PROTOCOL

EURL–*SALMONELLA* PROFICIENCY TEST FOOD-FEED 2019 DETECTION OF *SALMONELLA* spp. IN FLAXSEED

Introduction

This protocol describes the procedures for the Proficiency Test (PT) Food-Feed 2019 on the detection of *Salmonella* spp. in flaxseed amongst the National Reference Laboratories (NRLs) for *Salmonella* in the EU. The samples are artificially contaminated with different levels of a *Salmonella* serovar.

Note that the samples are transported with cooling packs and need to be stored at 5°C (± 3 °C) upon arrival.

The prescribed method is EN ISO 6579-1:2017 (Microbiology of the food chain - Horizontal method for the detection, enumeration and serotyping of *Salmonella* - Part 1: Detection of *Salmonella* spp.) and the underlying EN ISO documents, e.g. the EN ISO 6887 series for preparation of test samples.
Especially the procedure of EN ISO 6887-4:2017 should be respected. To avoid confusion about flaxseed as type of product and observing the practical aspect of this combined food-feed PT, the laboratories are asked to prepare the test samples in this PT as follows:

1. Add the BPW to the 25 gram test sample (instead of weighing accurately the sample into a pre-dispense volume of BPW)
2. Resuscitate the sample for 20 to 30 minutes at 18 °C to 27 °C (room temperature)
3. Mix for 60 s ± 5 s with a homogenizer

*Note: mincing, grinding or crushing the test samples in advance is not necessary

Additionally, laboratories (who are interested) can also perform a second detection method (e.g. PCR, qPCR, ELISA, etc.) to analyse the samples, if this is (routinely) used in their laboratories. These results can be reported in addition to the results of the prescribed method in the result form. Only the results obtained with EN ISO 6579-1:2017 are used to assess the performance of the NRL.
Please report relevant details of the method(s) used in the result form.

Objective

The main objective of the Proficiency Test is to evaluate the performance of the NRLs for *Salmonella* for their ability to detect *Salmonella* spp. at different contamination levels in flaxseed.

Outline of the study

Each participant will receive one box containing 2 large plastic safety bags, packed with cooling elements. The plastic safety bags contain 21 sample bags, consisting of:

- 18 samples of flaxseed (25 grams per sample) artificially contaminated with different levels of a *Salmonella* serovar (coded B1-B18);
- 2 (empty) sample bags, to be used for the control samples, being only BPW (coded C1), and the (own) positive control of the participating laboratory (coded C2).
- 1 sample bag containing a small electronic temperature recorder (coded with labcode).

Upon arrival: all 21 sample bags have to be stored at 5°C (± 3 °C) until the day of analyses (25 March 2019).

The sample bag containing a small electronic temperature recorder will measure the temperature during transport and storage of the samples at the laboratory. The safety bag and temperature recorder will be coded with your lab code. **You are urgently requested to return this complete sample bag with temperature recorder (and lab code) to the EURL-*Salmonella*, at the day your laboratory starts the study (25 March 2019)**. For this purpose a return envelope with a preprinted address label of the EURL-*Salmonella* is included.

Each box will be sent as biological substance category B (UN3373) by door-to-door (for non-EU-MS sometimes door-to-airport) courier service DHL. Please contact EURL-*Salmonella* when the parcel has not arrived at your laboratory by 21 March 2019 (this is 3 working days after the day of mailing).

The performance of the study will start on Monday 25 March 2019.

The media to be used for this study will not be supplied by the EURL

Relevant documents for this Proficiency Test are:

- Protocol 'EURL–*SALMONELLA* PROFICIENCY TEST FOOD-FEED 2019 DETECTION OF *SALMONELLA* spp. IN FLAXSEED' (this document)
- Short guidance on the electronic submission of data in the result form for the EURL-*Salmonella* Proficiency Test on the detection of *Salmonella* spp. in food, animal feed or in samples from the primary production stage
- EN ISO 6579-1:2017. Microbiology of the food chain - Horizontal method for the detection, enumeration and serotyping of *Salmonella* - Part 1: Detection of *Salmonella* spp.
- ISO 6887-1 & 4:2017 Microbiology of food and animal feeding stuffs – Preparation of test samples, initial suspension and decimal dilutions for microbiological examination. Part 1: General rules for the preparation of the initial suspension and decimal dilutions Part 4: Specific rules for the preparation of miscellaneous products

Reporting

All data have to be reported through the result form. The link will be sent by e-mail to the participants.

Submission of data has to be finalised on **16 April 2019** (23:59 h CET) at the latest. **Mind that the result form is no longer accessible after this deadline!** In case you foresee problems with the deadline, please contact us beforehand. The EURL will prepare a summary report soon after the study to inform all NRLs on the overall results.

In case of deviating results, the EURL-*Salmonella* may ask the NRLs to send additional information or to perform additional tests. Therefore, we ask to conserve one *Salmonella* confirmed colony from each positive sample (B1-B18 and C1-C2).

The EURL will prepare a summary report soon after the study to inform all NRLs on the overall results.

Time table EURL–*Salmonella* Proficiency Test Food-Feed 2019
Detection of *Salmonella* spp. In flaxseed

Week (2019)	**Dates**	**Subject**
11	11 – 16 March	Mailing of the protocol and instructions for the result form to the NRLs by e-mail. Sending the link for the result form to the participants by e-mail.
12	18 March	Mailing of parcels to the NRLs as Biological Substance Cat. B (UN3373) by door-to-door courier service. Preparation of media by the NRLs
13	25 March	Performance of the study
16	Before 17 April	Deadline for completing the electronic submission of results: **16 April 2019** (23:59h CET) After this deadline the result form will be closed.

If you have questions or remarks about this study, or in case of problems, please contact:

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