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The Freshwater Marine Continuum – Downstream Threats of Cyanotoxins

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A Wake-Up Call: Freshwater Cyanotoxins in Marine Mammals

Freshwater microcystin discharged to Monterey Bay National Marine Sanctuary via riverine sources and entered marine food webs

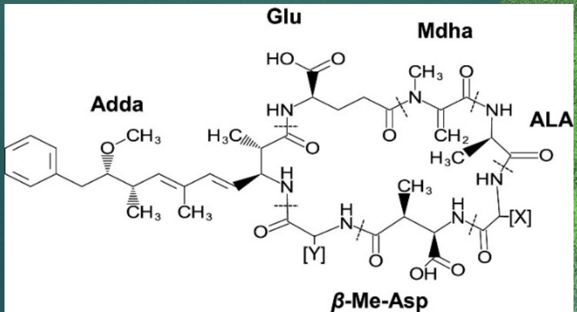
http://www.google.com/search?q=clams&client

Miller et al. 2010

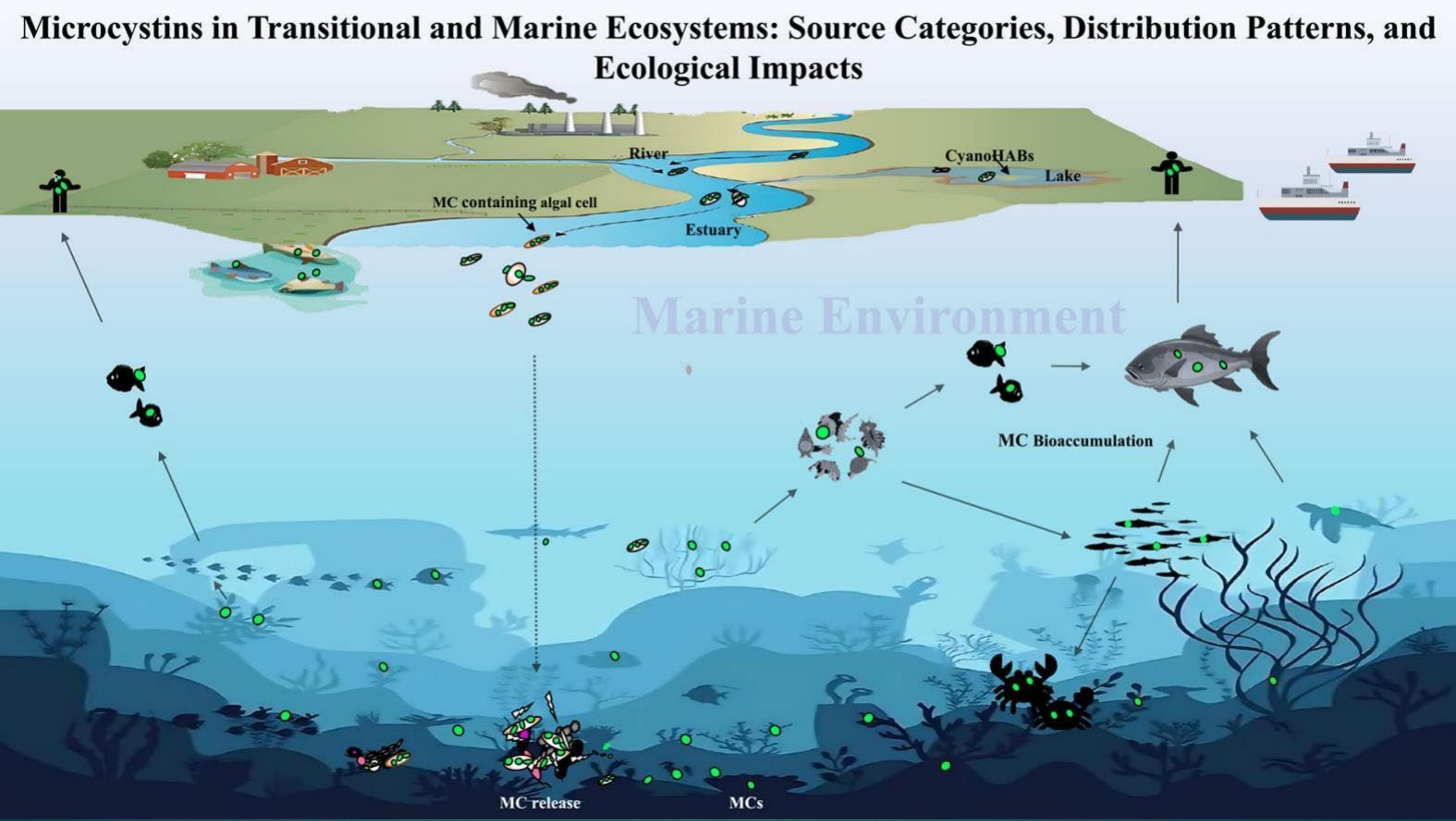
http://www.seaotterresearch.o

Why Focus on Microcystin?

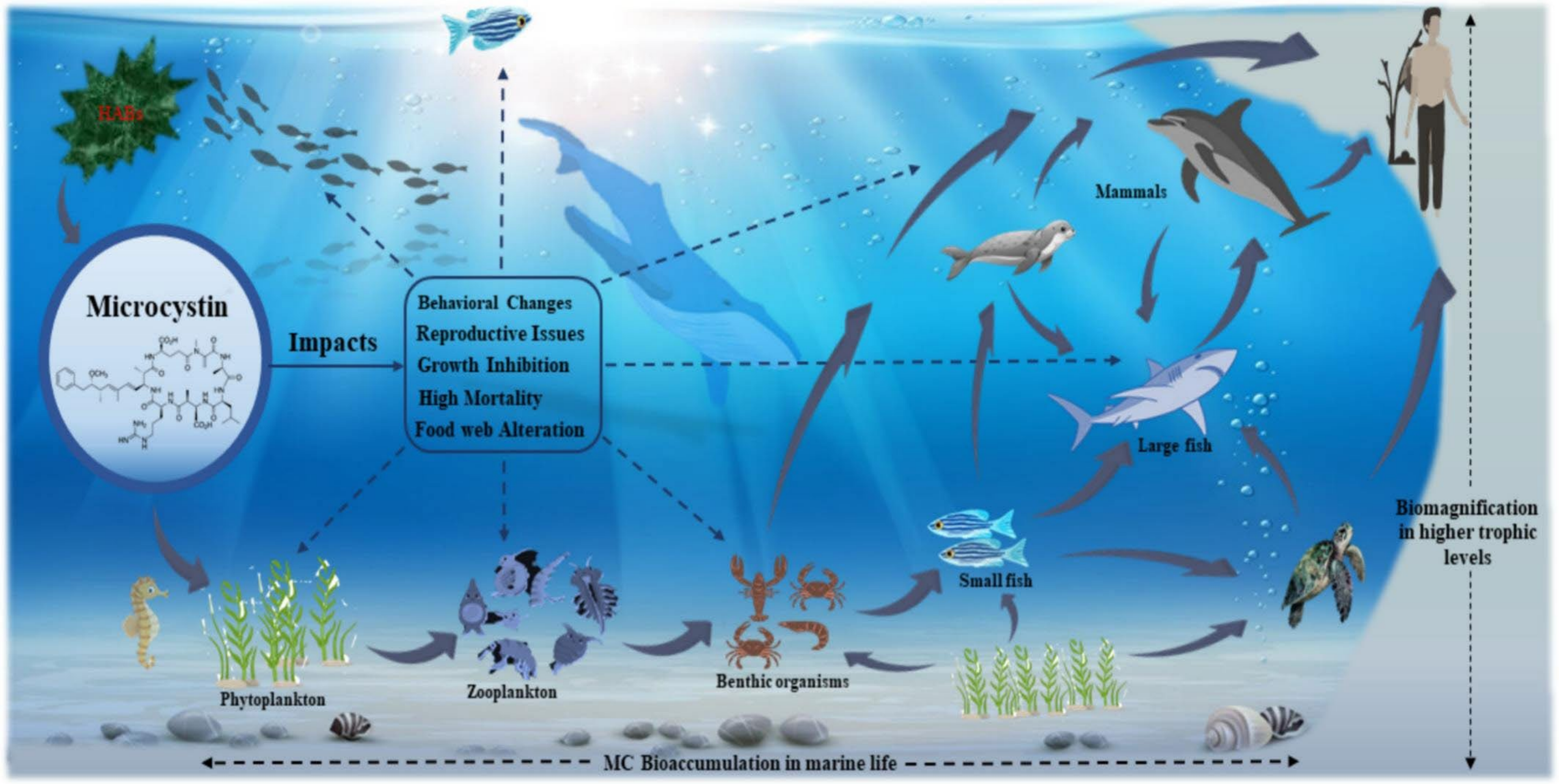
- Globally, most frequently detected cyanotoxin
- Cyanotoxin most often observed in transitional environments
- Demonstrated persistence during downstream transport




North Atlantic Ocean
South Pacific Ocean
South Atlantic Ocean
Indian Ocean
MC detected from CyanoHABs in coastal/estuarine waters
CyanoHABs in coastal/estuarine waters
MC detected from freshwater discharge
Global Microcystin Distribution Patterns in the Marine Ecosystems
Visualized according to continental affiliation
Latitude (N+/S-)
Longitude (E+/W-)
Continent
Africa
Asia
Europe
North America
South America
Australia



Yousaf et al. 2025

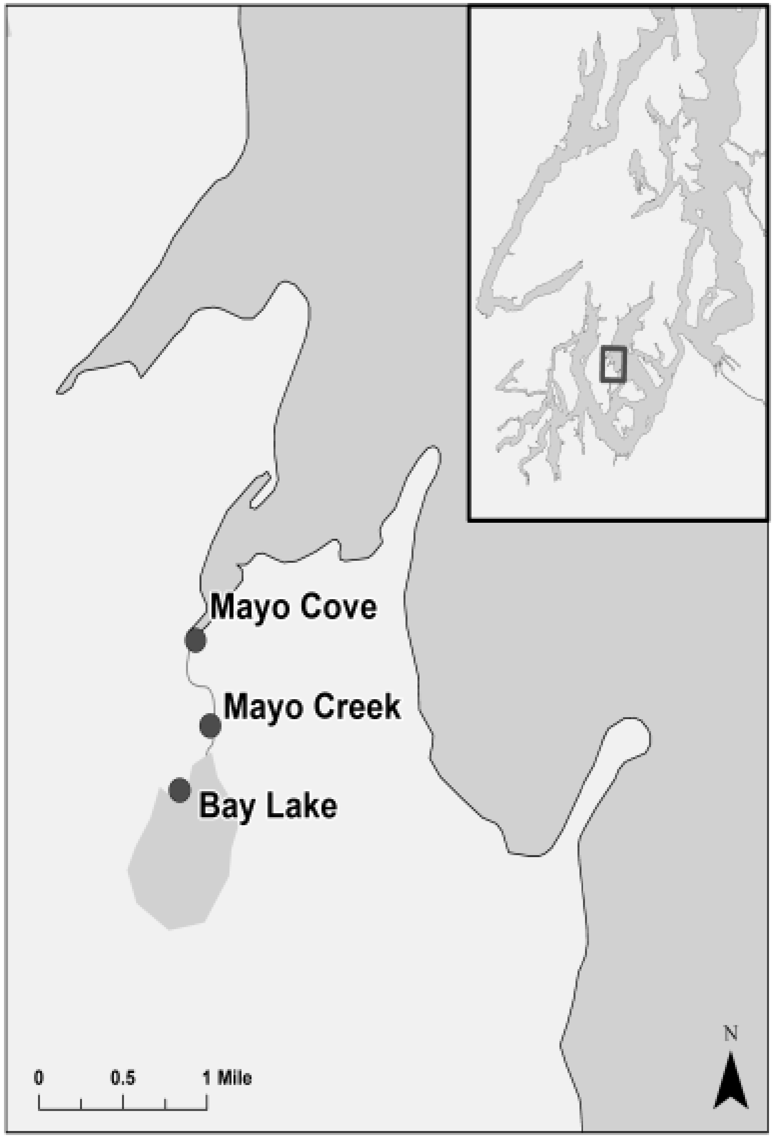


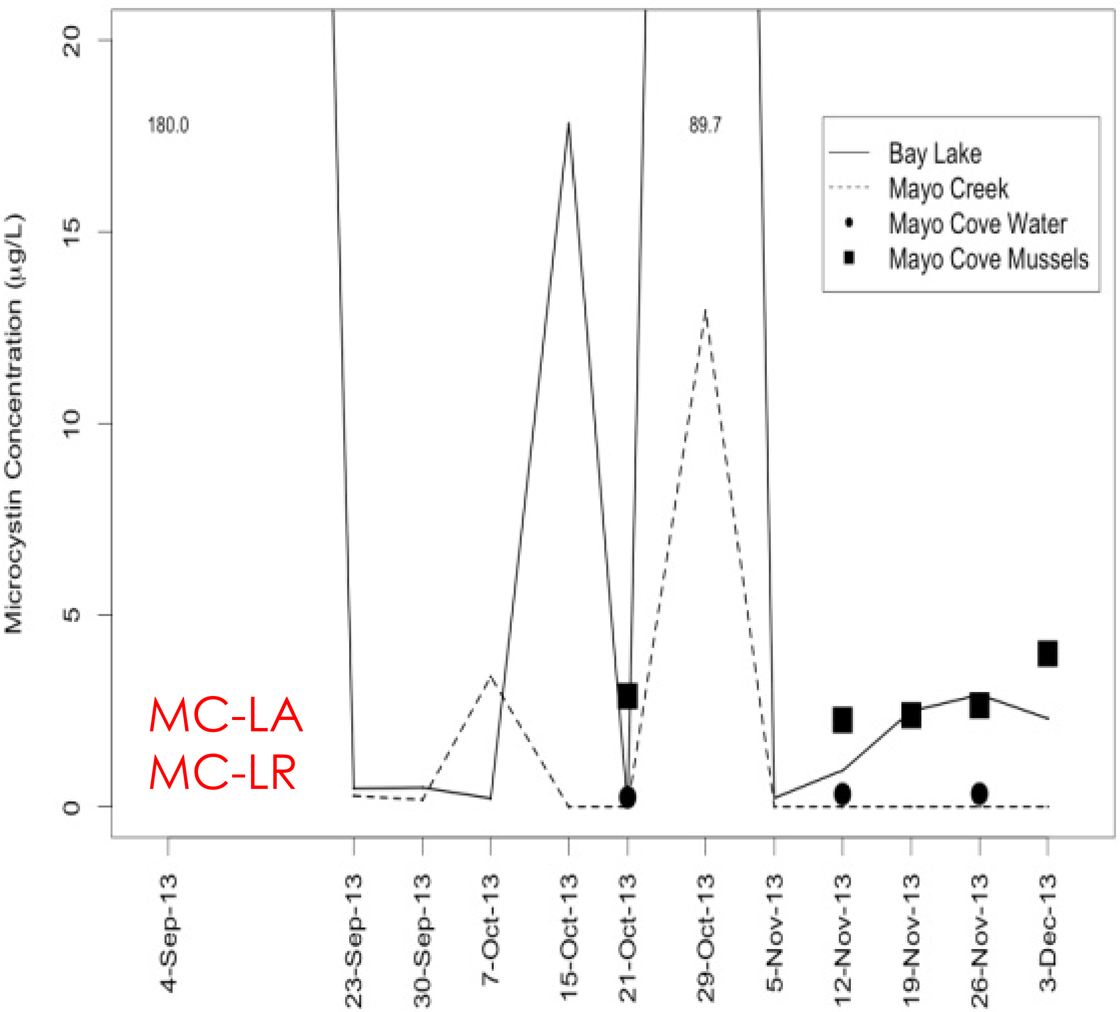
Yousaf et al. 2025

MC Persistence in Shellfish

- Rapid accumulation in mussels (within 24 hours), with persistence for weeks (>27 days) (Camacho-Muñoz et al. 2021)
- Microcystins persisted in oysters for ≥4 weeks, with detectable levels at 8 weeks (Gibble et al. 2016)
- Eastern oysters accumulate microcystin faster, reach higher levels, and depurate more slowly (weeks to months) than clams (days) (Straquadine et al. 2022)

Puget Sound: A Case Study in Freshwater–Marine Transport





The San Francisco Estuary
From the freshwater Sacramento–San Joaquin Delta (Delta) to the Pacific Ocean

San Francisco Estuary

- Delta is a vital ecosystem and water supply with complex hydrology
  - *Microcystis* occurs annually and accounts for most microcystin production
- Microcystins have also been detected throughout the Bay and on the coast

Study Overview

Knowledge gap

- Do MCs accumulate in Delta shellfish?
  - Asian clams (*Corbicula fluminea*), widespread – can reach densities in excess of 10,000 organisms m2
  - Introduced signal crayfish (*Pacifastacus leniusculus*), opportunistic can eat Asian clams
- Are MCs from the Delta transported downstream to San Francisco Bay?
  - Do these toxins accumulate in shellfish species along the salinity gradient?

Objectives

1. Compare ELISA and LC-MS methodologies for MC measurements in shellfish
2. Determine if MCs accumulate in Delta clams and crayfish
3. Determine if MCs accumulate in shellfish along the salinity gradient

Introduced Asian clams filter water containing cyanobacteria. Signal crayfish are a trophic level higher. Crabs, shrimp, crayfish occur in the transition zone

Field Methods

Delta Clams and Crayfish

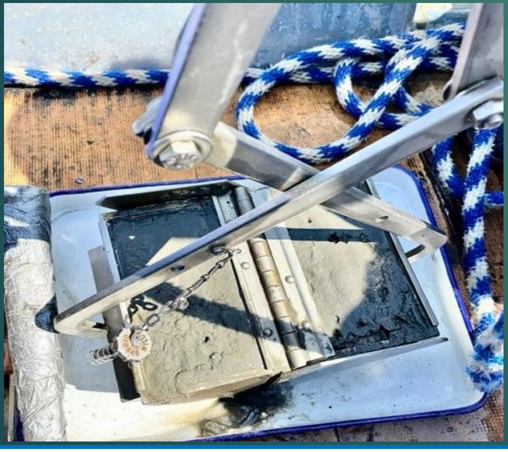
Estuary Crabs and Shrimp

2020 to 2022

2023 to 2025

2 x a month (June – Oct), 1 x a month (Nov – May)

- **Asian clams each date**
- **Crayfish opportunistically**
- **Water grab samples (toxins)**



- **Targeting crabs, shrimp, crayfish**
- **Water grab samples (toxins and qPCR)**
- **SPATT samplers**

For Delta collected approximately 50-100 clams per site to composite and 30 total crayfish. For Estuary Crabs/shrimp not at all locations in all months.

Lab Methods

Corbicula Fluminea
(Asian clam)

Delta Clams and Crayfish

- Remove shell, composite, freeze dry
- Extract with MeOH
- ELISA, LC-MS on subset (8 MC variants/MMPB)

Estuary Crabs, Shrimp, and Crayfish

- Remove crab/crayfish shell, keep shrimp whole
- MC and domic acid every date
- LC-MC (4 MC variants), ELISA on subset

A series of lab techniques were conducted to measure MCs in shellfish

Objective 1: Method Comparison ELISA vs LC-MS Delta Clams and Crayfish

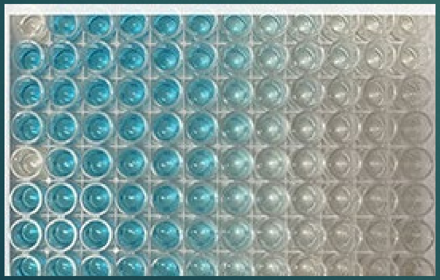
All Delta samples were analyzed by ELISA, a subset were analyzed by LC-MS. All Estuary samples were analyzed by LC-MS and a subset will be analyzed with ELISA. Both are common cyanotoxin detection methods.

Analytical Methods

- Variety of analytical methods available to measure microcystins
- Most common: ELISA (enzyme linked immunosorbent assay) and LC-MS.

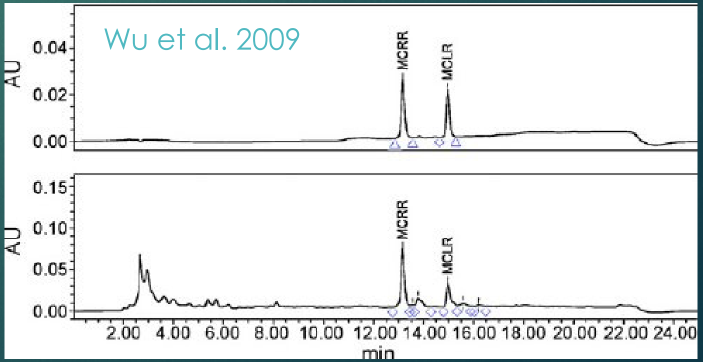
ELISA

- Relatively inexpensive
- Measures all microcystin congeners
- Easily accessible for many user groups
- Considered semi-quantitative (EPA protocols available)



LC-MS

- Expensive
- Target specific congeners (Usually ~4-12)
- Only found in some laboratories
- Considered more quantitative than ELISA



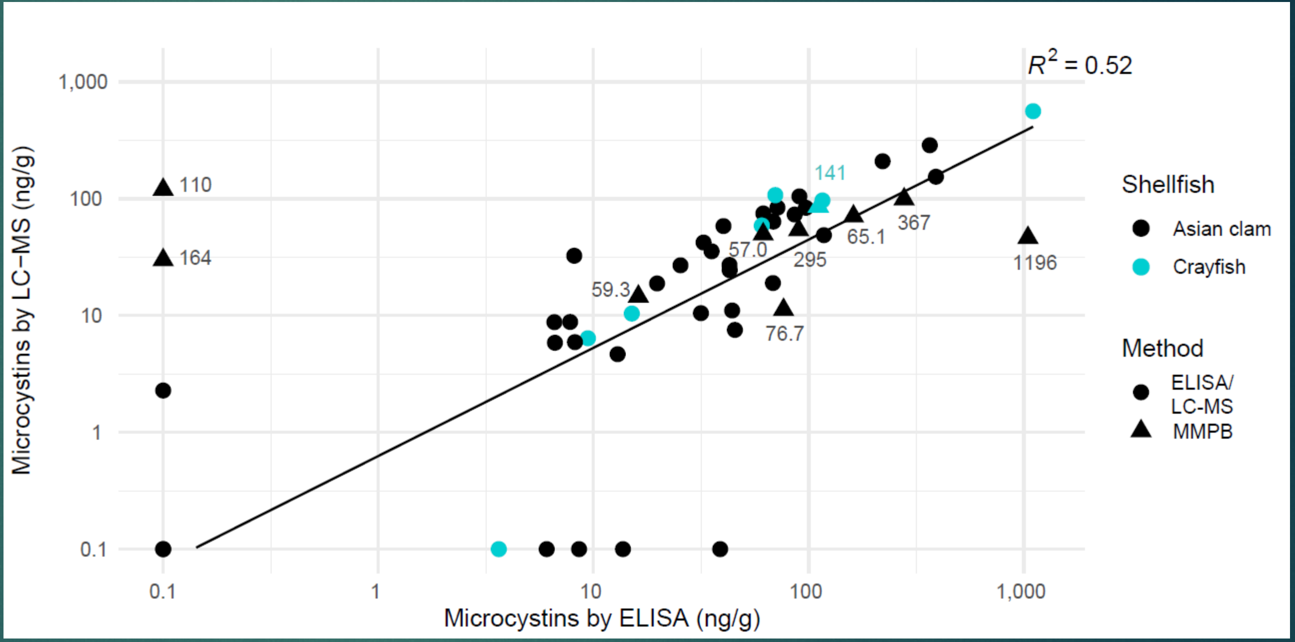
Method Comparison ELISA vs LC-MS

- Initial LC-MS results did not match up with ELISA results
  - BakerBond C18 columns (Mekebri et al. 2009)
  - Over or underestimated MCs
- Different clean-up step added to extraction process
  - Strata X Solid Phase Adsorption (polymeric resin, finer particle size)
    - Good agreement between methods

We encountered issues when following established LC-MS microcystin extraction and clean-up procedures. A new clean-up column improved analysis.

Method Comparison ELISA vs LC-MS

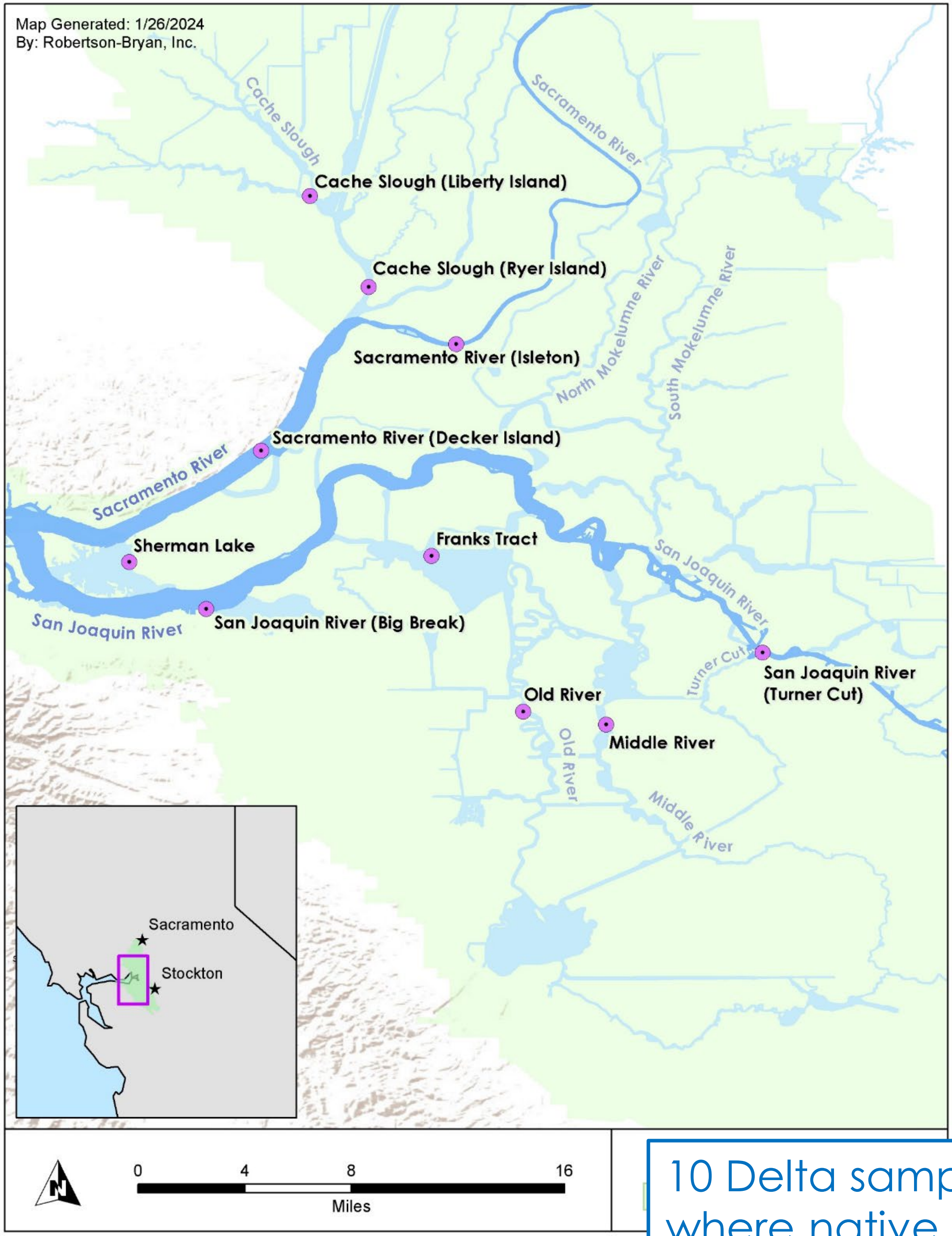
- Subset analyzed with LC-MS
- 9 clam and 1 crayfish sample analyzed with MMPB



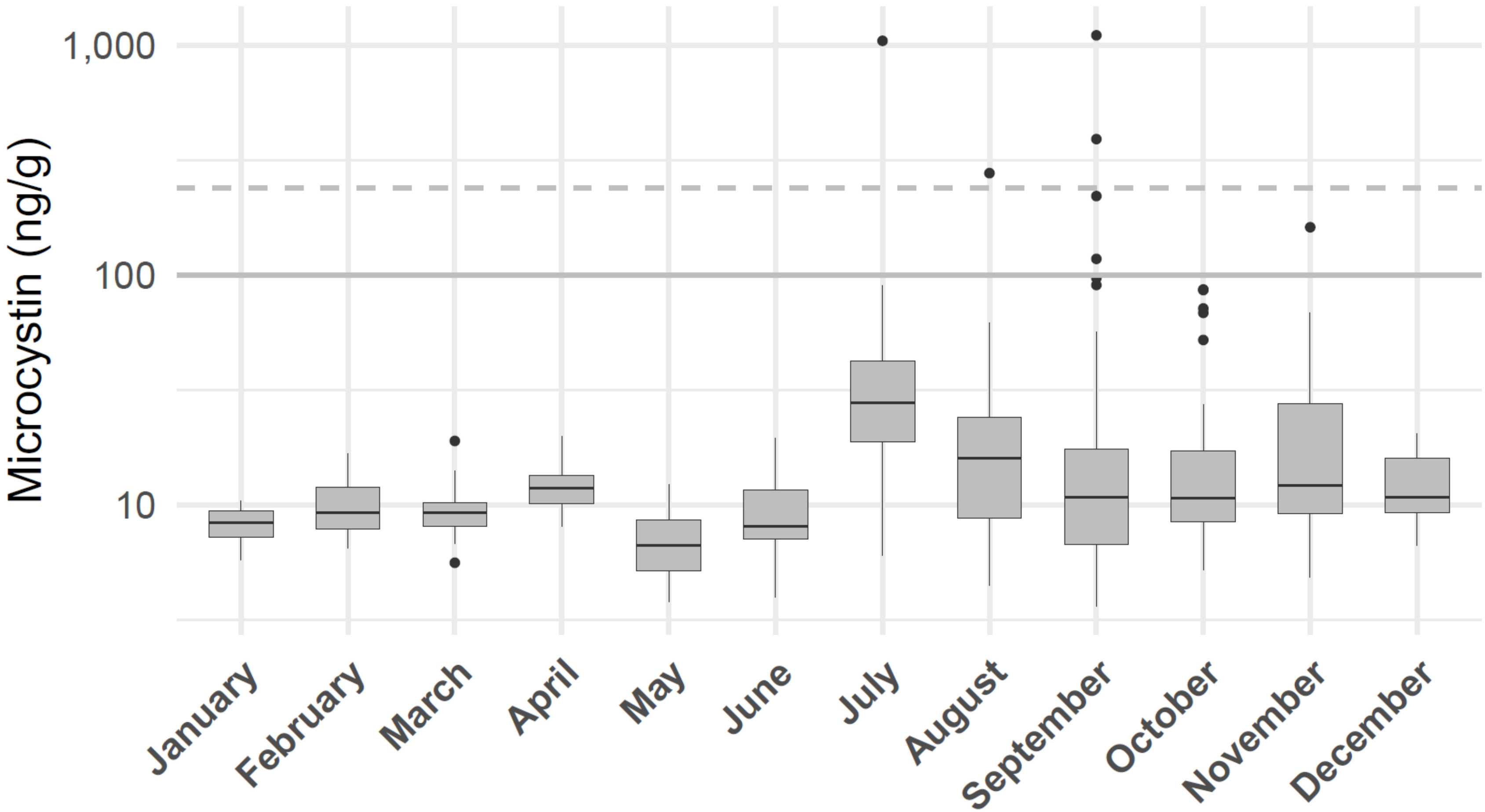
MMPB method, a more costly method, measures all freely available and protein bound MC congeners. There was generally good agreement between ELISA and LC-MS except for a few outliers.

Objective 2: Microcystin in Delta shellfish and water

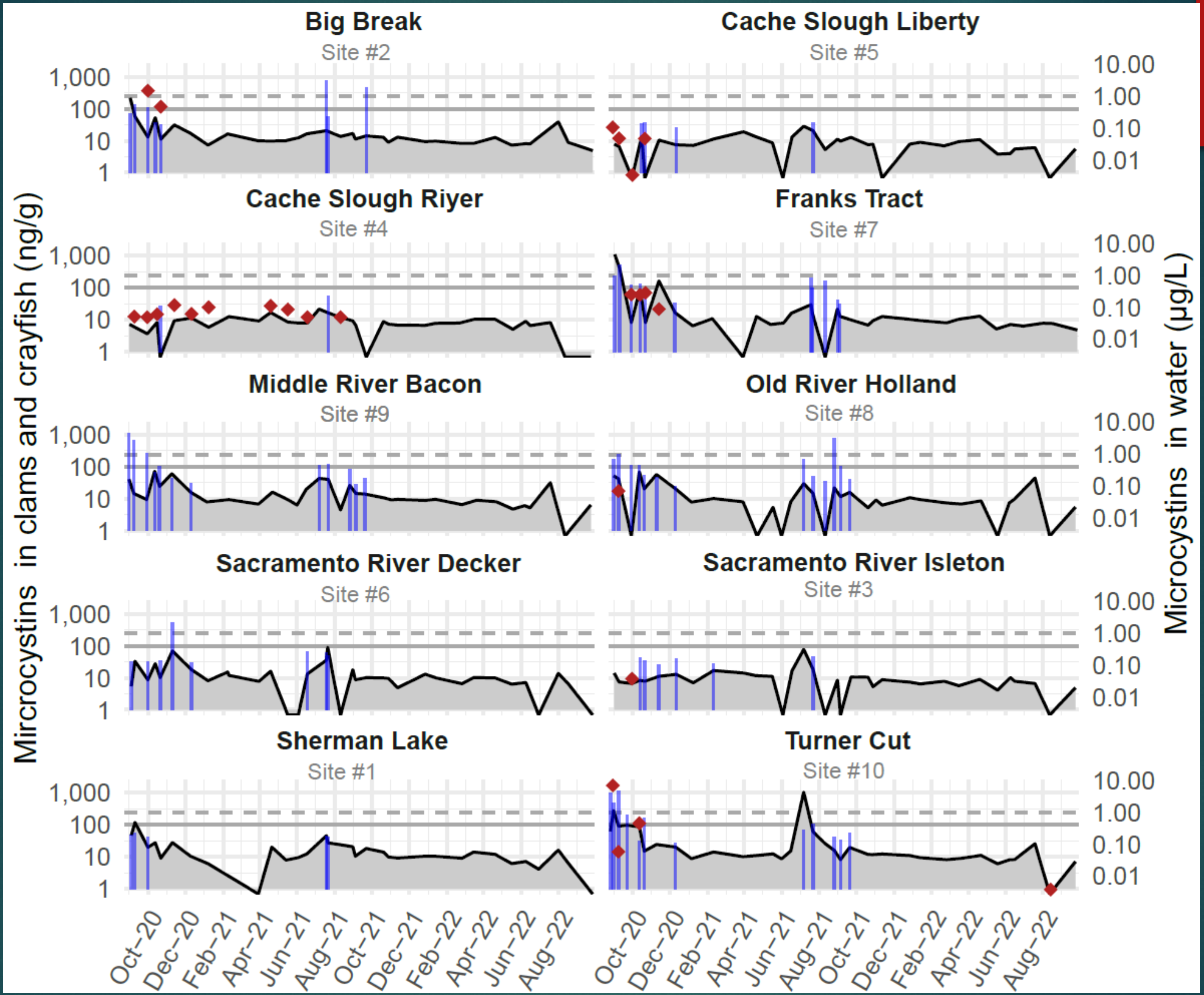
Asian Clam/ Crayfish Sampling Sites



10 Delta sampling sites. Chosen, based on where native fish species are known to feed

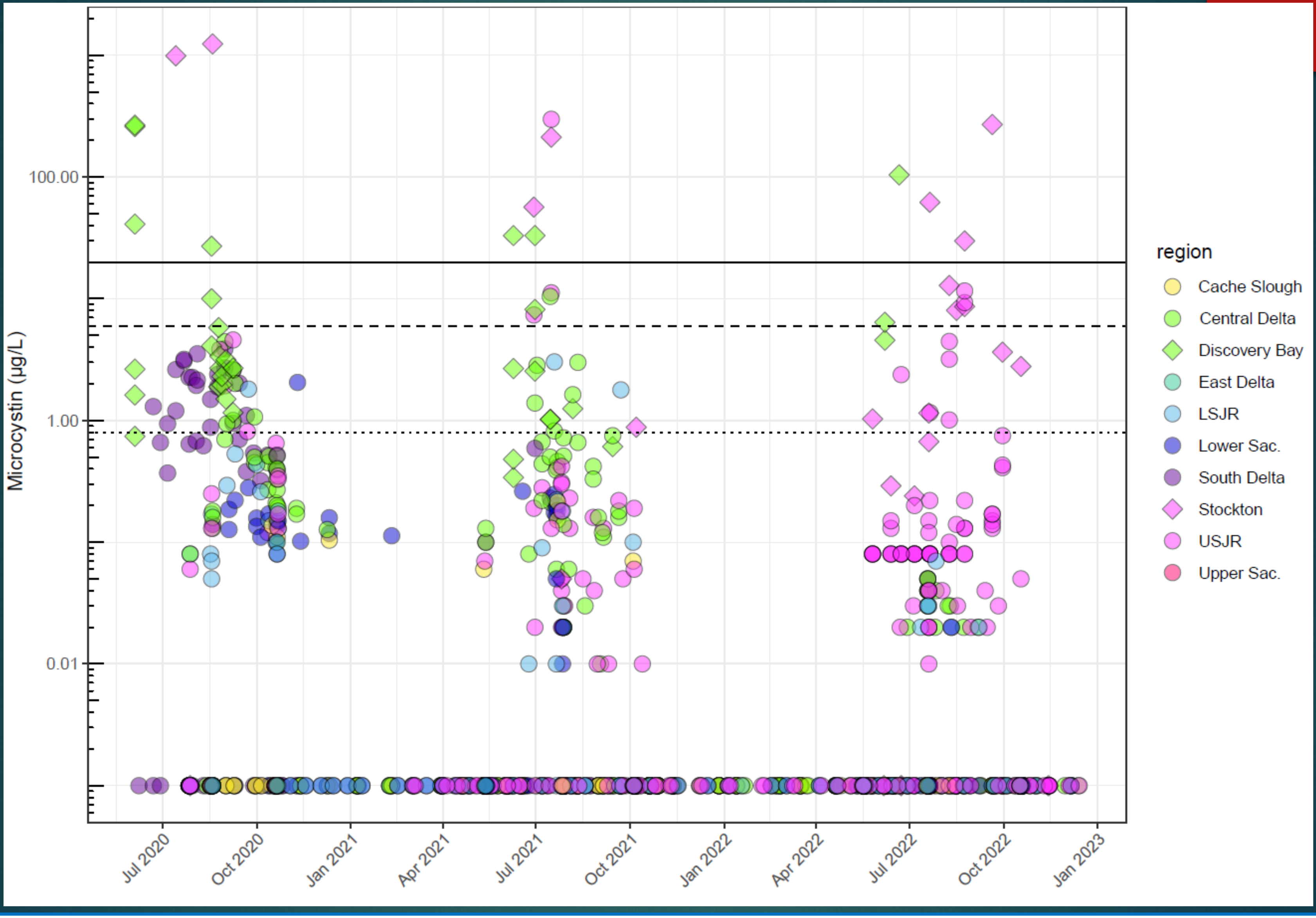


Microcystins (dry weight) in Asian clams highest from July - November



MCs were persistent in Asian clams at all sites year round and concentrations increased when water column MCs increased. MC's were not present in 2022 water grab samples from the study sites.

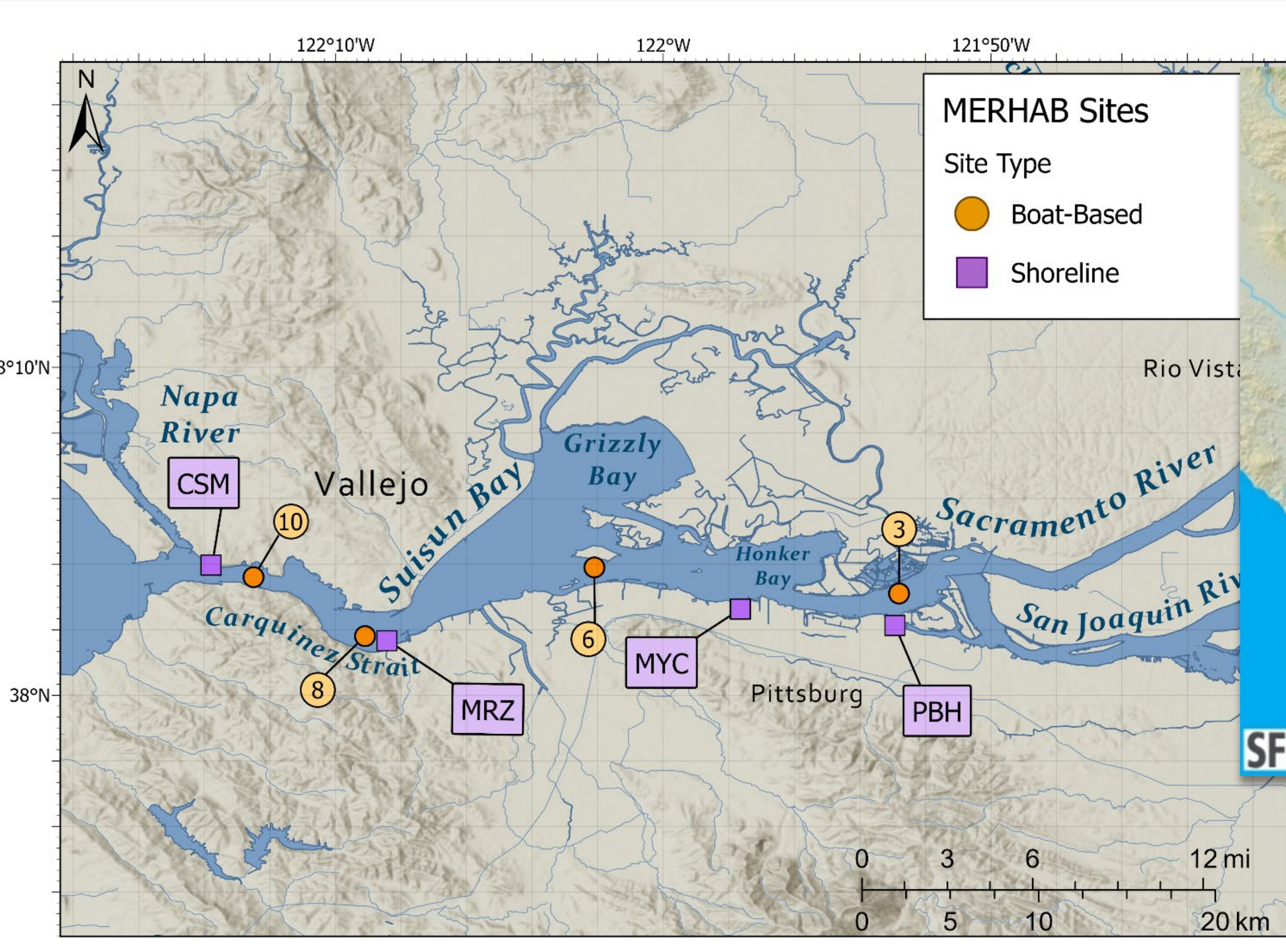
Microcystins in Delta Waters from 2020 to 2022



A compilation of all available Delta microcystin data shows the toxin was present in waters in all three study years.

Objective 3: Microcystins Across the Salinity Gradient

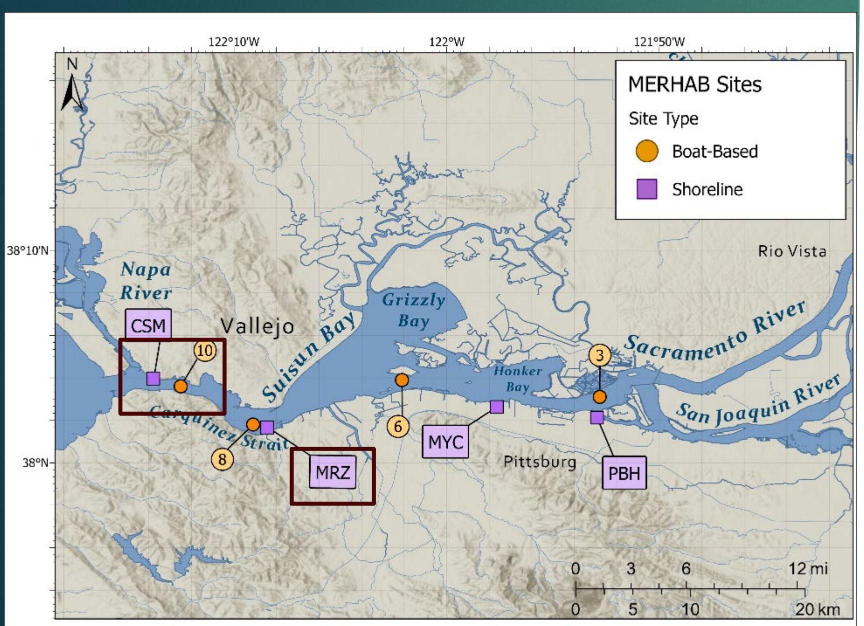
Shellfish results presented in wet weight.

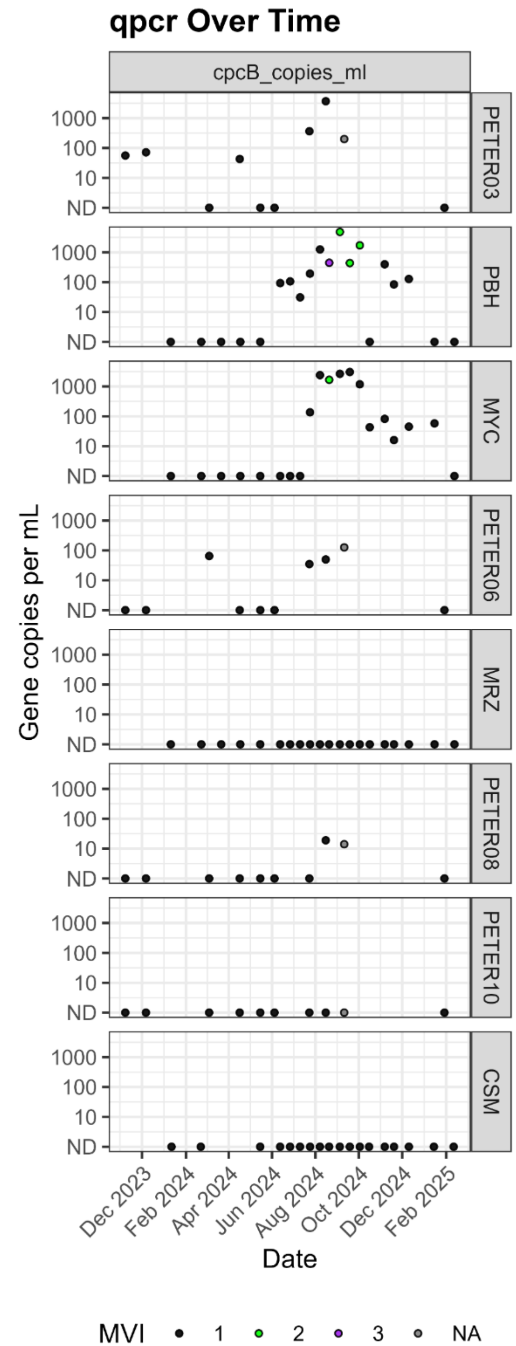

MERHAB Sites
Site Type
Boat-Based
Shoreline
122°10'W
122°W
121°50'W
38°10'N
38°N
N
Napa River
Vallejo
Suisun Bay
Grizzly Bay
Honker Bay
Carquinez Strait
Sacramento River
San Joaquin River
Rio Vista
Pittsburg
CSM
MRZ
MYC
PBH
10
8
6
3
0 3 6 12 mi
0 5 10 20 km


Sacramento
Legal Delta
Suisun Bay
San Pablo Bay
Stockton
Central Bay
Oakland
San Francisco
South Bay
Lower South Bay
San Jose
N
0 5 10
Miles
SFEI AQUATIC SCIENCE CENTER

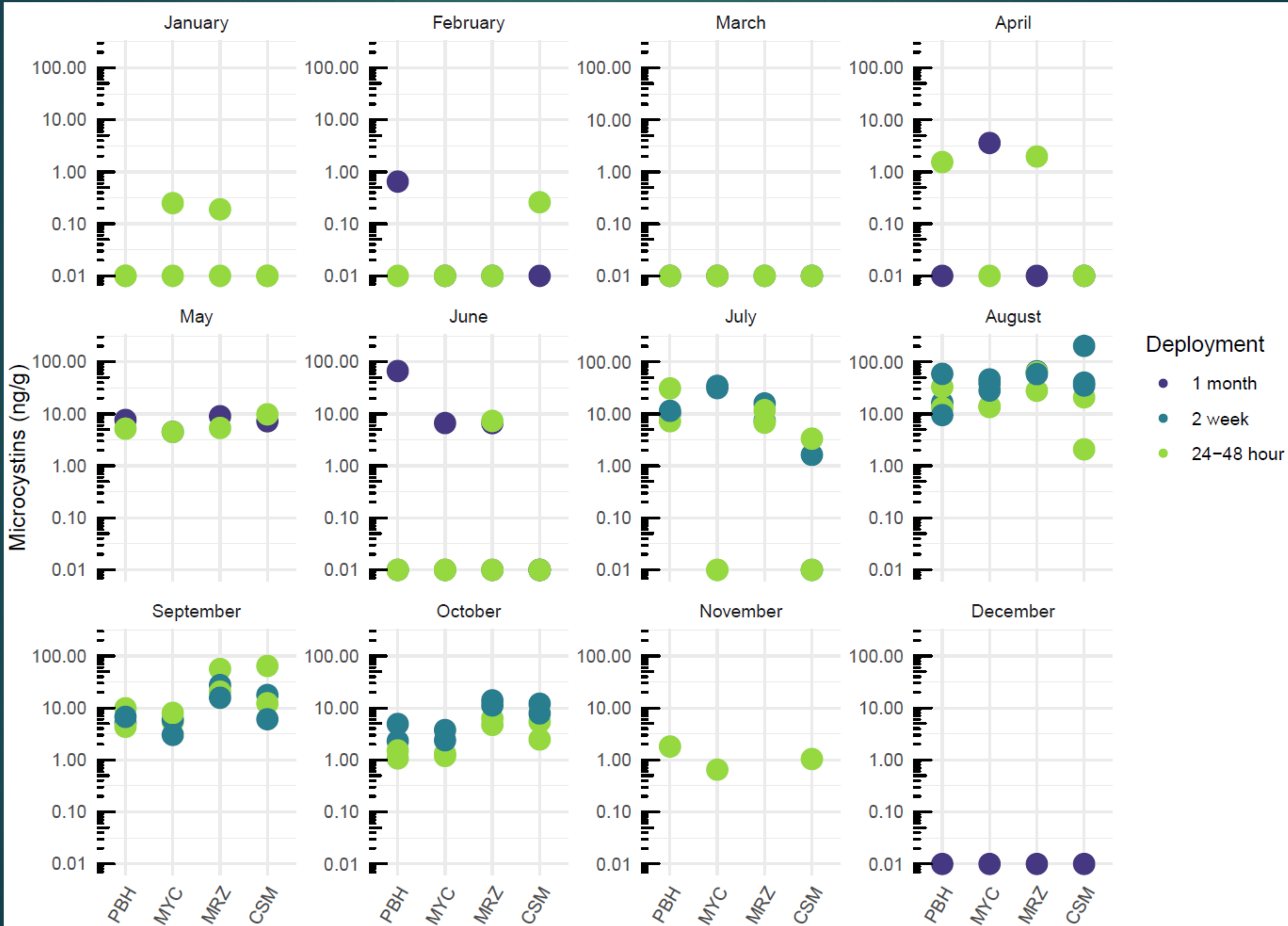
Microcystis across the salinity gradient

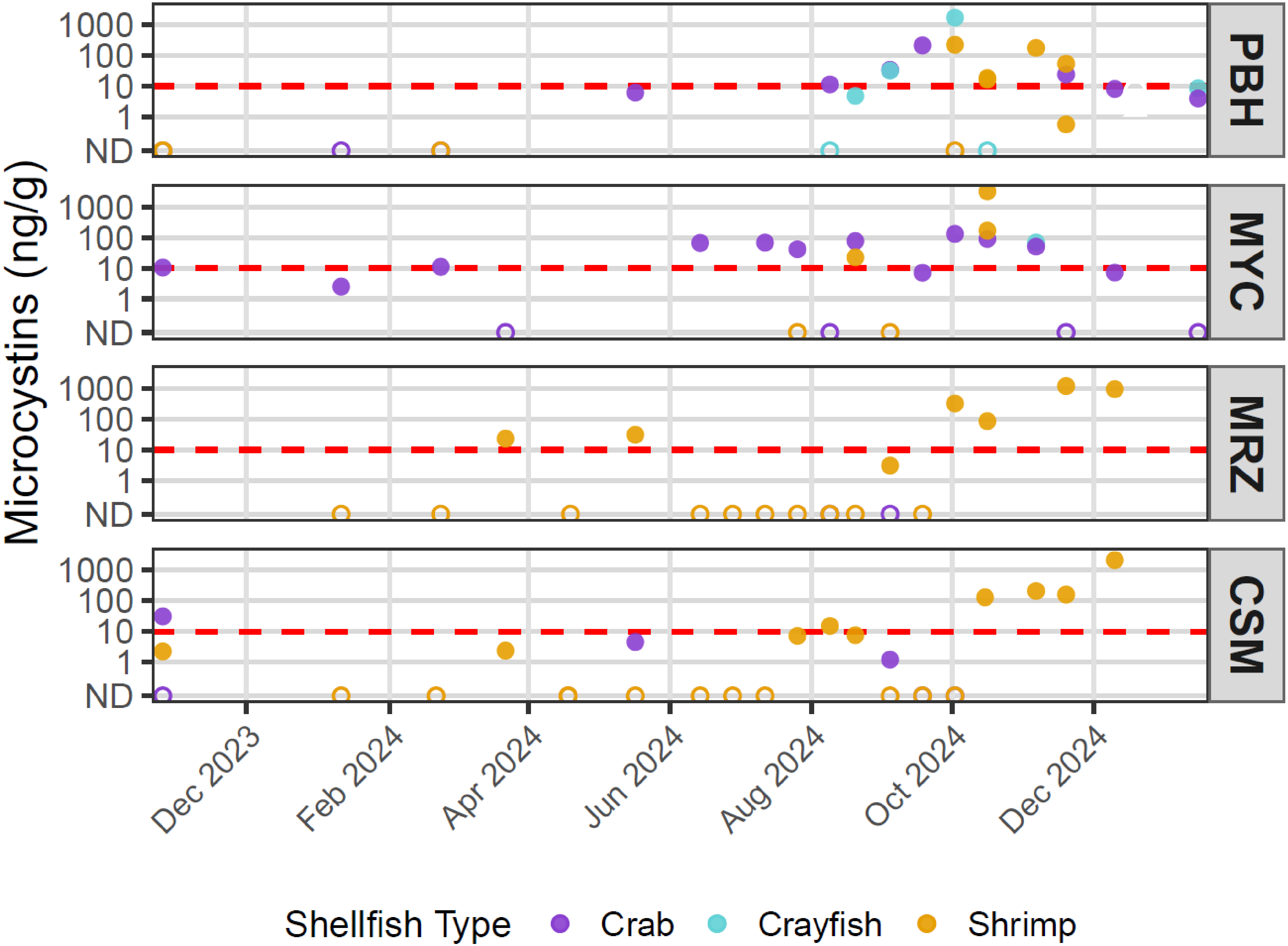
- *Microcystis* detected at most sites except western most sites (and MRZ)



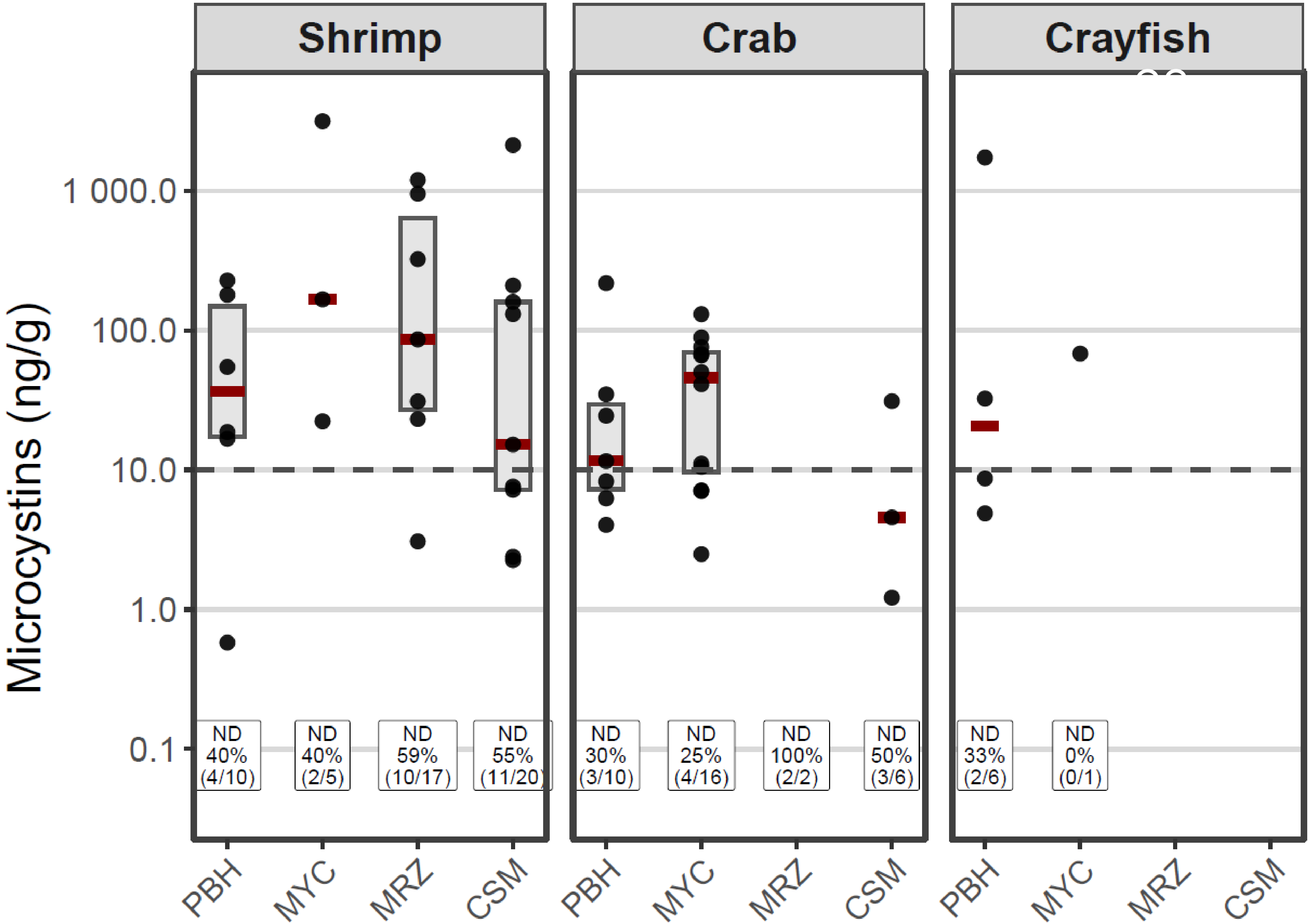


Microcystins in SPATT samplers





Microcystins (ng/g)
1000
100
10
1
ND
PBH
MYC
MRZ
CSM
Dec 2023
Feb 2024
Apr 2024
Jun 2024
Aug 2024
Oct 2024
Dec 2024
Shellfish Type
Crab
Crayfish
Shrimp



Shrimp
Crab
Crayfish
Microcystins (ng/g)
1 000.0
100.0
10.0
1.0
0.1
ND 40% (4/10)
ND 40% (2/5)
ND 59% (10/17)
ND 55% (11/20)
ND 30% (3/10)
ND 25% (4/16)
ND 100% (2/2)
ND 50% (3/6)
ND 33% (2/6)
ND 0% (0/1)
PBH
MYC
MRZ
CSM
PBH
MYC
MRZ
CSM
PBH
MYC
MRZ
CSM

Conclusions

Objective 1

- Analyzing animal tissues for MCs can be challenging
- Matrix specific issues with analyzing Asian clams via LC-MS
- Strata-X offers a promising option

Objective 2

- Widespread MC contamination of shellfish across the Delta
- We confirm the utility of shellfish for sentinel MCs monitoring in the Delta

Objective 3

- MC contamination along the salinity gradient
- Further study is necessary to confirm transport of toxins vs in situ production

Next steps

- Analyze 2025 shellfish samples
- Determine if domoic acid from the Bay is contaminating shellfish along the salinity gradient

Final Thoughts

- MCs produced in freshwater can extend beyond their source and persist across the freshwater to marine continuum
- Transport (through flow/tides) can allow MC accumulation in biota (and sediments) far from visible blooms
- These findings highlight the need for monitoring approaches that match the scale of the system

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Questions?

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Microcystins in the benthic food-web of the Sacramento-San Joaquin Delta, California

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Stockton Channel Turning Basin, July 2012
Photo Courtesy of Cyle Moon

References

Brown, A., Foss, A., Miller, M.A. and Gibson, Q., 2018. Detection of cyanotoxins (microcystins/nodularins) in livers from estuarine and coastal bottlenose dolphins (Tursiops truncatus) from Northeast Florida. *Harmful Algae*, *76*, pp.22-34.

Camacho-Muñoz D, Waack J, Turner AD, Lewis AM, Lawton LA, Edwards C. Rapid uptake and slow depuration: Health risks following cyanotoxin accumulation in mussels?. Environmental Pollution. 2021 Feb 15;271:116400.

Gibble, C.M., Peacock, M.B. and Kudela, R.M., 2016. Evidence of freshwater algal toxins in marine shellfish: Implications for human and aquatic health. *Harmful Algae*, *59*, pp.59-66.

Preece, E.P., Moore, B.C. and Hardy, F.J., 2015. Transfer of microcystin from freshwater lakes to Puget Sound, WA and toxin accumulation in marine mussels (Mytilus trossulus). *Ecotoxicology and environmental safety*, *122*, pp.98-105.

Preece, E.P., Hardy, F.J., Moore, B.C. and Bryan, M., 2017. A review of microcystin detections in Estuarine and Marine waters: Environmental implications and human health risk. *Harmful algae*, *61*, pp.31-45.

Preece, E.P., Otten, T.G., Cooke, J. and Kudela, R.M., 2024. Microcystins in the benthic food-web of the Sacramento-San Joaquin Delta, California. *Science of The Total Environment*, *946*, p.174250.

Straquadine, N.R., Kudela, R.M. and Gobler, C.J., 2022. Hepatotoxic shellfish poisoning: Accumulation of microcystins in Eastern oysters (Crassostrea virginica) and Asian clams (Corbicula fluminea) exposed to wild and cultured populations of the harmful cyanobacteria, Microcystis. *Harmful Algae*, *115*, p.102236.

Yousaf, M., Wang, J., Rehman, A. and Li, Z., 2025. Microcystins in transitional and marine ecosystems: Source categories, distribution patterns, and ecological impacts. *Marine Pollution Bulletin*, *220*, p.118432.