Drinking Water Microbiology March 2020

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Proficiency testing Drinking water Microbiology March 2020

Parameters included

Coliform bacteria and *Escherichia coli* with membrane filter method (MF) Coliform bacteria and *Escherichia coli*, (rapid methods with MPN) Suspected thermotolerant coliform bacteria with MF (not assessed) Intestinal enterococci with MF/MPN *Pseudomonas aeruginosa* with MF/MPN Culturable microorganisms (total count) 3 days incubation at 22±2 °C Culturable microorganisms (total count) 2 days incubation at 36±2 °C



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Abbreviations and explanations

Microbiological media

CCA	Chromocult Coliform Agar [®] (Merck; EN ISO 9308-1:2014)
Colilert	Colilert [®] Quanti-Tray [®] (IDEXX Inc.; EN ISO 9308-2:2014)
Enteroler	t Enterolert [®] Quanti-Tray [®] (IDEXX Inc.)
LES	m-Endo Agar LES (according to SS 028167)
m-Ent	m-Enterococcus Agar (Slanetz & Bartley; accord. to EN ISO 7899-2:2000)
m-FC	m-FC Agar (according to SS 028167)
PACN	Pseudomonas Agar base/CN agar (with cetrimide and nalidixic acid; according to EN ISO 16266:2008)
D 11	e ,
Pseudale	rt Pseudalert [®] Quanti-Tray [®] (IDEXX Inc.; ISO 16266-2:2018)
YEA	Yeast extract Agar (according to EN ISO 6222:1999)

Other abbreviations

MF	Membrane filter (method)
MPN	"Most Probable Number" (quantification based on statistical distributions)
ISO	"International Organization for Standardization" and their standards
EN	European standard from "Comité Européen de Normalisation" (CEN)
NMKL	"Nordisk Metodikkomité for næringsmidler" and their standards
DS, NS, S	SFS, SS National standards from Denmark, Norway, Finland and Sweden

Legend to method comparison tables

- N total number of laboratories that reported methods and numerical results
- n number of results except false results and outliers
- Mv mean value (with outliers and false results *excluded*)
- Med median value (with outliers and false results *included*)
- CV coefficient of variation = relative standard deviation in percentage of the mean, calculated from square root transformed results
- F number of false positive or false negative results
- < number of low outliers
- > number of high outliers
- total number of results for the parameter
- 601 remarkably low result
- 278 remarkably high result or CV or many deviating results

Explanations to histograms with accepted and deviating results

- result without remark
- false negative result
- outlier
- \downarrow 34 average without deviating results
- * over a bar means that the result is beyond the x-axis limit

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General information on results evaluation

The proficiency testing program organised by the Swedish Food Agency is accredited against EN ISO/IEC 17043. This standard prescribes that results should be grouped based on the methods used. Therefore it is mandatory for participants to inform about method data. This report presents, for each parameter, method data where differences are present or could be expected.

The method information gathered is sometimes difficult to interpret. Sometimes there is inconsistency between the standard referred to and the information regarding various method details. Results from laboratories with ambiguous details are either excluded or placed in the group "Other/Unknown" in the tables, together with results from methods used only by individual laboratories. Thus, to get an as appropriate evaluation as possible of the results, it is important that correct standards and method details are reported.

Outliers and false results are not included in the calculation of mean value and measure of dispersion for the various method groups. The numbers of low and high outliers, as well as false results, are instead explicitly given in tables together with the group means etc. The mean and measure of dispersion are not shown for groups with four or fewer results, other than exceptionally when it is specifically mentioned. However, all results are shown in the method histogram when possible.

The histograms and calculation of outliers are described on page 31 under "Processing of numerical results" with further reference to the scheme protocol [1].

Results of the PT round

General outcome

Test items were sent to 78 laboratories, 32 in Sweden, 39 in other Nordic countries (Faeroe Islands, Greenland and Åland included), 3 more from EU, and 1 from the rest of Europe and 3 from outside Europe. Results were reported from 77 laboratories.

The percentages of false results and outliers are compiled in table 1.

Microorganisms and parameters of analyses are also compiled in table 1. For the MF analyses the parameters *suspected* coliform and thermotolerant coliform bacteria could be reported (shaded in table 1 and table 3). The results from suspected colonies are only used for interpretations and discussions, not for assessment.

All reported results are compiled in **annex A** and results for each laboratory are also shown on our website after logging in (<u>https://www2.slv.se/absint/</u>).

Standardized z-scores for all evaluated results are given in **annex B** and photographs with examples of colony appearance on various media are presented in **annex C**.

Table 1 *Microorganisms in each sample and percentages of deviating results (F%: false positive or false negative, X%: outliers); parameters with shaded rows are not assessed*

Sample	Α			В			C		
Percentage of laboratories with 0 deviating results 1 deviating result 2 deviating results >2 deviating results	5% 3%	79%		2% 1% 18% 7	9%		4% 3%	71%	
No. of evaluable results	498			493			495		
No. of deviating results $*$	16	(3 %)		16 ((3 %)		22	(4 %)	
Microorganisms	Escherichia colt Klebsiella oxyto Clostridium per Streptomyces sp Pseudomonas fl	oca fringe o.		Escherichia coli weak) Citrobacter freur Clostridium bifer Phialophora fast Rhodotorula min	ndii menta igiata	ins	Klebsiella pneu Acremonium str Hanseniaspora Sphingomonas s Staphylococcus	rictum uvaru sp.	т
Analysis	Target org.	F%	X%	Target org.	F%	X%	Target org.	F%	X%
Coliform bacteria (MF)	E. coli K. oxytoca	0	0	E. coli C. freundii	4	4	K. pneumoniae	2	4
Susp. thermotolerant coliform bact. (MF)	E. coli	_	-	E. coli	_	-	K. pneumoniae	_	-
E. coli (MF)	E. coli	5	2	{ <i>E. coli</i> }	#	0	—	4	_
Coliform bacteria (rapid method)	E. coli K. oxytoca	0	2	E. coli C. freundii	0	0	K. pneumoniae	0	4
E. coli (rapid meth.)	E. coli	2	2	_	0	—	—	0	-
Presumptive C. perfringens (MF)	C. perfringens	2	5	C. bifermentans	2	0	_	3	-
C. perfringens (MF)	C. perfringens	6	10	[C. biferment.]	17	0	—	7	_
Actinomycetes (MF) 25 °C	Streptomyces sp.	4	11	_	0	_	—	0	-
Moulds (MF) 25 °C	—	11	_	Ph. fastigiata	3	5	A. strictum	37	3
Yeasts (MF) 25 °C	_	5	—	Rh. minuta	0	3	H. uvarum	5	5
Culturable micro- 22 °C organisms (total count), 3 days	P. fluorescens (Strepto. sp.) (E. coli) (K. oxytoca)	0	0	E. coli C. freundii	0	0	K. pneumoniae (S. warneri) {Sphingo. sp.}	0	0
Slow-growing 22 °C bacteria (total count), 7 days	P. fluorescens (Strepto. sp.) (E. coli) (K. oxytoca)	0	0	E. coli C. freundii (R. minuta)	6	3	Sphingo. sp. K. pneumoniae (S. warneri)	0	0

* In total 41of 78 laboratories (53 %) reported at least one deviating result

21 zero results (38 %) that is not stated as false negative when detection is based on activity of β -glucuronidase

- Organism missing or numerical result irrelevant

() The organism contributes with only very few colonies

[] The organism may be presumptively false positive on the primary growth medium

{ } The organism may give different results depending on method or definition used

Coliform bacteria (MF)

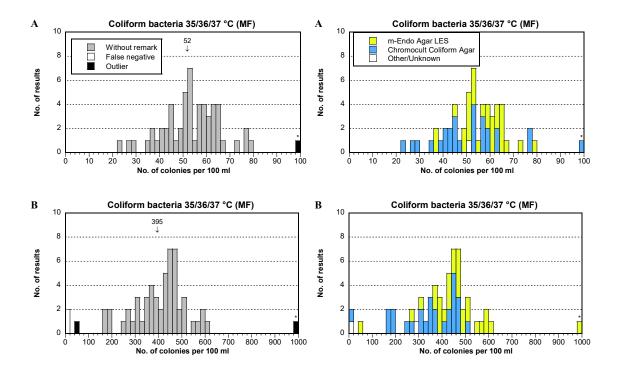
Approximately the same number of laboratories used CCA and LES. The proportion that use CCA has now stabilized since the standard EN ISO 8308-1 from 2014 was issued.

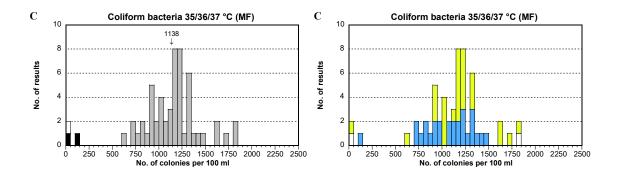
One out of two laboratories within the group Other/Unknown has used trypton glucose yeast extract agar (TGE) and incubated at room temperature for 7 days. The other laboratory reported the use of Colilert, still referring to ISO 9308-1:2014 with "Amendment". This medium is not in accordance with the standard.

As often before, CCA gave lower average result than LES, at least in sample A and B. The dispersion (CV) is at the same time larger for CCA in these samples. The results for CCA are the same as those reported as obtained by the use of the standard EN ISO 9308-1:2014 (with or without Amendment A1:2017).

In total five coliform bacteria, including E. coli, were present in the samples.

Medium	N			Α						В						С			
Medium	IN	n	Mv	CV	F	<	>	n	Mv	CV	F	<	>	n	Mv	CV	F	<	>
Total	57	55	52	12	0	0	1	53	395	14	2	1	1	54	1138	11	1	2	0
m-Endo Agar LES	28	28	56	9	0	0	0	26	448	10	0	1	1	27	1181	11	0	1	0
Chromocult C Agar	27	26	48	14	0	0	1	26	351	16	1	0	0	26	1072	10	0	1	0
Other/Unknown	2	1	_	_	0	0	0	1	_	-	1	0	0	1	_	_	1	0	0





Sample A

- A strain of *Escherichia coli* and a strain of *Klebsiella oxytoca* were included. They appeared with for coliform bacteria typical colonies on the MF media at 37 °C, a metallic sheen on LES and blue and pinkish red, respectively, on CCA.
- The distribution of the results was good with a small dispersion (CV; see page 34). One high outlier was present.

Sample B

- One strain each of *E. coli* and *Citrobacter freundii* were present as coliform bacteria. They appeared with for coliform bacteria typical colonies with a metallic sheen on LES. However, on CCA at 37 °C the colonies of *E. coli* were atypical pinkish with a violet centre while the colonies of *C. freundii* were typical pinkish.
- The distribution of the results was good with a small dispersion. Two false negative results and one low and one high outlier were present.
- It can be seen from both the table and the histogram that the results preferentially were lower for CCA compared to LES. Nothing else were noteworthy, however see the text under sample C.

Sample C

- No *E. coli* but the coliform bacteria, *Klebsiella pneumoniae*, was present. This strain appeared with for coliform bacteria, typical colonies on MF media at 37 °C, i.e. with metallic sheen on LES and pinkish on CCA.
- The distribution of the results was good with a small dispersion. One false negative result and two low outliers were present. The laboratory with the false negative result as well as one of those with a low outlier, had correspondingly low results and outcome in sample B. Thus, there seem to be a more general problem present.

Suspected thermotolerant coliform bacteria (MF)

No evaluation in relation to performance is done for what is called suspected (not confirmed) colonies of a parameter. Therefore, no identification of outliers is done. The *medians* are then more robust than the means and are given in the table and in histograms. **Thus, the parameter is not included in the performance assessment**.

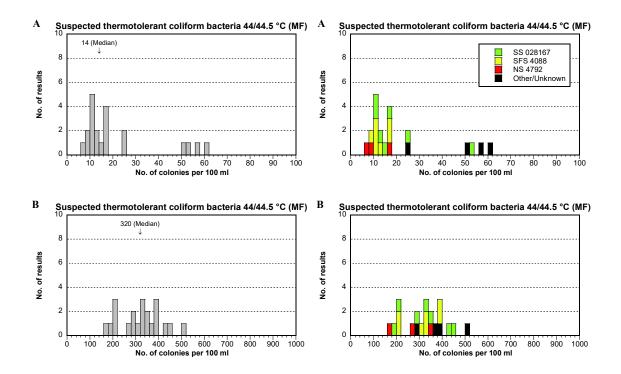
The only primary growth media used to identify suspected thermotolerant coliform bacteria at 44 or 44.5 °C is m-FC, based on various national standards. Two of the results within the group Other/Unknown are from Icelandic laboratories that have reason to state "ISO 9308-1:1990, modified" as their reference. The primary incubation is there done at 37 °C and only the confirmation at 44 °C. Thus, this is not an analysis of <u>suspected</u> thermotolerant coliform bacteria according to the definition in the instruction and is probably the reason to their high results in all samples, most obvious in sample A and B. Also the other two results per sample in the group

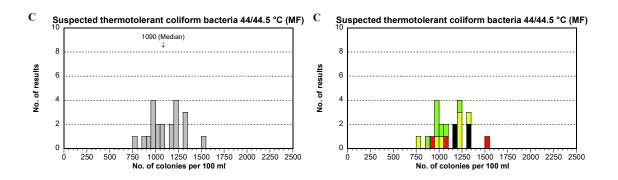
Stondard Mathed	N			Α					В					С			
Standard, Method	IN	n	Med	CV	F	< >	n	Med	CV	F <	< >	n	Med	CV	F	<	>
Total	21	21	14	-	—		21	320	—			21	1090	_	—	—	-
ISO 9308-1:2000	0	0	_	-	_		0	_	_			0	_	_	_	_	_
SS 028167	7	7	14	_	_		7	320	_			7	983	_	_	_	—
SFS 4088	7	7	10	_	_		7	320	_			7	1200	_	_	_	—
NS 4792	3	3*	8	_	_		3*	270	_			3*	1090	_	_	_	—
Other/Unknown	4#	4^*	54	_	_		4^*	370	_			4*	1246	_	_	_	_

Med = Median; used here instead of mean value because it describes "suspected" colonies

^{*t*} Probably, all these laboratories have used < 44 °C in their primary incubation step, see the text

* Median are given for comparison despite few results





Other/Unknown seem to have used a "none-thermotolerant method", as they claim they don't test suspected thermotolerant coliform bacteria specifically. Thus, they have probably reported results from incubation at 36 ± 2 °C. The strain of *K. oxytoca* in sample A seem here at least to be included in the result from one of the Icelandic laboratories and in the results from the other two laboratories in the group Other/Unknown. For the strain *C. freundii* in sample B it is more difficult to see. Both these none-thermotolerant strains may sometimes grow at near 44 °C but usually not at 44 °C.

Sample A

- Only the strain of *E. coli* appears as a typical suspected thermotolerant coliform bacteria, with blue colonies on m-FC agar at 44/44.5 °C. *K. oxytoca* will also grow with typical colonies if a lower temperature is used for a medium. This seems to be applicable to the four highest results, out of which three belong to the group Other/Unknown.
- The distribution of the results, except the four high ones, was good.

Sample B

- The strain of *E. coli* appears with blue colonies on m-FC at 44/44.5 °C. The strain of *C. freundii* does not normally grow at this temperature. Several of the highest results are probably obtained by use of a lower incubation temperature, as mentioned above (see sample A), but it is more difficult to see here where the number of *C. freundii* colonies only comprise 1/3 of the coliform bacteria.
- The distribution of the results was quite good.

Sample C

- *K. pneumoniae* was the only coliform bacterium in the sample. It appears also as a typical suspected thermotolerant coliform bacterium with blue colonies on m-FC agar at 44/44.5 °C.
- The distribution of the 21 results was generally good.

Escherichia coli (MF)

To identify and quantify *E. coli*, confirmation is required when colonies are isolated from the primary cultivation media LES or m-FC. Depending on the method, tests for indole production and/or β -glucuronidase activity from oxidase negative presumptive strains are usually performed. A violet to blue colony on CCA indicates positive β -glucuronidase activity and is reckoned as a confirmed *E. coli*.

The primary growth media are CCA and LES that are used at 36 ± 2 °C and m-FC that is used at 44/44.5 °C. In addition to incubation temperature, the results are also grouped by standard. For ISO 9308-1:2014 the incubation is at 36 ± 2 °C on CCA. For the standards from the Nordic countries (SS, SFS and NS) the majority of the results are from 36 ± 2 °C on LES but some are also from 44/44.5 °C on m-FC. Only one Finnish laboratory have stated the standard SFS 4088 (m-FC) instead of SFS 3016 for the analysis of *E. coli*.

It is obvious that the results in sample A based on Finnish standard were much higher than those based on other standards. In sample B, the results with the Swedish standard were the highest. See the discussion below. There is an indication that the results based on ISO 9308-1:2014 are the lowest even when all results are compared. It is more obvious from the second table, where incubation temperature is 36 ± 2 °C, that results (without deviating ones) are lower with CCA compared to with LES.

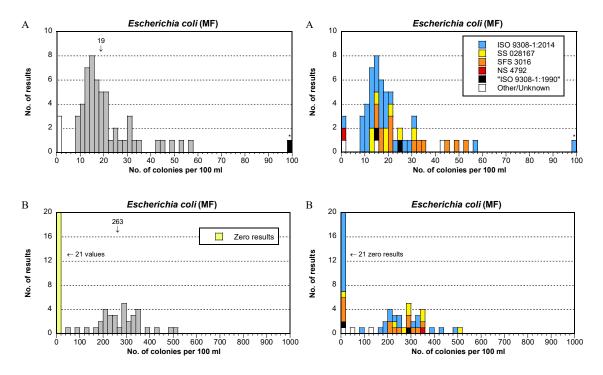
Origin &Standard	Ν			Α						В						С			
Origin & Standard	1	n	Mv	CV	F	<	$^{\prime}$	n	Mv	CV	F	<	<	n	Mv	CV	F	<	>
Total	57	53	19	25	3	0	1	35	263	20	21	0	0	55	0	_	2	_	—
<u>Colony origin</u>																			
$36 \pm 2 \ ^{\circ}\text{C}$	42	39	19	26	2	0	1	25	274	19	16	0	0	42	0	_	0	_	_
44/44.5 °C	6	5	17	13	1	0	0	4*	269	11	2	0	0	6	0	_	0	_	_
36 ± 2 & 44/44.5 °C	9	9	23	24	0	0	0	6	216	28	3	0	0	7	0	_	2	_	_
Other/Unknown	0	0	_	_	_	_	_	0	-	_	-	_	_	0	_	_	_	_	—
<u>Standard</u>																			
ISO 9308-1:2014	30	28	16	24	1	0	1	15	259	20	14	0	0	30	0	_	0	_	_
SS 028167	8	8	18	16	0	0	0	7	317	14	1	0	0	8	0	_	0	_	_
SFS 3016 (4088)	13	13	27	24	0	0	0	9	270	9	4	0	0	13	0	_	0	_	_
NS 4792	1	0	_	_	1	0	0	1	_	_	0	0	0	1	_	_	0	_	_
"ISO 9308-1:1990"	2	2	_	_	0	0	0	1	_	_	1	0	0	1	0	_	1	_	_
Other/Unknown	3	2	-	_	1	0	0	2	-	_	1	0	0	2	0	_	1	_	_

All results

Results from the analysis of "coliform bacteria" MF at 36 ± 2 °*C*

Madium	N			Α						В						С			
Medium	IN	n	Mv	CV	F	<	\vee	n	Mv	CV	F	<	$^{\prime}$	n	Mv	CV	F	< 3	>
Total	44 [#]	41	18	26	2	0	1	26	273	19	17	0	0	44	0	-	0		_
m-Endo Agar LES	15	15	22	27	0	0	0	12	332	13	3	0	0	15	0	_	0	_	_
Chromocult C Agar	27	26	17	25	0	0	1	13	236	18	13	0	0	27	0	_	0	_	_
Other/Unknown	2	0	_	_	2	0	0	1	-	_	1	0	0	2	0	_	0	_	_

Compare the table above – two more laboratories referring to ISO 9308-1:2014 (incubation at 36±2 °C) have been included, although one is claiming the use of both both 36 and 44 °C and the other only 44 °C



Sample A

- A typical strain of *E. coli* was present together with another coliform bacterium, *K. oxytoca*. The latter doesn't show any β -glucuronidase activity but is indole positive and, thus, could sometimes be taken for *E. coli* after confirmation.
- The distribution of the results was fairly good except a "tail" of 5 high results. The dispersion was, therefore, median (CV; see p. 34). Three false negative results and one high outlier were present.
- The high results, corresponding to the average of coliform bacteria (see p. 6), could be the case when a positive indole reaction is the criterion for *E. coli*. *K. oxytoca* could then have been included after growth in broth at 44 °C and a positive reaction in the indole test.
- For one of the three zero results, "Colilert" has been stated, for another a "medical standard" and for the last the incubation was at 44.5 °C. Thus, it could be the primary incubation that caused all the zero results.

Sample B

- A strain of *E. coli* with weak β -glucuronidase activity was included together with another coliform bacterium, *C. freundii*. The colonies of *C. freundii* are typical for a coliform bacterium, with a metallic sheen on LES and pink on CCA. The colony appearance of *E. coli* is typical on LES and m-FC that are based on lactose fermentation. However, on the chromogenic enzyme-based medium CCA the colony colour is atypical for *E. coli*. On this medium the colonies are pinkish with a more or less evident violet hue in the middle. It seems that these colonies are often (see below) interpreted as coming from another coliform bacterium than *E. coli*, leading to a zero result for *E. coli*. Further confirmation for *E. coli* is not

required on CCA. However, for colonies from LES and m-FC confirmation is necessary to discern *E. coli* from other coliforms.

- Twenty-one zero results were reported. The distribution was otherwise in general fairly good with a relatively small dispersion (CV = 20 %). There is a tendency to a tail with other low results. The dispersion was higher with ISO 9308-1:2014 than with other media. In the histogram the average is given for the results except the zero results.
- Fourteen of the 21 zero results were obtained with CCA, five by use of the Nordic standards based on lactose fermentation and confirmation and two by use of other methods (out of which one stated ISO 9308-1:2014 but claimed "Colilert" as medium).
- The strain of *E. coli* is producing gas in lactose broth at 44 °C, is positive when testing for indole production but show a weak β -glucuronidase activity. A negative outcome is probably an interpretation when β -glucuronidase activity is the only decisive criterion for *E.coli*. This applies to confirmation with MUG reagent in broth as well as to the use of enzyme-based chromogenic media like CCA.
- Zero results obtained due to an interpretation of the β -glucuronidase activity as negative is acceptable even though they are indicated as false negative in the table and in Annex A. However, zero results by other reasons than negative β glucuronidase activity should be seen as real false negative ones.

Sample C

- A strain of the thermotolerant coliform bacterium, *K. pneumoniae*, was present. Its colonies have a typical metallic sheen on LES and are blue on m-FC, both media based on lactose fermentation. The colonies are typical pink on the chromogenic enzyme-based medium CCA. The strain is indole-negative and is lacking activity of β -glucuronidase, making it impossible to mistake it for *E. coli* even after confirmation from LES and m-FC.
- No false positive result was reported.

Coliform bacteria & E. coli (rapid methods, MPN)

The rapid method used for both these parameters was exclusively Colilert[®] Quanti-Tray[®] from the manufacturer IDEXX Inc. with incubation at 35, 36 or 37 °C. Out of the about 50 laboratories that reported Colilert some used trays with 51 wells, while others used trays with 97 wells (a few of which, probably incorrectly, have reported 96 wells). The laboratories often analysed both diluted and undiluted samples. Yellow wells (ONPG positive; β -galactosidase activity shown) will be interpreted as coliform bacteria and yellow wells also exhibiting fluorescence (MUG positive; β glucuronidase activity shown) will be interpreted as *E. coli*.

When comparing different temperatures and maximum lengths of incubation it is clear that the differences are small and inconsistent. No differences based on these criteria are therefore given. This time, not even the group (one laboratory) with incubation for 24 hours showed lower results, which has been the case previously.

There is usually only small differences in the results based on number of wells on the trays. However, a somewhat lower average for coliform bacteria can at least this time be seen is sample B when using 51 wells.

There is no indication of interpretation difficulties in any sample.

Dringinlo	Ν			Α						В						С			
Principle	IN	n	Mv	CV	F	<	>	n	Mv	CV	F	<	>	n	Mv	CV	F	<	>
Total, Rapid meth.	52	51	59	8	0	0	1	49	450	11	0	0	0	48	1257	7	0	1	1
Colilert-18, 51 wells	9	8	56	4	0	0	1	7	390	10	0	0	0	7	1237	3	0	0	0
Colilert-18, 97 wells	40	40	60	9	0	0	0	39	458	10	0	0	0	38	1256	7	0	1	1
Colilert-18, 51 & 97	1	1	_	_	0	0	0	1	_	_	0	0	0	1	_	_	0	0	0
Colilert-24, ? wells	1	1	_	_	0	0	0	1	_	_	0	0	0	1	_	_	0	0	0
Wrong method [#]	1	1	_	_	0	0	0	1	_	_	0	0	0	1	_	_	0	0	0

Coliform bacteria, Rapid method with MPN

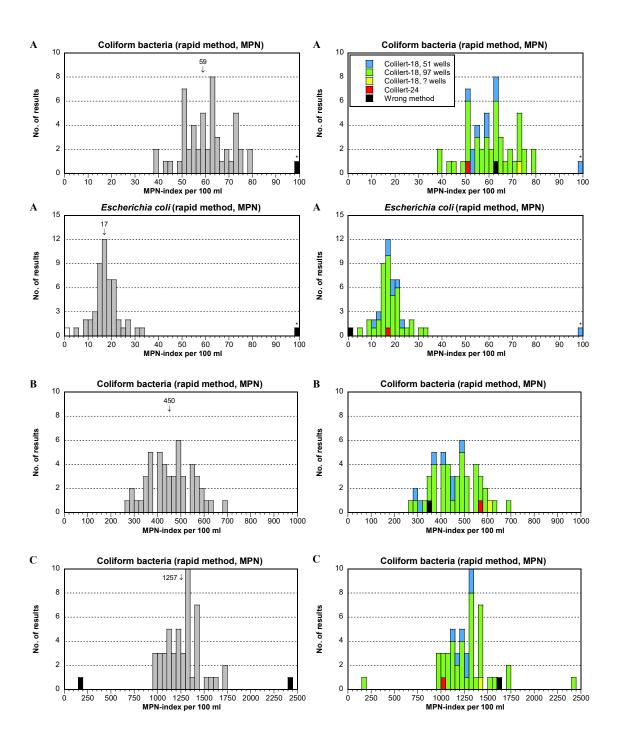
E. coli, Rapid method with MPN

Principle	N			Α						В						С		
rmcipie	14	n	Mv	CV	F	<	\vee	n	Mv	CV	F	<	>	n	Mv	CV	F	< >
Total, Rapid meth.	52	50	17	15	1	0	1	49	0	_	2	0	0	52	0	_	0	
Colilert-18, 51 wells	9	8	17	11	0	0	1	9	0	_	0	0	0	9	0	_	0	
Colilert-18, 97 wells	41	41	17	16	0	0	0	38	0	_	2	0	0	41	0	_	0	
Colilert-18, 51 & 97	0	0	_	_	_	_	_	0	_	_	_	_	_	0	0	_	_	
Colilert-24, ? wells	1	1	-	_	0	0	0	1	0	_	0	0	0	1	0	_	0	
Wrong method [#]	1	0	-	_	1	0	0	1	0	-	0	0	0	1	0	_	0	

No rapid kit method but a multiple tube method based on lactose fermentation,

Sample A

The strains of *E. coli* and *K. oxytoca* grow in the medium and possess the enzyme β -galactosidase. Therefore, they are detected as coliform bacteria by methods based on this enzyme (ONPG positive) e.g. Colilert[®]-18/24 Quanti-Tray[®] where ONPG is a substrate.



- Only the strain of *E. coli* possesses the enzyme β -glucuronidase and is detected as *E. coli*.
- The distributions of the results were good and the dispersion (CV; see p. 34) very small for coliform bacteria and small for *E. coli*. There was one high outlier from the same laboratory in each of the analyses. There was also one false negative result for *E. coli*.

- The averages for coliform bacteria was, as often, higher (13 %) with the rapid method compared to the MF method. Unusually it was 11 % lower for *E. coli*. This was probably caused by the fact that *K. oxytoca* in some cases were included when the MF method was used (compare p. 6 and 10).

Sample B

- The two coliform bacteria *E. coli* and *C. freundii* were present. Both have the enzyme β -galactosidase (ONPG positive) and are detected as coliform bacteria.
- The strain of *E. coli* possesses β -glucuronidase but the activity is so low that the fluorescens is usually interpreted as negative. Two non-zero results were present, still quite low compared to with the MF method. Here, in the light of the other results, they are judged as false positive ones.
- The average result for coliform bacteria was here 14 % higher than it was with the MF methods (compare p. 6).
- The distribution of the results was good with small, almost very small, dispersion in average. No deviating results were present for coliform bacteria but two false positive results for *E. coli*.

Sample C

- The strain of *K. pneumoniae* is the only coliform bacterium growing in the medium. It has the enzyme β -galactosidase and is detected as a coliform bacterium by methods based on this enzyme (ONPG positive) e.g. Colilert[®]-18/24 Quanti-Tray[®] where ONPG is a substrate.
- The strain of *K. pneumoniae* is lacking the enzyme β -glucuronidase and is <u>not</u> detected as *E. coli*.
- The distribution of the results was very good with very small dispersion in average. One low and one high outlier were present.
- The average result for coliform bacteria was here 10 % higher than it was with the MF methods (compare p. 6).

Presumptive and confirmed Clostridium perfringens (MF)

The analysis of *Clostridium perfringens* has some years ago been performed differently in different countries and laboratories. The parameter to be analysed is the sum of spores and vegetative cells of *C. perfringens*. In Sweden presumptive *C. perfringens* are accepted, which is why that parameter is presented separately.

In an annex [6] to the European Drinking Water Directive from 1998 [4] it is stated that the standard EN ISO 14189:2016 with its national editions must be used after October 2017. The identical version ISO 14189 from 2013 can alternatively be used. After the presumptive test on TSC agar (TSC) a test for acid phosphatase activity should be performed on isolated colonies as confirmation for *C. perfringens*.

Before 2017 there was no international standard stated as reference method in the EU Directive [4]. A specific method was instead explicitly included into the directive, the use of m-CP Agar incubated at 44 °C. The method includes a confirmation step with ammonia vapour, where a red coloration of colonies indicates *C. perfringens*.

Due to the hesitation in many countries to use the m-CP method, the use of a standard still under process (ISO/CD 6461-2:2002-12-20), based on TSC, was accepted as an alternative by the responsible group under the EU Commission, until a finished standard was available. Small adjustments in the draft were approved during the standardization process.

In the spring 2018, 24 % of the laboratories still used either of the old methods. The figure is now 19 % (10 of 53 laboratories) which is comparable to in the spring 2019 (18 %). Only one laboratory has now stated the use of the m-CP for *Clostridium perfringens*. That result was not different from those by other methods.

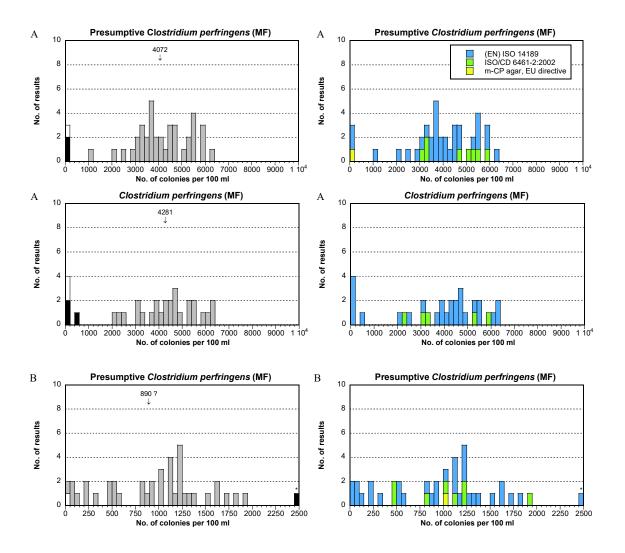
There is a tendency to lower results with (EN) ISO 14189 compared to ISO/CD 6461-2:2002-12-20 for presumptive *C. perfringens* in samples A and B. For *C. perfringens* in sample A the outcome is the opposite, with no clear reason. By checking the method data, it is clear that four of the laboratories referring to the older

Standard/Method	N #			Α						В						С			
Standard/Method	IN	n	Mv	CV	F	<	$^{\prime}$	n	Mv	CV	F	<	>	n	Mv	CV	F	<	>
Total	53	38	4072	15	1	2	0	40	890	37	1	0	0	39	0	_	1	_	-
(EN) ISO 14189	43	30	3978	15	0	2	0	31	866	41	1	0	0	30	0	_	1	_	_
ISO/CD 6461-2:2002	9	8	4434	13	0	0	0	8	972	23	0	0	0	8	0	_	0	_	_
m-CP agar, EU-direct.	1	0	_	_	1	0	0	1	_	_	0	0	0	1	0	_	0	_	_

Presumptive Clostridium perfringens MF

Standard/Method	N#			Α						В						С			
Standard/Wiethod	IN	n	Mv	CV	F	<	>	n	Mv	CV	F	<	>	n	Mv	CV	F	<	>
Total	53	26	4281	15	2	3	0	25	0	_	5	_	-	28	0	_	2	—	-
(EN) ISO 14189	43	21	4396	13	2	3	0	21	0	_	4	_	-	23	0	_	2	_	_
ISO/CD 6461-2:2002	9	5	3811	20	0	0	0	4	0	_	1	_	_	5	0	_	0	_	_
m-CP agar, EU-direct.	1	0	-	-	_	_	_	0	_	_	_	_	_	0	_	_	_	_	_

[#] The sum of laboratories that have reported results for presumptive C. perfringens, and/or C. perfringens



method has stated the use of acid phosphatase, i.e. they have in fact used (EN) ISO 14189. The other five laboratories have used other confirmation techniques.

Sample A

- A strain of *C. perfringens* was included. The colour of the colonies on TSC could vary from pale grey-brown to completely black depending on the condition and reduction potential of the medium.
- A few false negative results and other low results could be noted for both the presumptive test and for *C. perfringens*.
- The distribution of the results was, as the two last years, relatively good for both presumptive and confirmed *C. perfringens*, without the earlier occurring tail of low results. The reason is probably that there is in principle no results for m-CP agar that earlier gave considerably lower results than TSC. The dispersion (CV) was also this time not higher than for other parameters, but small (see p. 34).
- The two lowest outliers for *C. perfringens* were reported already in the presumptive test. No presumptive test was done in cases where other low deviating results occurred. The strain of *C. perfringens* used here sometimes gives

a weak colour reaction (dark purplish brown rather than violet) in the acid phosphatase test, usually not obvious until after 4 minutes. Still most laboratories identified the strain as *C. perfringens*.

Sample B

- No *C. perfringens* was included but instead a strain of *C. bifermentans*. The strain appeared on TSC with small, black to almost transparent presumptive colonies. Confirmation reveals that they are not *C. perfringens*.
- There is only a weak tendency to a Poisson distribution of the presumptive results due to the many low values. The dispersion (CV) was large to very large implying that no outliers could be identified. One false negative result and one high outlier were obtained.
- As many as 5 false positive results were present in the analyses of *C. perfringens*. Either has no confirmation been made and the colonies then been taken for *C. perfringens* or have the confirmation outcome been misinterpreted.

Sample C

- No presumptive *C. perfringens* was included. Still, 1 false positive result was present for presumptive *C. perfringens* and two for confirmed.

Moulds and yeasts (MF)

Out of the 38 laboratories that analysed moulds and yeasts, 26 reported that they used the Swedish standard SS 028192. In addition to Sweden, it is used in Finland under their own national designation SFS 5507. Sometimes it is modified regarding media composition as for example dichloran (DRBC) is used.

Various names are reported for the media linked to the use of SS 028192 and SFS 5507. These are: Cooke Rose Bengal Agar base, Rose Bengal Agar, Rose Bengal Chloramphenicol Agar and Dichloran Rose Bengal Chloramphenicol Agar (DRBC). According to the original standard, dichloran should not be an ingredient (and thus DRBC should not be used) but instead Rose Bengal and the two stronger inhibitory substances chlortetracycline and chloramphenicol. Both of them are used by at least 16 of the 20 Swedish laboratories. Here is shown what the laboratories have really stated, and a separation is made for those that have used any form of "Rose Bengal Agar" (RBC) and the five laboratories from various countries stating DRBC in conjunction with SS 028192 or SFS 5507 – or in one case "Standard methods" [5] – (DRBC "Water").

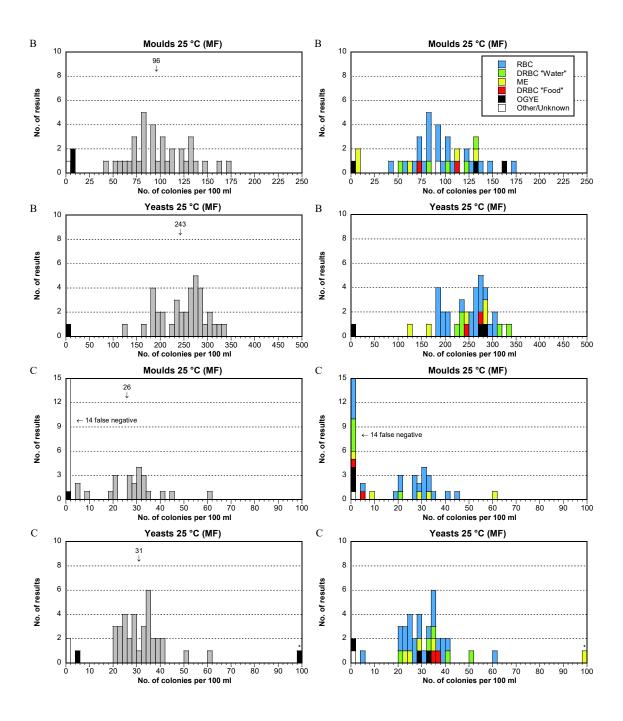
Two Norwegian laboratories instead stated NMKL 98:2005, modified to be used with DRBC. This comprises the group DRBC "Food" in the tables. Four Finnish laboratories, one in conjunction with NMKL 98:2005, together with a laboratory from Tanzania used Malt Extract Agar. These five laboratories are placed in the group ME. Two Finnish and one Danish laboratory using Oxytetracycline Glucose Extract Agar based on other methods/standards are placed in the group OGYE. In several of these groups there are so few results (<5) that it is not meaningful to discuss possible differences. But the mean values are still given for comparison.

Standard/Method	N			Α					В						С			
Standaru/Method	IN	n	$\mathbf{M}\mathbf{v}$	CV	F	< >	n	Mv	CV	F	<	>	n	Mv	CV	F	<	>
Total	38	34	0	_	4		35	96	16	1	2	0	23	26	25	14	1	0
RBC	22	20	0	_	2		22	93	16	0	0	0	17	27	19	5	0	0
DRBC "Water"	5	5	0	_	0		5	94	18	0	0	0	1*	21	_	4	0	0
ME	5	3	0	_	2		3*	98	_	0	2	0	4^*	29	_	1	0	0
DRBC "Food"	2	2	0	_	0		2*	89	_	0	0	0	1*	5	_	1	0	0
OGYE	3	3	0	_	0		2*	145	_	1	0	0	0	_	_	2	1	0
Other/Unknown	1	1	0	_	0		1	_	_	0	0	0	0	_	_	1	0	0

Yeasts MF

Standard/Method	N			Α					В						С			
Stanuaru/Wiethou	IN	n	Mv	CV	F	< >	n	Mv	CV	F	<	>	n	Mv	CV	F	<	$^{\prime}$
Total	38	36	0	_	2		37	243	10	0	1	0	33	31	13	2	1	1
RBC	22	21	0	_	1		22	238	9	0	0	0	20	31	14	0	1	0
DRBC "Water"	5	5	0	_	0		5	262	10	0	0	0	5	35	16	0	0	0
ME	5	4	0	_	1		5	212	18	0	0	0	4*	28	_	0	0	1
DRBC "Food"	2	2	0	_	0		2^*	255	_	0	0	0	2*	35	_	0	0	0
OGYE	3	3	0	_	0		2*	275	_	0	1	0	2*	30	_	1	0	0
Other/Unknown	1	1	0	_	0		1	-	_	0	0	0	0	_	_	1	0	0

* Mean value is given for comparison despite few results



This time, DRBC "Water" does not appear to give higher results for neither moulds nor yeasts compared to RBC in sample B, but possibly for yeasts in sample C. In the four Finnish cases where ME were used, the ME medium has been supplemented with a selective substance (chloramphenicol or streptomycin). Only the Tanzanian laboratory seems to use ME unselectively. The average results for ME seem to be somewhat lower for yeast in both sample B and C. However, there are too few results for a conclusion. There were relatively many deviating results when OGYE was used.

Sample A

- Neither moulds nor yeasts were included. Still, four false positive result was reported for moulds and two false positive results for yeasts. In one case for yeasts, where only 1 colony was found, it can be due to a contamination from the laboratory air. Such results should not be seen as false positive ones.

Sample B

- The mould *Phialophora fastigiata* and the yeast *Rhodotorula minuta* were included, with the yeast colonies being about twice as many. No apparent problem could be seen and the distributions of the results were good with small dispersions (CV; see p. 34) in both cases. There is a tendency to a second peak with fewer colonies for the yeasts.
- One false negative result and two low outlier were present for the moulds and one low outlier for the yeasts.

Sample C

- The mould *Acremonium strictum* and the yeast *Hanseniaspora uvarum* were included in about the same concentrations. With the exception of the many zero results for moulds and other low deviating results, the result distributions were relatively good. The relative dispersion (CV) of the accepted results was medium for moulds and small for yeasts.
- Except for the fourteen false negative results four moulds and the two for yeasts, only single outliers were present, one low for moulds and one low and one high for yeasts.
- The reason to the zero results is probably the small, undeveloped colonies with very pale mycelium without mature spores (colourless colonies) after 7 days of incubation. This highlights the importance of using magnification when reading the plates.

Actinomycetes (MF)

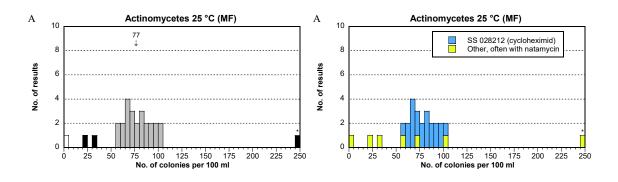
The analysis of actinomycetes is included because it is a prescribed method for drinking water monitoring according to the Swedish regulations. Therefore, it is mainly Swedish laboratories that perform the analysis. They do it according to the Swedish standard for actinomycetes in water, SS 028212 (1994). Seven Finnish laboratories that have performed the analysis based on other methods are placed in the group Other. Six of these have stated that they used natamycin as the selective substance instead of cycloheximide. The last laboratory has stated the use of Rose Bengal, chloramphenicol and chlortetracycline as in the standard for fungi in water SFS 5507, but has incubated for up to 14 days. The base agar medium varies also within the group Other but is in all cases different from Actinomycete Isolation Agar (ACTA) that is the base medium in the Swedish standard. The Finnish laboratories noted their results after 7 and 14 days.

The averages but also the dispersions (CV) of the two groups ACTA and Other in sample A are very similar. However, there were many deviating results in the group Other. As always, the outcome applies to the specific target strains that are present, and cannot be considered to be generally valid.

All results

Medium/Standard	N			Α						В					С		
Medium/Standard	IN	n	Mv	CV	F	<	\vee	n	Mv	CV	F	< >	n	Mv	CV	F	< >
Total	28	24	77	9	1	2	1	28	0	_	0		28	0	_	0	
ACTA (SS 028212)	21	21	77	8	0	0	0	21	0	_	0		21	0	_	0	
Other	7	3*	75	_	1	2	1	7	0	_	0		7	0	_	0	

* Mean value is given for comparison despite few results



Sample A

- One actinomycete from the group *Streptomyces* sp. was included. The distribution of the results was good and the average dispersion very small (see p. 34).
- One false negative result and two low and one high result was present, all from the group Other, where methods other than the Swedish standard were used.

Samples B and C

- These samples contained no actinomycetes and there were no false positive results.

Culturable microorganisms 22 °C, 3 days

Seventy of the 73 laboratories performing the analysis reported EN ISO 6222:1999 as method, which prescribes the use of Yeast extract Agar (YEA). Seven laboratories used Plate Count Agar instead stating the use of EN ISO 6222:1999. One laboratory used YEA in conjunction with Standard methods [5]. The majority of the laboratories have claimed counting both bacterial and fungal colonies. Ten laboratories state that they don't count fungi and two more that they count yeasts but not moulds.

Since all except three laboratories refer to EN ISO 6222:1999, differences among method variants are relevant to discuss only for these. Results are shown for culture media and magnification at reading.

As usual, it is difficult to find any consistent method differences. In sample A Plate Count Agar gave, as sometimes before, lower result than YEA. Also the dispersion (CV) for these results were smaller this time. Sometimes, PCA has given higher results and/or larger dispersion. Probably, this depends on the organisms that are present in the samples. For samples B and C it is impossible to see any differences.

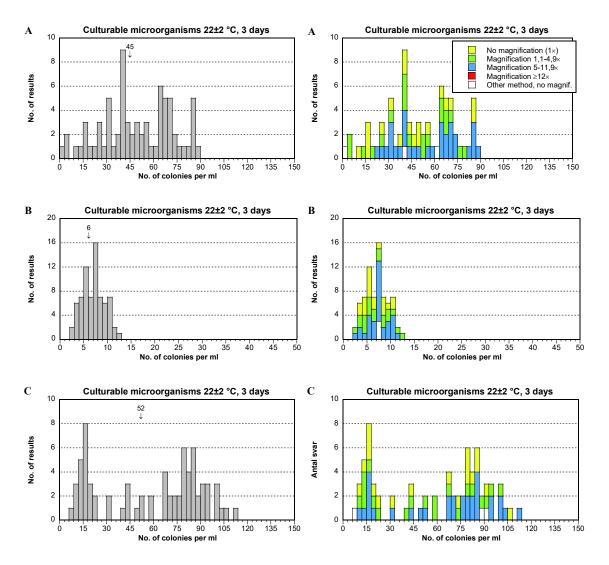
There were some differences present in the result averages of sample A and C in relation to magnification. They were highest in the group with the highest used magnification $(5-11.9\times)$. The dispersion was there also the smallest, in particular in sample A. The culturable microorganisms were probably easy to count in samples A and B. However, in sample C small colonies of *Sphingomonas sp.* seem to have occurred already after 3 days of incubation. Often they are not visible until the fourth day of incubation, when magnification is used.

Sample A

- The colonies consist mainly of a strain of *Pseudomonas fluorescens* but also the other bacteria may appear with single colonies.
- The distribution of the results was not good but dispersed with a tendency to two peaks. No outliers could therefore be identified.

Cuour of regults	Ν			Α						В						С			
Group of results	1	n	Mv	CV	F	<	\vee	n	Mv	CV	F	<	>	n	Mv	CV	F	<	>
Total, all results	73	73	45	27	0	0	0	73	6	19	0	0	0	73	52	34	0	0	0
EN ISO 6222	70	70	46	26	0	0	0	70	6	19	0	0	0	70	52	33	0	0	0
<u>Medium</u>																			
Yeast extract Agar	63	63	47	26	0	0	0	63	6	18	0	0	0	63	52	34	0	0	0
Plate Count Agar	7	7	38	18	0	0	0	7	5	29	0	0	0	7	52	31	0	0	0
Other /Unknown	0	0	-	_	_	_	_	0	_	_	_	_	—	0	_	_	_	_	—
Magnification																			
None	19	19	41	28	0	0	0	19	6	19	0	0	0	19	48	36	0	0	0
1.1–4.9×	20	20	41	33	0	0	0	20	6	23	0	0	0	20	46	36	0	0	0
5–11.9×	31	31	54	19	0	0	0	31	7	18	0	0	0	31	59	30	0	0	0
> 12×	0	0	_	_	_	_	_	0	_	_	_	_	_	0	_	-	_	_	—
Other method	3	3*	26	_	0	0	0	3*	7	_	0	0	0	2*	52	_	0	0	0

* Mean value is given for comparison despite few results



- The two peaks are likely an effect of that laboratories with lower magnification (<5×) having counted fewer colonies of *P. fluorescens* than those with higher magnification. In general, the results are lower compared to the results from the Swedish Food Agency (see p. 32). The general outcome is not affected by the other strains in the sample since they only contribute with occasional colonies.

Sample B

- The colonies are comprised of all bacterial and fungal strains except *C. bifermentans*, but in practice mainly *E. coli* and *C. freundii*. No effect of magnification can be seen with the few colonies present.
- The distribution of the results was good with small dispersion (see p. 34). No deviating results were present.

Sample C

- The colonies mainly consisted of *K. pneumoniae* but for some laboratories probably also of *Sphingomonas sp.* Moulds and yeasts, as well as *Staphylococcus warneri*, may contribute with occasional colonies.

- The distribution of the results was not good but dispersed with two distinctive peaks. No outliers could be identified because of this. The common average is in the middle between the two peaks and gives no meaningful information.
- The first peak (modus: 13-15 cfu/ml) is indicating results were only *K. pneumonia* have been counted. This is clearly seen from the outcome of 13 cfu/ml for coliform bacteria with the rapid method. The second more dispersed peak (modus: 79-87 cfu/ml) is then obtained primarily as the colony sum of *K. pneumonia* and *Sphingomonas sp.* As the colonies of the latter must be very small after only 3 days of incubation (usually not visible until the fourth day) the interpretation is that the differences in outcomes are caused by that strain. The magnification has an impact to some extent, as in sample A. A second cause may be that the colonies are counted after somewhat varying incubation times, implying no visible colonies or a varying number of visible colonies. Of course, indirectly, visibility is also an effect of magnification.
- Also during the 1990s, small slow-growing bacteria were a problem by giving varying results in the analyses of "total aerobic count" after 3 days of incubation at 20-21 °C compared to after 2 days of incubation (that was used in Sweden before 2003). Even then the magnification at reading was reckoned as part of the problem.

Slow-growing bacteria 22 °C, 7 days

Thirty-two laboratories have performed the analysis of slow-growing bacteria. The parameter is mandatory to monitor according the Swedish Drinking water ordinances and therefore there is a method stated for it for Swedish laboratories. Today a modification of the method in the standard EN ISO 6222:1999 based on the medium yeast extract agar (YEA) is used. The modification includes: incubation at 22 ± 1 °C for 7 days, at least 4× (preferentially 10×) magnification should be used when reading the plates and only bacterial colonies should be counted.

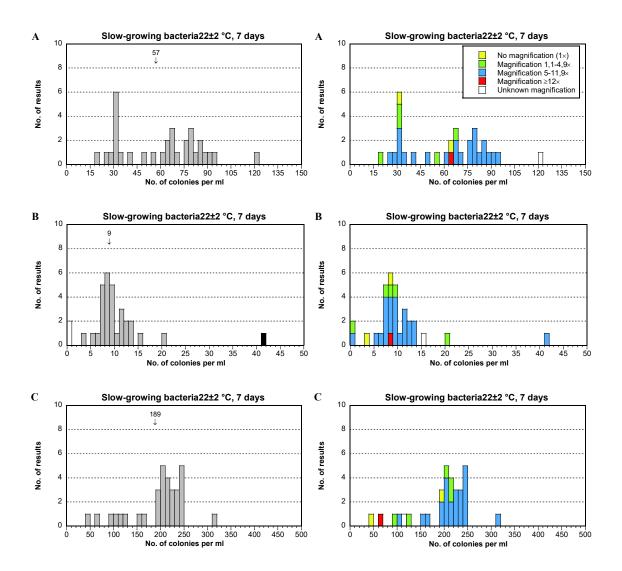
Within ISO, work is ongoing to initiate a standardization of a method for the parameter "slow-growing microorganisms". Currently, the proposal is to use a more nutrient depleted medium than YEA, namely "Reasoner's 2 Agar" (R2A). A consequence is that the majority of the laboratories that here not used YEA instead used R2A, which therefore is one of the groups in the table and the histograms. One laboratory has not stated the medium they used.

Twenty-one laboratories claim they don't include fungal colonies, while six (three with R2A) are claiming they are including both moulds and yeasts and five (one with R2A) that they are including yeasts only.

For both sample A and C it is clear that the few laboratories using R2A have reported lower results in average compared to those using YEA. However, these differences probably don't pertain to the medium used as they are even more pronounced when different magnifications are compared. In average 20 more colonies (50 %) were counted when the magnification $5-11.9 \times$ was used compared to when lower or no

Chann of hoghit	Ν			Α						В						С			
Group of result	IN	n	Mv	CV	F	<	>	n	Mv	CV	F	<	>	n	Mv	CV	F	<	>
Total, all results	32	32	57	23	0	0	0	29	9	17	2	0	1	32	189	18	0	0	0
<u>Medium</u>																			
Yeast extract Agar	26	26	60	21	0	0	0	25	9	12	0	0	1	26	203	14	0	0	0
"Reasoner's 2 Agar"	5	5	52	31	0	0	0	3*	13	_	2	0	0	5	159	15	0	0	0
Other/Unknown	1	1	_	_	0	0	0	1	_	_	0	0	0	1	_	_	0	0	0
Magnification																			
None	2	2^{*}	46	_	0	0	0	2*	5	_	0	0	0	2*	105	_	0	0	0
1.1–4.9×	5	5	38	25	0	0	0	4^*	10	26	1	0	0	5	166	19	0	0	0
5–11.9×	23	23	61	21	0	0	0	21	9	13	1	0	1	23	214	10	0	0	0
\geq 12×	1	1	-	-	0	0	0	1	-	-	0	0	0	1	_	_	0	0	0
Unknown	1	1	-	_	0	0	0	1	_	_	0	0	0	1	_	_	0	0	0

* Mean value is given for comparison despite few results



magnification was used in sample A. In sample C, however, about 65 more colonies (44 %) were counted when the higher magnification was used. Further, the dispersions (CV) were smaller both in samples A and C when the higher magnification was used, and even so in sample B.

In both samples A and B the absolute difference in recovery are not particularly large between culturable microorganisms and slow-growing bacteria, 10 and 33 % higher, respectively, for the latter group. However, in sample C the number of counted colonies is in average more than tripled. Only this sample contained the so called slow-growing bacteria and the outcome indicates the problem with equal evaluation of them. The colonies are often small and therefore difficult to count without magnification, which explains the higher absolute difference in recovery between the methods in sample C.

Sample A

- The colonies consist mainly of a strain of *P. fluorescens* but also the other bacteria may appear with occasional colonies.
- The distribution of the results was dispersed with a tendency to two peaks, approximately in the same way as for culturable microorganisms. Also the dispersion was approximately the same, which is medium. No outliers could be identified because of the wide dispersion.
- Here, as for culturable microorganisms, low magnification has been negative to the quantification of colonies.

Sample B

- The colonies are comprised of all bacterial and fungal strains except *C. bifermentans*, but in practice mainly *E. coli* and *C. freundii*. Perhaps, yeast colonies may also have been counted after 7 days of incubation. They are usually difficult to distinguish visually form bacterial colonies. No effect of magnification can be detected with this low number of rather big colonies.
- The distribution of the results was good with small average dispersion (see p. 34). Two false negative results and one high outlier were present.

Sample C

- The colonies mainly consisted of *Sphingomonas sp.* but also to some degree of *K. pneumoniae* and perhaps some occasional yeast colonies.
- The distribution of the results was much better than for culturable microorganisms with only one peak and some lower results. The dispersion (CV) was here on average small compared to large for culturable microorganisms. No outliers could be identified.
- Out of the 21 highest results only four are obtained by lower magnification than 5–11.9×. Out of the six lowest results only two are obtained by higher magnification than 4.9×. In general, this indicates the impact the magnification has for proper quantification of the strain of a slow-growing bacteria included here. It is most plausible that this also pertains many other typical slow-growing bacteria giving small colonies.

Outcome of the results and laboratory assessment

General information about reported results

The distributions of results for the respective analysis are shown in the histograms. A box plot (see below) gives a summarizing image of all the results of a laboratory, except false results. The number of false results and outliers are given below the plot for each laboratory. These values are highlighted with bold text on yellow background in annex A. The limit values for lowest and highest accepted results are given for each analysis in the summarizing lines at the end of annex A, together with the measurement uncertainty of the mean.

Base for assessment of the performance

The laboratories are not grouped or ranked in relation to their performances. The performance of an individual laboratory can be broadly assessed by the numbers of false results and outliers.

Generally, the laboratories that did not report their results in due time need to themselves compare their results with those of all other laboratories, by looking in tables, figures and annex A.

Mixed up results and other practical errors

Thirteen laboratories have more than one deviating result. When whole samples seem to have been mixed up, the corresponding sample numbers are crossed out in annex A. No laboratory seems to have mixed up neither samples nor individual results for a parameter this time. A few laboratories may have performed individual incorrect calculations from their colony readings to the final concentrations.

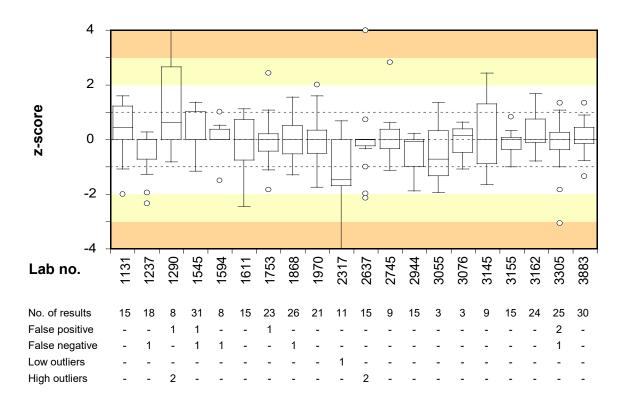
Z-scores, box plots and deviating results for each laboratory

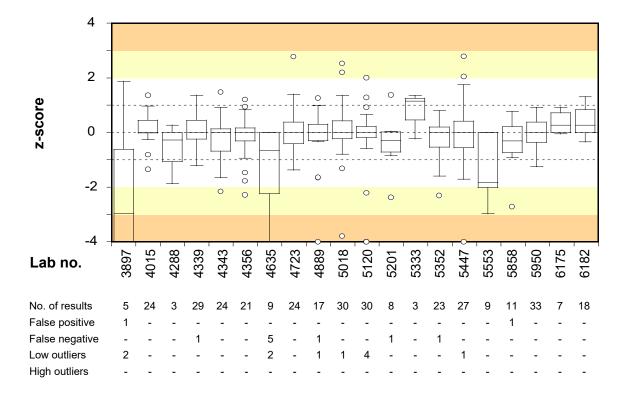
The square root transformed results of the laboratories are calculated to standard scores, z-scores, to be comparable between analyses. They are reported in annex B but are not further evaluated here. They are given explicitly to facilitate the follow-up process for laboratories using z-scores in control charts etc. See the explanation to annex A and the scheme protocol [1] for interpretation and calculation of z-scores.

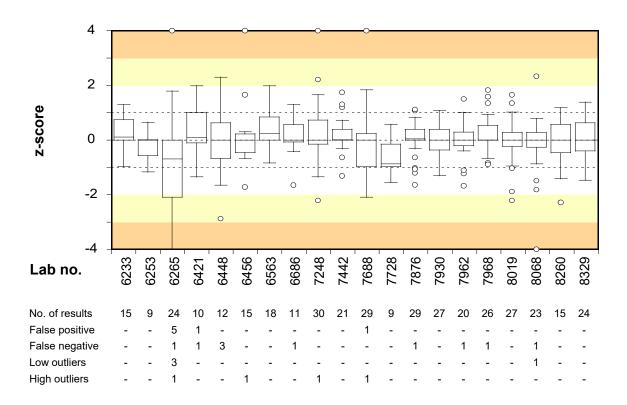
The z-scores are the base for the box plots. The range of the z-scores for each laboratory is shown by a rectangle (box) and lines and/or circles above and beneath the box. The smaller the range from lowest to highest value is in the plot and the more centred around zero the values are, the better the agreement is between the laboratory's results and the means from all laboratories.

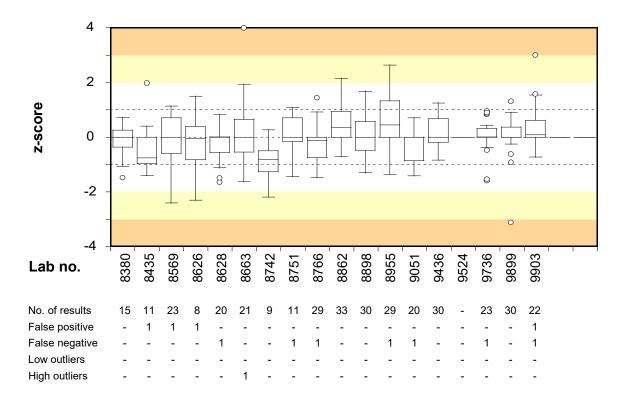
Box plots and numbers of deviating results for each participating laboratory

- *z*-scores are calculated from the formula z = (x mv) / s (see annex A).
- A correct result "zero" will get z = 0 when there is no target organism present.
- False results do not generate z-scores and are not included in 'No. of results'.
- The outliers are included in the plots after recalculation to standardised values with the same standard deviation (s) as the rest of the results for each parameter.
- *z*-scores > +4 and < -4 have in the plots been set to +4 and -4, respectively.
- The numbers of false positive and false negative results are given in the table under the plots together with the numbers of outliers.
- *The horizontal line in each box indicates the median for the laboratory.*
- The box includes 25 % of the results above and below the median. The lines protruding from the box and/or the circles embrace the remaining 50 % of the results, false results excluded.
- A circle is for technical reasons shown when a result is to a certain degree deviating* from the rest. This alone does not mean it is an outlier.
- The background is divided into coloured fields to simplify localization of the laboratory results.
- * < [smallest value of the box $1.5 \times$ (largest value of the box smallest value of the box)] or > [largest value of the box + $1.5 \times$ (largest value of the box smallest value of the box)]









Test material, quality controls and processing of data

Description of the test material

The round comprised three test items with different microorganism compositions. The test material was manufactured and freeze-dried in portions of 0.5 ml in small vials, according to the description by Peterz and Steneryd [2]. The simulated water samples were prepared by dissolving the content of the vials in 800 ml of sterile diluent. The composition and approximate concentrations in the samples obtained at the Swedish Food Agency are listed in table 2. The participating laboratories were assigned to perform the analyses according to the methods routinely used by them.

The test material is primarily suited to the EN ISO methods for analyses of drinking water referred to in the European Drinking water directive [4] and its updates [6]. Alternative methods and other standards can usually be used without any problem.

Sample ¹	Microorganisms	Strain co	llection no.	cfu/100 ml ²
		SLV (own)	Reference ³	•
A	Escherichia coli	084	From water	21
	Klebsiella oxytoca	553	From water	47
	Clostridium perfringens	442	CCUG 43593	3900
	Streptomyces sp.	548	DSMZ, identified	78
	Pseudomonas fluorescens	535	CCUG 45106	120*
В	Escherichia coli	295	From water	320
	Citrobacter freundii	424	From water	190
	Clostridium bifermentans	009	CCUG 43592	360
	Phialophora fastigiata	504	CBS, verified	120
	Rhodotorula minuta	506	CBS 970818	250
С	Klebsiella pneumoniae	186	CCUG 45102	1500
	Acremonium strictum	502	CBS, verified	26
	Hanseniaspora uvarum	555	CF SQE 77 $^{\#}$	31
	Sphingomonas sp.	547	CCUG 36955	160^{*}
	Staphylococcus warneri	189	CCUG 45143	<1 *

Table 2 Microorganisms present in the samples

1 The links between the samples and the randomised sample numbers are shown in annex A; the analyses were performed at the times given in note 1 of table 3

2 cfu = colony forming units; * indicates cfu per ml

3 Origin or typing collection no.; CCUG: Culture Collection University of Gothenburg, Sweden; CBS: Centraalbureau vor Schimmelcultures, Utrecht, Holland; DSMZ: Deutsche Sammlung von Mikroorganismen und Zellkulturen GmbH, Braunschweig, Germany; – or "From water" indicate a strain from our own "culture collection that has not yet been typed at another culture collection

Designation of an older culture collection

Quality control of the test material

It is essential to have a homogeneous sample mixture and a uniform volume in all vials in order to allow comparison of all freeze-dried samples derived from one sample mixture. The volume was checked by weighing 2 % of the number of vials produced from the sample mixtures. The largest differences between vials were 4, 6 and 7 mg in mixture A, B and C, respectively. The largest accepted difference is 15 mg (3 %).

Table 3 Contents (cfu) and measures of homogeneity (I2 and T, see reference 1) in
relevant sample volumes for the various parameters in the samples; shaded rows are
not used for performance assessing

Analysis parameter				Sar	nple	e ¹			
Method standard for analysis		Α			B			\mathbf{C}^{2}	
	cfu	I ₂	Т	cfu	I2	Т	cfu	I2	Т
Coliform bacteria (MF) <i>m-Endo Agar LES according to SS 028167</i>	68	1.2	1.3	51 ^a	1.1	1.3	15 ^b	0.7	1.5
Suspected thermotolerant colif. bact. (MF) <i>m-FC Agar, 44 °C according to SS 028167</i>	18	0.4	1.3	24 ^a	1.4	1.6	14 ^b	2.5	2.6
Escherichia coli (MF) m-Endo Agar LES according to SS 028167	21	0.8	1.5	24 ^a	1.4	1.6	_	_	_
Presumptive Clostridium perfringens (MF) TSC Agar according to SS-EN ISO 14189:2016	38	1.7	1.5	35 ^{a,d}	5.5	2.4	—	_	_
Moulds (MF) Rose Bengal Agar with both chloramphenicol and chlortetracycline according to SS 028192	_	_	_	12	1.8	2.1	26	2.5	1.9
Yeasts (MF) Rose Bengal Agar with both chloramphenicol and chlortetracycline according to SS 028192	_	_	_	25	0.9	1.5	31	2.8	1.3
Actinomycetes (MF) Actinomycete Isolation Agar with cycloheximide according to SS 028212	39°		1.4	—	_	_	—	_	_
Culturable microorg., 3d 22 °C (pour plate) Yeast extract Agar according to SS-EN ISO 6222:1999	127	9.1	1.6	5	1.0	2.6	15	1.4	1.8
Slow-growing bacteria, 7d 22 °C (pour plate) Yeast extract Agar according to SS-EN ISO 6222:1999	_	_	_	_	_	_	173	1.5	1.2

1 10 vials analysed in duplicate, normally100 ml for MF and 1 ml for pour plate, analysed 21, 19 and 17 weeks ahead of the testing round for the sample A, B and C, respectively

a Determined for the volume 10 ml

b Determined for the volume 1 ml

c Determined for the volume 50 ml

d The sample mixture was not homogeneous for the strain of *C. bifermentans*, which is acceptable for a strain that is false positive for *C. perfringens*

- No target organism and thus no analysis

Table 3 presents the results from the organizer in the form of concentration means (cfu) and the measures (I₂ and T; see reference 1) used to assess homogeneity from duplicate analyses of 10 vials from each mixture the first time a mixture is used or duplicate analyses from 5 vials in a stability check when a mixture is used a second time. The results relate to the volume that was used for counting the colonies. The criterion used for a sample mixture to be considered homogenous is that I₂ and T *not simultaneously* are higher than 2. According to that criterion, all mixtures were homogeneous regarding the parameters that could be analysed, with the exception of the false positive strain *C. bifermentans* and also of the suspected thermotolerant coliform bacteria in sample C that are not included in performance assessment.

Processing of numerical results

Most histograms have "tails" in either or both directions, due to values that do not belong to a normal distribution. For drinking water, ten log transformation of results is seldom routine. With low concentrations as there, calculations are here instead performed after square root transformations of the results, which give the best normal distributions by decreasing the significance of the high deviating results. Very deviating values will still be present in most analyses and are identified as outliers (black bars). False negative results are presented with white bars in the histograms.

Outliers are identified by use of Grubbs' test according to a modification by Kelly [3]. A level of 1 % is set as the risk to incorrectly assess a result as being an outlier. Although the method is objective, there is a prerequisite that the results are normally distributed in order to obtain correct outliers at the 1 % level. A zero result that is a low outlier is considered a false negative result. In special situations, e.g. when many zero results are reported and in some borderline cases, subjective adjustments will be are made based on the knowledge of the sample mixture's content in order to set the right limits. False results and outliers are not included in the calculations of mean values and measures of distribution.

The coefficient of variation (CV) for square root transformed results is given as a measure of dispersion. When the dispersion is <10 % it is regarded as very small, 10-20 % as small, 20-30 % as medium, 30-40 % as large and >40 % as very large.

The calculation of uncertainty of measurement of the assigned value is described in the scheme protocol [1]. The assigned value for an analysis is here calculated from the square root transformed results and is the square root of "Mean" in Annex A. It is there denoted as mv. Hence, also the measurement uncertainty will be expressed as a square root value. The standard uncertainty of measurement (u) correspond to the standard deviation of the assigned value (s) divided by the number of results squared-root transformed, i.e.: $u = s/\sqrt{n_{mv}}$ where n_{mv} is the number of results in annex A, except the deviating ones. Here is the relative uncertainty (u_{rel}) used and expressed as per cent after division by the mean value mv and multiplication by 100.

More about result processing and recommendations on follow-up work are given in the scheme protocol [1]. A PDF of that document is available on the website <u>https://www2.slv.se/absint</u>.

References

- 1. Anonymous 2018. Scheme protocol, Microbiology, Drinking water & Food, 5th ed. Swedish Food Agency (formerly National Food Agency), Sweden.
- 2. Peterz, M., Steneryd, A.-C. 1993. Freeze-dried mixed cultures as reference samples in quantitative and qualitative microbiological examinations of food. J. Appl. Bacteriol. 74:143-148.
- 3. Kelly, K. 1990. Outlier detection in collaborative studies. J. Assoc. Off. Chem. 73:58-64.
- 4. Anonymous 1998. Council Directive 98/83/EC of 3 November 1998 on the quality of water intended for human consumption. Official Journal of the European Communities. 5.12.98, L 330/32-54 (national translations available).
- 5. Standard Methods for the Examination of Water and Wastewater, <u>http://www.standardmethods.org/</u>
- 6. Anonymous 2015. Commission Directive (EU) 2015/1787 of 6 October 2015 amending Annexes II and III to Council Directive 98/83/EC on the quality of water intended for human consumption. Official Journal of the European Union. 7.10.2015, L 260/6-17 (national translations available).

Annex A Results of the participants, $cfu/100 \ ml$ (see also the note[#]). Susp. = suspected on membrane filter before confirmation. Results given as <, <1, <2, <10 and <100 are treated as zero. The fields with other results given as < 'value' and results given as > 'value' are yellow, and those results are not included in calculations or evaluations, as are also not results in shaded columns. A hyphen indicate that no result has been reported. Figures written in bold in yellow fields indicate outliers, false positive and false negative results. Underlined zero values indicate results characterized as 'False negative ?'. Crossed out sample numbers in a row indicate that the samples probably are mixed up.

Lab no.	Sample	Suspected coliform bacteria (MF)			Coliform	bacter	ia (MF)	Susp. thermotolerant coliform bact. (MF)			Ε.	coli (M	F)		orm bac		E. coli ("rapid" MPN		
	ABC	A	B B	F) C	Α	в	с	A	m bact. B	.(MF) C	A	в	с	("rapid" MPN) A B C			Α	в	с
1131	1 2 3	-	-	-	-	-	-	-	-	-	-	-	-	64	517	1300	25	<1	<1
1237	3 2 1	-	-	-	41	180	700	-	-	-	13	<1	<1	62	>200	>200	16	<1	<1
1290 1545	1 3 2 2 3 1	- 56	- 380	- 1340	6900 56	310 380	1300 1340	24	- 285	- 1090	2100 24	< 285	< 0	- 72	436	- 1140	22	110	-
1594	3 2 1	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
1611 1753	2 1 3 3 2 1	67	480	1030	67	480	1030	8	204	1000	17	290	0	67 49	548 291	1416 1733	12 21	0 0	0
1868	1 2 3	63	- 455	- 1350	63	455	1350	-	-	-	28	0	0	51	517	1553	15	0	0
1970	2 1 3	50	380	1300	50	380	1300	50	380	1300	14	0	0	-	-	-	-	-	-
2317 2637	123 312	-	-	-	35	163	750	-	-	-	8	0	0	1000	- 450	- 1200	222	- <1	- <1
2745	2 3 1	57	360	1300	57	360	1300	57	360	1300	57	160	0	-	-			-	-
2944 3055	321 213	-	-	-	-	-	-	-	-	-	-	-	-	53	288	1298	18	0	0
3035	3 2 1	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
3145	1 2 3	-	-	-	-	-	-	-	-	-	-	-	-	51	488	1733	26	0	0
3155 3162	2 3 1 3 1 2	-	-	-	53	405	1060	-	-	-	14	-	<1	50 55	- 428	1212 1200	15 20	-	<1 0
3305	3 2 1	52	560	1200	52	560	1200	-	-	-	14	340	<1	50	560	950	5	90	<1
3883	123	64	400	1027	64	400	1027	-	-	-	18	280	< 1	59	417	1405	15	< 1	< 1
3897 4015	3 1 2 3 1 2	45	48	48	45	48	48		-	-	42	45	45	- 63	462	- 1357	- 16	0	0
4288	1 2 3	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
4339 4343	1 3 2 3 2 1	58	380	1020	58	380	1020	17	300	1210	35	0	0	58 75	365 411	1120 980	18 19	0 0	0 0
4356	3 1 2	63	540	1000	63	540	1000	17	320	1200	21	324	0	39	365	1203	17	0	0
4635 4723	3 1 2 1 3 2	>1	<1	<1	>1	<1	<1	-	-	-	<1	<1	<1	- 65	- 411	- 1063	- 20	-0	-0
4723 4889	2 1 3	-	-	-	53	360	110		-	-	20	0	0	64	580	980	20	0	0
5018	2 3 1	65	440	1170	65	440	1170	-	-	-	52	0	0	73	687	1120	14	0	0
5120 5201	3 1 2 1 2 3	55	420	1300	55 27	420	1300 1150	10	390	1300	44 12	0 426	0	63	460	1200	20	0	0
5333	1 3 2	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
5352 5447	1 2 3 2 3 1	-	-	-	57 76	330 440	950 1200	-	-	-	19 30	330 310	0 0	-	-	-	-	-	-
5447 5553	1 2 3	-	-	-	22	176	720	-	-	-	30 8	96	0	-	-	-	-	-	-
5858	2 1 3	-	-	-	45	300	960	-	-	-	13	<1	<1	-	-	-	-	-	-
5950 6175	1 3 2 3 2 1	62	345	1291	62	345	1291	10	188	1209	16	218	0	63 59	488 >200	1300 >200	15 22	0 0	0
6182	2 3 1	53	440	1193	53	440	1193	-	-	-	19	<1	<1	68	548	1306	17	<1	<1
6233 6253	2 1 3 3 2 1	47	450	1309	47	450	1309	-	-	-	11	240	0	73 54	461 450	1414 1260	17 13	0 0	0
6265	3 1 2	43	430	1050	43	430	1050	-	-	-	14	250	0	63	350	1600	0	0	0
6421	321	-	-	-	51	260	1700	8	350	1500	0	350	0	-	-	-	-	-	-
6448 6456	3 1 2 1 3 2	-	-	-	50 49	280 7171	1800 1600	-	-	-	0 14	120 230	0 <1	53	480	- 1300	16	<1	<1
6563	2 3 1	60	509	1191	60	509	1191	60	509	1191	12	509	<1	72	613	1413	14	<1	<1
6686 7248	1 2 3 2 3 1	- 53	- 430	- 1600	- 53	- 430	- 1600	- 10	- 430	- 1000	- 30	- 344	- <1	62.4 50	306 370	1184 1112	20.7 30	<1 <1	<1 <1
7442	1 2 3	76	448	1219	76	448	1219	-	-	-	27	0	0	60	482	1260	17	0	0
7688 7728	2 3 1 1 3 2	36	610	1200	36	610	1200	-	-	-	11	488	0	43	272	1300	9	0	0
7876	1 3 2 3 1 2	62	- 460	- 1150	41 62	270 460	1100 1150	17	200	- 960	11 20	180 340	0 <1	68	- 561	- 1081	19	- <1	- <1
7930	3 1 2	59	510	1200	59	510	1200	-	-	-	23	230	<1	50	410	1300	11	<1	<1
7962 7968	321 213	53 59	580 425	1170 1200	53 59	580 425	1170 1200	10	330	980	20 16	0 250	0 0	44 62	548 450	1046 1400	15 26	0 0	0
8019	321	52	427	910	52	427	910	-	-	-	16	0	0	70	624	1316	19	0	0
8068 8260	2 3 1 2 1 3	- 39	- 472	- 1460	49 39	470 472	1200 1460	10	200	780	49 18	310 338	0 0	59	370	180	9	0	0
8260 8329	2 1 3 2 3 1	-	4/2	-	- 39	4/2	- 1400	-	-	-	-	- 550	-	74	- 548	- 1300	- 21	0	0
8380	1 3 2	61	480	1200	61	480	1200	-	-	-	20	215	0	57	370	1075	18	0	0
8435 8569	3 1 2 1 2 3	- 37	- 374	- 620	42 37	350 374	810 620	17	170	1090	11 14	190 0	0 0	- 38	- 411	- 1414	- 14	-0	-0
8626	3 2 1	50	580	1160	50	580	1160	25	290	1160	25	290	1160	-	-	-	-	-	-
8628 8663	2 3 1 2 1 3	- 79	- 300	- 900	45 79	340 300	870 900	6 13	270 390	920 1200	14 32	0 210	0 0	- 73	490	2400	- 18	-0	-0
8742	1 3 2	-	-	- 300	29	190	900 940	-	- 390	- 1200	9	200	<1	-	-		-	-	-
8751	231	-	-	-	-	-	-	-	-	-	-	-1	-	50	560	1013	16	<1	<1
8766 8862	1 3 2 2 1 3	50 56	373 473	1118 1400	50 56	373 473	1118 1400	12	320	970 -	13 13	<1 0	<1 0	54 78	437 596	1120 1540	12 17	<1 0	<1 0
8898	1 3 2	64	459	946	64	459	946	-	-	-	15	279	0	54	342	1047	11	0	0
8955 9051	1 2 3 1 3 2	-	-	-	60 58	470 255	1800 836	-	-	-	30 16	280 0	0 0	79 61	340 325	1300 1175	33 14	0 0	0
9436	3 1 2	64	- 518	945	64	518	945	14	455	855	12	389	<1	57	433	1195	16	<1	<1
9524	2 1 3	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
9736 9899	1 3 2 2 3 1	45	- 462	- 1138	45	- 462	- 1138	-	-	-	- 17	0	-	62 59	485 507	1304 1420	17 16	0 0	0
9903	1 2 3	-	-	-	73	467	1158	52	342	983	19	220	0	-	-	-	-	-	-
Mean CV (%)					52 12	395 14	1138 11				19 25	263 20	0	59 8	450 11	1257 7	17 15	0	0
UV (70)					12	14	11				20	20	-	Ö	11	(10	-	-

False positive and false negative values are excluded, as well as other outliers, in the summarizing calculated results at the end of the table. The mean value (Mean) is the square of the mean value for the square root transformed results (mv). The coefficient of variation (CV) is the standard deviation (s) in percentage of the mean value for the square root transformed results. As means to calculate the z-values of your own, the appropriate values of mv and s are given at the end of the table. The x-values of a laboratory are obtained as the square roots of each reported result, respectively. z = (x - mv) / s. $u_{rel,mv}$ is the relative standard uncertainty of mv in per cent. For calculation see the scheme protocol [1]; also briefly described in the text.

Presumptive C. Clostridium perfringens (MF) perfringens (MF)				Мо	ulds (Mi	F)	Yeasts (MF)			Actinomycetes (MF)				plate co		Slow-gro	Lab no.				
A	B B	C C	A	B	(MF) C	Α	в	с	Α	в	с	Α	в	с	22 °(C,3 day B	s″ C	22 °	C,7 day B	s″ C	
5800	64	<1	<u> </u>	-		<u>.</u>	-	-	-		-	<u>.</u>	-	-	82	4	103	85	7	310	1131
-	-	-	<1	<1	<1	-	-	-	-	-	-	-	-	-	50	5	17	65	8	190	1237
-	-	-	6100	1300	<	- 0	400	-	-	-	-	-	-	-	-	-	-	-	- 9	-	1290
5900	1900	0	5900	0	0	0	130 130	0	0	310 270	41 33	75 58	0 0	0 0	25	9	95	31	9	215	1545 1594
-	-	-	-	-	-	-	-	-	-	- 210	-	-	-	-	5	4	12	-	-	-	1611
5500	560	0	-	-	-	1	100	33	0	230	32	88	0	0	39	7	32	34	7	199	1753
3400	1118	0	-	-	-	0	73	0	0	182	34	70	0	0	71	5	18	-	-	-	1868
3000	1200	0	3000 410	0	0 0	0	50	21	0	330	50	-	-	-	52 15	4 4	79 79	-	-	-	1970 2317
2035	900	<1	2035	<1	<1	-	-	-	-	-	-	_	-	-	66	6	23	_	-	-	2637
-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	48	5	44	-	-	-	2745
-	-	-	4200	0	0	-	-	-	-	-	-	-	-	-	19	5	57	20	8	93	2944
-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	10 63	10 4	30 57	-	-	-	3055 3076
_	-	-	-	-	-	-	-	-	-	-	-	-	-	-	84	3	16	-		-	3145
-	-	-	4700	<1	<1	-	-	-	-	-	-	-	-	-	28	7	85	-	-	-	3155
5500	450	0		-	-	0	90	32	0	300	38	97	0	0	78	11	43	78	11	225	3162
3600	850	- < 1	3600	<1	-	9	130	28 29	<1 < 1	280	29	<1 67	<1	<1	65	7 10	40	- 68	- 12	-	3305
4600	800		-	-	-	< 1	84	29	-	180	35	67	<1 -	< 1	68	-	70	- 00	12	241	3883 3897
3772	1365	0	-	-	-	0	59	26	0	203	31	82	0	0	73	6	111	80	10	241	4015
-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	39	7	7	-	-	-	4288
5400	1220	0	5400	0	0	0	80	10	0	230	35	-	-	-	40	9	72	30	7	217	4339
4400 5300	38 1000	0 0	- 5300	-	-0	0	82	18	0	200	20	68	0	0	66 12	5 6	17 13	84	7	249	4343 4356
-		-	-	-	-	0	0	1	0	5	0	-	-	-	40	4	16	-	-	-	4635
3636	3636	0	-	-	-	0	82	21	0	182	25	82	0	0	87	7	15	90	9	216	4723
0	1000	0	-	-	-	-	-	-	-	-	-	-	-	-	39	3	92	-	-	-	4889
3800 49	1300 1600	0 0	3800 49	0 0	0 0	0	60 79	32 5	0 0	240 250	25 4	34 24	0 0	0 0	32 41	10 8	68 107	-	-	-	5018 5120
-	-	-	-	-	-	-	-	-	-	200	-		-	-	32	5	52	-		-	5201
-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	40	10	100	-	-	-	5333
5100	1020	0	-	-	-	0	120	0	0	220	33	66	0	0	50	2	11	41	11	162	5352
3900	1800	0	3900 2260	0 0	0 0	0	6	60	0	164	22	70	0	0	71	5	11	30	20	120	5447 5553
-	-	-	4550	830	<1	-	-	-	-	-		-	-	-	3	- 8	82	-	-	-	5858
3000	318	0	3000	0	0	0	109	30	0	273	25	61	0	0	71	7	82	71	11	205	5950
-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	60	7	88	-	-	-	6175
-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	84	7	94	89	8	239	6182
-	-	-	-	-	-	-	-	-	-	-		-	-	-	70 54	7 8	89 19	-	-	-	6233 6253
26	120	130	26	120	130	19	9	8	15	120	890	-	-	-	42	3	68	30	3	45	6265
-	-	-	4500	2400	0	-	-	-	-	-	-	-	-	-	32	9	100	-	-	-	6421
-	-	-	-	-	-	0	90	0	0	320	0	-	-	-	2 51	7 5	92 9	-	-	-	6448 6456
_	-	-	-	-	-	-	-	-	-	-	-	-	-	-	56	7	77	61	7	200	6563
5800	<1	<1	-	-	-	-	-	-	-	-	-	-	-	-	76	6	68	-	-	-	6686
3400	1700	<1	-	-	-	<1	40	35	<1	260	21	75	<1	<1	44	10	83	32	41	230	7248
3700 4600	524 940	0 0	- 4600	-0	-0	9	- 70	- 27	0	- 190	- 22	94 400	0 0	0 0	84 68	6 3	16 100	-	-	-	7442 7688
	-	-	-300	-	-	-	-	-	-		-		-	-	15	5	74	-	-		7728
3700	1150	<1	-	-	-	<1	100	<1	<1	260	41	80	<1	<1	31	3	78	30	6	200	7876
5500	530	<1	5500	<1	<1	<1	125	21	<1	265	27	-	-	-	38	5	85	-	-	-	7930
- 3300	- 1200	-0	- 3300	-0	-0	0 0	65 160	0	0 0	190 280	35 28	-	-	-	47 26	9 10	85 90	-	-	-	7962 7968
4000	82	0	4000	0	0	0	70	5	0	240	35	-	-	-	43	10	22	-	-	-	8019
-	-	-	4700	0	0	0	100	0	0	230	20	-	-	-	38	8	83	-	-	-	8068
4700	1100	0	-	-	-	-	-	-	-	-	-	-	-	-	63	5	14	64	8	68	8260
4400	210	0	-	-	-	0	80	30	0	180	37	70	0	0	86 16	5 7	43 80	81	5	230	8329 8380
1 1	-	-	- 4800	0	1100		-	-	-	-	-]	-	-	25	12	80 20]	-	1	8380
5200	484	0	5200	484	0	-	-	-	-	-	-	-	-	-	57	8	85	75	13	247	8569
-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	40	2	14	-	-	-	8626
-	-	-	4300 2500	0	0	0	110	0	0	270	36	-	-	-	39	3	85 15	25	8	190	8628
2500	1200	0	∠300 -	0	0	-	-	-	-			-	-	-	67 34	8 7	15 17	-	-	1	8663 8742
-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	56	9	95	54	<10	205	8751
2800	1600	<1	-	-	-	<1	145	<1	<1	273	29	65	<1	<1	21	6	71	27	9	104	8766
6200	1200	0	6200	0	0	0	173	31	0	282	27	90 64	0	0	65 72	9	78	67	8	224	8862
4545	1135	0	- 6200	-	-	0	91 110	30 0	0 0	300 280	23 35	64 100	0 0	0 0	72 85	11 6	76 84	80 120	13 15	229 110	8898 8955
4300	245	0	0200	0	0	-	-	-	-		-	-	-	-	31	4	80		-	-	9051
3300	800	<1	-	-	-	<1	136	45	<1	273	28	86	<1	<1	64	8	66	95	9	249	9436
-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-		-	-	-	-	9524
4091 1167	1522 1148	0 0	-	-	-	0 0	124 93	40 26	0 0	265 254	39 24	57 96	0 0	0 0	46 71	7 7	12 52	48 77	0 12	159 200	9736 9899
3240	1290	Ő	-	-	-	Ő	99	0	209	294	61	100	Ő	0	65	7	50	68	9	216	9903
4072	890	0	4281	0	0	0	96	26	0	243	31	77	0	0	45	6	52	57	9	189	Mean
15	37	-	15	-	-	-	16	25	-	10	13	9	-	-	27	19	34	23	17	18	CV (%)

Lab no.	Sample	Suspec bac	cted co teria (N		Coliform bacteria (MF)			Susp. th colifor			Ε.	coli (M	F)		orm bac apid" Mi		E. coli ("rapid" MPN)			
	ABC	Α	В	C	Α	В	C	Α	В	C	Α	В	C	Α	В	С	Α	В	С	
n		35	36	36	56	57	57	21	21	21	57	56	57	52	49	50	52	51	52	
Min		36	0	0	22	0	0	6	170	780	0	0	0	38	272	180	0	0	0	
Max		79	610	1600	6900	7171	1800	60	509	1500	2100	509	1160	1000	687	2400	222	110	0	
Median		56	444	1181	53	427	1165	14	320	1090	17	280	0	60	450	1279	17	0	0	
Mean					52	395	1138				19	263	0	59	450	1257	17	0	0	
CV (%)					12	14	11				25	20	-	8	11	7	15	-	-	
False po					0	0	0				0	0	2	0	0	0	0	2	0	
False ne					0	2	1				3	21	0	0	0	0	1	0	0	
Outliers					0 1	1	2 0				0	0	0	0	0	1	0	0	0	
Outliers	, nign				1	1	0				1	0	0	1	0	1	1	0	0	
Low limi		36	0	0	22	163	620	6	170	780	8	45	0	38	272	950	5	0	0	
High lim	it OK	79	610	1600	79	610	1800	60	509	1500	57	509	0	79	687	1733	33	0	0	
mv (√Mean)					19.887						16.219	0.000	-	21.219		4.118	0.000	0.000	
s (CV*mv/	100)				0.854	2.790	3.769				1.113	3.203	0.000	0.642	2.267	2.528	0.616	0.000	0.000	
u _{rel,mv} (% (100*s/ 1					1.6	1.9	1.5				3.5	3.3		1.2	1.5	1.0	2.1			
x (√Resul	t)																			
z ([x-mv]/s)																			

cfu/ml
* The 21 zero-resultats is not reckoned as false negative results but as accepted zero results when the detection is based on β-glucuronidase activity; see the text

	Presumptive C. Clostridium perfringens (MF) perfringens (MF)					Мо	ulds (M	F)	Y	easts (M	F)	Actino	mycetes	s (MF)		plate c C, 3 day		Slow-gr 22 °	Lab no.		
Α	В	С	Α	В	C	Α	В	С	Α	В	С	Α	В	С	Α	В	С	Α	В	С	
															=0						r
4	I 41	40 0	31 0	30 0	30 0	38 0	38 0	38 0	38 0	38 5	38 0	28 0	28 0	28 0	73 2	73 2	73 7	32 20	32 0	32 45	n Min
6200		130	6200	2400	1100	19	173	60	209	330	890	400	0	0	87	12	111	120	41	45 310	Max
4046		0	4525	0	0	0	93	29	0	260	31.5	75	0	0	48	7	68	66	9	210	Median
4072		0	4281	0	0	0	96	26	0		31	77	0	0	45	6	52	57	9	189	Mean
1	5 37	-	15	-	-	-	16	25	-	10	13	9	-	-	27	19	34	23	17	18	CV (%)
() 0 I 1	1 0	0 2	5 0	2 0	4 0	0 1	0 14	2 0	0 0	0 2	0	0 0	0 0	0 0	0 0	0 0	0	0 2	0 0	False pos. False neg.
	2 0	0	2	0	0	0		14	0	1	2	2	0	0	0	0	0	0	2	0	Outliers <
	0	Ő	0	0	0	0	2 0	0	0	0	1	1	Ő	0	0	Ő	0	Ő	1	0	Outliers >
116		0	2035	0	0	0	40	5	0	120	20	57	0	0	2	2	7	20	3	45	Low limit
6200	3636	0	6200	0	0	0	173	60	0	330	61	100	0	0	87	12	111	120	20	310	High limit
63.812	2 29.839	0.000	65.426	0.000	0.000	0.000	9.792	5.092	0.000	15.575	5.578	8.773	0.000	0.000	6.745	2.515	7.200	7.577	3.007	13.748	mv
9.516	6 10.983	0.000	9.490	0.000	0.000	0.000	1.563	1.291	0.000	1.620	0.741	0.775	0.000	0.000	1.852	0.477	2.439	1.732	0.525	2.411	s
2.4	5.8		2.8				2.7	5.3		1.7	2.3	1.8			3.2	2.2	4.0	4.0	3.2	3.1	u _{rel.mv} (%)
																					x
																					z

l ah no	Sample	Suspected coliform	Coliform bacte	ria	Sucn 4	hermoto	lorant	F	coli (M	E)	Colif	orm bac	toria	E. coli		
Lab IIO.	-	bacteria (MF)	(MF)			m bact.			-			apid" M				
1131	ABC 123	A B C	A B	С	Α	В	С	Α	В*	С	A 0.451	B	C 0.235	A 1.431	B 0.000	C
1237	3 2 1		-0.963 -2.319 -1	1.931				-0.713	0.000	0.000	0.255	0.070	0.233	-0.192	0.000	0.000
1290 1545	132 231		4.000 -0.817 0					4.000 0.449	0.000 0.207	0.000	1 207	-0.149	0.674	0.928		0.000
1545	2 3 1 3 2 1		0.302 -0.141 0	J.762				0.449	0.207	0.000	1.207	-0.149	-0.071	0.928		0.000
1611	2 1 3		1.123 0.725 -0	0.436				-0.248	0.253	0.000	0.740	0.966	0.858	-1.062	0.000	0.000
1753 1868	321 123		0.833 0.518 0	0.798				0.802	0.000	0.000		-1.835 0.670	2.440 1.562	0.753 -0.399	0.000 0.000	0.000 0.000
1970	2 1 3		-0.181 -0.141 0	0.615				-0.591	0.000	0.000	0.000	0.070		0.000	0.000	0.000
2317 2637	123 312		-1.533 -2.552 -1	1.685				-1.411	0.000	0.000	4 000	-0.002	-0 324	4 000	0.000	0.000
2745	231		0.379 -0.327 0	0.615				2.831	-1.115	0.000	4.000	-0.002	-0.524	4.000	0.000	0.000
2944	321										-0.670	-1.874	0.224	0.202	0.000	0.000
3055 3076	2 1 3 3 2 1															
3145	1 2 3										-0.886	0.385		1.591	0.000	0.000
3155 3162	231 312		0.064 0.085 -0).313				-0.591		0.000	-0.995 -0.458	-0.234	-0.256	-0.399 0.574	0.000	0.000 0.000
3305	321		-0.017 1.354 0						0.693		-0.995	1.079	-1.835	-3.055		0.000
3883 3897	123 312		0.906 0.041 -0 -0.606 -4.000 -4						0.161	0.000	-0.045	-0.352	0.800	-0.399	0.000	0.000
4015	3 1 2		-0.000 -4.000 -4					1.071	-2.570		0.354	0.122	0.545	-0.192	0.000	0.000
4288	123		0.457 0.444 0	477				4 000	0.000	0.000	0 4 4 7	0.000	0 700	0.000	0.000	0 000
4339 4343	1 3 2 3 2 1		0.457 -0.141 -0	J.4//				1.363	0.000	0.000		-0.933 -0.417		0.202	0.000 0.000	0.000 0.000
4356	3 1 2		0.833 1.201 -0	0.561				0.165	0.556			-0.933		0.008		0.000
4635 4723	312 132								0.000	0.000	0.548	-0.417	-1.130	0.574	0.000	0.000
4889	2 1 3		0.064 -0.327 -4					0.066	0.000		0.451	1.264	-1.644	0.574	0.000	0.000
5018 5120	231 312		0.979 0.390 0 0.223 0.218 0					2.527 2.008	0.000 0.000	0.000	1.299 0.354	2.202 0.101		-0.612 0.574	0.000 0.000	0.000
5201	1 2 3			0.047				-0.840	1.380		0.004	0.101	0.024	0.074	0.000	0.000
5333	1 3 2		0.070 0.047 0	. 770				0.000	0.000	0.000						
5352 5447	123 231		0.379 -0.617 -0 1.747 0.390 0					-0.036 0.969	0.608 0.433	0.000						
5553	1 2 3		-2.968 -2.373 -1	1.832				-1.411	-2.005	0.000						
5858 5950	2 1 3 1 3 2		-0.606 -0.920 -0 0.759 -0.471 0						0.000 -0.454		0.354	0.385	0.235	-0.399	0.000	0.000
6175	321										-0.045			0.928	0.000	0.000
6182 6233	231 213		0.064 0.390 0).213).649					0.000		0.835 1.299	0.966 0.111		0.008 0.008	0.000 0.000	0.000 0.000
6253	3 2 1		-0.433 0.473 0	J.049				-0.972	-0.221	0.000		-0.002	0.040	-0.833	0.000	0.000
6265	3 1 2		-0.782 0.305 -0					-0.591	-0.127		0.354	-1.108	1.796		0.000	0.000
6421 6448	321 312			1.989 2.306					0.777	0.000						
6456	1 3 2		-0.264 4.000 1	1.662				-0.591	-0.329	0.000		0.305		-0.192		
6563 6686	231 123		0.609 0.959 0	0.206				-0.840	1.980	0.000	1.207 0.295	1.562 -1.644		-0.612 0.700	0.000 0.000	0.000 0.000
7248	231			1.662				0.969	0.727		-0.995	-0.875	-0.836	2.205	0.000	0.000
7442 7688	123 231		1.747 0.458 0 -1.435 1.725 0	0.313				0.716	0.000 1.833		0.056	0.325	0.014	0.008	0.000 0.000	0.000 0.000
7728	1 3 2		-0.963 -1.239 -0						-0.875		-1.735	-2.005	0.200	-1.015	0.000	0.000
7876	3 1 2			0.047						0.000	0.835	1.088		0.390	0.000	
7930 7962	312 321).240).125						0.000 0.000	-0.995	-0.428 0.966	0.235	-1.302 -0.399	0.000	0.000 0.000
7968	2 1 3			0.240					-0.127			-0.002		1.591	0.000	0.000
8019 8068	321 231		-0.017 0.279 -0 -0.264 0.643 0					-0.358 2.337	0.000 0.433	0.000	1.022	1.659 -0.875	0.323	0.390	0.000 0.000	0.000 0.000
8260	2 1 3			1.187						0.000						
8329 8380	231 132		0.684 0.725 0	1 240				0.066	-0 486	0 000	1.389 -0.250	0.966			0.000 0.000	0.000
8435	3 1 2		-0.872 -0.423 -1	1.400				-0.972	-0.760	0.000						
8569 8626	123 321		-1.338 -0.196 -2 -0.181 1.504 0					-0.591 0.540	0.000	0.000	-2.407	-0.417	0.848	-0.612	0.000	0.000
8628	231		-0.181 1.504 0						0.253	0.000						
8663	2 1 3		1.946 -0.920 -0	0.991				1.130	-0.540	0.000	1.299	0.405	4.000	0.202	0.000	0.000
8742 8751	1 3 2 2 3 1		-2.154 -2.188 -0	010.				-1.257	-0.649	0.000	-0.995	1.079	-1.437	-0.192	0.000	0.000
8766	1 3 2		-0.181 -0.206 -0						0.000		-0.563	-0.139	-0.789	-1.062	0.000	0.000
8862 8898	2 1 3 1 3 2		0.302 0.667 0 0.906 0.551 -0					-0.713 -0.473	0.000 0.151		1.747 -0.563	1.409 -1.202		0.008	0.000	0.000 0.000
8955	123		0.609 0.643 2	2.306				0.969	0.161	0.000	1.834	-1.226	0.235	2.639	0.000	0.000
9051 9436	1 3 2 3 1 2		0.457 -1.405 -1 0.906 1.030 -0						0.000 1.094			-1.408 -0.181		-0.612 -0.192		0.000 0.000
9524	213		0.000 1.000 -0					-0.040	1.094	0.000	-0.200	-0.101	-0.000			
9736	132		0.606 0.570 0	1 000				0.240	0.000	0.000		0.355 0.573			0.000	
9899 9903	2 3 1 1 2 3		-0.606 0.576 0 1.543 0.618 0	0.000 0.078				-0.248	0.000 -0.433	0.000	-0.045	0.073	0.079	-0.192	0.000	0.000
n		0 0 0	56 55	56	0	0	0	54	56	55	52	49	50	51	49	52
Min		5 6 0	-2.968 -4.000 -4	4.000	Ŭ	U	0	-1.411	-2.970		-2.407	-2.085	-4.000		0.000	0.000
Max			4.000 4.000 2	2.306				4.000	1.980	0.000	4.000	2.202	4.000	4.000	0.000	0.000
Median			0.064 0.279 0						0.000			-0.002			0.000	0.000
Mean SD			0.071 0.000 -0					0.074 1.130	0.000 0.786			0.000 1.000		0.078	0.000 0.000	0.000 0.000
																0.000
z<-3 -3≤z<-2			0 1 3 4	2 1				0	0 2	0	0	0	1 0	1	0	0
-3≤z<-2 2 <z≤3< th=""><th></th><th></th><th>0 0</th><th>2</th><th></th><th></th><th></th><th>4</th><th>2</th><th>0</th><th>2</th><th>1</th><th>2</th><th>2</th><th>0</th><th>0</th></z≤3<>			0 0	2				4	2	0	2	1	2	2	0	0
z>3			1 1	0				1	0	0	1	0	1	1	0	0

Annex B Z-scores calculated from the laboratory results. Susp. = Suspected on the membrane filters before confirmation. z = (x - mv) / s. Z-scores are calculated also for outliers (excluding false negative results) in the same way as ordinary z-scores. From false

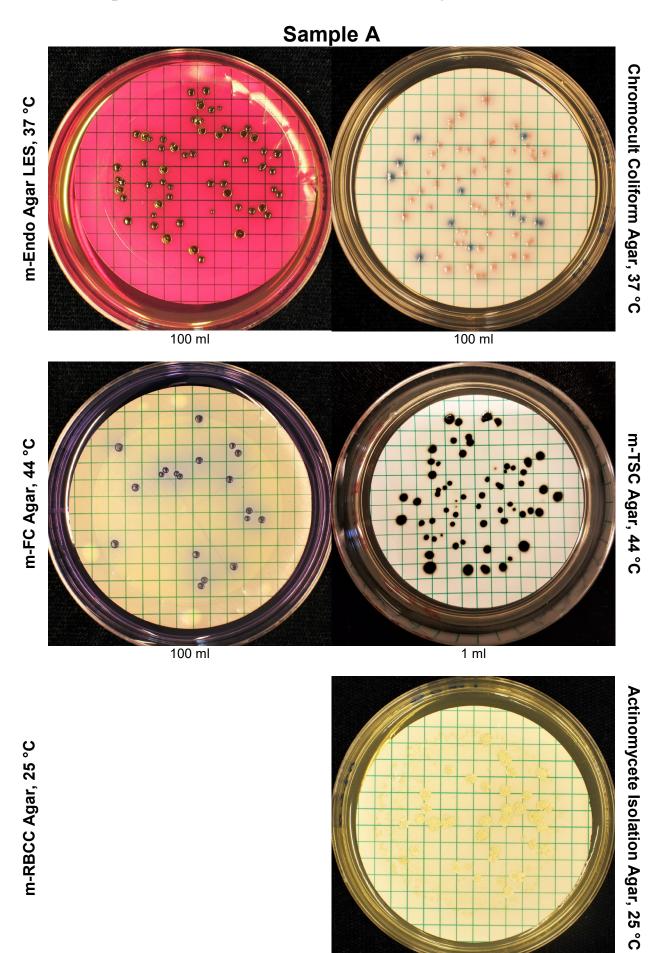
* The 21 original zero results has been given the z-score zero (0.000)

positive results no z-scores can be calculated. Z-scores form outliers are not real z-scores but a practical means to express also the results from the outliers. Very low and high values are here limited to -4 and +4, respectively.

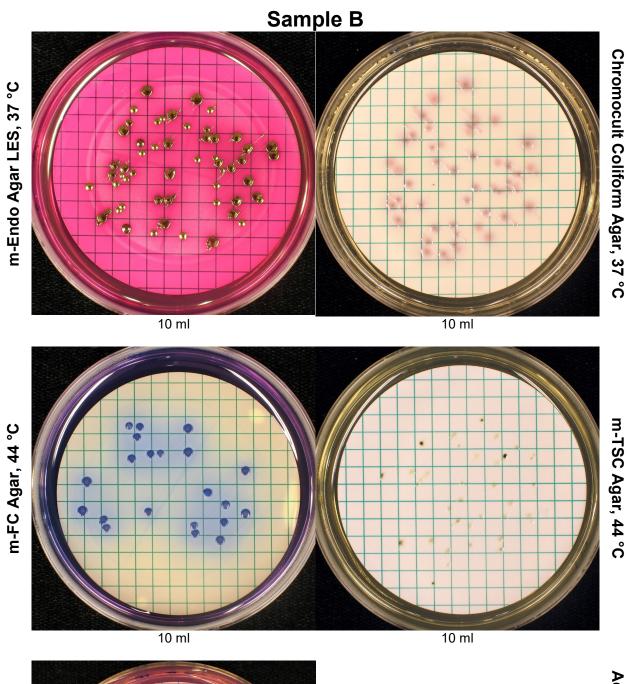
Presumptive C. Clostridium perfringens (MF) perfringens (MF)				Moulds (MF)			Yeasts (MF)			Actinomycetes (MF)				l plate c °C, 3 da		Slo [®] bacteria	Lab no.				
A	B	C	A	B	C C	Α	в	С	Α	в	С	Α	в	С	A	B	C	A	B B	C	
1.297	-1.989	0.000		0.000												-1.078 -0.583	1.209 -1.262		-0.689 -0.341	1.601 0.015	1131 1237
1.366	1.252	0.000	1.336 1.200	0.000	0.000 0.000		1.030 1.030		0.000 0.000	1.254 0.529	1.114 0.225	-0.146 -1.493	0.000 0.000	0.000 0.000		1.017 -1.078		-1.160	-0.014	0.380	1290 1545 1594 1611
1.088 -0.578 -0.950	-0.562 0.328 0.437	0.000 0.000 0.000	-1 123	0.000	0 000	0.000		0.506	0.000	-0.253 -1.286 1.599	0.106 0.341 2.015	0.784 -0.524		0.000 0.000	-0.270 0.908	0.275 -0.583 -1.078	-0.633 -1.213	-1.008	-0.689	0.149	1753 1868 1970
-1.965	0.015		-4.000 -2.141	0.000	0.000 0.000	0.000	1.140	0.000	0.000	1.000	2.010				-1.551 0.745 0.099	-1.078 -0.136 -0.583	0.692 -0.986 -0.233				2317 2637 2745
			-0.065	0.000	0.000										-1.935 0.644	-0.583 1.357 -1.078 -1.639	-0.707 0.143	-1.792	-0.341	-1.702	2944 3055 3076 3145
1.088 -0.401		0.000	0.330 -0.572	0.000	0.000	0.000	-0.195 1.030	0.438 0.155		1.077 0.715		1.388	0.000 0.000	0.000 0.000	-0.785 1.127 0.711	1.680 0.275	-0.359	0.725	0.589	0.520	3155 3162 3305
	-0.142 0.647						-0.401 -1.350			-1.332 -0.819	0.456	-0.758 0.364	0.000	0.000			0.478 1.368	0.387		0.737 0.737	3883 3897 4015
1.016	0.463	0.000	0.849	0.000	0.000	0.000	-0.542		0.000	-0.253	0.456				-0.270 -0.227	0.275 1.017	-1.868 0.527	-1.212	-0.689	0.408	4288 4339
0.945	-2.156 0.162	0.000	0.777	0.000	0.000	0.000	-0.471	-3.170	0.000	-0.884		-0.680		0.000	-1.772 -0.227	-0.583 -0.136 -1.078	-1.474 -1.312		-0.689	0.843	4343 4356 4635
-0.369 -0.228	2.773 0.162 0.566	0.000 0.000 0.000	-0.399	0.000			-0.471 -1.309			-1.286 -0.051	-0.780	0.364 -3.796	0.000			0.275 -1.639 1.357	0.981	1.103	-0.014	0.394	4723 4889 5018
-4.000	0.925	0.000	-4.000	0.000	0.000	0.000	-0.578	-2.212	0.000	0.146	-4.000	-4.000	0.000	0.000	-0.185 -0.588 -0.227	0.657 -0.583 1.357	1.289 0.004 1.148				5120 5201 5333
	0.191 1.146	0.000 0.000	-0.314 -1.885		0.000 0.000		0.744 -4.000	2.056		-0.458 -1.709		-0.837 -0.524	0.000 0.000	0.000 0.000		-2.305 -0.583			0.589 2.790		5352 5447 5553
-0.950	-1.093	0.000	0.214 -1.123	0.000	0.000 0.000	0.000	0.415	0.299	0.000	0.585	-0.780	-1.242	0.000	0.000	0.908 0.541		0.761 0.894		0.589		5858 5950 6175
															0.876 0.326	0.657			-0.341		6182 6233 6253
-4.000	-1.720		-4.000 0.175		0.000		-4.000 -0.195	-1.753	0.000	-2.851 1.428	4.000				-0.588 -2.879	1.017 0.275	0.981	-1.212	-2.429	-2.920	6265 6421 6448
1.297		0.000													0.399	-0.583 0.275 -0.136	-1.722 0.646 0.429	0.135	-0.689	0.164	6456 6563 6686
-0.314	1.037 -0.633 0.075	0.000 0.000 0.000	0.253	0.000	0.000		-2.218 -0.912		0.000	0.339		1.190	0.000	0.000 0.000 0.000		1.357 -0.136 -1.639		-1.108	4.000	0.588	7248 7442 7688
-0.314		0.000	0.921		0.000	0.000	0.133		0.000	0.339	1.114	0.221		0.000	-1.551 -0.636		0.575 0.669 0.828	-1.212	-1.063	0.164	7728 7876 7930
-0.669	0.437	0.000	-0.841	0.000	0.000	0.000 0.000	-1.106 1.828		0.000 0.000	-1.105 0.715	0.456 -0.387				0.060 -0.889	1.017 1.357	0.828 0.938				7962 7968
0.499	-1.892	0.000	-0.230 0.330	0.000 0.000	0.000 0.000	0.000			0.000	-0.051 -0.253	-1.493	0.504			-0.314 0.644	-0.583	0.783 -1.418		-0.341		8019 8068 8260
	-1.397			0.000	0.000	0.000	-0.542	0.299	0.000	-1.332	0.081	-0.524	0.000	0.000	-1.483 -0.942	1.989	0.715 -1.119		-1.469		8329 8380 8435
	-0.714			0.000		0.000	0.445		0.000	0.529	0.570				-0.227 -0.270	0.657 -2.305 -1.639	-1.418 0.828		1.139 -0.341		8569 8626 8628
-1.451			-1.626	0.000	0.000	0.000	4 400		0.000	0.505	0.000	0.047	0.000	0.000	-0.494 0.399	0.657 0.275 1.017	-1.262 1.044	-0.132	0.017	0.237	8663 8742 8751
	0.925 0.437 0.351	0.000		0.000		0.000 0.000	1.439 2.150 -0.162		0.000 0.000	0.585 0.752 1.077	-0.516 -1.056	0.921 -0.997	0.000 0.000 0.000	0.000 0.000	0.711 0.940	-0.136 1.017 1.680	0.669 0.622	0.351 0.789	-0.014 -0.341 1.139	0.506 0.575	8766 8862 8898
	-1.292 -0.142		1.403	0.000 0.000		0.000	0.445 1.196	1.252		0.715 0.585			0.000		-0.636	-0.136 -1.078 0.657	0.715		1.649 -0.014		8955 9051 9436
-3.116	0.835	0.000				0.000	0.859			0.434	-0.917	1.322	0.000	0.000	0.908	0.275	0.004		0.870		9524 9736 9899
-0.724		0.000				0.000					3.013	1.583	0.000			0.275			-0.014		9903
	40 -2.156 2.773		29 -4.000 1.403		28 0.000 0.000		37 -4.000 2.150			38 -4.000 1.599	36 -4.000 4.000	27 -4.000 4.000	28 0.000 0.000	28 0.000 0.000		73 -2.305 1.989	73 -1.868 1.368		30 -2.429 4.000		n Min Max
	0.247 0.000 1.000	0.000	0.016 -0.414 1.559		0.000 0.000 0.000	0.000	-0.162 -0.216 1.336	-0.132	0.000	0.281 -0.105 1.181	0.000	-0.141	0.000 0.000 0.000	0.000	0.000	0.275 0.000 1.000	0.000	0.000	-0.014 0.133 1.224	0.000	Median Mean SD
3 0 0 0	0 1 1 0	0 0 0	3 1 0 0	0 0 0 0	0 0 0	0 0 0	2 1 1 0	1 2 1 0	0 0 0	1 1 0 0	1 0 1 2	2 0 0 1	0 0 0	0 0 0 0	0 3 0 0	0 2 0 0	0 0 0 0	0 0 0	0 1 1 1	0 2 0 0	<u>Sum</u> 18 24 15 9

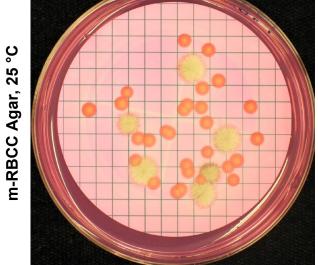
Annex C – photos

100 ml, 7 days



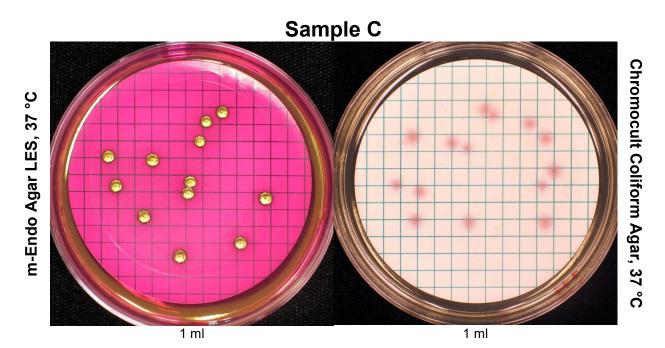
42 PT Microbiology – Drinking water, March 2020



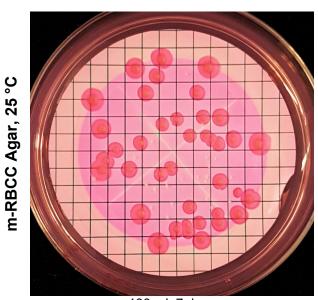


10 ml, 7 days

Actinomycete Isolation Agar, 25 °C



−Fr dar, 41 °C T mI



100 ml, 7 days

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m-TSC Agar, 44 °C

PT reports published 2019

Proficiency Testing – Food Microbiology, January 2019, by Jonas Ilbäck

Proficiency Testing – Drinking Water Microbiology, March 2019, by Tommy Šlapokas

Proficiency Testing - Food Microbiology, April 2019, by Jonas Ilbäck

Proficiency Testing – Drinking Water Microbiology, September 2019, by Tommy Šlapokas

Proficiency Testing - Food Microbiology, October 2019, by Jonas Ilbäck

PT reports published 2020

Proficiency Testing – Food Microbiology, January 2020, by Jonas Ilbäck

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 laboratories carry out some form of internal quality assurance, but their analytical work also has 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 proficiency test, identical test material is analysed by a number of laboratories using their routine methods. The laboratories report their results to the organiser that evaluates them and compiles them in a report.

The National Food Agency's PT program offers

- > External and independent evaluation of laboratories analytical competence.
- Improved knowledge of analytical methods with respect to various types of organisms.
- Expert support.
- > Tool for inspections regarding accreditation.
- Free extra material for follow-up analyses

For more information, visit our website: https://www2.slv.se/absint

The National Food Agency's reference material

As a complement to the proficiency testing but without specific accreditation, National Food Agency also produces reference material (RM) for internal quality control: a total of 8 RM for food and drinking water microbiological analyses, including pathogens, are available.

Information available on our website: https://www.livsmedelsverket.se/en/RM-micro