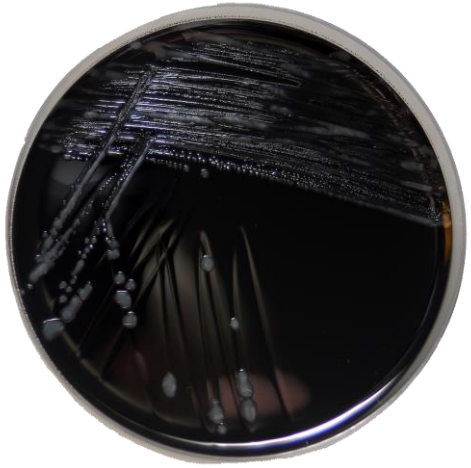
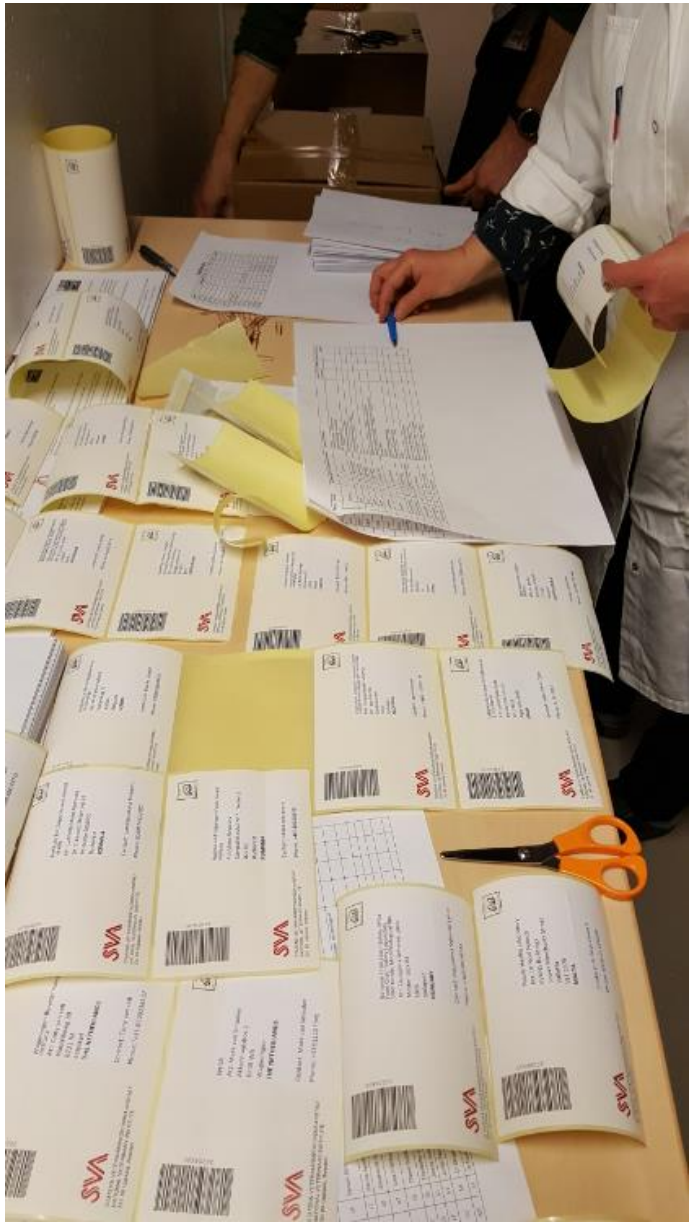
RESULTS OF PROFICIENCY TESTS NO. 26 AND 27

Helena Höök

EURL-*Campylobacter*

Workshop 2020



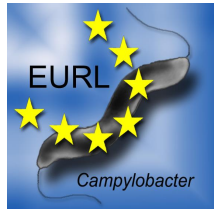


Thank you for your participation and for providing information in the questback reports!

EURL Campylobacter

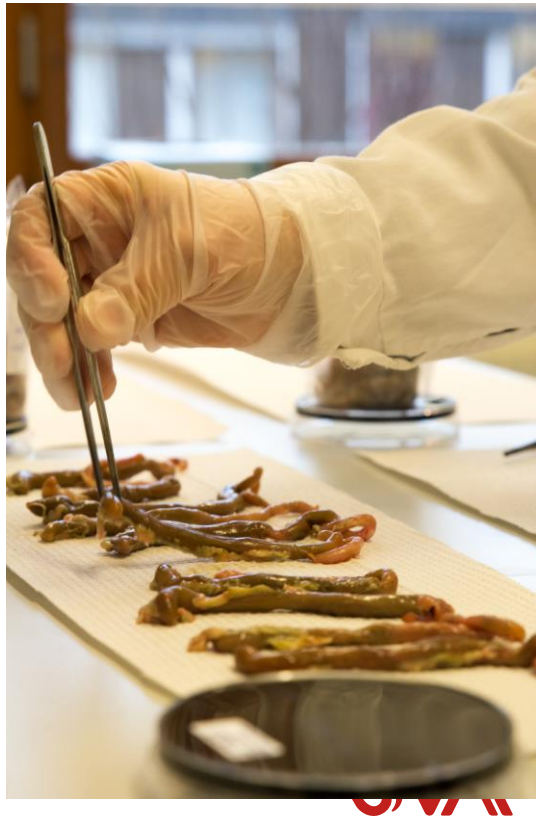
NUMBERS OF PARTICIPANTS

Year	2020	2019	2018	2017	2016	2015	2014	2013	2012
	PT 26	PT 23	PT 21	PT 19	PT 17	PT 15	PT 13	PT 11	PT 9
Enumeration	**33**/38	35	37	36	36	36	35	36	33
	PT 27	PT 24	PT 22	PT 20	PT 18	PT 16	PT 14	PT 12	PT 9
Detection & species id	**29**/31	33	31	34	33	32	36	34	36

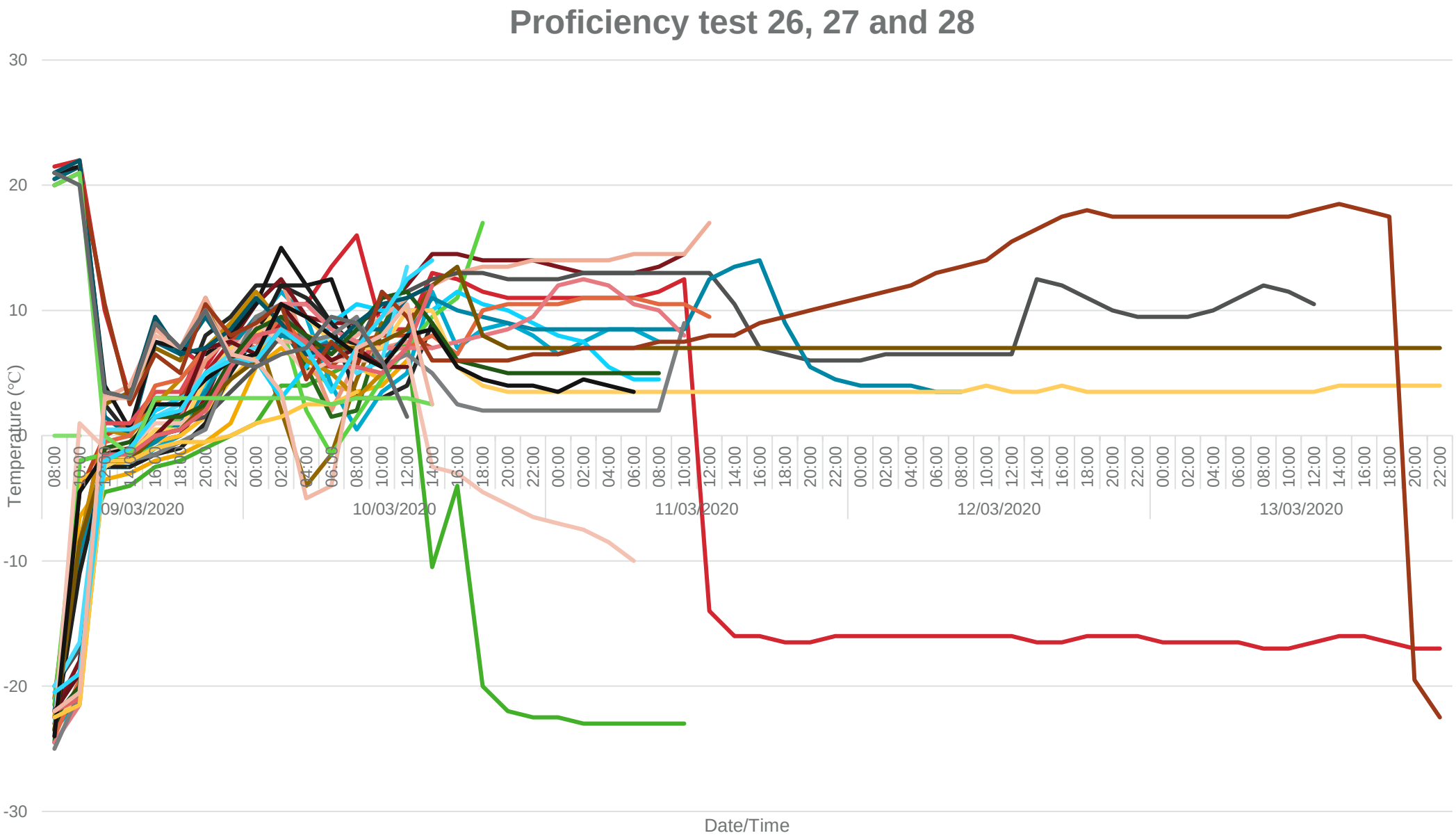


CAMPYLOBACTER-FREE MATRICES

- Chicken skin (PT 26)
- Caecal material (PT 27)
- All from a producer with no *Campylobacter*-positive broiler flocks for several months, and a slaughterhouse with very low level of *Campylobacter-positive* flocks
- Chicken skin and ceacal material tested negative for presence of *Campylobacter*



TEMPERATURE DURING TRANSPORT



PT 26 – ENUMERATION (AND SPECIES IDENTIFICATION) IN CHICKEN SKIN

PROFICIENCY TEST NO. 26

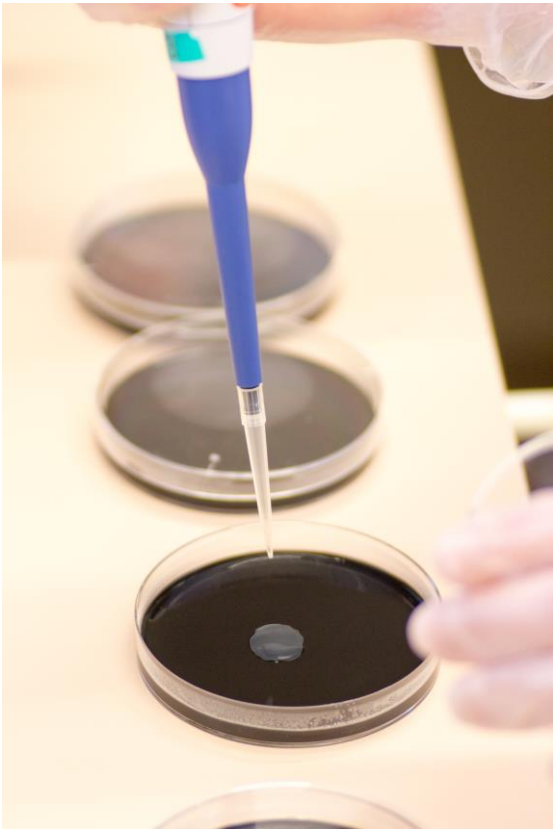
The objective was to assess the performance of the NRLs to enumerate (and voluntary species identify) *Campylobacter* in chicken skin.

- Enumeration and confirmation of *Campylobacter* spp. in chicken skin
- Species identification of *Campylobacter* (voluntary)
- Recommended method ISO 10272-2:2017, but other methods allowed
- Should allow enumeration of between 10 and 10^5 cfu *Campylobacter*/g chicken skin

PT 26: CONTENTS AND PROCEDURE

- Chicken skin (about 120 g) to be divided into 10 portions of 10 g
- 10 vials with freeze-dried sample (with or without *Campylobacter*)
- Homogenise and make a initial dilution of 10^{-1}
- Follow the method(s) of choice for
  - enumeration
  - species identification (voluntary)

of *Campylobacter* spp.



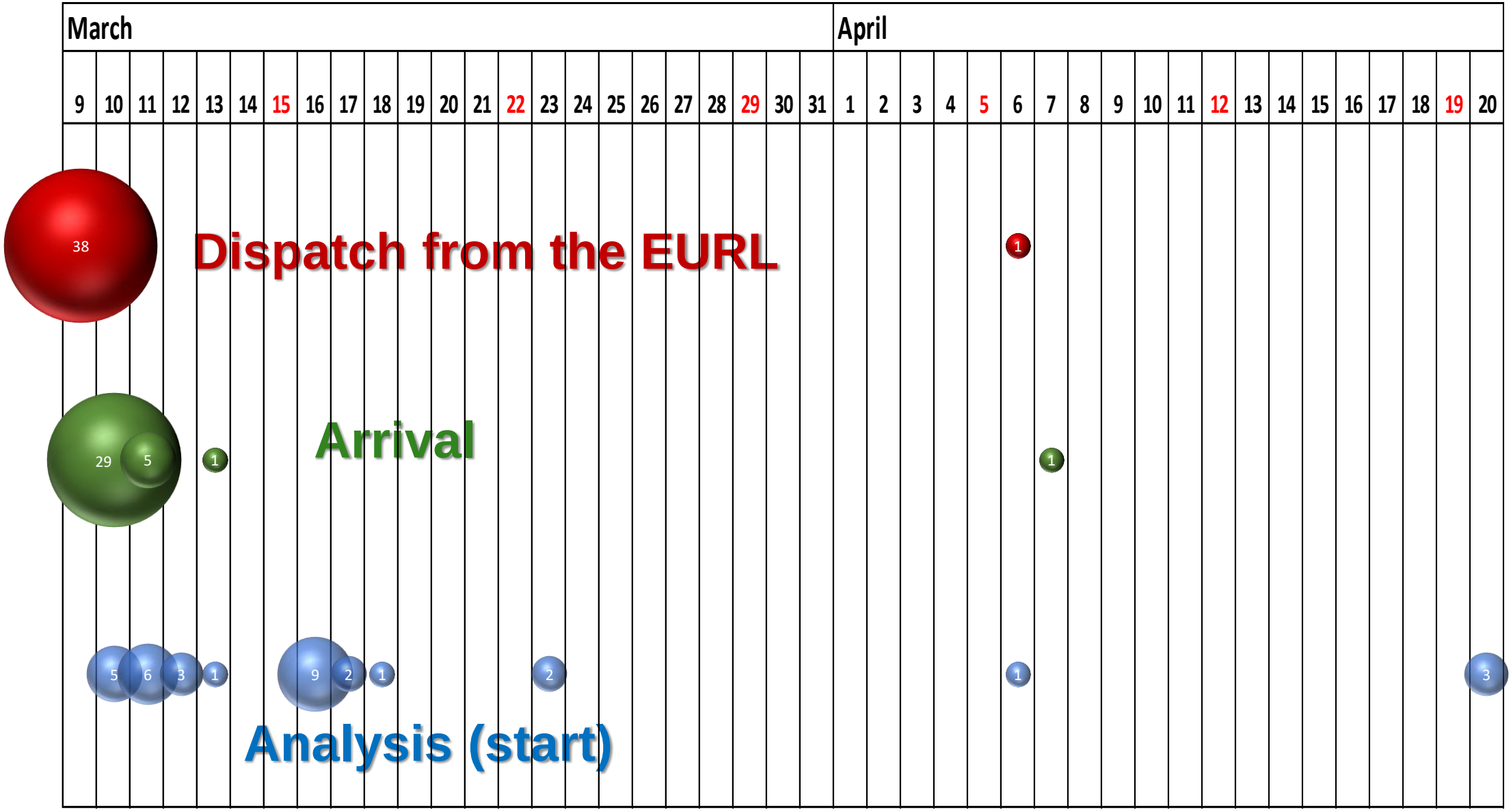
PT 26: QUALITY CONTROL

- Vials produced by the National Food Agency or the EURL
- Vials tested for homogeneity and stability by the producer and in triplicates by EURL
- Enumerations with chicken skin for control of *Campylobacter* levels and homogeneity
- Tested four times, once before and three times after dispatch (last time at last day for start of analysis)

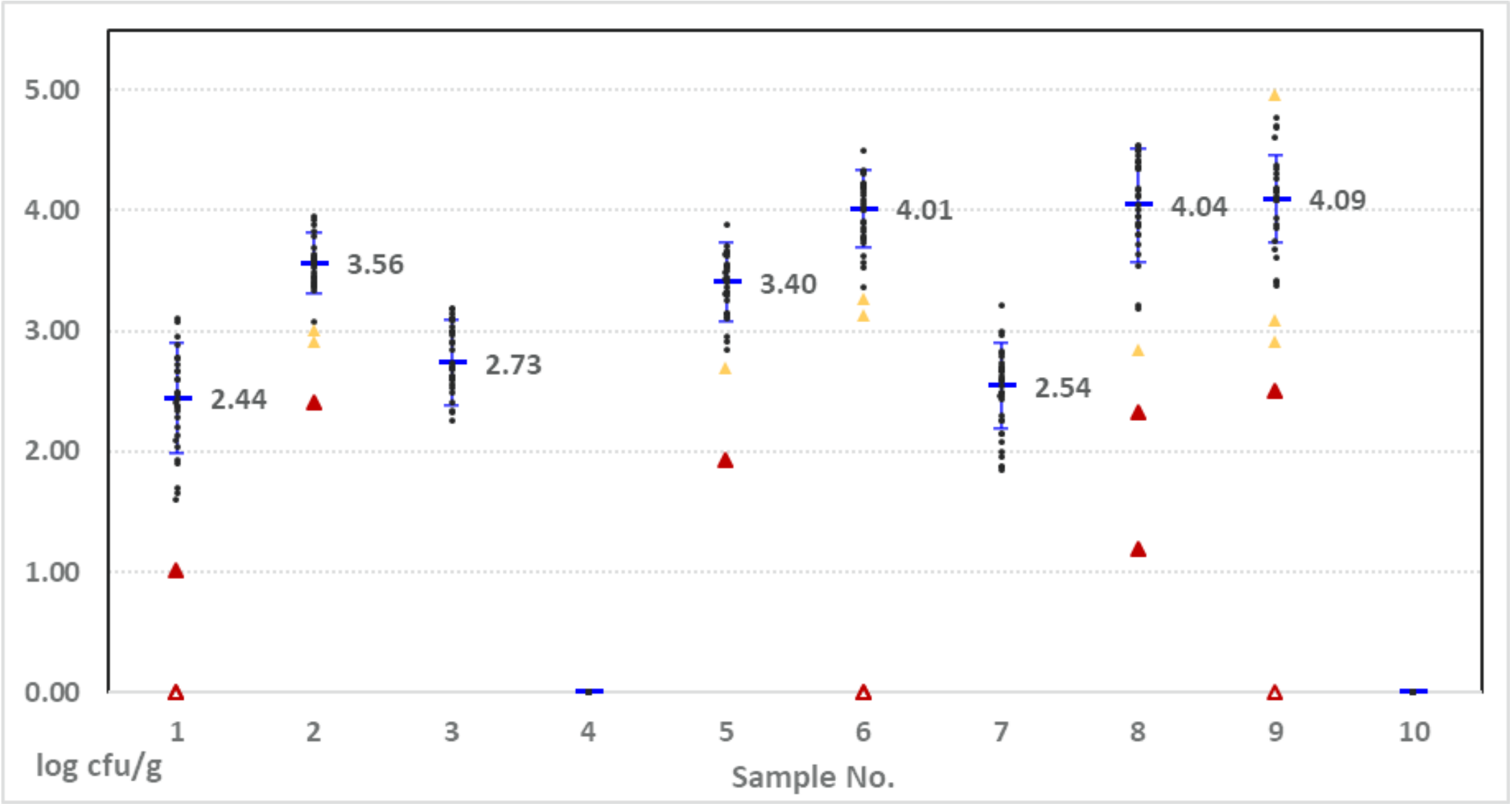
DESCRIPTION OF THE 10 VIALS IN PT 26

Sample No.	Species	Level (log cfu/vial)		Batch No.
1	***C. coli***	3.90		SLV334
2	***C. jejuni***	4.82		SLV305
3	***C. jejuni*** + *Escherichia coli*	3.53	4.00	SLV313
4	*Escherichia coli*		3.41	SLV159
5	***C. jejuni***	4.60		SLV336
6	***C. lari***	5.10		SLV335
7	***C. jejuni***	3.71		SLV306
8	***C. coli***	5.50		SLV333
9	***C. jejuni***	6.12		SVA038
10	Negative	5.67		SLV287/SLV272

PT 26: TIME TO ARRIVAL & START OF ANALYSIS



PT 26: RESULTS OF ENUMERATION



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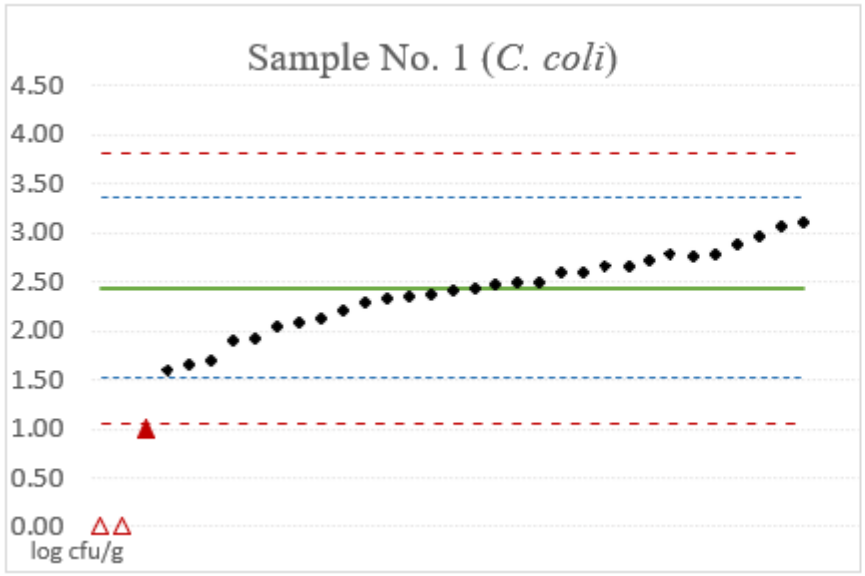

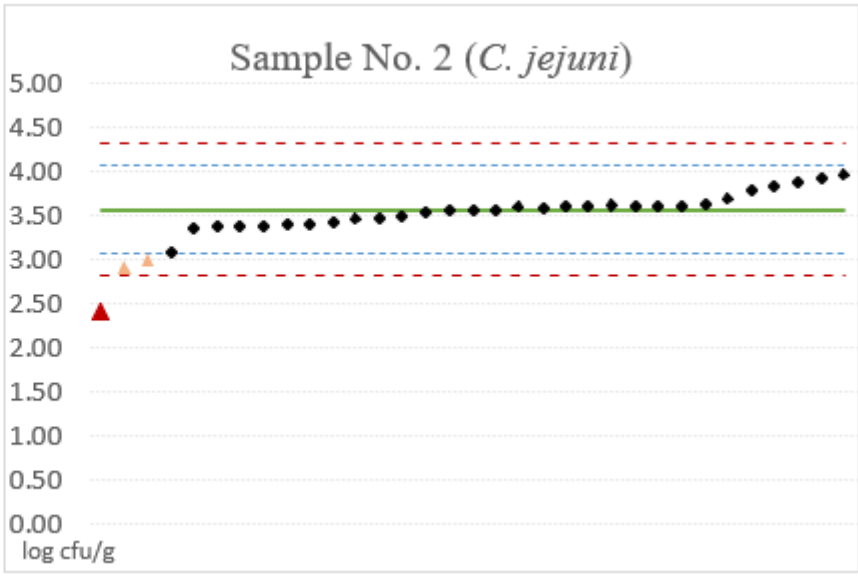

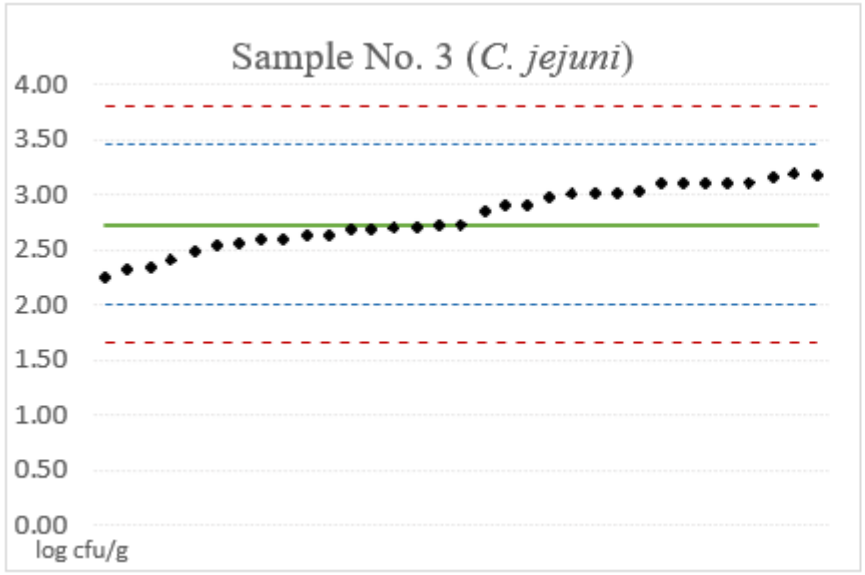

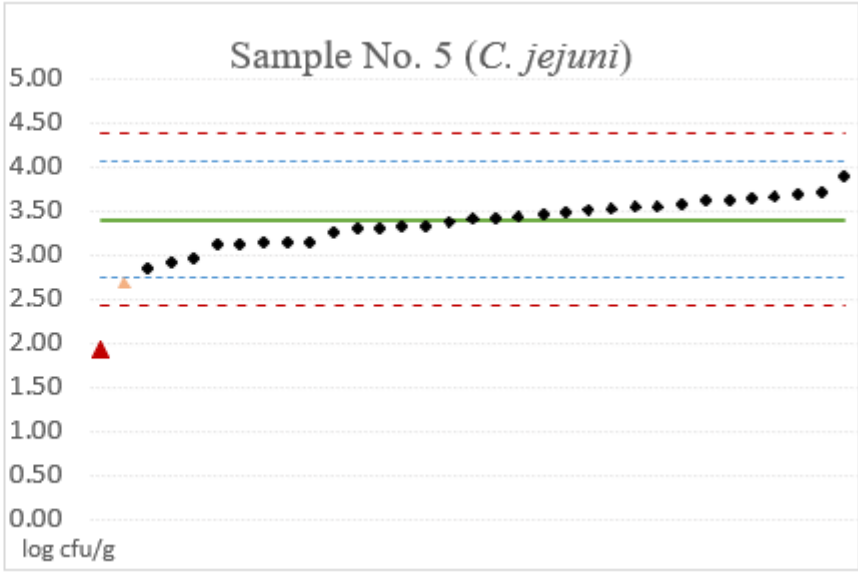

EURL Campylobacter

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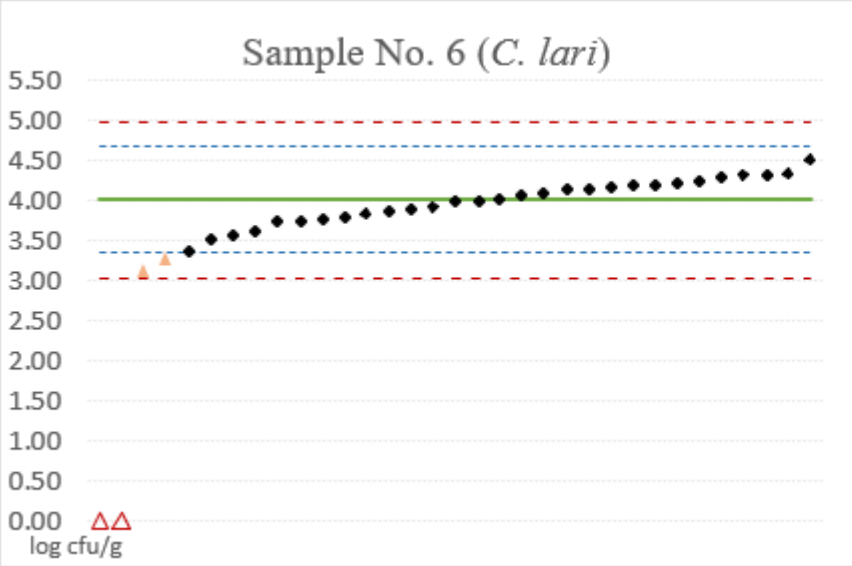
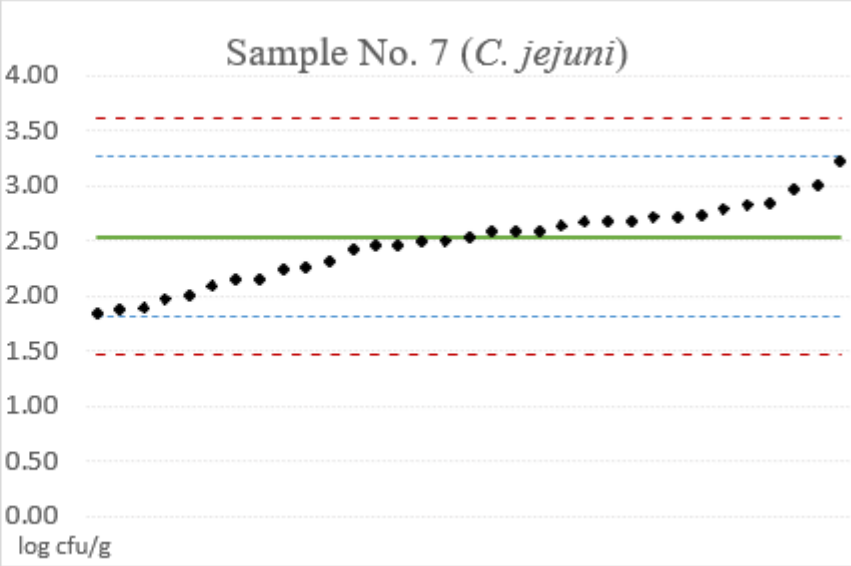
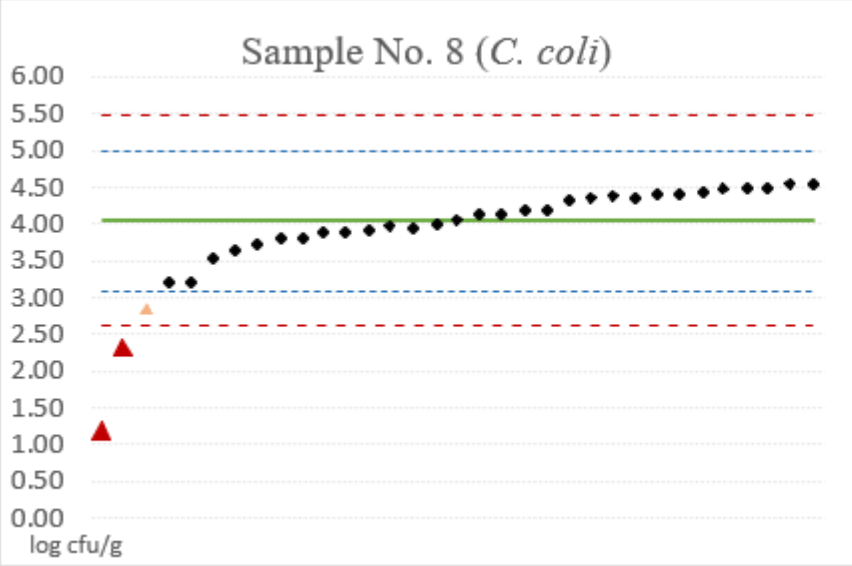
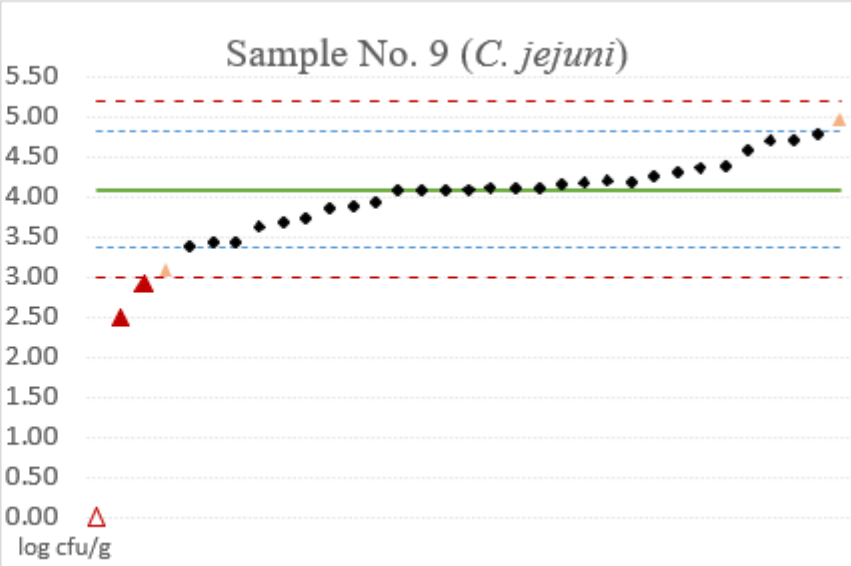






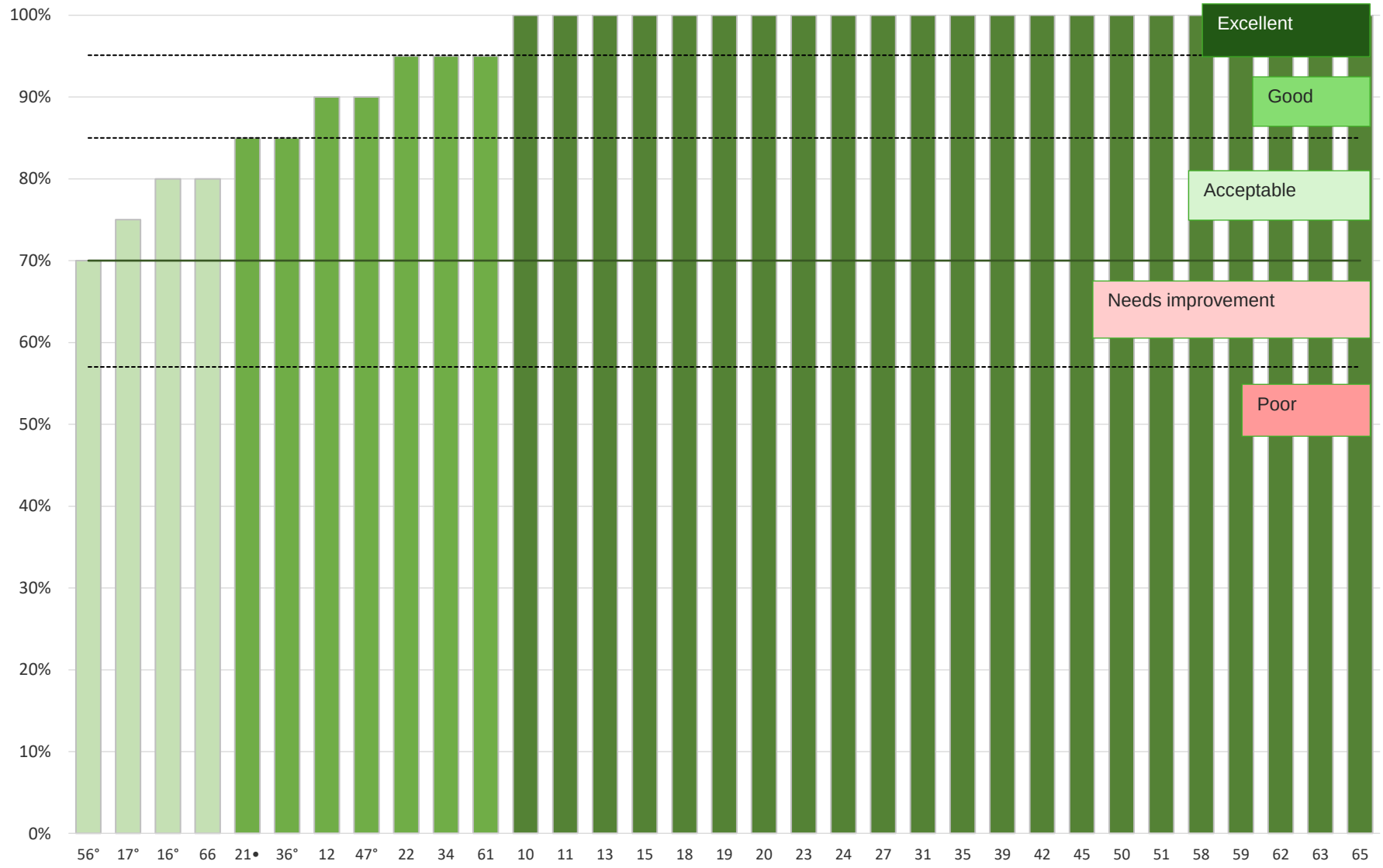


HOW WAS PERFORMANCE CALCULATED?

- The Median Absolute Deviation (MADe) to calculate performance
- σMADe = MADe × 1.4826
- *Campylobacter*-containing samples
  - Results within participants' median ±2σMADe = 2 points
  - Results between ±2σMADe and ±3σMADe = 1 point
  - Results outside ±3σMADe = 0 points
- *Campylobacter*-negative samples
  - No *Campylobacter* reported = 2 points
  - False positive result = 0 points
- The maximum score (2 points for each sample) was 20 points
- Calculate the score for each participant

Grade	Scoring limits	
Excellent	20	95.1–100%
Good	17–19	85.0–95.0%
Acceptable	14–16	70.0–84.9%
Needs improvement	12–13	57.0–69.9%
Poor	<12	<57.0%

PERFORMANCE PT 26



PT 26: SPECIES IDENTIFICATION (VOLUNTARY)

Content of sample (vial)	*C. jejuni*	*C. coli*	*C. lari*	*Camp* spp.	Other / No growth
1. ***C. coli***		**25**			1
2. ***C. jejuni***	**26**				
3. ***C. jejuni*** + *E. coli*	**26**				
4. *E. coli*					**26**
5. ***C. jejuni***	**26**				
6. ***C. lari***			**24**	1	1
7. ***C. jejuni***	**26**				
8. ***C. coli***	1	? **25**			
9. ***C. jejuni***	**24**	1			1
10. Negative					**26**

PT 27 – DETECTION AND SPECIES IDENTIFICATION OF *CAMPYLOBACTER*

EURL Campylobacter

PROFICIENCY TEST NO. 27

The objective was to assess the performance of the NRLs to detect and identify *Campylobacter* species in chicken caecal contents.

- Detection of *Campylobacter* spp. in chicken caecal contents
- Species identification of *Campylobacter*
- 10 samples mimicking a sample pooled from up to 30 chicken caeca
- Recommended method ISO 10272-1:2017, but other methods allowed
- A direct procedure (procedure C in the ISO method) was recommended for the analysis

PT 27: CONTENTS AND PROCEDURE: CHICKEN CAECAL CONTENTS

- 10 numbered plastic tubes with 6 ml of caecal material
- 10 freeze-dried vials (with or without *Campylobacter* and/or other bacteria)
- Mix each vial with the corresponding caecal content tube up to a total volume of 10 ml
- Follow the method(s) of choice for
  - detection
  - species identification

} of *Campylobacter* spp.

DESCRIPTION OF THE 10 VIALS IN PT 27

Sample No.	Content in vial	Batch No.	Level	log cfu/vial
11	***C. jejuni***	SVA021	Low	4.28
12	Negative			
13	*E. coli*	SVA045		
14	***C. jejuni*** + *E. coli*	SVA041	Low	4.78
15	***C. jejuni***	SVA036	High	5.52
16	***C. coli***	SVA033	Low	4.85
17	***C. lari***	SVA044	Low	4.77
18	***C. coli*** + *E. coli*	SVA043	High	5.39
19	***C. jejuni***	SVA039	Low	4.79
20	***C. coli***	SVA037	Low	4.32

EURL Campylobacter

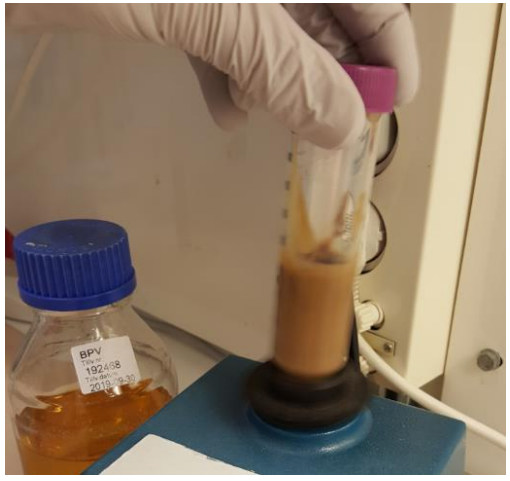
PT 27: QUALITY CONTROL

- Vials produced by EURL or the National Food Agency (negatives)
- Tested for homogeneity and stability by the producer

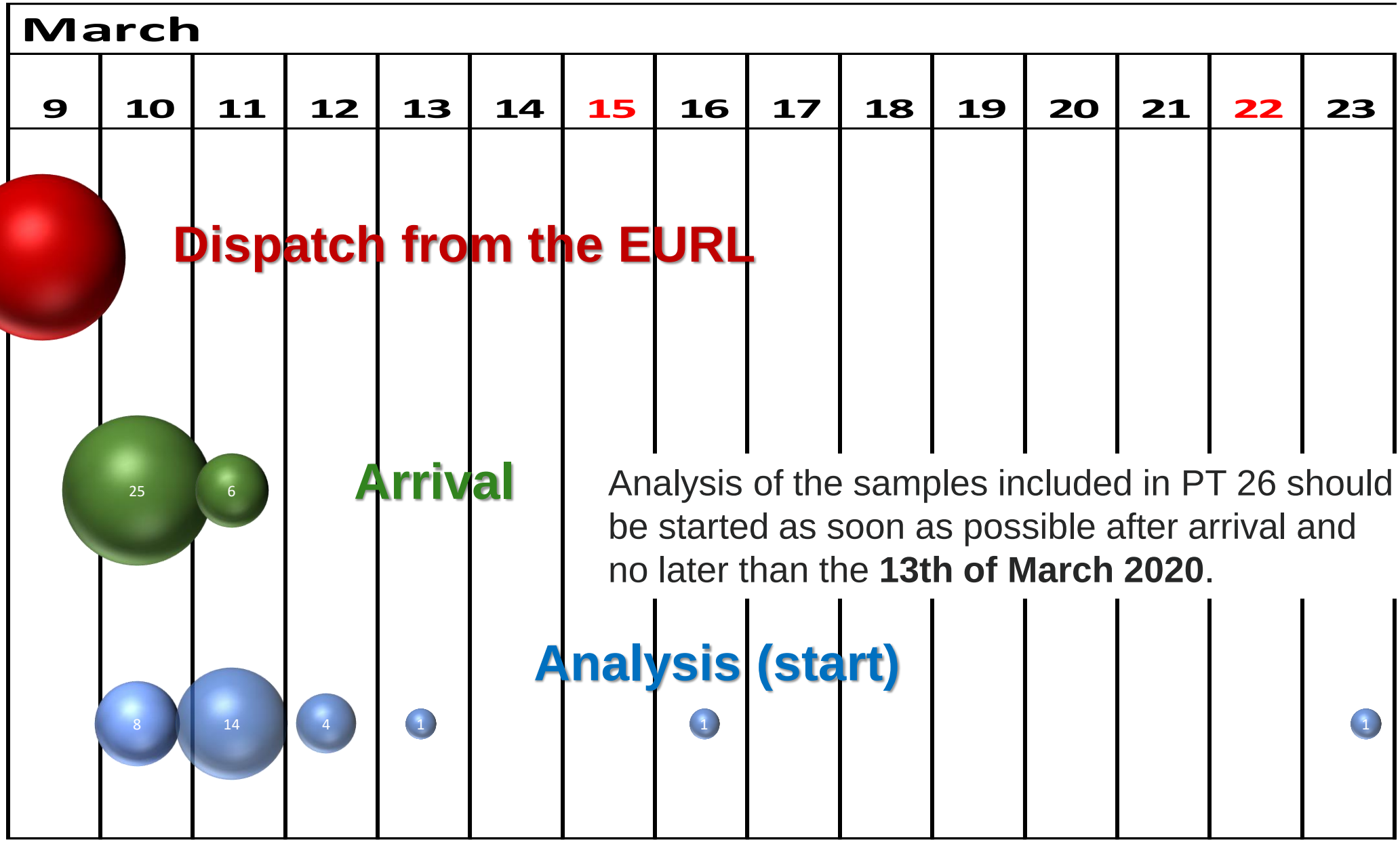
- Vials together with matrix were analysed according to ISO 10272-1:2017, procedure C (direct plating)
- Tested three times, once before and twice after dispatch

PT 27: PREPARATION OF THE MATRIX: CAECAL CONTENT

- *Campylobacter*-free caeca were cut and placed in a stomacher bag and mixed with Buffered Pepton Water.
- 6 ml of the suspension were added to a plastic tube, one for each sample
- The samples were freeze-stored three days.

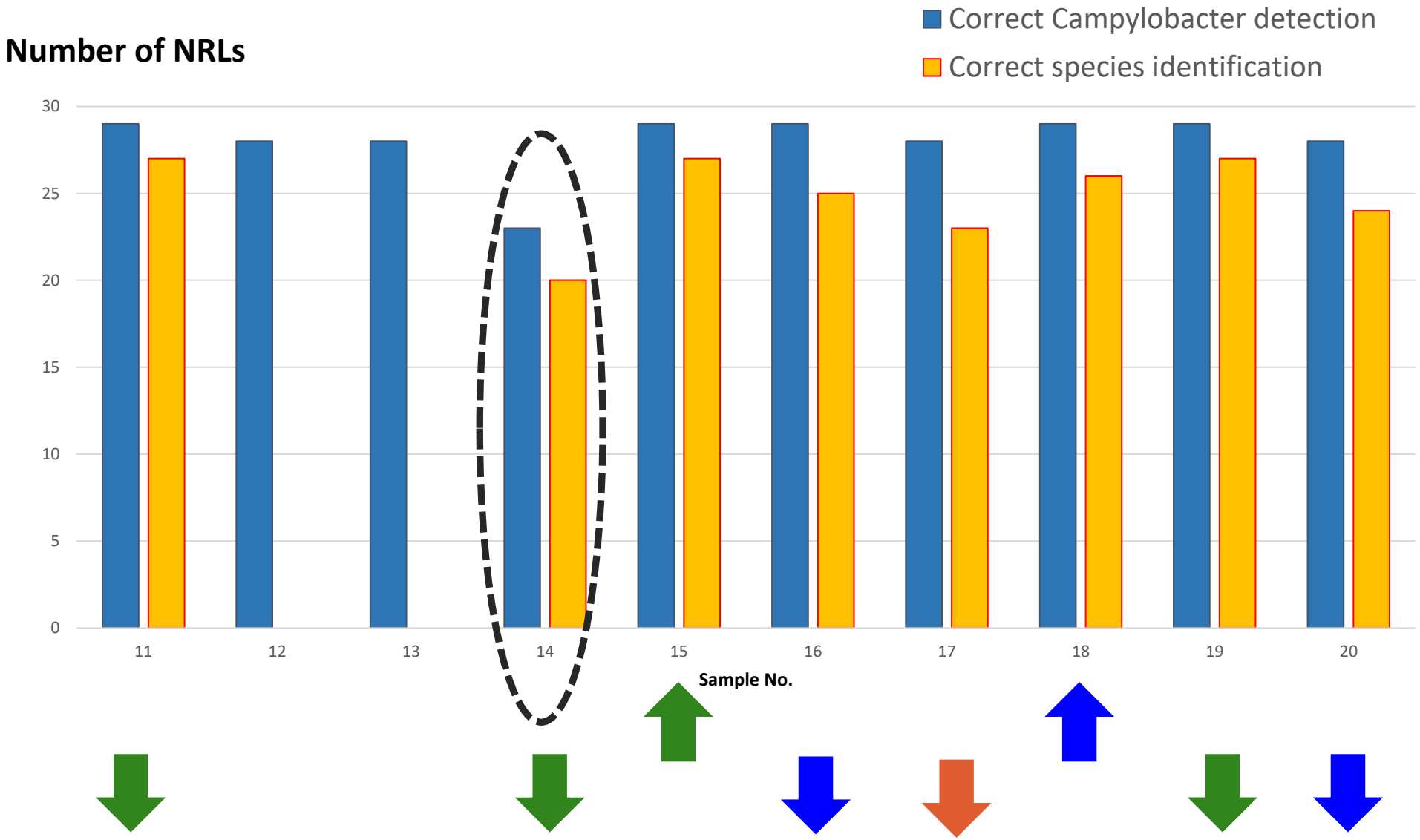


PT 27: TIME TO ARRIVAL & START OF ANALYSIS

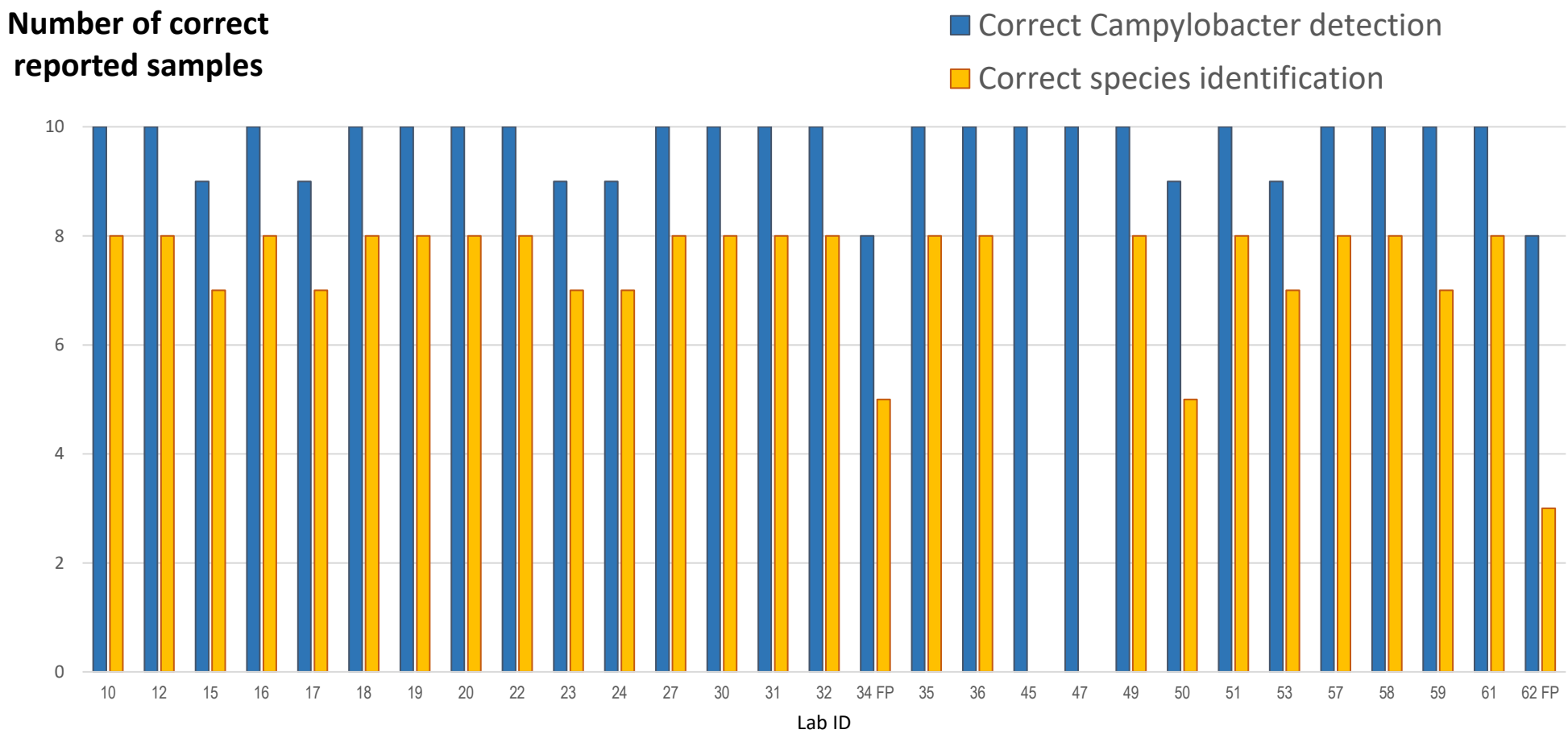


Analysis of the samples included in PT 26 should be started as soon as possible after arrival and no later than the **13th of March 2020**.

PT 27: CORRECT REPORTED RESULTS PER SAMPLE

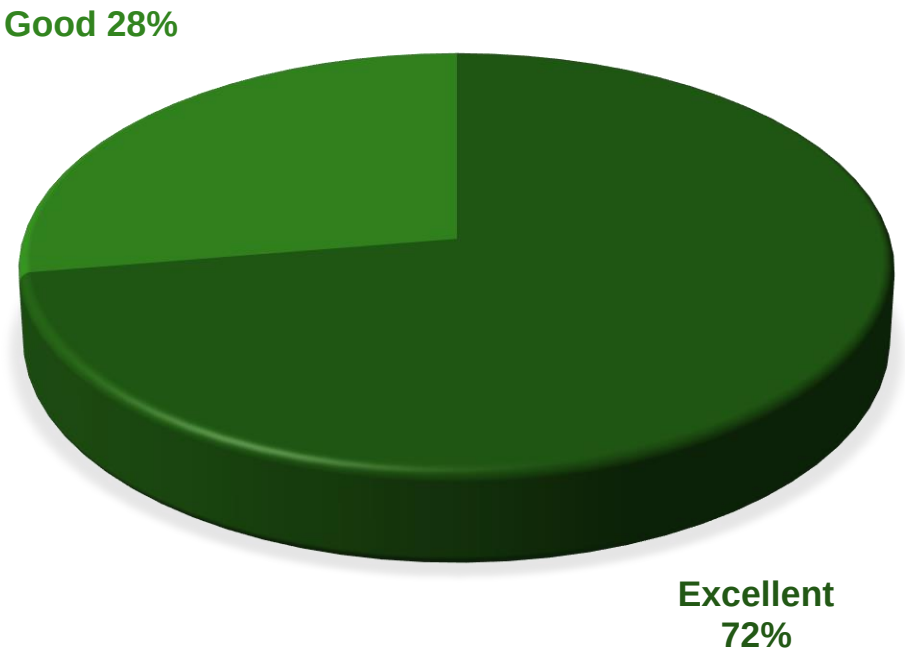


PT 27: CORRECT REPORTED RESULTS PER LAB

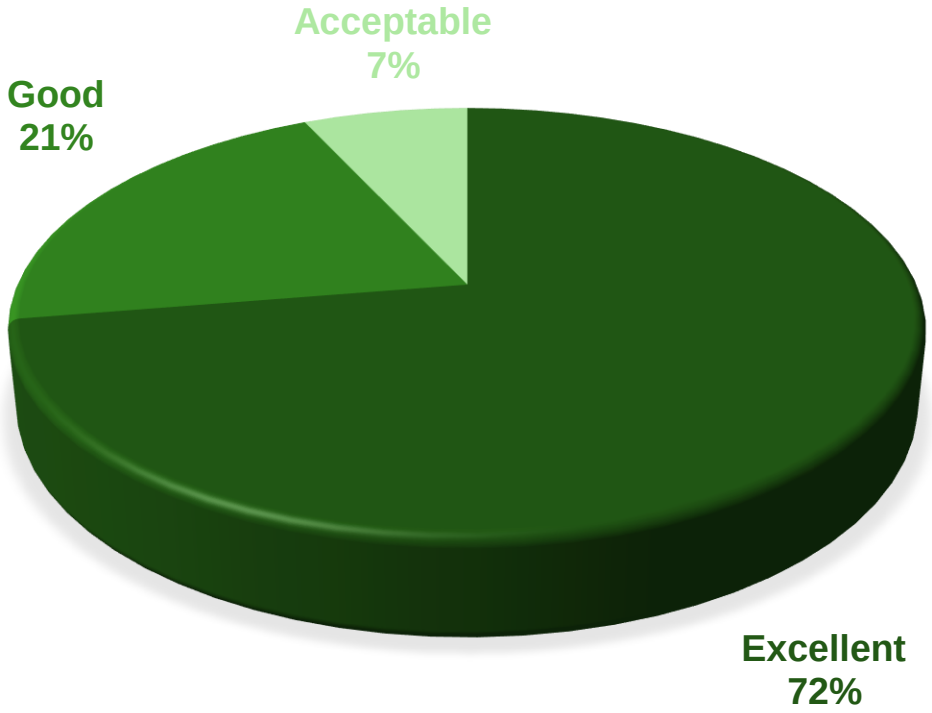


PT 27: PERFORMANCE – SENSITIVITY AND ACCURACY IN DETECTION OF *CAMPYLOBACTER*

SENSITIVITY IN DETECTION

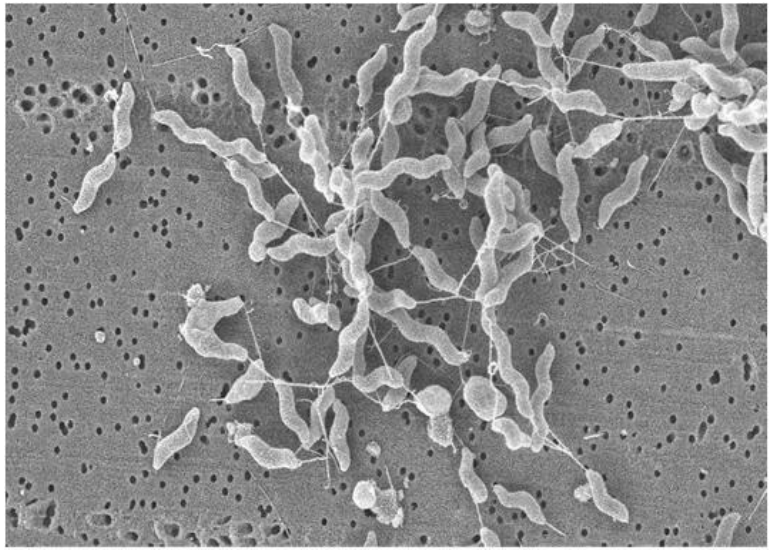


ACCURACY IN DETECTION OF POSITIVE AND NEGATIVE SAMPLES



PT 27: REPORTED SPECIES IDENTIFICATION

Sample No.	Bacterial species	*C. jejuni*	*C. coli*	*C. lari*	*Campylobacter* spp. but unable to identify species	Growth of other, not *Campylobacter*	No growth at all
11	***C. jejuni***	27					
12	Negative	1				12	14
13	*E. coli*				1	24	2
14	***C. jejuni*** + *E. coli*	20			1	6	
15	***C. jejuni***	27					
16	***C. coli***		25	2			
17	***C. lari***	1	2	23			1
18	***C. coli*** + *E. coli*		26		1		
19	***C. jejuni***	27					
20	***C. coli***	1	24	1			1



THANK YOU!

QUESTIONS?