

ISO 22174:2024-08 (E)

Microbiology of the food chain - Polymerase chain reaction (PCR) for the detection and quantification of microorganisms - General requirements and definitions

Contents		Page
Foreword		v
1	Scope	1
2	Normative references	1
3	Terms and definitions	1
3.1	General terms	2
3.2	Terms related to the extraction and purification of DNA/RNA	3
3.3	Terms related to reverse transcription of RNA to DNA	4
3.4	Terms related to DNA amplification by PCR/RT-PCR	4
3.5	Terms related to controls	5
3.6	Terms related to qPCR	6
3.7	Terms related to dPCR	7
4	Principle	8
4.1	General	8
4.2	Laboratory sample	9
4.3	Sampling, transport and storage	9
4.4	Preparation of test sample	9
5	Microbial enrichment and virus concentration	9
5.1	Microbial enrichment	9
5.2	Virus concentration	9
6	Nucleic acid preparation	10
6.1	General	10
6.2	Prevention of amplification of DNA from dead cells	10
6.3	Nucleic acid extraction, release and purification	10
6.4	Nucleic acid quality and quantity	10
7	PCR amplification	11
8	Detection and confirmation of amplicons	11
9	General environmental laboratory requirements	12
9.1	General	12
9.2	Laboratory setup	12
9.2.1	General	12
9.2.2	Control of flows	13
9.2.3	Cleaning of laboratory	14
9.2.4	Environmental monitoring for nucleic acid contamination	14
10	Reagents and consumables	14
11	Equipment	14
12	Procedure	15
12.1	Enrichment and sample treatment	15
12.2	Amplification	16
12.2.1	General	16

12.2.2	Control reaction	16
12.2.3	Detection of amplicon	18
12.2.4	Data analysis	18
12.3	Evaluation	19
12.3.1	Qualitative evaluation	19
12.3.2	Quantitative evaluation	20
12.4	Test report	21
13	Performance characteristics of PCR-based methods	21
14	Validation and verification of PCR-based methods	21
14.1	General	21
14.2	Validation	21
14.3	Verification	22
	Annex A (informative) Fluorescence signals and amplification curve	23
	Bibliography	26