Survey - Inventory for the EURL- *Campylobacter* Workshop 2022

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Co-funded by the European Union

Survey questionnaire

Invitation to all participants registered

(50)

Received

30 responses

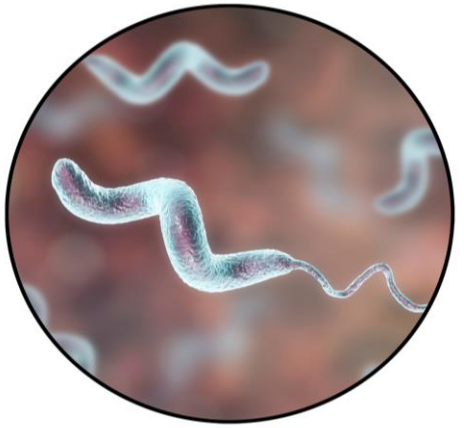
from 29 NRLs

28 MS-NRLs

Survey started 26th of August

Survey ended 16th of September

Two topics



ISO 10272:2017

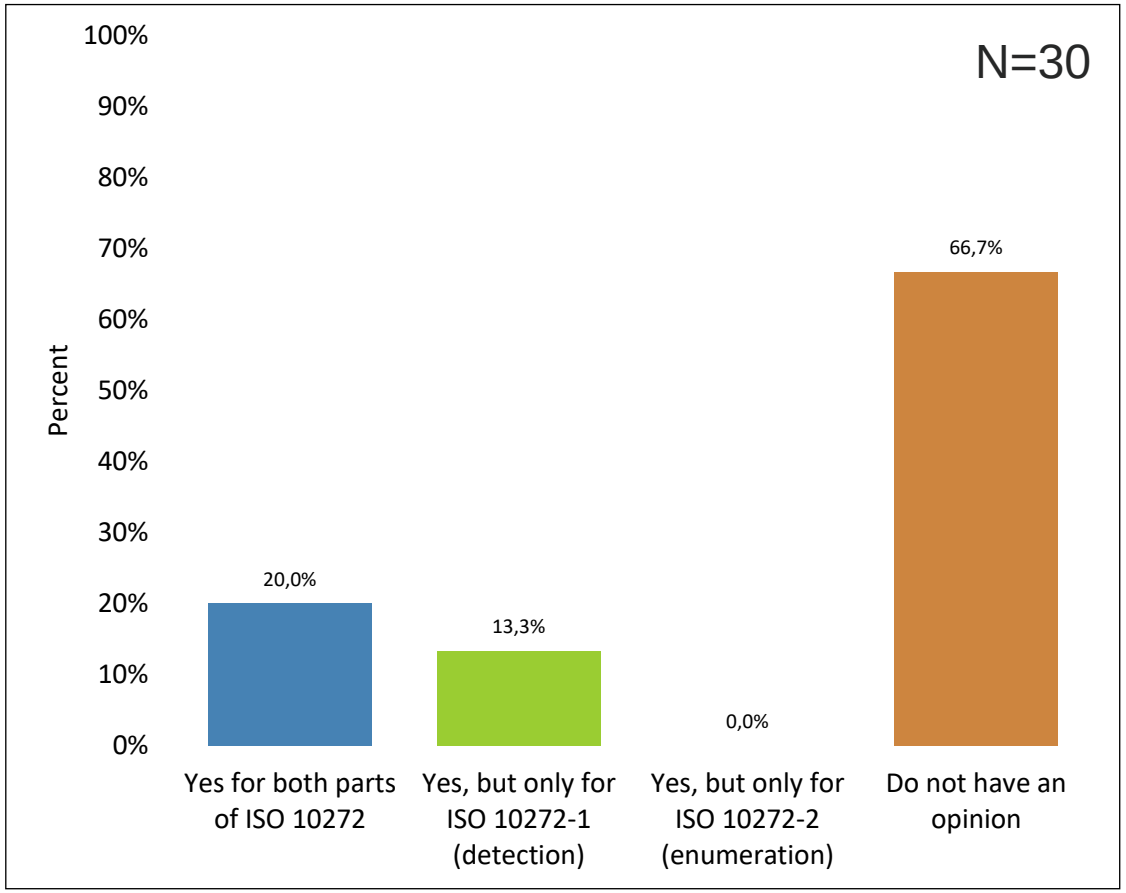
Questions in the survey for this topic were to collect information and opinions for the benefit of CEN/TC 463 Working Group 3 ‘*Campylobacter*’.

Activities for the EURL-*Campylobacter* work programme 2023-2024

To collect information on needs related to analytical methods, PTs, training courses and opinions for upcoming workshops

Topic: **ISO 10272:2017**

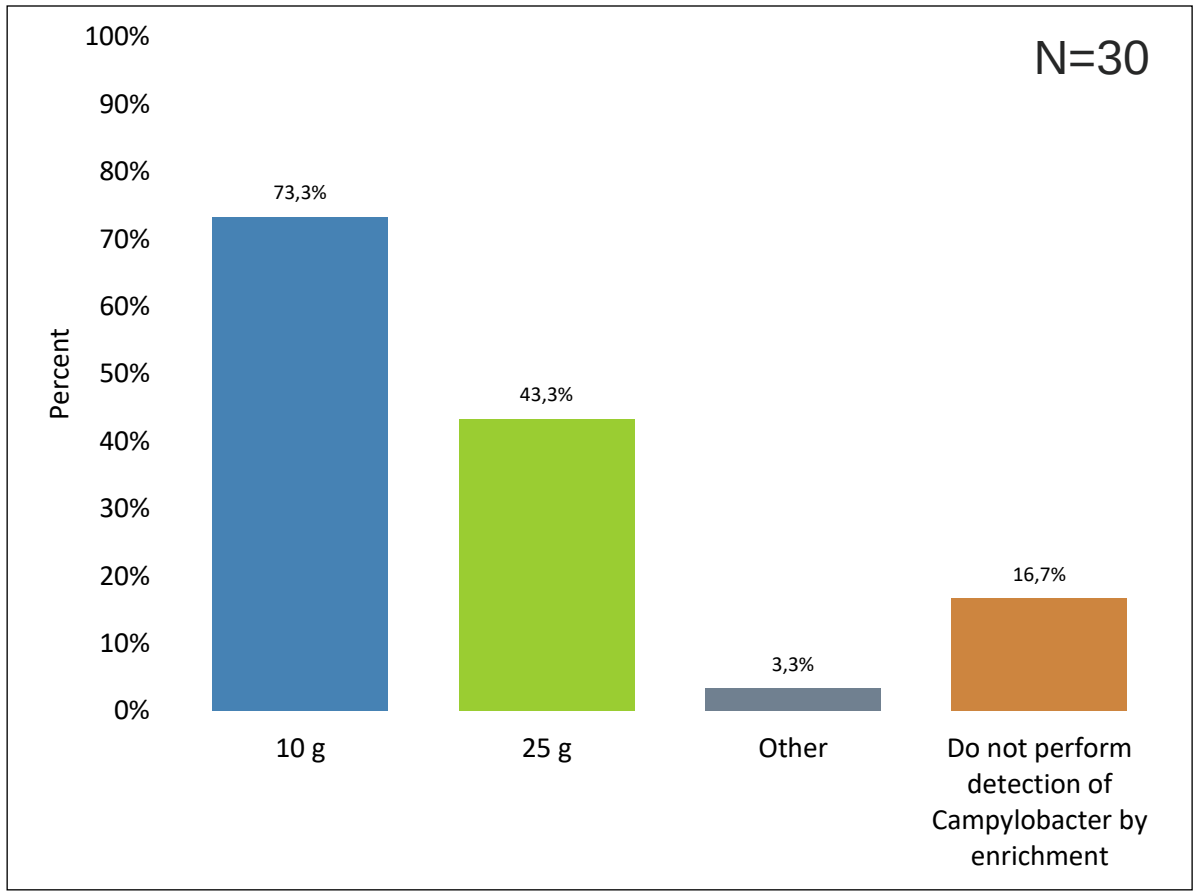
Question 1: Every five years, the need for a review of an ISO standard is assessed. ISO 10272 was reviewed 2017 and the need is currently assessed through an ISO ballot. In your opinion, is a full revision of the standard needed?



Question 1: If yes, please motivate your response

- Alternative or second selective media to mCCDA for enumeration
- Alternative or second selective media for detection
- Alternative selective broth for samples with low levels of *Campylobacter* and high levels of background flora
- Further research work on enrichment media is needed to improve detection, especially for *C. coli*
- Flexibility for time and period of incubation (e. g. 37 - 41,5 ºC, 48 - 72h)

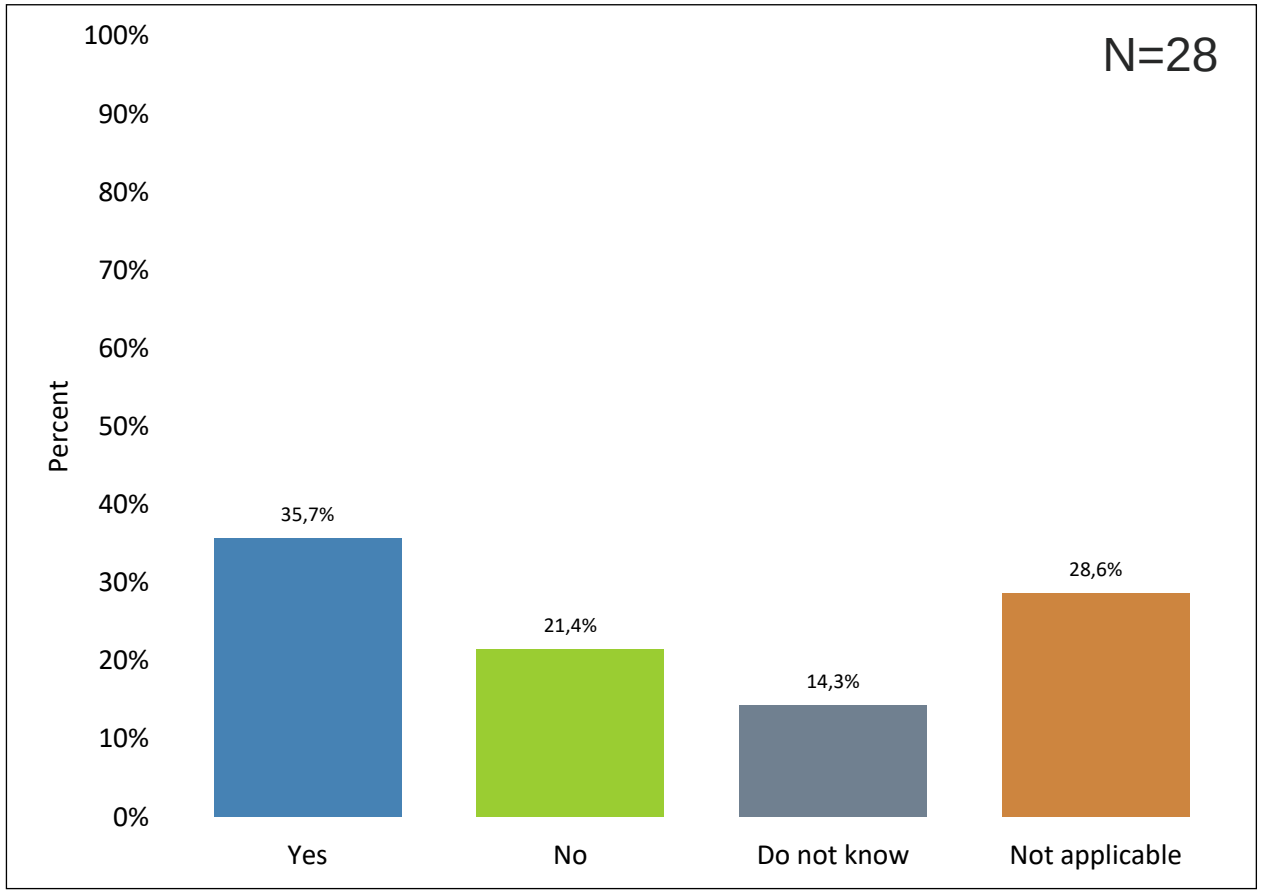
Question 2: What is the size of the test portion used at your laboratory for detection of *Campylobacter* by enrichment?



If other, please specify size of test portion:

Faeces – 1 g

Question 3: Whould you use a different size of test portion than 10 g if there were validation data supporting this in ISO 10272. If yes, please specify in which curcumstanses or for what type of matrices.



Question 3: If yes, please specify in which circumstances or for what type of matrices.

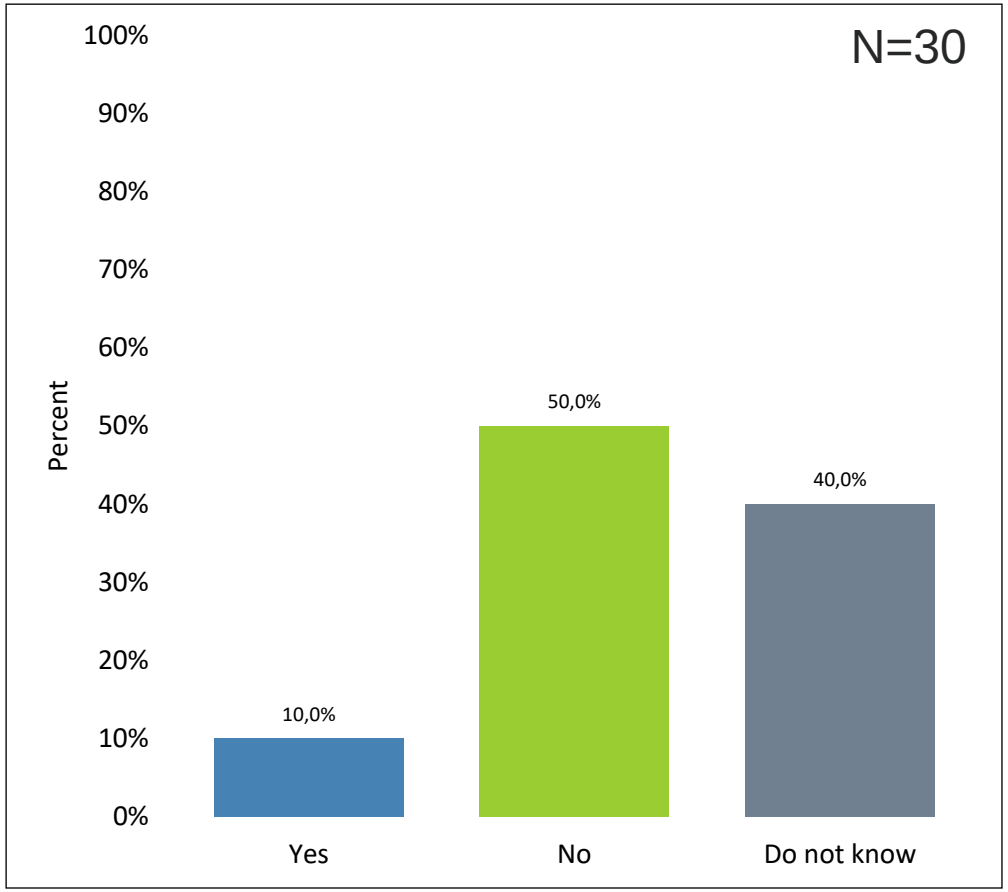
- milk and cheese – 25 g
- Swabs (surface samples)
- Raw milk
- Samples with low levels of *Campylobacter* (aligned with other standards)
- Depends what the competent authority decides for official control (10 g or 25 g)

Question 4: Do you experience problems reaching the expected productivity ratio ($P_R \geq 0,5$) for performance testing of mCCDA with blood agar as the reference medium (according to ISO 11133)?

Total count of colonies mCCDA

$$P_R = \frac{N_S}{N_O}$$

Total count of colonies blood agar



Question 4: comments

- Ready-to-use mCCDA (tested by the manufacturer) → do not perform productivity test
- Productivity of home-made versus ready-to-use mCCDA → home-made media better, although in some cases the expected productivity ratio couldn't be reached.
- The quality of ready-to-use mCCDA from different manufacturers vary. e.g Oxoids is more selective than Mercks. Although *C. jejuni* grows better on Mercks than on Oxoids.

Topic: Activities for the EURL-*Campylobacter* work programme 2023-2024

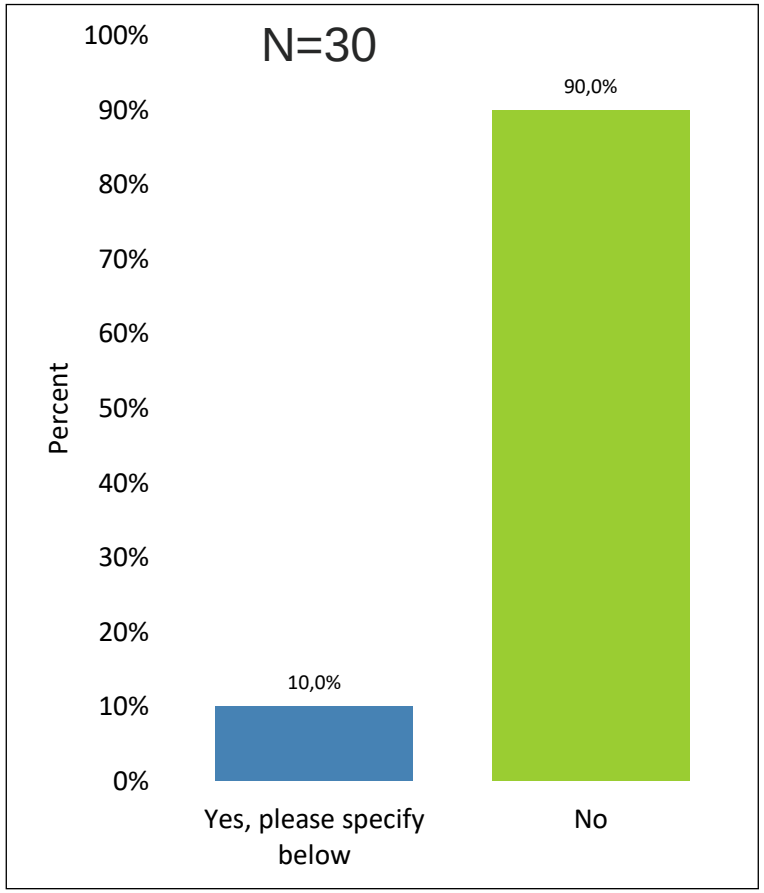
Question 1: Are there any specific studies comparing or validating analytical methods for detection, identification or characterisation of *Campylobacter* that would be particularly useful to you? Please describe.

- Molecular methods for detection and identification of:
  - *Campylobacter* in general
  - Unusual *Campylobacter* species
  - *Campylobacter* from different matrices (stools, wild animals, waste waters)
- Using MALDI-TOF MS for confirmation and identification
- Butzler agar compared with mCCDA
- Cluster analysis of WGS data

Question 2: Are there any specific analytical methods for which you need more guidance? Please specify

- Direct detection versus enrichment methods
- Enumeration of *Campylobacter*
- Real-time PCR for absolute quantification
- PCR methods for detection of *Campylobacter*
  - -in dairy samples after 24-48h storage
  - -in surface swabs (to check packaging)

Question 3:
As before, we plan to annually provide a PT for enumeration (mandatory), and a PT for detection (mandatory PT with 18 samples in years a third PT is not offered, otherwise voluntary and with 10 samples). The PT for enumeration will include a matrix related to food, and the PT for detection a matrix related to primary production. Do you have any general comments on the outline of these PTs on enumeration and detection?



Comments:
Sufficient amount of matrix to be able to perfom enrichment in both Bolton and Preston broth and also for direct method

If the frequency of PT´s can be reduced, or alternate the mandatory PT tests - one year enumeration, second year detection

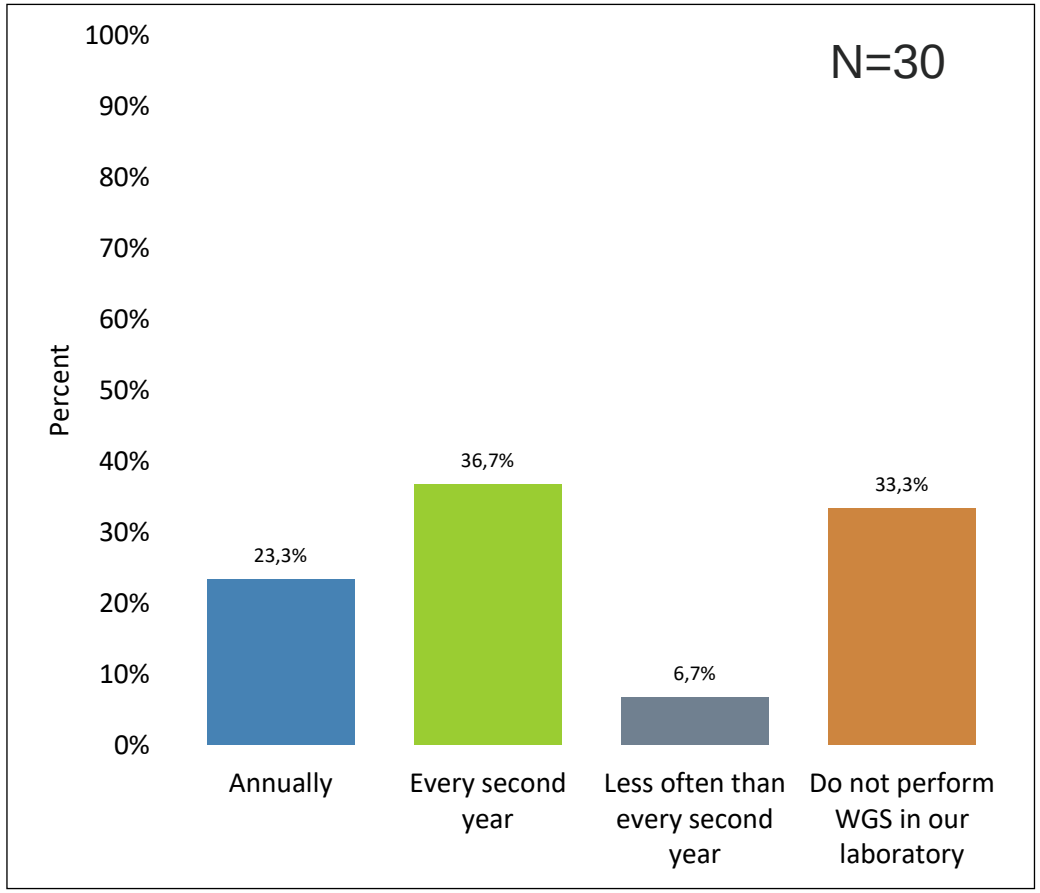
Question 4: Regarding the matrix in the PT for enumeration; every second year we aim to provide a PT for enumeration with chicken skin (due to the PHC). Which other matrices would be particularly useful for your laboratory?

- Milk, milk products
- Broiler feaces/caeca
- Meat (poultry, pork, beef)
- Fish products
- Water
- Vegetables

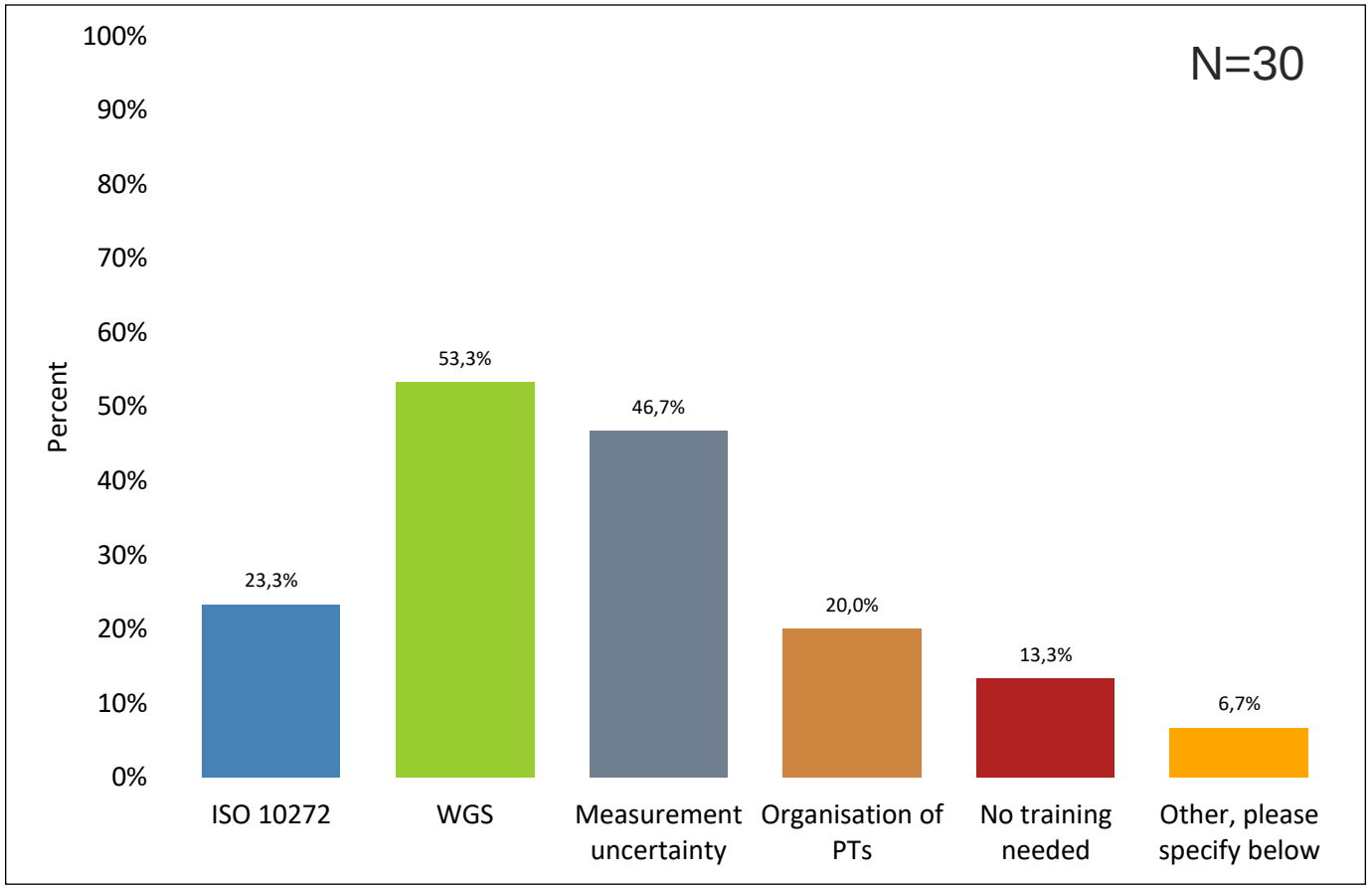
Question 5: Regarding the matrix in the PT for detection; which matrices would be particularly useful for your laboratory?

- Feaces (chicken, bovine)
- Caeca (broiler)
- Meat (poultry, pork, beef)
- Milk and milk products
- Farm environments (swabs or boot socks)
- Swab samples of carcasses
- Seafood
- Waste water

Question 6:
We have now organised three PTs for whole genome sequencing (WGS) and would like to know how often it is useful for your laboratory to participate in PTs for WGS-based methods (sequence quality and/or cluster detection) for *Campylobacter*? There are now several providers of PTs for WGS in various projects, and what we need to know is how often you consider it useful to receive a PT on WGS from us?

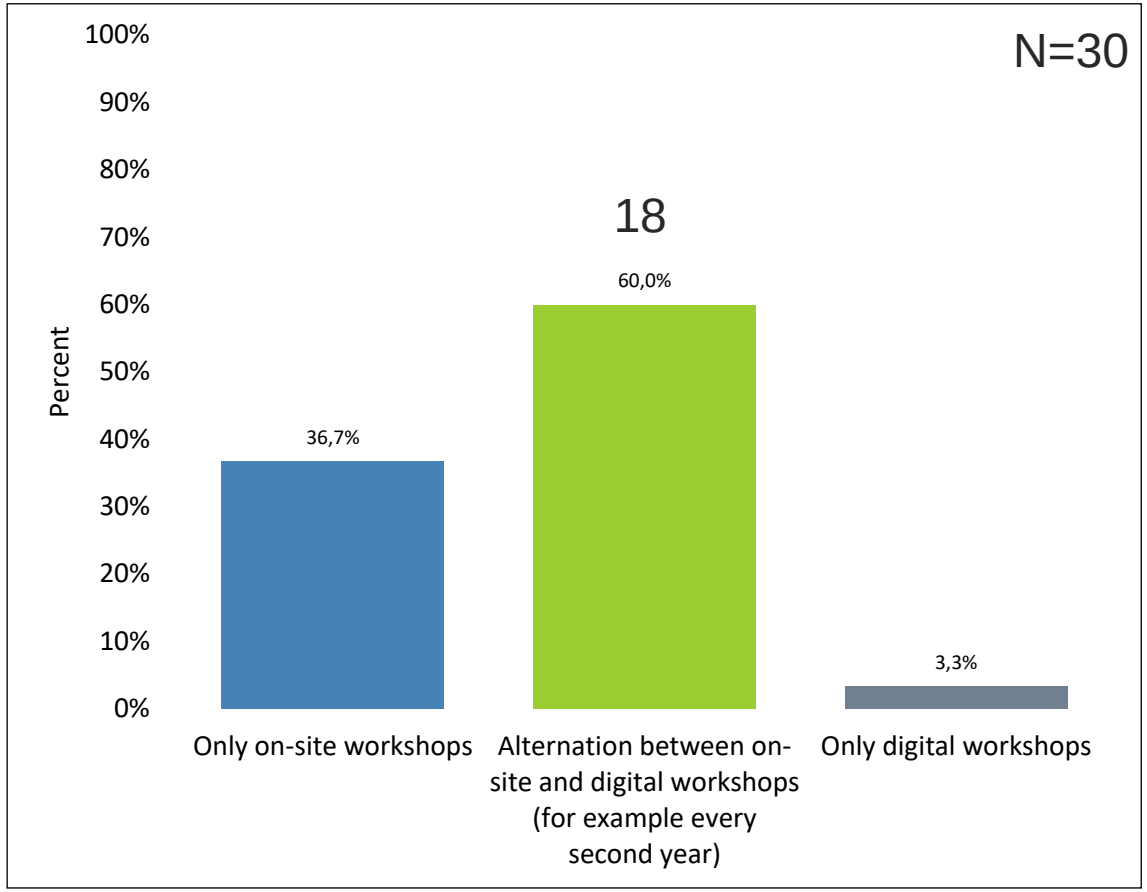


Question 7: For which of the following topics do you foresee the need for staff of your laboratory to participate in training in the two coming years?



Question 9:
Hybrid workshops are challenging to organise, for administrative reasons and for difficulties to achieve equal quality between digital and on-site participation. For upcoming workshops (and if the situation allows us to choose), would you prefer only on-site workshops or alternation between on-site and digital workshops (for example every second year).

Advantage on-site	Advantage digital
Get to know new colleagues	When it's not possible to send any representative in person
Networking	
Stay more focused	
Exchange experiences	



Follow-up to responses and continuous discussions

Follow-up to responses – future workshops

- With the improved technology and the big willingness and experience of the network to attend and engage in digital meetings, we consider it reasonable to alternate future workshops between physical meetings and digital meetings to save time and resources.

- But since travel still may be affected to some extent by covid-19 this year, we plan to organise a physical workshop again next year before starting the alternation between digital and physical workshops.

Follow-up to responses - PTs

- Since enumeration of *Campylobacter* is specified in a regulation (microbiological criteria) we see the need to continue to have one mandatory (for each MS) PT for enumeration every year.

- Due to the generally very good results for detection we will continue to define the PT for detection as voluntary every other year (with only 10 samples) and mandatory every other year (with 18 samples as recommended in ISO 22117).

- We believe it is most relevant to continue to allow to choose ”routinly used method” – majority uses ISO 10272. Keep in mind to send enough matrix so it is possible to chose procedure.

- Frequency of WGS-PTs: every second year seems to be a relevant frequency both for NRLs and the EURL.

Follow-up to responses – test portion size

- 25 g is a test portion size used by several NRLs for detecting *Campylobacter* and therefore validating the ISO 10272-1 (detection) for 25 g seems relevant for our network.

- In order to validate ISO 10272-1 for larger test portion size according to ISO 16140-2 (ISO 17468 Microbiology of the food chain — Technical requirements and guidance on establishment or revision of a standardized reference method):
  - 18 samples per matrix (6 negative, 6 low levels and 6 high levels)
  - Matrices: chicken skin, raw milk, frozen spinach, frozen minced meat, fresh produce (cabbage?)
  - Need 10 valid datasets (15 participants per matrix)

Follow-up to responses – test portion size

- For ISO 10272-1 (detection) it is difficult to use results from PTs to validate the method because:
  - validation study requires very low levels – reasonable for evaluation of the method performance but not for laboratory performance,
  - only food matrices (and laboratories needing PT for detection usually needs animal samples)
  - Difficult to combine with identification PT (one strain per matrix)

- To conduct a new validation study for ISO 10272-1 we think it should be organised as an ILS – and before the end of 2027
- For additional validation of ISO 10272-2 (additional food category), PT results can be used.

Year	Purpose	Type of PT/ILS	Matrix	Number of samples	Mandatory/ Voluntary
2023	PT	Enumeration	Chicken meat	10	**Mandatory**
	PT	Detection	Sock samples	18	**Mandatory**
2024	PT	Enumeration	Chicken skin	10	**Mandatory**
	PT	Detection	Chicken caecal samples	10	Voluntary
	PT	Whole genome sequence quality and cluster detection	-	2 + larger dataset	Voluntary
2025	PT	Enumeration	Chicken skin	10	**Mandatory**
	PT	Detection	Environmental swab samples	18	**Mandatory**
2026	PT	Enumeration	Cabbage	10	**Mandatory**
	PT	Detection	Raw milk	10	Voluntary
	PT	Whole genome sequence quality and cluster detection	-	?	Voluntary
	Validation	ILS	Cabbage, chicken skin, raw milk, frozen spinach, frozen minced meat	18 per matrix	Voluntary

Proposal for years 2023-2026

Topics for tomorrow's group discussions

1. Discuss the proposed outline of PTs and ILS studies in 2023 to 2026 in terms of feasability, relevance and alternative suggestions.

2a. Discuss possible areas to be further studied/changed in a future revision of the standard ISO 10272:2017.

2b. Discuss your first experiences with the harmonised protocol for isolation of *Campylobacter* for AMR monitoring.

Group dicussions for NRLs

- Seven participants in each group.
- You will be divided into groups when you come in the morning or be directed to different Teams break-out rooms
- Select one presenter in each group.
- Each group will have maximum ~3 min to present the main conclusions

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Thank you for your attention!

Questions?