MODELING TEMPERATURE AND TIME-BASED RISK OF *Vibrio parahaemolyticus* IN *Crassostrea virginica*: APPLICATIONS FOR DELAWARE'S OYSTER AQUACULTURE INDUSTRY

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Abstract

In Delaware, the Department of Natural Resources and Environmental Control (DNREC) enforces a *Vibrio parahaemolyticus* (Vp) Control Plan from June through September to reduce consumer risk of vibriosis associated with raw oyster consumption.[1] The plan emphasizes post-harvest temperature controls, including icing, shading, and depuration, to limit oyster bacterial growth. Vp proliferates rapidly at temperatures above 10°C, with reported growth rates of 0.022 log MPN/h at 15°C and 0.093 log MPN/h at 30°C.[2] Although oysters containing fewer than 10,000 CFU/g are legally marketable in the United States and Canada, illness has been documented at concentrations as low as 100–1,000 CFU/g.[3]

To mitigate this risk, DNREC requires resubmergence of oysters following air exposure or contamination with bird feces, allowing natural depuration through filtration of clean water.[1] While primarily designed to remove sewage-associated bacteria, depuration has demonstrated variable success against naturally occurring *Vibrio*, suggesting potential for refinement as a Vp control intervention.[4] This study evaluates the effectiveness of current Vibrio control measures like depuration, and seeks to identify the minimal post-harvest intervention duration needed to achieve meaningful reductions in Vp levels.

Introduction

- Eastern oysters (*Crassostrea virginica*) are ecologically vital and economically important, with Americans consuming over 60 million pounds annually. [5–7]
- As filter feeders, oysters can accumulate pathogenic bacteria such as *Vibrio parahaemolyticus*, a leading cause of seafood-associated illness in the U.S. (~80,000 cases/year).[8]
- Although post-harvest time and temperature guidelines exist, questions remain about how Vp responds to real-world harvest conditions (e.g., sun emersion, short depuration period).
- Understanding *Vibrio* growth dynamics under practical harvest conditions will inform improved post-harvest management strategies, enhance seafood safety, and support long-term industry sustainability.

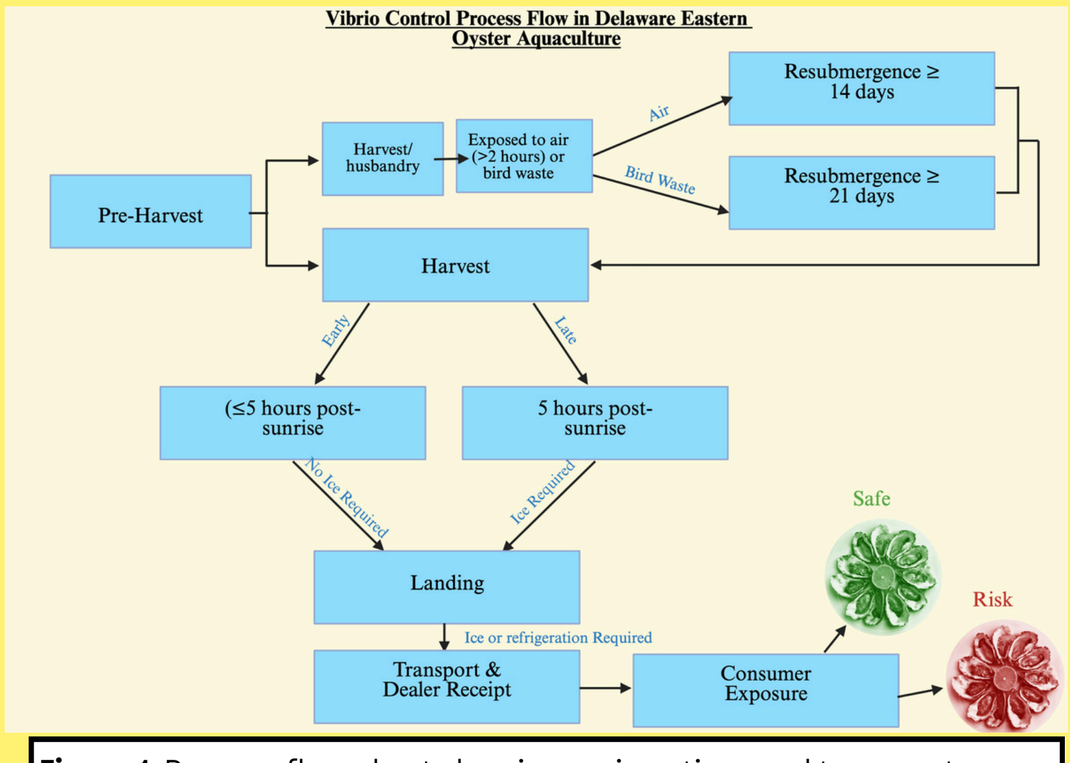


Figure 1. Process flow chart showing various time and temperature control measures implemented in DNREC's aquaculture oyster Vp prevention plan from 2024.

Materials and Methods

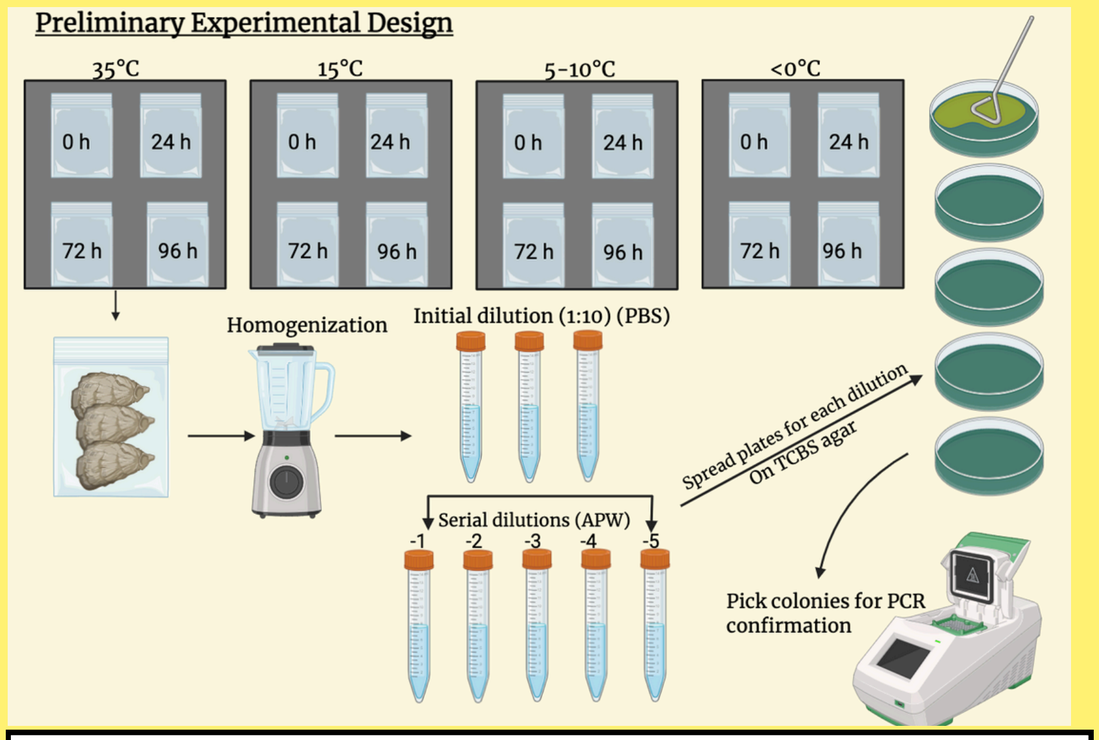


Figure 2. Design of preliminary experiment with oysters in October of 2025, observing Vp growth in oysters held at <0°C, 5-10°C, 15°C, and 35°C for 0, 24, 72, and 96 hours.

Preliminary Results and Discussion

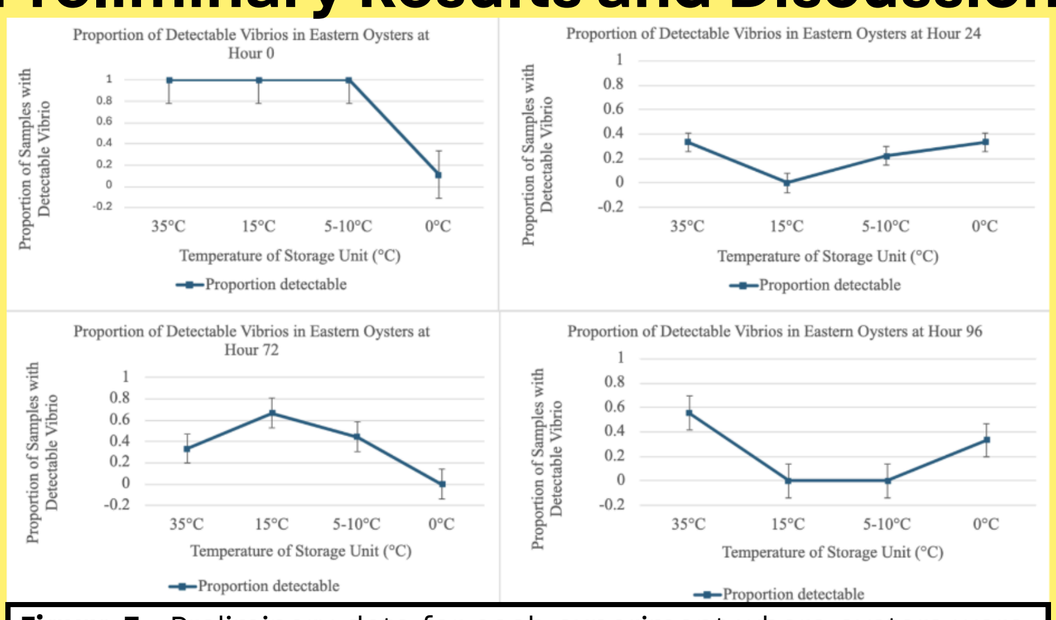
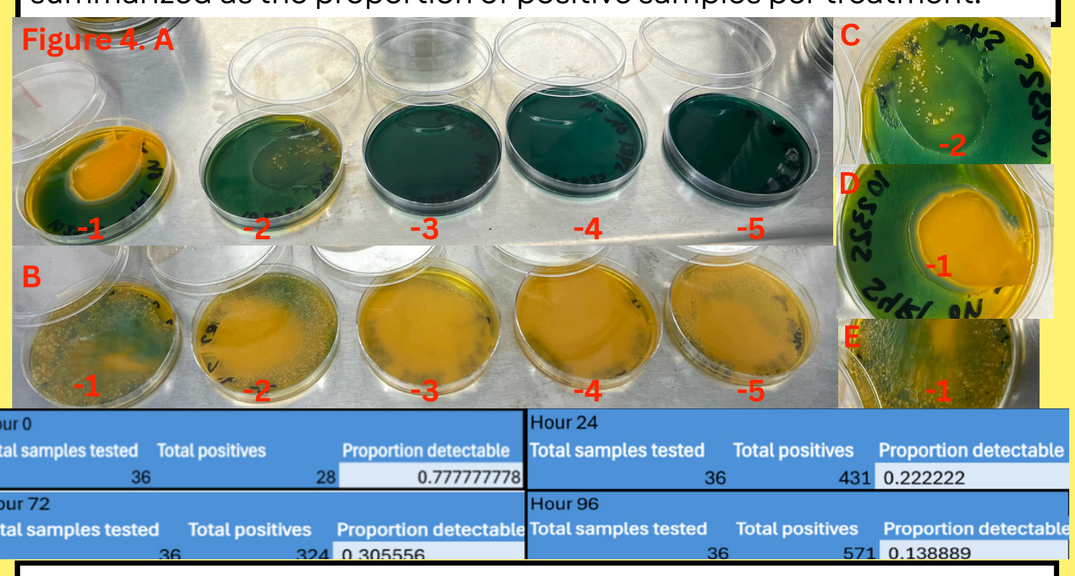


Figure 3. Preliminary data for each experiment where oysters were held at 0°C, 5-10°C, 15°C and 35°C for 0, 24, 72 and 96 hours. Samples were analyzed using a presence-absence framework, where samples with non-zero CFU were classified as "detectable" and summarized as the proportion of positive samples per treatment.



Hour 0			Hour 24		
Total samples tested	Total positives	Proportion detectable	Total samples tested	Total positives	Proportion detectable
36	28	0.777777778	36	431	0.222222
Hour 72			**Hour 96**		
Total samples tested	Total positives	Proportion detectable	Total samples tested	Total positives	Proportion detectable
36	324	0.305556	36	571	0.138889

Figure 4. Thiosulfate Citrate Bile Salts Sucrose (TCBS) agar plates of "Hour 0" samples from <0°C storage group (**A,C**, **D**) and "24 hour" sample from 35°C group (**B**, **E**).Vp and *Vibrio vulnificus* appear as blue-green colonies, while sucrose –fermenting strains like *Vibrio cholerae* appear as yellow colonies.

Consclusion

Temperature-Dependent

- Detection of Vp increased with elevated storage temperatures (15–35 °C)
- High temperatures led to persistent detection across multiple timepoints
- Refrigeration and freezing (<10 °C) resulted in rapid suppression of detectable *Vibrio*

Time-Dependent

- Overall detection decreased over time
- Prolonged high-temperature exposure sustained *Vibrio* presence in a subset of oysters
- Early post-harvest conditions strongly influenced detection outcomes
- Time alone is not a reliable predictor of *Vibrio* risk

Biological Variability

- Substantial individual oyster variability observed
- Detection patterns were non-linear and heterogeneous
- A subset of oysters maintained detectable levels despite overall trends

Post-Harvest Control Implications

- Reinforces the importance of immediate temperature control
- Supports interventions such as:
  - Rapid icing
  - Refrigeration
  - Depuration
  - Strict time–temperature management

Future Plans

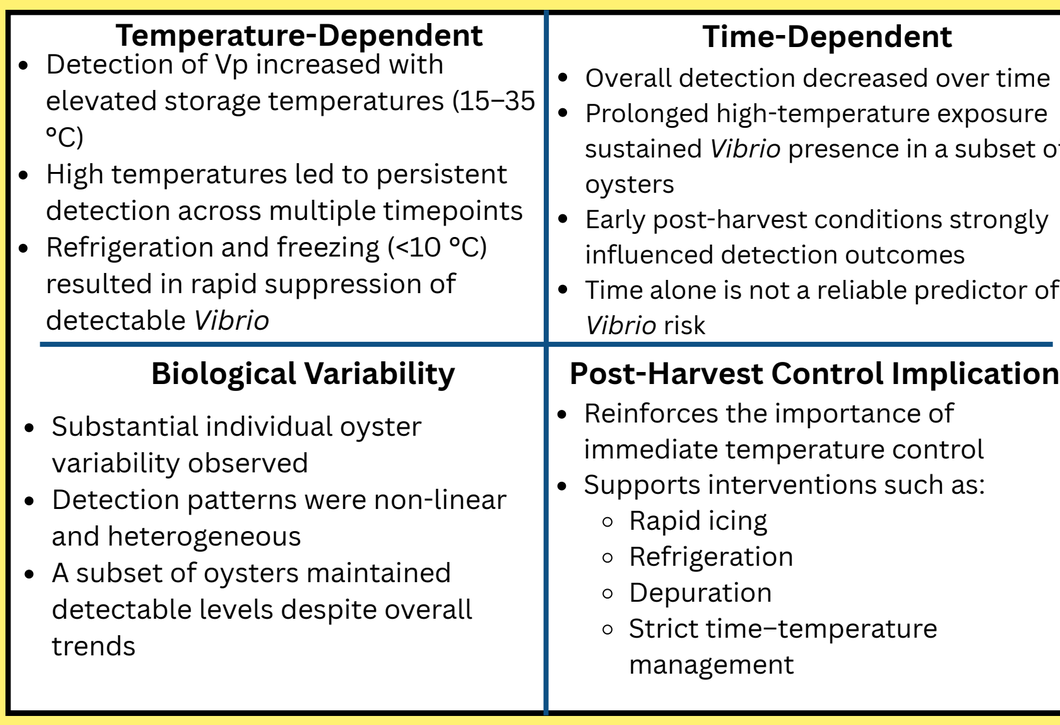


Figure 5. Experimental design for time-temperature modeling of *Vibrio* growth in oysters. Oysters will be subjected to icing, air exposure, or sunlight emersion (0–4 h), followed by 7 and 14-day depuration. Samples will be homogenized (1:10 Phosphate Buffered Saline), serially diluted in Alkaline Peptone Water, plated on TCBS agar plates, and confirmed by PCR. Experiments are to be conducted seasonally.

References

Acknowlegements

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