HACCP Controls to Prevent the Growth of *Vibrio parahaemolyticus* to Unacceptable Levels in Live Oysters Destined for Raw Consumption

Registered establishments are responsible for the development of effective controls for significant hazards through the use of a hazard analysis and implementation of a HACCP plan.

This document is intended to assist with the development of a HACCP Plan with respect to:

- The identification of *Vibrio parahaemolyticus* (Vp) as a significant hazard in live oysters intended for raw consumption and
- The development of effective controls to prevent unacceptable levels of Vp in the final product.

Controls for Vp other than the examples outlined in this document may be appropriate to specific operations. Registered establishments may use other control measures in their HACCP Plan provided data is available to validate the effectiveness of the control measure and critical limits.

This document was developed using information on harvest conditions and practices followed in British Columbia, which is the region where *Vibrio parahaemolyticus* (Vp) is known to be a significant hazard in oysters. Should Vp be identified as a significant hazard in other regions, controls should be tailored to suit the environmental conditions and harvest practices specific to those regions.

A full understanding of potential hazards is necessary to conduct a hazard analysis. Appendix 1 contains details on the biology of Vp and the illness that it causes. These details will assist in understanding this specific hazard.

A hazard, according to the CODEX definition, is both the specific agent and/or a condition with the potential to cause harm. The agent in this case is Vp. The conditions that enhance Vp growth and therefore increase potential for harm are:

- A rise in Vp levels in the growing waters during warmer weather and
- Exposure time of the shellstock to elevated temperatures

A product description is necessary for the hazard analysis. This document is specific to oyster shellstock for the raw consumption market.

A process flow diagram, which includes all the steps involved in producing the oyster shellstock, is necessary. If steps such as harvest, transport, and wet storage are under the direct control of the processor, they need to be identified in the process flow diagram. If these steps are not directly controlled by the processor, the use and verification of a Supplier Quality Assurance (SQA) agreement is an integral part of the Quality Management Program Plan.

A hazard analysis must be performed for each step identified in the process flow diagram. The hazard analysis must identify the potential hazard (Vp presence at unacceptable levels and/or growth of Vp to unacceptable levels). The potential significance of the hazard (Vp) and conditions that enhance its presence must be determined in the Hazard Analysis. Justification for inclusion or exclusion of Vp as a significant hazard as well as the associated preventative measures must be identified at each step of the hazard analysis procedure.

Where Vp is determined to be a significant hazard, there must be one or more control measures to prevent, reduce to an acceptable level, or eliminate the hazard. Each control measure that is designed to prevent, reduce to an acceptable level, or eliminate the occurrence of Vp is considered, by definition, to be a Critical Control Point (CCP). In this case the control measure will form part of a CCP and a HACCP Plan Worksheet describing the CCP needs to be developed.

Examples of processing steps are listed in Appendix 2, along with the resulting CCP determination of significance of Vp and examples of control measures that could prevent a Vp hazard.

Specific control measure examples are detailed in subsequent Appendices. Those control measures that are essential for the safety of the product are identified as requiring a CCP and the development of a HACCP Plan.
Appendix 1
Understanding *Vibrio parahaemolyticus*

**Illness**

Vp may cause gastroenteritis in humans and is associated with the consumption of raw, improperly cooked, or cooked and re-contaminated fish and shellfish.\(^6\) The incubation period ranges from 4 to 96 hours\(^6\) and the onset of disease is very sudden with explosive diarrhoea. Other symptoms include nausea, vomiting, headache, fever and chills. Symptoms typically subside within 48 to 72 hours but may last up to a week and treatment of most cases primarily includes rehydration.\(^1\)

**Biology and Ecology of Vp**

Being a mesophilic, salt tolerant bacteria, Vp will grow well in seafood stored at ambient temperature.\(^1\) Growth at the optimum temperature, 37°C, can be very rapid and doubling times of 9-10 minutes have been reported.\(^2\)

Rapid and efficient cooling (time and temperature control) is one of the most important control parameters in preventing Vp gastroenteritis. Cooling to 5°C will prevent growth.\(^1\)

Vp occurs in estuaries throughout the world and is easily isolated from coastal waters as well as from sediment, suspended particles, plankton, but not in the open sea. Vp has an annual cycle: the bacteria survive in the sediment during the winter and are released into the water associated with the zooplankton when the water temperature rises.\(^7\)

Vp is commonly isolated from bivalve molluscs. Levels fluctuate with temperature, with the higher numbers being isolated in the warmer months.\(^1\) Vp is typically undetectable in seawater when water temperatures are less than 10°C, although it can be cultured from sediments year-round at temperatures as low as 1°C.\(^12\)

Water temperatures on intertidal leases during the summer months may remain in the range favourable to Vp growth. Oysters from deepwater leases, which remain totally submerged, have also demonstrated Vp levels that greatly exceed 100 MPN/g, likely due to Vp growth in the marine environment.

The study by Nordstrom et al.\(^5\) demonstrates that Vp levels in live oysters are at a peak at the end of the low tide cycle after the oysters are out of the water and are exposed to warm air and the sun. The study also demonstrates that Vp levels in the oysters seem to return to the Vp level before they were exposed to the low tide cycle. Similar results were found by Buenaventura, Schallié, et al.\(^9\) and Herwig and Cheney\(^10\).

The USFDA Risk Assessment\(^8\) document provides references to support that depuration, using UV treatment, has little effect on Vp levels in naturally infected oysters or clams. The European Commission’s Scientific Opinion\(^7\) document comes to a similar conclusion.

While these last two paragraphs seem contradictory, it is possible that in the Nordstrom study, the new growth which results from the rise in Vp level after exposure during the low tide cycle, is more readily purged so that Vp levels return to those levels more indicative of the background level of Vp in the growing waters. It is possible that the USFDA references are indicating that UV treated systems will not readily remove background Vp levels that are already entrenched in the oyster.

In any case, validation data is necessary to support that any system (depuration, relaying, and wet storage) is capable of consistently reducing Vp levels to an acceptable level.

**Microbiological Standard**

Health Canada has set the microbiological standard for Vp in oyster shellstock intended for raw consumption at the retail level as follows: In a sample size of 5 units, no sample unit may exceed 10,000 MPN/g and no more than one sample unit may exceed 100 MPN/g.\(^11\) Oyster shellstock for raw consumption which exceeds these limits may result in action in accordance with a Health Risk 2 risk.
Appendix 2
Hazard Analysis and Control Measures at each Processing Step

Harvesting & Transportation

During the warmer months of the year (usually May to September/October), Vp can be present in growing waters at elevated levels and thus contamination of shellstock with Vp at harvest is considered a significant hazard. During this time there is a greater potential for exposure to elevated temperatures at harvest and during transport, which can accelerate the rate of Vp growth. The potential growth of Vp during each non-refrigerated step, from harvest to refrigerated holding, is also considered a significant hazard.

Preventative measures include:
- Monitoring of Vp levels for each harvest site per the current Communiqué “2013 B.C. Oyster Vibrio parahaemolyticus (Vp) Control Requirements”. See Appendix 3.
- Standard procedures to ensure site specific samples are representative. See Appendix 4.
- Standardized handling procedures for harvesting the oysters, particularly from intertidal harvest sites, to ensure the length of time they are exposed to elevated temperatures will not promote the growth of the pathogen during harvest or transport. See Appendix 5.
- The use and verification of a Supplier Quality Assurance Agreement for activities not under the direct control of the processor. Please see “Criteria for an Acceptable Supplier Quality Assurance Agreement” at http://www.inspection.gc.ca/english/fssa/fispoi/qual/sqaqfe.shtml

Holding Oyster Shellstock on Wet Storage Sites

Harvest lots are sometimes wet stored at secondary harvest sites prior to receiving into a registered establishment. The Vp levels, both from the initial harvest site and all wet storage sites, and the potential growth at any site is considered to be a significant hazard when harvesting is done in the warmer months, which are usually from May to September/October.

If wet storage is used to reduce Vp levels, then Vp is a significant hazard that requires the control of a CCP, including validation data that demonstrates the consistency and effectiveness of the system, as per Appendix 6.

Preventative measures include:
- Monitoring of Vp levels for each harvest lot (at initial harvest site and at secondary storage sites) per the current Communiqué “2013 B.C. Oyster Vibrio parahaemolyticus (Vp) Control Requirements.” See Appendix 3.
- Standard procedures to ensure site specific samples are representative. See Appendix 4.
- Standardized handling procedures for harvesting the oysters to ensure the length of time they are exposed to elevated temperatures will not promote the growth of the pathogen. See Appendix 5.
- Monitoring and control of the time and temperature exposure of the harvested oysters until they are refrigerated. See Appendix 5.
- Any controls undertaken by a third party harvester would form part of a Supplier Quality Assurance agreement.

Receiving at the Registered Establishment

Vp at the receiving step is considered a significant hazard, regardless of whether the harvest and/or transportation steps were under the direct control of the registered establishment.

Preventative measures include those listed above for harvesting, transportation, and wet storage, as appropriate to the harvest lot received at the registered establishment. The time and temperature of the shellstock at receipt are recorded as per Appendix 5.

Processing Steps: Cleaning, Sorting, Sizing, Packaging, and Labelling

When refrigeration of the oyster shellstock that rapidly reduces shellstock temperature to 10 °C or less occurs prior to these processing steps and the product remains at 10 °C or less until the completion of all of these steps, the potential for Vp growth will not be considered a significant hazard.
When oyster shellstock is received and processed prior to refrigeration, the potential growth of this pathogen during each non-refrigerated step is considered a significant hazard.

The preventative measures include:

- Monitoring and control of the time and temperature exposure of the harvested oysters until they are refrigerated as per Appendix 5.
- Recording the time and temperature at the beginning of each step as per Appendix 5.

**In Plant Wet Storage**

If a wet storage system is used to reduce Vp levels, then Vp is a significant hazard that requires the control of a CCP, including validation data that demonstrates the consistency and effectiveness of the system, as per Appendix 6.

If a wet storage system is used only to hold shellstock that meets the harvest criteria at receiving, then Vp growth is a significant hazard at this step when water temperatures exceed 10°C.

Preventative measures include:

- Monitoring and control of the time and temperature exposure of the harvested oysters until they are refrigerated and oyster shellstock temperatures are less than 10°C, as per Appendix 5.

**Refrigerated Storage**

Cooling equipment must be effective in rapidly reducing the temperature of bulk stored warm product to 10°C or less. With small coolers that have a very limited capacity to absorb a large thermal load, the time the product remains at temperatures greater than 10°C must be considered in the overall time and temperature exposure of the shellstock.
Appendix 3
Monitoring & Control of Vp Levels for Each Harvest Lot

As per the current season’s Communiqué “2013 B.C. Oyster *Vibrio parahaemolyticus* (Vp) Control Requirements,” unless a post-harvest processing step validated to reduce Vp levels to equal or less than 100 MPN/g is applied, oysters intended for sale in the shell may only be harvested from sites where Vp does not exceed 100 MPN/g.

Monitoring of Vp levels for harvest sites and wet storage sites constitutes a CCP and the details outlined on the HACCP Plan Worksheet must be included in the registered establishment’s QMP.

CFIA has six different environmental Vp indicator sites. When Vp results exceed 100 MPN/g, oysters from the affected area must not be harvested for the oyster shellstock market unless registered establishments can provide ongoing Vp sample monitoring data demonstrating that the Vp levels do not exceed 100 MPN/g for the specific harvest sites.

Sampling procedures need to be included in the registered establishment’s QMP, as per Appendix 4.

Appendix 4
Standard Operating Procedures for Site Specific Vp Monitoring Samples

Vp sample monitoring results need to be representative of the location and method of harvest and sampled in a consistent manner. Examples on how to achieve this include:

- Samples from deep water harvest sites are taken from a depth that represents the product to be harvested
- Samples from intertidal or beach harvest sites are taken from the water on a high tide, a receding tide or after the incoming tide has covered the oysters for a period of time per Appendix 5
- If both deep water and beach/intertidal shellstock are harvested from a site, the sample must represent the “worst case scenario,” or both types of shellstock harvested must be sampled.
- The status of the tide at harvest forms part of the record for the sample result
- 10 to 12 oysters are required per sample
- During shipping the temperature of samples is between 4°C and 10°C. To maintain cool temperatures, the use of gel packs is encouraged but care is required to avoid direct contact between the oysters and gel packs.
- Shellstock samples are sent to the laboratory immediately; within 24 hours of sampling is desirable.
Appendix 5
Oyster Shellstock Temperature

The following practices will assist in reducing the length of time that oyster shellstock is exposed to elevated temperatures to reduce the likelihood of Vp growth.

1. For intertidal or beach harvest, oysters are harvested at high tide, on a falling tide or after the incoming tide has covered the oysters for a period of time, as specified in the registered establishment's standard operating procedures. The timing of the harvest to the tide cycle is chosen to minimize the internal temperature of the oysters at the time of harvest as suggested by the work of Nordstrom et al.\(^5\)

2. Harvested oysters are protected from the sun.

3. When water or oyster temperatures at a harvest site are equal to or greater than 15°C, harvested oysters are placed into a temperature controlled environment within 1 hour of removal from the water.

4. When water or oyster temperatures at a harvest site are less than 15°C, harvested oysters are placed into temperature control within 4 hours of removal from the water.

5. Oysters are rapidly cooled to 10°C or less as quickly as possible, and this temperature is maintained to minimize post harvest growth of Vp.

6. Processors need to be aware of the temperature of product leaving the processing plant, as refrigerated transport is not an effective means of reducing oyster temperature.

Temperature Control as a CCP

After harvest, the only effective means of minimizing the hazard of Vp growth is to keep the oyster shellstock cool. Temperature control at all steps from harvest through processing, until immediately prior to shipping, would constitute a CCP and records of these temperatures must be maintained. The above points 3, 4 and 5 specify critical limits that should be used. The type of information outlined on the HACCP Plan Worksheet must be detailed in the registered establishment's QMP.

Temperature data must be recorded to demonstrate that the temperature environment is controlled from the harvest site through all transportation steps to the registered establishment. Procedures to ensure that temperature is consistently measured and is representative of the harvest lot need to be included in the QMP.

The temperature record must follow the product and include the following information:
- Temperature measurements are taken along the harvest and transportation chain prior to delivery to the registered establishment, starting with the harvest site water temperature or the shellstock temperature at the time of harvest;
- The record includes the time, location, and person responsible for each temperature measurement;
- Temperatures must be representative of the entire harvest lot. Note: temperatures are most representative if taken from more than one oyster and at more than one location in the lot (the centre being the worst case scenario). The internal meat temperature, rather than the shell temperature, should be measured.

The afore-noted points must also be documented and verified if harvest and/or transport are covered by a Supplier Quality Assurance agreement.
Appendix 6
Wet Storage and Depuration

If a wet storage system is relied on to reduce Vp levels, then Vp is a significant hazard that requires the control of a CCP. Prior to use, validation must be undertaken to demonstrate the consistency and effectiveness of the wet storage/depuration system. After validation has demonstrated that the system can consistently decontaminate the oyster shellstock, monitoring and record keeping must occur to demonstrate that the process parameters, determined during validation, have been met.

Validation data demonstrating that the system is consistently able to reduce Vp to an acceptable level is expected to include the following:

- Vp levels from samples taken when the shellstock is placed in the tank (zero hour)
- Details of the harvest site, tide cycle, and temperature record as per Appendix 5
- Any other factors that might influence the Vp level in the time zero sample

- Consistency of the wet storage or depurating conditions, such as
  - the length of time the oysters are held;
  - the temperature of the holding system;
  - water flow or tide cycle as appropriate to the system;
  - density of oysters;
  - arrangement of the oysters in the tanks;
  - appropriate water quality variables such as salinity, ammonia & oxygen levels.

- Vp levels from samples taken at the completion of the wet storage/depuration period when levels are expected to be less than 100 MPN/g

Refer to the CSSP Manual Chapters 5 and 10 for more information on wet storage, depuration, and validation of decontamination processes.
References


4. CODEX. Recommended International Code of Practice - General Principles of Food Hygiene. CAC/RCP 1-1969, Rev. 4-2003


6. USFDA Center for Food Safety & Applied Nutrition. The Bad Bug Book. [http](http://www.fda.gov/Food/FoodSafety/FoodborneIllness/FoodbornePathogensNaturalToxins/BadBugBook/ucm070452.htm)


