PRELIMINARY 2025 Analysis of *Alexandrium* Cysts in Bed Sediment of the Salish Sea

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INTRODUCTION

Alexandrium are a genus of dinoflagellate that produce cysts that lay dormant in marine bed sediments. These cysts can remain viable for extended periods of time and accumulate within the sediment until germination as shown in Figure 1. *Alexandrium* cysts are particularly harmful as they can form harmful algal blooms (HABs) that are linked to paralytic shellfish poisoning (PSP). PSP is particularly harmful to both human health as well as marine health.

In this study, University of Washington Tacoma researchers used marine bed sediment samples collected by the Washington Department of Ecology's Marine Sediment Monitoring Team (Weakland et al. 2023) from various locations throughout the Salish Sea as part of the 2023-2028 Puget Sound Sediment Monitoring Program. We hypothesized that there is *Alexandrium* cysts present in the Salish Sea.

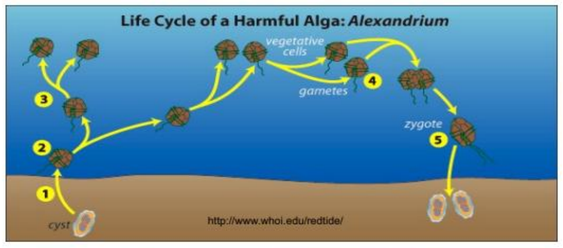

Fig. 1. The lifecycle of *Alexandrium catanella.* Images (from left to right) show resting cyst phase in visible light, chained vegetative cells, and stained cyst under epifluorescent light. Modified from Greengrove 2021 .

QUICK FACTS

>Saxitoxin is produced by *Alexandrium* can cause PSP in humans and marine mammals. (Lefebvre et al. 2025).

>Warmer seasurface temperatures positively correlate with *Alexandrium* blooms (Moore et al. 2015)

>Ongoing cyst monitoring helps establish future HAB hotspots (Trainer and King 2023).

METHODS

FIELD SAMPLING

Alexandrium cyst sediment was gathered through the 2023-2028 Puget Sound Sediment Monitoring Program run by the Washington Department of Ecology Marine Sediment Monitoring Team (Weakland et al. 2023) Analyzed samples were scooped from the top 0-1 cm of van Veen collections from 11 different stations located throughout the Salish Sea. Both 2025 and 2026 samples were kept at 4°C in the dark until analysis.

LABORATORY PREPARATION

- Preparation of samples were adapted from the Yamaguchi et al. 1995 method for determining total cyst counts in sediment.
- **Subsampled** 5mL from each station sample.
- **Diluted** subsample 1:5 with filtered sea water (FSW).
- **Diluted** 10mL of diluted subsample further to 45mL total volume with FSW.
- **Sonicated** 1 minute to break down outer cyst membranes.
- **Sieved** with 90µm and 20µm sieves in series to further isolate cysts (90µm>cyst dimensions> 20µm).
- **Preserved** recovered material from the 20µm sieve using 14mL FSW and 0.75ml diluted formalin.
- **Etched** preserved sample using 10mL methanol.
- **Stained** etched sample with 10mL FSW and 2ml Primulin.
- **Rinsed** stained sample three times with 10mL FSW using vortexing, centrifugation, and aspiration.
- **Loaded** Sedgwick-Rafter counting chamber with 1mL of stained sample.
- **Counted** stained cysts under epifluorescence microscope at 100X magnification using fluorescein isothiocyanate (FITC) filter set.

DISCUSSION

- Port Gardner had the highest cyst accumulation for wet and dry sediment samples (35 cyst/cc and 59 cyst/cc respectively).
- Commencement Bay and Nisqually National Wildfire Refuge had the lowest (0 cyst/cc).
- As median grain size increased cyst accumulation increased in dry sediment and vice versa in wet.
- Outlier shown in Fig. 6 might suggest errors in procedure or alter correlation between median grain size and cyst accumulation.
- No significant correlation between TOC% and cyst accumulation.

RESULTS

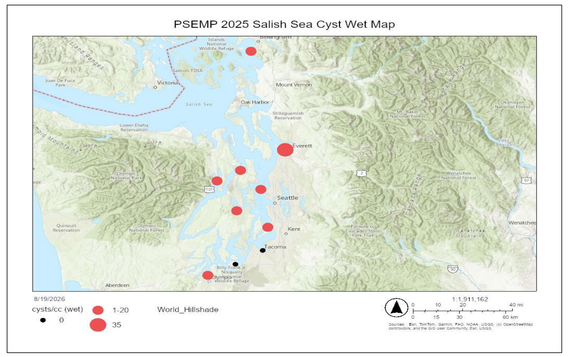

Fig. 2. 2025 cyst distribution in wet sediment sampled from the Salish Sea

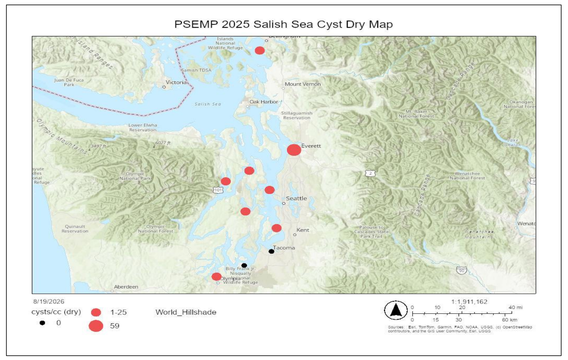

Fig. 3. 2025 cyst distribution in dry sediment sampled from the Salish Sea

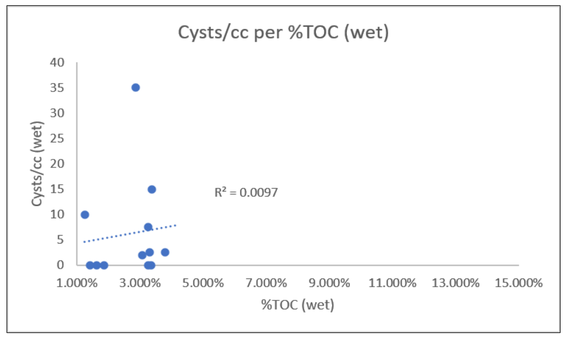

Fig. 4. Linear regression of %Total organic content of 10 samples from 2025 and their cyst counts in wet sediment.

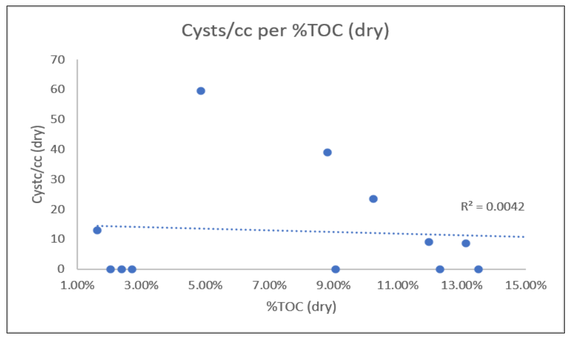

Fig. 5. Linear regression of %Total organic content of 10 samples from 2025 and their cyst counts in dry sediment.

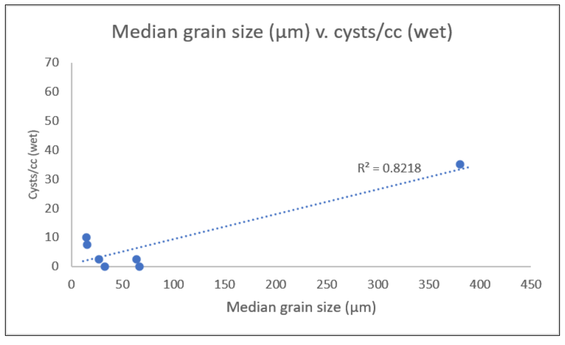

Fig. 6. Linear regression of median grain sizes of 10 samples from 2025 and their cyst counts in wet sediment.

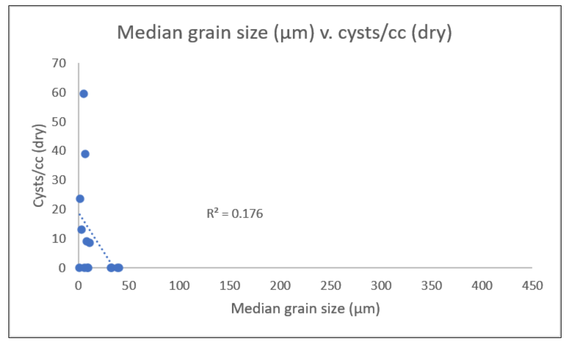

Fig. 7. Linear regression of median grain sizes of 10 samples from 2025 and their cyst counts in dry sediment.

Graphs' Correlation Summary

	Median Grain Size (µm)	TOC (wet)	TOC (dry)
Cysts/cc (wet)	Positive correlation	No correlation	No correlation
Cysts/cc (dry)	No correlation	No correlation	No correlation

REFERENCES