



SLOVENSKI STANDARD
SIST EN ISO 18743:2015/oprA1:2022
01-november-2022

Mikrobiologija v prehranski verigi - Ugotavljanje prisotnosti ličink *Trichinella* v mesu z digestivno metodo - Dopolnilo A1: Revizija besedila in manjših tehničnih vprašanj, posodobitev referenc in vključitev značilnosti delovanja metode z medlaboratorijsko študijo (ISO 18743:2015/DAM 1:2022)

Microbiology of the food chain - Detection of *Trichinella* larvae in meat by artificial digestion method - Amendment 1: Revision of text and minor technical issues, references update, and inclusion of performance characteristics of the method by interlaboratory study (ISO 18743:2015/DAM 1:2022)

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Microbiologie de la chaîne alimentaire - Recherche des larves de *Trichinella* dans la viande par une méthode de digestion artificielle - Amendement 1: Révision du texte et des questions techniques mineures, mise à jour des références et ajout des caractéristiques de performance de la méthode par une étude interlaboratoires (ISO 18743:2015/DAM 1:2022)

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07.100.30 Mikrobiologija živil Food microbiology

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ISO/TC 34/SC 9

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Microbiology of the food chain — Detection of *Trichinella* larvae in meat by artificial digestion method

AMENDMENT 1: Revision of text and minor technical issues, references update, and inclusion of performance characteristics of the method by interlaboratory study

ICS: 07.100.30

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ISO/CEN PARALLEL PROCESSING



Reference number
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This document was prepared by Technical Committee ISO/TC 34, *Food products*, Subcommittee SC 9, *Microbiology*.

Any feedback or questions on this document should be directed to the user's national standards body. A complete listing of these bodies can be found at www.iso.org/members.html.

2, third reference

Replace the text with the following:

World Organisation for Animal Health (OIE), Chapter 3.1.21. — “Trichinellosis”, Manual of Diagnostic Tests and Vaccines for Terrestrial Animals. ed. 2021

3.1, note 1 to entry

Replace the text with the following:

These larvae measure 0,70 mm to 1,10 mm in length and 0,02 mm to 0,04 mm in width.

4.1, first and second line

Replace the text with the following:

(see OIE, Manual of Diagnostic Tests and Vaccines for Terrestrial Animals. ed. 2021, Chapter 3.1.21—“Trichinellosis”, page 649)

ISO 18743:2015/DAM 1:2022(E)*4.2, after the paragraph*

Add the following text as a new paragraph:

In case predilection sites are not available, it is recommended to collect at least twice the minimum amount.

4.8, second sentence

Replace the text with the following:

Knowledge of the basic morphological characteristics of *Trichinella* larvae, including size (0,70 mm to 1,10 mm in length and 0,02 mm to 0,04 mm in width.) and shape is required to examine the sediment (see Figure C.1 and Figure C.2).

5.3

Replace text with the following:

Pepsin (Powdered or granular: 1: 10,000 NF, 1: 12,500 BP, 2,000 FIP; liquid: 660 U/ml).

5.3

Add the following text after the Note:

Liquid pepsin shall be brought at room temperature before use.

5.5

Replace text with the following:

Sodium hypochlorite or **similar disinfectant** of proven efficacy.

6.3

Replace text with the following:

Calibrated scale, for weighing samples and/or pepsin (accuracy $\pm 0,1$ g).

6.10

Replace text with the following:

Funnels (glass, plastic, or steel), of an appropriate size to accommodate the sieve

6.17, last paragraph

Replace text with the following:

Non-disposable equipment must be regularly cleaned by rinsing with hot water, to ensure removal of traces of fat and tissues from previous analyses. In case of positive results in the previous analysis, see decontamination process described at 12 (Safety Measures).

7, last paragraph

Replace text with the following:

Muscle samples should be tested as soon as possible or may be stored for up to 48 h at 2 °C to 8 °C to slow decomposition. Samples shall not be frozen.

8, last sentence

Replace the text with the following and add the following text:

For pools with a lower total muscle mass (up to 50 g), the digest fluid volume and ingredients may be adjusted accordingly to a minimum of 1 l. One liter conical glass separatory funnels and 2 l beakers can be used.

9.2

Replace the text with the following:

For blending/grinding, a small amount (50 ml to 100 ml per 100 g meat) of digest fluid or tap water (45 °C ± 2 °C) can be added to the meat in the blender/grinder to facilitate homogenization. Blending/grinding should be continued until the meat is chopped or minced thoroughly but shall not last so long as to harm the larvae.

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9.3, point a)

Replace the text with the following:

add 16 ml of 25 % hydrochloric acid (see Clause 5) to a glass beaker containing 2 l of tap water preheated to 47 °C ± 2 °C;

9.3, point c)

Replace the text with the following:

add 10 g ± 0,2 g of powdered or granular pepsin (1: 10,000 NF) or 30 ml ± 0,5 ml of liquid pepsin (660 U/ml).

9.3, NOTE 2 last sentence

Replace the text with the following:

For example, if a 37 % stock solution of hydrochloric acid (molar concentration: 12,1) is used, 11 ml is added to 2 l of preheated tap water.

9.3, NOTE 2

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Add the following text after last sentence:

If necessary, the washing step can be repeated.

9.7, point e)

Replace the text with the following:

The final 20 ml of digest fluid in the Petri dish shall be allowed to stand up to 1 min for any larvae to settle before microscopic examination.

9.8, point c)

Replace the text with the following:

Examine the digest fluid in the Petri dish grid by grid with a stereomicroscope or trichinoscope at a 10 X to 20 X magnification for at least 5 min, ensuring the whole Petri dish is examined or until a larva is detected. Examination shall be done systematically, taking care to avoid movement of fluid in the Petri dish.

11

Replace the text with the following:

Results shall be expressed as “presence” or “absence” of *Trichinella* larvae.

12

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Replace the text with the following:

Fluids (digestive fluids, supernatant fluids, rinses, etc.) as well as glassware and other equipment, which could be contaminated with *Trichinella* larvae, shall be decontaminated by heating to 70 °C for at least 1 min, or by chemical alternative methods (e.g., sodium hypochlorite at a final concentration of 0,01 % of active chlorite, at 3 h minimum, or ethanol at final concentration 70 % for at least 1 min) before discarding or washing.

A.1 second paragraph

Replace the text with the following:

For pigs, muscle samples shall be taken from the diaphragm pillars (crus) or from the masseter. In the absence of predilection site muscles, a greater amount of muscle sample (at least twice the minimum recommended amount) must be taken from other striated muscles near bones or tendons.

B.1 first paragraph

Add the following text at the end of first paragraph:

The testing of frozen samples is unreliable, dependent on the species of *Trichinella* infecting, as well as the time and temperature of freezing. The method cannot be used to rule out the presence of dead larvae, which do not survive digestion and should be used in surveillance studies only.

Annex E

Add the following text as a new annex E.

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