# Proficiency testing Food Microbiology

April 2023

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# **Abbreviations**

#### Media

ALOA Agar for Listeria according to Ottaviani & Agosti

APW 2% Alcaline peptone water, 2 % NaCl

BA Blood agar

BcsA Bacillus cereus selective agar

BEA Bile esculin agar
BGA Brilliant green agar

BGLB Brilliant green lactose bile broth

BP Baird-Parker agar

BPW Buffered peptone water

BS Bromthymol blue saccharose agar CBC Oxoid Brilliance™ *Bacillus cereus* agar CIN Cefsulodin irgasan novobiocin agar Compact Dry EC Compact Dry™ *E. coli* and coliforms Compact Dry ETB Compact Dry™ Enterobacteriaceae Compact Dry ETC Compact Dry™ Enterococcus Compact Dry TC Compact Dry™ Total Count **COMPASS** COMPASS® Enterococcus agar

CT-SMAC Cefixime tellurite sorbitol MacConkey agar

DG18 Dikloran glycerol agar

DRBC Dikloran Rose-Bengal chloramphenicol agar

EC E. coli broth

ENT Slanetz & Bartley Enterococcus agar

HEA Hektoen enteric agar

IA Iron agar

ISA Iron sulphite agar

ITC Irgasan ticarcillin potassium chlorate broth

KEAA Kanamycin esculin azide agar

LMBA Listeria monocytogenes blood agar

LSB Lauryl sulphate broth

LTLSB Lactose tryptone lauryl sulphate broth

mCCDA Modified charcoal cephoperazone deoxycholate agar

mCP Membrane Clostridium perfringens agar

MKTTn Muller-Kauffmann tetrathionate/novobiocin broth

MPCA Milk plate count agar
MRB Modified Rappaport broth

MRS de Man, Rogosa and Sharpe agar

MRS-aB de Man, Rogosa and Sharpe agar with amphotericin MRS-S de Man, Rogosa and Sharpe agar with sorbic acid

MSRV Modified semi-solid Rappaport-Vassiliadis enrichment media

mTSB Modified tryptone soya broth

MYP Mannitol egg yolk polymyxin agar NAP Nitrite actidione Polymyxin agar OCLA Oxoid Brilliance™ Listeria agar

OGYE Oxytetracyclin glucose yeast extract agar

OPSP Oleandomycin, Polymixin, Sulphadiazine, Perfringens agar

PAB Perfringens agar base
PDA Potato dextrose agar

PALCAM Polymyxin acriflavine lithium chloride ceftazidime aesculin mannitol agar

Petrifilm AC 3M™ Petrifilm™ Aerobic Count Petrifilm CC 3M™ Petrifilm™ Coliform count Petrifilm Disk 3M™ Petrifilm™ Staph Express Disk Petrifilm EB 3M™ Petrifilm™ Enterobacteriaceae 3M<sup>™</sup> Petrifilm<sup>™</sup> *E. coli*/Coliform count Petrifilm EC/CC Petrifilm EL 3M™ Petrifilm™ Environmental Listeria Petrifilm LAB 3M™ Petrifilm™ Lactic acid bacteria Petrifilm RAC 3M™ Petrifilm™ Rapid Aerobic Count

Petrifilm REC 3M™ Petrifilm™ Rapid *E. coli*/Coliform count

Petrifilm SEC 3M™ Petrifilm™ Select *E. coli*Petrifilm Staph 3M™ Petrifilm™ Staph Express

PEMBA Polymyxin pyruvate egg yolk mannitol bromothymol blue agar

PSB Peptone sorbitol bile salts broth

PCA Plate count agar

RPFA Baird-Parker agar with rabbit plasma fibrinogen

SFA Sugar-free agar

RVS Rappaport-Vassiliadis Soy peptone broth Saubouraud Saubouraud chloramphenicol agar

SC Sulphite cycloserine agar

SFP Shahidi-Ferguson Perfringens agar

SMAC Sorbitol MacConkey agar SP Salt Polymyxin broth

SSDC Salmonella/Shigella sodium deoxycholate calcium chloride agar

TBX Tryptone bile X-glucuronide agar

TCBS Thiosulphate citrate bile salts sucrose agar

TGE Tryptone glucose extract agar

TEMPO AC
TEMPO® Aerobic count
TEMPO BC
TEMPO® Bacillus cereus
TEMPO CAM
TEMPO CC
TEMPO® Campylobacter
TEMPO CC
TEMPO® Coliform count
TEMPO EB
TEMPO® Enterobacteriaceae

TEMPO EC TEMPO® E. coli

TEMPO RYM TEMPO® Rapid Yeast/Mould

TEMPO STA TEMPO® Coagulase-positive staphylocci

TEMPO YM TEMPO® Yeast/Mould

TGE Tryptone glucose extract agar

TS Tryptose sulphite agar

TSA Tryptic soya agar

TSC Tryptose sulphite cycloserine agar **TSBY** Tryptone soya broth with yeast extract XLD

Xylose lysine deoxycholate agar

VRB Violet red bile agar

VRBG Violet red bile glucose agar

YGC Yeast extract glucose chloramphenicol agar

# Organisations

**AFNOR** French National Standardization Association

AOAC AOAC INTERNATIONAL

ATCC American Type Culture Collection

CBS Centraalbureau voor Schimmelcultures (Westerdijk Institute)

CCUG **Culture Collection University of Gothenburg** 

IDF **International Dairy Foundation** 

ISO International Organization for Standardization NMKL Nordic-Baltic Committee on Food Analyses

NordVal NordVal International - NMKL

SLV Livsmedelsverket/Swedish Food Agency, Sweden

Fohm Public Health Agency of Sweden

# Analyses in this PT round

# Quantitative analyses

Aerobic microorganisms, 30  $^{\circ}\mathrm{C}$ 

Psychrotrophic microorganisms

Enterobacteriaceae

Escherichia coli

Presumptive Bacillus cereus

Coagulase-positive staphylococci

Lactic acid bacteria

Clostridium perfringens

Anaerobic sulphite-reducing bacteria

Aerobic microorganisms in fish products

H<sub>2</sub>S-reducing bacteria in fish products

Yeasts

Moulds

# Method

# Reporting of results and method information

It is the responsibility of the individual participants to correctly report results according to the instructions. Incorrectly reported results, for example results reported for the wrong sample, cannot be correctly processed. Incorrectly reported results are as a general rule excluded but may – after manual assessment by the Swedish Food Agency in each individual case – still be included and processed.

It is also mandatory for the participants to report method information for all analyses. This method information is sometimes contradictory or difficult to interpret. For example when participants state a medium that is not included in the standard method they refer to, or when manual comments by the participant contradict the reported method information. In such cases, the reported method information provided by the participants is generally used in method comparisons "as it is". Alternatively, method data that are difficult to interpret may be excluded or added to the group "Other", together with results from methods and media that are only used by 1–2 participants.

# Standard deviation and assigned value

Evaluation of the participants' results and statistical calculations are carried out on the log<sub>10</sub> transformed results. Results reported by participants as "> value" are not evaluated. Results reported as "< value" are treated as zero (negative result).

A robust statistical approach is used to determine the mean value and standard deviation. Algorithm A with iterated scale as described in ISO 13528:2022 [1] is used to determine the robust mean  $(m_{PT})$  and robust standard deviation  $(s_{PT})$  of the participants' results. Results that are obviously erroneous are excluded prior to determining  $m_{PT}$  and  $s_{PT}$  (blunder removal). For evaluated parameters, the assigned value consists of  $m_{PT}$ . It is regarded as the true, normative value.

For small datasets, there is an increased uncertainty associated with determining the robust mean  $(m_{PT})$  and robust standard deviation  $(s_{PT})$  of the participants' results. Therefore, when fewer than 12 participants have reported evaluated results, the statistical measures for performance evaluation will be provided *only as an information* to the participants.

#### **Outliers**

Outliers are results that deviate from the other results in a way that cannot be explained by normal variation. Results within  $m_{PT} \pm 3s_{PT}$  are considered acceptable, whereas results outside this interval are considered as outliers. When fewer than 12 participants have reported results, as well as in some individual cases, subjective adjustments are made to set acceptance limits based on prior knowledge of the samples contents.

#### Results from different methods

Non-robust median values (Med) and standard deviations (s) are calculated to assist in the evaluation of the results from different methods. These are shown in tables in the report, in connection with the respective analyses. In these instances, Med and s are calculated from the respective method groups' results, with outliers and false results excluded. For method groups with fewer than 5 results, only the number of false results and outliers are provided.

# Measurement uncertainty for the assigned values

The standard uncertainty  $(u_{PT})$  of the assigned value  $(m_{PT})$  is estimated from the standard deviation  $(s_{PT})$  and the number of evaluated results (n):

$$u_{\rm PT} = 1.25 \times \frac{s_{\rm PT}}{\sqrt{n}}$$

The measurement uncertainty is considered negligible compared to the standard deviation (which is used for evaluating the participants' results) when:

$$u_{\rm PT} < 0.3 s_{\rm PT}$$

#### **Z-scores**

To allow comparison of the results from different analyses and samples, results are transformed into standard values (z-scores). Z-scores are calculated as:

$$z = \frac{x_{\rm lab} - m_{\rm PT}}{s_{\rm PT}}$$

where  $x_{lab}$  is the result of the individual participant.

Z-scores for individual analyses are shown in Annex 2 and can be used as a tool by participants when following up on the results. For quantitative analyses, a z-score is either positive or negative, depending on whether the participants result is higher or lower than  $m_{PT}$ .

In evaluations of the analytical results, the following guidelines can be used:

 $|z| \le 2$  indicates that the result is acceptable

2 < |z| < 3 indicates a warning that the result may be deviating, and might motivate an action in the follow-up process

 $|z| \ge 3$  indicates that the result is regarded as deviating and should lead to an action in the follow-up process

# Table legends

N number of participants that reported results for the analysis

number of participants with satisfactory result (false results and outliers excluded)

 $m_{\rm PT}$  assigned value, robust mean value in  $\log_{10}$  cfu ml<sup>-1</sup>

 $s_{\rm PT}$  robust standard deviation

 $u_{\rm PT}$  standard uncertainty of the assigned value

- F number of false positive or false negative results
- < number of low outliers
- > number of high outliers
- results deviating more than  $1 s_{PT}$  from m, or unusually many deviating results.

# Figure legends

- results within the interval of acceptance
- outlier
- ☐ false negative result
- \* value outside the x-axis scale

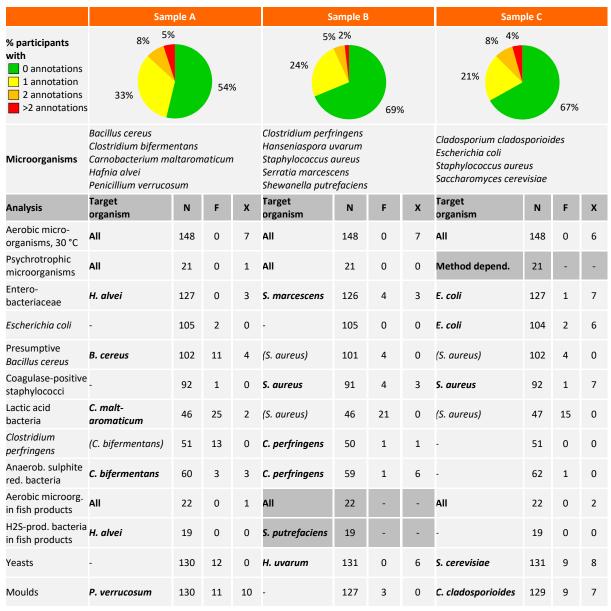
# Results

#### General outcome

Samples were sent to 168 participants; 38 in Sweden, 114 in Europe, and 16 outside of Europe. Of the 160 participants that reported results, 102 (64 %) provided at least one result that received an annotation. In the previous PT round with similar analyses (April 2022) the proportion was 62 %.

Individual results are listed in Annex 1 and on the website: <a href="https://www2.slv.se/absint">https://www2.slv.se/absint</a>. Z-scores for individual results are listed in Annex 2.

**Table 1**. Composition of the test material and proportion of deviating results (N: number of reported results, F: false positive or false negative, X: outliers)



<sup>-</sup> no target organism or no value; *microorganism* = main target organism; (*microorganism*) = false positive before confirmation

The results are not evaluated

# Aerobic microorganisms, 30 °C

# Sample A

H. alvei and C. maltaromaticum were the main target organisms.

In total, 148 participants reported results. Four low and three high outliers were reported.

# Sample B

S. putrefaciens and S. aureus were the main target organisms.

In total, 148 participants reported results. Three low and four high outliers were reported.

# Sample C

E. coli and S. aureus were the main target organisms.

In total, 148 participants reported results. Four low and two high outliers were reported.

#### General remarks

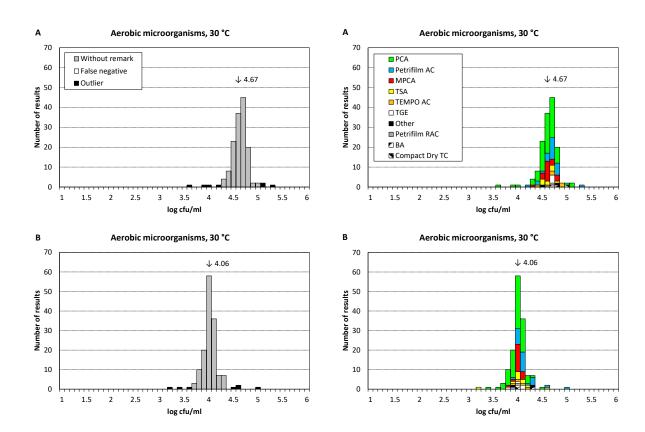
Most participants followed either NMKL 86:2013, ISO 4833-1:2013 or used 3M Petrifilm AC. The withdrawn NMKL 86:2006 and ISO 4833:2003 were used by eleven and six participants, respectively.

Both NMKL 86:2013 and ISO 4833-1:2013 are based on incubation on PCA or MPCA at 30 °C for 72 h. Users of Petrifilm AC can use different times/temperatures, depending on the method validation. For example, AOAC® prescribes incubation at 35 °C for 48 h while AFNOR prescribes 30 °C for either 48 h or 72 h, depending on which product that is analysed. ISO 4833-1:2013 was last reviewed by ISO in 2019 and remains current. An amendment with a clarification on the scope of the method is available (ISO 4833-1:2013/Amd 1:2022). NMKL 86:2013 was last reviewed by NMKL in 2022 and remains current.

The majority of the participants incubated on PCA, but Petrifilm AC was also common. Incubation on MPCA was mainly done by laboratories within the dairy industry. Incubation on TSA was mainly done by users of a company-specific method. A few participants used TEMPO AC, which is based on MPN (Most Probable Number). With this method, the sample is incubated in a card that contains different-sized wells. A substrate in the medium emits fluorescence when hydrolysed by the microorganisms. The number of microorganisms is determined statistically by the number and size of the fluoresceing wells.

**Table 2.** Results from analysis of aerobic microorganisms, 30 °C.

Medium			Samp	le A						Samp	le B						Samp	le C			
iviedium	N	n	m <sub>PT</sub>	<b>S</b> PT	F	<	>	N	n	m <sub>PT</sub>	<b>S</b> PT	F	<	>	N	n	m <sub>PT</sub>	SPT	F	<	>
All results	148	141	4.67	0.13	0	4	3	148	141	4.06	0.12	0	3	4	148	142	4.32	0.10	0	4	2
PCA	77	72	4.66	0.13	0	3	2	77	74	4.04	0.13	0	2	1	77	72	4.28	0.09	0	4	1
Petrifilm AC	26	24	4.72	0.12	0	1	1	26	24	4.12	0.12	0	0	2	26	25	4.39	0.10	0	0	1
MPCA	20	20	4.66	0.11	0	0	0	20	20	4.08	0.08	0	0	0	20	20	4.32	0.10	0	0	0
TSA	9	9	4.68	0.11	0	0	0	9	8	4.04	0.08	0	1	0	9	9	4.34	0.06	0	0	0
TEMPO AC	5	5	4.79	0.17	0	0	0	5	4	-	-	0	0	1	5	5	4.39	0.10	0	0	0
TGE	4	4	-	-	0	0	0	4	4	-	-	0	0	0	4	4	-	-	0	0	0
Other	2	2	-	-	0	0	0	2	2	-	-	0	0	0	2	2	-	-	0	0	0
Petrifilm RAC	2	2	-	-	0	0	0	2	2	-	-	0	0	0	2	2	-	-	0	0	0
BA	2	2	-	-	0	0	0	2	2	-	-	0	0	0	2	2	-	-	0	0	0
Compact Dry TC	1	1	-	-	0	0	0	1	1	-	-	0	0	0	1	1	-	-	0	0	0



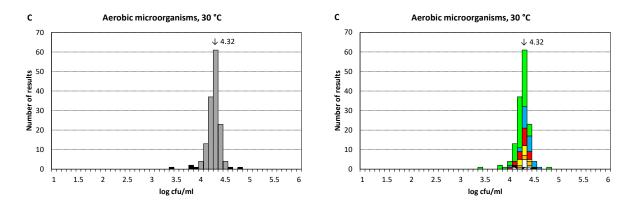


Figure 1. Results from analysis of aerobic microorganisms, 30 °C.

# Psychrotrophic microorganisms

# Sample A

H. alvei and C. maltaromaticum were the main target organisms.

In total, 21 participants reported results. One low outlier was reported.

# Sample B

S. putrefaciens and S. aureus were the main target organisms. In the quality control at the Swedish Food Agency (ten days incubation on PCA at 6.5 °C), two types of colonies were observed; larger white colonies and very small transparent colonies.

In total, 21 participants reported results. No outliers or false negative results were reported.

# Sample C

In the Swedish Food Agency's quality control, no colonies were observed on PCA after ten days incubation at 6.5 °C. Similar results were found for the participants; 9 of the 11 participants that incubated at low temperatures (6–7 °C) reported zero results. In contrast, the remaining 10 participants (of which most incubated at 15–22 °C) reported results in the range of 3.48–4.32 log<sub>10</sub> cfu ml<sup>-1</sup>, corresponding to *E. coli* and *S. aureus* in the sample.

Despite the stark differences in results, participants are not encouraged to repeat the analysis. Rather, since the differences in results align very well with the choice of method, they are likely to reflect the "true" results of the respective methods.

#### The results for sample C are not evaluated, and no z-scores are calculated for the analysis.

Note: The assigned value ( $m_{PT}$ ) and standard deviation ( $s_{PT}$ ) provided in Annex 1 are calculated only from the positive results, and can thus be used by participants that want to evaluate their own performance. They should however be used carefully, and only as a guidance, since the measurement uncertainty of the assigned value is not negligible ( $u_{PT}$  = 0.118).

#### General remarks

In summary, the results are difficult to evaluate. This is mainly due to the differences between the methods, but also due to the fact that several participants used modified methods. Users of NMKL 86:2013 typically incubated for 10 days at 6.5 °C, but 20 h at 17 °C followed by 3 days at 7 °C was also used. For psychrotrophic microorganisms in milk, ISO 6730:2005/IDF 101:2005 stipulates incubation at 6.5 °C. The other method for milk, ISO 8552:2004/IDF 132:2004, instead estimates the number of psychrotrophic microorganisms in a rapid method based on incubation at 21 °C. Both of these have been replaced by ISO 17410:2019, which stipulates 6.5 °C as the primary incubation temperature.

Several methods were referenced by only one participant each. These included NMKL 74:2000, ISO 4833-1:2013, ISO 6730:2005/IDF 101:2005 and NMKL 184:2006. NMKL 74:2000 has been replaced

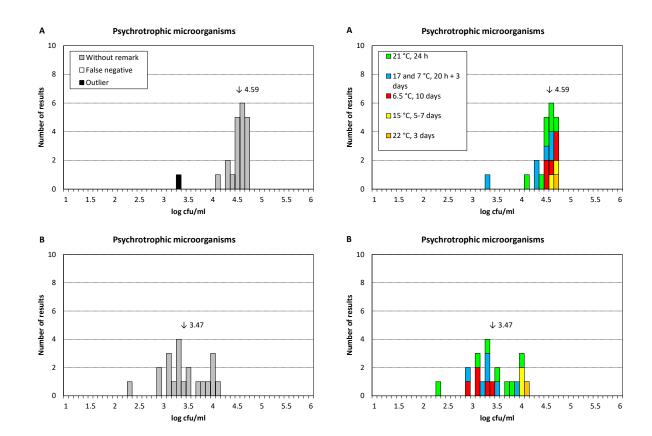
by NMKL 86:2013. NMKL 184 is mainly adapted for analysis of aerobic count and specific spoilage organisms in fish and fish products.

Fourteen of the 21 participants incubated on PCA, three on MPCA, two on Petrifilm AC and two on Long & Hammer agar.

**Table 3.** Results from analysis of psychrotrophic microorganisms.

Incubation			Sam	ple A						Sam	ple B						Sam	ole C*			
IIICUDALIOII	N	n	m <sub>PT</sub>	<b>S</b> PT	F	<		N	n	m <sub>PT</sub>	<b>S</b> PT	F	<	>	N	n	m <sub>PT</sub>	<b>S</b> PT	F	<	>
All results	21	20	4.59	0.16	0	1	0	21	21	3.47	0.47	0	0	0	21	21	3.94*	0.33*	-	-	-
21 °C, 24 h	7	7	4.57	0.20	0	0	0	7	7	3.51	0.58	0	0	0	7	-	-	-	-	-	-
17 and 7 °C, 20 h + 3 days	6	5	4.53	0.14	0	1	0	6	6	3.31	0.34	0	0	0	6	-	-	-	-	-	-
6.5 °C, 10 days	5	5	4.66	0.08	0	0	0	5	5	3.16	0.21	0	0	0	5	-	-	-	-	-	-
15 °C, 5-7 days	2	2	-	-	0	0	0	2	2	-	-	0	0	0	2	-	-	-	-	-	-
22 °C, 3 days	1	1	-	-	0	0	0	1	1	-	-	0	0	0	1	-	-	-	-	-	-

<sup>\*</sup> The sample is not evaluated. The values are shown only as an information to the participants. The measurement uncertainty of the assigned value is not negligible.



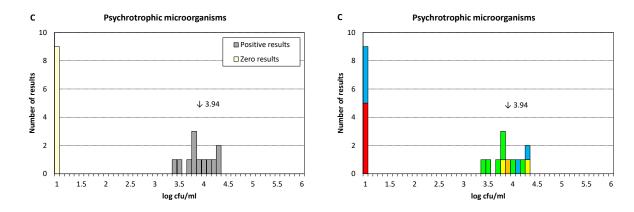


Figure 2. Results from analysis of psychrotrophic microorganisms

# Enterobacteriaceae

# Sample A

*H. alvei* was target organism. On VRBG, it forms typical red colonies that are surrounded by a bile salt precipitation zone. The strain is oxidase negative.

In total, 127 participants reported results. Two low and one high outliers were reported.

# Sample B

S. marcescens was target organism. On VRBG, it forms small pink/red colonies that are surrounded by a less prominent bile salt precipitation zone. The strain is oxidase negative.

In total, 126 participants reported results. Two low and one high outliers were reported, as well as four false-negative results.

# Sample C

*E. coli* was target organism. On VRBG, it forms dark red colonies that are surrounded by a distinct bile salt precipitation zone. The strain is oxidase negative.

In total, 127 participants reported results. Two low and five high outliers were reported, as well as one false negative result.

#### General remarks

Enterobacteriaceae are Gram-negative and oxidase-negative bacteria that ferment glucose with the production of acid by-products. On VRBG they therefore form pink/red colonies, with or without a bile salt precipitation zone. The appearance is similar on Petrifilm EB, which also includes a colour indicator for acid by-products and a plastic film for detection of gas production.

Most participants followed either NMKL 144:2005 (42 %), a method with Petrifilm EB (24 %), or an ISO method (24 %). Among the latter, the majority followed ISO 21528-2:2017, which is based on colony-count. In comparison, ISO 21528-1:2017 is based on MPN. The latter method is recommended when the expected level of Enterobacteriaceae is lower than 100 cfu g<sup>-1</sup>. Both ISO standards were last reviewed by ISO in 2022 and remain current. Ten participants still followed either of the previous – and now withdrawn – ISO 21528-2:2004 and ISO 21528-1:2004.

For sample B, the median value for participants that incubated on TSA/VRB, or that used TEMPO EB, differed by more than one  $s_{PT}$  from the assigned value. However only five and six participants used these media, respectively, so it might not reflect a true difference in the results between these methods.

Confirmation was performed by 61 % of the participants, and most often consisted of an oxidase test.

**Table 4.** Results from analysis of Enterobacteriaceae.

Medium			Samp	le A						Samp	le B						Samp	le C			
Medium	N	n	m <sub>PT</sub>	<b>S</b> PT	F	<	>	N	n	m <sub>PT</sub>	<b>S</b> PT	F	<	>	N	n	m <sub>PT</sub>	S <sub>PT</sub>	F	<	>
All results	127	124	4.14	0.15	0	2	1	126	119	2.63	0.17	4	2	1	127	119	3.80	0.14	1	2	5
VRBG	80	77	4.12	0.13	0	2	1	80	75	2.60	0.17	2	2	1	80	74	3.80	0.15	1	2	3
Petrifilm EB	32	32	4.21	0.15	0	0	0	32	31	2.67	0.12	1	0	0	32	31	3.84	0.10	0	0	1
TSA/VRBG	6	6	4.13	0.16	0	0	0	5	5	2.38	0.25	0	0	0	6	6	3.71	0.11	0	0	0
TEMPO EB	6	6	4.03	0.17	0	0	0	6	6	2.84	0.15	0	0	0	6	6	3.81	0.17	0	0	0
Compact Dry ETB	2	2	-	-	0	0	0	2	1	-	-	1	0	0	2	2	-	-	0	0	0
Other	1	1	-	-	0	0	0	1	1	-	-	0	0	0	1	0	-	-	0	0	1

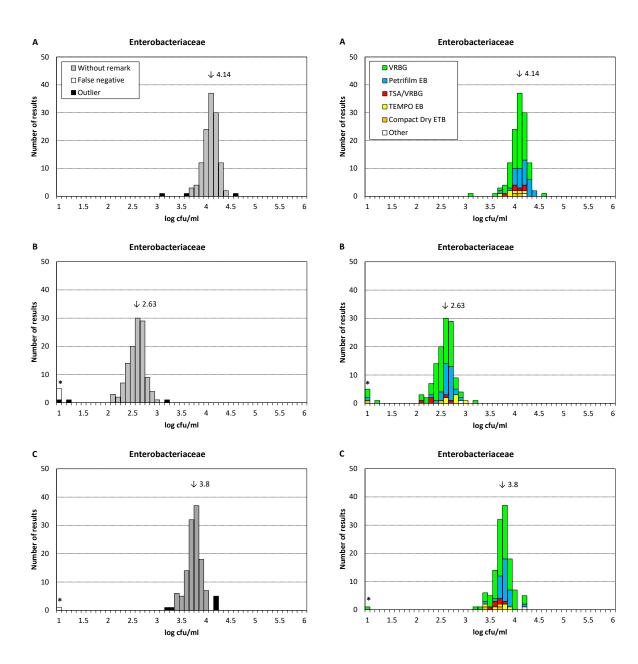


Figure 3. Results from analysis of Enterobacteriaceae

# Escherichia coli

# Sample A

No target organism was present in the sample.

In total, 105 participants reported results. Two false positive results were reported.

## Sample B

No target organism was present in the sample.

In total, 105 participants reported results. No false positive results were reported.

# Sample C

E.~coli was target organism. On TSA/VRB, it forms typical dark red colonies surrounded by a distinct bile salt precipitation zone. The strain is positive for indole production and β-glucuronidase activity, and produces gas in LTLSB.

In total, 104 participants reported results. Four low and two high outliers were reported, as well as two false negative results.

### General remarks

In total, 32 % of the participants used a method based on 3M<sup>TM</sup> Petrifilm<sup>TM</sup>. NMKL 125:2005 and ISO 16649-2:2001 were in comparison used by 23 % and 22 % of the participants, respectively. A few of the participants that followed NMKL 125:2005 and ISO 16649-2:2001 used media other than those stipulated by the respective standards, for example Petrifilm EC/CC or Brilliance EC/CC. 16649-2:2001 was last reviewed by ISO in 2019, and remains current.

ISO 7251:2005 and NMKL 96:2009 were used by four and one participants, respectively. ISO 7251 is an MPN-based method for the detection *E. coli*. NMKL 96 is also based on MPN, and is adapted for analysis of coliform bacteria, thermotolerant coliform bacteria and *E. coli* in fish and seafood. ISO 7251:2005 was last reviewed by ISO in 2019, and remains current.

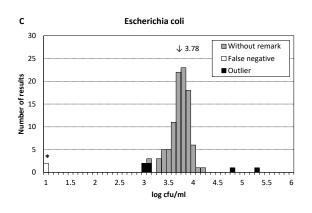
The definition of *E. coli* differs between methods, which may sometimes cause problems with non-typical strains. Users of ISO 16649-2:2001 typically count as *E. coli* those bacteria that form blue (i.e. β-glucuronidase positive) colonies on TBX, without further confirmation. Similarly, Petrifilm EC/CC and Petrifilm SEC are also based on media that detect *E. coli* β-glucuronidase. The latter media however also have a plastic film that facilitates detection of gas production due to lactose fermentation. In comparison, users of NMKL 125:2005 typically count as *E. coli* those bacteria that form typical dark red colonies surrounded by a red precipitation zone on VRB. Users often, but not always, perform further confirmation test, for example tests for the production of gas and indole. Other media, e.g. Brilliance<sup>TM</sup> EC/CC, facilitate detection and distinction between coliform bacteria and *E. coli*, based their differences in β-glucuronidase and β-galactosidase activity.

Despite these differences, altogether, the results from the different methods and media were similar. This is true also when considering differences in the incubation temperature, where participants typically incubated either at 41.5-44 °C (53 %) or at 35-37 °C (44 %).

 Table 5. Results from analysis of Escherichia coli.

Medium		9	Sampl	e A					:	Sampl	е В						Samp	ole C			
iviedium	N	n	m <sub>PT</sub>	S <sub>PT</sub>	F	<	>	N	n	m <sub>PT</sub>	S <sub>PT</sub>	F	<	>	N	n	m <sub>PT</sub>	S <sub>PT</sub>	F	<	>
All results	105	103	-	-	2	-	-	105	105	-	-	0	-	-	104	96	3.78	0.20	2	4	2
Petrifilm EC/CC <sup>1</sup>	23	23	-	-	0	-	-	23	23	-	-	0	-	-	24	24	3.84	0.15	0	0	0
TBX	24	23	-	-	1	-	-	24	24	-	-	0	-	-	24	22	3.62	0.20	0	2	0
TSA/VRB <sup>2</sup>	18	18	-	-	0	-	-	18	18	-	-	0	-	-	18	18	3.86	0.15	0	0	0
Petrifilm SEC	15	15	-	-	0	-	-	15	15	-	-	0	-	-	15	13	3.83	0.12	1	0	1
Other	12	12	-	-	0	-	-	12	12	-	-	0	-	-	10	8	3.84	0.23	0	1	1
TEMPO EC	6	6	-	-	0	-	-	6	6	-	-	0	-	-	6	6	3.95	0.15	0	0	0
Rapid'E.coli 2	3	3	-	-	0	-	-	3	3	-	-	0	-	-	3	3	-	-	0	0	0
VRB	2	1	-	-	1	-	-	2	2	-	-	0	-	-	2	1	-	-	1	0	0
Brilliance EC/CC	2	2	-	-	0	-	-	2	2	-	-	0	-	-	2	1	-	-	0	1	0

<sup>&</sup>lt;sup>2</sup> Petrifilm EC/CC includes one participant that used Petrifilm REC



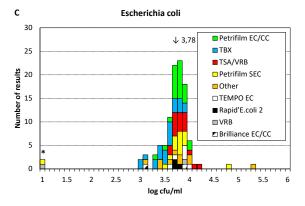


Figure 4. Results from analysis of Escherichia coli

<sup>&</sup>lt;sup>1</sup> TSA/VRB includes one participant that incubated on TSA/VRBG

# Presumptive Bacillus cereus

# Sample A

*B. cereus* was target organism. On BA, it forms typical grey colonies with a narrow zone of haemolysis. On BcsA, it forms typical blue colonies surrounded by a blue zone of precipitation. With some chromogenic media, the blue colour may be less prominent.

In total, 102 participants reported results. Two low and two high outliers were reported, as well as eleven false negative results. The false results were almost exclusively reported by users of NMKL67:2010 and ISO 7932:2004.

## Sample B

No target organism was present in the sample. S. aureus may form atypical colonies on BA and BcsA.

In total, 101 participants reported results. Four false positive results were reported.

## Sample C

No target organism was present in the sample. S. aureus may form atypical colonies on BA and BcsA.

In total, 102 participants reported results. Four false positive results were reported.

#### General remarks

Most participants followed either NMKL 67:2010 (35 %) or ISO 7932:2004 (31 %). The new NMKL 67:2021 – which replaces NMKL 67:2010 – was in contrast only followed only by twelve participants (12 %). Notably, users of the new NMKL 67:2021 reported no false results or outliers, whereas false results and outliers were reported by users of the old NMKL 67:2010 for all three samples. The new version of NMKL 67 stipulates a different primary incubation medium. ISO 7932:2004 was last reviewed by ISO in 2021, and remains current.

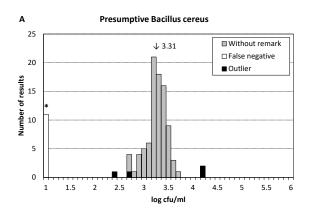
The most common selective media were MYP and BcsA. On BcsA, presumptive *B. cereus* form bluish colonies that are surrounded by a blue zone of precipitation, due to lecithinase activity on egg yolk present in the medium. On MYP, presumptive *B. cereus* form large pink colonies that are normally surrounded by a zone of precipitation, again as a consequence of lecithinase activity. MYP and BcsA were often used together with a non-selective medium, typically BA. On BA, *B. cereus* forms large, irregular grey colonies, surrounded by a distinct zone of haemolysis.

Other fairly common selective media were Brilliance<sup>TM</sup> B. cereus, PEMBA and Compact Dry X-BC. Brilliance<sup>TM</sup> B. cereus contains the substrate X-Gluc, which is cleaved by *B. cereus* β-glucuronidase. Colonies of *B. cereus* are therefore white with a blue/green centre on this medium. Similarly, the chromogenic and selective agents in Compact Dry X-BC cause *B. cereus* to form blue/green colonies, whereas other bacteria normally form white colonies. Compact Dry X-BC may give somewhat lower results compared to the reference method ISO 7932:2004, something that is mentioned in both the NordVal 045 and MicroVal 2011-LR41 validations. PEMBA is almost identical to BcsA in its

composition, and was similarly often used together with BA. In contrast, Brilliance $^{TM}$  B. cereus and Compact Dry X-BC were most often used as standalone media.

 Table 6. Results from analysis of presumptive Bacillus cereus.
 Only the main (selective) media are shown.

Medium			Sam	ple A						Samp	le B						Samp	le C			
Medium	N	n	m <sub>PT</sub>	S <sub>PT</sub>	F	<	>	N	n	m <sub>PT</sub>	S <sub>PT</sub>	F	<		N	n	m <sub>PT</sub>	S <sub>PT</sub>	F	<	>
All results	102	87	3.31	0.19	11	2	2	101	97	-	-	4	-	-	102	98	-	-	4	-	-
MYP	33	30	3.29	0.24	2	0	1	33	32	-	-	1	-	-	33	32	-	-	1	-	-
BcsA	25	20	3.26	0.21	4	1	0	24	22	-	-	2	-	-	25	22	-	-	3	-	-
BA	13	11	3.37	0.23	1	1	0	13	12	-	-	1	-	-	13	13	-	-	0	-	-
Brilliance™ B. cereus	7	7	3.34	0.09	0	0	0	7	7	-	-	0	-	-	7	7	-	-	0	-	-
PEMBA	6	4	-	-	2	0	0	6	6	-	-	0	-	-	6	6	-	-	0	-	-
Compact Dry X-BC	5	5	3.30	0.12	0	0	0	5	5	-	-	0	-	-	5	5	-	-	0	-	-
Other	5	3	-	-	1	0	1	5	5	-	-	0	-	-	5	5	-	-	0	-	-
ТЕМРО ВС	3	3	-	-	0	0	0	3	3	-	-	0	-	-	3	3	-	-	0	-	-
BACARA™	3	3	-	-	0	0	0	3	3	-	-	0	-	-	3	3	-	-	0	-	-
COMPASS® B. cereus	2	1	-	-	1	0	0	2	2	-	-	0	-	-	2	2	-	-	0	-	-



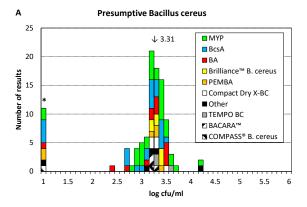


Figure 5. Results from analysis of presumptive Bacillus cereus.

# Coagulase-positive staphylococci

# Sample A

No target organism was present in the sample.

In total, 92 participants reported results. One false positive result was reported

## Sample B

*S. aureus* was target organism. This particular strain often grows slow on RPFA, and notably not all colonies may display a typical appearance and/or a distinct precipitation zone after 24 h incubation at 37 °C. Incubation for 48 h is therefore usually necessary to allow all bacteria in the sample to form typical colonies.

In total, 91 participants reported results. Three low outliers were reported, as well as four false negative results. All of the low outliers and false negative results were reported by participants that incubated for 48 h. Thus, the slow-growing characteristics of the particular *S. aureus* does not appear to have caused any problem for the participants.

# Sample C

S. aureus (different strain than in sample B) was target organism. On RPFA, it forms typical grey colonies surrounded by a precipitation zone.

In total, 92 participants reported results. Five low and two high outliers were reported, as well as one false negative result.

#### General remarks

Most participants incubated on BP. On this medium, *S. aureus* form characteristic convex, shiny colonies that have a grey/black colour due to reduction of tellurite in the medium. The colonies are usually surrounded by a clear zone, due to proteolysis of egg yolk in the medium (lecithinase activity). An opaque halo may also form near the colony, due to precipitation caused by lipase activity. Participants that used BP typically performed a confirmation based on coagulase activity, for example a tube coagulase test or the use of RPFA as a secondary medium.

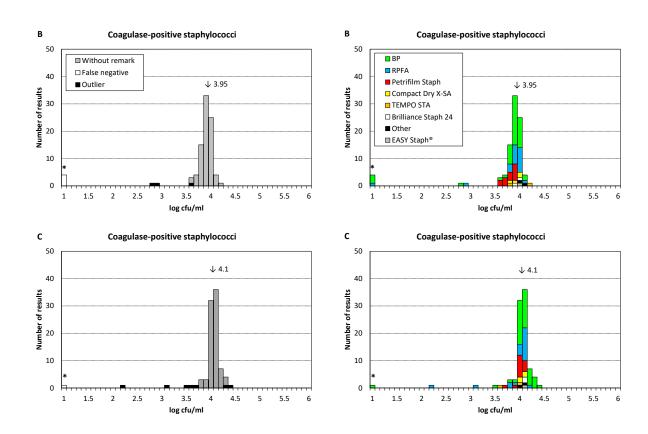
The second most common media were RPFA and Petrifilm Staph. With RPFA, the coagulase activity is tested directly in the medium. Petrifilm Staph is based on a modified Baird-Parker agar. It also contains a chromogenic indicator that causes *S. aureus* to form red/purple colonies. Participants that used Petrifilm Staph typically performed a confirmation with Petrifilm Staph Express Disk. This is based on detection of extracellular DNase, which is produced by the majority of coagulase-positive *S. aureus*, but also by the coagulase-positive staphylococci *S. intermedius* and *S. hyicus*. Toluidin blue O in the disks visualises DNase activity as a pink zone around the colonies.

Most participants (40 %) followed NMKL 66:2009, where incubation is done either with BP and/or RPFA. In comparison, ISO 6888-1:2021 stipulates BP, whereas 6888-2:2021 stipulates the use of RPFA. Most participants that followed an ISO method, referenced the withdrawn versions ISO 6888-1:1999

and ISO 6888-2:1999. One participant followed the MPN-based ISO 6888-3:2003, which is adapted for use when low numbers of stressed coagulase-positive staphylococci are expected. This was last reviewed by ISO in 2022 and remains current.

**Table 7.** Results from analysis of coagulase-positive staphylococci.

Markey			Sam	ole A						Sam	ple B						Sam	ple C			
Method	N	n	m <sub>PT</sub>	S <sub>PT</sub>	F	<	>	N	n	m <sub>PT</sub>	S <sub>PT</sub>	F	<	>	N	n	m <sub>PT</sub>	S <sub>PT</sub>	F	<	>
All results	92	91	-	-	1	-	-	91	84	3.95	0.09	4	3	0	92	84	4.10	0.09	1	5	2
ВР	46	45	-	-	1	-	-	44	40	3.97	0.09	3	1	0	45	41	4.11	0.09	1	1	2
RPFA	21	21	-	-	0	-	-	22	20	4.00	0.08	1	1	0	22	20	4.11	0.10	0	2	0
Petrifilm Staph	14	14	-	-	0	-	-	14	13	3.86	0.09	0	1	0	14	13	4.07	0.06	0	1	0
Compact Dry X-SA	4	4	-	-	0	-	-	4	4	-	-	0	0	0	4	4	-	-	0	0	0
TEMPO STA	2	2	-	-	0	-	-	2	2	-	-	0	0	0	2	1	-	-	0	1	0
Brilliance Staph 24	2	2	-	-	0	-	-	2	2	-	-	0	0	0	2	2	-	-	0	0	0
Other	2	2	-	-	0	-	-	2	2	-	-	0	0	0	2	2	-	-	0	0	0
EASY Staph®	1	1	-	-	0	-	-	1	1	-	-	0	0	0	1	1	-	-	0	0	0



 $\textbf{Figure 6.} \ \ Results \ from \ analysis \ of \ coagulase-positive \ staphylococci.$ 

# Lactic acid bacteria

## Sample A

*C. maltaromaticum* was target organism. On MRS-aB, it forms typical shiny, white colonies. The strain is catalase-negative and Gram-positive.

In total, 46 participants reported results. Two low outliers were reported, as well as 25 false negative results.

Compared to other lactic acid bacteria, the strain of *C. maltaromaticum* has a higher sensitivity to low pH, as for example in the media MRS, MRS-S and Rogosa agar. Users of these media often reported false negative results.

## Sample B

No target organism was present in the sample. S. aureus may cause false positive results.

In total, 46 participants reported results. Of these, 21 reported a false positive result.

False positive results are likely due to detection of *S. aureus*. Strains of *S. aureus* have in previous PT rounds formed small colonies on MRS and MRS-aB. In the initial quality control at the Swedish Food Agency, small transparent colonies were observed on MRS-aB at a concentration of 3.9 log<sub>10</sub> cfu ml<sup>-1</sup>. In the subsequent confirmation these were catalase positive, and they were therefore not considered as lactic acid bacteria.

# Sample C

No target organism was present in the sample. S. aureus may cause false positive results.

In total, 47 participants reported results. Of these, 15 reported a false positive result. Similar to sample B, false positive results are likely due to detection of *S. aureus*.

#### General remarks

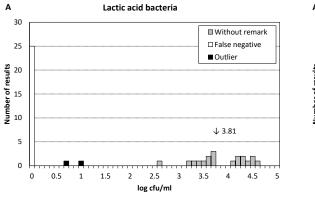
Most of the participants followed NMKL 140, either NMKL 140:2007 (39 %), or the withdrawn NMKL 140:1991 (6 %). In comparison, ISO 15214:1998 was used by 23 % of the participants. It was reviewed by ISO in 2021, and remains current. One participants followed ISO 7889/IDF 117:2003, which is a method for characteristic microorganisms in yoghurt at 37 °C.

Lactic acid bacteria constitute a heterogeneous group of microorganisms, and therefore have different optimal medium, pH and incubation conditions. Therefore, the choice of media will often have an impact on the results. For example, MRS-aB (pH 6.2) is a less selective medium that allows the growth of a more wide range of lactic acid bacteria. This may however also result in the appearance of more false positive colonies compared to the more acid media MRS and MRS-S (pH 5.7). Such differences between media and incubation conditions underline the importance of performing a confirmation test in uncertain cases, especially when using a less selective medium.

Both the ISO and the NMKL methods recommend confirmation of uncertain colonies. Lactic acid bacteria are Gram positive and normally catalase-negative. Confirmation of some kind was here performed by roughly half (55 %) of the participants. It often consisted of a catalase test or microscopy. For sample A, 8 false positive results were reported by the 23 participants that performed a catalase test, and 17 false positive results were reported by those that did not.

 Table 8. Results from analysis of lactic acid bacteria.

Medium			San	nple A						Sam	ıple I	3					Sam	ıple (			
iviedium	N	n	m <sub>PT</sub>	<b>S</b> PT	F	<	>	N	n	m <sub>PT</sub>	SPT	F	<	>	N	n	m <sub>PT</sub>	SPT	F	<	>
All results	46	19	3.81	0.68	25	2	0	46	25	-	-	21	-	-	47	32	-	-	15	-	-
MRS	25	8	4.03	0.68	17	0	0	25	12	-	-	13	-	-	26	15	-	-	11	-	-
MRS-aB	7	7	4.21	0.42	0	0	0	7	4	-	-	3	-	-	7	5	-	-	2	-	-
Rogosa	5	0	-	-	5	0	0	5	5	-	-	0	-	-	5	5	-	-	0	-	-
Petrifilm LAB	4	2	-	-	0	2	0	4	0	-	-	4	-	-	4	2	-	-	2	-	-
MRS-S	2	1	-	-	1	0	0	2	2	-	-	0	-	-	2	2	-	-	0	-	-
TEMPO LAB	2	1	-	-	1	0	0	2	2	-	-	0	-	-	2	2	-	-	0	-	-
Other	1	0	-	-	1	0	0	1	0	-	-	1	-	-	1	1	-	-	0	-	-



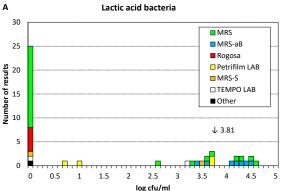


Figure 7. Results from analysis of lactic acid bacteria.

# Clostridium perfringens

# Sample A

No target organism was present in the sample. It did however contain *C. bifermentans*, which is false positive for the analysis. It forms black colonies on TSC, but can be distinguished from *C. perfringens* after confirmation, for example since *C. bifermentans* is motile.

In total, 51 participants reported results. Of these, 13 reported a false positive result.

The reported concentrations of the false positive results corresponded to that of C. bifermentans in the sample (2.3  $\log_{10}$  cfu ml<sup>-1</sup>).

## Sample B

*C. perfringens* was target organism. It forms black colonies on TSC. The strain is non-motile and ferments lactose. It grows on BA under anaerobic conditions, but not under aerobic conditions.

In total, 50 participants reported results. One low outlier was reported, as well as one false negative result.

## Sample C

No target organism was present in the sample.

In total, 51 participants reported results. No false positive results were reported.

#### General remarks

Most participants followed either NMKL 95:2009 (59 %) or ISO 7937:2004 (33 %). One participant followed the withdrawn NMKL 95:1997 and one participant analysed according to NMKL 56:2015 (Sulphite-reducing Clostridia). ISO 7937:2004 was reviewed by ISO in 2015 and remains current. It is however scheduled to be replaced by ISO 15213-2 ("Enumeration of *Clostridium perfringens* by colonycount technique"), which is currently under development. No obvious differences could be seen in the results from the different methods.

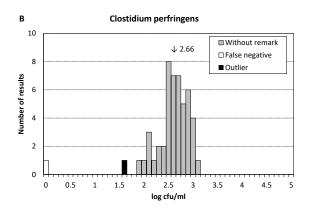
In line with ISO 7937:2004 and NMKL 95:2009, the majority (92 %) of the participants reported the use of TSC. On TSC, *C. perfringens* form black colonies after anaerobic incubation at 37 °C. The media ISA and SC were used by one each. Comparisons with TSC are difficult to make – due to the low number of users of these media – but TSC has by some been recommended for the analysis of *C. perfringens* in food samples [2, 3].

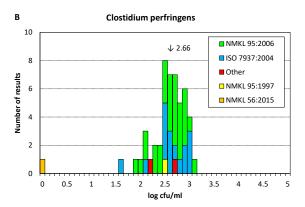
Two common methods for confirmation of *C. perfringens* are motility test and test for lactose fermentation; *C. perfringens* is non-motile and forms acid and gas as a consequence of lactose fermentation. *C. perfringens* can also be confirmed since it forms a double haemolytic zone upon anaerobic incubation on BA. In total, 90 % of the participants stated they performed some kind of confirmation. Common confirmation methods were motility test, test for lactose fermentation, test for haemolysis on BA, and test for absence of growth in aerobic conditions.

*C. perfringens* normally grows both at 37 °C and at 44 °C. Here, the majority of the participants (94 %) incubated at 37 °C, while only a few (6 %) incubated at 44 °C. It is therefore difficult to say if the choice of incubation temperature had an impact on the outcome, but it does not appear to be the case.

**Table 9.** Results from analysis of *Clostridium perfringens*.

Method			Sam	ple A	١					Sam	ple B						Samp	ole C			
ivietnod	N	n	m <sub>PT</sub>	S <sub>PT</sub>	F	<	>	N	n	m <sub>PT</sub>	S <sub>PT</sub>	F	<	>	N	n	m <sub>PT</sub>	S <sub>PT</sub>	F	<	>
All results	51	38	-	-	13	-	-	50	48	2.66	0.29	1	1	0	51	51	-	-	0	-	-
NMKL 95:2006	30	20	-	-	10	-	-	30	30	2.70	0.30	0	0	0	30	30	-	-	0	-	-
ISO 7937:2004	17	16	-	-	1	-	-	16	15	2.66	0.24	0	1	0	17	17	-	-	0	-	-
Other	2	1	-	-	1	-	-	2	2	-	-	0	0	0	2	2	-	-	0	-	-
NMKL 95:1997	1	1	-	-	0	-	-	1	1	-	-	0	0	0	1	1	-	-	0	-	-
NMKL 56:2015	1	0	-	-	1	-	-	1	0	-	-	1	0	0	1	1	-	-	0	-	-





**Figure 8.** Results from analysis of *Clostridium perfringens*.

# Anaerobic sulphite-reducing bacteria

# Sample A

C. bifermentans was target organism. It forms typical black colonies on ISA.

In total, 60 participants reported results. Two low and one high outliers were reported, as well as three false negative results.

## Sample B

*C. perfringens* was target organism. It forms black colonies on ISA. The black colour may be more distinct after 48 h incubation, compared to after 24 h incubation.

In total, 59 participants reported results. Four low and two high outliers were reported, as well as one false negative result.

# Sample C

No target organism was present in the sample.

In total, 62 participants reported results. Of these, one reported a false positive result.

### General remarks

The majority of the participants (63 %) followed a version of NMKL 56. However most followed the withdrawn NMKL 56:2008 (42 %) or even NMKL 56:1994 (3 %), and few (18 %) followed the current NMKL 56:2015. ISO 15213:2003 was used by 16 % of the participants. This was last reviewed by ISO in 2015, and remains current. It is however scheduled to be replaced by ISO 15213-1 ("Enumeration of sulphite-reducing *Clostridium* spp. by colony-count technique"), which is currently under development. Four participants followed ISO 7937:2004 ("Horizontal method for the enumeration of *Clostridium perfringens*"), which will be replaced by the future ISO 15213-2 ("Enumeration of *Clostridium perfringens* by colony-count technique").

Both NMKL 56:2015 and ISO 15213:2003 prescribe pour-plate methods with ISA, which was consequently the medium most frequently used by the participants (42 %). With ISA, black colonies (possibly surrounded by a black zone) are considered as sulphite-reducing bacteria. The black colour of the colonies comes from iron sulphide, which is formed as a precipitate of  $Fe^{3+}$  in the medium, and  $H_2S$  that is produced by the reduction of sulphite. Growth of anaerobic bacteria that only produce hydrogen (and not  $H_2S$ ) may sometimes result in a diffuse and unspecific blackening of the medium.

Other media that were used were TSC (24 %), SFP (19 %), PAB (6 %) and TS (3 %). These media are often used when identifying *C. perfringens*, and it should therefore be mentioned that for that purpose, colonies should be confirmed using the methods in for example NMKL 95.

**Table 10.** Results from analysis of anaerobic sulphite-reducing bacteria.

Method			Sam	ple A						Sam	ple B						Samp	ole C			
Method	N	n	m <sub>PT</sub>	<b>S</b> PT	F	<	>	N	n	m <sub>PT</sub>	SPT	F	<	>	N	n	m <sub>PT</sub>	S <sub>PT</sub>	F	<	>
All results	60	54	2.34	0.26	3	2	1	59	52	2.74	0.24	1	4	2	62	61	-	-	1	-	-
NMKL 56:2008	25	21	2.40	0.28	2	1	1	26	22	2.70	0.27	1	2	1	26	25	-	-	1	-	-
NMKL 56:2015	11	10	2.34	0.33	0	1	0	11	10	2.86	0.17	0	1	0	11	11	-	-	0	-	-
ISO 15213:2003	10	10	2.35	0.15	0	0	0	8	8	2.73	0.16	0	0	0	9	9	-	-	0	-	-
EB-SM-3013	6	6	2.26	0.17	0	0	0	6	6	2.77	0.10	0	0	0	6	6	-	-	0	-	-
ISO 7937:2004	4	3	-	-	1	0	0	4	4	-	-	0	0	0	4	4	-	-	0	-	-
Other	3	3	-	-	0	0	0	3	1	-	-	0	1	1	4	4	-	-	0	-	-
NMKL 56:1994	1	1	-	-	0	0	0	1	1	-	-	0	0	0	2	2	-	-	0	-	-

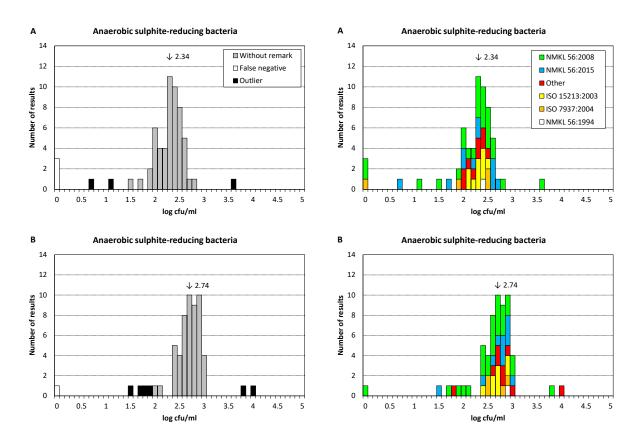


Figure 9. Results from analysis of anaerobic sulphite-reducing bacteria.

# Aerobic microorganisms in fish products

# Sample A

H. alvei and C. maltaromaticum were the main target organisms.

In total, 22 participants reported results. One high outlier was reported.

# Sample B

S. putrefaciens and S. aureus were the main target organisms.

During the initial quality control at the Swedish Food Agency the parameter did not fulfil the requirements for homogeneity, and a larger than usual distribution of the results can thus be expected. No values have therefore been considered as outliers.

In total, 22 participants reported results. All participants reported positive results. The majority of these were close to the assigned value (4.04 log<sub>10</sub> cfu ml<sup>-1</sup>) and only one of the results could be seen as a potential low outlier.

The results for sample B are not evaluated, and no z-scores are calculated for the analysis.

# Sample C

E. coli and S. aureus were the main target organisms.

In total, 22 participants reported results. Two low outliers were reported.

#### General remarks

Nineteen of the 22 participants followed the method for aerobic microorganisms and specific spoilage organisms in fish and fish products, NMKL 184:2006. This prescribes a pour-plate method with IA, which was consequently the medium most frequently used by the participants (86 %). Two and one participants followed ISO 4833-1:2013 and NMKL 86:2006, respectively, and thus incubated on PCA.

**Table 11.** Results from analysis of aerobic microorganisms in fish products.

Method			Sam	ple A						Sam	ple B*						Sam	ple C			
Wethod	N	n	m <sub>PT</sub>	S <sub>PT</sub>	F	<		N	n	m <sub>PT</sub>	S <sub>PT</sub>	F	<	>	N	n	m <sub>PT</sub>	S <sub>PT</sub>	F	<	>
All results	22	21	4.59	0.19	0	0	1	22	22	4.04*	0.24*	-	-	-	22	20	4.18	0.23	0	2	0
NMKL 184:2006	19	18	4.63	0.15	0	0	1	0	0	-	-	-	-	-	19	17	4.26	0.13	0	2	0
ISO 4833-1:2013	2	2	-	-	0	0	0	0	0	-	-	-	-	-	2	2	-	-	0	0	0
NMKL 86:2006	1	1	-	-	0	0	0	0	0	-	-	-	-	-	1	1	-	-	0	0	0

<sup>\*</sup> The sample is not evaluated. The values are shown only as an information to the participants.

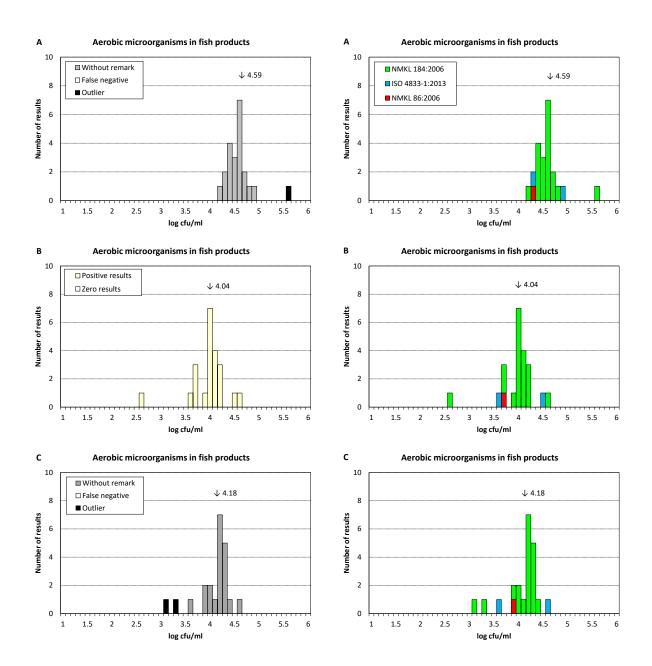


Figure 10. Results from analysis of aerobic microorganisms in fish products.

# H<sub>2</sub>S-reducing bacteria in fish products

# Sample A

H. alvei was target organism. It forms typical black colonies on IA.

In total, 19 participants reported results. All reported results were correct.

# Sample B

S. putrefaciens was target organism.

During the initial quality control at the Swedish Food Agency the parameter did not fulfil the requirements for homogeneity, and a larger than usual distribution of the results can thus be expected. No values have therefore been considered as outliers.

In total, 19 participants reported results. All of these were close to the assigned value (3.35 log<sub>10</sub> cfu ml<sup>-1</sup>), and no potential outliers could be identified.

The results for sample B are not evaluated, and no z-scores are calculated for the analysis.

# Sample C

No target organism was present in the sample.

In total, 19 participants reported results. All reported results were correct.

#### General remarks

All of the 19 participants followed the method for aerobic microorganisms and specific spoilage organisms in fish and fish products, NMKL 184:2006, and all of the participants incubated on IA. With IA, H<sub>2</sub>S-producing bacteria form black colonies.

Table 12. Results from analysis of H<sub>2</sub>S-reducing bacteria in fish products.

Method			Sam	ple A						Sam	ole B*						Samp	ole C			
Method	N	n	m <sub>PT</sub>	S <sub>PT</sub>	F	<	>	N	n	m <sub>PT</sub>	<b>S</b> PT	F	<	>	N	n	m <sub>PT</sub>	S <sub>PT</sub>	F	<	>
All results	19	19	4.23	0.13	0	0	0	19	19	3.35*	0.34*	-	-	-	19	19	-	-	0	-	-
NMKL 184:2006	19	19	4.26	0.12	0	0	0	0	0	-	-	-	-	-	19	19	-	-	0	-	-

<sup>\*</sup> The sample is not evaluated. The values are shown only as an information to the participants.

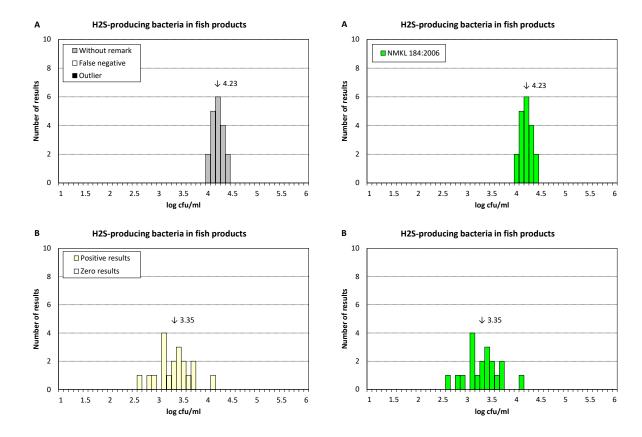


Figure 11. Results from analysis of H<sub>2</sub>S-reducing bacteria in fish products.

# Yeasts and moulds

# Sample A

No target organism was present for yeasts. *P. verrucosum* was target organism for moulds; it forms typical colonies on both DG18 and DRBC.

In total, 130 participants reported results for yeasts. Of these, 12 reported a false positive result.

In total, 130 participants reported results for moulds. Eight low and two high outliers were reported, as well as 11 false negative results.

Four participants reported false positive results for yeasts, and simultaneously false negative results for moulds.

# Sample B

*H. uvarum* was target organism for yeasts. It forms typical pink colonies on DRBC. No target organism was present for moulds.

In total, 131 participants reported results for yeasts. Five low and one high outliers were reported.

In total, 127 participants reported results for moulds. Three false positive results were reported.

# Sample C

S. cerevisiae was target organism for yeasts. C. cladosporioides was target organism for yeasts.

In total, 131 participants reported results for yeasts. Three low and five high outliers were reported, as well as nine false negative results.

In total, 129 participants reported results for moulds. Three low and four high outliers were reported, as well as nine false negative results.

Five participants reported false negative results for both yeasts and moulds.

#### General remarks

In essence, the same participants analysed both yeasts and moulds, and they generally reported identical methods for both parameters. In order, the most common methods were NMKL 98:2005, ISO 6611:2004/IDF 94:2004, 3M<sup>™</sup> Petrifilm<sup>™</sup> and ISO 21527-1:2008 / ISO 21527-2:2008. In addition, two participants followed ISO 7954:1987 ("General guidance for enumeration of yeasts and moulds"), which has been replaced by ISO 21527-1:2008 and ISO 21527-2:2008. Both ISO 21527-1:2008 and ISO 21527-2:2008 were last reviewed by ISO in 2018 and remain current.

With NMKL 98:2005, participants mainly used either DRBC and/or DG18. With ISO 6611:2004 / IDF 94:2004, which describes the enumeration of yeasts and moulds in milk and milk products, participants instead mainly used YGC. ISO 21527-1:2008 and ISO 21527-2:2008 stipulate the use of DRBC and

DG18, respectively. In general, DRBC is recommended for food with water activity  $a_w > 0.95$  (e.g. fresh fruit, vegetables, meat and milk products) while DG18 is recommended for food with  $a_w \le 0.95$  (e.g. dried fruit, dried meat, grains and nuts). Outliers and false results were for the most part evenly distributed between the main methods and media that were used.

Note: Two participants used TEMPO YM, which gives a <u>combined</u> value for yeasts and moulds. These results were excluded from calculations of the assigned value and standard deviations. Due to the low number of participants that use this methods, results from TEMPO YM need to be evaluated by the individual participants themselves.

**Table 13.** Results from analysis of yeasts.

Medium			Samp	le A						Samp	le B						Samp	le C			
Medium	N	n	m <sub>PT</sub>	S <sub>PT</sub>	F	<	>	N	n	m <sub>PT</sub>	S <sub>PT</sub>	F	<	>	N	n	m <sub>PT</sub>	S <sub>PT</sub>	F	<	>
All results	130	118	-	-	12	-	-	131	125	3.31	0.20	0	5	1	131	114	2.02	0.18	9	3	5
YGC	37	36	-	-	1	-	-	38	36	3.28	0.20	0	2	0	37	34	2.01	0.14	1	0	2
DG18 + DRBC	23	22	-	-	1	-	-	22	22	3.36	0.15	0	0	0	23	22	2.02	0.17	0	1	0
DG18	16	16	-	-	0	-	-	16	16	3.38	0.25	0	0	0	16	13	2.07	0.24	1	2	0
Other <sup>1</sup>	15	12	-	-	3	-	-	15	13	3.30	0.19	0	2	0	15	10	2.11	0.12	4	0	1
Petrifilm YM	14	11	-	-	3	-	-	14	13	3.31	0.19	0	0	1	14	11	2.00	0.17	2	0	1
DRBC	10	10	-	-	0	-	-	11	10	3.22	0.21	0	1	0	11	9	2.08	0.24	1	0	1
Petrifilm RYM	10	6	-	-	4	-	-	10	10	3.30	0.16	0	0	0	10	10	2.02	0.21	0	0	0
OGYE	5	5	-	-	0	-	-	5	5	3.42	0.22	0	0	0	5	5	1.98	0.08	0	0	0

For individual methods:  $m_{PT}$  = median value and  $s_{PT}$  = standard deviation for the particular method (outliers and false results excluded).

<sup>&</sup>lt;sup>1</sup> Other media includes e.g. Compact Dry YM, TEMPO YM, MEA, Saubouraud and PDA.

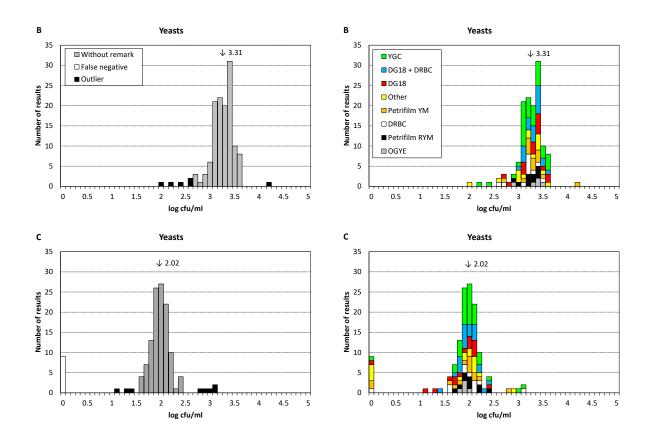


Figure 12. Results from analysis of yeasts.

**Table 14.** Results from analysis of moulds.

Medium			Samp	ole A					9	Sampl	е В						Samp	le C			
iviedium	N	n	m <sub>PT</sub>	SPT	F	<	>	N	n	m <sub>PT</sub>	SPT	F	<	>	N	n	m <sub>PT</sub>	SPT	F	<	>
All results	130	109	2.67	0.15	11	8	2	127	124	-	-	3	-	-	129	113	1.64	0.19	9	3	4
YGC	39	36	2.71	0.12	1	1	1	38	38	-	-	0	-	-	38	34	1.60	0.22	1	2	1
DG18 + DRBC	22	21	2.70	0.15	0	0	1	21	20	-	-	1	-	-	22	21	1.68	0.17	0	0	1
DG18	17	14	2.71	0.09	1	2	0	17	17	-	-	0	-	-	17	17	1.70	0.15	0	0	0
Petrifilm YM	15	10	2.69	0.18	2	3	0	15	15	-	-	0	-	-	15	13	1.71	0.19	1	0	1
Other <sup>1</sup>	13	8	2.73	0.16	3	2	0	13	12	-	-	1	-	-	13	7	1.65	0.19	4	1	1
DRBC	11	11	2.67	0.16	0	0	0	10	10	-	-	0	-	-	11	10	1.63	0.11	1	0	0
Petrifilm RYM	8	4	-	-	4	0	0	8	7	-	-	1	-	-	8	6	1.72	0.16	2	0	0
OGYE	5	5	2.74	0.21	0	0	0	5	5	-	-	0	-	-	5	5	1.67	0.26	0	0	0

For individual methods:  $m_{PT}$  = median value and  $s_{PT}$  = standard deviation for the particular method (outliers and false results excluded).

 $<sup>^{\</sup>rm 1}$  Other media includes e.g. Compact Dry YM, TEMPO YM, MEA, Saubouraud and PDA.

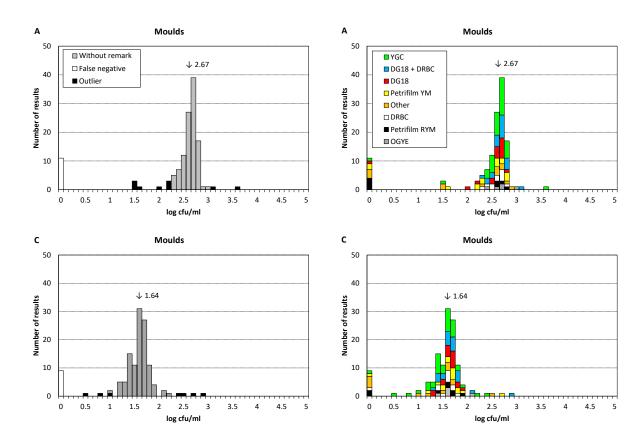


Figure 13. Results from analysis of moulds.

# Outcome of the results of individual participants - assessment

#### Reporting and evaluation of results

The results of all participants are listed in Annex 1, together with the minimum and maximum accepted values for each analytical parameter. Outliers and false results are highlighted in yellow and red, respectively, with bold font.

Participants are not grouped or ranked based on their results. The performance of an individual participant can be broadly assessed by the numbers of outliers and false results, and by the z-scores.

Information on the results processing and recommendations for follow-up work are given in the Scheme Protocol [4].

Samples for follow-up analyses can be ordered at: www.livsmedelsverket.se/en/PT-extra

#### Box plots and numbers of deviating results for each participant

Box plots are based on the z-scores listed in Annex 2, and give a comprehensive view of the performance of each participant. The range of z-scores is indicated by the size of the box and, for most participants, by lines and/or circles above and beneath the box. A small range of values, centred around zero, indicates that the results of the individual participant are in general close to  $m_{PT}$  for the different analyses. For each participant, the number of false results and outliers are also listed in the tables below the box plots.

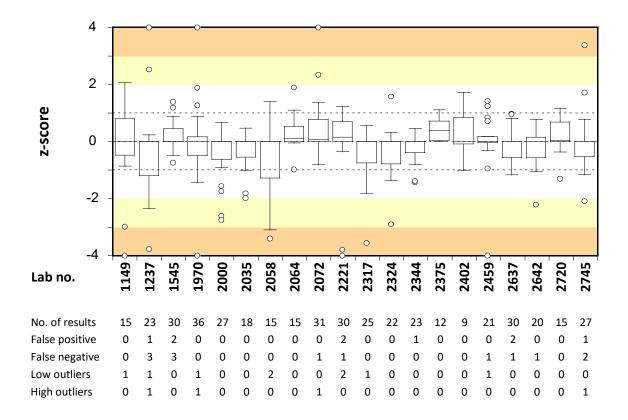
Outliers are included in the figures after being calculated to z-scores in the same way as for other results. Correct results for qualitative analyses and correct negative results for quantitative analyses without target organism are given a z-score of 0. False results do not generate any z-scores, and are not included in "No. of results".

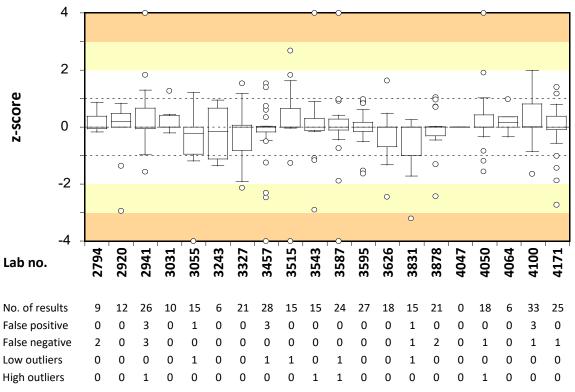
The participant's median value is illustrated by a horizontal line in the box. Each box includes 50 % of a participant's results (25 % of the results above the median and 25 % of the results below the median). The remaining 50 % are illustrated by lines and circles outside the box. A circle is for technical reasons shown in the plot when a value deviates to certain degree\* from the other values. This does not by itself indicate that the value is an outlier.

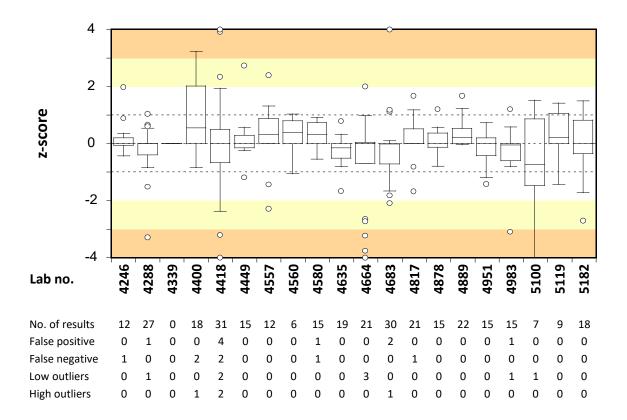
Z-scores >+4 and <-4 are positioned at +4 and -4, respectively, in the plot. The background is divided by lines and shaded fields to simplify identifying the range in which the results are located.

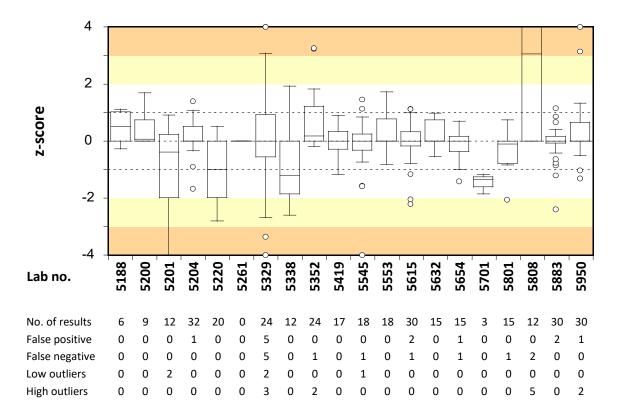
```
* < [lowest value in the box -1.5 \times (highest value in the box – lowest value in the box)] 
or
```

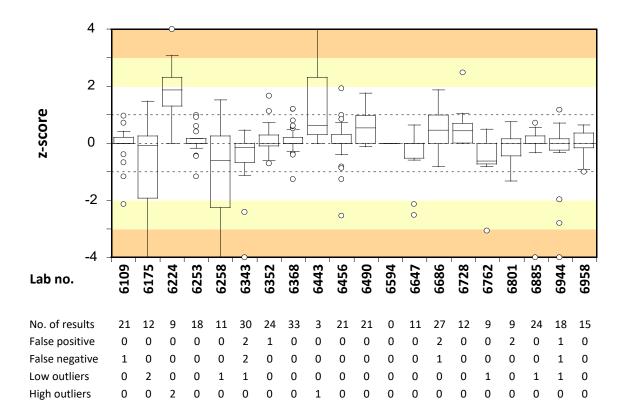
> [highest value in the box + 1.5  $\times$  (highest value in the box – lowest value in the box)].

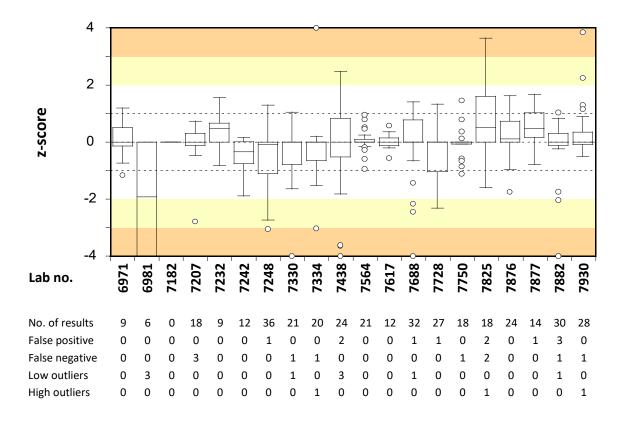


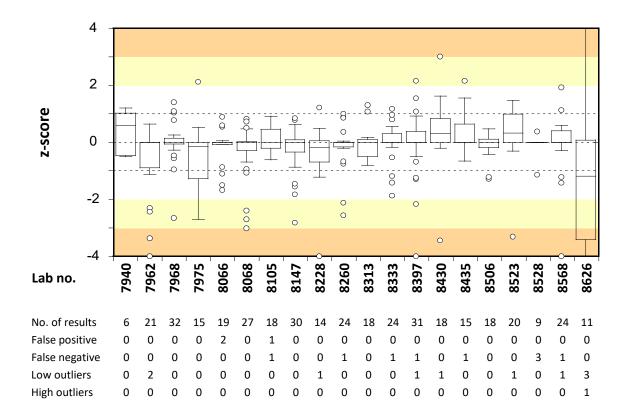


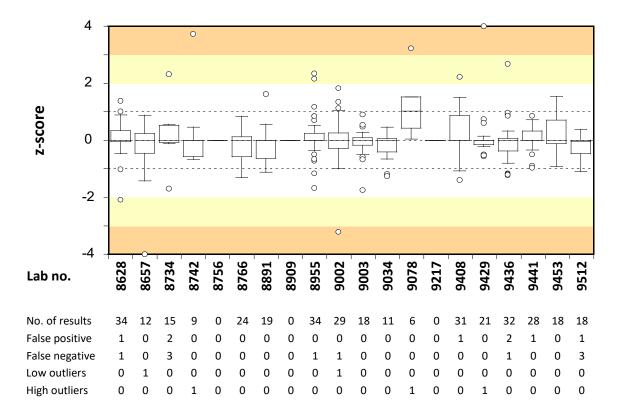


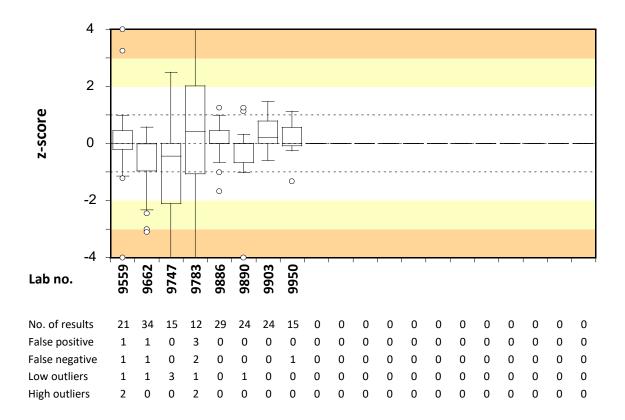












## Test material and quality control

#### Test material

Each participant received three samples with freeze-dried microorganisms, designated A–C. The test material was freeze-dried in 0.5 ml portions in glass vials, as described by Peterz and Steneryd [5]. Before analysing the samples, the contents of each vial should be reconstituted in 254 ml of sterile diluent. The microorganism content of the samples and the concentrations determined at the Swedish Food Agency are listed in the table below.

**Table 15.** Microorganisms and approximate concentrations in the samples.

			Strair	1	
Sample	Microorganism	SLV no.1	Origin	Reference <sup>2</sup>	log <sub>10</sub> cfu ml <sup>-1</sup>
А	Bacillus cereus	SLV-517	Cream sauce	CCUG 44739	3.5
	Clostridium bifermentans	SLV-009	Fish	CCUG 43592	2.3
	Carnobacterium maltaromaticum	SLV-519	Smoked salmon	CCUG 46539	4.2
	Hafnia alvei	SLV-015	Minced meat	CCUG 45642	4.3
	Penicillium verrucosum	SLV-544	Soil	CBS 112488	2.9
В	Clostridium perfringens	SLV-442	-	CCUG 43593	2.9
	Hanseniaspora uvarum	SLV-555	-	-	3.5
	Staphylococcus aureus	SLV-539	Mastitis	-	4.1
	Serratia marcescens	SLV-040	Pond water	-	2.7
	Shewanella putrefaciens	SLV-520	-	CCUG 46538	3.8
С	Cladosporium cladosporioides	SLV-488	Meat	CBS 812.96	2.0
	Escherichia coli	SLV-524	Chicken	CCUG 47554	3.9
	Staphylococcus aureus	SLV-350	-	CCUG 45099	4.2
	Saccharomyces cerevisiae	SLV-375	Baker's yeast	-	2.4

 $<sup>^{\</sup>rm 1}$  Internal strain identification no. at the Swedish Food Agency

<sup>&</sup>lt;sup>2</sup> Culture collection. ATCC: American Type Culture Collection, CBS: Centraalbureau voor Schimmelcultures (Westerdijk Institute), CCUG: Culture Collection University of Gothenburg, Sweden; Fohm: Public Health Agency of Sweden.

#### Quality control of the samples

In order to allow comparison of the freeze-dried samples, it is essential to have aliquots of homogeneous test material and equal volume in all vials. Quality control is performed on 10 randomly chosen vials in conjunction with manufacturing of the samples or on 5 vials if an "old" sample mixture was used and the last quality control was performed more than 6 months ago. Homogeneity of a test material is approved if, for each analysis, the values obtained for the test for "Index of dispersion" between vials  $(I_2)$  and the test for reproducibility (T) do not simultaneously exceed 2.0 and 2.6, respectively. (For definitions of  $I_2$ , and T, see references [6] and [7] respectively.)

**Table 16.** Concentration mean (m),  $I_2$  and T values from the quality control of the samples; m is expressed in  $log_{10}$  cfu (colony forming units) per ml of sample.

Analysis and marked		A <sup>1</sup>	_	_	B <sup>1</sup>	_	_	C <sup>2</sup>	
Analysis and method	m	I <sub>2</sub>	T	m	I <sub>2</sub>	T	m	I <sub>2</sub>	T
Aerobic microorganisms, 30 °C NMKL method no. 86:2013	4.77	1.29	1.34	4.64	0.91	1.48	4.37	1.33	1.60
Psychrotrophic microorganisms NMKL method no. 86:2013	4.70	0.36	1.19	4.38	0.98	1.52	-	-	-
Enterobacteriaceae NMKL method no. 144:2005	4.18	0.81	1.23	2.69	1.28	1.37	3.85	1.54	1.49
Escherichia coli NMKL method no. 125:2005	-	-	-	-	-	-	3.98	1.98	1.50
Presumptive <i>Bacillus cereus</i> NMKL method no. 67:2021	3.52	0.32	1.21	-	-	-	-	-	-
Coagulase-positive staphylococci NMKL method no. 66:2009	-	-	-	4.07	0.82	1.26	4.18	0.87	1.23
Lactic acid bacteria NMKL method no. 140:2007	4.22	3.40	2.13	-	-	-	-	-	-
Clostridium perfringens NMKL method no. 95:2009	-	-	-	2.84	0.97	1.27	-	-	-
Anaerobic sulphite-reducing bacteria NMKL method no. 56:2015	2.30	0.67	1.44	2.96	2.81	1.40	-	-	-
Aerobic microorganisms in fish products NMKL method no. 184:2006	4.67	0.55	1.23	4.24	5.53	1.42	4.01	1.88	2.31
H2S-producing bacteria in fish products NMKL method no. 184:2006	4.31	1.88	1.85	3.78	4.33	1.73	-	-	-
Yeasts NMKL method no. 98:2005 (DRBC)	-	-	-	3.47	2.10	1.62	2.37	2.01	1.78
Moulds NMKL method no. 98:2005 (DRBC)	2.93	4.53	1.88	-	-	-	1.96	1.56	2.65

<sup>-</sup> No target organism or no value

 $<sup>^{1}</sup>$  n = 5 vials analysed in duplicate

<sup>&</sup>lt;sup>2</sup> n = 10 vials analysed in duplicate

### References

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**Annex 1.** Results of the participating laboratories

Lab		Aerobio oorgan			hrotro		Enterd	bacter	iaceae	Esch	erichia	coli		sumpti			ulase-po		Lactic	acid ba	acteria		ostridiur			obic sulp		A microo	erobic rganis			-produc			Yeasts		ı	Moulds	
no.		30 °C		micro	organ	isms							Васі	llus cer	eus	Sta	phyloc	DCCI				pε	erfringen	15	reau	ing bact	eria	fish	produ	cts	р	roduct	5						
	Α	В	С	Α	В	С	Α	В	С	Α	В	С	Α	В	С	Α	В	С	Α	В	С	Α	В	С	Α	В	С	Α	В	С	Α	В	С	Α	В	С	Α	В	C
1149	3.65	3.7	4.23	-	-	-	4.18	2.78	4	<1	<1	3.7	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	- 1	-	-	-	-	-	0	3.2	2.4	2.78	0	1.85
1237	5	4	4.15	-	-	-	4.15	0	3.48	0	0	3.32	-	-	-	0	3.85	4.04	0	3.15	0	-		-	2.4	2.4	0		2.65	3.3	-	-	-	0	3.15	0		-	-
1545	4.79	4.1	4.34		-	-	4.34	2.72	3.8	<1	<1	3.78	3.36	<1	<1	<1	4	4.2	<1	3.11	2.11	<0	2.79	<0	2.3	2.85	<0		-	-		-	-	<1	3.21	<1	2.56	<1	<1
1970		3.99	4.18	3.34	3.3	0	3.93	2.43	3.63	0	0	3.88	3.43	0	0	0	3.91	4.26	3.79	0	0	0	2.91	0	3.68	2.76	0	4.46	4	4.23	4.15	2.95	0	0		2.08	2.67	0	1.76
2000 2035	4.34	3.85	4.04	-	-	-	3.91 3.85	2.52 2.49	3.71	0	0	3.6	3.18	0 <1	0 <1	0	3.93	4.11	4.26	0	0	0 <1	2.66	0	2.08	2.63	<1	- 1	-	-	-	-	-	0 <1		1.99	2.68	0 <1	1.61 1.7
2058	- 151	3.65	4.15	-			3.03	2.49	3.54	0	0	3.53	5.2	<1	<1							0	2.57 2.5	<1 0	2.08	2.03	<1							0	3.34 3.4	2.11	2.62 <b>2.2</b>	0	1.7
2064	4.51	4.09	4.13	_			4.13	2.75	4 07	-	-	-	3.52	0	0							-	-	-	_	-								0	3.43		2.67	0	1.46
2072	4.72	4.2	4.38	4.71	4.11	3.96	4.2	2.65	3.91	<1	<1	3.88	3.52	<1	<1	<1	-	4.3	<1	<1	<1	<1	2.54	<1	2.54	2.54	<1		_	-				<1		3.15	2.84	<1	1.8
2221	4.81	4.16	4.34	-	-	-	4.16	2.67	3.88	<1	<1	3.71	3.43	<1	<1	<1	4.05	2.24	<1	3.48	2.1	<1	2.92	<1	2.47	1.81	<0	- 1	-	-	-	-	-	<1	3.55	2.16	2.85	<1	1.73
2317	4.69	3.97	4.3	-	-	-	4.08	2.54	3.88	0	0	3.85	2.95	0	0	0	3.83	4.11	-	-	-	0	1.64	0	2.11	-	-	- 1	-	-	-	-	-	0	3.23	1.97	2.41	0	1.36
2324	4.56	3.95	4.18	-	-	-	3.97	2.12	3.81	<1	<1	3.79	3.3	<1	<1	<1	3.91	4.08	-	-	-	-	-	-	>1	>1	<1	- 1	-	-	-	-	-	<1	3.62	1.88	2.71	<1	1.7
2344	4.49	4.04	4.28	-	-	-	4.02	2.54	3.74	0	0	3.51	3.23	-	0	0	3.99	4.11	-	-	2.15	0	-	0	2.37	-	0	- 1	-	-	-	-	-	0	-	1.97	2.72	-	1.65
2375		4.06	4.33	-	-	-		2.76	3.86	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	- 1	-	-	-	-	-	0	3.38	2.04	2.78	0	1.76
2402		4.05	4.49	-	-	-	4.06	2.45	3.92	0	0	3.98	-	-	-	-	-		-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-		-	-	-	-
2459	4.64	4.08	4.46	-	-	-	4.26	2.84	3.82	<1	<1	3.72	<1	<1	<1	<1	3.94	4.16	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	<1	3.12		2.7	<1	1.67
2637 2642	4.7 4.7	3.98 4.08	4.32 4.3	4.48	3.14	3.53	4.23 4.25	2.57	3.64 3.82	<1 0	<1 0	3.72 3.91	<1	<1	<1	<1 0	3.84 4	4.08 4.11	4.36	2.79	2.52	<1	2.94	<1	2.04	2.97	<1	- 1	-	-	-	-	-	<1 0		1.86 1.83	2.52	<1 0	1.56 1.23
2720	4.7	4.08	4.3	4.46	5.14	3.33	4.25	2.67	3.9	-	-	5.91	3.26	<1	<1	-	4	4.11								-								<1		2.03	2.8	<1	1.23
2745		3.94	4.27	_	_		4.18	2.76	3.68	0	0	3.75	2.9	0	0	0	4.11	4.13	0	0	0	0	2.59	0	_	_	_			_	.			2.51			0	0	1.51
2794	4.66	4.04	4.32	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-		_	-				0		2.18	0	0	0
2920	4.7	4.11	4.18	-	-	-	4.2	2.77	3.83	0	0	3.88	-	-	-	-	-	-	-	-	-	-	-	-	2.52	2.02	0	- 1	-	-	-	-	-	-		-	-	- '	-
2941	4.7	4.16	4.27	-	-	-	4.12	2.54	3.66	0	0	3.78	3.53	0	0	0	0	4.14	0	3.87	1.45	-	-	-	-	0	2.79	- 1	-	-	-	-	-	0	3.67	3.17	2.87	0	1.35
3031	-	-	-	-	-	-	-	-	-	0	0	3.8		-	-	0	3.94	4.08	-	-	-	-	-	-	-	-	-	4.67	4.05	4.28	4.4	3.18	0	-	-	-	-	-	-
3055	4.67	3.96	4.2	-	-	-		2.46		-	-	-	3.54	2.08	0	-	-	-	-	-	-	-	-	-	-	-	-	- 1	-	-	-	-	-	0	2.46	1.95	2.49	0	1.48
3243	4.53	4.14	4.18	-	-	-	4.12	2.79	3.77	-	-	-	-	-	-	-		-	-	-	-	-	-	-	-	-	-	- 1	-	-	-	-	-	-	-	-	-	-	-
3327	4.57	3.83	4.26	-	-	-	3.93	2.53	3.71	0	0	3.36	-	-	-	0	4	4.23		17	- 0	-	1.05	-	2.11	2.94	0	1.6	-	4.25	4 20	- 20	-	0	3.32		2.85	0	1.78
3457 3515	4.66 4.91	4.12	4.27 4.48	-			4.36 4.18	2.63	3.79 3.62	0	0	3.94	3.33	0	0	0	3.93 4.2	4.15 <b>3.68</b>	1	1.7	-		1.95	-		-		4.6	4.13	4.35	4.28	3.28	0	2.49	3.28	2.28	2.32	0	1.41
3543	4.79	4.65	4.40	_			3.98	2.62	3.88	-	-	3.54	3.08	<1	<1	<1	3.95	4.09					-	-	1.59	2.7	<1							2.78	3.63	2.21			
3587	4.66	4.07	4.35	-	-		4.27	2.69	3.94	0	0	4.86	3.22	0	0	0	2.89	4.09	-	-	-	-	-	-	2.29	2.79	0		-	-				0	3.3		2.56	0	1.29
3595	4.7	4.07	4.3	-	-	-	4.12	2.78	3.82	<1	<1	3.72	3.01	<1	<1	<1	3.97	4.15	-	-	-	<1	2.94	<1	1.92	2.94	<1	-	-	-	-	-	-	<1	3.21	1.93	2.7	<1	1.59
3626	4.7	3.9	4.2	-	-	-	4.1	2.2	3.7	0	0	4.1	3.1	0	0	0	3.9	4.1	-	-	-	0	2.8	0	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
3831	4.69	4.06	4.22	-	-	-	-	-	-	0	0	3.15	-	-	-	-	-	-	0	2.24	0	-	-	-	-	-	-	- 1	-	-	-	-	-	0			2.41	0	1.43
3878	4.767	3.903	4.422	-	-	-	4.279	2.595	3.767	<1	<1	3.92	3.217	0	0	<1	3.716	4.063	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	<1	3.216	2.027	<1	0	0
4047 4050	- 4.72	- 4.29	- 4.16	-	-	-	4.14	- 2.7	3.75	-	-	-	4 24	-	-	-	-	-	0	-	- 0	-	-	-	-	-	-	- 1	-	-	-	-	-	- 0	- 3.51	1.07	- 2.70	-	1.42
4064	4.72	4.29	4.16	-			4.14	2.7	3.85				4.24	-	-					-	-					-								-	3.31	1.6/	2.79	-	1.42
4100	4.82	4.06	4.44	4.62	3.32	<1	4.28	2.79	3.8	<1	<1	3.61	3.26	<1	<1	-			<1	3.2	2.06	1.74	2.19	- <1	2.4	2.94	<1	4.73	4.23	4.33	4.34	3.77	<1	<1	3.48	2	2.72	- <1	1.75
4171	4.68	4.23	4.26	-	-	-	4.11	2.83	3.41	<0,60	<0,60	3.41	3.41	<0	<0	<1	>1,00	>1,00	<1	<1	<1	-	-	-	2.08	2.91	<0	-	-	-	-	-	-	<0		1.76	2.71	<0	1.79
4246	4.68	4.3	4.3	-	-	-	4.12	2.78	3.8	0	0	3.85	-	-	-	0	0	4.06	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
4288	4.48	4.01	4.28	-	-	-	4.08	2.72	3.33	0	0	3.91	3.51	0	0	0	3.92	4.15	-	-	-	2.18	2.42	0	2.17	2.69	0	-	-	-	-	-	-	0	3.3	1.92	2.74	0	1.7
4339	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-		-	-	-	-	-	-	-	-	-	-	-
4400	4.85	4.34	4.56	-	-	-	4.23	2.95	4.26	<1	<1	4.07	3.73	<1	<1	-	-	-	-	-	-	-		-	-	-	-	1	-	-	-	-	-	<1		2.11	<1	<1	<1
4418	5.18	4.12	3.45	4.64	3.54	4.3	4.11	2.51	3.57	0	0	3.8	0	3.38	3.62	0	4.13	4.3	-	-	-	0	2.5	0	0	2.5	0	4.95	4.57	4.6	-	-	-	2.95			2.39	1.41	1.2
4449	4.66 4.7	4.08 4.35	4.34 4.45	-	-	-	4.11 4.28	2.42	3.85 3.91	0	-	3.87	3.26	0	0	0	3.81	-	-	-	-	-	-	-	-	-	-	- 1	-	-	-	-	-	0	3.42	1.98	2.74	0	2.15
4557 4560	4.7	4.33	4.45		-		4.26	2.76	5.91	-	0	3.87				-	5.61	3.9																<0,5	3.46	1 83	2 83	- <5	1.79
4580	4.72	4.17	4.39		_		4.25	2.77	3.86	0	0	3.74	-	_	_	_	-		-	-	-	-		-			-		-	-	-		_	2.79		2.08	0	0	1.54
4635	4.46	3.98	4.3	-	-	-	4.07	2.59	3.72	-	-	-	3.46	<1	<1	<1	3.87	4.08	-	-	-	-		-	>1	>1	<1		-	-	-	-	-	<1	3.37	1.9	2.65	<1	1.65
4664	4.74	4.12	4.05	-	-	-	4.28	2.76	3.41	<1,26	<1,26	3.04	-	-	-	<1	3.92	3.75	-	-	-	-		-	2.86	2.64	0		-	-		-	-	0		1.43	2.64	0	1.65
4683	4.59	3.9	4.15	-	-	-	4.3	2.62	3.78	<1	<1	3.63	2.9	<1	<1	<1	3.9	3.94	2.67	3.78	2.3	<1	3	<1	2.25	4	<1		-	-	-	-	-	<1	3.2	2.04	2.65	<1	1.47
4817	4.57	4.1	4.33	-	-	-	-	-	-	<1	<1	3.45	<1	<1	<1	<1	4.02	4.2	-	-	-	<1	3	<1	-	-	-		-	-	-	-	-	<1	3.64	2.07	2.74	<1	1.75
4878	4.63	4.03	4.27	-	-	-	4.2	2.65	3.82	-	-	-	3.54	0	0	-	-	-	-	-	-	-	-	-	-	-	-		-	-	-	-	-	0	3.15	2.08	2.76	0	1.73
4889	4.72	4.11	4.34	-	-	-	4.26	2.62	4.04	0	0	3.93	3.3	0	0	0	4.04	4.11	-	-	-	-	-	-	2.66	2.81	0		4.15	4.3	4.26	3.4	0	-	-	-	-	-	-
4951 4983	4.71 4.71	3.89	4.39	-	-	-	4.2	2.65	3.7 3.79	<1	<1	3.92	2 5 4	-	-	-	-	-	-	-	-	-	-	-	-	-	-	- 1	-	-	-		-	<1	3.07	2	2.65 <b>2.2</b>	<1	1.5 1.53
4983	4./1	4.02	4.27		-	-	4.02	2.62	5.79			-	3.54	0	0	-	-	-	-		-		-	-				-		-	-	-	-	2.6	3.17	2.13	2.2	0	1.55

**Annex 1.** Results of the participating laboratories

Lab	mic	Aerob			:hrotro		Enterd	obacteri	iaceae	Esch	erichia	coli		sumpti			ılase-po		Lactic	acid ba	cteria		ostridiun			obic sul	-		Aerobic organis			-produc			Yeasts			Moulds	
no.		30 °C		micro	oorgan	isms							Bacii	llus cer	eus	sta	phyloc	occi				pe	rfringen	s	redu	cing bac	teria	fish	n produ	ıcts	p	roducts	5						
	Α	В	С	Α	В	С	Α	В	С	Α	В	С	Α	В	С	Α	В	С	Α	В	С	Α	В	С	Α	В	С	Α	В	С	Α	В	С	Α	В	С	Α	В	С
5100			4.47	-	-	-	-		-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-		-	-	-	-	-	-	-	-	3.16	2.2	2.79	-	1.48
5119			4.46	-	-	-		2.81		<1	<1	4		-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	<2	3	2	<2	3	2
5182		2 4.018	3 4.415	4.75	-	-	4.274	2.398	4.014	<1	<1	3.927	2.781	<1	<1	-	-	-	-	-	-	-	-	-	-	-	-	4.00	-	-	4.20	-	-	<1	3.441	1.707	2.797	<1	1.38
5188 5200		-	-	4.75	3.34	<2	-	-	-	<0,48	<0,48	4.04	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	4.69	4.08	4.3	4.38	3.41	<2	0	3.643	2 060	2 705	- <1	1.653
5200		4.17	4.3	-			4.23		3.73			3.56				-	2.94	3.85					-											-	2.2	1.97	2.63	-	1.45
5204				4.32	3.95	4.15	4.34	2.74	3.92	<1	<1	3.79	3.35	<1	<1	<2	3.86	4.07	3.59	<2	<2	2.36	2.92	<1	2.58	3	<1	-	-		-	-	-	<1		2.06	2.73	<1	1.7
5220	4.42	3.83	4.25	-	-	-	3.85	2.43	3.42	0	0	3.4	-	-	-	0	3.97	4.02	-	-	-	-	-	-	-	-	-	4.37	3.68	3.65	-	-	-	0	2.75	1.6	2.75	0	1.48
5261		-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
5329				-	-	-				3.65	<1	<1	<1	2.73	3.44	4.21	3.99	<1	<1	<1	<1	-	-	-	-	-	-	-	-	-	-	-	-	2.15	3.49	<1	1.54	<1	2.52
5338				-	-	-	3.91	2.96	3.68	-	-	-	-	-		-	-	-	-	-	-	-	-	-	-	-	-	-	-		-	-	-	0	2.98	1.93	2.41	0	1.68
5352				-	-	- 2.00	4.32	0	4.26	0	0 <1	4.01 3.76	3.3	0	0	0	4.11	4.38	-	-	-	0	3.14	0	-	-	-	-	-		-	-		0	3.67	1.99	2.7	0	1.8
5419 5549				4.64	3.89	3.86			3.65 0	<1	<1	3.76	3.47	0	0	0	3.95	4.07					-	-										<1 0		2.17	2.74	<1 0	1.64 1.91
5553		0.07		_				2.51		<1	<1	3.62	3.42	<1	<1	<1	4.02	4.23		-		<1	3	<1		-					-			-	5.10	-	-	-	-
5615				-	-	-	4.26	2.49	3.96	<1	<1	4	3.08	<0	<0	<1	3.91	4.08	<1	2.95	2.04	<0	2.65	<0	2.46	2.68	<0	-	-		-	-	-	<0	3.28	2.08	2.36	<0	1.23
5632	4.8	4	4.38	-	-	-	-	-	-	<1	<1	3.9	3.2	<2	<2	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	<1	3.5	2.2	2.7	<1	1.8
5654	4.67	4.08	4.28	-	-	-	4.2	2.57	3.89	-	-	-	0	0	1.11	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	0	3.22	2.15	2.52	0	1.38
5701				-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
5801				-	-	-	4.08	2.49	3.77	-	-	-	<1	<1	<1	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	<1	3.14			<1	1.6
5808 5883			4.53 4.23	-	-	-	4.13	2.74	-	- <1	1	2.00	<b>4.24</b> 3.34	<1 0	<1 0	- <1	3.94	3.89	2.75	2.96	2.48	- 0	2.44	-	2.64	2.44	- 0	-	-		-	-	-	<1 0	4.21	2.88	<1	<1 0	<1 1.67
5950				4.75	3.44	0	4.13	2.74	3.8 3.82	<1	<1 0	3.86 3.97	3.41	0	0	<1	3.94	3.89	3.75	0	2.48	-	2.44	-	2.64	2.44	0	4.4	4.69	4.23	4.32	4.14	0	0	3.18 3.27	2.18	2.72 <b>3.15</b>	1.95	2.91
6109		4.11		-	-	-	-	2.01	-	<0,6	<0,6	3.36	3.23	0	0	-	-		<1	<1	<1	-		-	2.04	2.82	<1	-	-	-	-	-	-	0		2.04	2.57	0	1.82
6175		4.1	4.3	-	-	-	4.1	2.2	3.6	-	-	-	-	-		-	-	-	-	-	-	-	-	-	-	-	-	-	-		-	-	-	0	3.6	1.3	2	0	1.7
6224	4.85	4.69	4.55	-	-	-	4.41	2.97	4.24	-	-	-	3.56	<2	<2	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
6253	4.68	4.08	4.32	-	-	-	-	-	-	-	-	-	3.08	<1	<1	<1	4.04	4.15	-	-	-	-	-	-	2.45	2.63	<1	-	-	-	-	-	-	<1	3.49	1.94	2.69	<1	1.61
6258				4.13	2.32	3.48	-		-	-		-		-	-	-		-	-	-	-	-		-		-	-	-	-	-	-	-	-	0	2.9	1.79	2.58	0	0.54
6343				-	-		4.04 4.08	2.58	3.74 3.75	0 <1	0 <1	3.87 3.75	3.11	0 <1	0 <1	0 <1	3.87	4 4.1	0	2.63	0	2.13	2.6 2.65	0 <1	2.39	1.76	0	-	-		-	-	-	0 <1	2.83	0	2.6	0	1.51
6352 6368				4.67	4.03	3.81		2.52		<1	V	3.75	3.17	0 <1	<1	<1 0	3.91 4.06	4.1	4.11	0	0	2.36	2.65	<1	2.39	2.88	0	4.54	4.15	4.29	4.26	3.52	0	<1 0		2.23		<1 0	1.95
6443		-		-	03	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	_	-	2.5	3.8	0	-	-	-	-	-	-	-	-	-	-	-	-
6456		4.07	4.19	-	-	-	-	-	-	-	-	-	2.81	<1	<1	<1	4.04	4.16	-	-	-	<1	2.75	<1	2.56	2.76	<1	-	-	-	-	-	-	<1	3.69	1.95	2.73	<1	1.48
6490	4.8	4.08	4.45	-	-	-	4.12	2.7	3.87	-	-	-	3.62	0	0	0	4.03	4.25	-	-	-	-	-	-	2.48	2.95	0	-	-	-	-	-	-	0	3.43	2.28	2.88	0	1.8
6594	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
6647					-	-	-		-		<0,48	>3,04	-	-	-	-	-	-	-	-	-	-		-	-	-	-	-			-		-	0	3.19		2.63	0	1.76
6686			4.38	4.78	4.04	4.32	4.41	2.85		<1	<1	3.81	-	-	-	<1	4	4.15	-	-	-	2.95	2.88	<1	-	-	-	4.75	4.2	4.08	4.3	3.15	<1	2.62	3.38	2.04	<1	<1	1.49
6728 6762							4.22 4.04	2.77 2.5	3.85 3.71	0 <1	0 <1	3.79 3.62				-	4	4.1																					
6801			-			-	-	-	-	-	-	-	-	-	-	<1	3.87	4.12			_	_			-		_		-		_	-		2.68	3.29	1.78	2.68	3.29	1.78
6885		4.11	4.33	-	-	-	4.09	2.66	3.87	0	0	3.89	3.37	0	0	0	3.98	4.16	-	-	-	-	-	-	1.18	2.74	0	-	-	-	-	-	-	0	3.26	2	2.67	0	1.67
6944	4.71	4.04	4.32	-	-	-	-	-	-	0	0	3.92	-	-	-	0	3.76	4.07	0.78	0.9	0	-	-	-	-	-	-	-	-	-	-	-	-	0	2.75	0	2.85	0	1.73
6958				-	-	-		2.72		-	-	-	3.27	0	0	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	0	3.11	2.01	2.65	0	1.55
6971		3.92	4.41	-	-	-	4.03	2.67	3.97	-	-	-	3.28	0	0	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	
6981		-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	0	2.06	1.91	1.53	0	1.04
7182 7207		4.07	4.39				4.07	2.14	3 76	-			3.41	- <1	<1	-			<1	- <1	- <1		-	-										- <1	3.26	<1	2.78	- <1	<1
7232					-			-	3.70				-	-	-	-			-1	-	-			-		-	-	_			_	-		0	3.4	2.2	2.78	0	1.73
7242				-	-	-	4.16	2.63	3.77	-	-		-	-		-	-			-		-	-	-	-						-		-	0	3.18	1.86	2.58	0	1.29
7248				4.53	3.25	0	3.76			0	0	3.88	3.26	0	0	0	3.72	4.03	4.59	3.79	0	0	2.52	0	2.04	2.48	0	4.56	3.96	4.16	4.01	3.63	0	0	3.46	1.76	2.87	0	1.68
7330				-	-	-	4.097	2.37	3.95	<1	<1	3.457	3.42	<1	<1	<1	<1	3.176	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	<1	3.041	2.176	2.829	<1	1.6
7334		4.02		-	-	-	-	-	-	0	0	>1	3.17	0	0	0	3.89	4.03	-	-	-	-	-	-	0	2.54	0	-	-	-	-	-	-	0	3.26	3.08	2.44	0	1.08
7438		4.18	4.4	-	-	-	4.08	2.69	3.28	3.93	<1	3.07	-	-	-	<1	4	3.56	-	-	-	2.3	2.51	<1	2.32	2.46	<1	-	-	-	-	-	-	<1	3.54	2.4	2.83	<1	2.1
7564 7617		4.08	- 4.26	-	-	-	4.11	- 2.62	- 3.79	<1 <1	<1 <1	3.88 3.85	3.46	<1	<1	<1 <1	3.99 4	4.18 4.08	-		-	<1	2.69	<1	2.3	2.69	<1	7			-			<1	3.19	1.85	2.71	<1	1.59
7688				4.59	3.16	<1	3.78	2.86	3.49	<1	<1	3.96	3.46	<1	<1	<1	4.05	4.08	4.64	3.88	<1	<1	2.81	<1	0.78	2.92	<1							<1	3.38	2.24	2.7	<1	1.88
7728				-	-	-	3.8		3.51	0	0	4.04	3.41	0	0	0	3.86	4.04	-	-	-	2.34	2.36	0	1.95	2.61	0		-		-	-	-	0		2.11		0	1.67

**Annex 1.** Results of the participating laboratories

Lab		Aerobio porgani			hrotro oorgan		Enterd	obacter	iaceae	Esch	herichia	coli		sumpti			ılase-po phyloco		Lactic	acid ba	acteria		lostridiun erfringen			obic sul		micro	Aerobio organis	ms in	bact	-produc	fish		Yeasts			Moulds	
no.		30 °C					_			_																		TISI	produ	icts	Р	roduct	5	_					
7750	A 52	Э 00	4.24	А	В	<u> </u>	А	В	C	А	В	L	A 2.46	0	0	A -	В -	C	0 0	0	0	Α -	- В	L	A 2.47	2.72	0	А	В	L C	A	В	L	Α	2.22	2.00	A 2.54	0	1.01
7750 7825	4.53 4.83	3.99 4.39	4.31	-	-	-	4 27	3.26	4.00	<1	-		3.46	U	U	-	-	-	<1	3.36	0	-	-	-	2.17	2.72	0	-	-	-		-		0	3.33	2.09 1.73	2.54	-	1.91
7876	4.65	4.39	4.42	-	-		4.37	2.32		<1	<1 <1	<1 3.94	3.41	<1	<1	- <1	3.99	4.16	<1	3.30	1.3	<1	2.73	- <1	-	-	-	-	-	-	-	-		<1 <1			2.59	<1 <1	1.53 1.83
7877	4.89	4.13	4.42	4.79	3.72	4.29		2.79	3.84	<b>\1</b>	<b>\1</b>	3.54	3.34	<1	<1	<b>\1</b>	3.33	4.10	-	-	-	<b>\1</b>	2.73	<b>\1</b>	-	-		-		-	-	-		2.65	3.40	1.88	2.72	<b>\1</b>	1.03
7882	4.69	4.08	4.42	4.79	3.72	4.29	4.34	2.79	3.86	0	0	3.84	3.3	0	0	0	4.02	4.09	3.7	3.19	2.88	2.61	0	0	2.61	1.57	0	-	-	-	-	-		0			2.65	0	1.66
7930	4.61	4.10	4.28				4.12	2.85	3.75	0	0	4.22	0	0	0	0	4.02	4.43	5.7	3.13	2.00	0	2.71	0	2.01	1.57	-	4.81	4.1	4.26	4.28	3.75	0	0		1.05	2.63	0	1.61
7940	4.83	4	4.42				4.07	2.76	3.86	-	-	4.22		-	-	-	03					-	2.71	-				4.01		4.20		5.75	-	-	5.41	1.55	-	-	1.01
7962	4.72	4.09	4 36	_	_	_	4.23	2.43	3.79	0	0	3.9	2.46	0	0	0	3.89	4.02	_	_	_		_	_	_	_		_		_	_	_		0	2.64	1.6	2.3	0	1.62
7968	4.81	4.04	4.26	_	_	_	4.16	2.46	3.78	0	0	3.83	2.79	0	0	0	3.92	4.11	4.55	0	0	0	2.68	0	2.22	3.08	0	_	_	_	4.22	3.32	0	0	3.46	2.06	2.68	0	1.67
7975	4.52	4.06	4.53	-	-	_	3.77	2.39	3.78	-		-	-			<1	3.69	3.93	-	-	-		-	-		-		-	-	-	-	-		<1	3.22	1.96	2.75	<1	1.71
8066		-		-	-	_	-	-	-	0	0	3.79	3.41	0	0	0	4.03	4.09	4.21	0	3.58	2.7	2.18	0	-	-	-	4.43	4.06	3.92	4.04	3.49	0	-	-	-	-		-
8068	4.78	4.03	4.3	-	-	_	4	2.62	3.81	<1	<1	3.79	2.78	0	0	<1	3.66	3.89	-	_	-	0	2.87	0	2.26	2.86	0	-	-	-	-	-	-	0	3.17	2.11	2.51	0	1.66
8105	4.76	4.17	4.4	-	-	_	-	-	-	0	0	3.7	3.24	0	0	0	3.97	4.08	-	-	-	-	_	-	-	-	-	-	-	-	-	-	-	2.55	3.48	1.91	0	0	1.6
8147	4.63	3.72	4.16	-	-	-	4.15	2.58	3.71	<1	<1	3.65	2.95	<1	<1	<2	4.02	4.13	3.6	<2	<1	<1	2.82	<1	2.56	2.82	<1	-	-	-	-	-	-	<1	3.43	2.04	2.45	<1	1.48
8228	4.62	4.03	4.35	4.57	4.04	4.05	3.96	2.71	3.7	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	<0	3.12	1.9	2.68	<0	0.8
8260	4.67	4.04	4.24	-	-	-	4.04	2.69	3.78	-	-	-	0	0	0	0	4.03	4.08	-	-	-	0	2.05	0	2.6	2.11	0	-	-	-	-	-	-	0	3.3	2.18	2.65	0	1.65
8313	4.61	4.08	4.24	-	-	-	4.02	2.59	3.69	-	-	-	3.56	0	0	-	-	-	-	-	-	-	-	-	2.38	3	0	-	-	-	-	-	-	0	3.3	1.92	2.67	0	1.66
8333	4.73	4.04	4.35	-	-	-	4.06	2.38	3.63	<0	<0	3.41	3.33	<0	<0	-	-	-	<1	<1	<1	-	-	-	2.34	2.87	<0	-	-	-	-	-	-	<0	3.36	2.17	2.85	<0	1.82
8397	4.71	4.19	4.41	-	-	-	3.95	2.41	4.02	0	0	3.78	0	0	0	0	4.15	4.11	3.34	0	0	-	-	-	2.44	2.93	0	4.65	3.74	3.15	4.17	3.15	0	0	3.48	1.95	2.34	0	1.72
8430	4.86	4.16	4.48	-	-	-	4.29	2.76	3.91	<1	<1	3.94	-	-	-	<1	3.62	4.11	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	<1	3.36	2.09	2.64	<1	2.2
8435	-	-	-	-	-	-	4.23	3	3.79	<1	<1	3.65	3.61	<1,7	<1,7	<2	<2	4.09	-	-	-	-	-	-	2.66	2.86	0	-	-	-	-	-	-	-	-	-	-		-
8506	4.51	4.01	4.35	-	-		-			0	0	3.54	3.27	0	0	-		-	-	-	-	-	-	-	2.37	2.71	0	-	-	-	-	-	-	0	3.4		2.61	0	1.67
8523	4.74	4.16	4.38	4.6	3.32	3.83	4.35	2.8	3.95	<1	<1	3.13	-	-	-	<1	3.96	4.11	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	<1	3.55	2.2	2.88	<1	1.74
8528	-	-	-	-	-	-	-	-	-	-	-	-	3.38	<1	<1	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	<1	3.08	<1	<1	<1	<1
8568		3.25	4.37	-	-	-		2.96 1.04	3.85	0	0	3.5	3.25	0	0	-	-	-	0	0	0	-	-	-	2.44	2.67	0	4.26		-	-	-	-	0	3.53	1.8	2.75	0	1.75
8626	4.08	3.72	3.85	-	-	-	4.2		3.82	0	0	<b>5.38</b> 3.85	2.26	-	-	-	4.02	4 1 2	-	2.16	0	-	2.05	0	2.7	2.04	0	4.36	3.72	3.9	4.2	2 57	-	-	2.4	2.16	2.64	-	1.66
8628 8657	4.65 4.63	4.05 4.04	4.27	-	-	-	3.99 3.93	2.63 <b>1.23</b>	3.81 3.82	0	U	3.85	3.26	U	U	0	4.03	4.13	U	3.16	U	0	2.95	U	2.7	2.84	U	4.2	4.28	4.25	4.2	3.57	0	0	3.4 3.48	2.16 1.92	2.64	0	1.66 1.7
8734	4.03	4.04	4.37				4.12	2.62	3.88										0	2.81	2.06		-					-						0		2.01	0	0	0
8742	4.66	4.51	4.37				4.12	2.02	3.88	<1	<1	3 66				<1	3 99	4 04	-	2.01	2.00													-	2.57	2.01	-	-	-
8756	-		-		_		_		_	-	-	-	_	_	_	-	-	-		_	_	_	_	_	_	_	_	_	_	_				_			-	_	_
8766	4.59	4.16	4.3	-	-	_	4	2.4	3.7	0	0	3.62	3.38	0	0	0	3.91	4.15	-	_	_	0	2.73	0	-	-	-	-	-	-	-	-		0	3.43	1.93	2.77	0	1.45
8891	4.59	4.06	4.48	-	-	-	4	2.52	3.88	0	0	3.83	3.32	0	0	0	3.84	4	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	0	3.15	-	2.59	0	-
8909	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	- 1	-
8955	4.65	4	4.28	-	-	-	4.26	2.66	3.9	<1	<1	3.88	3.08	<1	<1	<1	3.97	4.3	<1	<1	<1	<1	2.18	<1	2.56	2.56	<1	4.65	4.08	4.45	4.15	3.15	<1	<1	3.32	1.99	3	<1	1.65
9002	4.91	3.94	4.26	4.59	3.11	0	4.08	2.86	3.95	0	0	3.98	3.3	0	0	0	3.93	4.15	0	0	0	-	-	-	2.36	1.95	0	-	-	-	-	-	-	0	3.53	1.85	2.74	0	1.46
9003	4.6	4	4.3	-	-	-	4.15	2.63	3.84	<1	<1	3.78	-	-	-	<1	3.78	4.04	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	<1	3.32	2.11	2.81	<1	1.74
9034	4.59	4.04	4.32	4.66	2.91	<1	4.15	2.6	3.62	<1	<1	3.83	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-		-
9078	4.68	4.18	4.47	-	-	-	4.29	2.7	4.26	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
9217	-		-	-	-	-	-			-	-	-	-	-	-	-		-	-	-	-	-		-		-	-	-	-	-			-	-					
9408	4.6	3.93	4.38	-	-	-	4.34	2.79	3.93	<1	<1	3.95	3.2	<1	<1	<1	3.95	4.03	-	-	-	2.42		<1	2.42	2.95	<1	4.67	3.77	4.01	4.43	2.83	<1	<1	3.42	2.43	2.81	<1	1.78
9429	4.61	4.15	4.26	4 20	-	-	-	-	-		-	2.67	3.28	<1	<1	<1	3.96	4.15	- 45	2.00	2.40	<1	2.66	<1	2.3	2.7	<1	-	-	-	-	-	-	<1	3.2	2	3.62	<1	1.6
9436 9441	5.02 4.56	4.07 4	4.35	4.39	2.92	<1	4.1	2.56	3.63	<1 <1	<1 <1	3.67 3.89	3.26	<1	<1	<1	3.87 3.94	4.05	4.45 4.3	3.96 3.85	<b>3.49</b> <1	<1 0	2.56 2.79	<1 0	<1 2.4	2.44 2.79	<1 0	4.53	4.04	4.32	4.11	3.38	<1	<1 0	3.32 3.26	2.19	2.82	<1 0	1.8 1.72
9453	4.71	3.95	4.28				4.18	2.48	3.91		\1	5.65	3.6	<1	<1	<1 <1	3.99	4.11 4.23	4.5	3.03		-	2.75	-	2.4	2.75	-	4.33	4.04	4.32	4.11	5.56		<1	3.49	2.18 1.94	2.76 2.78	<1	1.62
9512	4.65	4.06	4.27				3.99	2.40	3.79				3.38	<1	<1	- 1	3.33	4.23	<1	<1	2.15													<1	3.09	1.85	<1	<1	<1
9559	4.74	4.18	4.34		_		4.61	2.79	3.74	0	0	3.85	3.38	0	0	0	3.83	4		-	-	_	_	_	_	_	_	_	_	_				2.06	3.08	0	1.57	0	2.45
9662	4.65	3.84	4.27	-	-	_	3.87	0	3.45	0	0	3.19	2.7	0	0	0	4	4.11	3.61	0	0	1.7	2.34	0	1.74	2.79	0	4.45	4.05	4.23	4.13	2.61	0	0	3.3	1.9	2.47	0	1.61
9747	4.56	4	3.9	-	-	-	3.15	2.81	4	-	-	-	3.22	0	0	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	0	2.75	2.48	2.2	0	1.38
9783	4.4	4.11	4.66	-	-	-	-	-	-	-	-	-	0	2.2	3	-	-	-	-	-	-	-	-	-	-		-	-	-	-	-	-	-	2.1	3.43	0	1.67	0	2.74
9886	4.66	4.08	4.32	4.51	3.51	3.74	4.11	2.45	3.56	<1	<1	3.65	3.42	<1	<1	<1	4.04	4.17	-	-	-	<1	2.79	<1	2.51	3.04	<1	-	-	-	-	-	-	<1	3.39	2.13	2.81	<1	1.54
9890	4.64	3.43	4.43	-	-	-	4.32	2.61	3.68	0	0	3.71	3.2	0	0	0	3.85	4.01	3.26	0	0	-	-	-	-	-	-	-	-	-	-	-	-	0	3.11	1.94	2.72	0	1.64
9903		4.15	4.34	-	-	-		2.58	3.9	<1	<1	3.99	3.19	<0	<0	<1	4.03	4.17	-	-	-	<0	3.08	<0	-	-	-	-	-	-	-	-	-	<0	3.51			<0	1.72
9950	4.82	4.03	4.39	-	-	-	4.19	2.6	3.88	-	-	-	-	-	-	-	-	-	0	0	0	-	-	-	-	-	-	-	-	-	-	-	-	0	3.33	1.78	2.66	0	1.85
N	148	148	148	21	21	21	127	126	127	105	105	104	102	101	102	92	91	92	46	46	47	51	50	51	60	59	62	22	22	22	19	19	19	130	131	131	130	127	129
n	141	141	142	20	21	21	124	119	119	103	105	96	87	97	98	91	84	84	19	25	32	38	48	51	54	52	61	21	22	20	19	19	19	118	125	114	109	124	113

**Annex 1.** Results of the participating laboratories

Lab no.	mic	Aerobi croorgan 30°C			chrotro roorgar		Enter	obacter	iaceae	Esch	erichia	coli		sumpti Illus cer			ulase-po phyloco		Lactic	acid ba	cteria		lostridiur erfringer			robic sul		micro	Aerobic organis n produ	ms in	bac	-produc teria in products	fish		Yeasts			Moulds	
	Α	В	С	Α	В	С	Α	В	С	Α	В	С	Α	В	С	Α	В	С	Α	В	С	Α	В	С	Α	В	С	Α	В	С	Α	В	С	Α	В	С	Α	В	С
Min	3.65	3.25	3.45	3.34	2.32	0	3.15	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	4.20	2.65	3.15	4.01	2.61	0	0	2.06	0	0	0	0
Max	5.34	5.03	4.81	4.79	4.11	4.32	4.61	3.26	4.26	3.93	0	5.38	4.24	3.38	3.62	4.21	4.20	4.43	4.64	3.96	3.58	2.95	3.14	0	3.68	4.00	2.79	5.60	4.69	4.60	4.43	4.14	0	2.95	4.21	3.17	3.62	3.29	2.91
Med	4.69	4.06	4.32	4.61	3.34	3.53	4.13	2.63	3.80	0	0	3.80	3.30	0	0	0	3.96	4.11	3.79	0	0	0	2.69	0	2.39	2.78	0	4.60	4.08	4.26	4.26	3.38	0	0	3.32	2.02	2.70	0	1.66
m <sub>PT</sub>	4.675	4.060	4.317	4.586	3.467	3.943	4.137	2.625	3.799	-	-	3.780	3.306	-	-	-	3.946	4.097	3.805	-	-	-	2.659	-	2.341	2.735	-	4.594	4.044	4.178	4.235	3.349	-	-	3.307	2.023	2.672	-	1.641
S <sub>PT</sub>	0.129	0.121	0.101	0.160	0.470	0.326	0.145	0.174	0.143	-	-	0.197	0.195	-	-	-	0.095	0.087	0.681	-	-	-	0.287	-	0.259	0.244	-	0.188	0.242	0.233	0.130	0.339	-	-	0.198	0.183	0.153	-	0.186
u <sub>PT</sub>	0.013	3 0.012	0.010	0.044	0.128	-	0.016	0.020	0.016	-	-	0.024	0.026	-	-	-	0.013	0.011	0.186	-	-	-	0.051	-	0.043	0.040	-	0.050	-	0.062	0.037	-	-	-	0.022	0.021	0.017	-	0.021
F+	0	0	0	0	0	-	0	0	0	2	0	0	0	4	4	1	0	0	0	21	15	13	0	0	0	0	1	0	-	0	0	-	0	12	0	0	0	3	0
F-	0	0	0	0	0	-	0	4	1	0	0	2	11	0	0	0	4	1	25	0	0	0	1	0	3	1	0	0	-	0	0	-	0	0	0	9	11	0	9
<	4	3	4	1	0	-	2	2	2	0	0	4	2	0	0	0	3	5	2	0	0	0	1	0	2	4	0	0	-	2	0	-	0	0	5	3	8	0	3
>	3	4	2	0	0	-	1	1	5	0	0	2	2	0	0	0	0	2	0	0	0	0	0	0	1	2	0	1	-	0	0	-	0	0	1	5	2	0	4
Lowe	r 4.29	3.70	4.01	4.11	2.06	0	3.70	2.10	3.37	0	0	3.19	2.72	0	0	0	3.66	3.84	1.76	0	0	0	1.80	0	1.56	2.00	0	4.03	3.32	3.48	3.84	2.33	0	0	2.71	1.47	2.21	0	1.08
Uppe	r 5.06	4.42	4.62	5.07	4.88	4.32	4.57	3.15	4.23	0	0	4.37	3.89	0	0	0	4.23	4.36	5.85	0	0	0	3.52	0	3.12	3.47	0	5.16	4.77	4.88	4.63	4.37	0	0	3.90	2.57	3.13	0	2.20

N = number of reported results n = results without annotation Min = lowest reported result Max = highest reported result Med = median value m<sub>PT</sub> = assigned value s<sub>PT</sub> = standard deviation u<sub>PT</sub> = measurement uncertainty

F+ = false positive < = low outlier F- = false negative > = high outlier Lower = lowest accepted value Upper = highest accepted value

False positive or false negative

Outside the acceptance limits

Results "larger than" are not evalutated

The parameter is not evaluated

The result not evaluated

u<sub>PT</sub> > 0,3 s<sub>PT</sub> and/or > 20 % outliers and/or fewer than 12 evaluated results

**Annex 2.** Z-scores of all participants

Lab	Aerobic microorganisms,	Psychrotrophic	Entero	bacteria	ceae	Esche	erichia	coli	Presum		icillus	_	gulase-po		Lactic :	acid bad	cteria		ostridiun		Anaerobic sulp		Aer microorg	obic anisms in		-produci teria in f	_		Yeasts		1oulds	
no.	30 °C	microorganisms							(	cereus		st	aphyloc	occi				p	erfringen	IS	reducing bact	teria	fish pr			roducts						
	A B C	A B C	Α	В	С	A	В	С	Α	В	С	Α	В	С	Α	В	С	A	В	С	A B	С	A I	3 C	Α	В	С	Α	в с	А	В	С
1149	<b>-4.000 -2.985</b> -0.861		0.293	0.889 1	1.405	0	0	-0.405																				0	-0.537 <b>2.05</b>	<b>7</b> 0.708	0	1.128
1237	<b>2.525</b> -0.498 -1.655		0.087	-2	2.236	0	0	-2.339				0	-1.016	-0.662			0				0.229 -1.375	0	1.000	-3.771				0	-0.789			
	0.896 0.331 0.232			0.544		0	0		0.278	0	0	0		1.186						0		0						0	-0.486	-0.733		
	-1.277 -0.581 -1.357	<b>-4.000</b> -0.355		-1.124 -1		0	0	0.511		0	0	0			-0.023	0	0	0	0.874	0	<b>4.000</b> 0.101	0	0.711	0.225	-0.651		0	0	1.277 0.31			0.643
2000 2035	-2.596 -1.741 -2.747			-0.606 -0		0	0		-0.646	0	0	0	-0.171	0.146	0.667	0	0	0	0.002	0	4 007 0 400							0	-0.386 -0.17			-0.166
	-1.277 <b>-3.399</b> -1.655		-1.975	-0.779 -1	1.816	0	0	-1.270	-0.544	U	U							0	-0.312 -0.556	0	-1.007 -0.432	U						0	0.169 0.47 0.471 -0.12			0.320 1.398
	0.197 0.248 0.430		-0.050	0.717 1	895	U	U		1.100	0	0							U	-0.330	U								0	0.622 0.09			-0.974
	0.353 1.160 0.629	0.774 1.368		0.141		0	0	0.511		0	0	0		2.341		0	0	0	-0.416	0	0.770 -0.801	0						0	-0.386 <b>4.00</b>			0.859
	1.051 0.828 0.232		0.156	0.256	0.565	0	0	-0.354		0	0	0	1.096	-4.000				0	0.909	0	0.499 -3.793	0						0	1.227 0.74		0	0.481
2317	0.120 -0.747 -0.165		-0.394	-0.491	0.565	0	0	0.358	-1.828	0	0	0	-1.227	0.146				0	-3.556	0	-0.891							0	-0.386 -0.28	87 -1.715	0 -	-1.513
2324	-0.889 -0.912 -1.357		-1.150	-2.907 C	0.075	0	0	0.053	-0.030	0	0	0	-0.382	-0.200								0						0	1.580 -0.77	7 0.250	0	0.320
2344	-1.432 -0.166 -0.364		-0.806	-0.491 -0	0.415	0	0	-1.372	-0.389		0	0	0.463	0.146				0		0	0.113	0						0	-0.28	37 0.315		0.050
	0.741 -0.001 0.132			0.774																								0	0.370 0.09	5 0.708	0	0.643
	0.353 -0.083 1.721			-1.009 0		0		1.020		_																		_				
	-0.268 0.165 1.423			1.234		0	0	-0.303		0	0		-0.065		0.014			_	0.070	_	4 462 0 062								-0.940 <b>-4.0</b> 0			0.158
	0.197 -0.664 0.033 0.197 0.165 -0.165	0.667 0.606	0.637	-0.319 -1	).145	0	0	-0.303 0.664		0	0	0	-1.121	0.146	0.814			U	0.979	U	-1.162 0.962	U							-1.142 -0.88 -0.789 -1.05			-0.435 - <b>2.214</b>
	1.129 0.663 -0.364	-0.007 -0.050		0.256		U	U		-0.235	0	0	U	0.308	0.140															1.177 0.04		-	-1.298
	<b>3.379</b> -0.995 -0.463			0.774 -0		0	0		-2.084		0	0	1.730	0.377		0	0	0	-0.242	0								ŭ	-0.587 -1.15			-0.705
	-0.113 -0.166 0.033																											0	0.723 0.85		0	
	0.197 0.414 -1.357		0.431	0.832	0.215	0	0	0.511													0.693 -2.932	0										
2941	0.197 0.828 -0.463		-0.119	-0.491 -0	0.976	0	0	0.002	1.152	0	0	0		0.493														0	1.832 4.00	0 1.298	0 -	-1.567
3031						0	0	0.104				0	-0.065	-0.200									0.406	0.440	1.270		0					
	-0.035 -0.829 -1.158			-0.951					1.203		0																	0	<b>-4.000</b> -0.39	96 -1.191	0 -	0.866
	-1.122 0.663 -1.357			0.947 -0		•	•	2 425				•	0.500	4 522							0.004 0.030							•	0.000 0.00	14 4 4 6 7	•	0.754
	-0.811 -1.907 -0.563 -0.113 -0.498 -0.463			-0.549 -0 0.026 -0		0		<b>-2.135</b> -0.100				0	0.568	0.608	4 000		0		-2.474	0	-0.891 0.839		0.033	0.741	0.348		0	U	0.068 -0.94 -0.134 1.40			-1.244
	1.827 0.497 1.622			-0.031 -1		0	0	0.816	0 124	0	0	0		-4.000	-4.000		U		-2.4/4	U			5.033	0.741	0.546		U		-0.134 1.40	3 -2.303	0	1.244
	0.896 4.000 0.728			-0.031			-		-1.160	0	0	0		-0.085							<b>-2.900</b> -0.145	0										
	-0.113 0.082 0.331			0.371		0	0		-0.441	0	0	0		-0.085							-0.196 0.224							0	-0.033 0.42	2 -0.733	0 -	-1.891
3595	0.197 0.082 -0.165		-0.119	0.889	0.145	0	0	-0.303	-1.519	0	0	0	0.251	0.608				0	0.979	0	-1.625 0.839	0						0	-0.486 -0.50	05 0.184	0 -	-0.273
	0.197 -1.327 -1.158		-0.256	-2.447 -(	0.696	0			-1.057	0	0	0	-0.488	0.031				0	0.490	0												
	0.120 -0.001 -0.960					0	0	-3.204									0												-0.990 0.25			-1.136
3878 4047	0.717 -1.302 1.046		0.974	-0.175 -0	0.226	0	0	0.715	-0.456	0	0	0	-2.430	-0.397														0	-0.456 0.02	4	0	
	0.353 1.906 -1.556		0.018	0.429 -0	2/15				4.000	0	0					0	0											0	1.025 -0.83	2 0 774	Λ.	-1.190
	-0.346 -0.001 0.232			0.084					4.000	U	U					U	U											U	1.025 -0.05	0.774	0	1.130
	1.129 1.989 1.225	0.210 -0.313		0.947		0	0	-0.863	-0.235	0	0								-1.637	0	0.229 0.839	0	0.725	0.655	0.809		0	0	0.874 -0.12	3 0.315	0	0.589
4171	0.042 1.409 -0.563		-0.188	1.177 -2	2.726	0	0	-1.881	0.535	0	0	0				0	0				-1.007 0.716	0						0	0.521 -1.43	0.250	0	0.805
	0.042 1.989 -0.165			0.889		0	0	0.358				0		-0.431																		
4288	-1.510 -0.415 -0.364		-0.394	0.544	3.286	0	0	0.664	1.049	0	0	0	-0.276	0.608					-0.835	0	-0.659 -0.186	0						0	-0.033 -0.55	9 0.446	0	0.320
4339																																
	1.361 2.321 2.416	0.225 0.455		1.867		0			2.179	0	0	_	1044	2 244				0	0.550	0	0.005	0	1 004	1.815				0	-0.839 0.47		0	2 270
	<b>3.922</b> 0.497 <b>-4.000</b> -0.113 0.165 0.232	0.335 0.155		-0.664 -1 -1.181 C		0	0	0.104	-0.235	0	0	U	1.941	2.341				U	-0.556	U	-0.965	U	1.894	1.815				0	- <b>3.208 4.00</b> 0.572 -0.23			-2.376 2.745
	0.197 <b>2.404</b> 1.324			0.774		0	0	0.460	-0.233	U	U	0	-1.438	-2.280														U	0.372 -0.23	0.440	U	2.743
4560	1.137 1.324		0.501	3,		ŭ	J	0.700				Ü	2.430															0	0.773 -1.05	0 1.036	0	0.805
	0.353 0.911 0.728		0.774	0.832	0.425	0	0	-0.201																					0.017 0.31			-0.543
4635	-1.665 -0.664 -0.165		-0.463	-0.204 -0	0.555				0.792	0	0	0	-0.804	-0.200								0						0	0.320 -0.66	8 -0.143	0	0.050
	0.508 0.497 <b>-2.648</b>			0.774 -2				-3.763				0	-0.276								<b>2.006</b> -0.391								-0.688 <b>-3.2</b> 3			0.050
	-0.656 -1.327 -1.655		1.118	-0.031 -0	0.135				-2.084		0	0			-1.666						-0.350 <b>4.000</b>	0							-0.537 0.09			-0.920
	-0.811 0.331 0.132		0.434	0.141	145	0	0	-1.677	1 202	0	0	0	0.779	1.186				0	1.188	0									1.681 0.25			0.589
	-0.346 -0.249 -0.463 0.353 0.414 0.232			0.141 C -0.031 1		0	0		1.203 -0.030	0	0	Λ	0.991	0 1/16							1.233 0.306	0	1450	0.526	0.194		0	U	-0.789 0.31	3 0.5//	0	0.481
	0.353 0.414 0.232 0.275 -1.410 0.728			0.141 -(		0		0.765	0.030	U	U	U	0.551	0.140							1.233 0.300	U	J.433	0.320	0.154		U	0	-1.192 -0.12	23 -0,143	0 .	-0.759
	0.275 -0.332 -0.463			-0.031 -0		Ü	·		1.203	0	0																	ŭ	-0.688 0.58			-0.597

#### **Annex 2.** Z-scores of all participants

Lab	Aerobic	Psychrotrophic	Fatamaka da		Facility of all		Presum	nptive Ba	acillus	Coa	gulase-p	ositive	Lastia			C	lostridium		Anaerobic sulphite-		Aerobic			produci	_		V		a l - l .	
Lab	microorganisms,	microorganisms	Enterobacteriace	ie	Escherich	iia coii		cereus		st	taphyloc	occi	Lactic	acid ba	acteria	р	erfringens		reducing bacteria		organis			eria in f	isn		Yeasts		Moulds	5
no.	30 °C			_																tist	n produ	cts		roducts						
	A B C	А В С	А В С		А В	С	Α	В	С	Α	В	С	Α	В	С	Α	в с		а в с	Α	В	С	Α	В	С	Α	B C	A	В	C
	<b>-4.000 -2.073</b> 1.523		4 405 4 060 0 0																								-0.738 0.96	0.774		-0.866
	0.353 -0.498 1.423 1.222 -0.349 0.977		-1.425 1.062 0.2		0 0		-2.696	0	_																	0	0.678 -1.72	0.000	_	-1.405
		1.035 0.370	0.939 -1.308 1.5	03	0 0	0.750	-2.090	U	0											0.513		0.536	1 116		0	U	0.078 -1.72	0.820	U	-1.405
5188 5200		1.025 -0.270			0 0	1.325														0.512		0.526	1.110		U	0	1.696 0.24	0.741	0	0.066
	0.818 0.911 -0.165		0.637 -0.4		0 0	-1.117					-4 000	-2.857														U	<b>-4.000</b> -0.28		U	-1.028
	-0.346 0.248 0.331	-1 670 1 028	1.393 0.659 0.8		0 0		0.227	0	0	0	-0.910			0	0		0.909 0	) (	0.924 1.085 0							0	0.723 0.20		0	0.320
5220	-1.975 -1.907 -0.662	1.070 1.020	-1.975 -1.124 <b>-2.6</b>		0 0			Ü	·	0		-0.893		Ü	Ü		0.505 0	´ ``	0.524 1.005 0	-1.190		-2.267					-2.805 -2.30		0	-0.866
5261	1.575 1.507 0.002		2.575 2.22			1.551				Ů	0.251	0.055								1.150		,				Ŭ	2.000	0.512	Ů	0.000
	<b>-2.673</b> -0.249 <b>4.000</b>		<b>-3.349</b> -0.549 <b>3.0</b>	85	0						0.463			0	0												0.925	-4.000	0	4.000
	<b>-2.596</b> -1.990 <b>-2.449</b>		-1.562 1.924 -0.8																							0	-1.646 -0.50		0	0.212
5352	1.051 0.828 0.927		1.256 3.2	25	0 0	1.173	-0.030	0	0	0	1.730	3.266				0	1.676 0	)								0	1.832 -0.17	3 0.184	0	0.859
5419	-1.122 -1.161 -0.960	0.335 0.900	-0.119 0.889 -1.0	146	0 0	-0.100	)																			0	-0.285 0.803	0.446	0	-0.004
5545	0.663 -1.576 -4.000		-0.188 -1.584				0.843	0	0	0	0.040	-0.316														0	-0.738 1.130	0.250	0	1.452
5553	0.508 0.580 1.721		1.668 -0.664 -0.1	.35	0 0	-0.812	0.587	0	0	0	0.779	1.533				0	1.188 0	)												
5615	0.353 0.994 0.629		0.843 -0.779 1.1	25	0 0	1.122	-1.160	0	0	0	-0.382	-0.200				0	-0.033 0	) (	0.461 -0.227 0							0	-0.134 0.31	-2.043	0	-2.214
5632	0.973 -0.498 0.629				0 0	0.613	-0.544	0	0																	0	0.975 0.96	0.184	0	0.859
5654	-0.035 0.165 -0.364		0.431 -0.319 0.6	35				0																		0	-0.436 0.694	-0.995	0	-1.405
	-1.354 -1.161 -1.854																													
	-0.811 0.746 <b>-2.052</b>		-0.394 -0.779 -0.2	:05				0	0																	0	-0.839 0.09		0	-0.220
	4.000 4.000 2.119						4.000	0	0																	0	4.000 4.000		0	
	0.275 -0.415 -0.861		-0.050 0.659 0.0		0 0		0.176	0	0	0	-0.065	-2.395			_	0	-0.765 0		1.156 -1.211 0						_	0	-0.638 0.858		0	0.158
	1.206 1.326 0.331	1.025 -0.057	0.018 -0.089 0.1		0 0		0.535	0	0				-0.507		0				1.316 1.044 0	-1.030		0.225	0.655		0	0	-0.184 0.470		•	4.000
	0.197 0.414 0.232		0.256 2.447 1.3		0 0	-2.135	-0.389	0	0					0	0			-	1.162 0.347 0							0	0.723 0.095 1.479 <b>-3.93</b>		0	0.966
	0.197 0.331 -0.165 1.361 <b>4.000 2.317</b>		-0.256 <b>-2.447</b> -1.3 1.874 1.982 <b>3.0</b>				1.306	0	0																	U	1.4/9 -3.93	-4.000	U	0.320
	0.042 0.165 0.033		1.074 1.302 3.0	03			-1.160		0	0	0.991	0 608							0.422 -0.432 0							0	0.925 -0.45	0 110	0	-0.166
	1.516 1.409 0.530	-2 861 -2 440					-1.100	U	U	U	0.551	0.008						,	0.422 -0.432 0							0	<b>-2.049</b> -1.26		0	-4.000
	-0.578 -0.415 -0.066	2.001 2.440	-0.669 -0.261 -0.4	15	0 0	0.460	-1.006	0	0	0	-0.804	-1.125			0		-0.207 0	) (	0.190 <b>-3.998</b> 0							0	-2.402	-0.471	0	-0.705
	0.275 0.331 0.331		-0.394 -0.606 -0.3		0 0		0 -0.698	0	0	0		0.031			-		-0.033 0									0	0.723 1.130		0	1.667
	-0.113 0.165 -0.364	0.523 1.198	-0.394 0.199 -1.2				-0.030	0	0	0	1.202		0.447	0	0				0.190 0.593 0	-0.286		0.483	0.194		0	0	0.622 0.803		0	-0.004
6443																		(	0.615 <b>4.000</b> 0											
6456	-0.811 0.082 -1.258						-2.547	0	0	0	0.991	0.724				0	0.316 0	) (	0.847 0.101 0							0	1.933 -0.39	0.381	0	-0.866
6490	0.973 0.165 1.324		-0.119 0.429 0.4	95			1.614	0	0	0	0.885	1.764						(	0.538 0.880 0							0	0.622 1.40	1.363	0	0.859
6594																														
	<b>-2.518</b> 0.082 -0.463				0 0																					0	-0.587 <b>-2.14</b>			0.643
6686	1.439 1.160 0.629	1.213 1.219	1.874 1.292 0.4		0 0						0.568						0.769 0	)		0.831		-0.419	0.502		0		0.370 0.09	i	0	-0.812
6728	1.051 <b>2.486</b> 0.530		0.568 0.832 0.3		0 0					0	0.568	0.031																		
	<b>-3.061</b> 0.497 -0.463		-0.669 -0.721 -0.6	26	0 0	-0.812	2				0.05																0.000 4			0.754
6801	0.360 0.444 0.433		0.335 0.400 0.4	0.5	0 0	0.500	0.330	0			-0.804								4 000 0 040							0	-0.083 -1.32		0	0.751
	-0.268 0.414 0.132 0.275 -0.166 0.033		-0.325 0.199 0.4		0 0		0.330	U	0		0.357 -1.966				0			-	<b>4.000</b> 0.019 0							0	-0.235 -0.12 -2.805	1.167	0	0.158 0.481
	0.275 -0.166 0.033		0.637 0.544 0.4		0 0	0.715	-0.184	0	0	U	-1.900	, -0.516	-4.000		U											0	-0.990 -0.06		0	-0.489
	0.508 -0.512 0.232		-0.738 0.256 1.1				-0.133		0																	U	-0.550 -0.00	-0.143	U	-0.465
6981	0.508 -1.101 0.527		-0.738 0.230 1.1	55			-0.133	U	U																	0	<b>-4.000</b> -0.61	4 000	0	-3.238
7182																										Ü	4.000	4.000	Ü	3.230
	0.663 0.082 0.728		-0.463 <b>-2.792</b> -0.2	75			0.535	0	0					0	0											0	-0.235	0.708	0	
	-0.811 0.663 -0.463																									0	0.471 0.96		0	0.481
	0.120 -0.995 -0.463		0.156 0.026 -0.2	.05																						0	-0.638 -0.88		0	-1.891
7248	-0.966 <b>-2.073 -3.045</b>	-0.354 -0.462	<mark>-2.593 -2.734</mark> -1.8	16	0 0	0.511	-0.235	0	0	0	-2.388	-0.778	1.152		0	0	-0.486 0	) -	1.162 -1.047 0	-0.179		-0.076	-1.727		0	0	0.773 -1.43	2 1.298	0	0.212
7330	-0.656 -0.912 -0.563		-0.277 -1.469 1.0	55	0 0	-1.641	0.587	0	0	0		-4.000														0	-1.338 0.830	1.029	0	-0.220
	0.197 -0.332 0.033				0 0		-0.698	0	0	0		-0.778							-0.801 0							0	-0.235 <b>4.00</b>		0	-3.023
	-1.820 0.994 0.828		-0.394 0.371 <b>-3.6</b>		0					0		-4.000							0.080 -1.129 0							0	1.177 <b>2.05</b>		0	2.476
7564					0 0		0.792	0	0	0		0.955				0	0.107 0	) -	0.157 -0.186 0							0	-0.587 -0.94	0.250	0	-0.273
	0.120 0.165 -0.563	0.022 0.552	-0.188 -0.031 -0.0		0 0	0.358		0		0		-0.200			_	_	0.535		4 000 0 757							0	0.270 4.55	0.00		4 200
	-1.432 1.409 -0.165	0.022 -0.653	-2.456 1.349 -2.1		0 0		0.792		0	0		0.608			0	0			<b>4.000</b> 0.757 0								0.370 1.18			1.290
//28	1.129 -1.824 -1.357		-2.318 -1.872 -2.0	126	0 0	1.325	0.535	0	0	0	-0.910	-0.662					-1.044 0		1.509 -0.514 0							U	-0.537 0.470	-0.929	0	0.158

#### **Annex 2.** Z-scores of all participants

Lab no.	Aerobic microorganisms, 30°C	Psychrotrophic microorganisms	Enterobacteriaceae	Escherich	nia coli	•	ive Bacillus reus		gulase-positi aphylococci	ve La	actic ac	id bacte	ria		ostridium rfringens		Anaerobic sulphite- reducing bacteria	Aero microorga fish pro	anisms in	bacteri	oducing ia in fish ducts		Yeasts	Mo	oulds	
	A В С	а в с	A В С	А В	С	Α	в с	Α	В	c ,	A	В (	С	Α	В	С	а в с	A E	3 C	Α .	в с	Α	в с	A	в с	ā
7750	-1.122 -0.581 -0.066					0.792						0 (	0				-0.659 -0.063 0					0	0.118 0.367	-0.864	0 1.45	i2
7825	1.206 <b>2.735</b> 1.026		1.599 <b>3.650 2.035</b>	0 0																		0	1.479 -1.595	-0.536	0 -0.59	<b>∌</b> 7
7876	-0.966 0.746 0.232		-0.256 -1.757 1.055	0 0	0.816	0.535	0 0	0	0.463 0.7	724				0	0.246	0						0	0.773 1.621	0.315	0 1.02	20
7877	1.672 0.165 1.026	1.276 0.538	1.393 0.947 0.285			0.176	0 0																0.471 -0.777			
7882	-1.742 0.828 0.430		-0.119 0.659 0.425	0 0	0.308	-0.030	0 0	0	0.779 -0.	085 -0.	155					0	1.040 <b>-4.000</b> 0					0	-0.235 <b>-2.031</b>	-0.143	0 0.10	)4
7930	-0.501 -0.498 -0.364		0.293 1.292 -0.345	0 0	2.241		0 0	0	0.885 3.8	343				0	0.177	0		1.150	0.354	0.348	0	0	0.521 -0.396	-0.471	0 -0.16	56
7940	1.206 -0.498 1.026		-0.463 0.774 0.425																							
7962	0.353 0.248 0.430		0.637 -1.124 -0.065	0 0	0.613	-4.000	0 0	0	-0.593 -0.	893												0	-3.359 -2.304	-2.436	0 -0.11	12
7968	1.051 -0.166 -0.563		0.156 -0.951 -0.135	0 0	0.257	-2.650	0 0	0	-0.276 0.3	146 1.0	093	0 (	0	0	0.072	0	-0.466 1.413 0			-0.113	0	0	0.773 0.204	0.053	0 0.15	8
7975	-1.199 -0.001 <b>2.119</b>		<b>-2.525</b> -1.354 -0.135					0	-2.705 -1.	933												0	-0.436 -0.341	0.512	0 0.37	3
8066				0 0	0.053	0.535	0 0	0	0.885 -0.	085 0.5	594	0			-1.672	0		-0.871	-1.107	-1.496	0					
8068	0.818 -0.249 -0.165		-0.944 -0.031 0.075	0 0	0.053	-2.701	0 0	0	-3.022 -2.	395				0	0.735	0	-0.312 0.511 0					0	-0.688 0.476	-1.060	0 0.10	14
8105	0.663 0.911 0.828			0 0	-0.405	-0.338	0 0	0	0.251 -0.	200													0.874 -0.614		0 -0.22	20
	-0.346 <b>-2.819</b> -1.556		0.087 -0.261 -0.626	0 0	-0.659	-1.828	0 0	0	0.779 0.3	377 -0.	301	0 (	0	0	0.560	0	0.847 0.347 0					0	0.622 0.095		0 -0.86	
	-0.423 -0.249 0.331	-0.103 1.219	-1.219 0.486 -0.696																			0	-0.940 -0.668		0 -4.00	
	-0.035 -0.166 -0.761		-0.669 0.371 -0.135				0 0	0	0.885 -0.	200				0	-2.126	0						0	-0.033 0.858		0 0.05	
	-0.501 0.165 -0.761		-0.806 -0.204 -0.766				0 0										0.152 1.085 0					0	-0.033 -0.559		0 0.10	
	0.430 -0.166 0.331		-0.531 -1.412 -1.186			0.124						0 (	-				-0.003 0.552 0					0	0.269 0.803		0 0.96	
	0.275 1.077 0.927		-1.287 -1.239 1.545	0 0	0.002		0 0		<b>2.152</b> 0.1		.683	0 (	0				0.384 0.798 0	0.299	-4.000	-0.497	0	0	0.874 -0.396		0 0.42	
	1.439 0.828 1.622		1.049 0.774 0.775		0.816			0	<b>-3.444</b> 0.3													0	0.269 0.367	-0.209	0 3.01	.5
8435			0.637 <b>2.154</b> -0.065			1.562		0	-0.	085							1.233 0.511 0									
	-1.277 -0.415 0.331			0 0		-0.184	0 0	_									0.113 -0.104 0					0	0.471 0.476		0 0.15	
	0.508 0.828 0.629	0.085 -0.313	1.462 1.004 1.055	0 0	-3.305			0	0.146 0.3	146												0	1.227 0.967		0 0.53	.5
8528			0.040 4.004 0.055		4 400	0.381							_										-1.142		0	
	0.430 -4.000 0.530		0.018 1.924 0.355			-0.287	0 0					0 (	U				0.384 -0.268 0	1 242	4 400			0	1.126 -1.214	0.512	0 0.58	9
	-4.000 -2.819 -4.000		0.431 -4.000 0.145		4.000				0.005.07			,	_			•	4 200 0 420 0	-1.243	-1.193			_	0.474 0.740	0.000		
	-0.191 -0.083 -0.463		-1.013 0.026 0.075	0 0	0.358	-0.235	0 0	0	0.885 0.3	3//		(	0	0	1.014	U	1.388 0.429 0	-2.094	0.311	-0.267	0	0	0.471 0.749		0 0.10	
	-0.346 -0.166 0.033		-1.425 <b>-4.000</b> 0.145																			•	0.874 -0.559		0 0.32	U
	0.197 <b>2.321</b> 0.530 -0.113 <b>3.730</b> -0.563		-0.119 -0.031 0.565	0 0	-0.608			0	0.463 -0.	662												U	-1.696 -0.069		U	
8756				0 0	-0.008			U	0.465 -0.	002																
	-0.656 0.828 -0.165		-0.944 -1.296 -0.696	0 0	-0.812	0.381	0 0	٥	-0.382 0.6	508				0	0.246	٥						0	0.622 -0.505	0.643	0 -1.02	28
	-0.656 -0.001 1.622		-0.944 -0.606 0.565	0 0			0 0		-1.121 -1.					U	0.240	U						0	-0.789		0 -1.02	.0
8909	0.050 0.001 1.022		0.544 0.000 0.505	0 0	0.257	0.073	0 0	·	1.121 1.	123												Ü	0.765	0.550	Ü	
	-0.191 -0.498 -0.364		0.843 0.199 0.705	0 0	0.511	-1.160	0 0	0	0.251 <b>2.</b> 3	841		0 (	n	0	-1.672	0	0.847 -0.719 0	0 299	1 171	-0.651	0	0	0.068 -0.178	2.149	0 0.05	:0
	1.827 -0.995 -0.563	0.022 -0.759	-0.394 1.349 1.055	0 0		-0.030			-0.171 0.6			0 (		·	1.072	Ŭ	0.075 -3.219 0	0.255	2.272	0.031			1.126 -0.941		0 -0.97	
	-0.578 -0.498 -0.165	0.022 0.755	0.087 0.026 0.285	0 0	0.002				-1.755 -0.			•					0.075					0	0.068 0.476		0 0.53	
	-0.656 -0.166 0.033	0.461 -1.185	0.087 -0.146 -1.256																							
	0.042 0.994 1.523		1.049 0.429 3.225																							
9217																										
9408	-0.578 -1.078 0.629		1.393 0.947 0.915	0 0	0.867	-0.544	0 0	0	0.040 -0.	778					-1.393	0	0.306 0.880 0	0.406	-0.720	1.501	0	0	0.572 <b>2.221</b>	0.905	0 0.75	1
9429	-0.501 0.746 -0.563					-0.133	0 0	0	0.146 0.6	508				0	0.002	0	-0.157 -0.145 0					0	-0.537 -0.123	4.000	0 -0.22	20
9436	<b>2.680</b> 0.082 0.331	-1.232 -1.164	-0.256 -0.376 -1.186	0 0	-0.558	-0.235	0 0	0	-0.804 -0.	547 0.9	946			0	-0.347	0	-1.211 0					0	0.068 0.912	0.970	0 0.85	9
9441	-0.889 -0.498 0.232			0 0	0.562			0	-0.065 0.3	146 0.7	726	(	0	0	0.456	0	0.229 0.224 0	-0.339	0.612	-0.958	0	0	-0.235 0.858	0.577	0 0.42	.7
9453	0.275 -0.912 -0.364		0.293 -0.836 0.775			1.511	0 0	0	0.463 1.5	533												0	0.925 -0.450	0.708	0 -0.11	12
9512	-0.191 -0.001 -0.463		-1.013 -0.146 -0.065			0.381	0 0					0										0	-1.091 -0.941		0	
9559	0.508 0.994 0.232		<b>3.249</b> 0.947 -0.415	0 0	0.358	0.381	0 0		-1.227 -1.														-1.142	-4.000	0 4.00	0
	-0.191 -1.824 -0.463		-1.837 <b>-2.446</b>	0 0	-3.000		0 0	0	0.568 0.3	146 -0.	287	0 (	0		-1.114	0	<b>-2.321</b> 0.224 0	-0.764	0.225	-0.805	0	0	-0.033 -0.668		0 -0.16	6
	-0.889 -0.498 <b>-4.000</b>		<b>-4.000</b> 1.062 1.405			-0.441	0 0															0	-2.805 2.493		0 -1.40	
	<b>-2.130</b> 0.414 <b>3.409</b>																						0.622		0 4.00	
	-0.113 0.165 0.033	-0.479 0.092	-0.188 -1.009 -1.676		-0.659		0 0		0.991 0.8					0	0.456	0	0.654 1.249 0					0	0.421 0.585		0 -0.54	
	-0.268 <b>-4.000</b> 1.126		1.256 -0.089 -0.836	0 0			0 0	0			.800	0 (	0									0	-0.990 -0.450		0 -0.00	
	0.197 0.746 0.232		0.431 -0.261 0.705	0 0	1.071	-0.595	0 0	0	0.885 0.8	340				0	1.467	0						0	1.025 1.021		0 0.42	
9950	1.129 -0.249 0.728		0.362 -0.146 0.565									0 (	0									0	0.118 -1.323	-0.078	0 1.12	8

|z| ≥ 3,0 ("Unacceptable" or "Action")

2,0 < |z| < 3,0 ("Warning")

#### **Annex 2.** Z-scores of all participants

Lab no.	mi	icroor	obic ganisi ) °C	ms,		chrotro oorgan		Ente	robacte	eriacea	e Es	herichi		Presu	mptive B cereus	acillus	Coag	ulase-po iphyloco	sitive occi	Lactio	acid ba	cteria		ostridiu erfringer			robic su icing ba			Aerobio oorganis h produ		b	2S-produ acteria ir produc	icing i fish ts		Yeasts			Moulds	;
	Α	E	В	С	A	В	С	Α	В	С	Α	В	С	Α	В	С	Α	В	С	Α	В	С	Α	В	С	Α	В	С	Α	В	С	Α	В	С	Α	В	С	Α	В	С

The parameter is not evaluated

The result is not evaluated

# Internal and external control for microbiological analyses of food and drinking water

All analytical activities require work of a high standard that is accurately documented. For this purpose, most participants carry out some form of internal quality assurance, but the analytical work also needs to be evaluated by an independent party. Such external quality control of laboratory competence is commonly required by accreditation bodies and can be done by taking part in proficiency testing (PT).

In a PT, identical test material is analysed by a number of participants. After reporting of results by the participants, the organiser evaluates the results and compiles them in a report.

#### The Swedish Food Agency's PT program offers

- External and independent evaluation of participants' analytical competence.
- Improved knowledge of analytical methods with respect to various types of organisms.
- Expert support.
- Tool for inspections regarding accreditation.

For more information, visit our website: <a href="https://www2.slv.se/absint">https://www2.slv.se/absint</a>

#### The Swedish Food Agency's reference material

As a complement to the proficiency testing, but without specific accreditation, the Swedish Food Agency also manufactures a number of reference materials (RM) for internal quality control of food and drinking water microbiological analyses, including pathogens.

For more information, visit our website: <a href="https://www.livsmedelsverket.se/en/RM-micro">www.livsmedelsverket.se/en/RM-micro</a>

