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Longitudinal study of paralytic shellfish toxins along Canada's coast

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ABSTRACT

Paralytic shellfish toxins (PST) in shellfish products have led to severe risks to human health. To monitor the risk, the Canadian Shellfish Sanitation Program has been collecting longitudinal PST measurements in blue mussel (*Mytilus edulis*) and soft-shell clam (*Mya arenaria*) samples in six coastal provinces of Canada. The spatial distributions of major temporal variation patterns were studied via Functional Principal Component Analysis. Seasonal increases in PST contamination were found to vary the most in terms of magnitude along the coastlines, which provides support for location-specific management of the time-sensitive PST contamination. In British Columbia, the first functional principal component (FPC1) indicated the variance among the magnitudes, while FPC2 indicated the seasonality of the PST levels. The temporal variations tended to be positively correlated with the abundance of dianoflagellates *Alexandrium* spp., and negatively with precipitation and inorganic nutrients. These findings indicate the underlying mechanism of PST variation in various geographical settings. In New Brunswick, Prince Edward, and Nova Scotia, the top FPCs indicated that the PST contamination differed mostly in the seasonal increase of the PST level during summer.

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1. 1. introduction

Paralytic Shellfish Toxins (PST), the neurotoxins produced by particular marine algae species including the dinoflagellates *Alexandrium* spp., can be of hazardous levels in shellfish through bio-accumulation (Bricelj and Shumway, 1998). Human consumption of such contaminated shellfish products would lead to Paralytic Shellfish Poisoning (PSP). This can be especially common in coastal regions where there is regular shellfish harvesting and consumption. PSP cases have been reported to cause illnesses globally, e.g., in southern China (Anderson et al., 1996), Alaska (Gessner and Middaugh, 1995), Venezuela (Barbera-Sánchez et al., 2004), and Mexico (Mee et al., 1986). In Canada, two cases occurred in Nova Scotia (NS) in 1977, and the symptoms of PSP have been found to include numbness from the lips, tongue, and fingertips to the legs, arms, and neck, which can get exacerbated into respiratory distress and muscular paralysis (Acres and Gray, 1978). In an effort to prevent such hazards placed on public health, monitoring of PST contamination has been widely conducted, e.g., by the Food and Drug Administration in the United States and the European Food Safety Authority in Europe.

The Canadian Shellfish Sanitation Program (CSSP, https://inspection.canada.ca/preventive-controls/fish/cssp/eng/1563470078092/1563470123546) has been monitoring for multi-decades the toxin contamination. The monitoring takes place in various shellfish product samples at shellfish harvesting sites along the coasts of British Columbia (BC), Quebec (QC), New Brunswick (NB), Prince Edward Island (PE), Nova Scotia (NS), and Newfoundland and Labrador (NL). Over time, the PST levels at sites were assessed, and shellfish harvesting schedules, e.g., closures and re-openings, of specific coastal regions were managed accordingly. Common assessments included the classification of the sites based on their PST contamination levels for the determination of the

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corresponding harvesting schedules and the assignment of risk levels based on the measurements at the sites. According to the risk ranking based on whether the contamination reached thresholds established by Health Canada, closure and reopening schedules were assigned to individual sites. The frequency of sampling was uneven, higher when the risk was high, lower (or even no sampling) when the risk was lower, which led to high dimensionality and irregular sparseness in data distribution and poses a challenge in pursuit of a macro-scale analysis to provide support for managing the harvesting sites and understanding the temporal and spatial PST variations across the coastal environments in each province.

Machine learning techniques have been widely applied to extract patterns among high-dimensional data. Among them, Principal Component Analysis (PCA) is commonly used for finding the largest modes of variation by obtaining the eigenvectors, known as the Principal Components (PCs), which explain decreasing proportions of variance in the data from the variance-covariance matrix in the high-dimensional feature variables (Jolliffe and Cadima, 2016). The projections onto the top PCs that explain most of the variance, therefore, provide an efficient explanation of the data in a low-dimensional space. Unfortunately, when dealing with longitudinal data, which consist of repeated measurements at typically sequential time points, PCA does not utilize the inherent correlation between these measurements. A significant loss of information would occur as a result. Therefore, PCA does not cater to a longitudinal study of the historical records of PST contamination over decades.

The functional version of PCA, Functional Principal Component Analysis (FPCA), overcomes this limitation. The conditional estimation algorithm (PACE) was proposed to further achieve robustness against both the irregular sparseness and the noises during FPCA (Wang et al., 2016, Yao et al., 2005). As demonstrated in the previous geospatial data analysis of fecal coliform (You et al., 2022, 2023), reducing the time resolution of the longitudinal measurements is effective in handling the sparseness and the noise. This approach was employed in this study for the extraction of the temporal variation patterns of the toxin contamination in each province. The visualization of their spatial distribution provides a direct illustration of risk dynamics and helps manage shellfish harvesting. Additionally, in BC, correlation analyses regarding the relationship between PST on one side and precipitation, dinoflagellates *Alexandrium* spp., nitrate or nitrite, and phosphate on the other revealed an underlying mechanism in the surrounding environments of each coastal region. The study provides a tool and the findings for determining optimal shellfish harvesting schedules according to the historical PST variations and influences from correlated factors in the surrounding environments along Canada's coast.

2. 2. data and methods

2.1. 2.1. data

The CSSP has been monitoring shellfish harvesting sites to ensure compliance with safety standards regarding marine pollutants including PST. As mentioned above, sampling schedules could vary at individual sites based on PST levels and assessed risks. Control of the harvesting areas was carried out according to specified regulations. Details of these procedures can be found in the CSSP Manual (https://inspection.canada.ca/food-guidance-by-commodity/fish/canadian-shellfish-sanitation-program/eng/1527251566006/1527251566942?chap=0#s7c3).

The data include measurements of PST (μg/100 g saxitoxin equivalence of all PST compounds) in blue mussel (*Mytilus edulis*) and soft-shell clam (*Mya arenaria*) samples collected at 1466 sites across 6 coastal provinces in Canada: BC, QC, NB, PE, NS, and NL, from 2000 to 2020. The reporting limits of the toxin measurements were 40 μg/100 g before 2010 but updated nationwide to 0 μg/100 g after 2012. During the period of implementation of this update from 2010 to 2012, variations between 0 and 40 μg/100 g existed across different regions. The reporting limits were adjusted to 40 μg/100 g across the entire 21 years in all regions to standardize the data for our analysis.

Additional data were 1) precipitation records (in mm; cumulative over five days prior to sampling) from Meteorological Service of Canada (MSC) at the closest weather station along the coasts, and 2) concentrations of the dinoflagellates *Alexandrium* spp. (cells mL^{-1}) and inorganic nutrients including nitrite, nitrate, and phosphate (μM) recorded at different locations inside Strait of Georgia from Esenkulova et al. (2021).

2.2. Preprocessing

Similar to our previous work (You et al., 2023), the 21-year data were aggregated into 53 weekly averages in one year to focus on the seasonal variation patterns. Sites without weekly averages greater than 40 μg/100 g were filtered out. During the winter seasons along the coasts in Atlantic provinces, sampling was impractical due to surface icing, and data became unavailable in the winter weeks. As a result, a province-specific period was investigated in each Atlantic province: Week 10–52 in QC, Week 1–52 in NB, and Week 15–50 in NS (PE and NL were excluded for lacking satisfactory data). Finally, since long periods of missing data in a one-year scale would cause unreliable imputation and extrapolation of the temporal patterns, sites were considered valid if (1) the longest period of missing weekly averages was not longer than 4 weeks and (2) there were at least 30% of available weekly averages in the one-year scale in BC and in the investigated period in each of the Atlantic provinces. These filtering processes retained 138, 23, 46, 36, and 20 eligible sites for PST in blue mussel samples from BC, QC, and NS and soft-shell clam samples from QC and NB, respectively.

2.3. Functional Principal Component Analysis

The PST level in blue mussel or soft-shell clam samples at a site over a year was considered as a function of week, i.e., $X_i(j)$, where i was the index of a site, and j referred to the week (1–53). Let the recorded measurements be realizations of these functions. μ, the mean function, was first obtained as a function of week. A covariance surface that integrated the covariance of two distinct weeks across the sites could then be acquired as

$$\mathrm{V}(s,t) = n^{-1}\sum_{i=1}^{n}\{X_i(s)-\mu(s)\}\cdot\{X_i(t)-\mu(t)\}$$

where s and t were two weekly numbers, and n was the number of sites.

From the covariance surface V, a set of K orthonormal eigen functions $\varphi_k(j), k = 1,\ldots,K$, referred to as the Functional Principal Components (FPCs), were obtained by solving

$$\int_T V(s,t)\varphi_k(s)ds = \lambda_k\varphi_k(t)$$

where J denoted the domain of weeks and λ_k was the eigen value paired with φ_k. The FPCs were ranked based on λ_k in decreasing order, and the k-th FPC explained a part of the total variance proportional to λ_k. Consequently, the top FPCs corresponded to the strongest modes of variation, while the last ones represented those that seemed barely informative. Focusing on projections of the raw data onto the top K FPCs could thus lead to an efficient summary of the variation among the data. An optimal choice of K would contribute to a decent percentage of the variance being explained while avoiding overfitting. Commonly, K is chosen so that the majority (e.g., $\geq$95%) of the variance is cumulatively explained by the top K FPCs. Subsequently, $X_i(j)$ could be approximated by

$$X_i(j) \approx \mu(j) + \sum_{k=1}^{K} \beta_{i,k} \cdot \varphi_k(j) \quad [1]$$

where $\beta_{i,k}$ was the k-th FPC score assigned to the i-th site and was acquired as

$$\beta_{i,k} = \int_J [X_i(j) - \mu(j)] \varphi_k(j) dj \quad [2]$$

where J was the domain of weeks. As a result, the longitudinal measurements at individual sites were summarized into the corresponding FPC scores with respect to the chosen FPCs.

When the longitudinal measurements are sparse and noisy, the application of FPCA demands adjustments for this extended applicability. Yao et al. (2005) proposed a conditional estimation approach, termed principal component analysis through conditional expectation (PACE), to achieve this goal. The PACE approach assumes there is a Gaussian error term of mean zero and constant variance at $X_i(j)$ regardless of the sites and the weeks. An estimated mean function and an estimated covariance surface are imputed, respectively, via univariate and bivariate kernel functions. The covariance surface is further smoothed through cross-validation. The FPCs are then estimated from the discretized smoothed covariance surface. Finally, the FPC scores are estimated conditional on the available realizations of X_i for $\mathrm{i} = 1, \ldots, \mathrm{n}$. This approach was employed in this study via the FPCA () function from the “fdapace” package in R (Gajardo etal, 2021).

2.4. Downstream analyses

Similarly, measurements of the precipitation, the dinoflagellates *Alexandrium* spp., and inorganic nutrients such as nitrate, nitrite, and phosphate were aggregated into weekly averages (nutrient data collected in the year 2015 were excluded due to the abnormal weather in that year), and the precipitation levels were further approximated by FPCA to have at least 95% of the variance explained.

Correlation analyses were performed to illustrate the characteristics reflected by the FPC scores and the correlation between the toxin and environmental factors (particularly precipitation, *Alexandrium* spp., nitrate or nitrite, and phosphate) at sites in each province. Regression models were fitted for the former purpose, with FPC scores as the predictors and the quantified characteristics as the responses, and the corresponding R^2 and p-value measured the goodness-of-fit of the reflection. Specifically, R^2 is calculated as 1 minus the proportion of the residual sum of squares (between the true and the predicted characteristics) to the total sum of squares (between the true characteristics and their mean values). Therefore, it is a numerical value between 0 and 1. The two boundaries, respectively, indicate that the FPC scores explain none or all of the variance in the characteristics. As a result, an R^2 value close to 1 is expected when we assume the FPC scores are indicators of the characteristics. On the other hand, the p-value shows the probability of perceived (or more extreme) results under the null hypothesis that the FPC scores cannot indicate the characteristics. Hence, a small p-value (e.g., ≤ 0.05) is another proof that the FPC scores are statistically significant indicators of the characteristics. For the latter purpose, Spearman correlation tests were performed to evaluate the rank-wise relationship between pairs of variables at each site at a distinct location. The magnitude and the sign of ρ respectively indicated the strength and the direction (i.e., positive or negative, respectively, corresponding to synchronization or reverse synchronization) of the relationship, of which the significance was indicated by an accompanying p-value.

After the FPCA model on the PST concentrations was built for each province, additional sites were checked by projecting their toxin levels onto the FPCs to estimate the corresponding FPC scoresbased on Eq. (2). Subsequently, according to Eq. (1), the variation of the toxin level at a site could be approximated. Among the sites that were filtered out during preprocessing, those found to have a positive Spearman correlation between the raw toxin measurements and the approximation were recruited. As a result, 280, 52, and 27 sites for blue mussel samples were recruited in BC, QC, and NS, respectively. There were also 35 sites for soft-shell clam samples recruited in NB. These sites were marked as “Ex” in order to distinguish them from those retained after preprocessing (labelled “In”) in the visualization described below to provide auxiliary signals.

Visualization of the results at individual sites was mapped onto their geographical locations via the “ggmap” package in R (Kahle and Wickham, 2013) to gain a spatial understanding of the variation of the toxin levels and its correlation with the factors along the coastal marine environments. An interactive website presenting such information is available at https://opms.uvic.ca.

3. Results

3.1. Exploration in BC

3.1.1. PST in blue mussel

Our analysis started with PST measurements in blue mussel samples collected from BC. The mean toxin level depicted a seasonal elevation mainly around Week 25 and a second but less intensive one around Week 40 (Fig. 1a). Most variance in toxin levels concentrated during these two periods, as explained by the top 2 FPCs (Fig. 1b).

3.1.2. Temporal variation patterns

The FPC1 (Fig. 1b) depicts noticeable elevated toxin levels during the period of Week 10–50. Based on Eq. (2), a higher FPC1 score would thus indicate a larger magnitude of elevated toxin level during this period, i.e., larger toxin measurements during these weeks were expected at the corresponding site. Via a regression model, the FPC1 scores significantly reflect the maximum PST levels of the sites during this period (Fig. 2a: $R^2 = 0.83$, p-value $<2.2\text{e-}16$).

During the same 40-week period, FPC2 appeared negative before Week 33 while positive afterwards (Fig. 1b). The negative and positive peaks of FPC2 respectively coincided with the first and the second peaks of the mean toxin level (Fig. 1a). This contrasting shape of the FPC2 illustrates two periods of opposite variations from Week 10–50. According to Eq. (2), negative and lower FPC2 scores would be obtained at sites with higher-than-average PST levels around Week 25, exactly as revealed by the fact that the sites with lower FPC2 scores had peak toxin levels around Week 25, whereas the above-average PST levels around Week 40, reflected by positive and higher FPC2 scores, were consistent with the fact that PST levels at sites with higher FPC2 scores peaked around Week 40 (Fig. 2b). These findings indicated that the FPC scores were quantification of the corresponding characteristics. The FPC1 scores positively relate to the increases of PST level at the corresponding sites from Week 10–50 (Fig. 2a), while the FPC2 scores indicate the specific windows (via the sign) and the intensity (via the magnitude) of the elevated PST contamination during this period (Fig. 2b).

3.1.3. Spatial distribution of temporal variation patterns

The spatial distributions of such quantified characteristics were studied by mapping the FPC scores to the geographical locations of the corresponding sites. There appeared to be geographical differences in the periods of high toxin levels at the sites investigated on BC coast (Fig. 3). Inside the Strait of Georgia, most of the sites appeared to have relatively low to medium FPC1 scores, while the high and medium FPC2 scores there indicated that their high toxin levels appeared in the fall; their risks were not the main concern. For example, the PST levels at sites located near Surrey, the Shoal Islands, the Denman Island, the Marina Island, the Copeland Islands, and the Hardy Island tended to peak around Week 40, but the overall contamination magnitude during Week 10–50 was relatively low at these locations. Outside the strait, the medium to high FPC1 scores and low FPC2 scores along the Pacific coast between Vancouver Island and the north of Prince Rupert indicated that

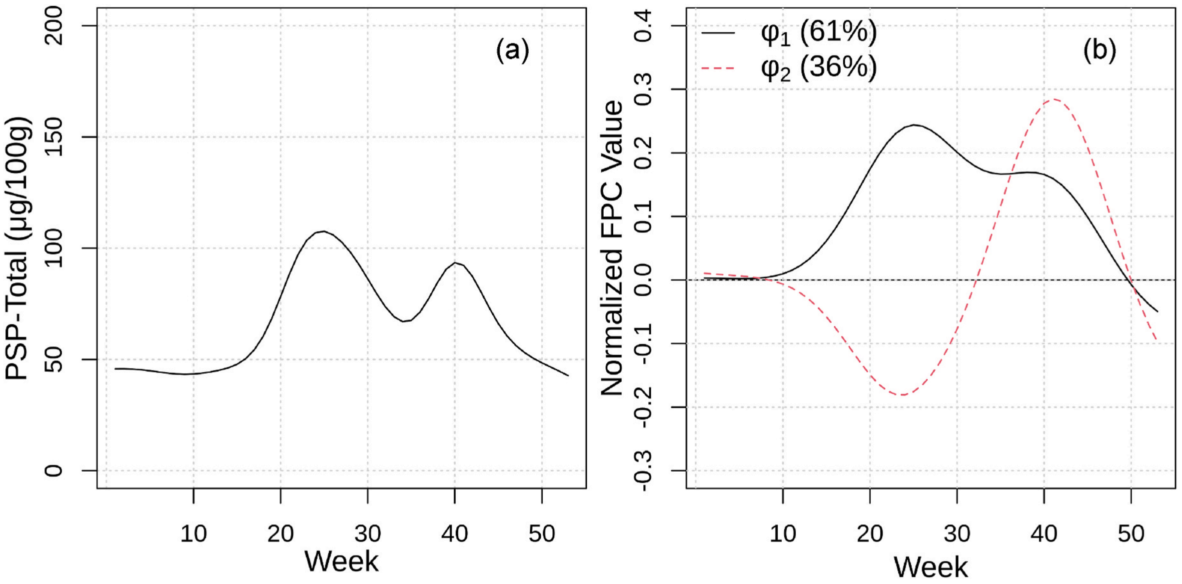

Fig. 1. FPCA results of the PST in blue mussel samples from BC: (a) the mean function of PST level; (b) the top 2 Functional Principal Components (FPCs), with proportions of variance explained in brackets.

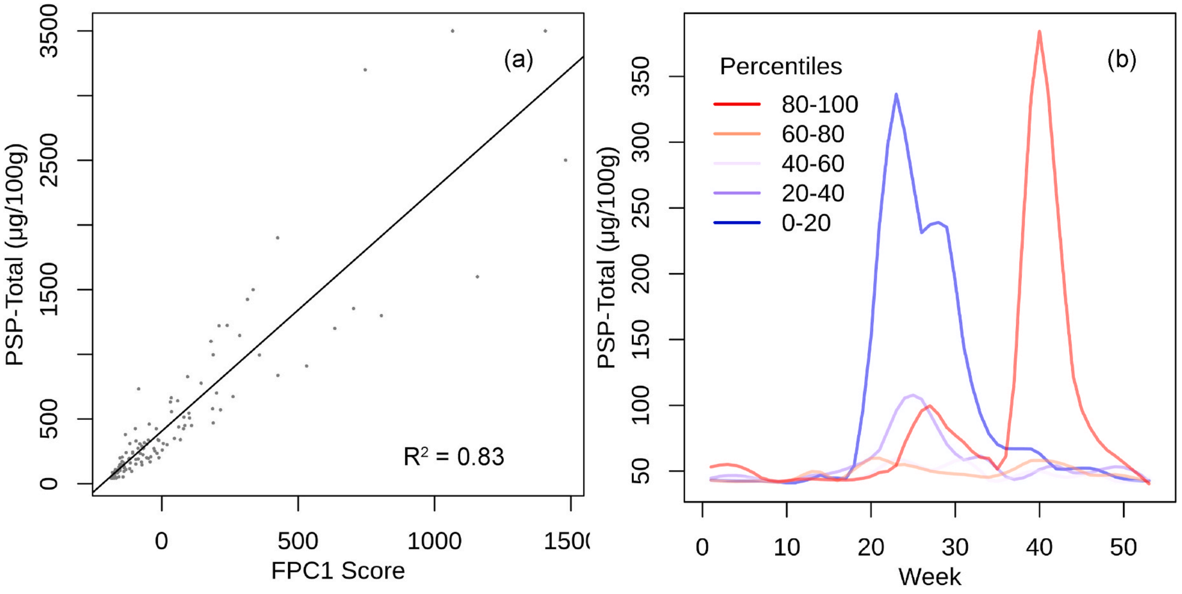

Fig. 2. Characteristics reflected by the FPC scores for PST in blue mussel samples from BC: (a) a higher FPC1 score reflected a higher maximum PST level at a site during Week 10–50; (b) according to the corresponding FPC2 scores, sites were sorted in increasing order and separated into 5 equal-sized bins based on the percentiles of their FPC2 scores as indicated by 5 sequential colors that were assigned to represent the relative magnitudes of the scores in the bins; the mean of the raw measurements at sites in each bin was smoothed by the Loess () function in R with the span parameter set to 0.2.

the relatively higher overall PST contamination at these sites and their peak toxin level appeared in the early summer. The risk levels at these sites would be a concern, as would the sites with high scores of both FPC1 and FPC2, which indicated their overall toxin levels were high and their maximum appeared in the fall, such as the regions surrounding the Broken Group and facing the Barkley Sound.

3.1.4. *Correlated environmental factors*

The spatial differences of the seasonal increased PST levels could relate to geographical features of surrounding environmental features. Available environmental factors including precipitation, *Alexandrium* spp., nitrate or nitrite, and phosphate were processed and investigated regarding their correlations with the PST level at nearby locations (the difference in latitude and longitude coordinates between the two parameters was less than 0.1). Negative correlations were found between PST and precipitation, most significantly along the west coast of Vancouver Island (Fig. 4a). As for the other factors, the potential correlations could require various time lags to take effect since they would not directly influence PST. Delays of 0-to-7 weeks were applied to the PST levels, i.e., the correlations were acquired between the other factors and the PST in weeks afterwards.

The dinoflagellates *Alexandrium* spp. in the Strait of Georgia were mostly found to be positively correlated with toxin levels (Fig. 4b). The strongest correlations were found throughout the seven weeks in the St. Vincent Bay (Site #21) and the Goliath Bay (Site #16) near the Jervis Inlet (Fig. 4e), which seemed to have influenced the Agamemnon Channel (Site #2) where there was a progressively more significant correlation in a few weeks of delay. The sites near the Malaspina

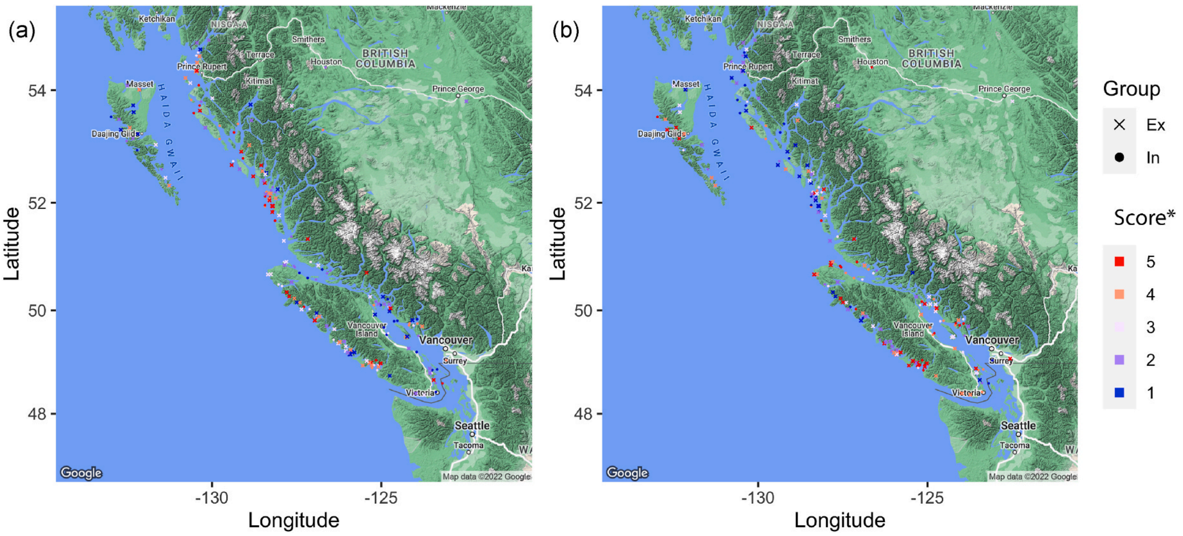

Fig. 3. Distribution of (a) FPC1 and (b) FPC2 scores regarding PST in blue mussel samples from BC: the scores were sorted in increasing order and separated into 5 equal-sized bins based on the percentiles of their FPC scores as indicated by 5 sequential colors that were assigned to represent the relative magnitudes of the scores in the bins; the shapes of the points indicate whether the sites were retained after preprocessing to build the FPCA model ("In") or excluded during preprocessing but brought back afterwards as the observations agreed with the FPCA findings ("Ex").

Provincial Park (Site #8) and the Patricia Bay (Site #20) were shown to have moderately positive correlations regardless of the delay applied to PST levels. Among the other sites, those near the Halfmoon Bay (Site #17), the Teakerne Arm Provincial Park (Site #13), Vesuvius (Site #12), the Powell River (Stie #6), Duncan (Site #9), Comox (Site #4), and Union Bay (Site #3) tended to positively correlate with PST levels in the first few weeks, whereas those near the Squirrel Cove (Site #11) and the Blubber Bay (Site #15) were found to have progressively stronger correlation as the delay prolonged.

The inorganic nutrients generally showed negative correlations with the PST (Fig. 4c and d). For all nutrients, the strongest negative correlations occurred near Vesuvius, where the surrounding locations appeared influenced (Site #1, #5: the Ganges Harbour, and #9); the negative correlations with the toxins were also consistent when a three-week delay was applied near the Finn Cove (Site #7) and the Malaspina Provincial Park. In the case of nitrate and nitrite, additional strong negative correlations appeared near the Squirrel Cove (Site #11) and the Jedediah Island Marine Provincial Park (Site #10). The p-values corresponding to the correlations are available in Supplementary Fig. S1.

3.2. Exploration in QC, NB, and NS

The same techniques were then applied to analyze the variation patterns among PST levels across the coastal environments in Atlantic Canada. The temporal variation patterns in the Atlantic provinces were similar across the coastlines (Fig. 5). In QC and NS, the FPC1 among the blue mussel samples in each province showed a seasonal increase during Week 25–40. Regarding the soft-shell clam samples from QC and NB, besides an identical period of increasing toxin levels, the FPC1 remains positive throughout the scale. Therefore, sites with higher FPC1 scores in each province tended to have larger magnitudes across the studied weeks and a higher seasonal elevation during Week 20-40 in their toxin levels (Fig. 6).

The spatial distributions of these patterns provide insights into the toxin levels at different locations along the coasts in these provinces (Fig. 7). Sites near Quebec city, those near St-Fabien, in between Portneuf-sur-Mer, Forestville, and Colombier that near or face Rimouski, and those surrounding the Bay of Fundy, e.g., from Meteghan towards Digby as well as the ones near Cape Chignecto Provincial Park, Kingsport, and Duncans Cove appeared to have relatively the highest seasonal increase during Week 25–40 based on their high FPC1 scores.

4. Discussion and conclusion

This study was guided to provide support for the management of shellfish harvesting sites along Canada's coast. Based on the multi-decade historical data, we aimed to find the temporal variation patterns and their spatial distributions as a tool for the governing agencies to come up with refined schedules for individual sites. With further analysis on how the environmental factors could potentially cause variations in paralytic shellfish toxins levels, the findings were expected to present evidence to enhance our understanding about the dynamics of PST contamination in the coastal marine environments. For this purpose, this paper illustrates the temporal-spatial variations and the potential influences of precipitation, *Alexandrium* spp., nitrate or nitrite, as well as phosphate on the PST content in blue mussels and soft-shell clams collected from the shellfish harvesting sites along the coastlines in BC, QC, NB, and NS. The application of Functional Principal Component Analysis to the weekly aggregation of toxin measurements extracts major temporal variation patterns as the top two FPCs, and the FPC scores assigned to each site provide a reflection of the magnitude and the seasonality of these temporal variation patterns in the dynamics of PST at each location.

The seasonal increases of PST centralized in the period from Week 25 to Week 40 are the major temporal variation of the differences in the PST levels along each coastline in BC, QC, NB, and NS. These weeks typically cover May to October, corresponding to the period from mid-spring to mid-fall in Canada. This is a period of higher surface water temperature and a higher amount of sunlight in a year, which are favorable to the growth of algae (Singh and Singh, 2015), including the dinoflagellates *Alexandrium* spp. that produce PST and are a positively correlated factor in our study. This relationship between warm temperatures and PST accumulation has also been suggested in Puget Sound, Washington, US, a region close to the south of Vancouver Island, where high PST levels exceeded the thresholds in blue mussels typically from July to November (Moore et al., 2009). The differences in the magnitudes of the seasonal increases during this period are attributable to the amount of water mixing, which typically brings up nutrients beneficial to algae growth (Baek et al., 2020). Therefore, larger magnitudes of seasonal increases of PST along the Pacific coast and the west coast of Vancouver Island in BC are observed in this study during the season of upwelling ocean currents along the Pacific coast of Canada (Freeland et al., 1984). In the Atlantic coastal environments, there are relatively higher increases in PST

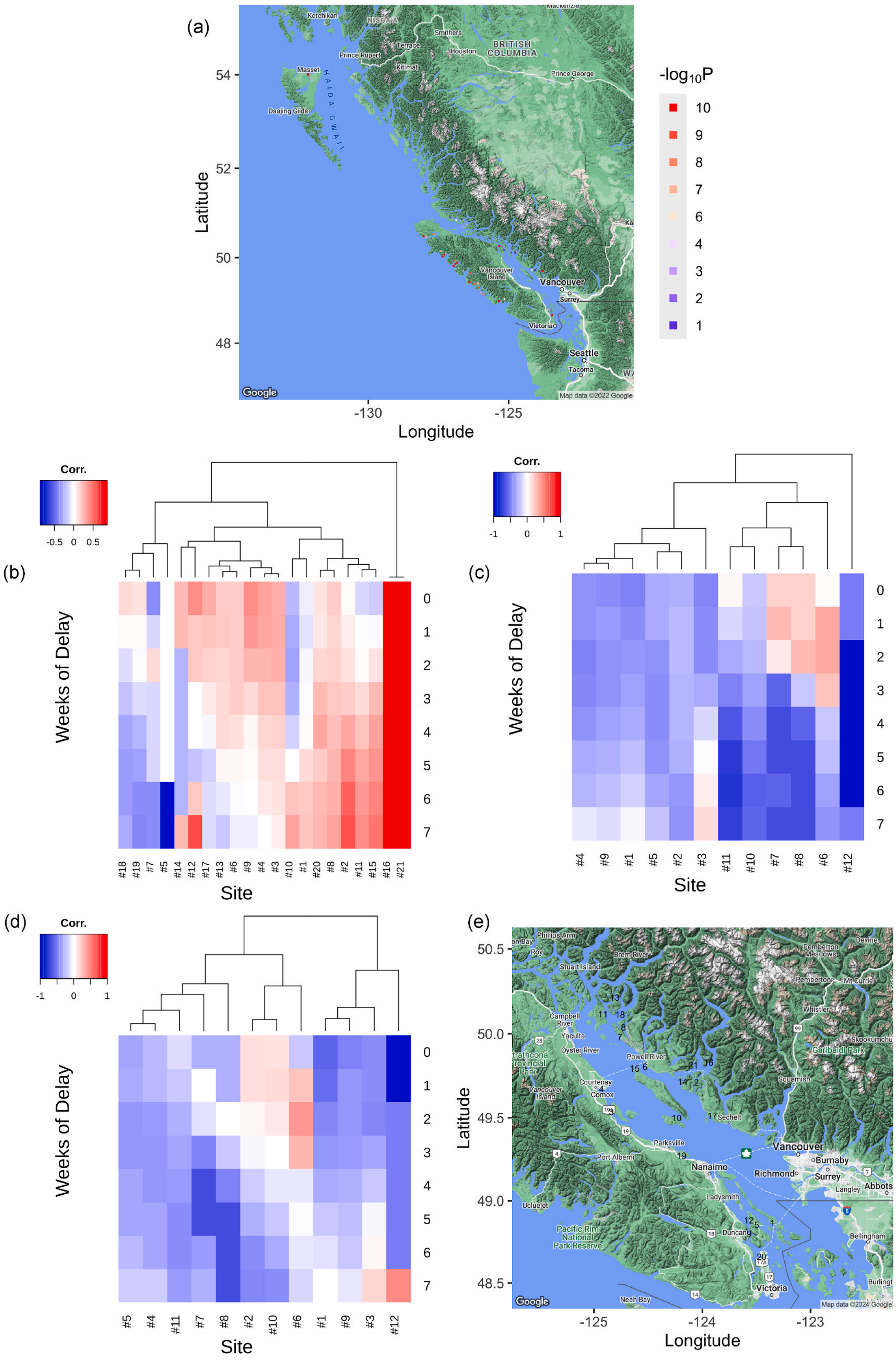

Fig. 4. Factors that were found to correlate with PST in blue mussel samples from BC: (a) correlation significance between the approximated PST levels and the approximated precipitation levels (note: only the significant correlations are plotted); the Spearman correlations between the 0-to-7-week delayed value of the approximated PST level at each site and (b) Alexandrium spp., (c) nitrate or nitrite, and (d) phosphate levels; the dendrograms depict the clustering results; (e) the geographical locations of the sites.

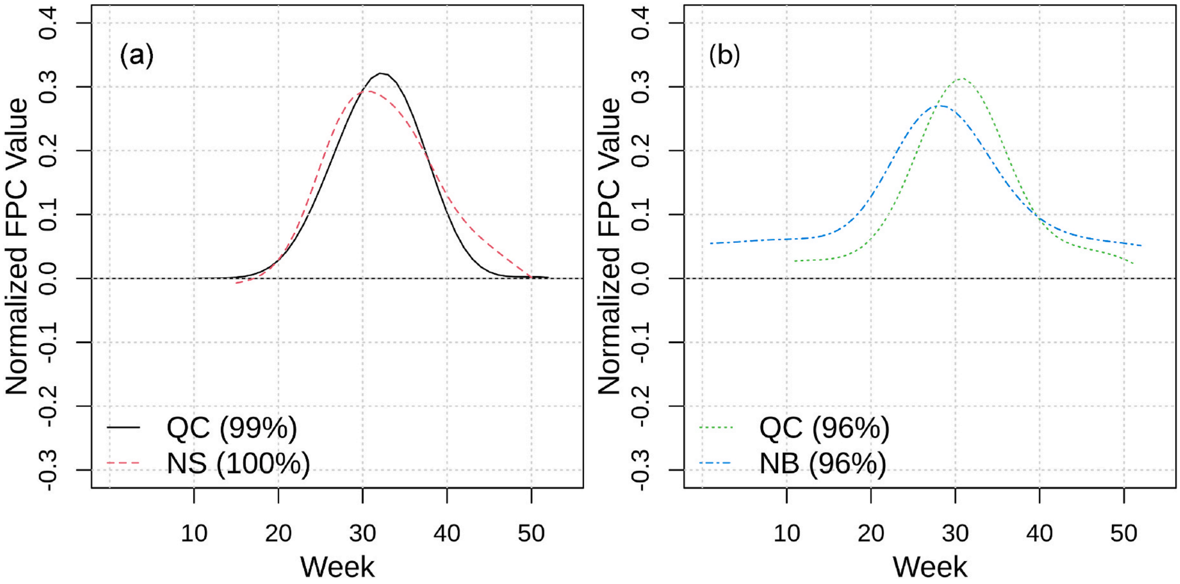

Fig. 5. FPC1 among (a) blue mussel samples in QC and NS, and (b) soft-shell clam samples in QC and NB; the proportion of variance explained by the FPC1 in each province is presented in the brackets.

around Week 30 in the Bay of Fundy, concurrent with the season of strong water mixing from May to September (Bumpus, 1960). Similarly, high PST in the region facing Rimouski is potentially influenced by the Rimouski Eddy (Blasco et al., 2003).

In BC, two specific short windows of relatively higher PST levels around Week 25 and Week 40, respectively, reflect different seasonal patterns in different geographical regions associated with distinct oceanographic characteristics. The highest PST contamination around mid-spring in the region along the north end of Vancouver Island is attributable to southeast ocean currents (Freeland et al., 1984), whereas the high mid-fall PST in the regions surrounding the Broken Group and those facing the Barkley Sound at the exit of Juan de Fuca Strait is attributable to ocean currents towards the northwest coast of southern Vancouver Island (Freeland et al., 1984). The difference in the seasonality and the directions of ocean currents in the different regions at warm temperatures would favor the growth of microalgae, including *Alexandrium* spp..

Recognizing these temporal patterns of increasing biotoxin levels allows for better timing of monitoring efforts and mitigation strategies. Based on the found seasons having elevated PST contamination, monitoring activities can be intensified during those periods. Pre-emptive closures of shellfish harvesting sites can also be implemented prior to the seasons. Moreover, understanding the duration and intensity of seasonal elevation periods can help in determining and aligning the appropriate duration of closures to shellfish harvesting or the application of other mitigation measures such as toxin removal techniques. The spatial distributions of biotoxin levels can help to identify “hotspot” areas where PST levels are relatively high in terms of seasonal contamination magnitude. By pinpointing these areas, monitoring efforts can be focused more intensively on these regions. These regions may require more frequent and targeted monitoring to effectively track changes in biotoxin levels. By doing so, the CSSP could save enormous time and effort whilst achieving more effective shellfish harvesting management.

Environmental factors could be significant contributors to the spatial distributions of the seasonal variations of PST (Tan and Ransangan, 2015). Precipitation has been a source of freshwater and, thus, a cause of decreased salinity in the surface marine water. In our study, precipitation appears negatively correlated with the toxin contamination along the west coast of Vancouver Island, where freshwater runoff is driven by precipitation and higher PST risks are associated with less freshwater input (Finnis et al., 2017). Regarding the inorganic nutrients, the limitations of nitrogen (Poulton et al., 2005) and phosphorus (Anderson et al., 1996) have been reported to contribute to the production of PST. This agrees with the negative correlations inside the Strait of Georgia, with weeks of delay applied to the toxin levels.

Inevitably, limitation of this study exists and awaits future research to resolve it. This study extracted temporal variation patterns and visualized their spatial distributions. These findings provide practical support for managing the location-specific shellfish harvesting schedules and contribute to understanding of dynamics of PST along Canada’ coast. However, data at a large number of sites were not used in this study due to the extreme sparsity. Though this was necessary to achieve consistent and reliable results, some of the sites does appear to show PST levels worthy of noticing. Leaving these data out of our analysis might cause biases to the findings, e.g., there could be minor shifts from the actual seasons of elevated PST contamination, and the sites with the highest seasonal PST levels might be veiled in this study due to lack of data. Therefore, one direction for future research would be to integrate the sparse but worthy data into the analysis. Based on findings in the current study, we can focus on the found seasons with elevated PST contamination. More sites are expected to be included when these shorter periods are of focus, and the variations can be studied in more details. Meanwhile, the mechanisms that drive PST variations can be further studied if more data are obtained. Firstly, the roles of other environmental factors, such as temperature, salinity, nutrient availability, and oceanographic conditions, remain intriguing for their influences on toxin production, accumulation, and degradation. Secondly, various PST-producers including *Alexandrium* spp. can have different characteristics in terms of their PST-producing abilities, and different types of shellfish can have different modes of PST intoxication and depuration. These aspects can help in understanding the differences among the variations in much more details. Furthermore, land-based nutrient runoff, coastal development, and pollution, and their effects on phytoplankton communities and biotoxin production can be explored as part of the mechanisms. Finally, there might be interactions between PST and other pollutants. For instance, the microplasitics that originate in disposal of plastic debris can not only be found as an airborne pollutant but also exist in water environments through river flows and fishing-related actions (Patidar et al., 2023; Sathyamohan et al., 2023). It has been shown that macroalgae suffer from increasing amount of microplastics when drifting while microalgal production can be affected

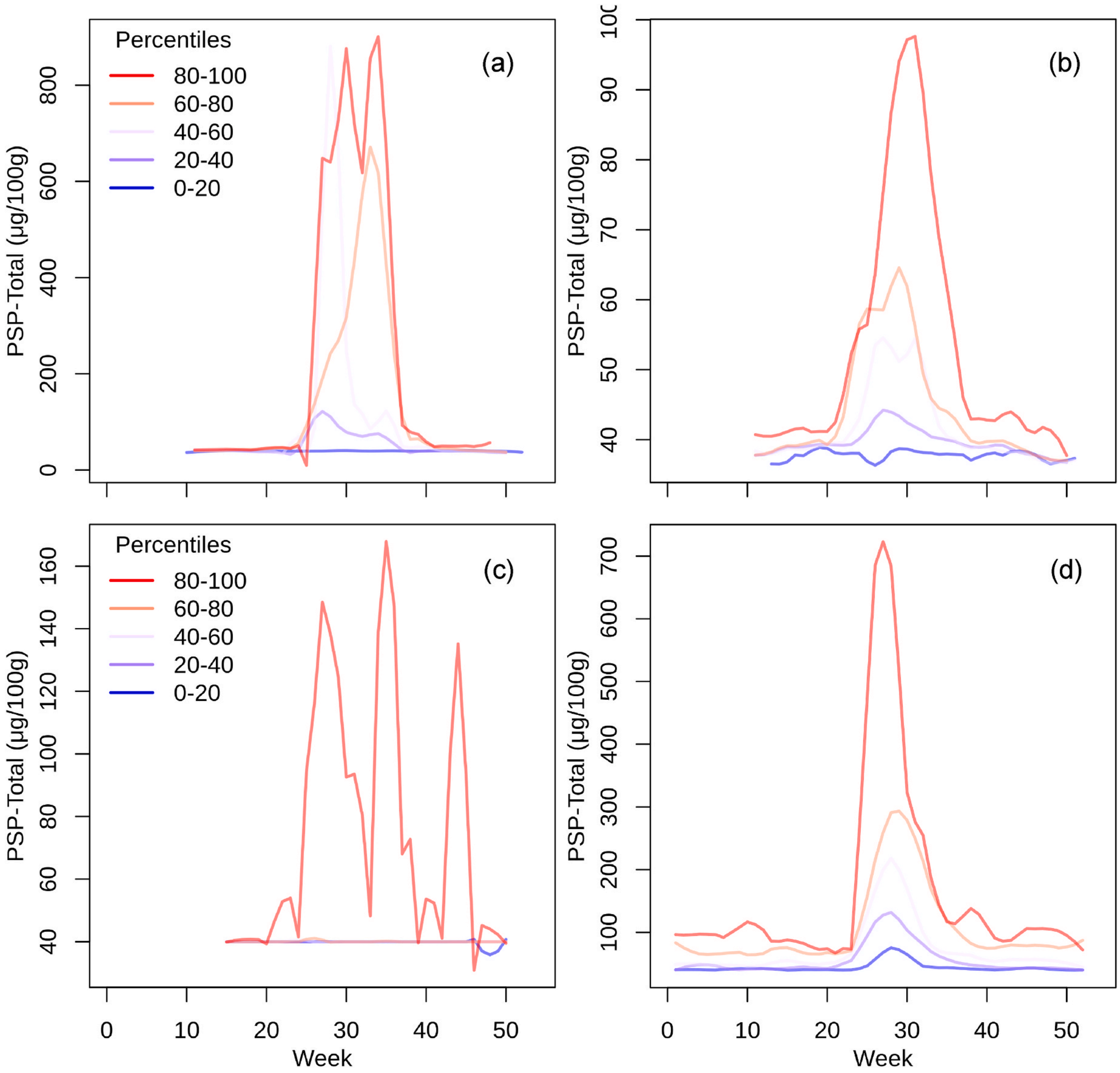

Fig. 6. Characteristics relfected by the FPC1 scores for PST in Atlantic provinces: sites were sorted in increasing order and separated into 5 equal-sized bins based on the percentiles of their FPC1 scores as indicated by 5 sequential colors that were assigned to represent the relative magnitudes of the scores in the bins; the mean of the raw measurements at sites in each bin was smoothed by the Loess () function in R with the span parameter set to 0.2, among blue mussel samples in QC (a) and NS (c), and softshell samples in QC (b) and NB (d).

in terms of quality and quantity by microplasitcs (Feng et al., 2020; Nava and Leoni, 2021). These findings suggest there could be potential influences of microplastics on the variations among PST levels in marine environments.

In conclusion, this longitudinal study investigates the temporal and spatial variation of paralytic shellfish toxins in blue mussels and soft-shell clams along Canada's coasts. FPC1 reveals the amplitude and FPC2 the seasonality of PST dynamics along the coast of British Columbia. Locations with higher FPC1 scores along the Pacific coast and the west coast of Vancouver Island have relatively higher overall PST contamination. The sites with extreme FPC2 scores in these regions and inside of the Strait of Georgia have a drastic seasonal contrast in the regional PST levels. Sites with high maximum FPC1 are the main risk concerns. The increase in PST is attributable to the growth of the PST-producing dianoflagellates *Alexandrium* spp. at warm temperatures and water mixing. In Atlantic Canada, increases in PST during the summer are the strongest in coastal regions with strong water mixing, such as the Bay of Fundy and the areas near Rimouski.

CRediT authorship contribution statement

Shuai You: Writing – review & editing, Writing – original draft, Visualization, Validation, Software, Formal analysis, Data curation, Conceptualization. **Li Xing:** Writing – review & editing, Methodology, Conceptualization. **Mary Lesperance:** Writing – review & editing, Supervision. **Youlian Pan:** Writing – review & editing, Supervision, Project administration, Methodology, Funding acquisition, Conceptualization. **Xuekui Zhang:** Conceptualization, Funding acquisition, Methodology, Project administration, Supervision, Writing – review & editing.

Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

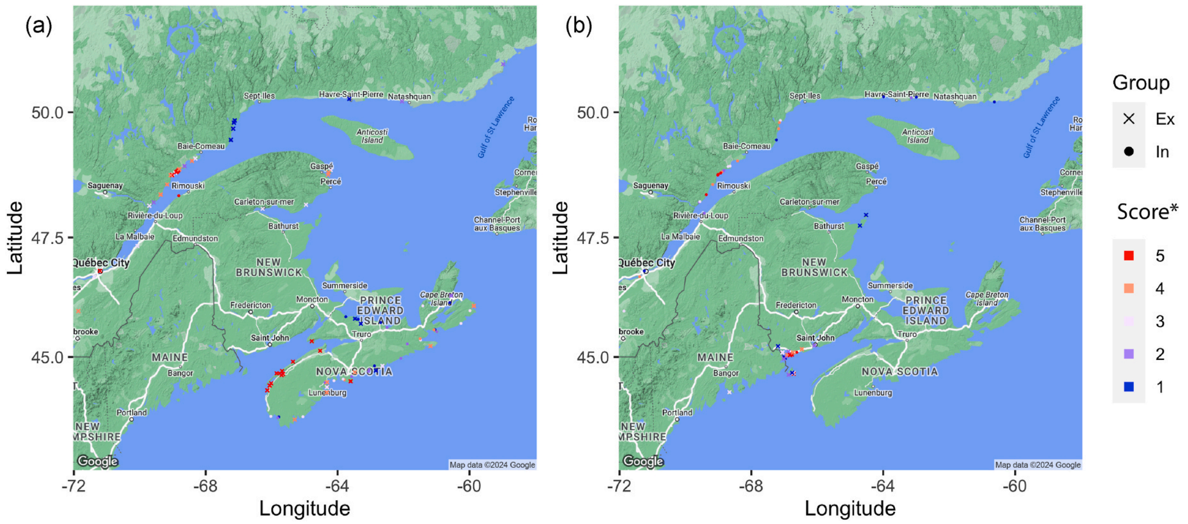

Fig. 7. Distribution of FPC1 scores regarding PST in (a) blue mussel and (b) soft-shell clam samples from Atlantic provinces: the scores in each province were sorted in increasing order and separated into 5 equal-sized bins based on the percentiles of their FPC1 scores as indicated by 5 sequential colors that were assigned to represent the relative magnitudes of the scores in the bins; the shapes of the symbols were used in the same way as in Fig. 3.

Data availability

The data that has been used is confidential.

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Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.envres.2024.118944.

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