ISO 7218:2024-06 (E)

Microbiology of the food chain - General requirements and guidance for microbiological examinations

Contents

Page

Foreword ..... **vii**

Introduction ..... **viii**

1 Scope **1**

2 Normative references **1**

3 Terms and definitions **1**

4 Premises **5**

4.1 General 5

4.2 Biosafety considerations 5

4.3 Laboratory design 5

4.4 Laboratory areas 5

4.4.1 General 5

4.4.2 Areas associated with samples and testing 6

4.4.3 General areas 6

4.5 Layout and fittings of the premises 6

4.5.1 Objectives 6

4.5.2 Fittings 7

4.5.3 Other arrangements for laboratory premises 7

4.5.4 Cleaning and disinfection 8

5 Personnel **8**

5.1 General 8

5.2 Competence 8

5.3 Verification of ongoing staff competence 9

5.4 Hygiene 9

6 Equipment and consumables **9**

6.1 General 9

6.2 Sterilization and other heating equipment 10

6.2.1 General 10

6.2.2 Autoclave 10

6.2.3 Culture media preparator 11

6.2.4 Steamers, including boiling-water baths 12

6.2.5 Sterilizing oven 12

6.2.6 Microwave oven 13

6.2.7 Hotplate, induction cooker and heating mantle 14

6.2.8 Gas burner or wire incinerator 14

6.3 Temperature controlled equipment and monitoring devices 15

6.3.1 General 15

6.3.2 Incubator 15

6.3.3 Thermostatically controlled bath 16

6.3.4 Heating blocks 17

6.3.5 Refrigerators and cold-storage rooms 18

6.3.6 Freezer and deep freezer/ultra-low temperature freezer 19

6.3.7 Temperature-monitoring devices, including automatic recorders 19

6.3.8 Balances and gravimetric diluters 20

6.4 Defined volume inoculation equipment 21

6.4.1 Pipettes and pipettors 21

6.4.2 Dispensers 22

6.4.3 Spiral platers 23

6.4.4 Serial diluters ... 24
6.5 Protective cabinets ... 24
6.5.1 Description ... 24
6.5.2 Use ... 25
6.5.3 Cleaning and disinfection ... 25
6.5.4 Maintenance and inspection ... 26
6.6 Homogenizers, blenders, mixers and shakers ... 26
6.6.1 Homogenizers and blenders ... 26
6.6.2 Vortex mixers ... 27
6.7 Stills, deionizers and reverse-osmosis units ... 28
6.7.1 Description ... 28
6.7.2 Use ... 28
6.7.3 Maintenance ... 28
6.7.4 Verification ... 28
6.8 Separation and concentration equipment ... 28
6.8.1 Immunomagnetic separator (IMS) ... 28
6.8.2 Centrifuge ... 29
6.8.3 Filtration systems ... 29
6.9 Modified atmosphere equipment ... 29
6.9.1 Description ... 29
6.9.2 Use ... 29
6.9.3 Maintenance ... 30
6.9.4 Verification ... 30
6.10 Other equipment ... 30
6.10.1 pH meter ... 30
6.10.2 Colony-counting device ... 31
6.10.3 Timers and timing devices ... 31
6.10.4 Optical microscope ... 32
6.10.5 Glass washers, glassware and other laboratory ware ... 32
6.10.6 Disposable equipment and consumables ... 33
6.10.7 Other equipment and software ... 34

7 Sterilization/decontamination and disposal of laboratory materials ... 34
7.1 Sterilization ... 34
7.1.1 General ... 34
7.1.2 Sterilization by dry heat ... 34
7.1.3 Sterilization by moist heat (steam) ... 34
7.2 Decontamination and disinfection ... 34
7.2.1 Decontamination of glassware and materials before use ... 34
7.2.2 Decontamination of glassware and materials after use ... 34
7.3 Waste management ... 35
7.4 Washing ... 35

8 Preparation and use of culture media and reagents ... 35

9 Laboratory samples ... 36
9.1 Sampling techniques and sampling plans ... 36
9.1.1 General ... 36
9.1.2 Sampling ... 36
9.2 Sample transport ... 36
9.3 Sample receipt ... 37
9.4 Sample handling ... 37
9.4.1 General ... 37
9.4.2 Storage before examination ... 38
9.4.3 Test portions ... 38
9.4.4 Storage of laboratory samples after examination ... 38
9.5 Pre-testing of samples ... 38

10 Examination ... 39
10.1 Hygienic precautions during sample preparation and examination ... 39
10.1.1 General ... 39
10.1.2 Basic precautions ... 39
10.1.3 Sample handling ... 39
10.1.4 Sample handling tools and implements ... 40
10.1.5 Spillages ... 40
10.1.6 Process controls ... 40
10.1.7 Aerosols ... 40

10.1.8 Molecular methods 41
10.2 Preparation of initial suspension and dilutions 41
10.2.1 General 41
10.2.2 Concentration 41
11 Enumeration (quantitative) methods 41
11.1 General 41
11.2 Enumeration using a solid medium 42
11.2.1 General 42
11.2.2 Pour plate technique 42
11.2.3 Surface plating techniques 43
11.2.4 Enumeration of yeasts and moulds 44
11.2.5 Incubation 45
11.2.6 Calculation and expression of results obtained with solid culture media 45
11.2.7 Calculations for enumeration methods 47
11.3 Enumeration using liquid media 54
11.3.1 Principle 54
11.3.2 General MPN procedure 54
11.3.3 Limitations of MPN 54
11.3.4 Inoculation procedure 55
11.3.5 Choice of MPN configuration 55
11.3.6 Incubation 56
11.3.7 Interpretation and expression of results 56
11.3.8 Determination of MPN values using MPN calculators 56
11.3.9 Rarity categories 57
11.4 Estimates of uncertainty of test results 57
12 Detection (qualitative) methods 58
12.1 General 58
12.2 Principle 58
13 Confirmation and identification methods 58
13.1 General 58
13.2 Preparation of a pure culture 59
13.3 Confirmation methods 59
13.3.1 Latex agglutination test 59
13.3.2 Nucleic acid hybridization or molecular amplification methods 59
13.3.3 Slide agglutination tests 60
13.4 Identification methods 60
13.4.1 Biochemical galleries 60
13.4.2 DNA sequencing 60
13.4.3 Mass spectrometry 61
14 Selection and characterization of control microorganisms 61
14.1 General 61
14.2 Characterization of microorganisms 62
14.2.1 General 62
14.2.2 Phenotypic characterization 62
14.2.3 Molecular characterization 62
14.3 Selection of control microorganisms 62
15 Test report 63
16 Laboratory quality control in microbiology 64
16.1 General 64
16.2 Internal quality control 65
16.2.1 General 65
16.2.2 Process controls 65
16.2.3 Replicate testing 66
16.2.4 Spiked samples 66
16.2.5 IQC assessment using control charts 66
16.3 External quality assessment 66

17 Validation and verification of microbiological methods **67**
17.1 General 67
17.2 Performance characteristics 67
17.3 Validation 67
17.4 Verification 68
Annex A (informative) **Properties of disinfectants** **69**
Annex B (informative) **Confidence intervals for colony count technique** **70**
Annex C (normative) **General confirmation tests** **73**
Bibliography **78**