REPORT OF THE *Vibrio vulnificus* POST-HARVEST TREATMENT VALIDATION/VERIFICATION WORKING GROUP

JULY 18, 2002

INTRODUCTION

This working group was conviened in part to respond to Issue 01-208 to provide guidance to states on appropriate procedures to validate Post-Harvest Treatment (PHT) processes for vibrios. Validation in the context of this issue is defined as the process of ensuring that a defined set of control measures is capable of achieving control over a specific hazard(s) in a specific food(s). Factors to consider in validation include: (1) Consistency of demonstrating repeated efficacy of specified control measures; (2) Determining that the process variabilities are within acceptable levels; (3) Determining the extent of the established science and process necessary for laboratory validation of control measures: (4) Adequacy of control measures requiring more than laboratory validation; (5) Resource constraints; and (6) Uncertainties associated with the validation of control measures.

Validation is typically conducted prior to the initiation of a new food safety system to assure that it is capable of achieving the desired food safety outcome. Validation is repeated only infrequently when changes made to the food safety system are significant enough to require revalidation. Alternatively, it could be required if there is a change in the level of the hazard(s) or there is the emergence of a previously unidentified hazard of concern.

Validation of food safety control measures is different from verification and routine monitoring. Further, validation is not a process of monitoring the on-going assurance that a critical control point is operating properly within specifications for the control of a hazard in a food product. Additionally, it is not the ongoing process of verifying whether a HACCP plan is operating correctly.

When developing the format for validating a PHT process for molluscan shellfish, the following was considered:

1. The number of samples that must be tested.
2. The initial (or zero hour) level of vibrios in the product to be subjected to the PHT.
3. The analytical method to be utilized.
4. The format of the MPN test (number of tubes in series, number of series, and amount of inoculum in tubes).
5. Determination of numerical value for an endpoint criteria.

6. Whether or not any nonconforming units will be allowed recognizing that a MPN is a population estimate, and therefore when a number of tests are run on a sample that has an actual number of cells of 2/g, some percentage of tests on that sample will indicate an MPN greater than three. For this reason, it is desirable when selecting a scheme to validate a PHT process, to permit a small number of samples to exceed the endpoint criteria numerical value.

PROPOSED VV WORKING GROUP INTERIM GUIDANCE FOR THE VALIDATION OF PHT PROCESSES

1. Data should be obtained from 30 samples (no compositing), comprised from 10 samples representing three different process runs, with each of the three runs being separated by a 24-hour period, e.g. three different days, as indicated in Attachment 1. This protocol is premised upon the logic used to validate low acid canned foods scheduled processes.

2. The initial vibrio load (zero hour) shall be determined by taking five subsamples and analyzing each subsample individually (no compositing of subs), recording the data, and subjecting the values to a mathematical technique to enumerate a "central tendency" value, e.g., arithmetic average, geometric average, etc. While it is recognized that compositing the five subs to determine the initial (zero hour) vibrio load would be advantageous in terms of practicality and utility – nevertheless – more evaluation on this issue needs investigating before it can be recommended. The initial load shall be 100,000/g as worse case scenario or if some lower initial levels are used the process will only be validated for those maximum initial levels. The zero hour level may be achieved through naturally occurring vibrio levels in shellfish and, where not practical, by time/temperature abuse. For Vibrio parahaemolyticus, the 03:K6 serotype shall be used for the initial load through an inoculation process.

3. Analytical methodology to determine vibrio levels should be the official methods previously endorsed by the ISSC.

4. A five tube, single dilution MPN with an inoculum of 0.1g per tube should be used.

5. The numerical value of the endpoint criteria, it was predetermined that this should represent the lowest sensitivity of the MPN method which is less than 3 per gram.

6. Because of the variability of the MPN test it is expected that there will be sample results with values above the designated endpoint.

7. Existing PHT process validations which have been concurred with by FDA, which may not have followed this recommended validation protocol should be grandfathered into acceptance.

NEXT STEPS

1. It is recognized, given known variations in MPN, that further study needs to be done to determine the best procedure to innumerate the initial virbio load, (zero hour) of raw material to be subjected to the PHT process. A small working group comprised of Drs. Blodgett, Cook, and Rainosek, will address this issue with a view toward determining the suitability of compositing the aforementioned five subsamples (Item 2, paragraph 2 of the proposed interim guidance). They will be asked to review the variability data to be supplied by some states, FDA, and possibly some industry members. Nevertheless, no proprietary data are to be examined. Further, the group recommends that when designing validation of PHT processes that consideration be given to assure, in the raw material to be subjected to the process, the diversity of factors known to impact the efficacy of the PHT process be reflected.

2. The Working Group will also begin to address and develop guidance for the verification procedures for PHT processes.

ATTACHMENT 1:

1. 30 = the number of samples to be tested (see item 2 in proposed ISSC interim guidance) with no compositing of the samples.

2. 5 = the number of tubes in the single dilution MPN series.

3. 0.1 = the amount (g) of sample inoculated into each tube.

4. 2 = the maximum number of positive tubes out of 5 for the SAMPLE to pass; if 3 or 4 are positive, the SAMPLE fails, however, if all 5 tubes are positive, the PROCESS is not validated.

PLAN

Maximum number of samples Out of 30 that can fail and the Process still be validated	Number of actual vibrios per gram in the sample	Probability (%) the process will be validated
3	2	95.2
	3	54.0
	4 **	10.6
	5	0.7

**Example interpretation: When each of the 30 samples have an actual number of vibrios per gram = 4, there is a 10.6% chance that the process will be validated.

Note: If the number of positive tubes is 2 out of 5, the MPN is 5.
If the number of positive tubes is 4 out of 5, the MPN is 16.
As indicated above, if all 5 tubes are positive, the PROCESS is not validated.