## ISO 16140-4:2020 (E)

Microbiology of the food chain — Method validation — Part 4: Protocol for method validation in a single laboratory

## **Contents**

	Fore	eword			
	Intro	oduction			
1	Sco	pe			
2	Nori	Normative references			
3	Tern	ns and definitions			
4	General principles of the single-laboratory detection or quantification method validation				
	4.1	General			
	4.2	Principles of the factorial approach			
	4.3	Principles of the conventional approach			
5	Tech	nnical protocol for validation — Factorial approach			
	5.1	Qualitative methods			
	5.1.1	Single-laboratory method validation study against a reference method			
	5.1.1.1	General considerations			
	5.1.1.2	Factorial, orthogonal method comparison study			
	5.1.1.2.1	Selection of samples			
	5.1.1.2.2	Selection of method factors			
	5.1.1.2.3	Experimental design			
	5.1.1.3	Calculation and interpretation for sensitivity			
	5.1.1.4	Calculation and interpretation of the RLOD			
	5.1.1.5	Inclusivity/exclusivity study			
	5.1.2	Single-laboratory method validation study without a reference method			
	5.1.2.1	General considerations			
	5.1.2.2	Factorial study			
	5.1.2.3	Calculation and interpretation for sensitivity			
	5.1.2.4 5.1.2.5	Calculation and interpretation of LOD50			
	5.1.2.5 5.2	Inclusivity/exclusivity study Quantitative methods			
	5.2 5.2.1	Single-laboratory method validation study against a reference method			
	5.2.1.1	General considerations			
	5.2.1.2	Selection of samples			
	5.2.1.3	Selection of method factors			
	5.2.1.4	Experimental design			
	5.2.1.5	Relative trueness			
	5.2.1.6	Accuracy profile			
	5.2.1.7	In-house precision (in-house repeatability and in-house reproducibility)			
	5.2.1.8	Inclusivity/exclusivity study			
	5.2.2	Single-laboratory method validation study without a reference method			
	5.2.2.1	General considerations			
	5.2.2.2	Selection of samples			
	5.2.2.3	Selection of method factors			
	5.2.2.4	Experimental design			
	5.2.2.5	Relative trueness			
	5.2.2.6	Accuracy profile			
	5.2.2.7 5.2.2.8	In-house precision (in-house repeatability and in-house reproducibility) Inclusivity/exclusivity study			
6		nnical protocol for validation — Conventional approach			

	6.1 6.1.1		Qualitative methods Single-laboratory method validation study against a reference method		
	6.1.1.1		General		
	6.1.1		Sensitivity study		
	6.1.1 6 1 1		RLOD study Inclusivity/exclusivity study		
	6.1.1.4 6.1.2 6.1.2.1		Single-laboratory method validation study without a reference method		
			General		
	6.1.2	2.2	Specificity		
	6.1.2.3 6.1.2.4 6.1.2.5 6.2		LOD50 study Calculation and interpretation for sensitivity Inclusivity/exclusivity study Quantitative methods Single-laboratory method validation study against a reference method		
	6.2.1 6.2.1.1		General		
	6.2.1.1		Relative trueness study		
	6.2.1.		Accuracy profile study		
	6.2.1.4		Limit of quantification study		
6.2.1		1.5	In-house precision study		
	6.2.1		Inclusivity/exclusivity study		
	6.2.2		Single-laboratory method validation study without a reference method		
	6.2.2		General		
	6.2.2 6.2.2		Relative trueness study Accuracy profile study		
	6.2.2		Limit of quantification study		
	6.2.2		In-house precision study		
	6.2.2	2.6	Inclusivity/exclusivity study		
7		Summ	nary of acceptability limits		
Annex	Α	(inforr	mative) List of factors and factor levels for factorial method validation		
Annex B		(informative) Calculation of in-house reproducibility for qualitative methods based on the LOD50 study described in 6.1.2.3			
Annex C		(informative) Example of a factorial single-laboratory method validation study for a quantitative method against a reference method			
	C.1		General		
	C.2		Study design		
	C.3		Calculations and summary of data		
C.3.		1	Summary of the results		
C.3			Relative trueness		
	C.3. C.3.		Accuracy profile		
	C.3.		Precision data Interpretation		
			·		
Annex	D		mative) Example of a factorial single-laboratory method validation study for a qualitative od against a reference method		
Annex E		(informative) Example of a factorial single-laboratory method validation study for the variability of the LOD50 for a qualitative method without a reference method			
Annex F		(informative) Determination of precision if the artificially contaminated samples are unstable			
	F.1		General		
	F.2		Adjustment of measurement values by using a linear trend		
	F.3		Adjustment of measurement values by using a reference method		
Annex G		(informative) Protocol for single-laboratory validation of alternative methods for microbiological confirmation and typing procedures			

7

Page count: 46