

# DETECTION OF CAMPYLOBACTER IN MILK

## A COLLABORATIVE STUDY

*EURL-Campylobacter* workshop 2018

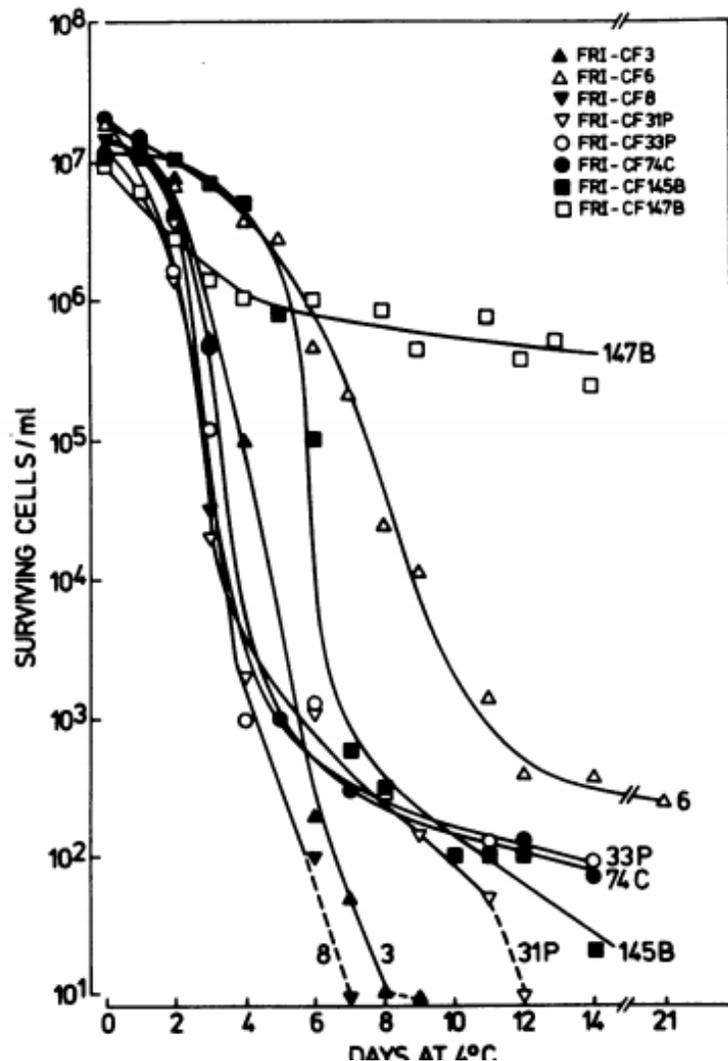
Hanna Skarin



# CAMPYLOBACTER IN MILK

- *Campylobacter* spp. - in the intestine of healthy cattle
- Risk for fecal contamination of milk
- Pasteurization - effective way to improve milk safety
- But – consumers ask for 'raw milk'
- Studies performed on prevalence in raw milk find *Campylobacter* in 0.4-12% of samples
- Type of samples: bulk tank milk (BTM) or in-line milk filters

# CAMPYLOBACTER SURVIVAL IN MILK



Doyle and Roman, 1982

- The LOD<sub>50</sub> in milk is higher than other matrices tested (ISO 10272-1)
- Methodology, poor survival, or viable but nonculturable (VBNC) cells

# MILK AND PH

- The pH in fresh raw milk is 6.5-6.7
- The pH is affected by growth of lactic acid bacteria
- A lower pH:
  - Gives lactic acid bacteria an advantage
  - Increases the activity of Lactoperoxidase, naturally present in milk. It has a bacteriocidal effect on gram negative bacteria. Highest activity at ~pH 6

# ISO 10272:2017 (AND 2006)

## 8 Preparation of test sample

Prepare the test sample from the laboratory sample in accordance with the specific International Standard dealing with the product concerned: see ISO 6887 (all parts).

*ISO 6887 (all parts), Microbiology of the food chain — Preparation of test samples, initial suspension and decimal dilutions for microbiological examination*

*ISO 6887 – Part 5: Specific rules for the preparation of milk and milk products*

# ISO 6887-5

ISO 6887-5:2010 "specific rules for preparation of milk and milk products"

Currently under revision

Version	Preparation	pH	Test sample (ISO 6887-5 + ISO 10272-1)
2010	Add 9x "diluent of general use"	Should be adjusted to neutral	1 ml milk + 9 ml diluent should be mixed with 90 ml buljong enrichment broth
ISO/DIS	Make dilutions or inoculate directly in enrichment broth	Should be adjusted to neutral	As above, or 10 ml milk is inoculated directly into 90 ml broth

# ISO 6887-5 VS BACTERIOLOGICAL ANALYTICAL MANUAL (BAM)

Reference	Preparation	pH	Test sample
2010	Add 9x "diluent of general use"	Should be adjusted to neutral	1 ml milk + 9 ml diluent should be mixed with 90 ml buljong enrichment broth
ISO/DIS	Make dilutions or inoculate directly in enrichment broth	Should be adjusted to neutral	As above, or 10 ml milk is inoculated directly into 90 ml broth
BAM	Centrifuge 50 ml and resovl the pellet (without milk fat) in 10 ml broth	Should be adjusted to neutral at the sampling site and again at the laboratory	10 ml disolved pellet in broth is inoculated into 90 ml broth (Bolton)

# SUMMARY OF THE SURVEY "METHODS FOR DETECTION OF CAMPYLOBACTER IN MILK"

Aim: To explore how the laboratories in the EURL-*Campylobacter* network analyse milk for detection of *Campylobacter*

Questionnaire sent out: 2018-09-07

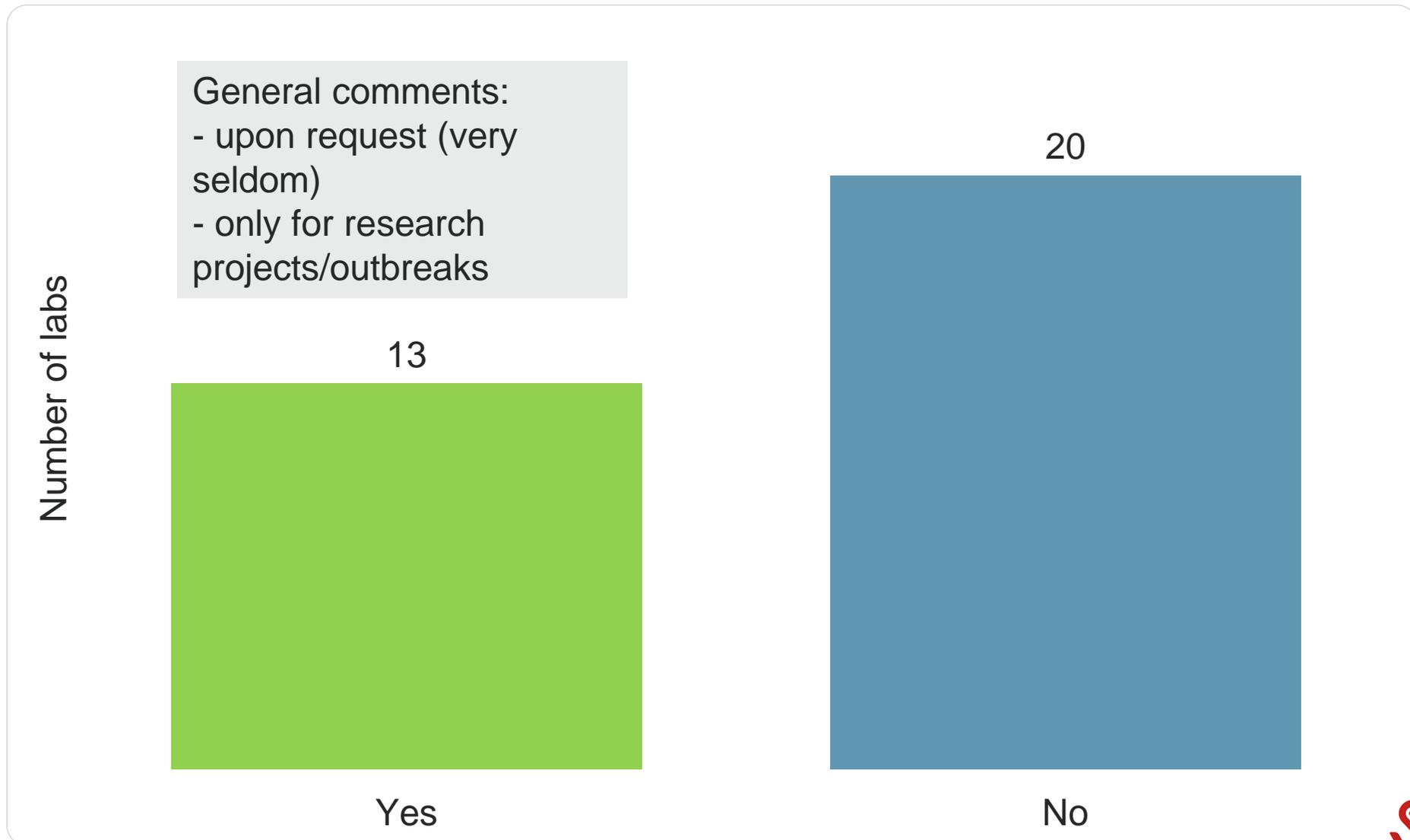
Last date to reply: 2018-09-28

Questionnaire sent to: the EURL-*Campylobacter* network (43)

Number of participants in the questionnaire: 33 (out of 43)

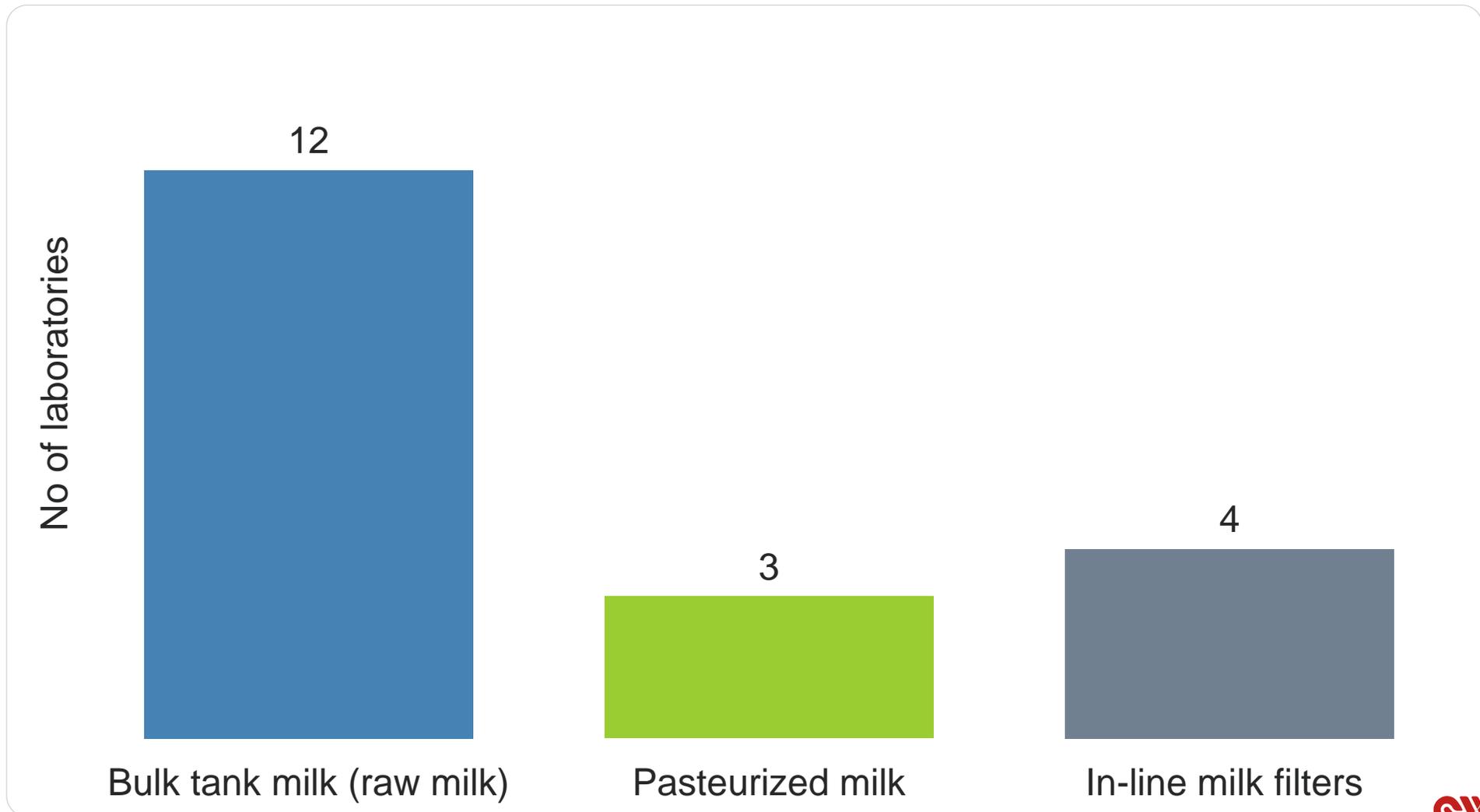
# Question no. 1. In your laboratory, do you analyse milk for Campylobacter?

N responding labs=33



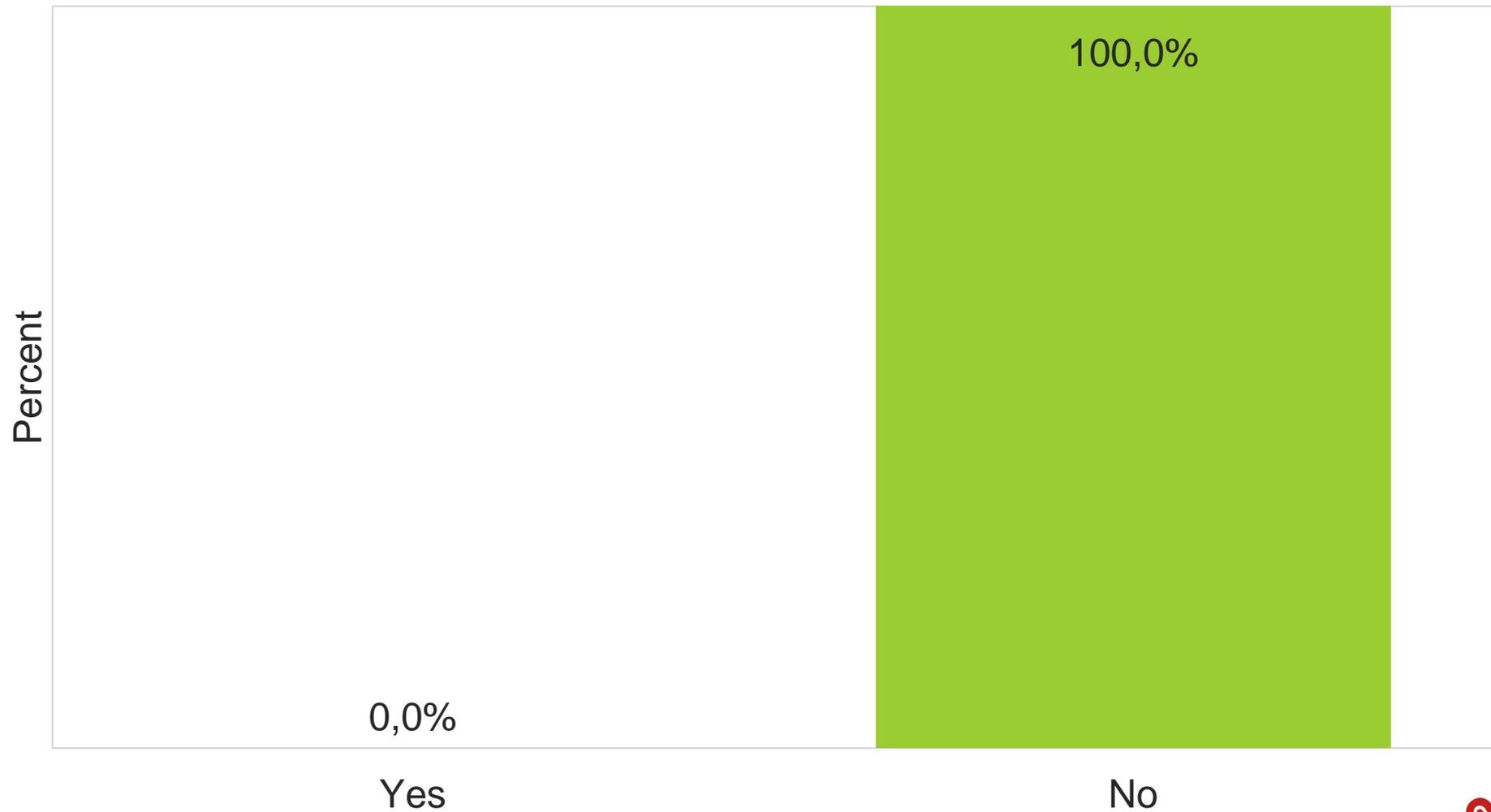
Question no. 2. If you analyse milk for Campylobacter, what type of samples do you receive (multiple choices)?

N responding labs=13



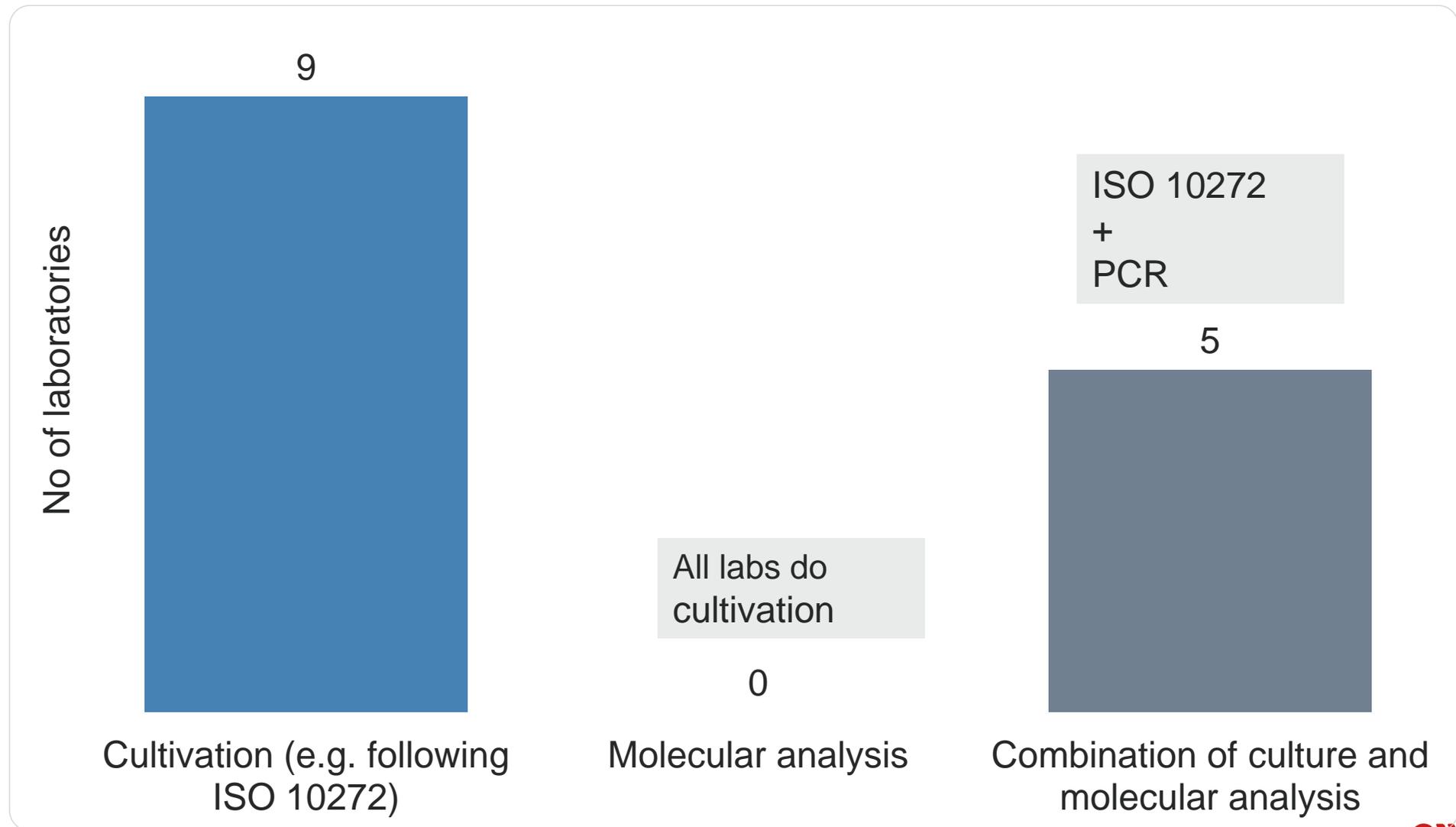
Question no. 3. Is the pH in the milk by any means adjusted into neutral prior to analysis (single choice)?

N responding labs=13



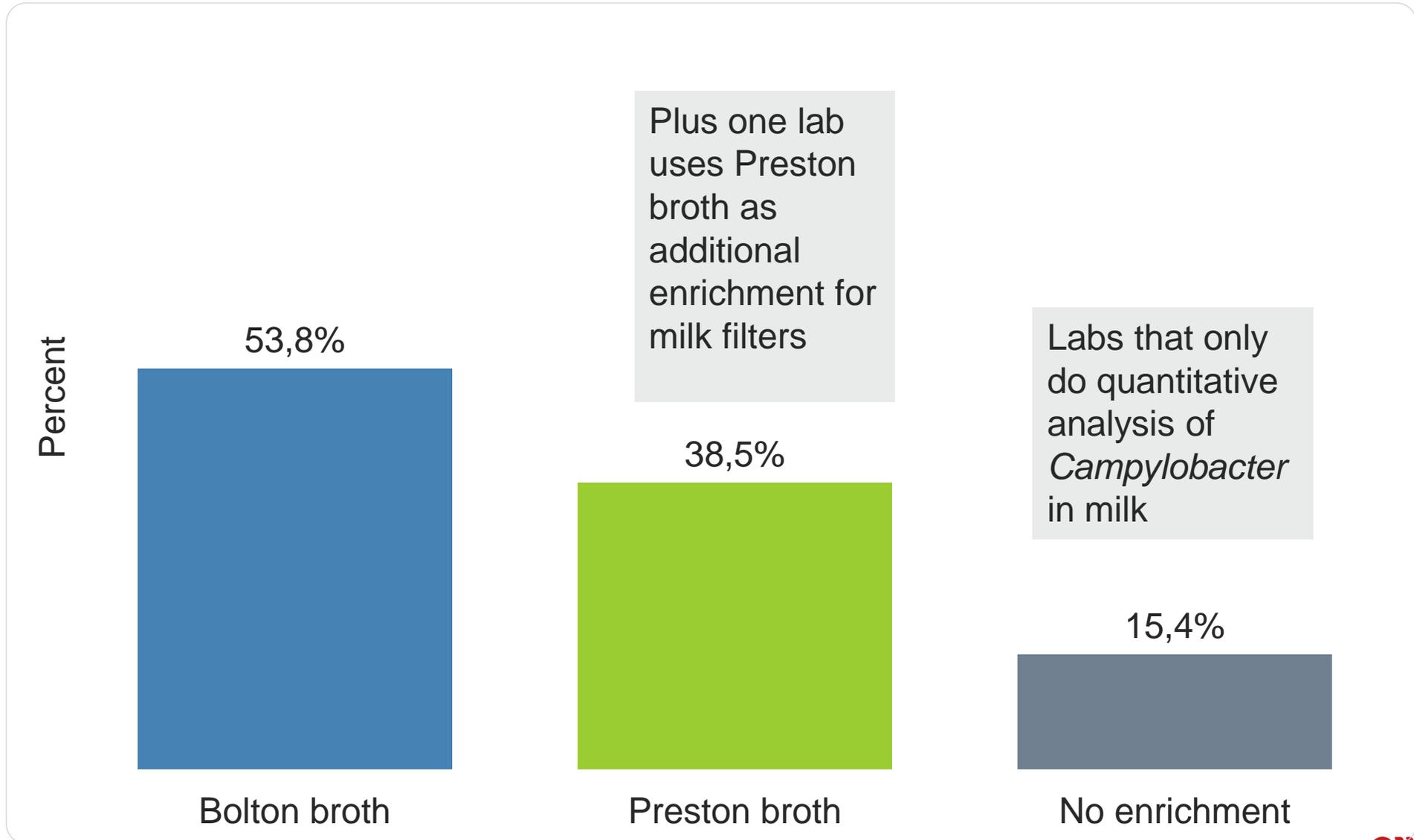
## Question no. 7. What principle of methodology do you use for the analysis of *Campylobacter* in milk (multiple choice)?

N responding labs=13

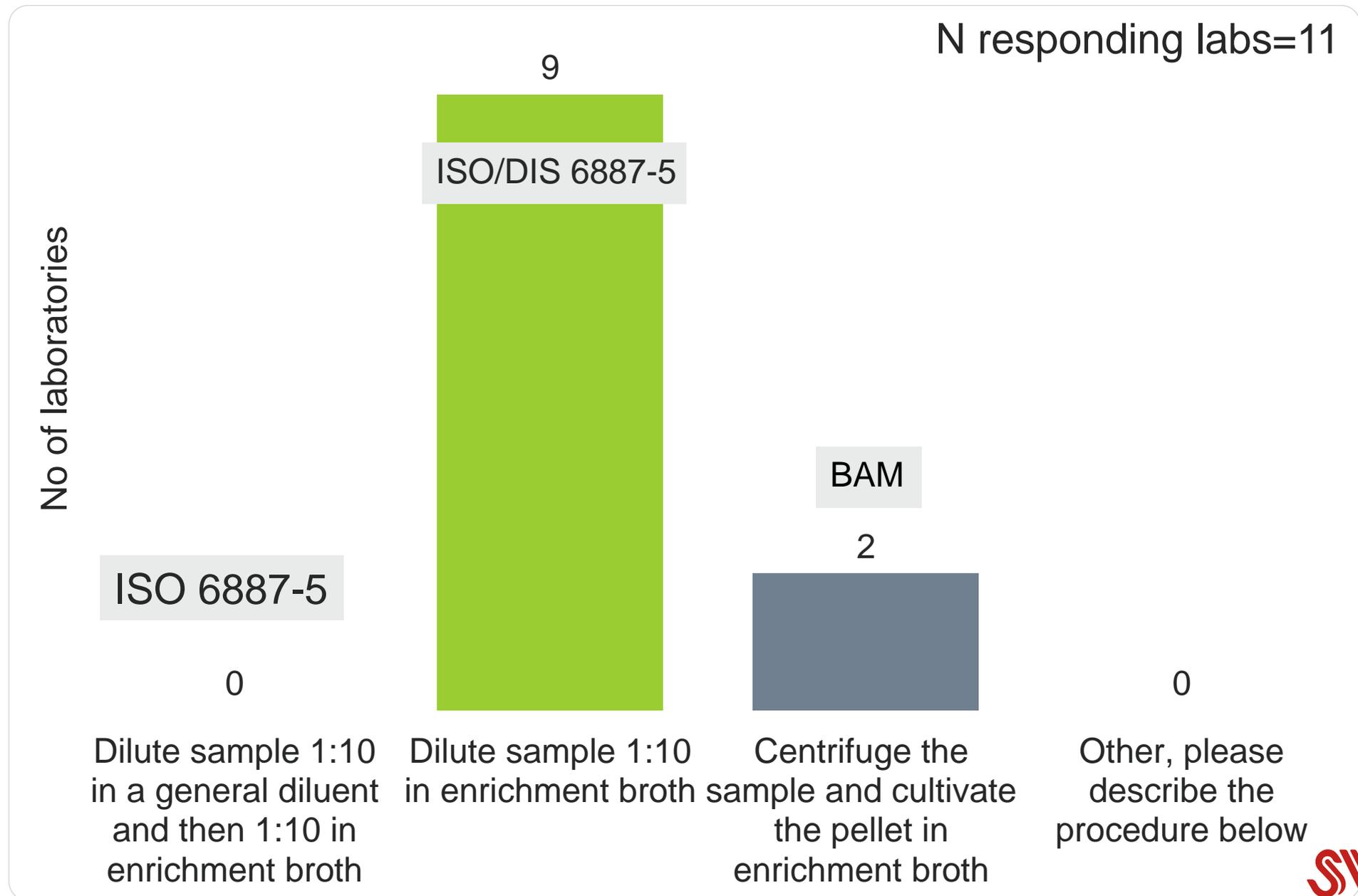


## Question no. 8. Which enrichment broth do you use for milk samples (multiple choice)?

N responding labs=13



## Question no. 9. How do you proceed when preparing a milk sample for enrichment of *Campylobacter* (multiple choice)?



# SUGGESTION FOR COLLABORATIVE STUDY

- Does pH adjustment have a positive impact on detection of *Campylobacter*?
- How well does *Campylobacter* survive/stay culturable in milk?
- Practical approach:
  - The EURL sends strains to participants
  - All follow same test protocol but spike domestic milk
  - Possibly including naturally contaminated samples

# TO BE CONTINUED

- Comments are welcome! Please e-mail to [hanna.skarin@sva.se](mailto:hanna.skarin@sva.se)
- The EURL will send out a summary of the background and a brief description proposal to a study plan
- You can comment on the study plan and tell us if you are interested in participating
- We discuss the study together and produce the study plan for 2019/2020