

1 **AOAC SMPR 2020.XXX; Draft Standard Method Performance Requirements (SMPRs®) for Quantitative**
2 **Microbiology Methods for Food and Environmental Samples; Version 4; March 11, 2020**
3

4 **Intended Use:**

5 Laboratory use by trained technicians.
6

7 **1. Purpose:**

8 AOAC SMPRs describe the minimum recommended performance characteristics and acceptance
9 criteria to be used during the evaluation of a method. The evaluation may be an on-site verification,
10 a single-laboratory validation, or a multi-site collaborative study. SMPRs are written and adopted by
11 AOAC as voluntary consensus standards and are used by AOAC in their evaluation of validation study
12 data for method being considered for *Performance Tested MethodsSM* or *AOAC Official Methods of*
13 *AnalysisSM* programs. This SMPR may also be used as acceptance criteria for verification at user
14 laboratories. (AOAC OMA, Appendix F, 2019)
15

16 **2. Applicability:**

17 Validation or verification of candidate methods used for the quantification or enumeration of
18 microorganisms in foods and environmental samples.
19

20 **3. Analytical Technique:**

21 Any analytical technique that can meet the performance requirements.
22

23 **4. Definitions:**

- 24 a. Candidate Method.—The method submitted for validation (Microbiology, 2019, 21st
25 Edition);
- 26 b. Reference Method.—Pre-existing recognized analytical method against which the candidate
27 method will be compared. (AOAC OMA, 2019)
- 28 c. Quantitative Method.—Method of analysis whose response is the amount (count or mass)
29 of the analyte measured either directly (e.g., enumeration in a mass or a volume), or
30 indirectly (e.g., color absorbance, impedance, etc.) in a specified test portion. (Microbiology,
31 2019, 21st Edition)
- 32 d. Enumeration.—The determination of viable microorganisms in a given test portion.
33 Enumeration can be performed directly or indirectly.
- 34 e. Equivalent.—The state or condition of being equal. The methods (candidate and reference)
35 would be considered equivalent within a specified confidence if the acceptance criteria are
36 satisfied.
- 37 f. Bias.—measurement bias; estimate of a systematic measurement error, or the systematic
38 difference between the quantitative assigned value and the average of measurement
39 replicate results (16140-1)
- 40 g. Confidence Interval.—The estimated range in which an obtained result should enclose the
41 actual concentration. For the purpose of this SMPR, a 90% confidence interval is used.
- 42 h. Most Probable Number (MPN) .—The maximum likelihood estimate of the contamination in
43 a given matrix using test portions from multiple levels
44

45 **5. Method Performance Requirements:**

46

| Parameters | Performance Requirement/Acceptance Criteria |
|---|--|
| Candidate Method to Reference Method Equivalence Acceptance Criteria | 90% confidence interval of the bias (difference between means) between two methods must fall within -0.5 to $0.5 \log_{10}$ for a given matrix at a given concentration |
| Number of Replicates* | Method developers may increase the number of replicates tested to improve the chance that the 90% confidence interval will all within the acceptable range (e.g. -0.5 to $0.5 \log_{10}$) |
| Evaluation of methods for different applications(e.g., pathogens vs. spoilage organisms) | For certain applications, a modification of the acceptance criteria may be appropriate. Any changes to the acceptance criteria should be reviewed and approved by the subject matter experts prior to submission of data. When narrowing the acceptance criteria, the range should be not be tighter than is -0.33 to $0.33 \log_{10}$. |

47 *A minimum of 5 replicates is required per contamination level when determining the bias and
 48 95% confidence intervals.
 49

50 **6. System Suitability and/or Analytical Quality Controls:**

- 51 a. Target and non-target organism controls shall be embedded in the validation or verification
- 52 study as appropriate.
- 53 b. Inhibition controls should be used for method verification for each new matrix as
- 54 appropriate.
- 55 c. Manufacturer must provide written justification if controls are not appropriate to an assay.

56
 57 **7. Validation Guidance:**

- 58 a. Validation studies should be conducted in accordance with procedures as outlined in
- 59 Appendix J, **Official Methods of Analysis of AOAC INTERNATIONAL: AOAC INTERNATIONAL**
- 60 *Methods Committee Guidelines for Validation of Microbiological Methods for Food and*
- 61 *Environmental Surfaces (2012)* or
- 62 b. ISO 16140-2:2016: Microbiology of the food chain—Method validation—Part 2: Protocol for
- 63 the validation of alternative (proprietary) methods against a reference method
- 64 Methods validated according to ISO 16140-2 or in a harmonized AOAC/ISO study, must be
- 65 statistically analyzed according to Appendix J and meet the performance requirements as
- 66 identified in this SMPR.
- 67 c. Collaborative studies and Inclusivity/Exclusivity (excluding total enumeration methods) are
- 68 required for candidate methods submitted for First Action *Official MethodsSM* review and
- 69 consideration.
- 70 d. Method Robustness, Stability, Product Consistency and Inclusivity/Exclusivity (excluding
- 71 total enumeration methods) Studies are required for *Performance Tested MethodsSM*
- 72 submission.

73

74 **8. Reference Methods**

- 75 a. The selection of the appropriate reference method will be determined based on the target
76 analyte and matrix being validated. Examples of acceptable reference methods but not
77 limited to are:
78 i. AOAC Official Methods of Analysis
79 ii. ISO standards
80 iii. US FDA Bacteriological Analytical Manual (BAM)
81 iv. USDA FSIS Microbiology Laboratory Guidebook (MLG)
82 v. Standard Method for the Examination of Dairy Products (SMEDP)
83 vi. Compendium of Microbiological Methods for the Examination of Foods (CMMEF)

84
85 **9. Maximum Time-to-Result:**

86 None mandated.

87
88 **10. References Cited:**

- 89 a. Appendix F, **Official Methods of Analysis of AOAC INTERNATIONAL: AOAC INTERNATIONAL**
90 *Guidelines for the Development of Standard Method Performance Requirements* (2016).
91
92 b. Appendix J, **Official Methods of Analysis of AOAC INTERNATIONAL: AOAC INTERNATIONAL**
93 *Methods Committee Guidelines for Validation of Microbiological Methods for Food and*
94 *Environmental Surfaces* (2012).
95
96 c. Appendix I, **Official Methods of Analysis of AOAC International: AOAC INTERNATIONAL**
97 *Methods Committee Guidelines for Validation of Biological Threat Agent Methods and/or*
98 *Procedures* (2012).
99
100 d. ISO 16140-2:2016 *Microbiology of the food chain—Method validation—Part 2: Protocol for*
101 *the validation of alternative (proprietary) methods against a reference method*
102
103 e. ISO 16140-1:2016 *Microbiology of the food chain—Method validation—Part 1: Vocabulary*