

Photo: Linda Svensson, EURL-Campylobacter/SVA

Proficiency tests (PT)

EURL for *Campylobacter* organise at least one proficiency tests (PT) each year for the National Reference Laboratories (NRL) to detect and quantify *Campylobacter* spp. in different types of matrices. The aim with the PTs is to provide NRLs with details of relevant analytical methods for analysing samples that mimic realistic diagnostic samples to be analysed for *Campylobacter* in the member states (MSs). The aim is also to assess the performance of the NRLs and thereby to identify potential analytical problems that could be solved by assistance from the EURL in order to improve the performance. In some countries the accreditation bodies request to see the results from the proficiency test for those specific countries. Therefore the selection of matrices has in recent years been influenced by the accreditation demands.







The type of matrices for the PTs are discussed with the NRLs at the preceding workshops. The demand on selected matrix are what is relevant for monitoring *Campylobacter* spp. or as a tracing source of infection in outbreak situations. Before sending out the proficiency tests, thorough analysis of the same batches of materials that is to be sent out to the NRLs has been made by the EURL staff.

Since 2007, 20 PTs have been organised. The different matrices have been: chicken meat, chicken skin, chicken caeca, milk filters, minced meat, sock samples and swab samples. In addition to *Campylobacter* spp., the PTs can also include other bacteria spp. that are relevant for the selected matrix. It is also essential that there are samples in the PTs that do not contain *Campylobacter*, as a test for cross-contamination in the laboratories, and to be able to calculate the performance.

Basically, the protocols for analysis (the Standard Operating Procedure, SOPs) have followed the standardized protocols of ISO 10272 Part 1 and Part 2: 2006 "Microbiology of food and animal feeding stuffs – Horizontal method for detection and enumeration of *Campylobacter* spp.". The PT protocols have also included mandatory or voluntary parts using different selective media and incubation times. This has been done in order to obtain more knowledge about analysing *Campylobacter* that are required for the ongoing revision of the standard.



The results of the PTs, including performance criteria and proposed limits for good performance, are presented and discussed at the EURL- *Campylobacter* workshops. Final reports are sent to the NRLs/participating laboratories and to DG-SANTÉ after the workshops.





Below are a list of proficiency test sent out to the NRLs by the EURL-Campylobacter.

Year	PT no.	Name of the proficiency test
2007	PT1	Detection and species identification of <i>Campylobacter</i> in chicken caecal contents
	PT2	Campylobacter detection in chicken caeca
2008	PT3	Detection and enumeration of <i>Campylobacter</i> spp. in broiler skin
	PT4	Species identification of <i>Campylobacter</i> by three PCR assays
2009	PT5	Detection and enumeration of <i>Campylobacter</i> spp. in broiler meat
2010	PT6	Identification of <i>Campylobacter</i> spp. in swabs samples
	PT7	Detection and enumeration of <i>Campylobacter</i> spp. in broiler meat
2011	PT8	Detection and enumeration of <i>Campylobacter</i> spp. in minced meat
2012	PT9	Detection and enumeration of <i>Campylobacter</i> spp. in chicken breast muscle
	PT10	Identification of <i>Campylobacter</i> spp. in swabs samples
2013	PT11	Enumeration (and voluntary detection after enrichment) of <i>Campylobacter</i> spp. in chicken breast muscle
	PT12	Detection and species identification of <i>Campylobacter</i> in sock samples
2014	PT13	Enumeration (and voluntary detection after enrichment) of <i>Campylobacter</i> spp. in minced meat
	PT14	Detection and species identification of <i>Campylobacter</i> spp. in milk filters





Continuation of the list of proficiency test sent out to the NRLs by the EURL-Campylobacter.

Year	PT no.	Name of the proficiency test
2015	PT15	Detection and enumeration of <i>Campylobacter</i> spp. in chicken meat
	PT16	Detection and enumeration of <i>Campylobacter</i> spp. in sock samples
2016	PT17	Enumeration (and voluntary detection after enrichment) of <i>Campylobacter</i> spp. in chicken skin.
	PT18	Detection and species identification of <i>Campylobacter</i> spp. in caecum samples.
2017	PT19	Detection and enumeration of <i>Campylobacter</i> in minced chicken meat
	PT20	Detection and species identification of <i>Campylobacter</i> spp. in swab samples
2018	PT21	Enumeration (and voluntary detection and species identification) of <i>Campylobacter</i> in chicken skin
	PT22	Detection and species identification of <i>Campylobacter</i> spp. in chicken faecal swab samples





Detection and species identification of *Campylobacter* in chicken caecal contents (2007)

Detection and species identification of *Campylobacter* spp. (live cultures) in swabs with chicken caecum contents. The aims were to test the capacity of the NRLs to detect *Campylobacter* in chicken faeces and to provide information about the logistics of transportation of samples from the EURL to the NRLs.



Mandatory/Voluntary:	Voluntary
No of samples:	5
No of participating laboratories:	24

Injection of live culture of Campylobacter spp. into chicken caecum. Photo: Linda Svensson, EURL-Campylobacter/SVA

PT2

Campylobacter detection in chicken caeca (2007)

Detection and species identification of *Campylobacter* spp. in chicken caeca, naturally contaminated or inoculated with live cultures of *Campylobacter jejuni* or *Campylobacter coli*. Also caeca without *Campylobacter* spp. were included. The aim was to test the capacity of the NRL to detect *Campylobacter* in chicken faeces.

Mandatory/Voluntary:	Mandatory
No of samples:	12
No of participating laboratories.	25





Detection and enumeration of Campylobacter spp. in broiler skin (2008)

Detection, species identification and enumeration of *Campylobacter* spp. in carcass inoculated or not inoculated with live cultures of *Campylobacter* spp.

The aims were to assess the performance of the NRLs to detect and enumerate *Campylobacter* in broiler skin samples, and to evaluate the standard operating procedures for this kind of *Campylobacter* analyses.

Mandatory/Voluntary:	Mandatory
No of samples:	14
No of participating laboratories.	31

PT4

Species identification of Campylobacter by three PCR assays (2008)

Identification of *Campylobacter* species by 3 PCR assays. Bacterial DNA, primers and protocols were provided by the EURL. The aim was to test the robustness of three traditional and commonly used PCR-based assays for genus/species identification of *Campylobacter* species.



Photo: Bengt Ekberg/SVA

Mandatory/Voluntary:	Voluntary
No of samples:	11
No of participating laboratories:	25





Detection and enumeration of Campylobacter spp. in broiler meat (2009)

Detection, species identification and enumeration of *Campylobacter* spp. (freeze dried bacteria) in chicken meat (breast fillets). The aim was to assess the performance of the NRLs to detect and enumerate *Campylobacter* in broiler meat samples.



Preparation of chicken meat for the proficiency test.

Photo: Linda Svensson, EURL-Campylobacter/SVA

Mandatory/Voluntary:	Mandatory
No of samples:	11
No of participating laboratories	: 35

PT6

Identification of Campylobacter spp. in swab samples (2010)

Detection and species identification of *Campylobacter* (live cultures) in swab samples. The aim was to obtain information about the NRLs ' capacities and methods used for identification of *Campylobacter spp*. of different species, including rare species and those that might be difficult to identify.

Mandatory/Voluntary:	Voluntary
No of samples:	10
No of participating laboratories:	33





Detection and enumeration of Campylobacter spp. in broiler meat (2010)

Detection, species identification and enumeration of *Campylobacter* spp. (freeze dried bacteria) in chicken meat (breast fillets). The aims were to assess the performance of the NRLs to detect and enumerate *Campylobacter* in broiler meat samples, to evaluate different protocols for *Campylobacter* analyses, and to make a first outline of what could be considered "good performance" in the PT.



Mandatory/Voluntary:	Mandatory
No of samples:	10
No of participating laboratories	34

Freeze dried bacteria used in the proficiency test. Photo: Linda Svensson, EURL-Campylobacter/SVA

PT8

Detection and enumeration of Campylobacter spp. in broiler meat (2011)

Detection, species identification and enumeration of *Campylobacter* spp. (freeze dried bacteria) in chicken meat (minced fillets). The aims were to assess the performance of the NRLs to detect and enumerate *Campylobacter* in minced meat samples and to evaluate different protocols for *Campylobacter* analyses.

Mandatory/Voluntary:	Voluntary
No of samples:	10
No of participating laboratories:	34





Detection and enumeration of *Campylobacter* spp. in chicken breast muscle (2012)

Detection, species identification and enumeration of *Campylobacter* spp. (freeze dried bacteria) in chicken meat (breast fillets). The aims were to assess the performance of the NRLs to detect and enumerate *Campylobacter* in broiler meat samples and to evaluate different protocols for *Campylobacter* analyses.

Mandatory/Voluntary:	Mandatory
No of samples:	12
No of participating laboratories.	: 36

PT10

Identification of Campylobacter spp. in swab samples (2012)

Detection and species identification of *Campylobacter* spp. (live cultures) in swab samples. The aim was to obtain information about the NRLs ' capacites and methods used for identification of *Campylobacter* of different species, including rare species and those that might be difficult to identify.



Photo: Linda Svensson, EURL-Campylobacter/SVA

Mandatory/Voluntary:	Voluntary
No of samples:	12
No of participating laboratories:	29





Enumeration (and voluntary detection after enrichment) of *Campylobacter* spp. in chicken breast muscle (2013)

Detection and enumeration of *Campylobacter* spp. (freeze dried bacteria) in chicken breast muscle samples. The aim was to assess the performance of the NRLs to detect and enumerate *Campylobacter* in breast muscle samples. Detection after enrichment and species identification of *Campylobacter* were included as voluntary parts of the PT.

Mandatory/Voluntary:	Mandatory (some parts voluntary)
No of samples:	10
No of participating laborator	ies: 36

PT12

Detection and species identification of *Campylobacter* spp. in sock samples (2013)

Detection and species identification of *Campylobacter* spp. (live cultures) in sock samples mixed with chicken litter and Cary Blair transport medium. The aim was to assess the performance of the NRLs to detect and species identify *Campylobacter* spp. in sock samples.



No of participating laboratories:

Mandatory/Voluntary:

No of samples:

Chicken litter used for proficiency test. Photo: Linda Svensson, EURL-Campylobacter/SVA



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Mandatory

12

29

Enumeration (and voluntary detection after enrichment) of *Campylobacter* spp. in minced meat (2014)

Enumeration (and voluntary detection after enrichment) of *Campylobacter* spp. (freeze dried bacteria) in minced meat. The aim was to assess the performance of the NRLs to detect and enumerate *Campylobacter* in minced meat. Species identification and detection of *Campylobacter* after enrichment were included as voluntary parts of PT 13.

Mandatory/Voluntary:	Mandatory (some parts voluntary)
No of samples:	10
No of participating laboratories:	35



Photo: Linda Svensson, EURL-Campylobacter/SVA

PT14

Detection and species identification of Campylobacter spp. in

milk filters (2014)

Detection and species identification of *Campylobacter* spp. (live cultures) in milk filters. The aim was to assess the performance of the NRLs to detect and species identify *Campylobacter* spp. in milk filters.

Mandatory/Voluntary:	Mandatory
No of samples:	18
No of participating laboratories	: 36





Detection and enumeration of Campylobacter spp. in chicken meat (2015)

Detection, species identification and enumeration of *Campylobacter* spp. (freeze dried bacteria) in chicken meat (breast fillets). The aim was to assess the performance of the NRLs to enumerate *Campylobacter* in chicken meat samples. Detection after enrichment and species identification of *Campylobacter* were included as a voluntary part of PT 15.

Mandatory/Voluntary:	Mandatory (some parts voluntary)	
No of samples:	10	
No of participating laboratories	: 36	

PT16

Detection and species identification of *Campylobacter* spp. in sock samples (2015)

Detection and species identification of *Campylobacter* spp. (live cultures) in sock samples mixed with chicken litter and Cary Blair transport medium. The aim was to assess the performance of the NRLs to detect and species identify *Campylobacter* in environmental samples taken in the vicinity of broiler houses.



Sock samples ready to be sent out to the NRLs.

Photo: Linda Svensson, EURL-Campylobacter/SVA

Mandatory/Voluntary:	Mandatory
No of samples:	18
No of participating laboratories	: 32





Detection and enumeration of Campylobacter spp. in broiler skin (2016)

Detection, species identification and enumeration of *Campylobacter* spp. (freezed dried bacteria) in broiler skin. The aim was to assess the performance of the NRLs to enumerate *Campylobacter* in broiler skin. Detection after enrichment and species identification of *Campylobacter* were included as a voluntary part of PT 17.

Mandatory/Voluntary:	Voluntary (some parts voluntary)
No of samples:	10
No of participating laboratories	: 34



Plastic container used for vials and broiler skin. Photo: Linda Svensson, EURL-Campylobacter/SVA

PT18

Detection and species identification of *Campylobacter* in chicken caecal contents (2016)

Detection and species identification of *Campylobacter* spp. (live cultures) in broiler caecum samples. The aim was to assess the performance of the NRLs to detect and species identify *Campylobacter* species in caecum samples taken at slaughter.

Mandatory/Voluntary:	Mandatory
No of samples:	18
No of participating laboratories.	: 33





Detection and enumeration of *Campylobacter* in minced chicken meat (2017)

Detection, species identification and enumeration of *Campylobacter* spp. (freezed dried bacteria) in minced chicken meat. The aim was to assess the performance of the NRLs to enumerate *Campylobacter* in minced meat. Detection after enrichment and species identification of *Campylobacter* were included as a voluntary part of PT 19.

Mandatory/Voluntary:	Mandatory (some parts voluntary)
No of samples:	10
No of participating laboratories	: 36

PT20

To detect and identify *Campylobacter spp*. in swab samples taken in transport crates (2017)

Detection and species identification of *Campylobacter* spp. (live cultures) in swab samples mixed with chicken caeca, Buffered Peptone Water (BPW) and Cary Blair transport medium. The aim was to assess the performance of the NRLs to detect and species identify *Campylobacter* species in samples taken in transport crate (transporting chicken to the slaughter house).

Mandatory/Voluntary:	Mandatory
No of samples:	18
No of participating laboratories.	34



Cutting of chicken caeca into stomacher bag

Photo: Linda Svensson, EURL-Campylobacter/SVA



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Enumeration (and voluntary detection and species identification) of *Campylobacter* in chicken skin (2018)

Enumeration of *Campylobacter* spp. (freezed dried bacteria) in chicken skin. The aim was to assess the performance of the NRLs to enumerate *Campylobacter* in chicken skin. As voluntary parts of PT21, Detection after enrichment and species identification of *Campylobacter* were included.

Mandatory/Voluntary:	Mandatory (some parts voluntary)	
No of samples:	10	
No of participating laboratories	: 37	

PT22

Detection and species identification of *Campylobacter* spp. in chicken feacal swab samples with voluntary educational samples (2018)

Detection and species identification of *Campylobacter* spp. (freezed dried bacteria) in faecal swabs containing Cary Blair transport medium mixed with chicken caeca. The aim was to assess the performance of the NRLs to detect and identify *Campylobacter* species from faecal swabs.

Additional four samples were sent to the NRLs as voluntary educational samples.



Faecal swabs with caecal material and Cary Blair Photo: Therese Jernberg, EURL-Campylobacter/SVA

Mandatory/Voluntary:	Voluntary
No of core samples:	18
No of participating laboratories:	31
V	Voluntary
No of educational samples:	4
No of participating laboratories:	.31



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