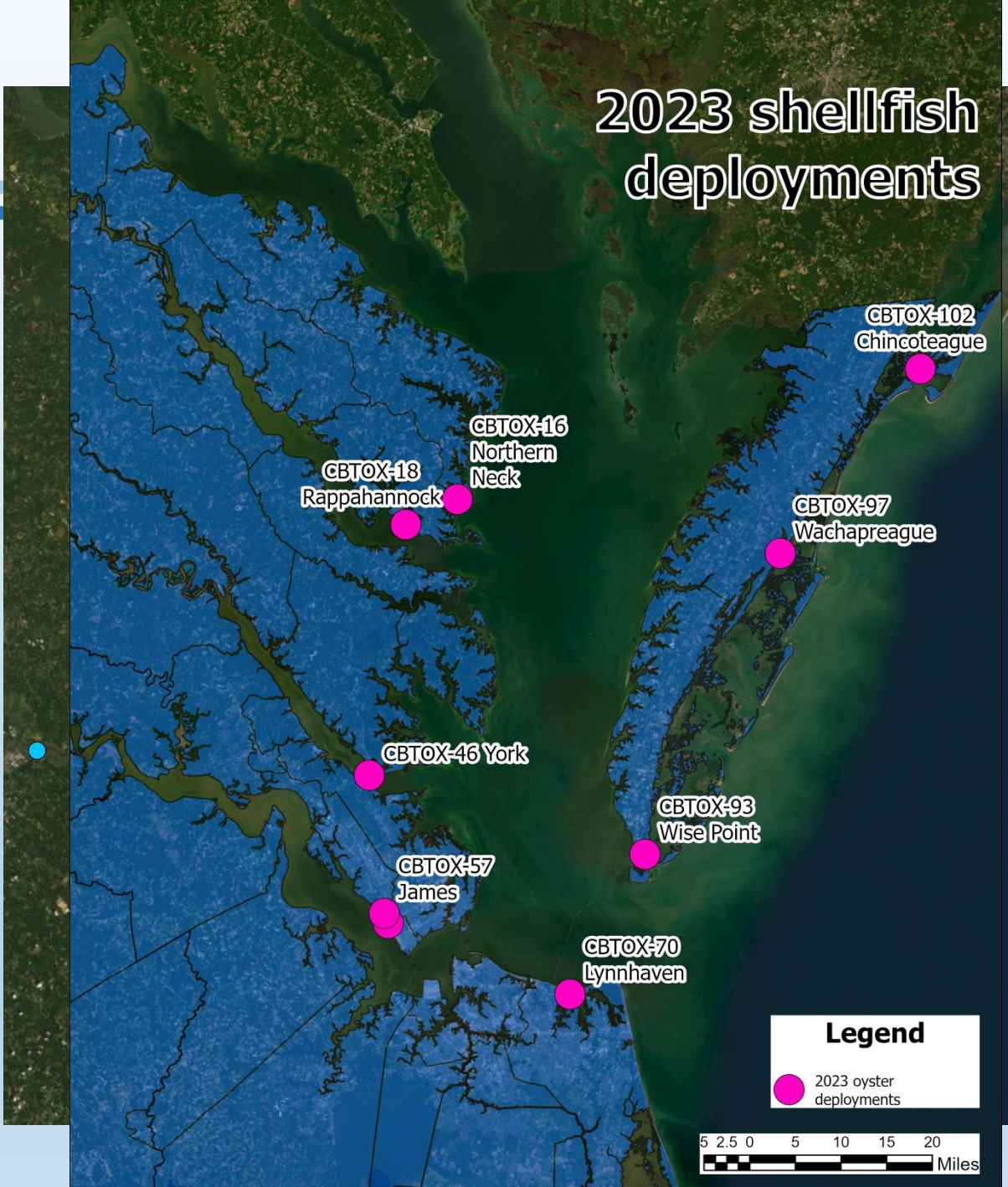
Alexandrium
monilatum

Margalefidinium
polykrikoides

Algal species	Impacts	Main Toxin	NSSP shellfish growing area closure level (toxin w/in meat)	working regional bloom density (cell density in water column)
<i>Alexandrium tamarense</i> species complex	Paralytic Shellfish Poisoning	Saxitoxin	80µg /100g	presence
<i>Karenia brevis</i>	Neurotoxic Shellfish Poisoning	Brevetoxin	0.8mg /kg	presence
<i>Dinophysis</i> spp.	Diarrhetic Shellfish Poisoning	Okadaic acid	0.16 mg/kg	≥5 cells/ml
<i>Pseudo-nitzschia</i> spp.	Amnesic Shellfish Poisoning	Domoic acid	2mg/100g	* ≥ 1,000 cells/ml
<i>Azadinium & Amphidoma</i> spp	Azspiracid Shellfish Poisoning	Azspiracid-a	0.16 mg/kg	TBD
<i>Alexandrium monilatum</i>	Fish/invertebrate mortality	Goniodomin A	NA	≥ 1,000 cells/ml
<i>Margalefidinium polykrikoides</i>	Fish/invertebrate mortality	icthyotoxin	NA	≥ 1,000 cells/ml
<i>Karlodinium veneficum</i>	Fish mortality	Karlotoxins	NA	≥ 10,000 cells/ml

Virginia Estuarine Phytoplankton monitoring

2023 shellfish deployments



- Chesapeake Bay Monitoring Program (DEQ/ODU)
 - 14 stations
 - 7-Chesapeake Bay monthly year-round
 - 7-Tidal tributaries monthly March-October
 - Full species composition

- VDH: Shellfish (DSSWH/ ODU/VIMS)
 - 69 stations
 - Monthly year-round
 - Targeted HAB identification
 - Targeted toxin screening (based on cell counts)
 - Targeted qPCR analyses-all samples

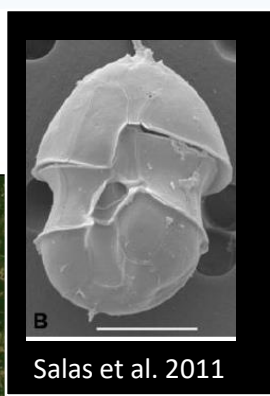


- CBTOX (VDH:DSS/ VIMS)
 - 12 stations (2017-2018)
 - 4 stations (2019-2020)
 - 7 stations (2021-2022)
 - 8 stations (2023)**
 - Bi-weekly/ monthly sampling
 - Full species composition
 - Routine toxin analyses – **oyster deployments**



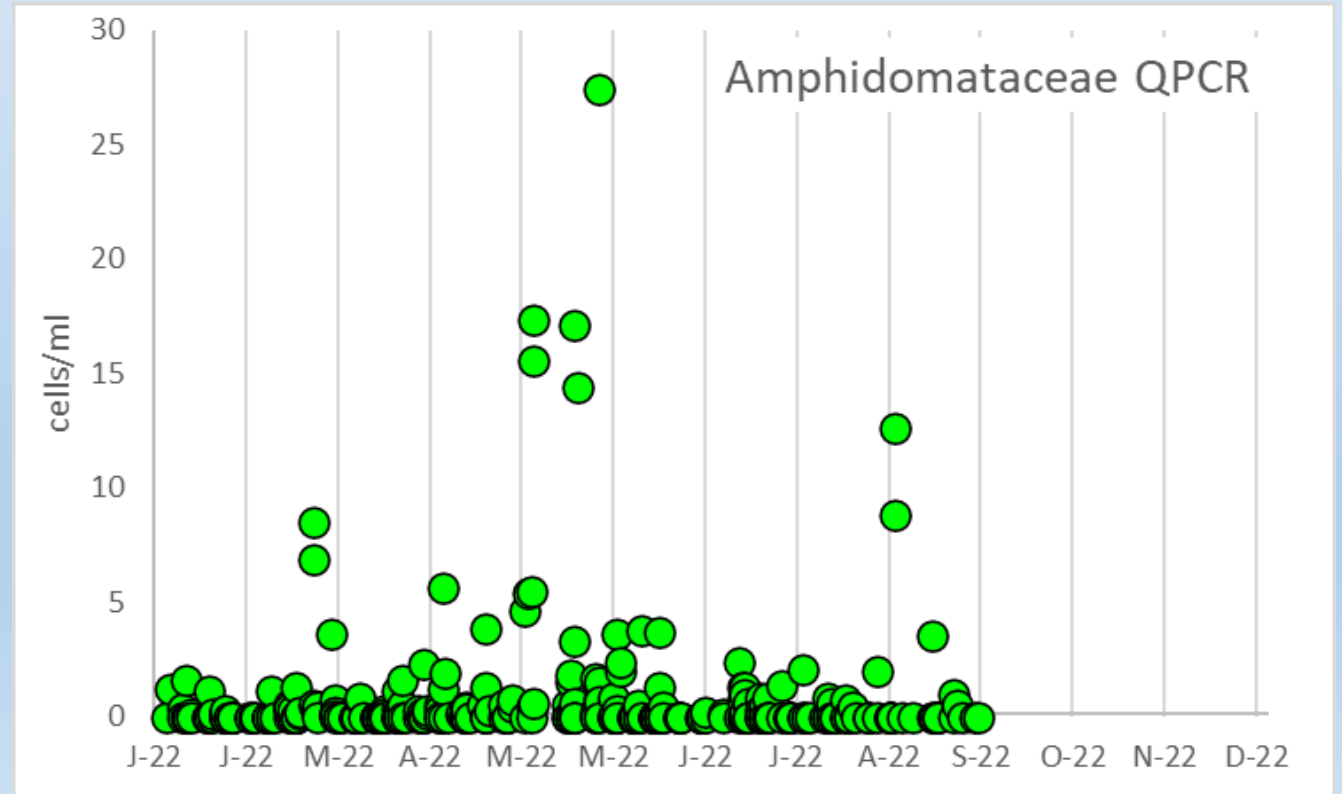
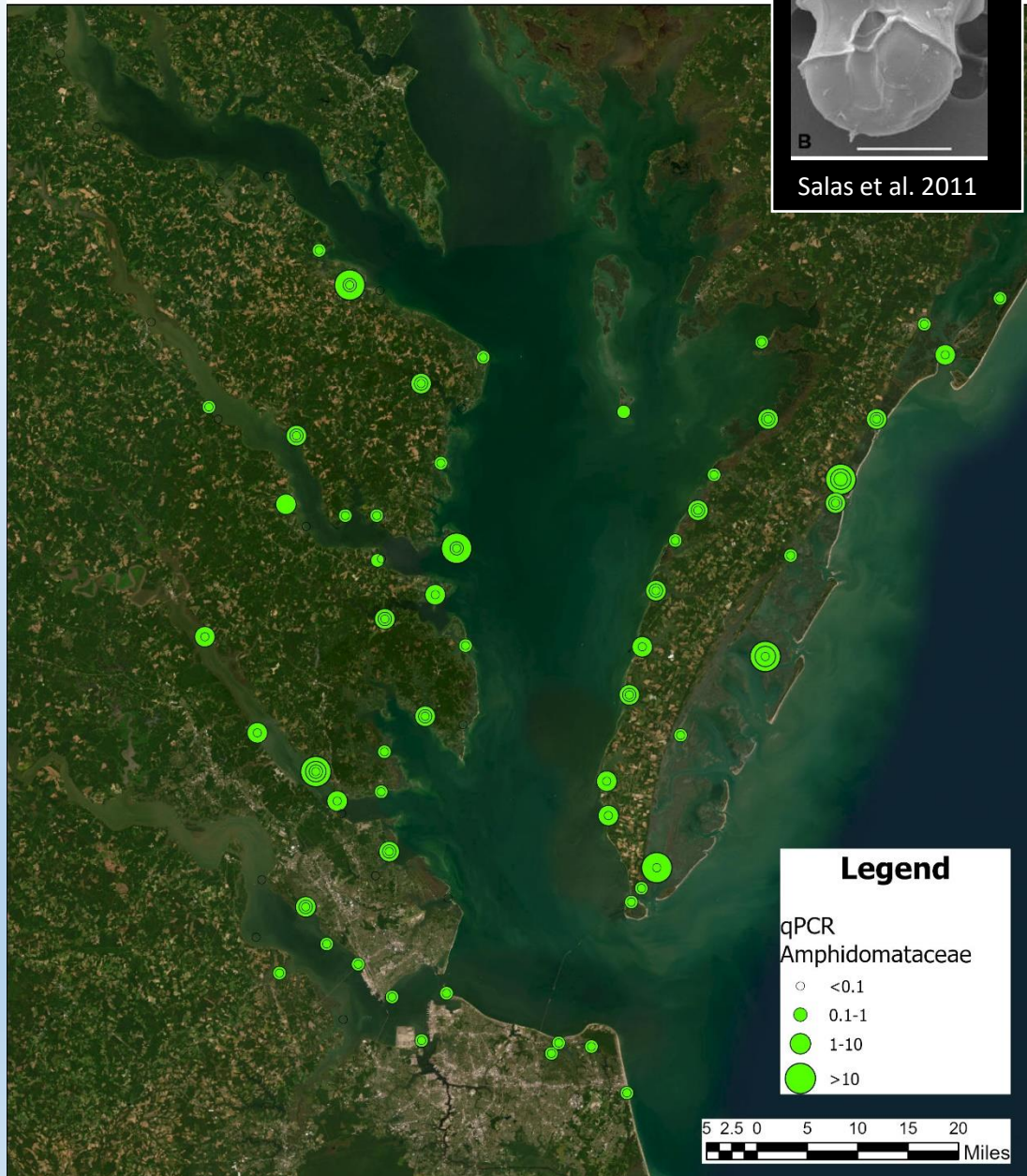
Additional monitoring: ODU and HRSD James River & research (Mulholland et al). VIMS (Reece, Smith, et al.)

Amphidomataceae

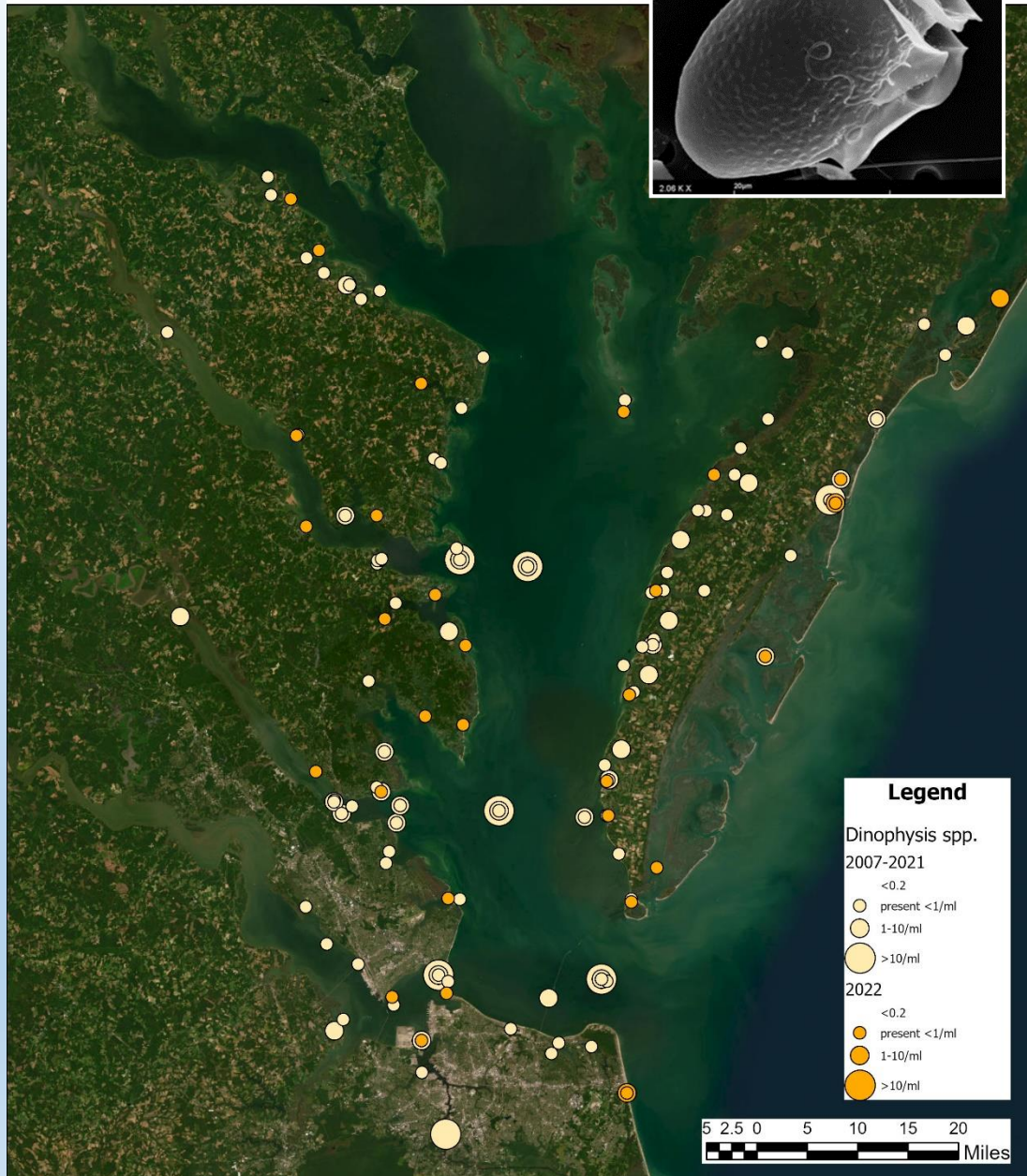
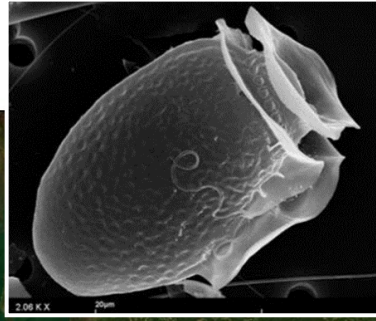


AZP- Amphidomataceae

- Dinoflagellate family: *Azadinium* & *Amphidoma* spp.
 - Not identified through microscopy. 2nd year of qPCR survey
 - qPCR >0.1 in ~ 35% of 2022 samples
 - <0.1- 27.4 cell equivalents/ml (14% >1/ml)
 - No commercial Azaspiracid test kits available
 - AZA-1 reported using SPATTs- 2017-2018 (CBTOX- Onofrio et al. 2021)
 - Continued Surveillance

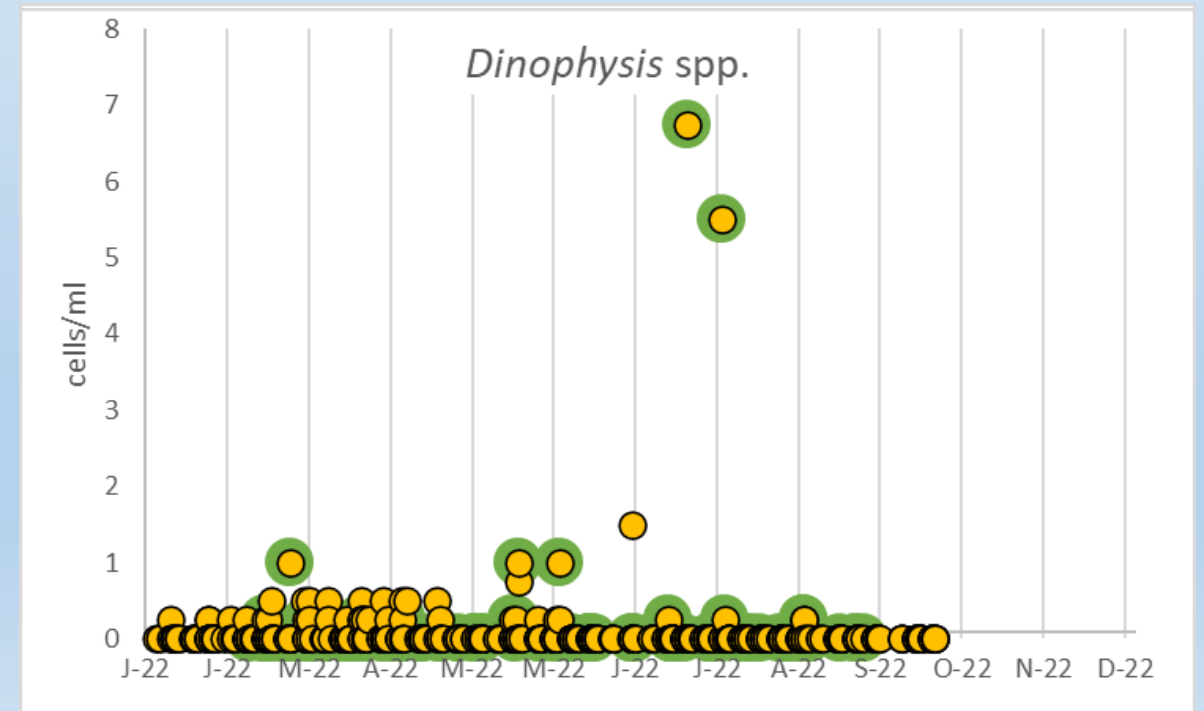


Dinophysis spp.

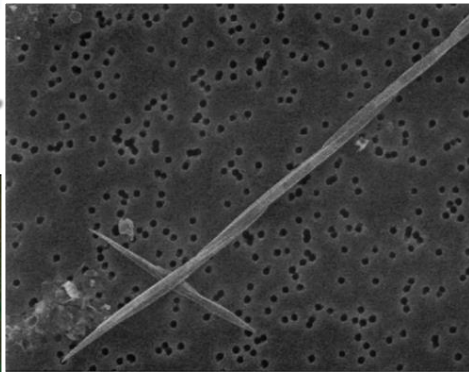


DSP- *Dinophysis*

- Widespread distribution in Chesapeake Bay and seaside E. Shore
 - Generally low cell densities
 - Present in ~ 11% of 2022 samples (<0.25 cells/ml)
 - 0.25-6.75 cells/ml (1% >1/ml)
- Okadaic Acid ELISA on 40 seawater samples & 4 SPATTs
- **0/107 sample above detection limit (<0.1ppb)**
 - 2021: 0.13ppb: 2.5 cells/ml
 - Widespread OA/DTXs reported using SPATTs- 2017-2018 (CBTOX- Onofrio et al. 2021)



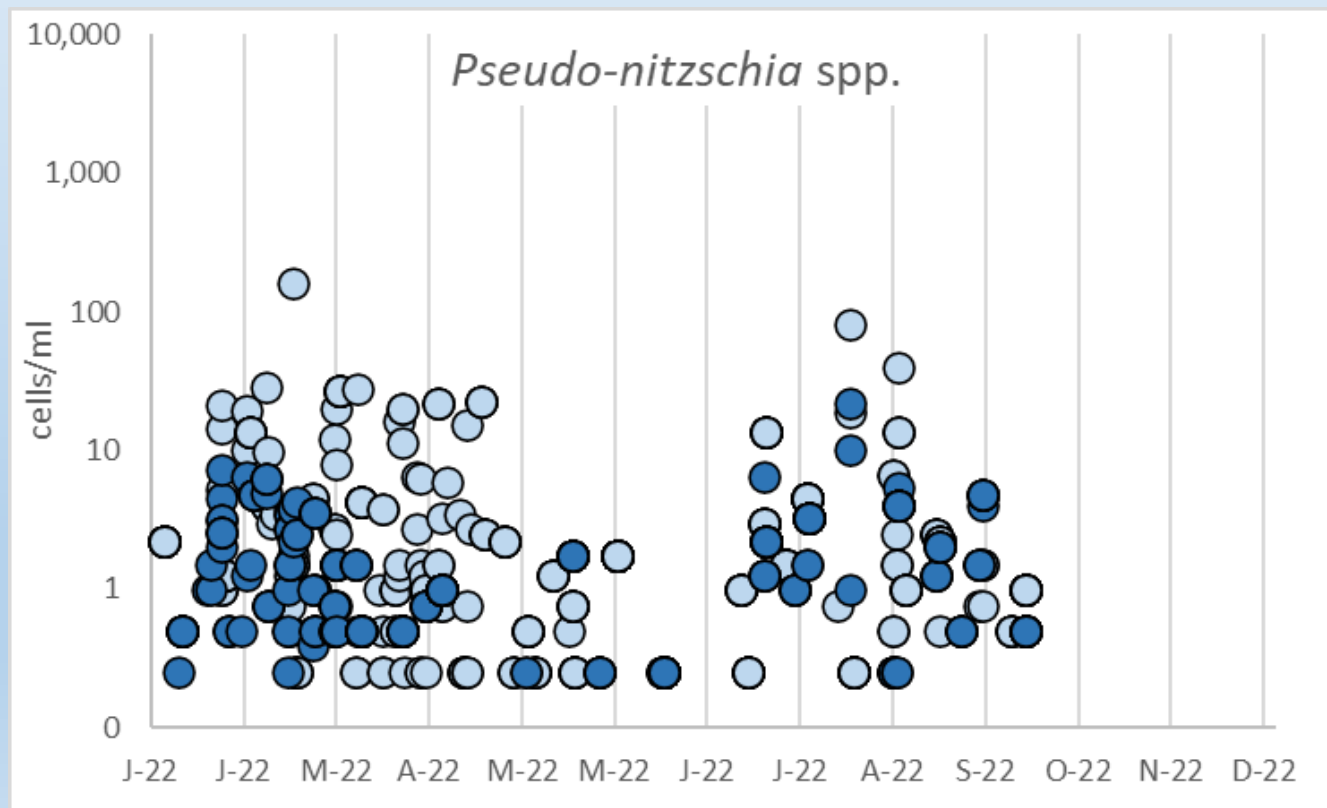
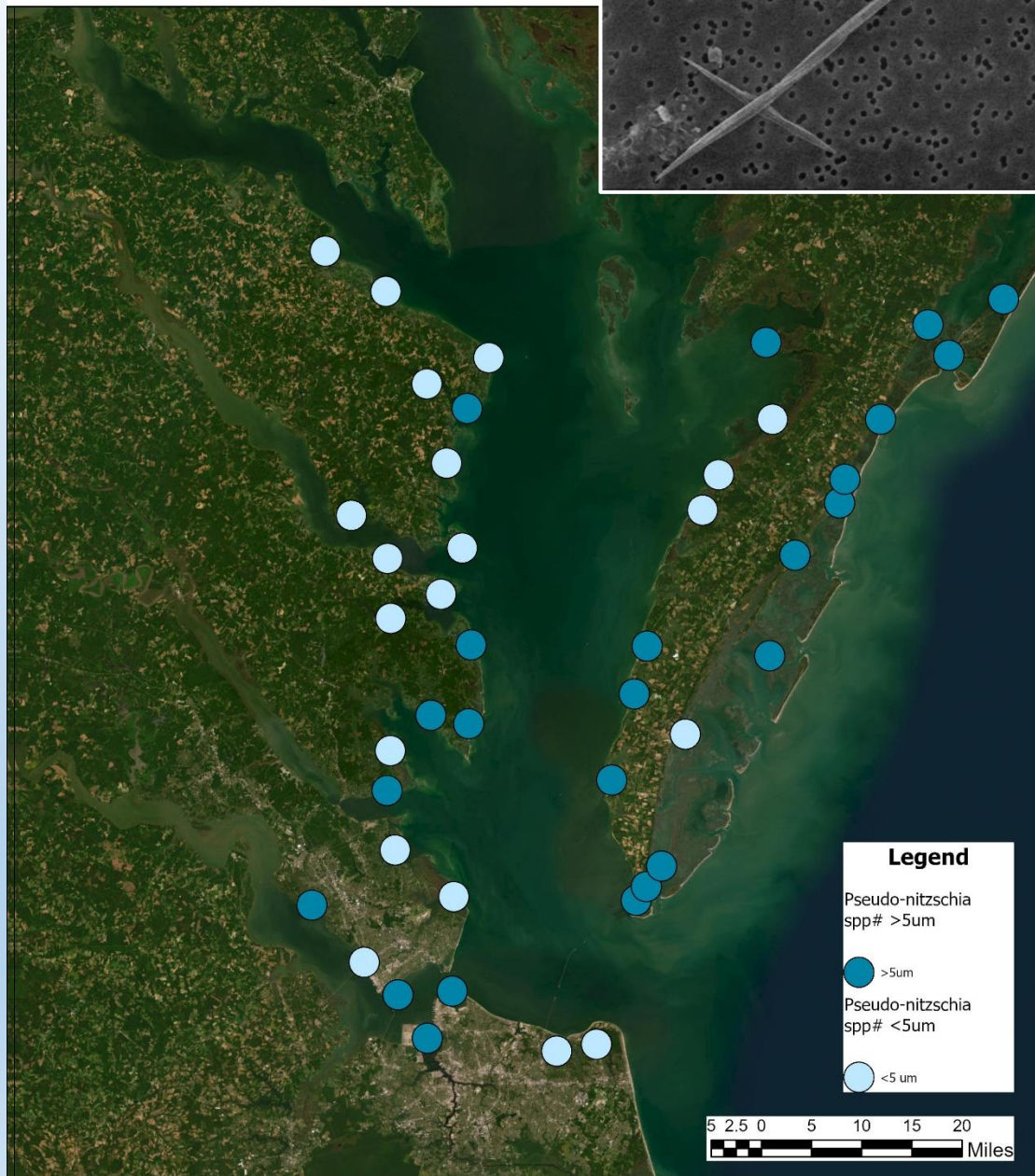
Pseudo-nitzschia spp.



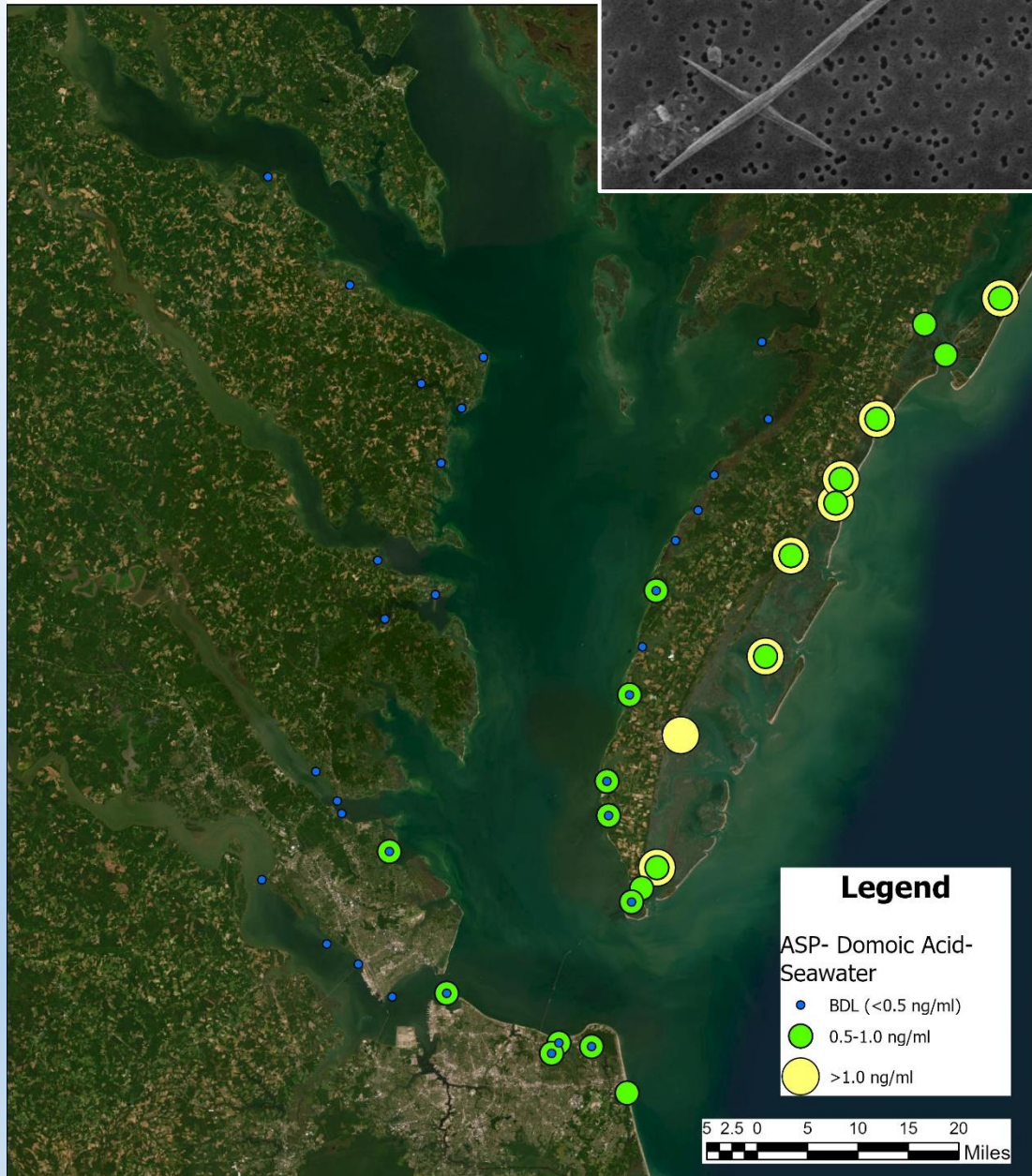
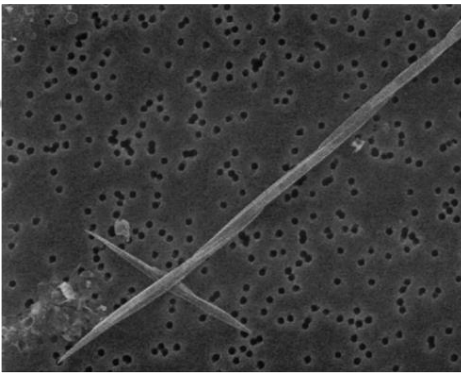
ASP- *Pseudo-nitzschia*

Widespread distribution in Chesapeake Bay and Seaside E. Shore

- Present in ~ 30% of 2022 samples: 0.5-163 cells/ml (<1% >50/ml)
- Sizes:
 - Larger: (>5 μ m width)- primarily coastal Atlantic sites- max ~23 cells/ml
 - Smaller (< 5 μ m width)- within Chesapeake Bay- max ~163 cells/ml

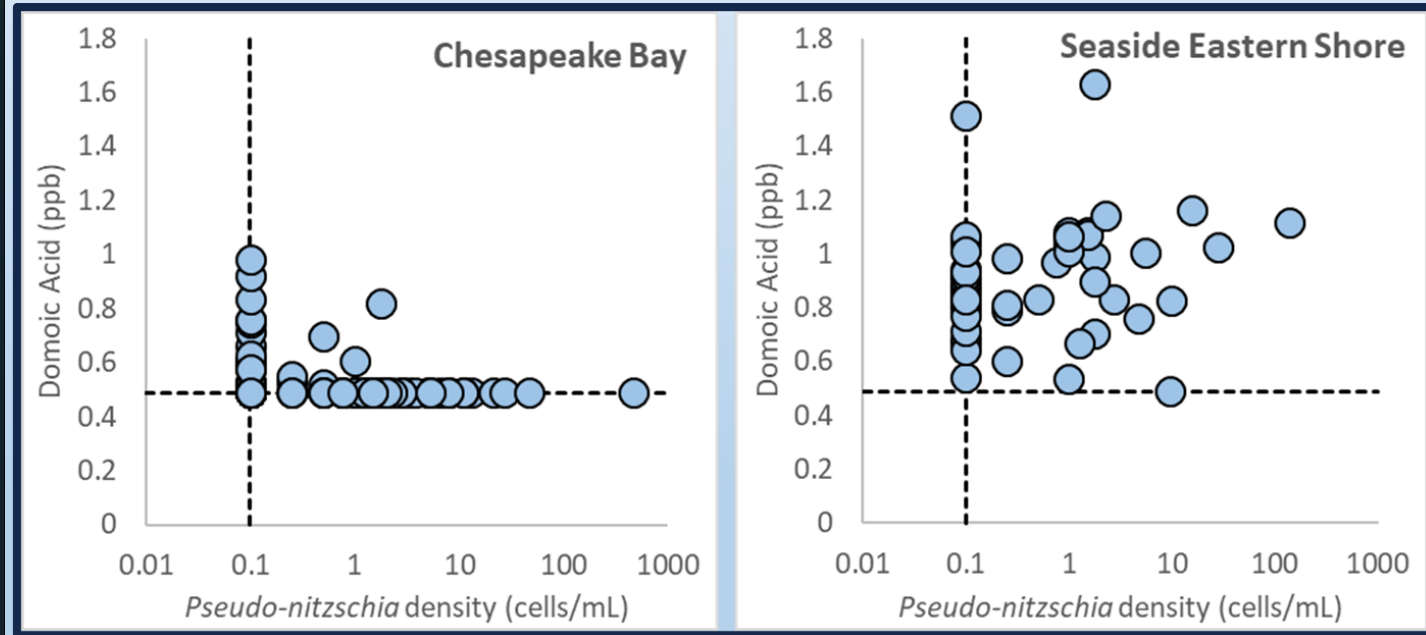


Pseudo-nitzschia spp.



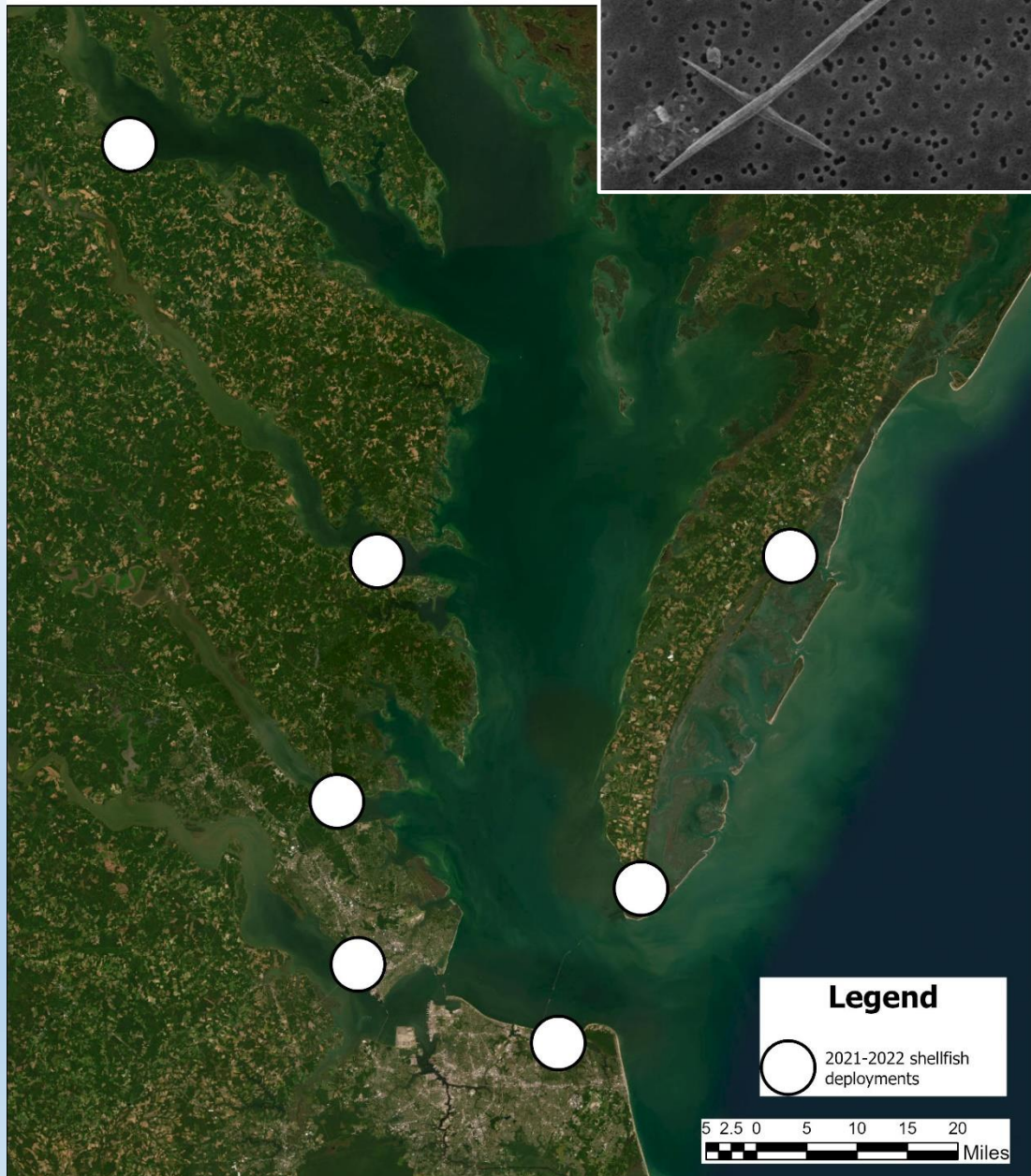
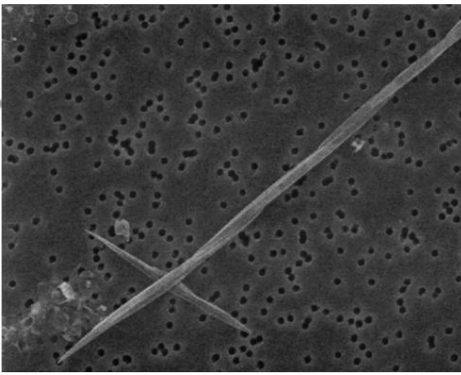
ASP- *Pseudo-nitzschia*

- Widespread distribution in Chesapeake Bay and Seaside E. Shore
 - Present in ~ 30% of 2022 samples: 0.5-163 cells/ml (<1% >50/ml)
- Domoic Acid ELISA on seawater samples and SPATTs
 - **60/132 2022 water samples tested above detection limit (0.5ppb)**
 - 0.51-1.42 ppb Domoic Acid in seawater
 - Primarily Seaside E. Shore, throughout year
 - Similar max in seawater as measured in CBTOX study (Pease/Smith et al)



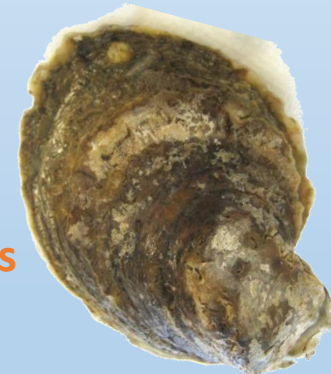
There was weak correlation between cell density and DA concentration, with considerable detections of DA when cells were not observed. Inside Chesapeake Bay there also were elevated concentrations of *Pseudo-nitzschia* observed in the absence of detectable DA.

Pseudo-nitzschia spp.



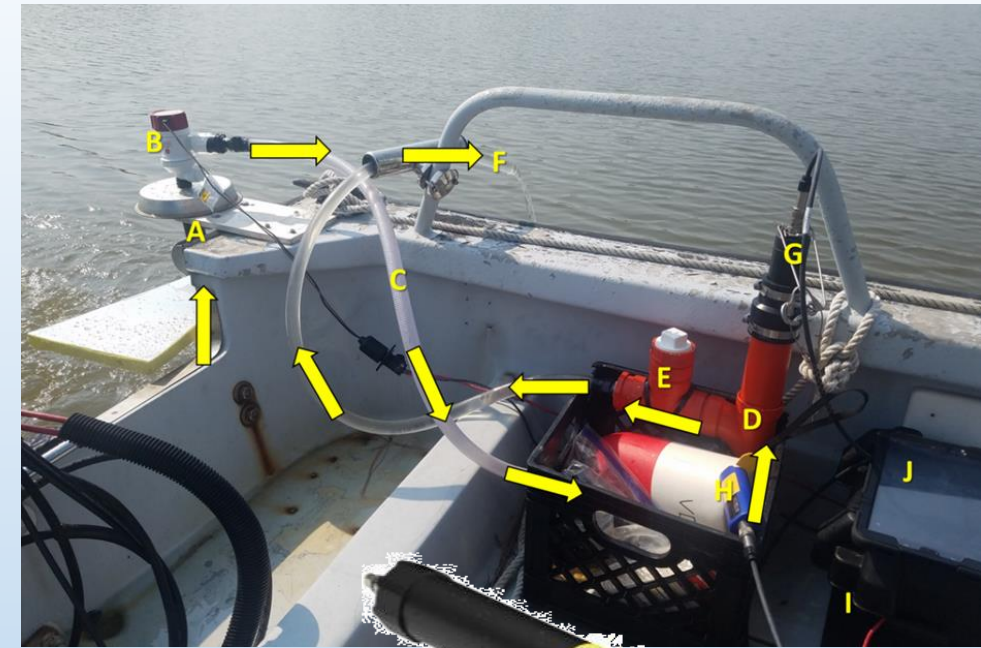
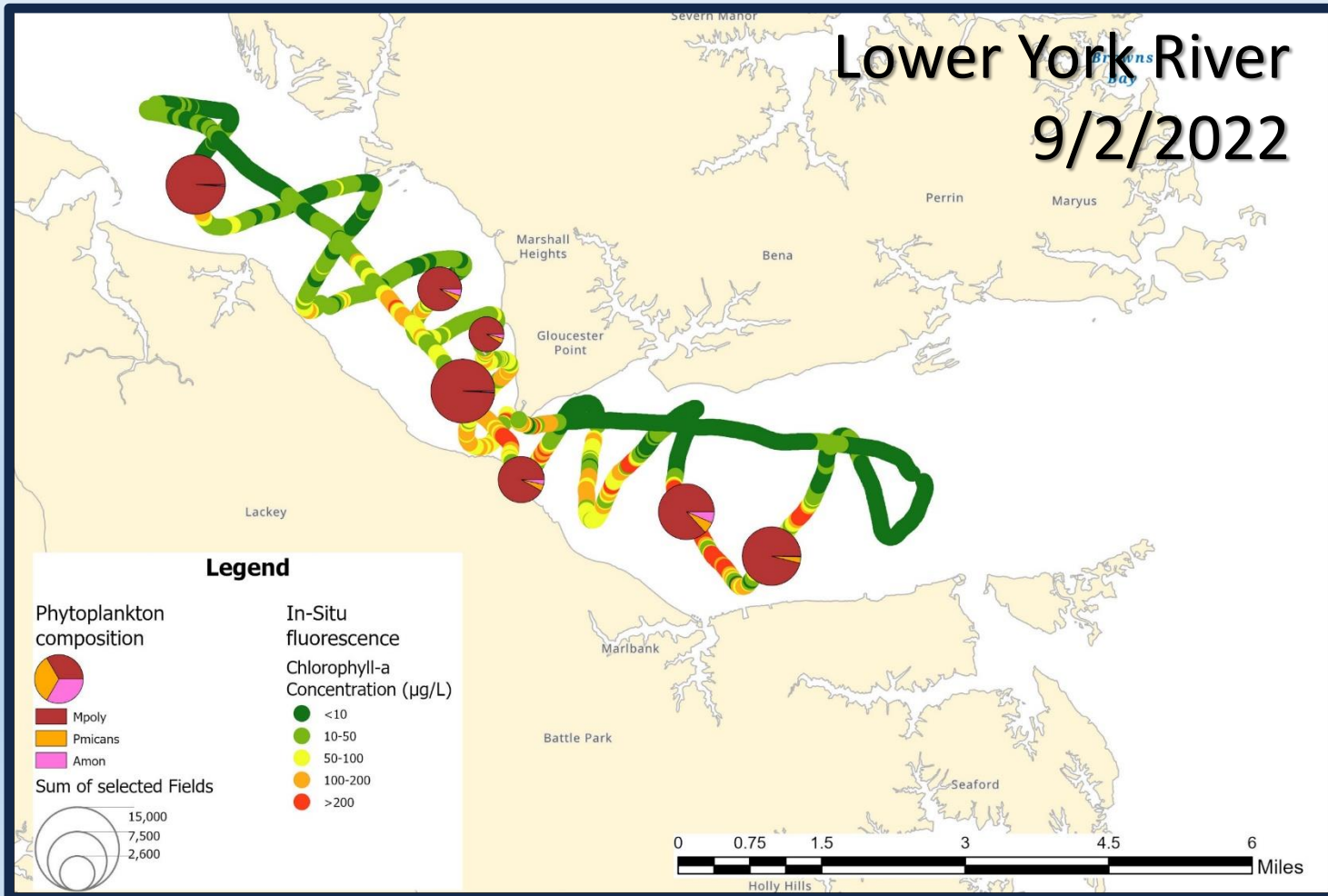
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 - Primarily Seaside E. Shore, throughout year
 - Similar max in seawater as measured in CBTOX study (Pease/Smith et al)
- **Shellfish Deployments:**
 - December 2021-February 2022: 7 sites
 - All samples (23) BDL (<0.1ppm in shellfish meat)
 - July-August 2022: 5 sites:
 - 1/11 samples positive for DA (0.356ppm)
 - **NSSP Closure criteria: 20ppm**
 - CBTOX shellfish DA max 0.579ppm
 - **December 2022- ongoing deployment at 8 sites**
 - **Ongoing monthly collections**
- Expanded seawater analyses and shellfish deployments
 - Additional sampling stations
 - Continued work to relate to cell densities/ qPCR (ODU/VIMS collaborations)



Flow-through

- ISSC Techniques and Tools for Toxin Management grant supported equipment (2020-2021).
- Field test during 2022 summer/fall bloom



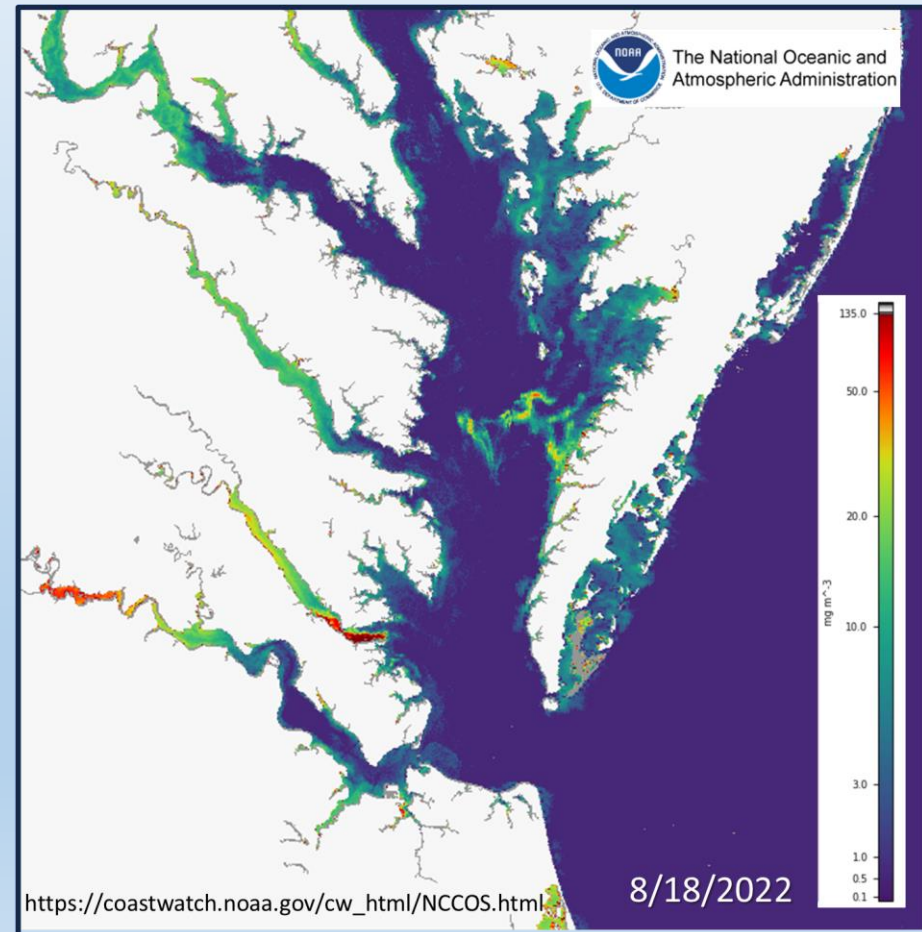
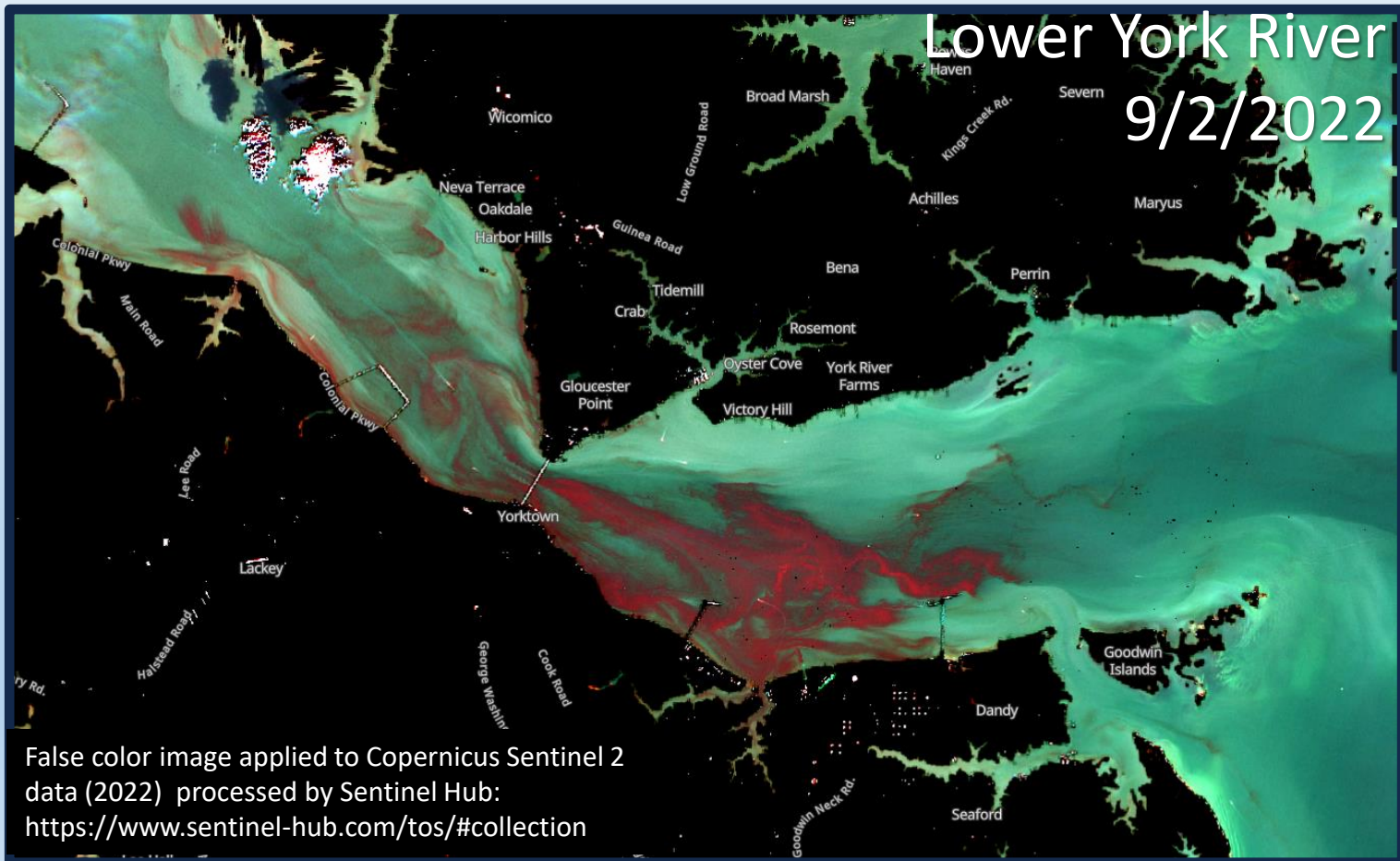
September 2022 Lower York bloom
Flow-through chlorophyll measurements

- <1 to >700 µg/L

M. polykrikoides 2400-8700 cells/mL
Alexandrium monilatum 440 cells/ml (max)
Prorocentrum micans 450 cells/mL (max)

Remote sensing

- Coordination and collaboration with NOAA, National Centers for Coastal Ocean Science (https://coastwatch.noaa.gov/cw_html/NCCOS.html)



Ongoing and upcoming projects

- Additional shellfish deployments and toxin testing
 - New sites, year-round studies
 - Additional routine seawater DA ELISAs
 - CAAS Cube and plate reader
- Revisiting cell/toxin relationships and thresholds
 - qPCR vs cell counts (Flow Cam and traditional)
 - Cell densities vs toxins (seawater)
 - Toxins: grab samples vs shellfish/SPATTs
 - Cyanotoxins
- Workgroups/meetings:
 - 2022 US HAB symposium: presentation
 - NOAA SeaGrant Workshop: Jan18-19 *Applying novel techniques to assess and forecast HABs in Chesapeake Bay to protect fisheries, aquaculture and human health*
 - VDH/DEQ HAB Technical Meetings (MOU finalization anticipated spring 2023)
 - ISSC Biotoxin Committee (ISSC March 2023)

