

VALIDATION CRITERIA

Ruggedness is the ability of the qualitative method to withstand relatively minor changes in analytical technique, reagents or environmental factors likely to arise in different test environments.

Procedure: This procedure is applicable for use with either growing waters or shellfish tissue. For each shellfish type of interest use a minimum of 10 – 12 animals. For each sample take two (2) aliquots of either the growing water sample or shellfish homogenate appropriately sized for your work. Spike both aliquots of each sample with a suitable concentration of the target analyte/measurand/organism of interest. Process both aliquots of sample as usual to determine whether the test response is positive or negative. For the second aliquot from each sample, however, use a different batch or lot of culture media and/or test reagents as appropriate to process this aliquot. Do a minimum of 30 samples using a variety of concentrations to spike the samples which span the range of the method's intended application. Use the same two batches or lots of culture media and/or test reagents to process each sample such that "batch or lot 1" is used to process the first aliquot of each sample and batch or lot 2" is used to process the second aliquot from each sample. Both aliquots of the same sample must be spiked with the same concentration of target analyte/measurand/organism of interest. If spatial or geographic influences on method performance are suspected, follow the above procedure for all samples suspected of being so influenced. If possible, process samples over a period of several days using different analysts to process and read "batch or lot 1" and batch or lot 2" samples.

Data:

Sample type _____

Sample #	Concentration	Positive results		Negative results	
		Batch/lot 1	Batch/lot 2	Batch/lot 1	Batch/lot 2
1					
2					
3					
4					
5					
6					
7					
8					
9					
10					
"					
"					
*n					

*n is the last sample tested.

Repeat for each shellfish tissue type of interest and/or samples suspected of being affected by spatial or geographic influences.

DATA HANDLING**Ruggedness**

In the day to day operations of the laboratory there will be changes in the batches/lots of culture media and/or test reagents used to process samples. Analysts may change and environmental factors may vary over time. None of these factors, however, should adversely impact test results if the method as implemented is sufficiently rugged to be used routinely for regulatory monitoring.

To determine whether the method as implemented is sufficiently rugged to withstand the types of changes anticipated to occur in routine use, the numbers of positive and negative results produced from the use of batch or lot 1 and batch or lot 2 culture media and/or test reagents will be compared. If the numbers of positive and negative results produced by batch or lot 1 and batch or lot 2 culture media and/or test reagents are identical, the method as implemented is not affected by relatively minor changes in the day to day operations of the laboratory and is sufficiently rugged as implemented for routine use. If the numbers of positive and negative results generated by

batch or lot 1 and batch or lot 2 culture media and/or test reagents are not alike then a two-sided statistical test comparing the numbers (proportions) of positive and negative results produced by each batch or lot of culture media and/or test reagents is undertaken to determine significance at the .05 level. From the number of positive and negative results produced by batch or lot 1 and batch or lot 2 culture media and/or test reagents determine the "observed contrast pair". The "observed contrast pair" is the smallest value from either the positive or the negative results of either batch or lot 1 or batch or lot 2 and the value from the same category of results (positive or negative) produced by the other batch or lot of culture media and/or test reagents. The "observed contrast pair" once determined is compared to statistical table values for the minimum contrast (a pair of values) required to demonstrate significance at the .05 level from a standard 2x2 table for equal sample numbers.

Data Summary:

Number of samples tested _____

Number of positive results from batch or lot 1 _____

Number of negative results from batch or lot 1 _____

Number of positive results from batch or lot 2 _____

Number of negative results from batch or lot 2 _____

The "observed contrast pair" if applicable _____

Is there a significant difference between batch or lot 1 and batch or lot 2 Y/N