

**Maine Department of Marine Resources
Public Health Division
Growing Area Classification Program**

Transitioning to Membrane Filtration for Seawater and Pollution Source Samples

The Maine Department of Marine Resources has chosen to switch to a fecal coliform method that was approved for use in the National Shellfish Sanitation Program (NSSP) at the Interstate Shellfish Sanitation Conference in 2003. This method is the Membrane Filtration (MF) for Fecal Coliforms using mTEC agar with a two hour resuscitation step. The MF method reduces staff laboratory time and labor costs. Overall it will reduce the time that some staff spend in the lab, freeing them up for more field work and report writing. Additionally, it will allow the laboratory to analyze more samples.

A number of studies have confirmed that the MF procedure using mTEC agar is comparable to the approved Multiple Tube Fermentation (MPN) procedures. The NSSP relies on two standards to determine classification of harvest areas; a geometric average (geomean) of the 30 sample data set and for systematic random sampling strategy a 90th percentile. The 90th percentile is basically a variability component and encompasses the variability of the test method plus an allowance for some variability due to changing conditions in the water. The variability or precision of the test method employed impacts the 90th percentile standard. The more precise the test method, the less variability one would expect when conducting repeated measurements. The less variability translates into a lower 90th percentile standard. The MF procedure is a more precise procedure than most of the MPN dilution series approved for the use in the NSSP program. The 90th percentile standard for MF is 31 compared to the less precise 3 dilution/3 tubes per dilution MPN 90th percentile standard of 49.

The results of over 2200 samples from a variety of shellfish growing areas and classifications from Mississippi, Florida, Alabama and Maine were analyzed for comparison between MPN and MF using mTEC media. It was determined that the geometric average for both methods was comparable. Both MPN and MF provided a geomean of 14. The data from Maine indicated that the MF 90th percentile of 31 was equivalent to 90th percentile of 49 for the 3 dilution/3 tubes dilution MPN utilized by MEDMR.

The geometric mean and the 90th percentile are calculated on 30 data points extending over a five year period. During the transition from MPN to MF, we will be accumulating MF data points. The statistical calculations will be a combination of MPN and MF data points. The FDA has determined that the best way to handle the data is to perform the calculations as always for the data set, but to compare the data set to a hybrid weighted 90th percentile. This hybrid standard is calculated by weighting the relative contributions of each method to the database. This will mean that as the number of MPN data points reduce and the number of MF data points increase the 90th percentile standard that the sample site is compared to will change over time. A table is attached to demonstrate the 90th percentile standard for the gradual change in data between the two methods. Once all 30 data points are analyzed using MF, the 90th percentile for approved classification will be 31 and for restricted (for depuration) will be 163. The geomean approved standard of 14 fecal coliforms per 100 ml and geomean restricted standard of 88 fecal coliforms per 100 ml will remain the same for both methods.

Reports that display 90th percentiles will show the number of data points derived from MF analysis and will show the appropriate 90th percentile standard for that MPN/MF combination for approved and restricted classifications.

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It must be remembered that this weighted standard is only used for data sets encompassing data from the two different test methods, MF and MPN (3 tube/3 dilution). If decisions are to be made on a single test result analyzed by the MF method or a multiple number of test results all exclusively analyzed by the MF method, the 90th percentile standard is 31 fecal coliforms per 100 ml.

Questions regarding the transition from MPN to MF should be directed to Mercuria Cumbo, Water Quality Laboratory Manager at mercuria.cumbo@maine.gov or 207.667.5654.

P90 Approved Standard when transitioning from MPN (3 tube/3 dilution) to MF mTEC		
# Tests by A1	# Tests by MF	Approved Standard
29	1	48
28	2	48
27	3	47
26	4	46
25	5	45
24	6	45
23	7	44
22	8	43
21	9	43
20	10	42
19	11	41
18	12	41
17	13	40
16	14	40
15	15	39
14	16	38
13	17	38
12	18	37
11	19	37
10	20	36
9	21	36
8	22	35
7	23	34
6	24	34
5	25	33
4	26	33
3	27	32
2	28	32
1	29	31
0	30	31

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P90 Restricted Standard when transitioning from MPN (3 tube/3 dilution) to MF mTEC		
# Tests by A1	# Tests by MF	Restricted Standard
29	1	294
28	2	288
27	3	282
26	4	277
25	5	271
24	6	266
23	7	260
22	8	255
21	9	250
20	10	245
19	11	240
18	12	235
17	13	230
16	14	226
15	15	221
14	16	217
13	17	212
12	18	208
11	19	204
10	20	200
9	21	196
8	22	192
7	23	188
6	24	184
5	25	180
4	26	177
3	27	173
2	28	170
1	29	166
0	30	163