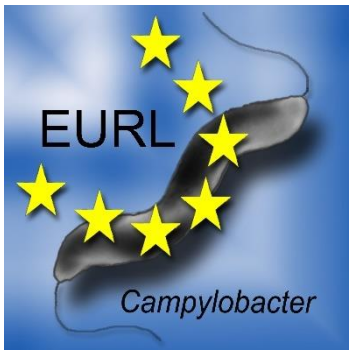

EURL-*CAMPYLOBACTER*

REPORT

PROFICIENCY TEST NUMBER 23

Enumeration of *Campylobacter* in chicken meat

Contents

Abbreviations

C.	*Campylobacter*
cfu	colony forming units
CR	central range
ed.	edition
EU	European Union
EURL	European Union reference laboratory
ISO	International Organization for Standardization
$\log_{10}$	logarithm to base 10 (common logarithm)
MADe	scaled median absolute deviation
MALDI-TOF MS	matrix-assisted laser desorption ionization-time of flight mass spectrometry
mCCD	modified charcoal-cefoperazone-deoxylate (agar)
MS	member state
NMKL	Nordic Committee on Food Analysis (Nordisk metodikkomite for levnedsmidler)
NRL	national reference laboratory
PCR	polymerase chain reaction
PT	proficiency test
spp.	species

Introduction

Proficiency test (PT) number 23 on enumeration of *Campylobacter* spp. in chicken meat was organised by the EU reference laboratory (EURL) for *Campylobacter* in March 2019. Thirty-five national reference laboratories (NRLs) in 28 EU member states (some member states have more than one NRL) and in Iceland, Norway, and Switzerland participated in the PT. The test results and operational details were reported to the EURL from all 35 NRLs. All 35 NRLs reported that they were accredited for detection of *Campylobacter* and 29 were also accredited for enumeration of *Campylobacter*. PT 23 included enumeration of *Campylobacter* in ten chicken meat samples mixed with the freeze-dried contents of vials with or without *Campylobacter* (Table 1). The objective was to assess the performance of the NRLs to enumerate *Campylobacter* in chicken meat. Species identification of detected *Campylobacter* was included as a voluntary part of PT 23.

Table 1. Contents of the ten vials distributed to the NRLs in proficiency test No. 23 (2019).

Sample No.	Species	Level (log cfu/vial)		Batch No.
1	*Campylobacter jejuni**	3.71		SLV306
2	*Campylobacter lari*	4.82		SLV248
3	Negative			SLV289
4	*Escherichia coli*		4.46	SLV150
5	*Campylobacter lari*	4.04		SLV299
6	*Campylobacter jejuni**	3.71		SLV306
7	*Campylobacter jejuni** and *Escherichia coli*	3.50	4.00	SLV313
8	*Campylobacter coli*	5.67		SLV287
9	*Campylobacter jejuni**	4.47		SLV305
10	*Campylobacter coli*	5.67		SLV287

*All *Campylobacter jejuni* strains were hippurate positive.

Terms and definitions

- *Campylobacter* spp.: Thermotolerant *Campylobacter* spp., i.e. which are able to grow at 41.5 °C, foremost (but not exclusively) *C. jejuni*, *C. coli*, *C. lari* and *C. upsaliensis*.
- Enumeration of *Campylobacter*: Determination of the number of *Campylobacter* colony forming units (cfu) per g.
- Confirmation of *Campylobacter* spp.: Microorganisms suspected to be *Campylobacter* spp. are confirmed as such by biochemical methods and/or by molecular methods.
- Species identification of *Campylobacter*: Identification of thermotolerant *Campylobacter* species with biochemical methods and/or by molecular methods.

Outline of the proficiency test

Preparation of the chicken meat

The chicken meat used as matrix in the PT was obtained from a broiler producer that had not delivered any *Campylobacter*-positive flocks to slaughter for more than one year. The broilers were slaughtered at a slaughterhouse with a very low general level of *Campylobacter*-positive flocks (less than 5 % during the previous year) and no positive flocks at all for two months before taking out and sending broiler carcasses to the EURL. Chicken skin and caecal samples from the broiler flock tested negative for presence of *Campylobacter*. The chicken meat was freeze-stored until distribution of the PT.

Production and quality control of the vials

The vials with freeze-dried bacterial cultures used in the PT were produced and tested for stability and homogeneity by the Swedish National Food Agency. Before choosing the vials for the PT, the EURL tested three vials of each batch with modified charcoal-cefoperazone-deoxylate (mCCD) agar. The results were noted as common logarithm values (log_{10}) of colony forming units (cfu) for analysis of each tested vial and values for the difference between the highest and lowest values. The vials chosen for the PT included vials with both high and low *Campylobacter* levels, and the maximum difference allowed was 0.50 log_{10} cfu. In addition, enumeration of *Campylobacter* spp. in chicken meat according to ISO 10272-2:2017 was performed by the EURL three times for each batch: before dispatching, just after dispatching and two weeks after dispatching, i.e. at the last time for start of the analysis by the participants. This was done to check for possible matrix effects as well as the stability of the vials and matrix together.

Distribution of the proficiency test

The PT samples were distributed from the EURL on 11th of March, 2019. The samples were placed in foam boxes along with freezing blocks. The foam boxes were packed in cardboard boxes for transportation and were sent from the EURL using courier service.

Each participant received a package containing:

- ten numbered vials; each containing freeze-dried material with or without *Campylobacter* spp., and
- one plastic bag with chicken meat (ca 120 g), to be divided into 10 g portions, one for each of the ten vials.

Twenty-seven NRLs received the PT within one day after the packages had been dispatched from the EURL, and eight NRLs two days after (Table 2). A Micro-T-Log was included in each shipment to record the temperature every second hour during transport.

Table 2. Dates of arrival and start of the analysis of proficieny test No. 23, 2019.

Arrival	**Number of NRLs**	**Start of analysis**	**Number of NRLs**
12th of March	27	12th of March	2
13th of March	8	13th of March	11
		14th of March	3
		17th of March	1
		18th of March	8
		19th of March	3
		20th of March	1
		21th of March	1
		25th of March	4
		26th of March	1

All results had to be reported in the Questback Essentials system by 15th of April, 2019. The analysis was recommended to be started the same week as the PTs were dispatched from the EURL. The NRLs were recommended to follow ISO 10272-2:2017 for performing PT 23. However, if their standard laboratory procedure followed a different method, they were allowed to use that method for the test. Instructions for preparation of an initial dilution of each sample were included in the packages, and were also sent out by e-mail a few days before he PT distribution. If the analysis could not be started the same week, the chicken meat was recommended to be stored in −20 °C for up to two weeks and the vials in −20 °C for one week or in −70 °C for two weeks. The dates for the start of analysis are presented in Table 2.

Used methods

Thirty-two NRLs reported to have followed the recommended method of ISO 10272-2:2017. Two NRLs reported to have used NMKL 119, 3rd ed. 2007, and one NRL an internal method.

Campylobacter spp. should be incubated in a microaerobic atmosphere, with oxygen content of 5%±2%, and carbon dioxide 10%±3%. The appropriate microaerobic atmosphere can be obtained by using commercially available microaerobic incubators, commercial gas-generating kits, or by using gas-jars, filled with the appropriate gas mixture prior to incubation. Of the 35 NRLs, 17 reported using commercial gas-generating kits, 12

microaerobic incubators, six the Anoxomat® system and three other methods (jars filled with gas mixture, zip-lock bags filled with gas or GENbox microaer-generator). Some of the NRLs used more than one system.

Assessing the performance of the NRLs

Assessment of performance in enumeration

The median values of the log-transformed cfu of *Campylobacter* spp. reported by all NRLs were used as assigned values for the eight *Campylobacter*-positive samples. The performance in enumeration was assessed by using scaled median absolute deviation (MADe) from the median values for calculating z-scores. The scaled MADe method is used to identify outlying counts when fewer than 50 participants undertake an enumeration (ISO 22117:2019). A scoring system was used for assessing the performance in enumeration of each sample, where results within median value ±2σMADe ($|z| \leq 2.0$) were given score 2, results between ±2σMADe and ±3σMADe ($2.0 < |z| < 3.0$) were given score 1 and results outside ±3σMADe ($|z| \geq 3.0$) were given score 0. For the two pairs of samples made from the same batches of vials (sample No. 1 and 6, and sample No. 8 and 10, respectively), it was checked that values falling outside the limits (i.e., given a score of 0 or 1) were doing so according to the limits for both samples. For the *Campylobacter*-negative samples a score of 2 were given when no campylobacters were reported, and a score of 0 when a false positive result was reported.

An overall assessment of all ten enumerations was performed by summarising all the scores for each NRL. A five-level grading scale was used for the overall assessment: excellent, good, acceptable, needs improvement and poor. “Excellent performance” was considered if all enumerations were within median values ±2σMADe and no campylobacters were reported in the two *Campylobacter*-negative samples, i.e. the total score was 20. “Good performance” was considered if the NRL had a score of 17–19. “Acceptable performance” was considered if the NRL had a score of 14–16. “Needs improvement” were given to NRLs with a score of 12–13 and those with a score of <12 were considered to have a “poor performance”.

Assessment of performance in species identification

The performance in correctly identifying the species for the samples where *Campylobacter* was detected, the sensitivity, was categorized on a five-level grading scale. The limits were set at the same levels of sensitivity as the scoring percentages for the enumeration performance grading.

Results

Proficiency test number 23 was distributed to 35 NRLs and all of them reported the results of the analysis. Seventeen laboratories started the analyses the same week the samples were dispatched from the EURL, thirteen NRLs the week after, and five NRLs two weeks after the PT was dispatched from the EURL (Table 2).

Enumeration of *Campylobacter* spp. (mandatory)

Of the 35 laboratories, 33 correctly reported *Campylobacter* spp. in all samples where *Campylobacter* spp. were included and not *Campylobacter* in the samples without *Campylobacter*. One false positive result, of sample No. 4, and one false negative result, of sample No. 5, were reported. The median values of the enumerations varied from 2.70 (sample No. 6) to 4.38 (sample No. 8 and 10) log cfu/g (Figure 1 and Figure 2a).

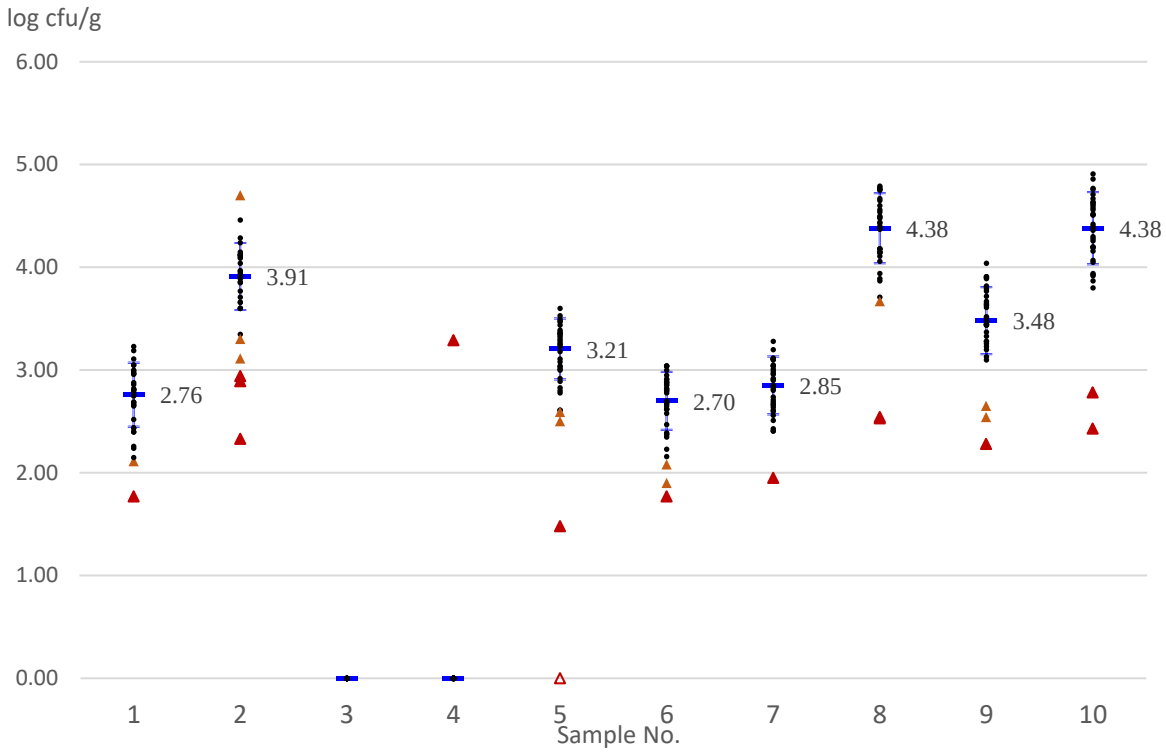

Figure 1. The number (log_{10} cfu/g) of *Campylobacter* spp. reported by 35 laboratories in PT 23 (2019). The samples reported as *Campylobacter* spp. not detected are shown as 0 in the figure. The median values are displayed in numbers and marked with horizontal lines. Vertical bars show the σMADe. Values outside the ±2σMADe and ±3σMADe limits are shown as small and large triangles, respectively.

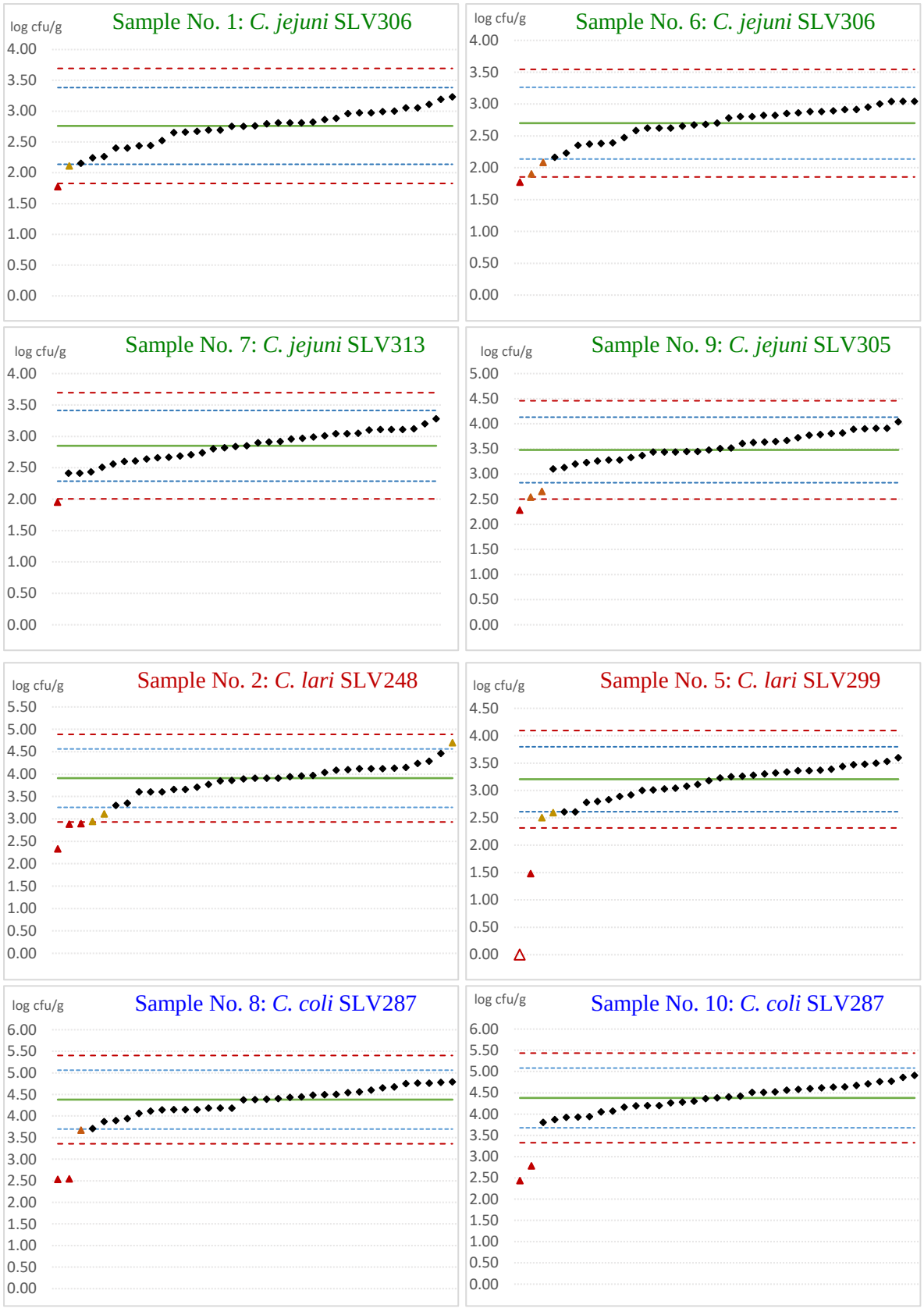

Figure 2a. The number (log_{10} cfu/g) of *Campylobacter* spp. reported for each of the eight *Campylobacter*-positive samples by 35 laboratories in PT 23 (2019). Results for samples made from the same batches of vials (sample No. 1 and 6, and sample No. 8 and 10, respectively) are placed beside each other. Samples reported as *Campylobacter* spp. not detected are shown as 0 in the figure. The median values and the ±2σMADe and ±3σMADe limits are shown as horizontal lines. Values outside any of the limits are shown as triangles.

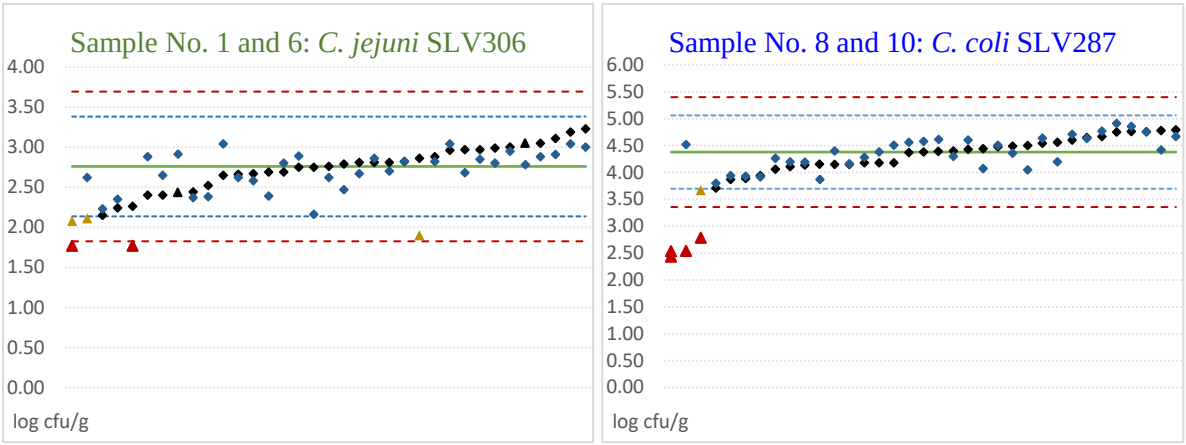

Figure 2b. The number (log_{10} cfu/g) of *Campylobacter* spp. reported for samples of *C. jejuni* and *C. coli* by 35 laboratories in PT 23 (2019). The results of the samples made from the same batches of vials are displayed in the same diagram, with the two results from the same laboratory at the same x axis point. Sample No. 1 and 8 are represented by black dots, and sample No. 6 and 10 by blue dots. The median values and the ±2σMADe and ±3σMADe limits are shown as horizontal lines. Values outside any of the limits are shown as triangles.

The within-laboratory variation was examined for the four samples made from the same two batches of vials: the *C. jejuni* SLV306 samples No. 1 and No. 6, and the *C. coli* SLV287 samples No. 8 and No. 10, respectively (Figure 2b). The median within-laboratory difference between the two samples was 0.15 log cfu/g for *C. jejuni* SLV306, and 0.12 log cfu/g for *C. coli* SLV287. The mean difference was 0.23 log cfu/g for both batches.

Performance in enumeration of *Campylobacter* spp.

The results of using the five-level grading scale for the overall assessment of the NRLs' enumeration of *Campylobacter* spp. are presented in Table 3 and Figure 3.

According to the assessment, 32 NRLs (29 of the MS-NRLs) fulfilled the criterion for excellent or good performance and one MS-NRL scored below the acceptable limit (Table 3 and Figure 3). The overall median percentage of scores was 100% (50% Central Range (CR): 90.0%–100%).

The underperforming NRL reported generally lower log values than average, but also that they had probably mixed-up the matrices between PT 23 and PT 24 (the detection PT received at the same time). The minced chicken meat intended for PT 24 was, in contrast to the chicken meat for PT 23, contaminated with *Candida* to make the detection somewhat more challenging. The matrix mix-up was judged as a probable explanation to the lower enumeration results and the following underperformance in this case.

The NRLs' enumeration results and z-scores for the eight *Campylobacter*-positive samples included in PT 23 are presented in Table 4.

Table 3. Overall performance of the NRLs' enumeration of *Campylobacter* spp. (n=35) in proficiency test No. 23 (2019).

Grade	Scoring limits for each performance grade	Number (proportion) of NRLs with performance at each level	
		All NRLs n=35	MS-NRLs n=32
Excellent	95.1–100%	21 (60%)	20 (62%)
Good	85.0–95.0%	11 (31%)	9 (28%)
Acceptable	70.0–84.9%	2 (6%)	2 (6%)
Needs improvement	57.0–69.9%	1 (3%)	1 (3%)
Poor	<57.0%	0 (0%)	0 (0%)

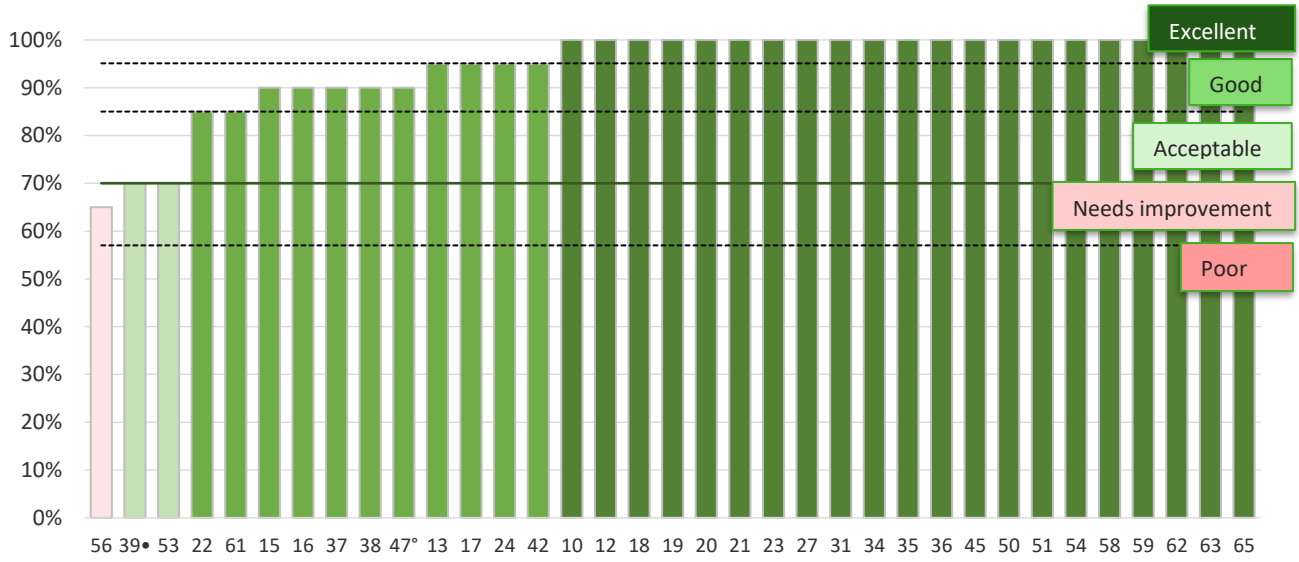

Figure 3. Distribution of the results of participating NRLs (n=35), represented by lab ID, in combined score for enumerations of the eight *Campylobacter*-positive samples and two *Campylobacter*-negative samples in PT 23 (2019). Limits for grading of the overall performance are marked by horizontal lines. Each ° stands for a false negative result, and • for a false positive result.

Table 4. Results from the enumeration and z-scores of *Campylobacter*-positive samples in proficiency test No. 23 (2019). Yellow shadowed cells indicate values outside median values ±2σMADe and z-scores ±2.0. Red shadowed cells indicate values outside median values ±3σMADe and z-scores ±3.0.

	Sample 1		**Sample 2**		**Sample 5**		**Sample 6**		**Sample 7**		**Sample 8**		**Sample 9**		**Sample 10**	
Lab id	log_{10} cfu/g	z-score	log_{10} cfu/g	z-score	log_{10} cfu/g	z-score	log_{10} cfu/g	z-score	log_{10} cfu/g	z-score	log_{10} cfu/g	z-score	log_{10} cfu/g	z-score	log_{10} cfu/g	z-score
10	3.05	0.93	3.35	-1.72	3.36	0.52	2.78	0.28	3.05	0.71	4.76	1.11	3.77	0.89	4.86	1.37
12	2.44	-1.04	4.29	1.15	3.36	0.51	2.91	0.76	2.80	-0.16	4.39	0.03	3.63	0.46	4.62	0.67
13	3.19	1.38	4.70	2.42	3.60	1.33	3.04	1.21	3.28	1.53	4.79	1.20	3.91	1.32	4.67	0.83
15	2.81	0.16	2.33	-4.84	2.92	-0.96	2.67	-0.11	2.99	0.50	3.94	-1.29	3.10	-1.17	3.92	-1.31
16	2.52	-0.77	3.71	-0.61	1.48	-5.82	2.38	-1.14	2.43	-1.49	4.15	-0.67	3.37	-0.34	3.87	-1.45
17	2.86	0.32	3.60	-0.95	3.25	0.15	1.90	-2.84	2.41	-1.56	4.40	0.06	3.20	-0.86	4.30	-0.23
18	2.81	0.16	4.24	1.01	3.37	0.56	2.86	0.57	3.10	0.89	4.56	0.53	3.89	1.26	4.20	-0.51
19	2.76	0.00	3.60	-0.95	2.89	-1.06	2.62	-0.28	2.84	-0.04	4.14	-0.70	3.44	-0.12	4.19	-0.54
20	2.81	0.16	4.13	0.67	2.61	-2.01*	2.70	0.00	2.74	-0.39	4.50	0.35	3.26	-0.67	4.05	-0.94
21	3.23	1.51	3.94	0.09	2.61	-2.01*	3.00	1.06	3.11	0.92	3.89	-1.44	3.67	0.58	3.93	-1.28
22	2.44	-1.03	2.88	-3.16	2.50	-2.38	2.37	-1.17	2.56	-1.03	4.06	-0.94	3.44	-0.12	4.26	-0.34
23	2.75	-0.03	4.12	0.64	2.78	-1.43	2.89	0.67	3.01	0.57	3.87	-1.50	3.33	-0.46	3.94	-1.25
24	2.66	-0.32	3.11	-2.45	3.11	-0.32	2.62	-0.28	2.82	-0.11	4.15	-0.67	3.23	-0.77	4.40	0.06
27	2.79	0.10	3.97	0.18	3.01	-0.66	2.47	-0.82	2.71	-0.50	4.18	-0.59	3.65	0.52	4.28	-0.28
31	2.75	-0.03	3.86	-0.15	3.08	-0.42	2.16	-1.92	2.60	-0.89	4.18	-0.59	3.64	0.49	4.38	0.00
34	3.05	0.93	4.46	1.69	3.03	-0.59	2.88	0.64	3.04	0.67	4.67	0.85	3.61	0.40	4.77	1.11
35	2.82	0.19	3.77	-0.43	3.44	0.79	2.82	0.43	3.11	0.92	4.60	0.65	3.90	1.29	4.71	0.94
36	2.97	0.67	3.91	0.00	3.47	0.89	2.68	-0.07	2.91	0.21	4.49	0.32	3.48	0.00	4.36	-0.06
37	2.11	-2.09	2.94	-2.97	3.00	-0.69	2.62	-0.28	2.41	-1.56	4.18	-0.59	3.28	-0.61	4.51	0.37
38	2.96	0.64	2.89	-3.13	3.26	0.19	3.04	1.21	3.20	1.24	4.76	1.11	4.04	1.72	4.76	1.08
39	2.26	-1.61	4.12	0.64	2.83	-1.26	1.77	-3.30	2.92	0.25	2.54	-5.40	3.91	1.32	4.52	0.40
42	2.65	-0.35	3.85	-0.18	2.59	-2.07	3.04	1.21	3.04	0.67	4.38	0.00	3.51	0.09	4.58	0.57
45	3.11	1.12	4.09	0.55	3.18	-0.08	2.91	0.75	2.67	-0.64	4.44	0.18	3.44	-0.12	4.07	-0.88
47	2.24	-1.67	3.91	0.00	<1.00	-7.45**	2.35	-1.24	2.61	-0.85	3.71	-1.96	3.28	-0.61	3.80	-1.65
50	2.69	-0.22	3.96	0.15	3.48	0.93	2.39	-1.10	2.85	0.00	4.15	-0.67	3.13	-1.07	4.16	-0.63
51	2.40	-1.16	3.89	-0.06	3.34	0.46	2.88	0.64	3.11	0.92	4.43	0.15	3.45	-0.09	4.60	0.63
53	2.88	0.39	3.30	-1.87	3.04	-0.56	2.82	0.43	2.66	-0.67	2.53	-5.43	2.28	-3.68	2.43	-5.55
54	2.40	-1.16	4.04	0.40	3.32	0.39	2.65	-0.18	2.64	-0.75	4.11	-0.79	3.45	-0.09	4.20	-0.51
56	1.77	-3.18	3.60	-0.95	2.80	-1.37	2.08	-2.20	2.51	-1.21	3.67	-2.08	2.54	-2.88	2.78	-4.55
58	2.99	0.74	3.66	-0.77	3.30	0.32	2.80	0.35	2.97	0.43	4.65	0.79	3.72	0.74	4.63	0.71
59	2.69	-0.22	3.91	0.00	3.23	0.08	2.80	0.35	2.96	0.39	4.75	1.09	3.52	0.12	4.91	1.51
61	2.15	-1.96	3.66	-0.77	3.39	0.62	2.23	-1.67	1.95	-3.19	4.48	0.29	2.65	-2.54	4.51	0.37
62	3.00	0.77	4.15	0.74	3.28	0.25	2.95	0.89	2.90	0.18	4.54	0.47	3.82	1.04	4.64	0.74
63	2.67	-0.29	4.12	0.64	3.50	0.99	2.58	-0.43	2.69	-0.57	4.37	-0.03	3.81	1.01	4.56	0.51
65	2.97	0.67	4.10	0.58	3.53	1.10	2.85	0.53	3.12	0.96	4.78	1.17	3.79	0.95	4.42	0.11
Median	**2.76**		**3.91**		**3.21**		**2.70**		**2.85**		**4.38**		**3.48**		**4.38**	
MADe	0.21		0.22		0.20		0.19		0.19		0.23		0.22		0.24	
σMADe	0.31		0.33		0.30		0.28		0.28		0.34		0.33		0.35	
±2σMADe	3.39	2.13	4.57	3.25	3.80	2.61	3.27	2.13	3.42	2.28	5.07	3.69	4.14	2.82	5.09	3.67
±3σMADe	3.70	1.82	4.89	2.93	4.10	2.31	3.55	1.85	3.70	2.00	5.41	3.35	4.46	2.50	5.44	3.32

*Rounded to –2.0 and considered on the limit, not exceeding it.

**Calculated from 1.00 log cfu/g.

Species identification of *Campylobacter* spp. (voluntary)

Thirty-two (91%) of the 35 NRLs reported results of species identification (Table 5). Sample No. 1, 2, 6 and 10 were correctly identified by all 32 NRLs.

Table 5. Species identification reported by 32 NRLs in the voluntary part of proficiency test No. 23 (2019).

		Number of NRLs reporting				
Content of sample (vial)		*Campylobacter jejuni*	*Campylobacter coli*	*Campylobacter lari*	*Campylobacter* spp. but unable to identify species	Other/No growth
1.	*Campylobacter jejuni*	32				
2.	*Campylobacter lari*			32		
3.	Negative					32
4.	*Escherichia coli*					32
5.	*Campylobacter lari*			31	1	
6.	*Campylobacter jejuni*	32				
7.	*Campylobacter jejuni* & *Escherichia coli*	30		1	1	
8.	*Campylobacter coli*	1	31			
9.	*Campylobacter jejuni*	30	1		1	
10.	*Campylobacter coli*		32			

The isolated *Campylobacter* spp. were identified by biochemical methods and/or molecular methods, polymerase chain reaction (PCR), matrix-assisted laser desorption ionization-time of flight mass spectrometry (MALDI-TOF MS) or whole genome sequencing (WGS). The biochemical methods included detection of catalase, hippurate hydrolysis, indoxyl acetate hydrolysis, sensitivity to nalidixic acid and cephalotin, and H_2S production in triple sugar iron medium.

Seventeen of the 32 NRLs reported that they used MALDI-TOF MS for the species identification, in six cases in combination with other techniques. Thirteen NRLs used PCR assays, in six cases in combination with other techniques. Nine NRLs reported to have used the multiplex PCR assay published by Wang *et al.* (2002). Other protocols reported to be used or adapted by more than one NRL were the PCR assays by Denis *et al.* (1999), Best *et al.* (2003) and Mayr *et al.* (2010). Ten NRLs used biochemical methods (at least detection of catalase), in six cases in combination with MALDI-TOF MS or PCR. One NRL used WGS for the species identification.

Twenty-three NRLs used one technique only (a set of biochemical tests regarded as one technique) and nine NRLs combined two techniques for the species identification.

Performance in identification of *Campylobacter* spp.

Of the 32 NRLs reporting results for species identification of *Campylobacter*, 30 fulfilled the criterion for excellent or good performance in identification of *Campylobacter* spp., and one scored below the acceptable limit (Table 6). The overall median sensitivity in correctly identifying *Campylobacter* spp. was 100% (50% CR: 100%–100%).

Table 6. Overall performance of NRLs' sensitivity in correctly identifying *Campylobacter* spp. in the voluntary part of PT 23 (2019).

		Identification of *Campylobacter* spp.	
Grade	**Sensitivity**	**Number of NRLs (%) All NRLs, n=32**	**Number of NRLs (%) MS-NRLs, n=29**
Excellent	95.1–100%	29 (91%)	26 (90%)
Good	85.0–95.0%	1 (3%)	1 (3%)
Acceptable	70.0–84.9%	1 (3%)	1 (3%)
Needs improvement	57.0–69.9%	1 (3%)	1 (1%)
Poor	<57.0%	0 (0%)	0 (0%)

Summary of the proficiency test number 23, 2019

Of the 35 laboratories 32 (81%) had good or excellent performance considering the enumeration which is about the same level as the four previous years (Table 7). Only one NRL (3%) scored below the acceptable limit.

Because of changes in the new version of ISO 22117, the limit for deeming an individual result as non-acceptable was slightly more generous (score 0 only if the absolute value of z-score was above 3) than previous year. However, although this changed the combined score in a few cases, it did not at all affect the grading.

Table 7. Overall performance of the NRLs' enumeration of *Campylobacter* spp. in proficiency test (PT) No. 23, 2019, compared to performance in PTs for previous years, as well as grades for the results of the NRLs.

	All samples (n=10)		Only *Campylobacter*-positive samples (n=8)		
Grade	**PT 23 (2019) Number of NRLs (%) n=35**	**PT 21 (2018) Number of NRLs (%) n=37**	**PT 19 (2017) Number of NRLs (%) n=36**	**PT 17 (2016) Number of NRLs (%) n=36**	**PT 15 (2015) Number of NRLs (%) n=36**
Excellent	21 (60%)	20 (54%)	22 (61%)	26 (72%)	17 (47%)
Good	11 (31%)	11 (30%)	9 (25%)	6 (17%)	12 (33%)
Acceptable	2 (6%)	3 (8%)	2 (6%)	1 (3%)	2 (6%)
Needs improvement	1 (3%)	1 (3%)	0 (0%)	2 (6%)	2 (6%)
Poor	0 (0%)	2 (5%)	3 (8%)	1 (3%)	3 (8%)

Species identification of *Campylobacter* was included as a voluntary part in PT 23, and 32 (91%) of the 35 laboratories reported results of species identification (Table 8). The performance was high (94% excellent or good) and only one NRL (3%) scored below the acceptable limit.

Table 8. Overall performance of NRLs' sensitivity in correct species identification of *Campylobacter* in proficiency test No. 23, 2019, compared to performance in proficiency tests (PT) for previous years, as well as grades for the results of the NRLs.

Grade	PT 23 (2019) Number of NRLs (%) n=32	PT 21 (2018) Number of NRLs (%) n=33	PT 19 (2017) Number of NRLs (%) n=31	PT 17 (2016) Number of NRLs (%) n=29	PT 15 (2015) Number of NRLs (%) n=27
Excellent	29 (91%)	29 (88%)	30 (97%)	27 (93%)	26 (96%)
Good	1 (3%)	3 (9%)	1 (3%)	1 (3%)	0 (0%)
Acceptable	1 (3%)	1 (3%)	0 (0%)	0 (0%)	0 (0%)
Needs improvement	1 (3%)	0 (0%)	0 (0%)	1 (3%)	0 (0%)
Poor	0 (0%)	0 (0%)	0 (0%)	0 (0%)	1 (4%)

The majority of the NRLs had excellent or good performance in both enumeration and species identification, meeting the requirements of being a NRL. A matrix mix-up was deemed to be a probable explanation to one NRL not reaching the acceptable criterion for enumeration.

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