ISO 23732:2021-07 (E)

Marine technology - Marine environment impact assessment (MEIA) - General protocol for observation of meiofaunal community

Contents

Page

Foreword iv

Introduction v

1 Scope 1

2 Normative references 1

3 Termsanddefinitions 1

4 Principle 2

5 Sampling 2
5.1 Method and equipment 2
5.2 Sub-sampling 3
5.3 Sample preservation 3
5.3.1 Preservation method before analysis 3
5.3.2 Preservation method after analysis 3
5.4 Recording sample information 3

6 Proceduresforimagingflowcytometry(morphologicalanalysis) 3
6.1 Pre-processing of samples 3
6.2 Observation using imaging flow cytometer 3
6.3 Image processing 3

7 Procedures for metagenomic analysis (molecular analysis) 4
7.1 General 4
7.2 Nucleic acids (DNA/RNA) extraction 4
7.3 PCR primers 4
7.4 PCR protocol 4
7.4.1 DNA 4
7.4.2 RNA (cDNA library production) 4
7.5 Check and purification of PCR amplicon 4
7.6 Index PCR 5
7.7 Purification of amplicon by magnetic beads 5
7.8 Concentration measurement 5
7.9 Quality check for amplicon by an automated electrophoresis system using microfluidic chip 5
7.10 Sequencing by a next generation sequencer 5
7.11 Data processing 5

8 Data and sample management 5

9 Combination of morphological analysis and molecular analysis (integrated analysis) 5

Annex A (informative) Example procedure for the analysis of the meiofaunal community byimagingflowcytometry 7

Annex B (informative) Example procedure for analysis of meiofaunal community by NGS 10

Annex C (informative) Example procedure for quantitative analysis of meiofaunal community by NGS **13**

Annex D (informative) Useful information for users **16**

Bibliography **17**