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Performance of cold chains and modeled growth of *Vibrio parahaemolyticus* for farmed oysters distributed in the United States and internationally

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ABSTRACT

Vibrio bacteria can accumulate in molluscan shellfish and cause human diseases. The United States (U.S.) has implemented *Vibrio* Control Plans to mitigate risks associated with these bacteria, which include time and temperature requirements for post-harvest processing and maintaining an unbroken cold chain. In this study, we tracked the performance of cold chains for U.S. farmed oysters distributed nationally and internationally using temperature sensors. Boxes and bags of oysters (n = 125) were shipped from farms in Washington State and the Chesapeake Bay to 143 unique businesses in 20 U.S. states, Washington D.C., and Hong Kong, China. Eighty-one percent of the temperature sensors were returned with usable data. The average product temperature among all participants was 4.4 ± 2.7 °C (40 ± 5 °F), which is 5.6 °C (10 °F) cooler than the 10 °C (50 °F) guidance criterium established by the U.S. government. There were spikes in temperature in some shipments: 18% of shipments (16/91) experienced oyster temperatures above 10 °C for one hour or more, and the median time spent out of temperature control was 2.5 h. We modeled *V. parahaemolyticus* abundance using temperature sensor data and 75% (68/91) of shipments had a net decrease in *V. parahaemolyticus* abundance in the cold chain. There are opportunities for improvements in cold chain performance in the shellfish industry and related businesses. In the discussion we provide recommendations for oyster producers related to product cooling, for businesses that handle shellfish, and for government and industry groups to develop guidance for shipping by air, among other issues.

1. Introduction

Vibrio parahaemolyticus and *V. vulnificus* are Gram-negative bacteria that are becoming more prevalent due to increased temperature from climate change (Baker-Austin et al., 2017; Deeb et al., 2018) and can accumulate in molluscan shellfish and cause human diseases. *V. vulnificus* is the leading cause of death related to seafood consumption in the United States (U.S.) and *V. parahaemolyticus* causes self-limiting gastroenteritis (Bross et al., 2007; Newton et al., 2012). Oysters are the most valuable marine aquaculture species in the U.S. (NOAA, 2017). Twenty of 23 shellfish-producing states in the U.S. have developed *Vibrio* Control Plans (VCPs) to mitigate risks associated with these bacteria, which include time and temperature requirements for post-harvest processing and maintaining an unbroken cold chain (NSSP, 2015).

Time and temperature indicators (TTI) are widely used in the food industry to assess the microbiological quality of products (Biji et al., 2015). In this study, we used temperature data loggers to track the temperature of U.S. farmed oysters grown in Washington State and the Chesapeake Bay (Virginia and Maryland), the largest farming regions in the U.S. (NOAA, 2017; USDA, 2014), and distributed nationally and internationally. We then modeled the concentration of *V. parahaemolyticus* in cold chains. Based on our findings, we provide recommendations for the shellfish industry and stakeholders.

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Table 1
Number of companies participating in the study and number of shipments of oysters with temperature sensors.

Supply chain	Washington State	Chesapeake Bay[a] (Love et al., 2019a)	Total
Shipments			
Domestic	60	63	123
International	2[b]	0	2
Total	62	63	125
Company type			
Producer	7	6	14
Wholesale	35[c]	3	38
Restaurant	29	28	57
Food retail	4	4	8
Consumer	1	2	3
Freight carrier[d]	21	6	27
Total	97	49	147

[a] Includes several participants outside the Chesapeake Bay region that were not included in (Love et al., 2019a).
[b] Two shipments to Hong Kong, China.
[c] One wholesaler was a participant in both Washington State and Chesapeake Bay supply chain studies.
[d] Washington State: 7 air freight, 7 ground freight, 4 freight forwarders, 2 direct-to-consumer freight; Chesapeake Bay: 1 air freight, 4 ground freight, 1 direct-to-consumer freight.

2. Methods

2.1. Study design and temperature sensor methods

Temperature sensors (Smart Buttons, ACR Systems Inc., British Columbia, Canada) were inserted into live oysters and also taped to the outside of oyster bags or boxes to track temperature of shipments throughout the supply chain from harvest to retail. Methods are reported previously (Love et al., 2019a). The Chesapeake Bay portion of the study ran from February to September 2017, and we tracked shipments of Eastern oysters (*Crassostrea virginica*) primarily to businesses within the Chesapeake Bay region (Delaware, Maryland, Pennsylvania, Virginia, Washington D.C.) and a few national shipments. The Washington State portion of the study ran from April to July 2018, and we tracked Pacific oysters (*C. gigas*) shipments to local, national, and international customers.

We excluded wild caught oysters, shucked oysters, and frozen half-shell oysters from the study, as well as any product harvested outside of the Chesapeake Bay or Washington State. We visited all producers in the study to observe on-farm processing and to measure the temperature of the harvest water, ambient air at harvest, and during processing.

2.2. Participant recruitment

Participants were recruited by chain sampling methods. We started by recruiting oyster producers and wholesalers and then recruited their customers and suppliers into the study. Participants were contacted by phone, email, or in-person and given a 1-page description of the study and a consent form. The inclusion criteria for respondents were: employed at a company in the shellfish supply chain, over 18 years of age, and an English speaker. Producers were offered financial compensation for boxes of oysters used in the study, and all participants who completed a short interview were given information about their own cold chain. The study was reviewed by the Johns Hopkins School of Public Health Institutional Review Board.

2.3. Statistical analyses

Data from temperature sensors were downloaded using Trendreader (ACR Systems Inc., British Columbia, Canada), analyzed in Excel (Microsoft, Redmond, WA) and graphed in Prism (v7, GraphPad, La Jolla, CA). One-way ANOVAs were used to compare the mean temperature among groups, and Tukey's Multiple Comparison Test for pairwise *post*-tests. *t*-tests were used to compare mean temperatures between groups. We calculated the number of shipments that were above 10 °C (50 °F) or below 1.7 °C (35 °F) for one hour or more, and divided

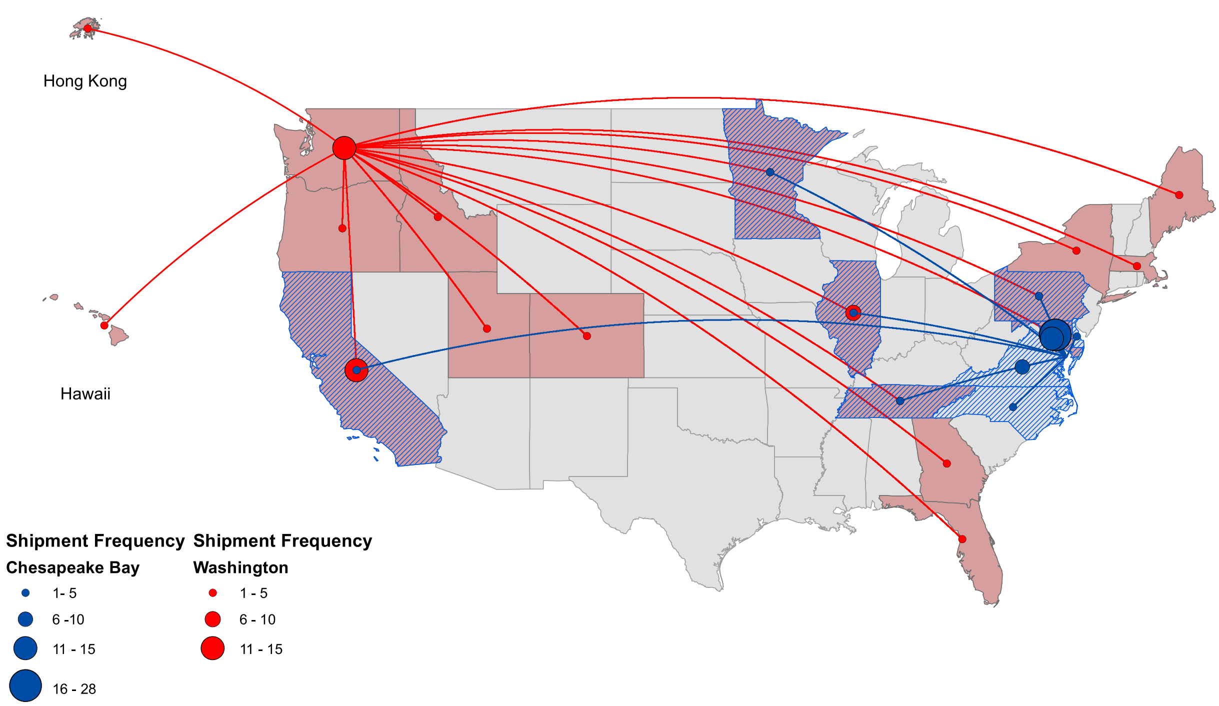

Fig. 1. Destinations for Chesapeake Bay oysters shipped in 2017 (blue, n = 63) and Washington State oysters shipped in 2018 (red, n = 62). Shipments were made to 20 states, Washington D.C., and Hong Kong, China. This figure includes all shipments made in the study, regardless of whether the temperature sensors were returned. In the Chesapeake Bay portion of the study, sensors from 51 of 63 shipments were returned, and in Washington State, sensors from 52 of 62 shipments were returned. (For interpretation of the references to colour in this figure legend, the reader is referred to the web version of this article.)

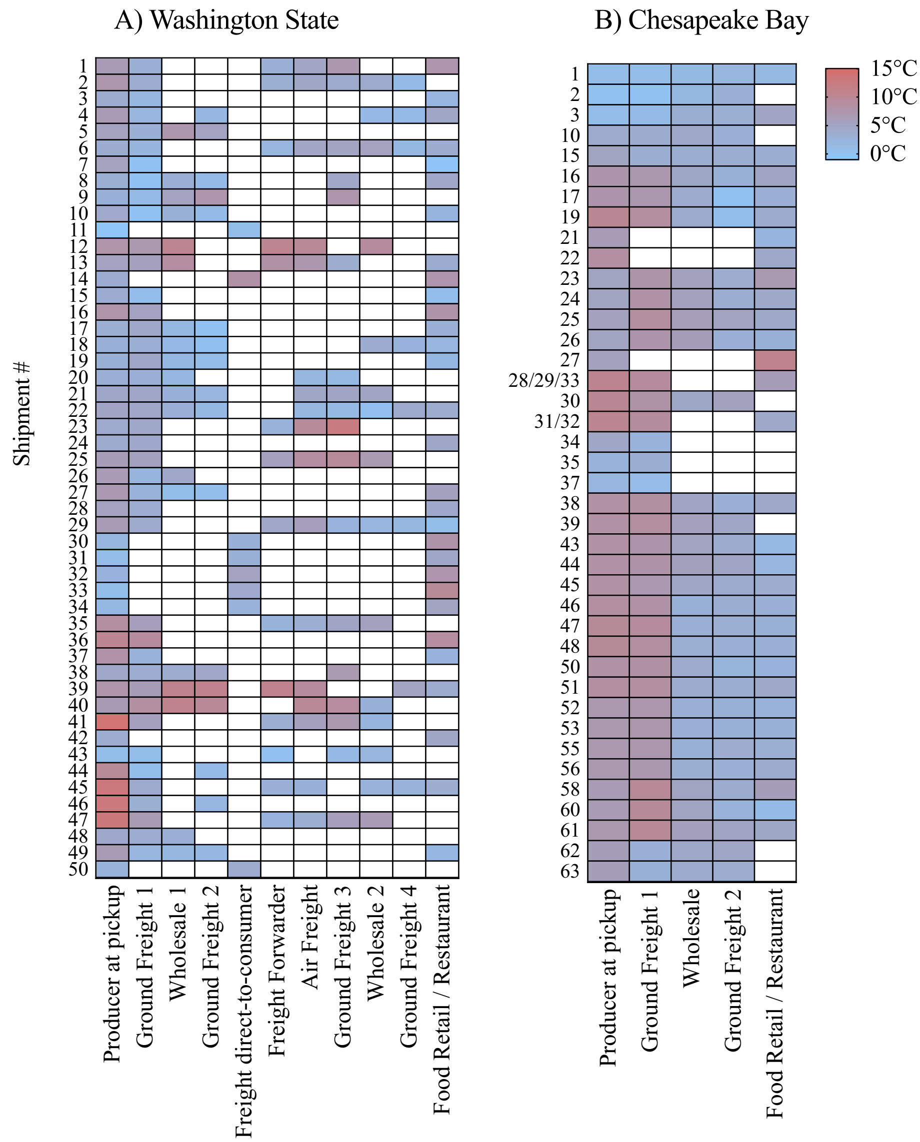

Fig. 2. Temperature gradient plots for all oyster shipments (rows) by stage of supply chain (columns) for A) Washington State oysters and B) Chesapeake Bay oysters. Each cell represents the average internal temperature of oysters. Supply chain stages that were not used are presented as empty cells.

that by the total number of shipments to determine the rate of shipments with temperature extremes. The U.S. National Shellfish Sanitation Program (NSSP) model ordinance recommends that shellfish products be held below 10 °C (NSSP, 2015). There is no consensus in the literature on the temperature that can kill oysters. Several sources report temperatures below 1.7 °C (35 °F), 1.1 °C (34 °F), 1.0 °C (33.8 °F), or 0 °C (32 °F) can kill oysters (Chiltern District Council, n.d.; ISSC, 2018; Marine Exension and Georgia Sea Grant, 2017), and we selected 1.7 °C (35 °F) as a conservative estimate.

2.4. *V. parahaemolyticus modeling*

We modeled the expected abundance of *V. parahaemolyticus* in oysters and the associated risk of gastroenteritis using internal oyster temperature data. Statistical models were based on the U.S. Food and Drug Administration's risk assessment of *V. parahaemolyticus* in raw oysters (FDA, 2005). The methods for *V. parahaemolyticus* modeling are described previously (Love et al., 2019a). In the Chesapeake Bay, models estimated *V. parahaemolyticus* abundance at harvest using water temperature. In Washington State, we used data from the *V. parahaemolyticus* shellfish monitoring program (courtesy of Washington State Department of Health) as a proxy for *V. parahaemolyticus* abundance at harvest. To protect the anonymity of participants we do not disclose the Washington State Department of Health sampling locations.

3. Results

3.1. *Study population*

Throughout the two year study, producers shipped 125 boxes or bags of oysters to customers in 20 states, Washington D.C., and Hong Kong, China (Table 1). Eighty-one percent of the temperature sensors were returned with usable data, and the return rates were similar for Washington State and Chesapeake Bay supply chains. Roughly equal numbers of shipments were made with Chesapeake Bay oysters as were made with Washington State oysters, however, the Washington State portion of the study had twice the number of participants. This was due to a change in the study methodology in Washington (as mentioned in Section 2.1) to include more national and international shipments.

Fig. 1 provides a map of the origin and final destination of shipments in this study. Participants were geographically dispersed and included 13 oyster producers from three states, 38 seafood wholesalers

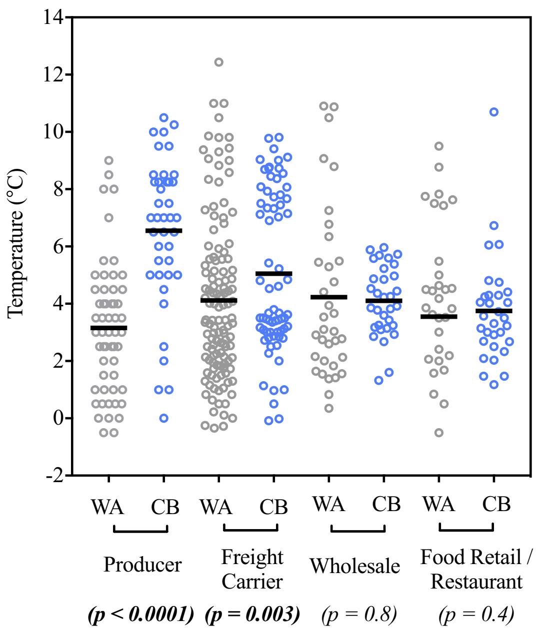

Fig. 3. Strip plot and mean (black bars) of internal oyster temperatures for Washington State (WA, grey) and Chesapeake Bay (CB, blue) farmed oyster supply chains. Sample sizes are reported in Table A.1. P values in bold are statistically significant. Y-axes are in °F and °C. (For interpretation of the references to colour in this figure legend, the reader is referred to the web version of this article.)

from 16 states, 63 grocery stores, seafood markets, and restaurants from 17 states, and 27 freight carriers, including 7 commercial airlines and 11 commercial trucking companies, that serviced markets ranging from local to international.

3.2. Summary of oyster cold chains

In Fig. 2 we provide a heat map indicating the average internal oyster temperature for each shipment at each stage of the supply chain. (Shipments are presented as rows of data in Fig. 2, which can be read from left to right as shipments move down the supply chain) Overall, oysters were maintained at an average temperature of 4.4 ± 2.7 °C (40 ± 5 °F) among all participants. Oysters harvested from the Chesapeake Bay were maintained 1.2 °C (2 °F) warmer in supply chains than oysters originating from Washington State (Fig. 3; see Table A.1 for means and p-values). These differences were primarily due to warmer oyster temperatures among Chesapeake Bay producers (3.4 °C warmer) and freight carriers (0.9 °C warmer) compared to Washington State producers and freight carriers (Fig. 3). There were no significant differences in oyster temperatures between wholesalers, food retailers, or restaurants who handled Washington State and Chesapeake Bay oysters.

3.3. Washington state oyster cold chains

Post-harvest cooling is a critical period to control the growth of *V. paraehemolyticus*. Washington State producers cooled oysters to an average temperature of 3.2 ± 2.3 °C (38 ± 4 °F) (Table A.1), and most farms achieved temperatures below 10 °C (50 °F) in three hours or less (Fig. 4). Once oysters left the farm they remained at a similar temperature across the remaining stages of the supply chain (ANOVA, p = .1).

Among freight deliveries, long-distance ground freight carriers (i.e., truck shipments > 24 h) maintained oysters at 1.6 ± 1.3 °C (35 ± 2 °F), which was significantly cooler than other forms of freight [freight forwarders (4.4 ± 3.3 °C; 40 ± 6 °F; p = .04), local ground freight carriers (4.2 ± 2.8 °C; 40 ± 5 °F; p = .009), or air freight carriers (5.5 ± 2.7 °C; 42 ± 5 °F; p = .001) (Fig. A.1)]. Oysters were warmer when freight carriers shipped in months with Vibrio Control Plans (VCPs) compared to a non-VCP month (*t*-test, p = .008, Fig. A.2). Among direct to consumer freight deliveries, one-day deliveries provide cooler oysters than two-day deliveries, however, even one-day deliveries can approach 10 °C (50 °F) if the receiver is located in a hot climate (Fig. A.3).

3.4. Chesapeake Bay oyster cold chains

An article by Love et al. provides key findings on Chesapeake Bay oyster cold chains (Love et al., 2019a). The current study pooled the Chesapeake Bay and Washington State datasets, including a reanalysis of the Chesapeake Bay data using five national shipments not included in the earlier publication (Love et al., 2019a). The major findings of the Chesapeake Bay study did not change after this reanalysis.

3.5. Washington state and Chesapeake Bay oyster shipments with high temperatures

Over the two year study, 18% of all shipments (16/91) and 16% of domestic shipments (14/89) exceeded 10 °C (50 °F) for one hour or more (Fig. 5a, Table A.2). The median amount of time these 16 shipments spent above 10 °C (50 °F) was 2.5 h (range: 1.3 to 62 h). The highest internal oyster temperature recorded in the study was 14.4 °C (58 °F). Washington State and Chesapeake Bay supply chains had similar rates of shipments over 10 °C.

Supply chain groups had the following rates of oyster shipments over 10 °C: freight carriers (7%), wholesalers (6%), producers (4%), and food retail/restaurants (2%). Freight carriers were higher than other groups mainly due to the higher rates of oysters over 10 °C that were shipped by air freight (35%) (Table A.2).

3.6. Washington state and Chesapeake Bay oyster shipments with low temperatures

Oysters can be at risk for freezing in cold chains. Roughly half of all shipments (48/91) had oyster temperature readings below 1.7 °C (35 °F) for one hour or more (Fig. 5b, Table A.3). The coldest internal oyster temperature recorded in the study was −2.2 °C (28 °F). Rates of near freezing were similar across all supply chain groups (Table A.3). Oyster shipments in the Washington State supply chains were often colder than oyster shipments in the Chesapeake Bay supply chains (Fig. 2, Table A.3).

3.7. Modeling V. parahaemolyticus abundance and health risks

Models of *V. parahaemolyticus* abundance and associated risks are presented for Washington State shipments (Fig. 6). The model found a net decrease in *V. parahaemolyticus* abundance throughout the supply chain (from harvest to food retail/restaurant) in 82% (41/50) of Washington State shipments. The highest modeled *V. parahaemolyticus* concentration in Washington State was 1135 *V. parahaemolyticus* per gram of oyster tissues, which corresponded to an illness rate of 0.84 per 100,000 servings. (A plot of modeled *V. parahaemolyticus* growth in Chesapeake Bay shipments is provided in (Love et al., 2019a)). In the Chesapeake Bay study, 66% (27/41) of shipments had a net decrease in *V. parahaemolyticus* abundance in the supply chain. The highest modeled *V. parahaemolyticus* concentration in the Chesapeake Bay was 115 *V. parahaemolyticus* per gram of oyster tissue or an illness rate of 0.07 per 100,000 servings (Love et al., 2019a). (We assumed a serving to be 12 raw oysters.). Overall, from both regions, 75% (68/91) of shipments had a net decrease in modeled *V. parahaemolyticus* abundance in the supply chain.

Fig. 7 provides the percent change in *V. parahaemolyticus*

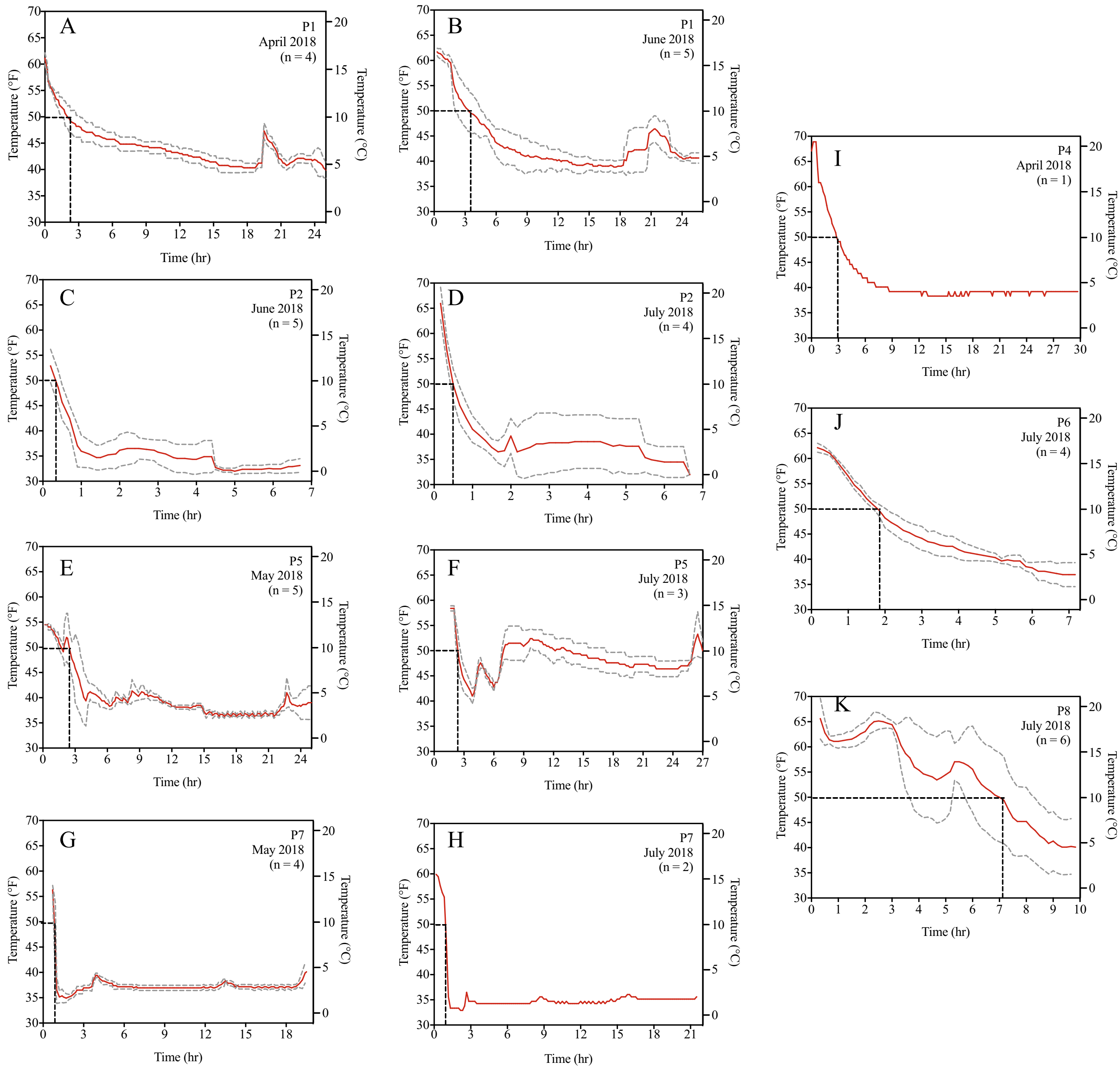

Fig. 4. Internal oyster temperature during harvest and on-farm processing at eight farms in Washington State. Graphs represent time from harvest to when the box or bag of oysters left the farm property. Y-axes are in °F and °C. Several farms had repeat visits (A and B; C and D; E and F; G and H). Grey lines indicate standard deviation, red lines indicate the mean. The sensors took readings at 10 min intervals. (For interpretation of the references to colour in this figure legend, the reader is referred to the web version of this article.)

abundance, by comparing the modeled *V. parahaemolyticus* concentrations at harvest to the modeled *V. parahaemolyticus* concentrations at the end of the supply chain.

4. Discussion

4.1. Main findings

Temperature control of shellfish during harvest, post-harvest processing, and throughout the distribution chain is essential to control *Vibrio* growth (Cook, 1994; Gooch et al., 2002). This study worked with a large and diverse cross-section of the shellfish industry to track the internal temperature of oysters in cold chains. On average, businesses in our study maintained oysters 5.6 °C (10 °F) cooler than the 10 °C (50 °F) guidance criterium established by the U.S. government (NSSP, 2015), however, temperature spikes occurred in some shipments. Temperature sensors indicated that 18% of oyster shipments exceeded 10 °C for an hour or more, and the median time spent out of temperature control was 2.5 h. Temperature exceedance rates were similar between Washington State and Chesapeake Bay cold chains, suggesting that there is internal validity to these findings. In the present study we report data by the origin of the oysters (Washington State or Chesapeake Bay), and in subsequent analyses we explored whether supply chain configuration (direct vs intermediated supply chains) affects temperature abuse, and report qualitative findings from interviews (Love et al., 2019b) The only other study of oyster cold chains we could identify was from Australia,

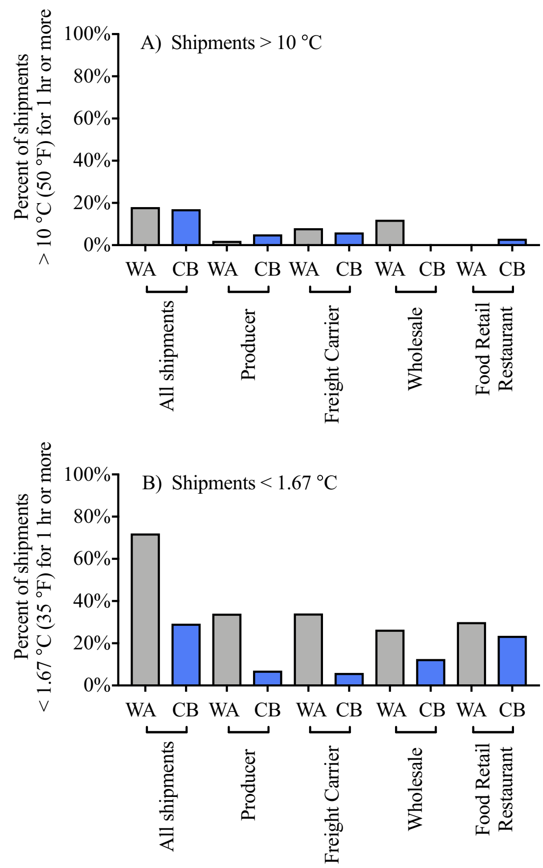

Fig. 5. Frequency of shipments and supply chain participants with internal oyster temperatures A) > 10 °C (50 °F) and B) < 1.67 °C (35 °F) for Washington State (WA, grey) and Chesapeake Bay (CB, blue) farmed oyster supply chains. Sample sizes are reported in Tables A.2 and A.3. (For interpretation of the references to colour in this figure legend, the reader is referred to the web version of this article.)

which found that 47% of shipments (21/45 shipments) were held above 10 °C (Madigan, 2008), which is more than twice the percentage found in the U.S.

Temperature control issues come in many different forms: from faulty mechanical refrigeration units, delays in ground or air transit, not icing products on docks or loading docks, or forgetting to refrigerate boxes upon arrival at restaurants. Two businesses in our study routinely had problems maintaining oyster temperatures and accounted for a third of all temperature exceedances in the study. These outliers, however, cannot be brushed aside. Temperature abuse anywhere in the supply chain can lead to food quality or food safety risks for consumers, and negative outcomes reflect poorly not just on the businesses involved but on the industry as a whole.

4.2. Freight carriers

Transportation is one area where participants reported issues with cold chain performance. Freight carriers in this study had higher rates of temperature abuse (both high and low temperatures) than other stages of the supply chain. Airlines had the highest rates of high-temperature abuse among all participants, and long distance ground freight had low-temperature abuse. Many consider that maintaining cold chains is a shared responsibility among all businesses, however, airlines have specific policies to the contrary. Airlines recommend packing perishable cargo to withstand 48 h outside refrigeration for domestic flights, and 72 h for international flights (American Airlines, 2018; Delta, 2018; United, 2018). Similar policies exist for direct to consumer freight carriers. Challenges in using air cargo shipments include the several hours of staging before the flight (including time spent on runways), no refrigeration under the belly of the plane for commercial passenger planes, and unexpected delays that can extend the time that oyster shipments go unrefrigerated. In addition, the temperature of walk-in refrigerators at airports may be set warmer than optimal for shellfish. In some cases, airlines choose those temperatures based on the most valuable cargo, and in the case of cut flowers, these plants require higher refrigeration temperatures than seafood (personal communication, air cargo representative).

4.3. Colder oysters in Washington State

Washington State oysters were maintained significantly cooler than Chesapeake Bay oysters, which is due to several possible factors: cooler harvest water temperatures, more restrictive VCP regulations, and colder temperatures in long distance freight trucks originating in Washington State. Washington State oysters had more instances of low-temperature abuse than Chesapeake Bay oysters as well. Two Washington State wholesalers in the study reported previously receiving frozen oysters, which confirms these risks exist outside of the study. In Australia, a country with an overall warmer climate, found just 18% of shipments (8/45 shipments) were below their temperature criteria of > 2 °C (35.6 °F) for Pacific oysters (*C. gigas)* or > 5 °C (41 °F) for Sydney Rock oysters (*Saccostrea glomerata*) (Madigan, 2008). Over-icing, freezing weather conditions, storage near a refrigeration condenser, or in a truck carrying frozen seafood can lead to cold abuse in seafood (Gokoglu and Yerlikaya, 2015; Madigan, 2008). The freezing point for oysters is −2.8 °C (27 °F) (Kolbe and Kramer, 2007) and others report that temperatures below about 1 °C can kill oysters (Chiltern District Council, n.d.; ISSC, 2018; Marine Exension and Georgia Sea Grant, 2017). There were mixed opinions among participants about what is a safe lower bound for refrigerating *C. gigas* oysters, and this issue could be resolved through laboratory studies.

4.4. V. parahaemolyticus modeling

Temperature data loggers combined with risk models provide a powerful tool for assessing cold chains. We found that 75% of oysters shipments had a net decrease in *V. parahaemolyticus* from harvest to food retail/restaurant. We attribute the findings of *V. parahaemolyticus* growth to either slow cooling during post-harvest processing or breakdowns in the cold chain. Harvesting and processing oysters in accordance with state VCP regulations did not always prevent *V. parahaemolyticus* growth according to the model. Conversely, when oyster temperatures were above VCP regulations at harvest, the models occasionally found no net growth of *V. parahaemolyticus* if other stages of the supply chain remained in compliance with VCP regulations. These 'exceptions to the rule' should be further explored.

The growth model we used was validated for *C. virginica* oysters (Parveen et al., 2013), but has not been validated for *C. gigas* oysters. The significance of our Washington State *Vibrio* model findings should be interpreted with caution until the model is validated. Others have developed *V. parahaemolyticus* models for oyster slurry (Yoon et al., 2008), which may not be relevant for whole live oysters, and *C. gigas* oysters (Fernandez-Piquer et al., 2013). Future work could compare these models to the U.S. Food and Drug Administration model under real world conditions, and attempt to validate the models using *V. parahaemolyticus* cultures at various stages of the supply chain.

4.5. Limitations

There were several limitations and issues to note. Our sampling was skewed toward warm seasons when temperature control is more challenging, therefore we expect that a random sample throughout the year would find lower rates of temperature abuse. Chefs and food retailers

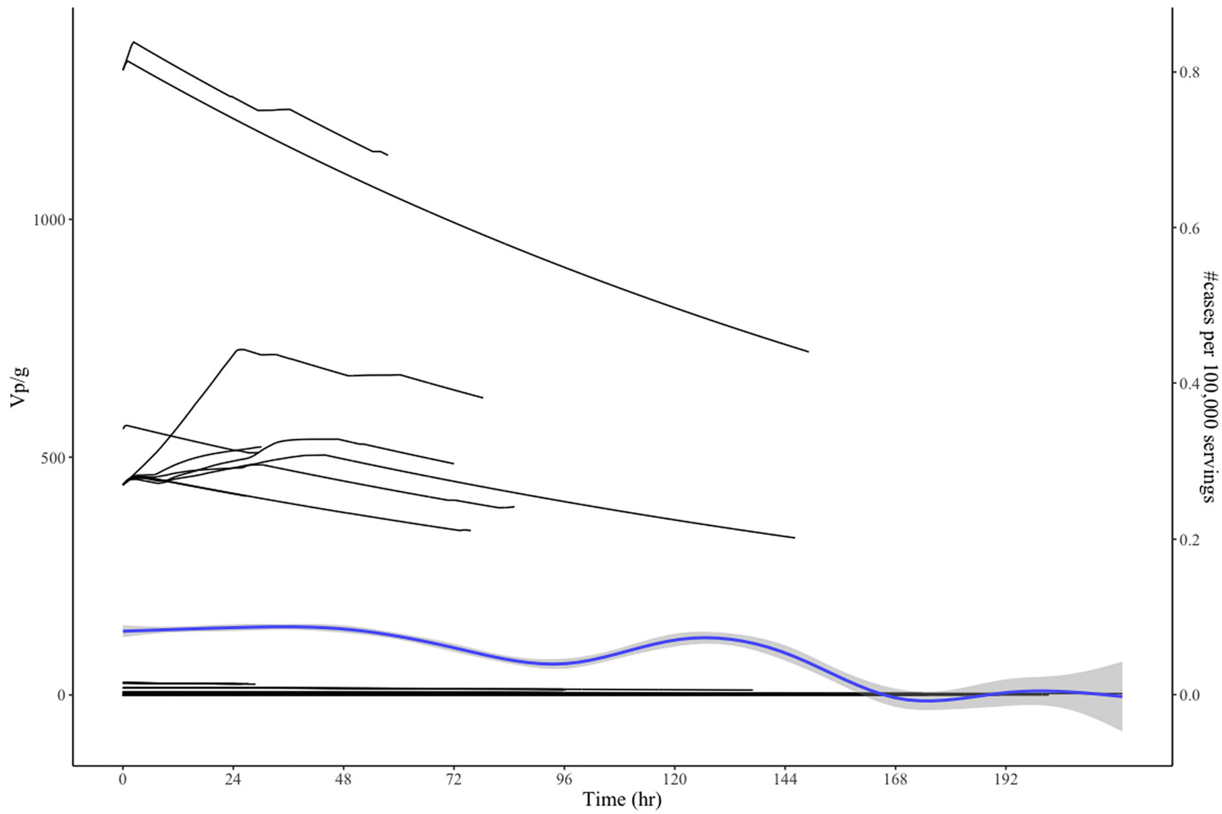

Fig. 6. Estimated *Vibrio parahaemolyticus* abundance per gram oyster tissue (left y-axis) and the risk of illness as cases per 100,000 servings (right y-axis) from oysters produced in Washington State and shipped locally, nationally, and internationally. X-axis represents time elapsed since harvest. Estimations of the left and right y-axes can be displayed simultaneously due to the linear approximation of the Beta-Poisson dose-response model. Vibrio abundance at harvest was estimated based on Washington State Department of Health *V. parahaemolyticus* monitoring, and growth in supply chains was calculated using iterative temperature-based models. The black lines represent the abundance or risk of individual oyster shipments. The blue line depicts the mean abundance or risk across all oysters estimated by using a generalized additive model and the grey band displays the corresponding 95% confidence interval. (For interpretation of the references to colour in this figure legend, the reader is referred to the web version of this article.)

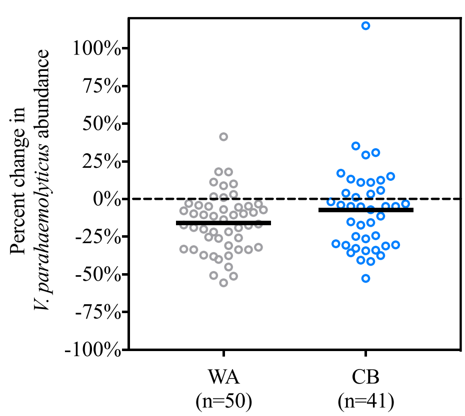

Fig. 7. Strip plot of modeled percent change in *Vibrio parahaemolyticus* abundance in Washington State oyster shipments (grey circles) and Chesapeake Bay oyster shipments (blue circles). The percent change was calculated as the final *V. parahaemolyticus* abundance (at food retail/restaurant) minus the initial *V. parahaemolyticus* abundance (at harvest) divided by the initial *V. parahaemolyticus* abundance. The dashed line represents no change in *V. parahaemolyticus* abundance. (For interpretation of the references to colour in this figure legend, the reader is referred to the web version of this article.)

indicated that up to 5% mortality was normal in a box of oysters, however, we did not assess whether warm or colder temperature in distribution caused these mortalities or any deterioration in food quality (i.e., changes in flavor, texture, colour, or liquor loss). We did not measure the actual amount of *V. parahaemolyticus* in oysters. We tracked just two shipments to Hong Kong, which limits our ability to make generalizations about international supply chains. Future work should focus on more international shipments.

4.6. Recommendations

Based on our findings, we offer a list of recommendations for the oyster industry; broken out into advice for oyster producers, businesses that handle shellfish, and policy.

4.6.1. Recommendations for oyster producers

- Review state *Vibrio* Control Plans (*Interstate Shellfish Sanitation Conference*, 2018) and strive to meet or exceed regulatory time and temperature requirements;
- Remember that the most critical windows in which to control the growth of *Vibrio* bacteria in the supply chain are immediately after harvesting and during post-harvest processing;
- Use ice slurries or layered ice for cooling, which have been found to be more effective to control the growth of *Vibrio* bacteria than mechanical refrigeration alone (Jones et al., 2017; Lydon et al., 2015; New Jersey Department of Environmental Protection, 2015).

4.6.2. Recommendations for businesses than handle shellfish

- Verify that Hazard Analysis and Critical Control Point (HACCP) plans are being followed and are working appropriately to reduce *Vibrio* bacterial growth caused by time and temperature abuse.

Regularly review procedures for monitoring, corrective action, verification, and recordkeeping systems (FDA, 2011);
- Use TTIs or temperature sensors within your facility and in shipments one-up and one-down in your supply chain to verify that procedures and practices are working properly and are in compliance with food safety guidelines. For more guidance on the use of TTIs and temperature sensors, please see our Extension Factsheet (Lane et al. 2019);
- Perform practice recalls to verify that there is one-up and one-down traceability in your supply chain.

4.6.3. Policy recommendations for government and industry

- Develop guidance for the shellfish industry regarding best practices for domestic and international air freight shipments;
- Develop tools to assist shippers in making packaging decisions. One option is an online calculator where shippers could manipulate input variables (e.g., package type, insulation R-value, starting temperature of oysters, starting temperature of frozen gel packs, estimated time of travel, ambient air temperature, etc.) to determine what combinations of variables would meet oyster temperature criteria during shipping;
- Establish a working group within the Interstate Shellfish Sanitation Conference to address issues related to cold chains and microbial growth;
- Validate the Food and Drug Administration *Vibrio* risk calculator for Pacific oysters (*C. gigas*).

Acknowledgements

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Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.ijfoodmicro.2019.108378.

References

American Airlines, 2018. Perishable Cargo.
Baker-Austin, C., Trinanes, J., Gonzalez-Escalona, N., Martinez-Urtaza, J., 2017. Non-cholera vibrios: the microbial barometer of climate change. Trends Microbiol. 25, 76–84.
Biji, K.B., Ravishankar, C.N., Mohan, C.O., Gopal, T.S., 2015. Smart packaging systems for food applications: a review. J. Food Sci. Technol. 52, 6125–6135.
Bross, M.H., Soch, K., Morales, R., Mitchell, R.B., 2007. *Vibrio vulnificus* infection: diagnosis and treatment. Am. Fam. Physician 76, 539–544.
Chiltern District Council, Safely Storing Oysters, Clams and Mussels, Fact Sheet and Hygiene Rating Improver.
Cook, D.W., 1994. Effect of time and temperature on multiplication of *Vibrio vulnificus* in postharvest Gulf Coast shellstock oysters. Appl. Environ. Microbiol. 60, 3483–3484.
Deeb, R., Tufford, D., Scott, G.I., Moore, J.G., Dow, K., 2018. Impact of climate change on *Vibrio vulnificus* abundance and exposure risk. Estuar. Coasts 1–15.
Delta, 2018. Fresh– Shipping Perishable, Time-Sensitive Products. https://www.deltacargo.com/Cargo/catalog/products/specialized-fresh.
FDA, 2005. Quantitative Risk Assessment on the Public Health Impact of Pathogenic *Vibrio parahaemolyticus* in Raw Oysters. US Department of health and Human Services.
FDA, 2011. Fish and fishery products hazards and controls guidance. In: Chapter 12: Pathogenic Bacteria Growth and Toxin Formation (Other than *Clostridium botulinum*) as a Result of Time and Temperature Abuse, Fourth edition. .
Fernandez-Piquer, J., Bowman, J.P., Ross, T., Estrada-Flores, S., Tamplin, M.L., 2013. Preliminary stochastic model for managing *Vibrio parahaemolyticus* and total viable bacterial counts in a Pacific oyster (*Crassostrea gigas*) supply chain. J. Food Prot. 76, 1168–1178.
Gokoglu, N., Yerlikaya, P., 2015. Seafood Chilling, Refrigeration and Freezing: Science and Technology. John Wiley & Sons.
Gooch, J., DePaola, A., Bowers, J., Marshall, D., 2002. Growth and survival of *Vibrio parahaemolyticus* in postharvest American oysters. J. Food Prot. 65, 970–974.
ISSC, 2018. Oysters. Interstate Shellfish Sanitation Conference. State Vibrio Plans.
Jones, J., Lydon, K., Kinsey, T., Friedman, B., Curtis, M., Schuster, R., Bowers, J., 2017. Effects of ambient exposure, refrigeration, and icing on *Vibrio vulnificus* and *Vibrio parahaemolyticus* abundances in oysters. Int. J. Food Microbiol. 253, 54–58.
Kolbe, E., Kramer, D., 2007. Planning for seafood freezing. In: Alaska Sea Grant College Program. University of Alaska Fairbanks, Fairbanks, Alaska.
Lane, B., Love, D.C., Kuehl, L., Hudson, B., 2019. Application of Time-Temperature Indicators and Time Temperature Data Loggers in the Seafood Industry. Virginia Cooperative Extension publication FST345NP. Available at www.pubs.ext.vt.edu/FST/FST-345/FST-345.html.
Love, D.C., Lane, R.M., Davis, B.J., Clancy, K., Fry, J.P., Harding, J., Hudson, B., 2019a. Performance of cold chains for Chesapeake bay farmed oysters and modeled growth of V*ibrio parahaemolyticus*. J. Food Prot. 82, 168–178.
Love, D.C., Lane, R.M., Kuehl, L.M., Hudson, B., Harding, J., Clancy, K., Fry, J.P., 2019b. Performance and Conduct of Supply Chains for United States Farmed Oysters. Aquaculture 515, 734569.
Lydon, K.A., Farrell-Evans, M., Jones, J.L., 2015. Evaluation of ice slurries as a control for postharvest growth of *Vibrio spp*. in oysters and potential for filth contamination. J. Food Prot. 78, 1375–1379.
Madigan, T.L., 2008. A Critical Evaluation of Supply-Chain Temperature Profiles to Optimise Food Safety and Quality of Australian Oysters. 2007/700. Australian Seafood Cooperative Research Center, pp. 1–84.
Marine Exension and Georgia Sea Grant, 2017. Safely Storing Oysters and Other Molluscan Shellfish. University of Georgia.
New Jersey Department of Environmental Protection, 2015. Techniques and practices for Vibrio reduction – Use of shading and rapid cooling (ice slurry) to control Vibrio growth. In: Interstate Shellfish Sanitation Conference.
Newton, A., Kendall, M., Vugia, D.J., Henao, O.L., Mahon, B.E., 2012. Increasing rates of vibriosis in the United States, 1996–2010: review of surveillance data from 2 systems. Clin. Infect. Dis. 54, S391–S395.
NOAA, 2017. Fisheries of the United States, 2016. National Marine Fisheries Service Office of Science and Technology.
NSSP, 2015. Guide for the control of Molluscan shellfish 2015 revision. In: Interstate Shellfish Sanitation Conference.
Parveen, S., DaSilva, L., DePaola, A., Bowers, J., White, C., Munasinghe, K.A., Brohawn, K., Mudoh, M., Tamplin, M., 2013. Development and validation of a predictive model for the growth of *Vibrio parahaemolyticus* in post-harvest shellstock oysters. Int. J. Food Microbiol. 161, 1–6.
United, 2018. Seafood. https://www.unitedcargo.com/shipping/productsAndShipping.jsp?name=ShippingSeafood&type=handling.
USDA, 2014. Census of Aquaculture 2013.
Yoon, K.S., Min, K.J., Jung, Y.J., Kwon, K.Y., Lee, J.K., Oh, S.W., 2008. A model of the effect of temperature on the growth of pathogenic and nonpathogenic *Vibrio parahaemolyticus* isolated from oysters in Korea. Food Microbiol. 25, 635–641.