National Shellfish Sanitation Program

Guide for the Control of Molluscan Shellfish

2007

Section IV. Guidance Documents
Chapter II. Growing Areas

.14 Calculating the 90th Percentile for End-Product Depurated Shellfish

Guide Contents

Process verification in depuration is performed continuously to ensure that the microbial contaminant load is being effectively reduced. Two indices of performance, the geometric mean and the 90th percentile have been developed to describe the effectiveness of the depuration process. Critical limits for these parameters have been established empirically by shellfish species. For soft clams (*Mya arenaria*), a geometric mean of 50 and a 90th percentile of 130 have been set. For hard clams, oysters, manilla clams and mussels, a geometric mean of 20 and a 90th percentile of 70 have been adopted.

Geometric means and 90th percentiles are determined daily or as end-product results become available from the analysis of the most recent ten (10) consecutive harvest lots per species, per restricted harvest area used. If the critical limits for either the geometric mean and/or the 90th percentile are exceeded, the process is considered to be unverified; and, additional sampling requirements must be instituted to ensure effective process control.

End-product depurated shellfish samples are analyzed using two different methods of recovery, a pour plate procedure and a single dilution MPN test. Calculation of the 90th percentile for these samples is complicated by the fact that fecal coliforms recovered by the MPN and ETCP methods follow different statistical distributions. To accommodate these differences and maintain a high likelihood for detecting an unacceptable amount of process variability without having to change or alter the formula used requires the use of nonparametric or "distribution free statistics." Using "distribution free statistics," the 90th percentile for end-product depurated shellfish samples is calculated by arraying the fecal coliform count data in ascending order and applying the formula (n + 1)P/100.

As an example of the use of this formula, the *Model Ordinance* requires that the 90th percentile of the fecal coliform analytical data be calculated from the most recent ten (10) consecutive harvest lots for each shellfish species depurated from each restricted harvest area. Fecal coliform count data, whether from the ETCP or MPN procedure for these ten (10) lots must be arrayed from the smallest to the largest value using the arithmetic (not logarithmically transformed) count data. Applying the formula, n would equal 10 for the ten (10) most recent consecutive harvest lots required by the *Model Ordinance*. P, the percentile of interest would be 90. Multiplying the formula out gives the position of the 90th percentile in the arrayed data. Performing these calculations, 10 + 1 = 11, 11 x 90 = 990/100 = 9.9. Thus, the 90th percentile for end-product depurated shellfish data is the value of the 9.9th sample in the ten (10) sample array.

Using the ten (10) samples as required by the *Model Ordinance*, the 90th percentile for end-product depurated shellfish samples would always be the value of the 9.9th sample in the ascending array of the arithmetic count data. To calculate this value from the arrayed data, interpolation between samples 9 and 10 is necessary. This is best illustrated using several samples.

Example 1

For soft clams, the ten (10) most recent consecutive harvest lots from a particular restricted harvest area produced the following end-product fecal coliform count data which has been arrayed in ascending order for ease in calculation.

	Sample #	FC Count (MPN/100 grams)	
	1	**8.9 (<9.0)**	
	2	**9.0**	
	3	**9.0**	
	4	**9.0**	
	5	**9.0**	
	6	**18**	
	7	**18**	
	8	**18**	
	9	**29**	
	10	**248**	

a. By convention and for the purpose of these calculations, fecal coliform counts that signify the upper or lower limit of sensitivity of the test (MPN or ETCP) shall be increased or decreased by one significant figure. For example <9.0 becomes 8.9, <17 becomes 16 and >248 becomes 250. Individual plates which are too numerous to count (TNTC) are considered to have >100 colonies per plate. A sample containing "TNTC" plates is collectively rendered as having a count of 10,000.

b. The 90th percentile for a ten (10) sample array is the 9.9th sample in the array. The value for the 9.9th sample in the array is interpolated by subtracting the value for sample #9 from the value for sample #10 in the array. This value is subsequently multiplied by 0.9 and then added to the value of sample #9 to give the value for the 9.9th sample in the array or the 90th percentile.

c. In this example, sample #9 which is 29 is subtracted from sample #10 which is 248 to give 219. 219 is subsequently multiplied by 0.9 to give 197.1. 197.1 is then added to the value of sample #9, which is 29 to give 226.1. Rounding this off to 226, the value of the 90th percentile becomes 226.

Example 2

Soft clams from another restricted harvest area produced the following end-product depurated fecal coliform counts which have been arrayed in ascending order for ease in calculation.

	Sample #	**FC Count (MPN/100 grams)**	
	1	**16 (<17)**	
	2	**16 (<17)**	
	3	**16 (<17)**	
	4	**17**	
	5	**17**	
	6	**33**	
	7	**50**	
	8	**50**	
	9	**67**	

	10	**84**	

In this example as above, the 90th percentile equals the value of the 9.9th sample in this ten (10) sample array. The value for the 9.9th sample in the array is interpolated by subtracting the value of sample # 9 which is 67 from the value of sample #10 which is 84 to give 17. 17 is then multiplied by 0.9 to give 15.3 which is added to the value of sample #9 which is 67 to give 82.3. Rounding this value off to 82, the value for the 90th percentile becomes 82.

Example 3

In this case, oysters from a restricted harvest area produced the following end-product depurated fecal coliform counts which have been arrayed in ascending order for ease in calculation.

	Sample #	**FC Count (MPN/100 grams)**	
	1	**8.9 (<9.0)**	
	2	**8.9 (<9.0)**	
	3	**8.9 (<9.0)**	
	4	**8.9 (<9.0)**	
	5	**9.0**	
	6	**9.0**	
	7	**9.0**	
	8	**18**	

	9	**88**	
	10	**88**	

In this example as in the other two, the 90th percentile equals the value of the 9.9th sample in the ten (10) sample array. Unlike the other two examples, however, the values for samples # 9 and #10 are identical making interpolation unnecessary in finding the value for the 9.9th sample in this array. This value is by convention identical to the value for samples #9 and #10. In this case, the value is 88.

Conditional Protocol

In examples 1 and 3 above, the values of the 90th percentiles calculated exceeded the critical limits set for the individual shellfish species depurated. Such high levels of variability when detected in the performance of the depuration process subsequently trigger the conditional protocol. Implementation of the conditional protocol requires the institution of a number of additional control measures designed to ensure adequate depuration. One such control measure involves the analysis of at least one (1) zero hour shellfish sample from each harvest lot. Like end-product depurated shellfish samples, the Elevated Temperature Coliform Plate Method may also be used for these analyses. However, the 12-tube, single dilution MPN test must not be used because of its limited effective count range (from 9 to 248). Instead, the 5-tube, 3-decimal dilution MPN test must be used to accommodate the expanded range in fecal coliform counts which may be encountered.