Interim summary report

EURL-*Salmonella* Proficiency Test Primary Production Stage 2025

Detection of *Salmonella* in chicken faeces samples

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1. Introduction

In June/July 2025, the European Union Reference Laboratory for *Salmonella* (EURL-*Salmonella*) organised a Proficiency Test (PT) for the detection of *Salmonella* in samples of the Primary Production Stage (PPS) for the National Reference Laboratories for *Salmonella* (NRLs-*Salmonella*). The matrix under analysis was chicken faeces. NRLs-*Salmonella* which analyse PPS samples were invited to participate in this PT.

In total 35 NRLs-*Salmonella* participated in this PT, originating from 27 EU Member States and 8 third countries (EU candidate Member State, members of the European Free Trade Association (EFTA), United Kingdom, and a non-European country).

2. Samples

The samples of this PT consisted of chicken faeces with different concentrations of *Salmonella* Typhimurium.
Each NRL-*Salmonella* had to analyse in total 14 samples:

- 4 negative samples of each 25 g chicken faeces (no *Salmonella* added);
- 6 samples of each 25 g chicken faeces with a low level of *Salmonella* Typhimurium (STm);
- 4 samples of each 25 g chicken faeces with a high level of *Salmonella* Typhimurium (STm).

The decoding of the samples can be found in Table 3, where also the overview of the results are presented with the corresponding samples.

On Monday 2 June 2025, the PT samples were shipped to the NRLs-*Salmonella*. During transport, the samples were kept at a low temperature using frozen cooling elements and the temperature during transport was registered by an electronic temperature device ('temperature probe'). The NRLs-*Salmonella* could start the analyses of the samples immediately after receipt and at latest by Tuesday 10 June 2025.

The level of natural background flora in the chicken faeces was tested at the EURL-*Salmonella* on 15 April 2025 (shortly after receipt of the chicken faeces) and on 10 June 2025, which was the latest date to start the PT. The total number of aerobic bacteria and the number of *Enterobacteriaceae* in the chicken faeces was examined according to EN ISO 4833-1:2013 and EN ISO 21528-2:2017, respectively. Table 1 shows the number of aerobic bacteria and *Enterobacteriaceae* in the chicken faeces.

Table 1 Number of aerobic bacteria and Enterobacteriaceae *per 25 g chicken faeces sample*

Date	Aerobic bacteria (cfu / sample)	*Enterobacteriaceae* (cfu / sample)
15 April 2025	$7,7 \times 10^{10}$	$1,3 \times 10^{9}$
10 June 2025[a]	$1,0 \times 10^{9}$	$1,1 \times 10^{7}$

[a] After storage at -20 °C for 5 weeks and at 5 °C for 4 weeks

Table 2 shows the (low and high) inoculation levels of the diluted culture of *Salmonella* Typhimurium used to artificially contaminate the chicken faeces samples. Also a five tube Most Probable Number (MPN) test was performed on the artificially contaminated PT samples with a low and high level STm. The MPN test was performed at the latest date to start the PT (10 June 2025), by the EURL-*Salmonella*.

Table 2 Number of Salmonella *Typhimurium in the inoculum for artificial contamination of the 25 g chicken faeces samples and in the PT samples after storage at 5 °C for 14 days*

Date	Low level STm (cfu / sample)	High level STm (cfu / sample)
27 May 2025 Inoculation of chicken faeces samples	21	79
10 June 2025[a] MPN of chicken faeces samples, inoculated with STm (95% confidence limit)	0,8 (0,2 – 2,5)	8,8 (2,8 – 27,5)

[a] After storage at 5 °C for 14 days

3. Results

The results per laboratory, when following EN ISO 6579-1:2017(/A1:2020), can be found in Table 3. Five laboratories also reported results of a second detection method. These results can be found in Table 4.

4. Performance analysis

The performance of the NRLs-*Salmonella* were judged on the results found with EN ISO 6579-1:2017(/A1:2020) only. The criteria for good performance used in the current EURL-*Salmonella* PT for the detection of *Salmonella* in chicken faeces samples are presented in Table 5.

Table 5 Criteria for good performance

Artificially contaminated samples	Percentage positive	# pos samples / total # samples
Negative samples	0%	0 / 4
Low level of *S.* Typhimurium	≥ 50%	≥ 3 / 6
High level of *S.* Typhimurium	≥ 75%	≥ 3 / 4

Thirty-four laboratories fulfilled the criteria for good performance. Of these 34 laboratories, seven (laboratory codes 12, 13, 14, 23, 24, 33 and 35) reported five of the six low level samples positive for *Salmonella*. Four laboratories (laboratory codes 1, 9, 25 and 28) reported four of the six low level samples as positive. One laboratory (laboratory code 24) detected *Salmonella* in only three out of four high level samples. All these results are within the criteria for good performance.
One laboratory did not fulfilled all criteria for good performance. Laboratory 33 detected *Salmonella* in a negative sample, which was assessed as unsatisfactory performance. The EURL-*Salmonella* will contact the laboratory.

Table 3 Results per participant for the detection of Salmonella *in the samples of the EURL-*Salmonella *PT PPS 2025, following EN ISO 6579-1:2017(/A1:2020)*

Sample number	**A1**	**A2**	**A3**	**A4**	**A5**	**A6**	**A7**	**A8**	**A9**	**A10**	**A11**	**A12**	**A13**	**A14**
Level of contamination	N	L	L	H	H	L	L	N	N	L	N	H	L	H
Laboratory code														
1	ND	D	D	D	D	ND[a]	D	ND	ND	ND[a]	ND	D	D	D
2	ND	D	D	D	D	D	D	ND	ND	D	ND	D	D	D
3	ND	D	D	D	D	D	D	ND	ND	D	ND	D	D	D
4	ND	D	D	D	D	D	D	ND	ND	D	ND	D	D	D
5	ND	D	D	D	D	D	D	ND	ND	D	ND	D	D	D
6	ND	D	D	D	D	D	D	ND	ND	D	ND	D	D	D
7	ND	D	D	D	D	D	D	ND	ND	D	ND	D	D	D
8	ND	D	D	D	D	D	D	ND	ND	D	ND	D	D	D
9	ND	D	D	D	D	ND[a]	D	ND	ND	ND[a]	ND	D	D	D
10	ND	D	D	D	D	D	D	ND	ND	D	ND	D	D	D
11	ND	D	D	D	D	D	D	ND	ND	D	ND	D	D	D
12	ND	D	ND[a]	D	D	D	D	ND	ND	D	ND	D	D	D
13	ND	D	D	D	D	D	D	ND	ND	D	ND	D	ND[a]	D
14	ND	D	D	D	D	D	D	ND	ND	D	ND	D	ND[a]	D
15	ND	D	D	D	D	D	D	ND	ND	D	ND	D	D	D
16	ND	D	D	D	D	D	D	ND	ND	D	ND	D	D	D
17	ND	D	D	D	D	D	D	ND	ND	D	ND	D	D	D
18	ND	D	D	D	D	D	D	ND	ND	D	ND	D	D	D
19	ND	D	D	D	D	D	D	ND	ND	D	ND	D	D	D
20	ND	D	D	D	D	D	D	ND	ND	D	ND	D	D	D
21	ND	D	D	D	D	D	D	ND	ND	D	ND	D	D	D
22	ND	D	D	D	D	D	D	ND	ND	D	ND	D	D	D
23	ND	D	D	D	D	D	D	ND	ND	D	ND	D	ND[a]	D
24	ND	D	D	D	D	D	D	ND	ND	D	ND	ND[a]	ND[a]	D
25	ND	D	ND[a]	D	D	ND[a]	D	ND	ND	D	ND	D	D	D
26	ND	D	D	D	D	D	D	ND	ND	D	ND	D	D	D

Sample number	A1	A2	A3	A4	A5	A6	A7	A8	A9	A10	A11	A12	A13	A14
Level of contamination	N	L	L	H	H	L	L	N	N	L	N	H	L	H
Laboratory code														
27	ND	D	D	D	D	D	D	ND	ND	D	ND	D	D	D
28	ND	D	D	D	D	ND[a]	D	ND	ND	D	ND	D	ND[a]	D
29	ND	D	D	D	D	D	D	ND	ND	D	ND	D	D	D
30	ND	D	D	D	D	D	D	ND	ND	D	ND	D	D	D
31	ND	D	D	D	D	D	D	ND	ND	D	ND	D	D	D
32	ND	D	D	D	D	D	D	ND	ND	D	ND	D	D	D
33	ND	D	D	D	D	ND[a]	D	D[b]	ND	D	ND	D	D	D
34	ND	D	D	D	D	D	D	ND	ND	D	ND	D	D	D
35	ND	D	ND[a]	D	D	D	D	ND	ND	D	ND	D	D	D

N = Negative

L = Low level of *S*. Typhimurium (inoculation level 21 cfu/sample)

H = High Level of *S*. Typhimurium (inoculation level 79 cfu/sample)

D = *Salmonella* detected

ND = *Salmonella* not detected

[a] = Deviating from expected result, but within the criteria for good performance

[b] = Deviating from the expected result and outside the criteria for good performance

Table 4 Results per participant for the detection of Salmonella *in the samples of the EURL-*Salmonella *PT PPS 2025, analysed with a second detection method. Only laboratories, which reported results of a second detection method, are presented in this table*

Sample number	**A1**	**A2**	**A3**	**A4**	**A5**	**A6**	**A7**	**A8**	**A9**	**A10**	**A11**	**A12**	**A13**	**A14**
Level of contamination	N	L	L	H	H	L	L	N	N	L	N	H	L	H
Laboratory code														
16	ND	D	D	D	D	D	D	ND	ND	D	ND	D	D	D
18	ND	D	D	D	D	D	D	ND	ND	D	ND	D	D	D
24	ND	D	D	D	D	D	D	ND	ND	D	ND	ND[a]	ND[a]	D
29	ND	D	D	D	D	D	D	ND	ND	D	ND	D	D	D
34	ND	D	D	ND[a]	D	D	D	ND	ND	D	ND	D	D	D

N = Negative

L = Low level of *S*. Typhimurium (inoculation level 21 cfu/sample)

H = High Level of *S*. Typhimurium (inoculation level 79 cfu/sample)

D = *Salmonella* detected

ND = *Salmonella* not detected

[a] = Deviating from expected result, but within the criteria for good performance

References

EN ISO 4833-1:2013. Microbiology of the food chain — Horizontal method for the enumeration of microorganisms — Part 1: Colony count at 30 °C by the pour plate technique. International Organization for Standardization, Geneva, Switzerland.

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