

DIN EN ISO 16140-2:2025-07 (E)

Microbiology of the food chain - Method validation - Part 2: Protocol for the validation of alternative (proprietary) methods against a reference method (ISO 16140-2:2016 + Amd 1:2024) (includes Amendment A1:2024)

Contents	Page
Foreword.....	v
A1 Foreword to Amendment 1 A1	vi
A1 Introduction A1	vii
1 Scope.....	1
2 Normative references.....	1
3 Terms and definitions.....	1
4 General principles for the validation of alternative methods.....	1
5 Qualitative methods — Technical protocol for validation.....	2
5.1 Method comparison study.....	2
5.1.1 General considerations.....	2
5.1.2 Paired or unpaired study.....	3
5.1.3 Sensitivity study.....	3
5.1.4 Relative level of detection study.....	9
5.1.5 Inclusivity and exclusivity study.....	12
5.2 Interlaboratory study.....	13
5.2.1 General considerations.....	13
5.2.2 Measurement protocol.....	14
5.2.3 A1 Summary of data and trueness calculations A1	15
5.2.4 A1 Interpretation of trueness data A1	18
6 Quantitative methods — Technical protocol for validation.....	20
6.1 Method comparison study.....	20
6.1.1 General considerations.....	20
6.1.2 Relative trueness study.....	20
6.1.3 Accuracy profile study.....	24
6.1.4 Limit of quantification study.....	28
6.1.5 Inclusivity and exclusivity study.....	29
6.2 Interlaboratory study.....	30
6.2.1 General considerations.....	30
6.2.2 Measurement protocol.....	31
6.2.3 Calculations, summary, and interpretation of data.....	32
Annex A (informative) Classification of sample types and suggested target combinations for validation studies	35
Annex B (normative) Order of preference for use of naturally and artificially contaminated samples in validation studies	51
Annex C (informative) General protocols for contamination by mixture and artificial contamination of foods	52
Annex D (informative) Models for RLOD calculations using data from the method comparison study	55
Annex E (normative) Points to be considered when selecting strains for testing inclusivity and exclusivity	57

Annex F (normative) Calculation of the relative level of detection (RLOD) using data from the interlaboratory study	59
Annex G (informative) Principle of the accuracy profile for validation of quantitative models	63
Annex H (informative) Application of the accuracy profile in the method comparison study	65
Annex I (informative) Example of the application of the accuracy profile for an interlaboratory study	68
Annex J (informative) Commercial sterility testing protocol for specific products	72
Bibliography	75