NF VALIDATION
Validation of alternative analysis methods
Application to the food industry

Summary report
according to the standard EN ISO 16140-2:2016

Quantitative method

RAPID'*E. coli* 2 (REC2)
Certificate # BRD 07/01 – 07/93
for the enumeration at 44°C of β-glucuronidase positive
Escherichia coli in human food products

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This report contains 57 pages, including 34 pages of appendices.
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Version 0

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Preamble

- **Protocols of validation:**

- **EN ISO 16140-1 and NF EN ISO 16140-2 (September 2016): Microbiology of the food chain — Method validation**
  Part 1: Vocabulary.
  Part 2: Protocol for the validation of alternative (proprietary) methods against a reference method.

- **Requirements regarding comparison and interlaboratory studies for implementation of the standard EN ISO 16140-2 (version 12).**

- **Reference method:**

- **NF ISO 16649-2 (July 2001):** horizontal method for the enumeration of β-glucuronidase positive *Escherichia coli*– Colony count technique at 44°C using 5-bromo-4-chloro-3-indolyl β-glucuronate

- **Application scope:**

- **All human food products** by a validation testing of a broad range of foods, including:
  - meat products,
  - dairy and egg products,
  - seafood products,
  - vegetal products,
  - ready-to-eat and ready-to-reheat products,

- **Certification body:**

- **AFNOR Certification** (https://nf-validation.afnor.org/).

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Definitions

- ## Method comparison study

The method comparison study is the part of the validation process that is performed in the expert laboratory. It consists of four parts:

- A comparative study of the results of the reference method to the results of the alternative method in a variety of different items (naturally and/or artificially) contaminated samples (so-called relative trueness study).
- A comparative study of the results of the reference method to the results of the alternative method in artificially contaminated samples using replicates of a single item per category. The data are analyzed using the accuracy profile (AP) approach (so-called AP study).
- An inclusivity/exclusivity study of the alternative method.

- ## Relative trueness study

The relative trueness study is a comparative study between the results obtained by the reference method and the results of the alternative method.

The relative trueness is the degree of correspondence between the response obtained by the reference method and the response obtained by the alternative method on identical samples.

- ## Accuracy profile study

The accuracy profile study is a comparative study between the results obtained by the reference method and the results of the alternative method.

The accuracy profile is the graphical representation of the capacity of measurement of the quantitative method, obtained by combining acceptability intervals and β-expectation tolerance intervals, both reported to different levels of the reference value.

- ## Inclusivity and exclusivity study

The inclusivity study is a study involving pure target strains to be detected or enumerated by the alternative method.

The exclusivity study is a study involving pure non-target strains, which can be potentially cross-reactive, but are not expected to be detected or enumerated by the alternative method.

- ## Interlaboratory study

The interlaboratory study is a study performed by multiple laboratories testing identical samples at the same time, the results of which are used to estimate alternative-method performance parameters.

The aim of an interlaboratory study is to determine the variability of the results obtained in different laboratories using identical samples.

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1. Introduction

The RAPID'*E. coli* 2 method is validated by AFNOR Certification under the NF VALIDATION mark with the certification number BRD 07/01 – 07/93 according to the standard ISO 16140-2:2016 for the enumeration of β-glucuronidase *Escherichia coli* at 44°C. The method is intended for all human food products since its initial validation.

The method has been initially validated according to the AFNOR rules in 1993, then renewed according to the same standard in 1997 and 2002.Table 1 summarizes the different steps of the validation that occurred since 2004, where the first validation according to both the ISO 16140 and the ISO 16649-2 standards occurred.

Table 1: Steps of the validation AFNOR certification

Study	Date	Standards	Expert Laboratory	Observation
Initial validation	December 2004	ISO 16140:2003 ISO 16649-2:2001	Eurofins IPL Nord	/
First renewal	November 2008	ISO 16140:2003 ISO 16649-2:2001	Eurofins IPL Nord	/
Second renewal	November 2012	ISO 16140/A1:2011 ISO 16649-2:2001	Eurofins IPL Nord	New statistical analysis of the interlaboratory study data
Third renewal	November 2016	ISO 16140-2:2016 ISO 16649-2:2001	Institut Scientifique d'hygiène et d'Analyse	Additional tests to fulfill the updated validation standard and reinterpretation
Fourth renewal	October 2020	ISO 16140-2:2016 ISO 16649-2:2001	Microsept	/
Fifth renewal	October 2024	ISO 16140-2:2016 ISO 16649-2:2001	Microsept	No additional tests

The present document is a summary report of the overall NF Validation certification of the RAPID'*E. coli* 2 method according to the standard ISO 16140-2:2016 for a broad range of foods.

The results set out in this report were produced during validation tests carried out by Eurofins IPL Nord and by the Institut Scientifique d'Hygiène et d'Analyse as part of NF Validation, in accordance with prevailing requirements.

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2. Protocols of the methods

2.1. Alternative method

2.1.1. Principle of the method

The method uses a chromogenic agar media for the enumeration of *E. coli*.

The principle of the agar media relies on the simultaneous revelation of two enzymatic activities: the β-D-glucuronidase (GLUC) and the β-D-galactosidase (GAL).

The agar media contains two chromogenic substrates:
- a specific substrate of the GAL which leads to a blue coloration of the colonies positive for this enzyme,
- a specific substrate of the GLUC which leads to a pink coloration of the colonies positive for this enzyme.

E. coli (GAL+/GLUC+) form violet to pink colonies.

2.1.2. Protocol of the method

The diagram summarizing the method is shown in Appendix A.

From an initial suspension ten-times diluted or directly from the samples if it's liquid, volumes of 1 mL are inoculated in Petri dishes. Several decimal dilutions can also be realized and poured.

The melted RAPID'*E. coli* 2 agar media at 44-47°C is then poured in Petri dishes. The medium is used in single layer to enhance readability and practicability.

The RAPID'*E. coli* 2 agar media are incubated at 44±1°C for 21±3 hours.

After incubation, the violet to pink colonies are enumerated as β-glucuronidase *Escherichia coli*.

2.1.3. Restrictions

There are no restrictions on use for the RAPID'*E. coli* 2 method.

2.2. Reference method

The ISO 16649-2:2001 standard, Horizontal method for the enumeration of β-glucuronidase positive *Escherichia coli*– Colony count technique at 44°C using 5 bromo-4-chloro-3-indolyl β-glucuronate, was used for the initial validation study, for the first and the second renewal studies and for the present renewal study.

The workflow of the reference method is presented in Appendix B.

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3. Methods comparison study

The study was conducted on a variety of samples and strains representative of food products. This is not an exhaustive list of the various matrices included in the application scope. For any remark on the alternative method, you can contact AFNOR Certification by connecting to the Internet page *http://nf-validation.afnor.org/contact-2/*.

3.1. Relative trueness study

3.1.1. Number and nature of the samples

Considering all the validation studies, 172 samples were analyzed giving 89 exploitable results.
The distribution of the samples per category, type and inoculation technique is given in table 2.

Table 2: number and nature of the samples analyzed for the relative trueness study (IVS: initial validation study, TRS: third renewal study)

Category	Type		Samples analyzed	Interpretable results
Meats products	a	Raw products	21	7
	b	Ready-to-reheat products	7	6
	c	Smoked and cured products	7	5
	Total		**35**	**18**
Dairy & egg products	a	Pasteurized and dehydrated products	10	5
	b	Raw milk products	10	5
	c	Desserts and egg products	12	11
	Total		**32**	**21**
Seafood products	a	Raw products	15	5
	b	Marinated and smoked products	12	10
	c	Ready-to-reheat products	14	5
	Total		**41**	**20**
Vegetal products	a	Raw products	15	5
	b	Pre-cut and pre-cooked products	10	5
	c	Processed products	8	5
	Total		**33**	**15**
Ready-to-eat & ready-to-reheat products	a	Ready-to-eat products	16	5
	b	Ready-to-reheat products	9	5
	c	Smoked and marinated products	6	5
	Total		**31**	**15**
Total			**172**	**89**

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3.1.2. Artificial contaminations

Naturally contaminated samples were analyzed preferably.
However, artificially contaminated samples were still analyzed, using seeding or spiking protocols as described in the standard NF EN ISO 16140-2:2016.

Among the interpretable results, forty-three correspond to naturally contaminated samples and forty-six to artificially contaminated samples. The artificial contaminations performed are presented in the Appendix C.

3.1.3. Protocols used during the study

The samples were analyzed by the reference and the alternative method. For the alternative method, the minimum incubation time of the Petri dishes was applied, namely 18 hours.

3.1.4. Results

Samples were analyzed by the reference and the alternative method so as to obtain at least 15 interpretable results per category and at least 5 per type.

Raw results are shown in Appendix D.

Three kinds of results are not considered as part of the statistical calculations:
- Those expressed with less than 4 colonies per Petri dish for at least one method or inoculation modality,
- those lower or higher than the quantification limits,
- Undetermined results.

All results are presented in scatter plots per category and for all categories in figure 1.

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Figure 1: Two-dimensional plots per category and for all categories

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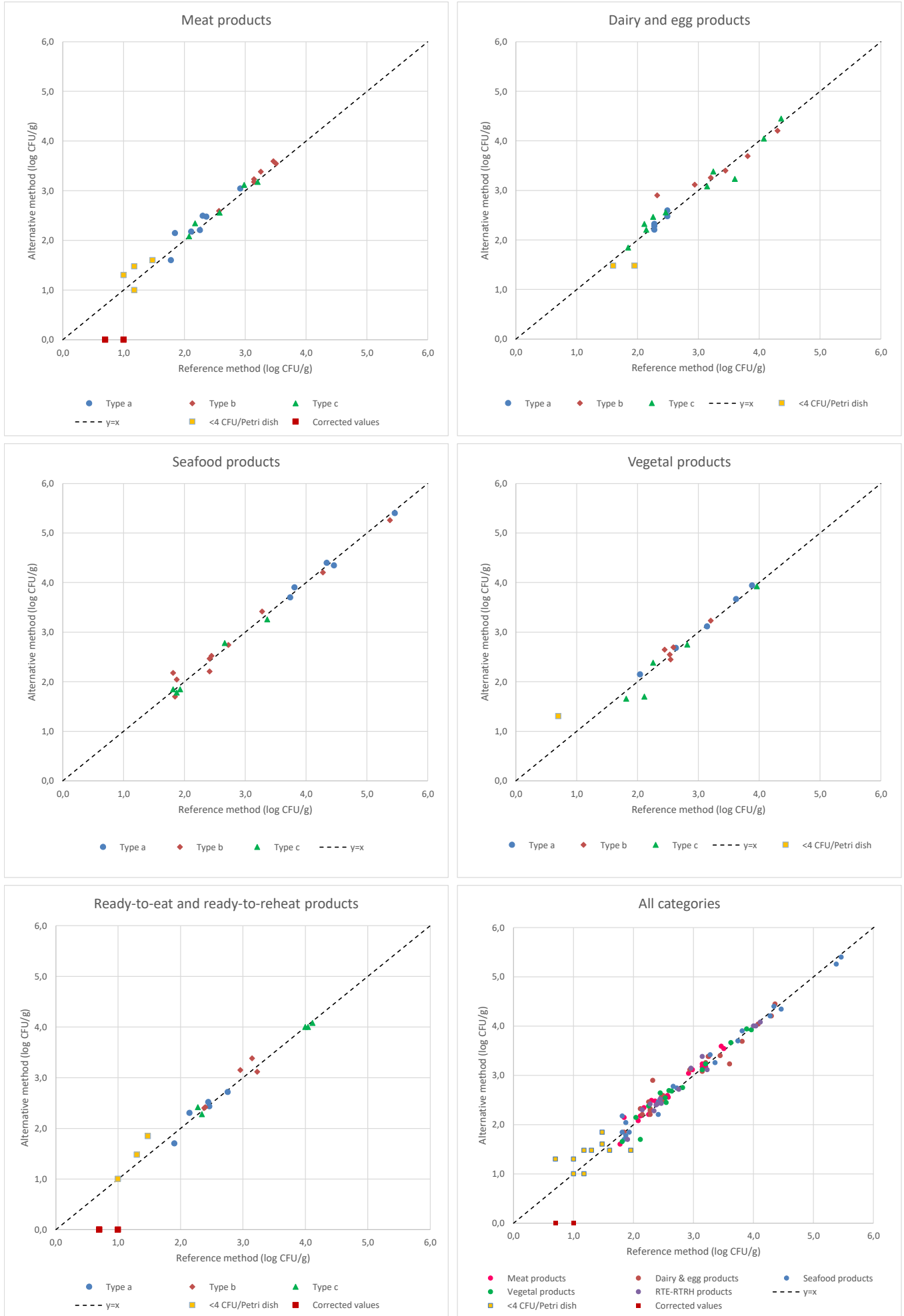
3.1.5. Calculation and interpretation of relative trueness study

The results obtained are analyzed using the Bland-Altman method.
Statistical calculations are presented in Appendix E, as well as the results excluded from the statistical analysis per category, type and modality of inoculation.

Table 3 presents the summary of the average differences and standard deviation differences per category and for all categories.

Table 3: values for the Bland-Altman difference plot

Category	n	Average difference = bias	Standard deviation differences	Lower Confidence Limit	Upper Confidence Limit
Meat products	18	0.07	0.11	-0,16	0,30
Dairy products	21	0.04	0.18	-0,34	0,42
Seafood products	20	0.00	0.14	-0,29	0,29
Vegetal products	15	-0.01	0.15	-0,33	0,31
Ready-to-eat & ready-to-reheat products	15	0.02	0.12	-0,24	0,16
All categories	**89**	**0.03**	**0.14**	**-0.25**	**0.31**

Overall, the average difference is equal to 0.03, showing no bias between the RAPID'*E. coli* 2 method and the reference method.
The average difference varies from -0.01 log CFU/g (vegetal products) to 0.07 CFU/g (meat products).

Upper and lower 95% confidence interval limits are lower than 0.5 log showing a good correlation between the two methods.

The Bland-Altman difference plots are presented for all categories in figure 2.

As on scatter plots:
- Each category is differentiated by a specific colour,
- Results expressed with less than 4 colonies per Petri dish for at least one method are indicated by a yellow square,
- Results lower or higher than the quantification limits for one method are indicated by a red square. The value of these results is corrected according to the EN ISO 16140-2:2016 requirements.

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Figure 2: Bland-Altman difference plot for all categories

- Meat products
- Vegetal products
- Corrected values
- Upper confidence limit
- Dairy & egg products
- RTE-RTRH products
- Bias
- Seafood products
- Values <4 CFU/Petri dish
- Lower confidence limit
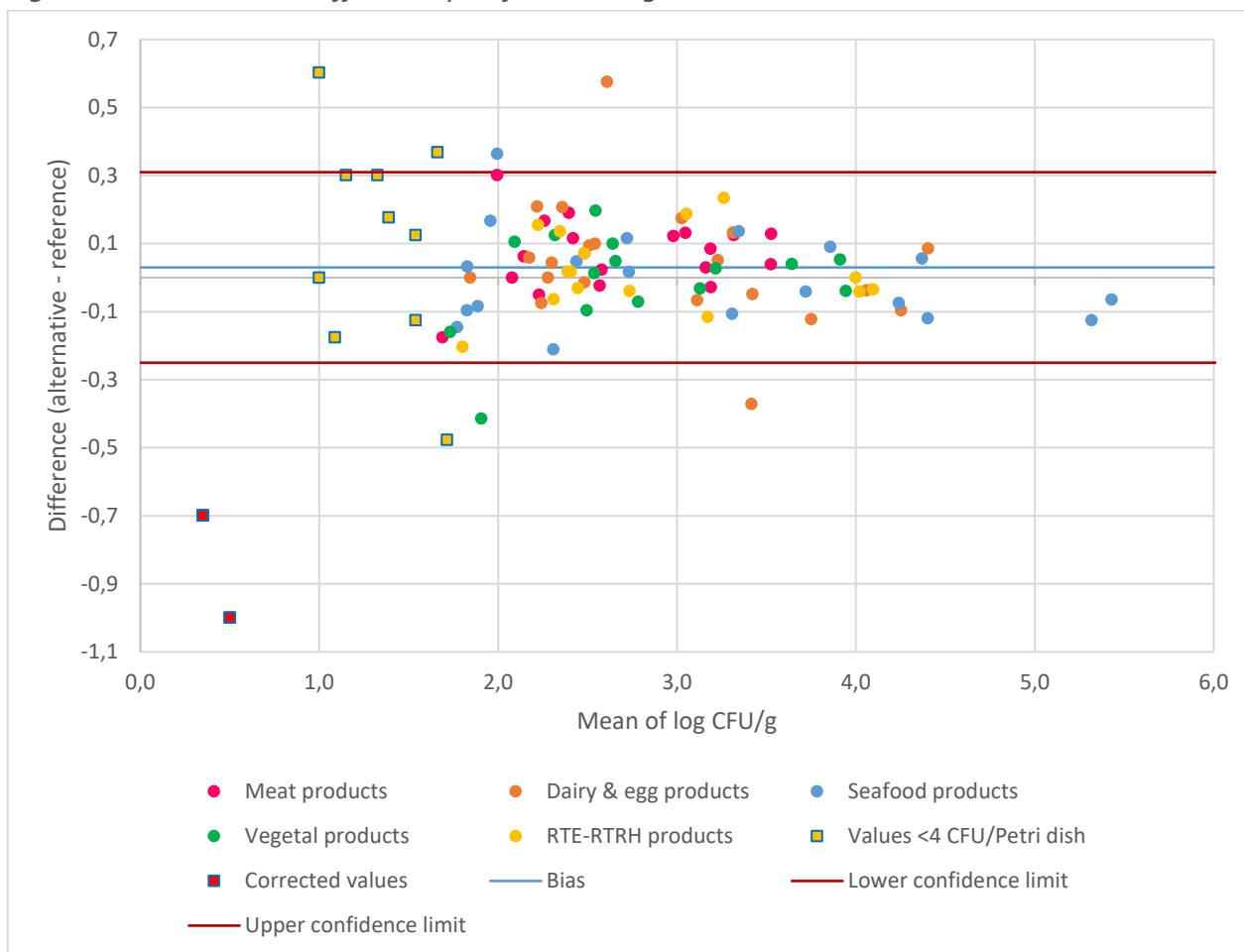
- Observations:

Samples for which the average difference is lower or higher than the confidence limits are listed in table 4.

Table 4: values outside the confidence limits on the Bland-Altman difference plot (green cases: values <4 CFU/Petri dish, yellow cases: values lower and higher than the quantification limits, blue: values higher than the confidence limits, red: values lower than the quantification limits)

Category	Type	#	RM CFU/g	AM CFU/g	RM log CFU/g	AM log CFU/g	Mean	Difference
Meat products	a	26	10	0	1,00	0,00	0,50	-1,00
	a	28	5	0	0,70	0,00	0,35	-0,70
Dairy and egg products	b	25	210	790	2,32	2,90	2,61	0,58
	c	72	4000	1700	3,60	3,23	3,42	-0,37
	c	20	90	30	1,95	1,48	1,72	-0,48
Seafood products	b	ISHA 31	65	150	1,81	2,18	1,99	0,36
Vegetal products	b	ISHA 15	5	20	0,70	1,30	1,00	0,60
	c	ISHA 6	130	50	2,11	1,70	1,91	-0,41
Composite foods	a	17	5	0	0,70	0,00	0,35	-0,70
	a	19	5	0	0,70	0,00	0,35	-0,70
	a	57	30	70	1,48	1,85	1,66	0,37
	b	ISHA 37	10	0	1,00	0,00	0,50	-1,00

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Twelve samples are outside the confidence limits: 8 concern corrected values or samples with less than 4 CFU/Petri dish, 2 are higher than the upper confidence limit and 2 are lower than the lower confidence limit.

3.1.6. Conclusion
The relative trueness study of the alternative method is satisfactory.

3.2. Accuracy profile study

3.2.1. Protocols
Five matrix-strain couples were tested by both methods. Two batches of a matrix, representative of each category, were inoculated with a strain of *Escherichia coli* at three levels (low, medium and high). For each sample, 5 replicates, represented by 5 different test portions, were tested by each method. This represents a total of 30 analyses per method.

The matrix-strain couples are presented in table 5.

Table 5: matrix-strain couples for the accuracy profile study

Category	Matrix	Strain	Origin of the strain	Contamination Level (log CFU/g)
Meat products	Ground beef	*E. coli* ESC.1.15	Ground beef 20% fat	
Dairy products	Raw milk cheese	*E. coli* ESC.1.5	Camembert	2.5
Seafood products	Fish fillet	*E. coli* ESC.1.31	Scallop shell	3.5
Vegetal products	Grated carrots	*E. coli* ESC.1.2	Grated carrots	
Composite foods	Piemontese salad	*E. coli* ESC.1.28	Salad pasta, surimi, mayonnaise	5.0

3.2.2. Results
Raw data are provided in Appendix F.

The statistical data and the accuracy profiles are shown in figure 3.

Statistical calculations were done according to the Excel spreadsheet named __AP_calculation_tool MCS 16140-2_clause 6-1-3-3 ver_31-07-2018.xlsx__ available at http://standards.iso.org/iso/16140.

The probability for the tolerance interval is set at 80% and the central value is the median.
The acceptability limit is set at AL = 0.5 $\log_{10}$ CFU/g or ml.

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Figure 3: Accuracy profiles per category
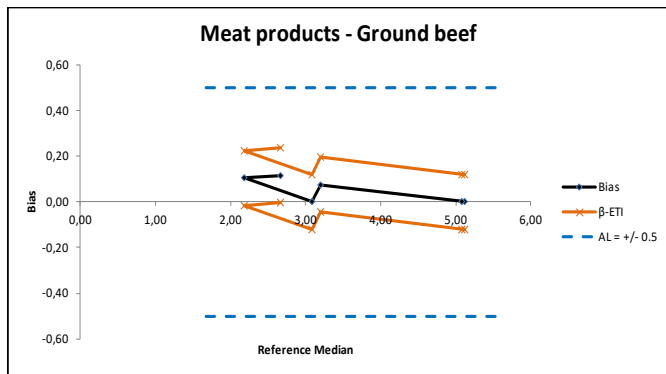
Meat products - Ground beef

Sample Name	Reference central value	Bias	Lower β-ETI	Upper β-ETI	β-ETI compared to AL=±0.5 Acceptable	β-ETI compared to final AL Acceptable
	2,66	0,115	-0,004	0,235	YES	YES
	2,18	0,103	-0,017	0,222	YES	YES
	3,08	0,000	-0,120	0,120	YES	YES
	3,20	0,075	-0,045	0,194	YES	YES
	5,11	0,000	-0,120	0,120	YES	YES
	5,08	0,000	-0,120	0,120	YES	YES

	Reference method	Alternative method	SD repeatability of reference method <= 0.125	Final AL
SD Repeatability	0,085	0,083	YES	+/- 0,500
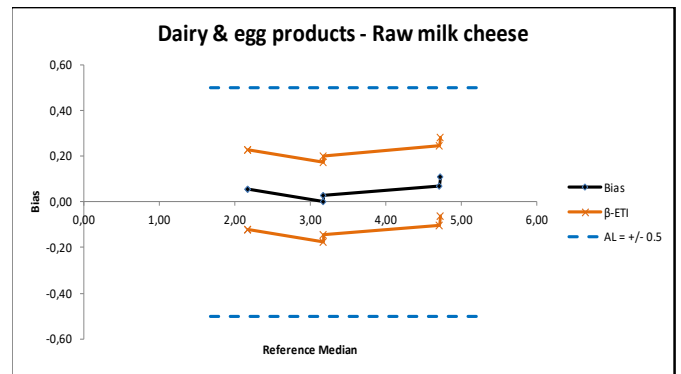
Dairy & egg products - Raw milk cheese

Sample Name	Reference Central value	Bias	Lower β-ETI	Upper β-ETI	β-ETI compared to AL=±0.5 Acceptable	β-ETI compared to final AL Acceptable
	2,18	0,054	-0,119	0,228	YES	YES
	2,18	0,054	-0,119	0,228	YES	YES
	3,18	0,000	-0,173	0,173	YES	YES
	3,18	0,028	-0,145	0,201	YES	YES
	4,71	0,071	-0,103	0,244	YES	YES
	4,72	0,110	-0,063	0,283	YES	YES

	Reference method	Alternative method	SD repeatability of reference method <= 0.125	Final AL
SD Repeatability	0,057	0,120	YES	+/- 0,500
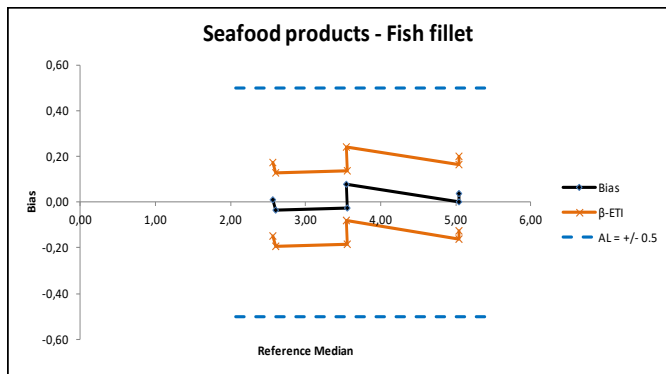
Seafood products - Fish fillet

Sample Name	Reference Central value	Bias	Lower β-ETI	Upper β-ETI	β-ETI compared to AL=±0.5 Acceptable	β-ETI compared to final AL Acceptable
	2,57	0,012	-0,150	0,173	YES	YES
	2,60	-0,034	-0,196	0,128	YES	YES
	3,56	-0,025	-0,187	0,137	YES	YES
	3,54	0,079	-0,083	0,241	YES	YES
	5,04	0,000	-0,162	0,162	YES	YES
	5,04	0,038	-0,124	0,199	YES	YES

	Reference method	Alternative method	SD repeatability of reference method <= 0.125	Final AL
SD Repeatability	0,072	0,112	YES	+/- 0,500
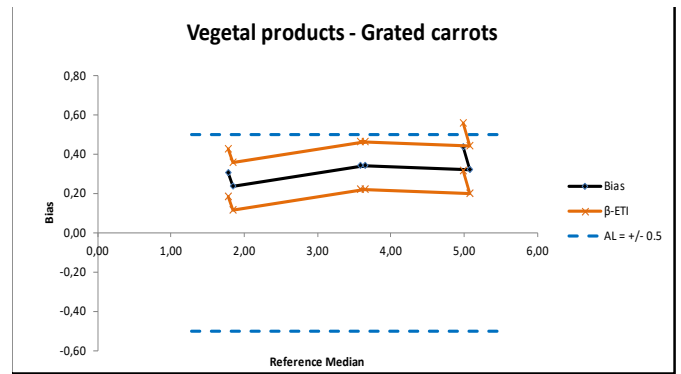
Vegetal products - Grated carrots

Sample Name	Reference Central value	Bias	Lower β-ETI	Upper β-ETI	β-ETI compared to AL=±0.5 Acceptable	β-ETI compared to final AL Acceptable
	1,78	0,301	0,180	0,422	YES	YES
	1,85	0,234	0,113	0,355	YES	YES
	3,64	0,339	0,218	0,460	YES	YES
	3,58	0,339	0,218	0,461	YES	YES
	5,08	0,319	0,197	0,440	YES	YES
	4,98	0,433	0,311	0,554	NO	NO

	Reference method	Alternative method	SD repeatability of reference method <= 0.125	Final AL
SD Repeatability	0,100	0,084	YES	+/- 0,500
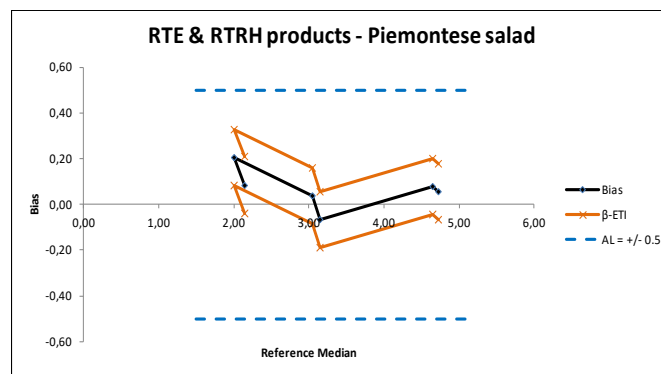
RTE & RTRH products - Piemontese salad

Sample Name	Reference Central value	Bias	Lower β-ETI	Upper β-ETI	β-ETI compared to AL=±0.5 Acceptable	β-ETI compared to final AL Acceptable
	2,15	0,084	-0,038	0,207	YES	YES
	2,00	0,204	0,081	0,327	YES	YES
	3,04	0,038	-0,085	0,160	YES	YES
	3,15	-0,067	-0,190	0,056	YES	YES
	4,65	0,079	-0,044	0,202	YES	YES
	4,72	0,054	-0,069	0,177	YES	YES

	Reference method	Alternative method	SD repeatability of reference method <= 0.125	Final AL
SD Repeatability	0,080	0,085	YES	+/- 0,500

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The tolerance intervals fall into the acceptability limits for all categories, except for the matrix "grated carots". For this matrix, the upper tolerance interval is higher than the acceptability limit of 0.5 log CFU/g for one of the two batches, which illustrates a higher enumeration by the alternative method.

3.2.3. Conclusion
The alternative method is accepted as being equivalent to the reference method.

3.3. Specificity / selectivity

3.3.1. Protocols
Fifty positive strains and fifty-four negative strains were tested in duplicate by the alternative method.

Results are shown in Appendix G.

3.3.2. Results
All the β-glucuronidase positive *Escherichia coli* strains cultivated on the agar media of the alternative method and gave typical colonies.

All the negative strains, when they cultivated on the agar media of the alternative method, showed a non-typical aspect, except for a strain of *Shigella sonnei* (β-glucuronidase positive) and two strains of *Salmonella arizonae* (lactose positive).
These three strains were tested with the reference method (pouring in TBX agar media). They also gave a typical aspect on the TBX agar media (blue colonies).

3.3.3. Conclusion
The inclusivity and the exclusivity of the alternative method are satisfactory.

3.4. Practicability
The following criteria are precised:

- **Storage conditions of the elements (+ time limit for unopened products):**
The RAPID'*E. coli* 2 is available in:
  - ready-to-use vials of 100 mL,
  - dehydrated powder vials of 500 g.

The storage temperature is indicated on the packages and in the user guides.
The storage temperature is 2-8°C for the ready-to-use agar media and 15–25°C for the dehydrated media.
The validity of the media is 14 months for the ready-to-use agar media and 39 months for the dehydrated agar media.
The expiry date is indicated on the vials.

- **Methods of use after first use (particularly existence of limit dates):**
For the dehydrated agar media, it is necessary to shake the products before every use.

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- Time-to-result:

The results are obtained in 18 to 24 hours for the two methods: the method EN ISO 16649-2 and the RAPID'*E. coli* 2method.

- Common steps with the reference method

The alternative method and the reference method have one common step: the dilution of the sample in an appropriate diluent.

3.5. General conclusion for the methods comparison study

The relative trueness study shows a good correlation between the alternative method and the reference method.

The accuracy profile study illustrates that the performances of the alternative method are comparable to those of the reference method.

The alternative method is specific and selective.

Time-to-result is equivalent to that of the reference method (one day).

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4. Interlaboratory study

4.1. Study organization

The interlaboratory study has been realized in November 2001. The samples were sent to 16 collaborators.

The collaborators received 8 samples of pasteurized milk (2 vials per contamination level; 4 contamination levels) to perform the analyses with the reference method NF ISO 16649-2 and with the alternative method REC2.

The strain used for the contamination of the samples was a β-glucuronidase positive *Escherichia coli* isolated from a pastry.

4.2. Control of experimental parameters

4.2.1. Contamination levels obtained after artificial contamination

The four contamination levels are detailed in the following table.

Table 6: Contamination levels of inoculated samples

Level	Targeted level (CFU/ml)	Real level (CFU/ml)
Level 0 (L_0)	0	0
Level 1 (L_1)	50	40
Level 2 (L_2)	500	400
Level 3 (L_3)	5000	4100

4.2.2. Temperatures during shipping and at reception, delivery times

The temperatures recorded at reception of the packages confirm that the samples were stored at correct temperatures (between 1.3°C and 6.4°C), except for collaborator C (9.2°C).

Among the 16 collaborators, 15 received the samples the day following the shipping. One collaborator (J) received the samples two days after the shipping. The temperature at reception was not communicated by this collaborator.

4.2.3. Conclusion

The collaborator J, which received the samples one day after the other collaborators, didn't realize the analyses.

The results obtained by the collaborator C were not taken into account because of the temperature during the shipping of the samples that was over the limits.

Finally, regarding the conditions of the delivery of the samples, the results of fourteen collaborators were kept.

4.3. Results

4.3.1. Expert laboratory

The results obtained by the expert laboratory are shown in the table below. Analyses were performed after a storage of the samples for 24 hours at 2°C and at 8°C and for 18 and 24 hours of incubation of the Petri dishes.

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Table 7: Results of the Expert Laboratory (in CFU/ml) after a storage of the samples for 24 h at 2°C and 8°C

Storage temperature	Level	Reference method				Alternative method			
		Incubation 18 h		Incubation 24 h		Incubation 18 h		Incubation 24 h	
		R1	R2	R1	R2	R1	R2	R1	R2
Storage at 2°C for 24 h	L_0	<1	<1	<1	<1	<1	<1	<1	<1
	L_1	37	39	37	39	47	35	47	35
	L_2	390	460	390	460	400	440	400	440
	L_3	4500	3800	4500	3800	4500	4700	4500	4700
Storage at 8°C for 24 h	L_0	<1	<1	<1	<1	<1	<1	<1	<1
	L_1	54	47	54	47	47	50	47	50
	L_2	380	430	380	430	440	550	440	550
	L_3	4500	4700	4500	4700	5500	3600	5500	3600

Results according to the ISO 16649-2 standard and according to the alternative method were in agreement.

The results after storage at 2°C and 8°C didn't show any significant evolution of the number of *Escherichia coli* per mL. In the same way, no difference appeared between the readings of the Petri dishes after 18 hours of incubation and after 24 hours of incubation.

4.3.2. Results obtained by the collaborators

The results of the 14 laboratories which realized the analysis are summarized in the tables 8 and 9 for the three levels of contamination.

Collaborator O did not give results for the low level of contamination (L_1) because of an issue during the pouring of the Petri dishes.

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Table 8 : results of the collaborators (CFU/ml)

Collaborator	Level 0				Level 1				Level 2				Level 3			
	Reference method		Alternative method		Reference method		Alternative method		Reference method		Alternative method		Reference method		Alternative method	
	R1	R2	R1	R2	R1	R2	R1	R2	R1	R2	R1	R2	R1	R2	R1	R2
A	<1	<1	<1	<1	20	45	19	19	110	100	360	360	3600	4300	3900	3900
B	<1	<1	<1	<1	17	19	34	31	120	350	440	450	2500	3000	4100	4300
D	<1	<1	<1	<1	19	36	48	32	490	490	550	450	5400	5800	6100	5200
E	<1	<1	<1	<1	39	37	32	50	370	430	490	490	3700	3800	5300	3800
F	<1	<1	<1	<1	10	45	60	60	350	320	464	340	4800	4500	6000	5500
G	<1	<1	<1	<1	28	38	37	34	370	510	470	360	3800	4400	3600	3500
H	<1	<1	<1	<1	37	19	44	44	450	200	510	360	6100	6300	5500	5100
I	<1	<1	<1	<1	40	45	53	48	550	460	560	440	5800	5700	6100	6900
K	<1	<1	<1	<1	40	41	35	48	570	520	550	510	5900	5800	6100	7800
L	<1	<1	<1	<1	28	43	50	39	550	400	560	500	7500	7500	5900	7000
M	<1	<1	<1	<1	38	35	44	42	510	570	640	560	5100	4900	5900	8300
N	<1	<1	<1	<1	25	21	38	42	330	340	360	490	4900	4700	5000	4500
O*	<1	<1	<1	<1	-	-	-	-	460	490	350	360	5800	6300	4500	4400
P	<1	<1	<1	<1	39	43	45	36	490	480	570	520	1500	5800	5600	4000

Table 9 : results of the collaborators (log CFU/ml)

Colla-borator	Level 0				Level 1				Level 2				Level 3			
	Reference method		Alternative method		Reference method		Alternative method		Reference method		Alternative method		Reference method		Alternative method	
	R1	R1	R1	R1	R1	R2	R1	R2	R1	R2	R1	R2	R1	R2	R1	R2
A	0	0	0	0	1,301	1,653	1,279	1,279	2,041	2,000	2,556	2,556	3,556	3,633	3,591	3,591
B	0	0	0	0	1,230	1,279	1,531	1,491	2,079	2,544	2,643	2,653	3,398	3,477	3,613	3,633
D	0	0	0	0	1,279	1,556	1,681	1,505	2,690	2,690	2,740	2,653	3,732	3,763	3,785	3,716
E	0	0	0	0	1,591	1,568	1,505	1,699	2,568	2,633	2,690	2,690	3,568	3,580	3,724	3,580
F	0	0	0	0	1,000	1,653	1,778	1,778	2,544	2,505	2,667	2,531	3,681	3,653	3,778	3,740
G	0	0	0	0	1,447	1,580	1,568	1,531	2,568	2,708	2,672	2,556	3,580	3,643	3,556	3,544
H	0	0	0	0	1,568	1,279	1,643	1,643	2,653	2,301	2,708	2,556	3,785	3,799	3,740	3,708
I	0	0	0	0	1,602	1,653	1,724	1,681	2,740	2,663	2,748	2,643	3,763	3,756	3,785	3,839
K	0	0	0	0	1,602	1,613	1,544	1,681	2,756	2,716	2,740	2,708	3,771	3,763	3,785	3,892
L	0	0	0	0	1,447	1,633	1,699	1,591	2,740	2,602	2,748	2,699	3,875	3,875	3,771	3,845
M	0	0	0	0	1,580	1,544	1,643	1,623	2,708	2,756	2,806	2,748	3,708	3,690	3,771	3,919
N	0	0	0	0	1,398	1,322	1,580	1,623	2,519	2,531	2,556	2,690	3,690	3,672	3,699	3,653
O*	0	0	0	0	/	/	/	/	2,663	2,690	2,544	2,556	3,763	3,799	3,653	3,643
P	0	0	0	0	1,591	1,633	1,653	1,556	2,690	2,681	2,756	2,716	3,176	3,763	3,748	3,602

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4.3.3. Conclusion

The results obtained by the collaborators were in agreement with those of the Expert Laboratory. The statistical interpretation has been realized with the results of 14 laboratories, except for L_1 for which the results of 13 laboratories were taken into account.

4.4. Statistical interpretations and calculations

4.4.1. Visual linearity checking

After the log10 transformation of all test results, data are plotted with the results of the reference method on the x-axis and the results of the alternative method on the y-axis (figure 4).

Figure 4: Scatter plot of reference-method versus alternative-method results for the interlaboratory study (dotted line: first bisecting line, vertical lines: medians of the measurements obtained with the reference method)
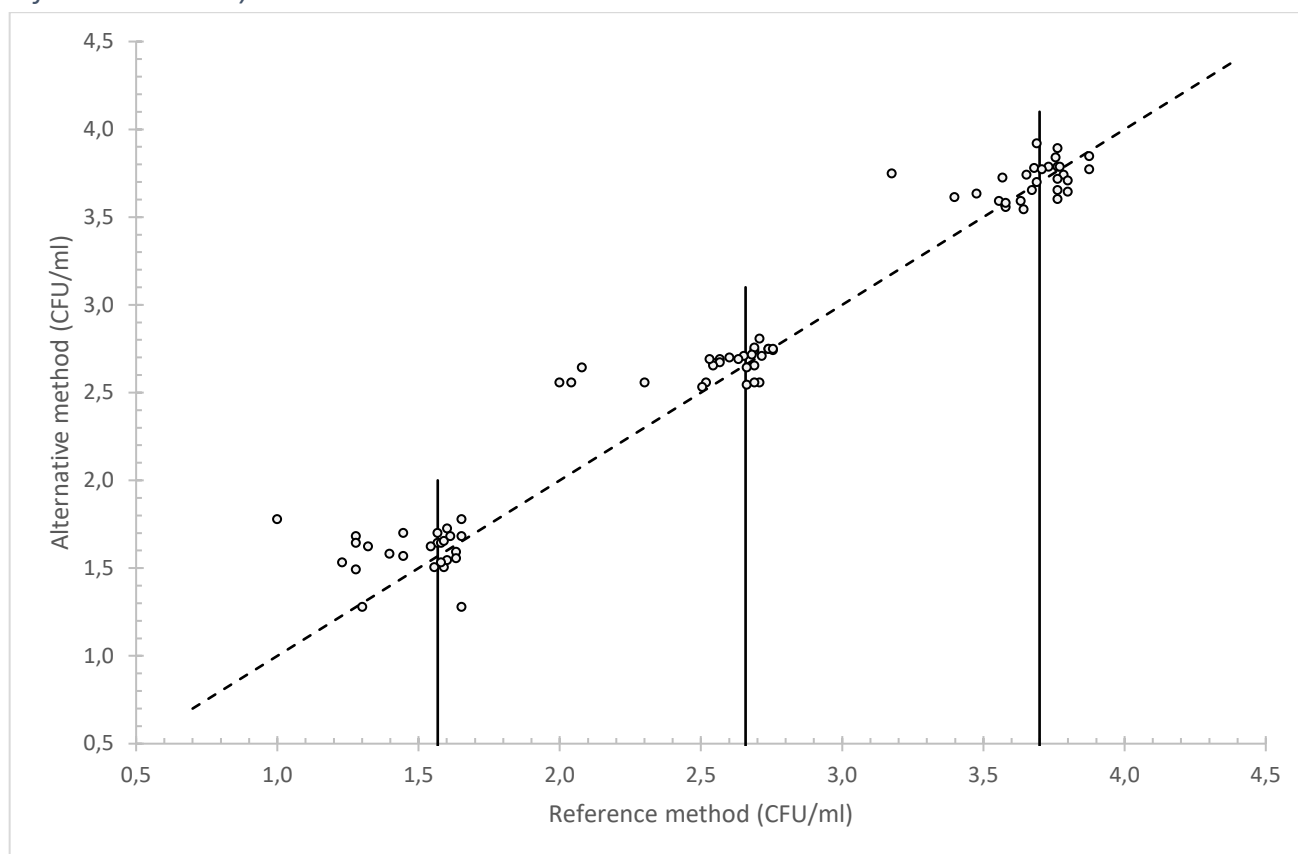
Data are well balanced around the median values of the reference method for each level, and a very slight positive bias is observed for the alternative method at all levels.

4.4.2. Calculation of the accuracy profile and interpretation

An accuracy profile is drawn according to the calculations provided in the Excel spreadsheet named [AP calculation tool_ILS_(clause_6-2-3_Calculations summary and interpretation of data)_ver 14-03-2016.xlsx](#), available at: http://standards.iso.org/iso/16140/-2/ed-1/en.
The results of the calculations are provided in table 10.
The graphical representation of the accuracy profile lies in figure 5.

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Table 10: Summary of the accuracy profile calculations (AL: acceptability limit, TI: tolerance interval, SD: standard deviation)

Tolerance probability (beta)	80%	80%	80%				
Acceptability limit in log (lambda)	0,50	0,50	0,50				
	Alternative method				**Reference method**		
Levels	**Low**	**Medium**	**High**		**Low**	**Medium**	**High**
Target value	**1,485**	**2,571**	**3,676**				
Number of participants (K)	13	14	14		13	14	14
Average for alternative method	1,597	2,662	3,711		1,485	2,571	3,676
Repeatability standard deviation (sr)	0,067	0,060	0,058		0,173	0,119	0,114
Between-labs standard deviation (sL)	0,106	0,055	0,085		0,000	0,179	0,097
Reproducibility standard deviation (sR)	0,125	0,081	0,103		0,173	0,215	0,150
Corrected number of dof	15,901	21,705	17,776		24,960	17,615	22,388
Coverage factor	1,381	1,356	1,370				
Interpolated Student t	1,337	1,322	1,331				
Tolerance interval standard deviation	0,1291	0,0830	0,1060				
Lower TI limit	1,424	2,552	3,570				
Upper TI limit	1,769	2,772	3,852				
Bias	**0,112**	**0,091**	**0,035**				
Relative Lower TI limit (beta = 80%)	**-0,061**	**-0,019**	**-0,106**				
Relative Upper TI limit (beta = 80%)	**0,285**	**0,201**	**0,176**				
Lower Acceptability Limit	**-0,50**	**-0,50**	**-0,50**				
Upper Acceptability Limit	**0,50**	**0,50**	**0,50**				
Pooled repro standard dev of reference	0,181						

Figure 5: Accuracy profile of the alternative method using β = 80% and λ = 0,50 log$_{10}$ unit

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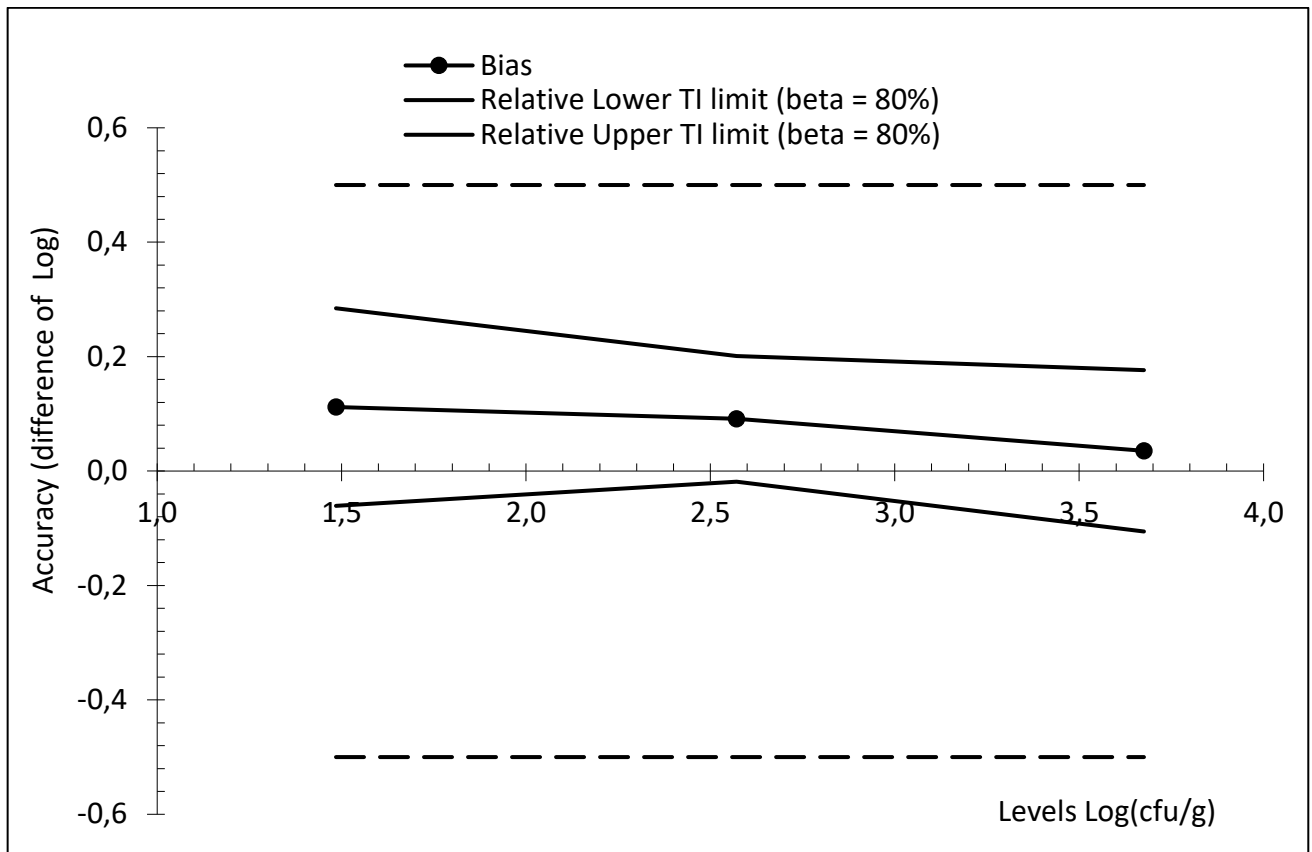
The AL is met for all the contamination levels: the tolerance interval limits of the alternative method are within the acceptability limits of 0.5 log CFU/g

4.5. General conclusion for the interlaboratory study

The tolerance intervals of all levels of contamination fall within the default acceptability limits (±0.5 log CFU/ml).
The alternative method is regarded as being equivalent to the reference method.

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5. General conclusion

The data and the interpretation of the methods comparison study and of the interlaboratory study fulfilled the requirements of the EN ISO 16140-2:2016 standard. The RAPID'*E. coli* 2 method is considered as equivalent to the reference method described in the ISO 16649-2:2001 standard.

Le Lion d'Angers, October 15, 2024.

Guillaume MESNARD
Technical deputy manager

François Le Nestour
Head of the Microbiology Department

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<u>APPENDICES</u>

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APPENDIX A

RAPID'*E. coli* 2 METHOD

TECHNICAL PROCEDURE

Preparation and dilution of the sample according to EN ISO 6887

1 ml in a sterile Petri dish
Pouring of about 15 ml of the molten RAPID'*E. coli* 2 agar medium
Mix the inoculum with the culture medium
Repeat this step with the following decimal dilutions

Incubation at 44±1°C for 21±3 h

Reading of the Petri dishes
Violet to pink colonies
Enumeration of the Petri dishes with less than 150 characteristic colonies

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APPENDIX B

ISO 16649-2
TECHNICAL PROCEDURE

Preparation and dilution of the sample according to EN ISO 6887

1 ml in a sterile Petri dish
Addition of the molten TBX agar medium (around 15 ml)
Repeat this step with the following decimal dilutions

Incubation at 44±1°C for 21±3 h
If the presence of stressed cells is suspected, incubation for an initial period of 4 h at 37°C
and then incubation at 44°C for 18 to 24 h

Reading
Blue colonies
Enumeration of the Petri dishes with less than 150 characteristic
colonies and less than 300 colonies in total

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APPENDIX C - Artificial contaminations

N° sample	Analysis date	Sample	Artificial contamination			
			Strain	Code	Origin	Injury protocol
61	Previous validation	Shrimp CA	/	/	/	/
66	Previous validation	Red Cabbage CA	/	/	/	/
73	Previous validation	Squid CA	/	/	/	/
74	Previous validation	Squid CA	/	/	/	/
90	Previous validation	Raw milk cheese (CA)	/	/	/	/
91	Previous validation	Smoked Salmon (CA)	/	/	/	/
92	Previous validation	Smoked Salmon (CA)	/	/	/	/
ISHA 14	ISHA 2017	Salmon Tagliatelle (CA)	*Escherichia coli*	ESC.1.33	Shrimp	Seeding : 4 °C/ 72h
ISHA 29-2	ISHA 2017	Cod Brandade (CA)	*Escherichia coli*	ESC.1.33	Shrimp	Seeding : 4 °C/ 72h
ISHA 31	ISHA 2017	Smoked Salmon (CA)	*Escherichia coli*	ESC.1.33	Shrimp	Seeding : 4 °C/ 72h
ISHA 32	ISHA 2017	Parmentier of duck (CA)	*Escherichia coli*	ESC.1.9	Ground beef 15 % fat	Seeding : 4 °C/ 72h
ISHA 33	ISHA 2017	"Croque monsieur" (CA)	*Escherichia coli*	ESC.1.6	Chicken ravioli	Seeding : 4 °C/ 72h
ISHA 34	ISHA 2017	Omelet (CA)	*Escherichia coli*	ESC.1.5	Camembert	Seeding : 4 °C/ 72h
ISHA 35	ISHA 2017	Smoked Chicken (CA)	*Escherichia coli*	ESC.1.9	Ground beef 15 % fat	Seeding : 4 °C/ 72h
ISHA 36	ISHA 2017	Smoked Veal (CA)	*Escherichia coli*	ESC.1.10	Marinated chicken	Seeding : 4 °C/ 72h
ISHA 37	ISHA 2017	Gratin (CA)	*Escherichia coli*	ESC.1.13	Stuffed croissant	Seeding : 4 °C/ 72h
ISHA 38	ISHA 2017	Smoked herring fillet (CA)	*Escherichia coli*	ESC.1.18	Smoked salmon rillette	Seeding : 4 °C/ 72h
ISHA 39	ISHA 2017	Smoked herring fillet (CA)	*Escherichia coli*	ESC.1.18	Smoked salmon rillette	Seeding : 4 °C/ 72h
ISHA 40	ISHA 2017	Egg Cream (CA)	*Escherichia coli*	ESC.1.5	Camembert	Seeding : 4 °C/ 72h
ISHA 42	ISHA 2017	Goat cheese (CA)	*Escherichia coli*	ESC.1.5	Camembert	Seeding : 4 °C/ 72h
ISHA 43	ISHA 2017	Emmental (CA)	*Escherichia coli*	ESC.1.5	Camembert	Seeding : 4 °C/ 72h
ISHA 44	ISHA 2017	Haddock fillet (CA)	*Escherichia coli*	ESC.1.18	Smoked salmon rillette	Seeding : 4 °C/ 72h
ISHA 45	ISHA 2017	Lingue fillet (CA)	*Escherichia coli*	ESC.1.18	Smoked salmon rillette	Seeding : 4 °C/ 72h
ISHA 46	ISHA 2017	Marinated mackerel (CA)	*Escherichia coli*	ESC.1.18	Smoked salmon rillette	Seeding : 4 °C/ 72h
ISHA 47	ISHA 2017	Smoked trout (CA)	*Escherichia coli*	ESC.1.18	Smoked salmon rillette	Seeding : 4 °C/ 72h
ISHA 48	ISHA 2017	Smoked Salmon (CA)	*Escherichia coli*	ESC.1.33	Shrimp	Seeding : 4 °C/ 72h
ISHA 49	ISHA 2017	Carpaccio (CA)	*Escherichia coli*	ESC.1.10	Marinated chicken	Seeding : 4 °C/ 72h
ISHA 50	ISHA 2017	Carpaccio (CA)	*Escherichia coli*	ESC.1.10	Marinated chicken	Seeding : 4 °C/ 72h
ISHA 51	ISHA 2017	Smoked mackerel (CA)	*Escherichia coli*	ESC.1.33	Shrimp	Seeding : 4 °C/ 72h
ISHA 52	ISHA 2017	Quiche Lorraine (CA)	*Escherichia coli*	ESC.1.6	Chicken ravioli	Seeding : 4 °C/ 72h
ISHA 54	ISHA 2017	Salad Mix (CA)	*Escherichia coli*	ESC.1.2	Grated carrot	Seeding : 4 °C/ 72h
ISHA 55	ISHA 2017	Grated carrots (CA)	*Escherichia coli*	ESC.1.2	Grated carrot	Seeding : 4 °C/ 72h
ISHA 56	ISHA 2017	Salad Bag (CA)	*Escherichia coli*	ESC.1.2	Grated carrot	Seeding : 4 °C/ 72h
ISHA 57	ISHA 2017	"Poêlée" of vegetables (CA)	*Escherichia coli*	ESC.1.2	Grated carrot	Seeding : 4 °C/ 72h
ISHA 58	ISHA 2017	Endive with Ham (CA)	*Escherichia coli*	ESC.1.6	Chicken ravioli	Seeding : 4 °C/ 72h
ISHA 59	ISHA 2017	Lasagna 3 meats (CA)	*Escherichia coli*	ESC.1.10	Marinated chicken	Seeding : 4 °C/ 72h
ISHA 60	ISHA 2017	Chicken Peas (CA)	*Escherichia coli*	ESC.1.13	Stuffed croissant	Seeding : 4 °C/ 72h
ISHA 61	ISHA 2017	Grilled Sausage (CA)	*Escherichia coli*	ESC.1.11	Chicken breast	Seeding : 4 °C/ 72h
ISHA 62	ISHA 2017	Parmentier of ham (CA)	*Escherichia coli*	ESC.1.11	Chicken breast	Seeding : 4 °C/ 72h
ISHA-63	ISHA 2017	Cod with sorrel sauce (CA)	*Escherichia coli*	ESC.1.31	Scallops	Seeding : 4 °C/ 72h
ISHA-64	ISHA 2017	Cod with tomato sauce (CA)	*Escherichia coli*	ESC.1.31	Scallops	Seeding : 4 °C/ 72h
ISHA-65	ISHA 2017	Salmon with sorrel sauce (CA)	*Escherichia coli*	ESC.1.31	Scallops	Seeding : 4 °C/ 72h
ISHA 66	ISHA 2017	Cucumber (CA)	*Escherichia coli*	ESC.1.2	Grated carrot	Seeding : 4 °C/ 72h
ISHA 67	ISHA 2017	Tomato (CA)	*Escherichia coli*	ESC.1.2	Grated carrot	Seeding : 4 °C/ 72h
ISHA 68	ISHA 2017	Apple (CA)	*Escherichia coli*	ESC.1.147	Cucumber	Seeding : 4 °C/ 72h
ISHA 69	ISHA 2017	Grape (CA)	*Escherichia coli*	ESC.1.147	Cucumber	Seeding : 4 °C/ 72h
ISHA 70	ISHA 2017	Red cabbage (CA)	*Escherichia coli*	ESC.1.147	Cucumber	Seeding : 4 °C/ 72h
ISHA 71	ISHA 2017	Basil Condiment (CA)	*Escherichia coli*	ESC.1.147	Cucumber	Seeding : 4 °C/ 72h
ISHA 71	ISHA 2017	Tabbouleh (CA)	*Escherichia coli*	ESC.1.6	Chicken ravioli	Seeding : 4 °C/ 72h
ISHA 72	ISHA 2017	Apple pie (CA)	*Escherichia coli*	ESC.1.17	Chicken ravioli	Seeding : 4 °C/ 72h
ISHA 73	ISHA 2017	Smoked breast of duck (CA)	*Escherichia coli*	ESC.1.17	Chicken ravioli	Seeding : 4 °C/ 72h
ISHA 74	ISHA 2017	Beef carpaccio (CA)	*Escherichia coli*	ESC.1.17	Chicken ravioli	Seeding : 4 °C/ 72h
ISHA 75	ISHA 2017	Beef marinated (CA)	*Escherichia coli*	ESC.1.17	Chicken ravioli	Seeding : 4 °C/ 72h

/: no information available

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Appendix D

Relative trueness study – Raw results

Caption:

Results are expressed in CFU/g or ml

ND: not determined

/ : information not available

    Estimated number

    Microorganisms are present but less than 40 per g or ml

    Results not countable or inferior to the limit of quantification

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Meat products

Type	#	Study	Sample	Reference method: NF ISO 16649-2				Alternative method: REC2 44°C		
				Dilution	CFU/ plate	CFU/ plate	Result (CFU/g or mL)	Dilution	CFU/plate	Result (CFU/g or mL)
a	1	IV	Meat offcuts	/ /	/ /	/ /	230	/ /	/ /	300
a	2	IVS	Ground beef	/ /	/ /	/ /	<10	/ /	/ /	<10
a	11	IVS	Leg of lamb	/ /	/ /	/ /	<10	/ /	/ /	<10
a	14	IVS	Turkey cutlet	/ /	/ /	/ /	130	/ /	/ /	150
a	26	IVS	Turkey steak	/ /	/ /	/ /	10	/ /	/ /	<10
a	27	IVS	Chicken fillet	/ /	/ /	/ /	10	/ /	/ /	20
a	28	IVS	Chicken fillet	/ /	/ /	/ /	5	/ /	/ /	<10
a	29	IVS	Beef kidneys	/ /	/ /	/ /	70	/ /	/ /	140
a	31	IVS	Sausage	/ /	/ /	/ /	30	/ /	/ /	40
a	32	IVS	Turkey cutlet	/ /	/ /	/ /	830	/ /	/ /	1100
a	33	IVS	Chopped steak	/ /	/ /	/ /	60	/ /	/ /	40
a	35	IVS	Veal cutlet	/ /	/ /	/ /	200	/ /	/ /	310
a	38	IVS	Veal cutlet	/ /	/ /	/ /	<10	/ /	/ /	<10
a	39	IVS	Turkey steak	/ /	/ /	/ /	15	/ /	/ /	30
a	64	IVS	Minced meat	/ /	/ /	/ /	ND	/ /	/ /	ND
a	65	IVS	Minced meat	/ /	/ /	/ /	ND	/ /	/ /	ND
a	82	IVS	Ground beef	/ /	/ /	/ /	ND	/ /	/ /	ND
a	83	IVS	Smoked ham	/ /	/ /	/ /	ND	/ /	/ /	ND
a	84	IVS	Horse Steak	/ /	/ /	/ /	ND	/ /	/ /	ND
a	ISHA 1	R2017	Groung beef	10 / 100	23 / 3	13 / 0	180	10 / 100	16 / 1	160
a	ISHA 25	R2017	Meatball	10 / 100	0 / 0	0 / 0	<10	10 / 100	0 / 0	<10
b	12	IVS	Blanquette of turkey	/ /	/ /	/ /	<10	/ /	/ /	<10
b	ISHA 2	R2017	Poultry bites	100 / 1000	30 / 3	29 / 1	2900	100 / 1000	38 / 5	3900
b	ISHA 30	R2017	Ravioli of poultry	100 / 1000	30 / 2	38 / 1	3200	100 / 1000	36 / 2	3500
b	ISHA 32	R2017	Parmentier of duck (CA)	10 / 100	34 / 5	38 / 5	370	10 / 100	37 / 6	390
b	ISHA 59	R2017	Lasagna 3 meats (CA)	100 / 1000	19 / 1	19 / 1	1800	100 / 1000	22 / 4	2400
b	ISHA 61	R2017	Grilled Sausage (CA)	100 / 1000	14 / 2	12 / 2	1400	100 / 1000	12 / 4	1500
b	ISHA 62	R2017	Parmentier of ham (CA)	100 / 1000	17 / 1	11 / 1	1400	100 / 1000	18 / 1	1700

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Meat products

Type	#	Study	Sample	Reference method: NF ISO 16649-2				Alternative method: REC2 44°C		
				Dilution	CFU/ plate	CFU/ plate	Result (CFU/g or mL)	Dilution	CFU/plate	Result (CFU/g or mL)
c	34	IVS	Chipolata	/	/	/	960	/	/	1300
				/	/	/		/	/	
c	3	IVS	Merguez	/	/	/	120	/	/	120
				/	/	/		/	/	
c	4	IVS	Veal Sausage	/	/	/	150	/	/	220
				/	/	/		/	/	
c	6	IVS	Herb Sausage	/	/	/	380	/	/	360
				/	/	/		/	/	
c	13	IVS	Merguez	/	/	/	15	/	/	10
				/	/	/		/	/	
c	ISHA 35	R2017	Smoked chicken (CA)	100	16	16	1600	100	14	1500
				1000	3	0		1000	3	
c	ISHA 36	R2017	Smoked veal (CA)	1000	0	0	<1000	1000	1	<1000
				10000	0	0		10000	0	

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Dairy and egg products

Type	#	Study	Sample	Reference method: NF ISO 16649-2				Alternative method: REC2 44°C		
				Dilution	CFU/ plate	CFU/ plate	Result (CFU/g or mL)	Dilution	CFU/plate	Result (CFU/g or mL)
a	L3	IVS	Nougat ice cream	/ /	/ /	/ /	<1	/ /	/ /	<1
a	L4	IVS	Chocolate ice cream	/ /	/ /	/ /	<1	/ /	/ /	<1
a	ISHA 18	R2017	Pasteurized milk brie	10 / 100	23 / 2	40 / 2	310	10 / 100	30 / 3	300
a	ISHA 19	R2017	Pasteurized milk brie	10 / 100	23 / 1	17 / 1	190	10 / 100	21 / 0	190
a	ISHA 20	R2017	Pasteurized milk brie	10 / 100	25 / 5	31 / 7	310	10 / 100	40 / 3	390
a	ISHA 42	R2017	Goat cheese (CA)	10 / 100	18 / 1	20 / 2	190	10 / 100	20 / 3	210
a	ISHA 43	R2017	Emmental (CA)	10 / 100	17 / 1	22 / 1	190	10 / 100	17 / 1	160
a	ISHA 41	R2017	Fresh cream	10 / 100	0 / 0	0 / 0	<10	10 / 100	0 / 0	<10
b	L1	IVS	Raw milk	/ /	/ /	/ /	<1	/ /	/ /	<1
b	L2	IVS	Raw milk	/ /	/ /	/ /	<1	/ /	/ /	<1
b	10	IVS	Reblochon	/ /	/ /	/ /	870	/ /	/ /	1300
b	24	IVS	Reblochon	/ /	/ /	/ /	40	/ /	/ /	30
b	25	IVS	Roquefort	/ /	/ /	/ /	210	/ /	/ /	790
b	36	IVS	Raw milk cheese	/ /	/ /	/ /	20000	/ /	/ /	16000
b	59	IVS	Raw milk cheese	/ /	/ /	/ /	2800	/ /	/ /	2500
b	78	IVS	Reblochon	/ /	/ /	/ /	6500	/ /	/ /	4900
b	90	IVS	Raw milk cheese (CA)	/ /	/ /	/ /	1600	/ /	/ /	1800
c	44	IVS	Strawberry cream	/ /	/ /	/ /	<10	/ /	/ /	<10
c	96	IVS	Chocolate cake	/ /	/ /	/ /	12000	/ /	/ /	11000
c	95	IVS	Versailles	/ /	/ /	/ /	23000	/ /	/ /	28000
c	93	IVS	Cream puffs	/ /	/ /	/ /	180	/ /	/ /	290
c	70	IVS	Coffee éclair	/ /	/ /	/ /	140	/ /	/ /	160
c	40	IVS	Banana cream	/ /	/ /	/ /	70	/ /	/ /	70
c	20	IVS	Coffee éclair	/ /	/ /	/ /	90	/ /	/ /	30
c	72	IVS	Peach cake	/ /	/ /	/ /	4000	/ /	/ /	1700
c	94	IVS	Apple pie	/ /	/ /	/ /	1770	/ /	/ /	2400
c	60	IVS	Apricot cake	/ /	/ /	/ /	290	/ /	/ /	360
c	ISHA 21	R2017	Almond cake	10 / 100	158 / 18	126 / 9	1400	10 / 100	116 / 14	1200
c	ISHA 40	R2017	Egg cream (CA)	10 / 100	10 / 3	15 / 1	130	10 / 100	21 / 2	210

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Seafood products

Type	#	Study	Sample	Reference method: NF ISO 16649-2				Alternative method: REC2 44°C		
				Dilution	CFU/ plate	CFU/ plate	Result (CFU/g or mL)	Dilution	CFU/plate	Result (CFU/g or mL)
a	7	IVS	Fish	/ /	/ /	/ /	<10	/ /	/ /	ND
a	8	IVS	Fish	/ /	/ /	/ /	<10	/ /	/ /	<10
a	9	IVS	Fish	/ /	/ /	/ /	<10	/ /	/ /	<10
a	22	IVS	Tuna	/ /	/ /	/ /	<10	/ /	/ /	<10
a	52	IVS	Fillet of Red mullet	/ /	/ /	/ /	22000	/ /	/ /	25000
a	53	IVS	Salmon Steak	/ /	/ /	/ /	29000	/ /	/ /	22000
a	54	IVS	Fish	/ /	/ /	/ /	290000	/ /	/ /	250000
a	80	IVS	Cod fillet	/ /	/ /	/ /	ND	/ /	/ /	ND
a	ISHA 44	R2017	Haddock fillet (CA)	1000 / 10000	7 / 1	6 / 1	6500	1000 / 10000	8 / 3	8000
a	ISHA 45	R2017	Lingue fillet (CA)	1000 / 10000	6 / 1	5 / 2	5500	1000 / 10000	5 / 1	5000
a	ISHA 7	R2017	Julienne fillet	10 / 100	0 / 0	0 / 0	<10	10 / 100	0 / 0	<10
a	ISHA 10	R2017	Raw Salmon	10 / 100	0 / 0	0 / 0	<10	10 / 100	0 / 0	<10
a	ISHA 11	R2017	Raw Salmon	10 / 100	0 / 0	0 / 0	<10	10 / 100	0 / 0	<10
a	ISHA 27	R2017	Petoncle Nuts	10 / 100	0 / 0	0 / 0	<10	10 / 100	0 / 0	<10
a	ISHA 29-1	R2017	Sole fillet	10 / 100	0 / 0	0 / 0	<10	10 / 100	0 / 0	<10
b	81	IVS	Smoked Marlin Fillet	/ /	/ /	/ /	<10	/ /	/ /	<10
b	55	IVS	Smoked salmon	/ /	/ /	/ /	240000	/ /	/ /	180000
b	91	IVS	Smoked Salmon (CA)	/ /	/ /	/ /	260	/ /	/ /	160
b	92	IVS	Smoked Salmon (CA)	/ /	/ /	/ /	1900	/ /	/ /	2600
b	ISHA 17	R2017	Marinated salmon	10 / 100	0 / 0	0 / 0	<10	10 / 100	0 / 0	<10
b	ISHA 31	R2017	Smoked Salmon (CA)	10 / 100	8 / 1	5 / 2	65	10 / 100	15 / 1	150
b	ISHA 38	R2017	Smoked herring fillet (CA)	10 / 100	9 / 0	6 / 0	75	10 / 100	11 / 1	110
b	ISHA 39	R2017	Smoked herring fillet (CA)	10 / 100	10 / 1	5 / 0	70	10 / 100	5 / 0	50
b	ISHA 47	R2017	Smoked trout (CA)	10 / 100	23 / 2	35 / 1	280	10 / 100	31 / 5	330
b	ISHA 48	R2017	Smoked Salmon (CA)	10 / 100	26 / 4	24 / 2	260	10 / 100	30 / 2	290
b	ISHA 51	R2017	Smoked mackerel (CA)	10 / 100	46 / 6	61 / 4	530	10 / 100	57 / 3	550
b	ISHA 46	R2017	Marinated mackerel (CA)	1000 / 10000	21 / 2	18 / 1	19000	1000 / 10000	15 / 2	16000
c	43	IVS	Cod fillet	/ /	/ /	/ /	<10	/ /	/ /	<10
c	61	IVS	Shrimp (CA)	/ /	/ /	/ /	2300	/ /	/ /	1800

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Seafood products

Type	#	Study	Sample	Reference method: NF ISO 16649-2				Alternative method: REC2 44°C		
				Dilution	CFU/ plate	CFU/ plate	Result (CFU/g or mL)	Dilution	CFU/plate	Result (CFU/g or mL)
c	63	IVS	Shrimp	/	/	/	ND	/	/	ND
				/	/	/		/	/	
c	79	IVS	Smoked salmon	/	/	/	ND	/	/	ND
				/	/	/		/	/	
c	ISHA 29-2	R2017	Cod Brandade (CA)	10	38	56	460	10	58	600
				100	3	5		100	8	
c	ISHA-63	R2017	Cod with sorrel sauce (CA)	10	8	5	65	10	7	70
				100	0	0		100	0	
c	ISHA-64	R2017	Cod with tomato sauce (CA)	10	7	8	75	10	6	60
				100	0	0		100	0	
c	ISHA-65	R2017	Salmon with sorrel sauce (CA)	10	6	11	85	10	7	70
				100	0	0		100	0	
c	ISHA 28	IVS	Cod Accras	10	0	0	<10	10	0	<10
				100	0	0		100	0	

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Vegetal products

Type	#	Study	Sample	Reference method: NF ISO 16649-2				Alternative method: REC2 44°C		
				Dilution	CFU/ plate	CFU/ plate	Result (CFU/g or mL)	Dilution	CFU/plate	Result (CFU/g or mL)
a	ISHA 66	R2017	Cucumber (CA)	100	42	38	4200	100	48	4600
				1000	5	7		1000	3	
a	ISHA 67	R2017	Tomato (CA)	100	87	64	7700	100	87	8700
				1000	9	8		1000	9	
a	ISHA 68	R2017	Apple (CA)	100	18	13	1400	100	12	1300
				1000	0	0		1000	2	
a	ISHA 69	R2017	Grape (CA)	10	11	12	110	10	13	140
				100	0	1		100	2	
a	ISHA 70	R2017	Red cabbage (CA)	10	39	45	430	10	48	480
				100	5	6		100	5	
b	89	IVS	Grated carrots	/	/	/	1600	/	/	1700
				/	/	/		/	/	
b	49	IVS	Grated carrots	/	/	/	<10	/	/	<10
				/	/	/		/	/	
b	ISHA 8	R2017	Grated carrots	10000000	0	0	<10 000 000	10000000	0	<10 000 000
				100000000	0	0		100000000	0	
b	ISHA 15	R2017	Carrots in bags	10	1	0	5	10	2	20
				100	0	0		100	0	
b	ISHA 22	R2017	Rocket in Bag	10	0	0	<10	10	0	<10
				100	0	0		100	0	
b	ISHA 23	R2017	Grated carrots	10	0	0	<10	10	0	<10
				100	0	0		100	0	
b	ISHA 54	R2017	Salad Mix (CA)	10	24	38	340	10	32	350
				100	6	7		100	6	
b	ISHA 55	R2017	Grated carrots (CA)	10	41	28	350	10	30	280
				100	4	4		100	1	
b	ISHA 56	R2017	Salad Bag (CA)	10	26	31	280	10	43	440
				100	2	2		100	5	
b	ISHA 57	R2017	"Poêlée" of vegetables (CA)	10	36	43	390	10	44	490
				100	4	3		100	10	
c	50	IVS	Macedonia	/	/	/	65	/	/	45
				/	/	/		/	/	
c	56	IVS	Fruit Compote	/	/	/	660	/	/	560
				/	/	/		/	/	
c	ISHA 12	R2017	Basil Condiment	10	26	11	180	10	23	240
				100	1	1		100	3	
c	ISHA 71	R2017	Basil Condiment (CA)	100	93	89	9200	100	84	8400
				1000	11	10		1000	8	
c	ISHA 6	R2017	Parsnip Purée	10	15	13	130	10	5	50
				100	1	0		100	0	
c	ISHA 4	R2017	Coco Pearl	10	0	0	<10	10	0	<10
				100	0	0		100	0	
c	ISHA 5	R2017	Creamy Cabbage Flowers	10	0	0	<10	10	0	<10
				100	0	0		100	0	
c	ISHA 9	R2017	Homemade cucumbers	100000	0	0	<10	1000	0	<10
				1000000	0	0		10000	0	

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RTE-RTRH products

Type	#	Study	Sample	Reference method: NF ISO 16649-2				Alternative method: REC2 44°C		
				Dilution	CFU/ plate	CFU/ plate	Result (CFU/g or mL)	Dilution	CFU/plate	Result (CFU/g or mL)
a	15	IVS	Tabbouleh	/	/	/	<10	/	/	<10
				/	/	/		/	/	
a	17	IVS	Savarin with chocolate	/	/	/	5	/	/	<10
				/	/	/		/	/	
a	18	IVS	"Eclair" with pastry cream	/	/	/	<10	/	/	<10
				/	/	/		/	/	
a	19	IVS	Pastas salad	/	/	/	5	/	/	<10
				/	/	/		/	/	
a	23	IVS	Chocolate Cream	/	/	/	<10	/	/	<10
				/	/	/		/	/	
a	46	IVS	Cream puffs	/	/	/	<10	/	/	<10
				/	/	/		/	/	
a	51	IVS	Apple pie	/	/	/	<10	/	/	<10
				/	/	/		/	/	
a	57	IVS	Strawberry	/	/	/	30	/	/	70
				/	/	/		/	/	
a	75	IVS	Cream puffs	/	/	/	<10	/	/	<10
				/	/	/		/	/	
a	76	IVS	Cream puffs	/	/	/	<10	/	/	<10
				/	/	/		/	/	
a	71	IVS	Tuna tomatoes	/	/	/	140	/	/	200
				/	/	/		/	/	
a	ISHA 3	R2017	Gabonese salad	10	6	10	80	10	5	50
				100	0	1		100	0	
a	ISHA 71	R2017	Tabbouleh (CA)	10	56	61	570	10	52	520
				100	5	4		100	5	
a	ISHA 72	R2017	Apple pie (CA)	10	26	31	290	10	27	270
				100	4	2		100	3	
a	ISHA 13	R2017	Tomato salad dressing	10	29	30	280	10	34	330
				100	2	1		100	2	
a	21	IVS	Tomato salad	/	/	/	<10	/	/	<10
				/	/	/		/	/	
b	ISHA 33	R2017	"Croque monsieur" (CA)	100	11	8	910	100	14	1400
				1000	1	0		1000	1	
b	ISHA 52	R2017	Quiche Lorraine (CA)	10	17	29	240	10	20	250
				100	5	2		100	7	
b	ISHA 60	R2017	Chicken Peas (CA)	100	12	13	1400	100	22	2400
				1000	2	3		1000	4	
b	ISHA 58	R2017	Endive with Ham (CA)	100	21	14	1700	100	13	1300
				1000	1	1		1000	1	
b	ISHA 53	R2017	Beef vegetables	10	24	27	250	10	26	260
				100	1	3		100	3	
b	ISHA 34	R2017	Omelet (CA)	10	2	1	20	10	3	30
				100	0	0		100	0	
b	ISHA 37	R2017	Gratin (CA)	10	1	0	10	10	0	<10
				100	1	0		100	0	
b	ISHA 14	R2017	Salmon Tagliatelle (CA)	10	1	0	10	10	1	10
				100	0	0		100	0	
b	ISHA 24	R2017	Royal noodles	10	0	0	<10	10	0	<10
				100	0	0		100	0	
c	ISHA 49	R2017	Carpaccio (CA)	10	16	19	190	10	28	260
				100	4	2		100	1	
c	ISHA 50	R2017	Carpaccio (CA)	10	22	23	220	10	18	190
				100	1	2		100	3	
c	ISHA 26	R2017	Carpaccio	10	0	0	<10	10	0	<10
				100	0	0		100	0	
c	ISHA 73	R2017	Smoked breast of duck (CA)	100	123	118	13000	100	120	12000
				1000	18	21		1000	12	
c	ISHA 74	R2017	Beef carpaccio (CA)	100	97	104	10000	100	100	10000
				1000	12	9		1000	11	
c	ISHA 75	R2017	Beef marinated (CA)	100	110	103	11000	100	102	10000
				1000	9	9		1000	10	

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Appendix E

Relative trueness study – Statistical calculations

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Appendix E

	Type	#	Study	Sample	RM CFU/g	AM CFU/g	RM log CFU/g	AM log CFU/g	Mean	Difference
Meat products	a	1	IV	Meat offcuts	230	300	2,36	2,48	2,42	0,12
	a	14	IVS	Turkey cutlet	130	150	2,11	2,18	2,15	0,06
	a	29	IVS	Beef kidneys	70	140	1,85	2,15	2,00	0,30
	a	32	IVS	Turkey cutlet	830	1100	2,92	3,04	2,98	0,12
	a	35	IVS	Veal cutlet	200	310	2,30	2,49	2,40	0,19
	a	ISHA 1	R2017	Groung beef	180	160	2,26	2,20	2,23	-0,05
	a	33	IVS	Chopped steak	60	40	1,78	1,60	1,69	-0,18
	b	ISHA 2	R2017	Poultry bites	2900	3900	3,46	3,59	3,53	0,13
	b	ISHA 30	R2017	Ravioli of poultry	3200	3500	3,51	3,54	3,52	0,04
	b	ISHA 32	R2017	Parmentier of duck (CA)	370	390	2,57	2,59	2,58	0,02
	b	ISHA 59	R2017	Lasagna 3 meats (CA)	1800	2400	3,26	3,38	3,32	0,12
	b	ISHA 61	R2017	Grilled Sausage (CA)	1400	1500	3,15	3,18	3,16	0,03
	b	ISHA 62	R2017	Parmentier of ham (CA)	1400	1700	3,15	3,23	3,19	0,08
	c	34	IVS	Chipolata	960	1300	2,98	3,11	3,05	0,13
	c	3	IVS	Merguez	120	120	2,08	2,08	2,08	0,00
	c	4	IVS	Veal Sausage	150	220	2,18	2,34	2,26	0,17
	c	6	IVS	Herb Sausage	380	360	2,58	2,56	2,57	-0,02
	c	ISHA 35	R2017	Smoked chicken (CA)	1600	1500	3,20	3,18	3,19	-0,03
	Average difference of the category									0,07
	Standard deviation of differences									0,11

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Appendix E

	Type	#	Study	Sample	RM CFU/g	AM CFU/g	RM log CFU/g	AM log CFU/g	Mean	Difference
Dairy and egg products	a	ISHA 18	R2017	Pasteurized milk brie	310	300	2,49	2,48	2,48	-0,01
	a	ISHA 19	R2017	Pasteurized milk brie	190	190	2,28	2,28	2,28	0,00
	a	ISHA 20	R2017	Pasteurized milk brie	310	390	2,49	2,59	2,54	0,10
	a	ISHA 42	R2017	Goat cheese (CA)	190	210	2,28	2,32	2,30	0,04
	a	ISHA 43	R2017	Emmental (CA)	190	160	2,28	2,20	2,24	-0,07
	b	10	IVS	Reblochon	870	1300	2,94	3,11	3,03	0,17
	b	25	IVS	Roquefort	210	790	2,32	2,90	2,61	0,58
	b	36	IVS	Raw milk cheese	20000	16000	4,30	4,20	4,25	-0,10
	b	59	IVS	Raw milk cheese	2800	2500	3,45	3,40	3,42	-0,05
	b	78	IVS	Reblochon	6500	4900	3,81	3,69	3,75	-0,12
	b	90	IVS	Raw milk cheese (CA)	1600	1800	3,20	3,26	3,23	0,05
	c	96	IVS	Chocolate cake	12000	11000	4,08	4,04	4,06	-0,04
	c	95	IVS	Versailles	23000	28000	4,36	4,45	4,40	0,09
	c	93	IVS	Cream puffs	180	290	2,26	2,46	2,36	0,21
	c	70	IVS	Coffee éclair	140	160	2,15	2,20	2,18	0,06
	c	40	IVS	Banana cream	70	70	1,85	1,85	1,85	0,00
	c	72	IVS	Peach cake	4000	1700	3,60	3,23	3,42	-0,37
	c	94	IVS	Apple pie	1770	2400	3,25	3,38	3,31	0,13
	c	60	IVS	Apricot cake	290	360	2,46	2,56	2,51	0,09
	c	ISHA 21	R2017	Almond cake	1400	1200	3,15	3,08	3,11	-0,07
	c	ISHA 40	R2017	Egg cream (CA)	130	210	2,11	2,32	2,22	0,21
	Average difference of the category									0,04
	Standard deviation of differences									0,18

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Appendix E

	Type	#	Study	Sample	RM CFU/g	AM CFU/g	RM log CFU/g	AM log CFU/g	Mean	Difference
Seafood products	a	52	IVS	Fillet of Red mullet	22000	25000	4,34	4,40	4,37	0,06
	a	53	IVS	Salmon Steak	29000	22000	4,46	4,34	4,40	-0,12
	a	54	IVS	Fish	290000	250000	5,46	5,40	5,43	-0,06
	a	ISHA 44	R2017	Haddock fillet (CA)	6500	8000	3,81	3,90	3,86	0,09
	a	ISHA 45	R2017	Lingue fillet (CA)	5500	5000	3,74	3,70	3,72	-0,04
	b	55	IVS	Smoked salmon	240000	180000	5,38	5,26	5,32	-0,12
	b	91	IVS	Smoked Salmon (CA)	260	160	2,41	2,20	2,31	-0,21
	b	92	IVS	Smoked Salmon (CA)	1900	2600	3,28	3,41	3,35	0,14
	b	ISHA 31	R2017	Smoked Salmon (CA)	65	150	1,81	2,18	1,99	0,36
	b	ISHA 38	R2017	Smoked herring fillet (CA)	75	110	1,88	2,04	1,96	0,17
	b	ISHA 39	R2017	Smoked herring fillet (CA)	70	50	1,85	1,70	1,77	-0,15
	b	ISHA 47	R2017	Smoked trout (CA)	280	330	2,45	2,52	2,48	0,07
	b	ISHA 48	R2017	Smoked Salmon (CA)	260	290	2,41	2,46	2,44	0,05
	b	ISHA 51	R2017	Smoked mackerel (CA)	530	550	2,72	2,74	2,73	0,02
	b	ISHA 46	R2017	Marinated mackerel (CA)	19000	16000	4,28	4,20	4,24	-0,07
	c	61	IVS	Shrimp (CA)	2300	1800	3,36	3,26	3,31	-0,11
	c	ISHA 29-2	R2017	Cod Brandade (CA)	460	600	2,66	2,78	2,72	0,12
	c	ISHA-63	R2017	Cod with sorrel sauce (CA)	65	70	1,81	1,85	1,83	0,03
	c	ISHA-64	R2017	Cod with tomato sauce (CA)	75	60	1,88	1,78	1,83	-0,10
	c	ISHA-65	R2017	Salmon with sorrel sauce (CA)	85	70	1,93	1,85	1,89	-0,08
	Average difference of the category									0,00
	Standard deviation of differences									0,14

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Appendix E

	Type	#	Study	Sample	RM CFU/g	AM CFU/g	RM log CFU/g	AM log CFU/g	Mean	Difference
Vegetal products	a	ISHA 66	R2017	Cucumber (CA)	4200	4600	3,62	3,66	3,64	0,04
	a	ISHA 67	R2017	Tomato (CA)	7700	8700	3,89	3,94	3,91	0,05
	a	ISHA 68	R2017	Apple (CA)	1400	1300	3,15	3,11	3,13	-0,03
	a	ISHA 69	R2017	Grape (CA)	110	140	2,04	2,15	2,09	0,10
	a	ISHA 70	R2017	Red cabbage (CA)	430	480	2,63	2,68	2,66	0,05
	b	89	IVS	Grated carrots	1600	1700	3,20	3,23	3,22	0,03
	b	ISHA 54	R2017	Salad Mix (CA)	340	350	2,53	2,54	2,54	0,01
	b	ISHA 55	R2017	Grated carrots (CA)	350	280	2,54	2,45	2,50	-0,10
	b	ISHA 56	R2017	Salad Bag (CA)	280	440	2,45	2,64	2,55	0,20
	b	ISHA 57	R2017	"Poêlée" of vegetables (CA)	390	490	2,59	2,69	2,64	0,10
	c	50	IVS	Macedonia	65	45	1,81	1,65	1,73	-0,16
	c	56	IVS	Fruit Compote	660	560	2,82	2,75	2,78	-0,07
	c	ISHA 12	R2017	Basil Condiment	180	240	2,26	2,38	2,32	0,12
	c	ISHA 71	R2017	Basil Condiment (CA)	9200	8400	3,96	3,92	3,94	-0,04
	c	ISHA 6	R2017	Parsnip Purée	130	50	2,11	1,70	1,91	-0,41
	Average difference of the category									-0,01
	Standard deviation of differences									0,15

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Appendix E

	Type	#	Study	Sample	RM	AM	RM	AM	Mean	Difference
					CFU/g	CFU/g	log CFU/g	log CFU/g		
Ready-to-eat and ready-to-reheat products	a	71	IVS	Tuna tomatoes	140	200	2,15	2,30	2,22	0,15
	a	ISHA 3	R2017	Gabonese salad	80	50	1,90	1,70	1,80	-0,20
	a	ISHA 71	R2017	Tabbouleh (CA)	570	520	2,76	2,72	2,74	-0,04
	a	ISHA 72	R2017	Apple pie (CA)	290	270	2,46	2,43	2,45	-0,03
	a	ISHA 13	R2017	Tomato salad dressing	280	330	2,45	2,52	2,48	0,07
	b	ISHA 33	R2017	"Croque monsieur" (CA)	910	1400	2,96	3,15	3,05	0,19
	b	ISHA 52	R2017	Quiche Lorraine (CA)	240	250	2,38	2,40	2,39	0,02
	b	ISHA 60	R2017	Chicken Peas (CA)	1400	2400	3,15	3,38	3,26	0,23
	b	ISHA 58	R2017	Endive with Ham (CA)	1700	1300	3,23	3,11	3,17	-0,12
	b	ISHA 53	R2017	Beef vegetables	250	260	2,40	2,41	2,41	0,02
	c	ISHA 49	R2017	Carpaccio (CA)	190	260	2,28	2,41	2,35	0,14
	c	ISHA 50	R2017	Carpaccio (CA)	220	190	2,34	2,28	2,31	-0,06
	c	ISHA 73	R2017	Smoked breast of duck (CA)	13000	12000	4,11	4,08	4,10	-0,03
	c	ISHA 74	R2017	Beef carpaccio (CA)	10000	10000	4,00	4,00	4,00	0,00
	c	ISHA 75	R2017	Beef marinated (CA)	11000	10000	4,04	4,00	4,02	-0,04
	Average difference of the category									0,02
	Standard deviation of differences									0,12

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Appendix E

Average difference of all categories	0,03
Standard deviation of differences	0,14

n =	89		
β =	0,95	**Lower confidence limit**	**Upper confidence limit**
T(0.025;97)=	1,99	-0,25	0,31

Results not used in the statistical interpretation

Category	Type	#	Study	Sample	RM CFU/g	AM CFU/g	RM log CFU/g	AM log CFU/g	Mean	Difference
Meat products	a	26	IVS	Turkey steak	10	0	1,00	0,00	0,50	-1,00
	a	27	IVS	Chicken fillet	10	20	1,00	1,30	1,15	0,30
	a	28	IVS	Chicken fillet	5	0	0,70	0,00	0,35	-0,70
	a	31	IVS	Sausage	30	40	1,48	1,60	1,54	0,12
	a	39	IVS	Turkey steak	15	30	1,18	1,48	1,33	0,30
	c	13	IVS	Merguez	15	10	1,18	1,00	1,09	-0,18
Dairy and egg products	b	24	IVS	Reblochon	40	30	1,60	1,48	1,54	-0,12
	c	20	IVS	Coffee éclair	90	30	1,95	1,48	1,72	-0,48
Vegetal products	b	ISHA 15	R2017	Carrots in bags	5	20	0,70	1,30	1,00	0,60
Composite foods	a	17	IVS	Savarin with chocolate	5	0	0,70	0,00	0,35	-0,70
	a	19	IVS	Pastas salad	5	0	0,70	0,00	0,35	-0,70
	a	57	IVS	Strawberry	30	70	1,48	1,85	1,66	0,37
	b	ISHA 34	R2017	Omelet (CA)	20	30	1,30	1,48	1,39	0,18
	b	ISHA 37	R2017	Gratin (CA)	10	0	1,00	0,00	0,50	-1,00
	b	ISHA 14	R2017	Salmon Tagliatelle (CA)	10	10	1,00	1,00	1,00	0,00

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Appendix F

Accuracy profiles – Raw results

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Meat products

Matrix	Strain	Level	Sample number	Reference method 16649-2						Alternative method REC2 44°C			
				Replicate 1		Replicate 2		CFU/g	log CFU/g	Dilution	CFU / Petri dish	CFU/g	log CFU/g
				Dilution	CFU / Petri dish	Dilution	CFU / Petri dish						
Meat - Batch 1 - TVC result: 4,0x10^3 CFU/g	ESC.1.15	300 CFU/g	L1T1R1	-1	83	-1	50	660	2,820	-1	52	500	2,699
				-2	9	-2	3			-2	3		
			L1T1R2	-1	41	-1	52	460	2,663	-1	61	600	2,778
				-2	3	-2	5			-2	5		
			L1T1R3	-1	44	-1	35	400	2,602	-1	53	630	2,799
				-2	4	-2	4			-2	16		
			L1T1R4	-1	43	-1	62	510	2,708	-1	60	620	2,792
				-2	4	-2	4			-2	8		
			L1T1R5	-1	45	-1	51	460	2,663	-1	48	470	2,672
				-2	3	-2	3			-2	4		
		3000 CFU/g	L1T2R1	-1	118	-1	96	1200	3,079	-1	103	1 100	3,041
				-2	20	-2	19			-2	16		
			L1T2R2	-1	90	-1	98	960	2,982	-1	82	910	2,959
				-2	11	-2	12			-2	18		
			L1T2R3	-1	110	-1	100	1100	3,041	-1	120	1 200	3,079
				-2	18	-2	17			-2	8		
			L1T2R4	-1	132	-1	100	1200	3,079	-1	118	1 200	3,079
				-2	14	-2	10			-2	10		
			L1T2R5	-1	114	-1	120	1200	3,079	-1	112	1 200	3,079
				-2	8	-2	18			-2	20		
		100000 CFU/g	L1T3R1	-3	110	-3	111	110000	5,041	-3	149	150000	5,176
				-4	10	-4	14			-4	18		
			L1T3R2	-3	102	-3	80	94000	4,973	-3	129	130000	5,114
				-4	16	-4	8			-4	14		
			L1T3R3	-3	125	-3	139	130000	5,114	-3	130	130000	5,114
				-4	11	-4	13			-4	16		
			L1T3R4	-3	156	-3	160	160000	5,204	-3	133	140000	5,146
				-4	18	-4	13			-4	22		
			L1T3R5	-3	145	-3	140	140000	5,146	-3	133	130000	5,114
				-4	15	-4	14			-4	13		

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Meat products

Matrix	Strain	Level	Sample number	Reference method 16649-2						Alternative method REC2 44°C			
				Replicate 1		Replicate 2		CFU/g	log CFU/g	Dilution	CFU / Petri dish	CFU/g	log CFU/g
				Dilution	CFU / Petri dish	Dilution	CFU / Petri dish						
Meat - Batch 2 - TVC result: 2,0x10³ CFU/g	ESC.1.15	300 CFU/g	L1T1R1	-1	14	-1	16	150	2,176	-1	19	190	2,279
				-2	1	-2	1			-2	2		
			L1T1R2	-1	17	-1	13	150	2,176	-1	19	200	2,301
				-2	1	-2	2			-2	3		
			L1T1R3	-1	15	-1	15	160	2,204	-1	18	190	2,279
				-2	3	-2	2			-2	3		
			L1T1R4	-1	15	-1	8	100	2,000	-1	19	190	2,279
				-2	0	-2	0			-2	2		
			L1T1R5	-1	22	-1	17	200	2,301	-1	14	140	2,146
				-2	0	-2	4			-2	1		
		3000 CFU/g	L1T2R1	-2	16	-2	18	1700	3,230	-2	12	1 100	3,041
				-3	2	-3	2			-3	0		
			L1T2R2	-2	17	-2	17	1600	3,204	-2	19	2 000	3,301
				-3	0	-3	1			-3	3		
			L1T2R3	-2	18	-2	19	1800	3,255	-2	20	1 400	3,146
				-3	1	-3	1			-3	6		
			L1T2R4	-2	13	-2	14	1300	3,114	-2	30	2 700	3,431
				-3	2	-3	0			-3	0		
			L1T2R5	-2	7	-2	14	1000	3,000	-2	18	1 900	3,279
				-3	0	-3	2			-3	3		
		100000 CFU/g	L1T3R1	-3	143	-3	106	120000	5,08	-3	137	140000	5,146
				-4	14	-4	10			-4	12		
			L1T3R2	-3	128	-3	101	110000	5,04	-3	121	120000	5,079
				-4	12	-4	5			-4	9		
			L1T3R3	-3	95	-3	106	98000	4,99	-3	143	150000	5,176
				-4	9	-4	6			-4	19		
			L1T3R4	-3	131	-3	138	140000	5,15	-3	115	110000	5,041
				-4	14	-4	14			-4	10		
			L1T3R5	-3	125	-3	141	130000	5,11	-3	91	90000	4,954
				-4	12	-4	14			-4	8		

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Dairy and egg products

Matrix	Strain	Level	Sample number	Reference method 16649-2						Alternative method REC2 44°C			
				Replicate 1		Replicate 2		CFU/g	log CFU/g	Dilution	CFU / Petri dish	CFU/g	log CFU/g
				Dilution	CFU / Petri dish	Dilution	CFU / Petri dish						
Cheese - Batch 1 - TVC result: 1,0x10^7 CFU/g	ESC.1.5	300 CFU/g	L1T1R1	-1	13	-1	18	150	2,176	-1	17	180	2,255
				-2	1	-2	2			-2	3		
			L1T1R2	-1	18	-1	19	170	2,230	-1	19	170	2,230
				-2	1	-2	0			-2	0		
			L1T1R3	-1	14	-1	18	160	2,204	-1	24	240	2,380
				-2	2	-2	1			-2	2		
			L1T1R4	-1	12	-1	12	120	2,079	-1	7	70	1,845
				-2	3	-2	0			-2	1		
			L1T1R5	-1	10	-1	16	120	2,079	-1	18	170	2,230
				-2	1	-2	0			-2	1		
		3000 CFU/g	L1T2R1	-1	143	-1	151	1500	3,176	-1	160	1 500	3,176
				-2	10	-2	15			-2	10		
			L1T2R2	-1	144	-1	148	1500	3,176	-1	136	1 400	3,146
				-2	25	-2	14			-2	14		
			L1T2R3	-1	122	-1	150	1400	3,146	-1	148	1 500	3,176
				-2	19	-2	15			-2	21		
			L1T2R4	-1	165	-1	142	1500	3,176	-1	184	1 700	3,230
				-2	14	-2	16			-2	13		
			L1T2R5	-1	162	-1	165	1700	3,230	-1	161	1 700	3,230
				-2	22	-2	26			-2	28		
		100000 CFU/g	L1T3R1	-3	42	-3	54	51000	4,708	-3	36	36 000	4,556
				-4	8	-4	7			-4	4		
			L1T3R2	-3	57	-3	44	52000	4,716	-3	60	64 000	4,806
				-4	8	-4	5			-4	10		
			L1T3R3	-3	56	-3	56	56000	4,748	-3	60	61 000	4,785
				-4	5	-4	5			-4	7		
			L1T3R4	-3	43	-3	44	42000	4,623	-3	48	50 000	4,699
				-4	2	-4	4			-4	7		
			L1T3R5	-3	48	-3	46	47000	4,672	-3	62	60 000	4,778
				-4	4	-4	6			-4	4		

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Dairy and egg products

Matrix	Strain	Level	Sample number	Reference method 16649-2						Alternative method REC2 44°C			
				Replicate 1		Replicate 2		CFU/g	log CFU/g	Dilution	CFU / Petri dish	CFU/g	log CFU/g
				Dilution	CFU / Petri dish	Dilution	CFU / Petri dish						
Cheese - Batch 2 - TVC result: >300 CFU/g	ESC.1.5	300 CFU/g	L1T1R1	-1	15	-1	12	130	2,114	-1	34	330	2,519
				-2	1	-2	1			-2	2		
			L1T1R2	-1	16	-1	16	150	2,176	-1	22	210	2,322
				-2	1	-2	1			-2	1		
			L1T1R3	-1	15	-1	14	150	2,176	-1	14	130	2,114
				-2	3	-2	0			-2	0		
			L1T1R4	-1	8	-1	23	150	2,176	-1	14	140	2,146
				-2	1	-2	1			-2	1		
			L1T1R5	-1	21	-1	27	230	2,362	-1	16	170	2,230
				-2	2	-2	0			-2	3		
		3000 CFU/g	L1T2R1	-1	151	-1	155	1500	3,176	-1	169	1 600	3,204
				-2	10	-2	11			-2	5		
			L1T2R2	-1	169	-1	143	1600	3,204	-1	154	1 600	3,204
				-2	18	-2	15			-2	17		
			L1T2R3	-1	141	-1	140	1400	3,146	-1	171	1 700	3,230
				-2	19	-2	18			-2	17		
			L1T2R4	-1	170	-1	160	1700	3,230	-1	198	2 000	3,301
				-2	19	-2	20			-2	22		
			L1T2R5	-1	140	-1	134	1400	3,146	-1	156	1 600	3,204
				-2	21	-2	12			-2	25		
		100000 CFU/g	L1T3R1	-3	41	-3	58	48000	4,681	-3	60	68 000	4,833
				-4	2	-4	4			-4	15		
			L1T3R2	-3	52	-3	70	59000	4,771	-3	63	60 000	4,778
				-4	4	-4	4			-4	3		
			L1T3R3	-3	54	-3	54	56000	4,748	-3	66	67 000	4,826
				-4	8	-4	7			-4	8		
			L1T3R4	-3	50	-3	52	51000	4,708	-3	48	47 000	4,672
				-4	5	-4	5			-4	4		
			L1T3R5	-3	53	-3	49	52000	4,716	-3	72	74 000	4,869
				-4	7	-4	5			-4	9		

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Seafood products

Matrix	Strain	Level	Sample number	Reference method 16649-2						Alternative method REC2 44°C			
				Replicate 1		Replicate 2		CFU/g	log CFU/g	Dilution	CFU / Petri dish	CFU/g	log CFU/g
				Dilution	CFU / Petri dish	Dilution	CFU / Petri dish						
Fish - Batch 1 - TVC result: 1,0x10^7 CFU/g	ESC.1.31	300 CFU/g	L1T1R1	-1	51	-1	36	440	2,643	-1	48	450	2,653
				-2	5	-2	4			-2	1		
			L1T1R2	-1	31	-1	43	370	2,568	-1	29	280	2,447
				-2	5	-2	3			-2	2		
			L1T1R3	-1	42	-1	35	370	2,568	-1	39	380	2,580
				-2	3	-2	1			-2	3		
			L1T1R4	-1	32	-1	33	320	2,505	-1	39	360	2,556
				-2	1	-2	4			-2	1		
			L1T1R5	-1	41	-1	25	320	2,505	-1	40	400	2,602
				-2	4	-2	1			-2	4		
		3000 CFU/g	L1T2R1	-2	35	-2	36	3300	3,519	-2	59	5 600	3,748
				-3	0	-3	1			-3	3		
			L1T2R2	-2	28	-2	35	3300	3,519	-2	16	1 900	3,279
				-3	4	-3	5			-3	5		
			L1T2R3	-2	38	-2	47	4300	3,633	-2	54	5 300	3,724
				-3	2	-3	8			-3	4		
			L1T2R4	-2	46	-2	30	4000	3,602	-2	34	3 400	3,531
				-3	8	-3	4			-3	3		
			L1T2R5	-2	33	-2	34	3600	3,556	-2	33	3 200	3,505
				-3	6	-3	6			-3	2		
		100000 CFU/g	L1T3R1	-3	112	-3	126	120000	5,079	-3	98	100 000	5,000
				-4	12	-4	19			-4	15		
			L1T3R2	-3	107	-3	108	110000	5,041	-3	107	110 000	5,041
				-4	15	-4	10			-4	9		
			L1T3R3	-3	78	-3	104	93000	4,968	-3	110	110 000	5,041
				-4	12	-4	11			-4	7		
			L1T3R4	-3	106	-3	105	110000	5,041	-3	123	130 000	5,114
				-4	10	-4	8			-4	16		
			L1T3R5	-3	96	-3	119	110000	5,041	-3	142	140 000	5,146
				-4	9	-4	11			-4	12		

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Seafood products

Matrix	Strain	Level	Sample number	Reference method 16649-2						Alternative method REC2 44°C			
				Replicate 1		Replicate 2		CFU/g	log CFU/g	Dilution	CFU / Petri dish	CFU/g	log CFU/g
				Dilution	CFU / Petri dish	Dilution	CFU / Petri dish						
Fish - Batch 2 - TVC result: 2,5x10⁷ CFU/g	ESC.1.31	300 CFU/g	L1T1R1	-1	71	-1	68	670	2,828	-1	65	650	2,813
				-2	5	-2	4			-2	6		
			L1T1R2	-1	39	-1	35	380	2,577	-1	30	300	2,477
				-2	6	-2	3			-2	3		
			L1T1R3	-1	41	-1	41	400	2,607	-1	40	370	2,568
				-2	2	-2	5			-2	1		
			L1T1R4	-1	25	-1	35	320	2,503	-1	25	250	2,398
				-2	6	-2	4			-2	3		
			L1T1R5	-1	44	-1	42	400	2,597	-1	39	390	2,591
				-2	0	-2	1			-2	4		
		3000 CFU/g	L1T2R1	-2	35	-2	32	3500	3,544	-2	42	4 200	3,623
				-3	5	-3	5			-3	4		
			L1T2R2	-2	23	-2	38	3100	3,490	-2	34	3 100	3,491
				-3	3	-3	4			-3	1		
			L1T2R3	-2	26	-2	22	2600	3,421	-2	38	3 600	3,556
				-3	6	-3	4			-3	2		
			L1T2R4	-2	48	-2	38	4200	3,631	-2	37	4 300	3,633
				-3	5	-3	3			-3	10		
			L1T2R5	-2	36	-2	42	3900	3,587	-2	37	4 600	3,663
				-3	2	-3	5			-3	3		
		100000 CFU/g	L1T3R1	-3	127	-3	141	130000	5,117	-3	112	110 000	5,041
				-4	13	-4	7			-4	12		
			L1T3R2	-3	110	-3	103	100000	5,017	-3	118	120 000	5,079
				-4	5	-4	11			-4	15		
			L1T3R3	-3	95	-3	103	100000	5,014	-3	117	110 000	5,041
				-4	12	-4	17			-4	8		
			L1T3R4	-3	108	-3	109	110000	5,043	-3	115	120 000	5,079
				-4	14	-4	12			-4	17		
			L1T3R5	-3	128	-3	107	110000	5,057	-3	135	130 000	5,114
				-4	6	-4	10			-4	13		

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RAPID E.coli2 - enumeration of E.coli

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Vegetal products

Matrix	Strain	Level	Sample number	Reference method 16649-2						Alternative method REC2 44°C			
				Replicate 1		Replicate 2		CFU/g	log CFU/g	Dilution	CFU / Petri dish	CFU/g	log CFU/g
				Dilution	CFU / Petri dish	Dilution	CFU / Petri dish						
Vegetables - Batch 1 - TVC result: 2,0x10² CFU/g	ESC.1.2	300 CFU/g	L1T1R1	-1	6	-1	2	40	1,602	-1	12	120	2,079
				-2	2	-2	0			-2	1		
			L1T1R2	-1	4	-1	5	45	1,653	-1	8	80	1,903
				-2	0	-2	0			-2	0		
			L1T1R3	-1	8	-1	4	60	1,778	-1	16	160	2,204
				-2	0	-2	0			-2	1		
			L1T1R4	-1	8	-1	6	70	1,845	-1	10	110	2,041
				-2	1	-2	1			-2	2		
			L1T1R5	-1	9	-1	8	85	1,929	-1	12	120	2,079
				-2	1	-2	0			-2	1		
		3000 CFU/g	L1T2R1	-2	34	-2	46	4 200	3,623	-2	106	10 000	4,000
				-3	7	-3	5			-3	7		
			L1T2R2	-2	49	-2	51	4 800	3,681	-2	102	9 600	3,982
				-3	3	-3	3			-3	4		
			L1T2R3	-2	38	-2	38	3 700	3,568	-2	96	9 800	3,991
				-3	4	-3	1			-3	12		
			L1T2R4	-2	48	-2	40	4 400	3,643	-2	76	7 700	3,886
				-3	5	-3	3			-3	9		
			L1T2R5	-2	50	-2	60	5 600	3,748	-2	84	8 500	3,929
				-3	7	-3	5			-3	9		
		100000 CFU/g	L1T3R1	-4	10	-4	13	120 000	5,079	-4	26	250 000	5,398
				-5	2	-5	2			-5	2		
			L1T3R2	-4	14	-4	12	120 000	5,079	-4	24	240 000	5,380
				-5	1	-5	0			-5	2		
			L1T3R3	-4	13	-4	10	110 000	5,041	-4	38	360 000	5,556
				-5	1	-5	0			-5	2		
			L1T3R4	-4	14	-4	11	170 000	5,230	-4	28	270 000	5,431
				-5	6	-5	7			-5	2		
			L1T3R5	-4	10	-4	13	110 000	5,041	-4	28	210 000	5,322
				-5	2	-5	0			-5	6		

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RAPID E.coli2 - enumeration of E.coli   50/57   October 15, 2024

Vegetal products

Matrix	Strain	Level	Sample number	Reference method 16649-2						Alternative method REC2 44°C			
				Replicate 1		Replicate 2		CFU/g	log CFU/g	Dilution	CFU / Petri dish	CFU/g	log CFU/g
				Dilution	CFU / Petri dish	Dilution	CFU / Petri dish						
Vegetables - Batch 2 - TVC result: $1,0 \times 10^3$ CFU/g	ESC.1.2	300 CFU/g	L1T1R1	-1	6	-1	8	70	1,845	-1	9	90	1,954
				-2	0	-2	0			-2	2		
			L1T1R2	-1	8	-1	4	60	1,778	-1	10	100	2,000
				-2	0	-2	0			-2	0		
			L1T1R3	-1	6	-1	7	65	1,813	-1	14	140	2,146
				-2	0	-2	0			-2	1		
			L1T1R4	-1	8	-1	7	75	1,875	-1	19	190	2,279
				-2	0	-2	0			-2	2		
			L1T1R5	-1	9	-1	9	90	1,954	-1	12	120	2,079
				-2	1	-2	0			-2	1		
		3000 CFU/g	L1T2R1	-2	34	-2	36	3 600	3,556	-2	96	9 500	3,978
				-3	5	-3	3			-3	8		
			L1T2R2	-2	43	-2	40	4 300	3,633	-2	97	9 400	3,973
				-3	7	-3	4			-3	6		
			L1T2R3	-2	34	-2	28	3 500	3,544	-2	81	8 300	3,919
				-3	9	-3	5			-3	10		
			L1T2R4	-2	39	-2	40	3 800	3,580	-2	84	8 200	3,914
				-3	2	-3	2			-3	6		
			L1T2R5	-2	45	-2	53	4 800	3,681	-2	80	7 900	3,898
				-3	3	-3	4			-3	7		
		100000 CFU/g	L1T3R1	-4	58	-3	55	55 000	4,740	-4	28	260 000	5,415
				-5	3	-4	5			-5	1		
			L1T3R2	-3	81	-3	112	96 000	4,982	-4	23	250 000	5,398
				-4	9	-4	10			-5	4		
			L1T3R3	-3	108	-3	80	91 000	4,959	-4	32	310 000	5,491
				-4	8	-4	5			-5	2		
			L1T3R4	-4	16	-4	9	130 000	5,114	-4	29	300 000	5,477
				-5	3	-5	1			-5	4		
			L1T3R5	-4	15	-4	12	130 000	5,114	-4	24	230 000	5,362
				-5	1	-5	0			-5	1		

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RAPID E.coli2 - enumeration of E.coli

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Composite foods

Matrix	Strain	Level	Sample number	Reference method 16649-2						Alternative method REC2 44°C			
				Replicate 1		Replicate 2		CFU/g	log CFU/g	Dilution	CFU / Petri dish	CFU/g	log CFU/g
				Dilution	CFU / Petri dish	Dilution	CFU / Petri dish						
Piemontese salad - Batch 1 - TVC result: 1,0x10.³ CFU/g	ESC.1.28	300 CFU/g	L1T1R1	-1	13	-1	11	110	2,041	-1	17	170	2,230
				-2	0	-2	0			-2	2		
			L1T1R2	-1	15	-1	17	160	2,204	-1	13	150	2,176
				-2	1	-2	2			-2	3		
			L1T1R3	-1	13	-1	15	140	2,146	-1	20	200	2,301
				-2	0	-2	2			-2	2		
			L1T1R4	-1	15	-1	10	120	2,079	-1	14	140	2,146
				-2	2	-2	0			-2	1		
			L1T1R5	-1	15	-1	13	150	2,176	-1	24	270	2,431
				-2	4	-2	1			-2	6		
		3000 CFU/g	L1T2R1	-1	104	-1	113	1 100	3,041	-1	114	1 200	3,079
				-2	9	-2	11			-2	13		
			L1T2R2	-1	113	-1	91	1 000	3,000	-1	95	970	2,987
				-2	13	-2	10			-2	12		
			L1T2R3	-1	112	-1	108	1 100	3,041	-1	113	1 200	3,079
				-2	15	-2	9			-2	16		
			L1T2R4	-1	148	-1	152	1 500	3,176	-1	148	1 500	3,176
				-2	13	-2	13			-2	16		
			L1T2R5	-1	20	-1	18	1 900	3,279	-1	156	1 600	3,204
				-2	3	-2	1			-2	23		
		100000 CFU/g	L1T3R1	-3	59	-3	55	55 000	4,740	-3	44	44 000	4,643
				-4	4	-4	3			-4	4		
			L1T3R2	-3	49	-3	46	47 000	4,672	-3	54	54 000	4,732
				-4	6	-4	2			-4	5		
			L1T3R3	-3	28	-3	38	34 000	4,531	-3	58	58 000	4,763
				-4	5	-4	3			-4	6		
			L1T3R4	-3	44	-3	46	45 000	4,653	-3	43	44 000	4,643
				-4	4	-4	5			-4	5		
			L1T3R5	-3	46	-3	48	45 000	4,653	-3	60	58 000	4,763
				-4	1	-4	5			-4	4		

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RAPID E.coli2 - enumeration of E.coli   52/57   October 15, 2024

Composite foods

Matrix	Strain	Level	Sample number	Reference method 16649-2						Alternative method REC2 44°C			
				Replicate 1		Replicate 2		CFU/g	log CFU/g	Dilution	CFU / Petri dish	CFU/g	log CFU/g
				Dilution	CFU / Petri dish	Dilution	CFU / Petri dish						
Piemontese saled - Batch 2 - TVC result: 1,5x10^2 CFU/g	ESC.1.28	300 CFU/g	L1T1R1	-1	7	-1	15	100	2,000	-1	17	160	2,204
				-2	0	-2	1			-2	1		
			L1T1R2	-1	7	-1	11	90	1,954	-1	13	160	2,204
				-2	0	-2	1			-2	5		
			L1T1R3	-1	8	-1	19	130	2,114	-1	18	190	2,279
				-2	0	-2	2			-2	3		
			L1T1R4	-1	10	-1	10	100	2,000	-1	17	160	2,204
				-2	0	-2	3			-2	1		
			L1T1R5	-1	10	-1	16	120	2,079	-1	16	150	2,176
				-2	0	-2	1			-2	1		
		3000 CFU/g	L1T2R1	-1	113	-1	106	1 100	3,041	-1	102	1 000	3,000
				-2	12	-2	9			-2	12		
			L1T2R2	-1	130	-1	142	1 400	3,146	-1	119	1 200	3,079
				-2	15	-2	19			-2	9		
			L1T2R3	-1	123	-1	167	1 500	3,176	-2	20	1 900	3,279
				-2	10	-2	19			-3	1		
			L1T2R4	-1	123	-1	119	1 200	3,079	-1	99	1 000	3,000
				-2	14	-2	9			-2	14		
			L1T2R5	-2	14	-2	18	1 600	3,204	-2	18	1 700	3,230
				-3	1	-3	2			-3	1		
		100000 CFU/g	L1T3R1	-3	53	-3	56	54 000	4,732	-3	60	60 000	4,778
				-4	5	-4	5			-4	6		
			L1T3R2	-3	52	-3	63	57 000	4,756	-3	65	66 000	4,820
				-4	5	-4	5			-4	8		
			L1T3R3	-3	38	-3	39	37 000	4,568	-3	55	54 000	4,732
				-4	2	-4	3			-4	4		
			L1T3R4	-3	44	-3	62	53 000	4,724	-3	61	59 000	4,771
				-4	5	-4	5			-4	4		
			L1T3R5	-3	55	-3	42	49 000	4,690	-3	62	61 000	4,785
				-4	5	-4	5			-4	5		

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RAPID E.coli2 - enumeration of E.coli

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APPENDIX F - Inclusivity

#	Study	Code	Strain	Origin	PCA at 30°C		REC2 at 44°C				TBX at 44°C			
					R1	R2	R1		R2		R1		R2	
					Nombre de colonies	Nombre de colonies	Couleur des colonies	Nombre de colonies	Couleur des colonies	Nombre de colonies	Couleur des colonies	Nombre de colonies	Couleur des colonies	Nombre de colonies
1		/	*Escherichia coli*	Rognons de porc	60	74	violet	65	violet	85	/	/	/	/
2		/	*Escherichia coli*	Chou rouge	47	70	violet	48	violet	82	/	/	/	/
3		/	*Escherichia coli*	Persil	89	88	violet	87	violet	88	/	/	/	/
4		/	*Escherichia coli*	Pâtisserie	123	122	violet	119	violet	128	/	/	/	/
5		/	*Escherichia coli*	Crépinette	56	106	violet	49	violet	111	/	/	/	/
6		/	*Escherichia coli*	Crépinette	111	145	violet	108	violet	139	/	/	/	/
7		/	*Escherichia coli*	Chair à saucisse	134	98	violet	130	violet	89	/	/	/	/
8		/	*Escherichia coli*	Tomate	87	84	violet	88	violet	89	/	/	/	/
9		/	*Escherichia coli*	Céleri rémoulade	79	105	violet	81	violet	115	/	/	/	/
10		/	*Escherichia coli*	Crème vanille	65	126	violet	60	violet	128	/	/	/	/
11	Etude de validation initiale	/	*Escherichia coli*	Merguez	143	96	violet	139	violet	90	/	/	/	/
12		/	*Escherichia coli*	Foie de porc	54	107	violet	60	violet	104	/	/	/	/
13		/	*Escherichia coli*	Lait cru	67	122	violet	66	violet	115	/	/	/	/
14		/	*Escherichia coli*	Fromage au lait cru	132	141	violet	123	violet	119	/	/	/	/
15		/	*Escherichia coli*	Moules	128	109	violet	134	violet	100	/	/	/	/
16		/	*Escherichia coli*	Chair à saucisse	76	56	violet	79	violet	67	/	/	/	/
17		/	*Escherichia coli*	Carottes râpées	97	89	violet	93	violet	89	/	/	/	/
18		/	*Escherichia coli*	Pâtisserie	104	76	violet	117	violet	70	/	/	/	/
19		/	*Escherichia coli*	Chou à la crème	57	98	violet	54	violet	97	/	/	/	/
20		/	*Escherichia coli*	Crème pâtissière	113	73	violet	118	violet	70	/	/	/	/
21		/	*Escherichia coli*	Taboulé	93	91	violet	95	violet	99	/	/	/	/
22		/	*Escherichia coli*	Crème chantilly	71	107	violet	65	violet	101	/	/	/	/
23		/	*Escherichia coli*	Lardons	132	99	violet	139	violet	95	/	/	/	/
24		/	*Escherichia coli*	Saumon fumé	96	141	violet	98	violet	135	/	/	/	/
25		/	*Escherichia coli*	Filet de rouget	153	38	violet	156	violet	40	/	/	/	/
26		/	*Escherichia coli*	Sandwich	49	99	violet	56	violet	100	/	/	/	/
27		/	*Escherichia coli*	Pâtisserie	100	116	violet	100	violet	121	/	/	/	/
28		/	*Escherichia coli*	Foie de porc	113	125	violet	119	violet	121	/	/	/	/
29		/	*Escherichia coli*	Rognons de porc	88	94	violet	84	violet	92	/	/	/	/
30		/	*Escherichia coli*	Lait cru	111	114	violet	109	violet	112	/	/	/	/
31		ESC.1.11	*Escherichia coli*	Chicken fillet	97	95	violet	98	violet	100	bleu	98	bleu	100
32		ESC.1.111	*Escherichia coli*	Fountain water	89	93	violet	79	violet	87	bleu	78	bleu	82
33	Etude de reconduction de 2016	ESC.1.12	*Escherichia coli*	Lamb necklace	54	60	violet	53	violet	49	bleu	49	bleu	49
34		ESC.1.13	*Escherichia coli*	Croissant stuffed	86	80	violet	80	violet	73	bleu	84	bleu	76
35		ESC.1.139	*Escherichia coli*	River water	85	85	violet	83	violet	78	bleu	83	bleu	80
36		ESC.1.140	*Escherichia coli*	River water	52	59	violet	45	violet	41	bleu	49	bleu	49
37		ESC.1.145	*Escherichia coli*	Crudeness	125	111	violet	117	violet	112	bleu	118	bleu	120
38		ESC.1.146	*Escherichia coli*	Cheese	47	47	violet	43	violet	39	bleu	39	bleu	44
39		ESC.1.15	*Escherichia coli*	Beef muscle	103	105	violet	99	violet	104	bleu	99	bleu	104

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RAPID E.coli2 - enumeration of E.coli
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APPENDIX F - Inclusivity

#	Study	Code	Strain	Origin	PCA at 30°C		REC2 at 44°C				TBX at 44°C			
					R1	R2	R1		R2		R1		R2	
					Nombre de colonies	Nombre de colonies	Couleur des colonies	Nombre de colonies	Couleur des colonies	Nombre de colonies	Couleur des colonies	Nombre de colonies	Couleur des colonies	Nombre de colonies
40	Etude de reconduction de 2016	ESC.1.18	*Escherichia coli*	"Rillettes" of cooked and smoked salmon	98	94	violet	89	violet	87	bleu	89	bleu	78
41		ESC.1.19	*Escherichia coli*	Shrimp ravioli	49	48	violet	50	violet	46	bleu	48	bleu	40
42		ESC.1.2	*Escherichia coli*	Grated carot	122	130	violet	111	violet	109	bleu	113	bleu	116
43		ESC.1.20	*Escherichia coli*	Meat for kebab	119	119	violet	115	violet	97	bleu	115	bleu	106
44		ESC.1.21	*Escherichia coli*	Beef granules with 20% of fat	38	45	violet	32	violet	29	bleu	40	bleu	40
45		ESC.1.26	*Escherichia coli*	Vegetable sauce	38	39	violet	34	violet	33	bleu	35	bleu	33
46		ESC.1.28	*Escherichia coli*	Mixed salad	123	126	violet	117	violet	118	bleu	118	bleu	113
47		ESC.1.3	*Escherichia coli*	Dairy industry	56	62	violet	42	violet	49	bleu	48	bleu	49
48		ESC.1.5	*Escherichia coli*	Cheese	56	53	violet	42	violet	49	bleu	51	bleu	56
49		ESC.1.50	*Escherichia coli*	Cheese	49	42	violet	49	violet	52	bleu	46	bleu	45
50		ESC.1.6	*Escherichia coli*	Chicken ravioli	112	108	violet	87	violet	89	bleu	101	bleu	104

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RAPID E.coli2 - enumeration of E.coli

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APPENDIX F - Exclusivity

#	Souche	Origine	PCA à 30°C		REC2 à 44°C				TBX à 44°C			
			R1	R2	R1		R2		R1		R2	
			Nombre de colonies	Nombre de colonies	Couleur des colonies	Nombre de colonies	Couleur des colonies	Nombre de colonies	Couleur des colonies	Nombre de colonies	Couleur des colonies	Nombre de colonies
1	*Citrobacter diversus*	Aliments animaux	146	132	bleu	118	bleu	120	/	/	/	/
2	*Citrobacter diversus*	Herbes séchées	136	143	bleu	110	bleu	152	/	/	/	/
3	*Citrobacter diversus*	Levure	164	112	bleu	160	bleu	123	/	/	/	/
4	*Citrobacter freundii*	Produit carné	39	55	bleu clair	40	bleu clair	54	/	/	/	/
5	*Citrobacter freundii*	Végétaux	38	59	bleu clair	44	bleu clair	59	/	/	/	/
6	*Citrobacter freundii*	Poisson	33	51	bleu clair	46	bleu clair	45	/	/	/	/
7	*Citrobacter freundii*	Lait	66	90	bleu clair	67	bleu clair	90	/	/	/	/
8	*Enterobacter amnigenus*	Brochette de poisson	96	71	bleu	87	bleu	76	/	/	/	/
9	*Enterobacter amnigenus*	Jambon	60	67	bleu	70	bleu	73	/	/	/	/
10	*Enterobacter cloacae*	Produit laitier	86	81	bleu	91	bleu	75	/	/	/	/
11	*Enterobacter cloacae*	Produit laitier	48	63	bleu	50	bleu	51	/	/	/	/
12	*Enterobacter cloacae*	Produit laitier	118	110	bleu	127	bleu	121	/	/	/	/
13	*Enterobacter cloacae*	Produit laitier	88	91	bleu	105	bleu	85	/	/	/	/
14	*Enterobacter sakazakii*	Aliments animaux	135	113	bleu	140	bleu	121	/	/	/	/
15	*Enterobacter sakazakii*	Pâtisserie	75	59	bleu	85	bleu	64	/	/	/	/
16	*Erwinia spp.*	Collection	23	112	blanc-gris	22	blanc-gris	102	/	/	/	/
17	*Escherichia coli* O157	Collection	36	103	bleu	36	bleu	97	/	/	/	/
18	*Escherichia hermanii*	Aliments animaux	56	102	bleu-gris	54	bleu-gris	100	/	/	/	/
19	*Escherichia hermanii*	Produit carné	50	143	bleu	65*	bleu	134	/	/	/	/
20	*Escherichia hermanii*	Produit laitier	22	89	bleu	10*	bleu	32	/	/	/	/
21	*Escherichia vulneris*	Produit carné	31	112	bleu	14	bleu	87	/	/	/	/
22	*Hafnia alvei*	Foie de porc	90	82	ng	0	ng	0	/	/	/	/
23	*Hafnia alvei*	Reblochon	80	75	blanc-gris	87	blanc-gris	73	/	/	/	/
24	*Hafnia alvei*	Persil	75	76	ng	0	ng	0	/	/	/	/
25	*Hafnia alvei*	Flétan	84	50	ng	0	ng	0	/	/	/	/
26	*Hafnia alvei*	Viande hachée	127	119	ng	0	ng	0	/	/	/	/
27	*Hafnia alvei*	Lait cru	109	134	ng	0	ng	0	/	/	/	/
28	*Hafnia alvei*	Echine de porc	126	97	ng	0	ng	0	/	/	/	/
29	*Hafnia alvei*	Rognons de porc	150	101	ng	0	ng	0	/	/	/	/
30	*Hafnia alvei*	Persil	123	59	ng	0	ng	0	/	/	/	/
31	*Hafnia alvei*	Brochette de poisson	134	128	ng	0	ng	0	/	/	/	/
32	*Hafnia alvei*	Concombre	121	107	ng	0	ng	0	/	/	/	/
33	*Hafnia alvei*	Tomate	150	89	ng	0	ng	0	/	/	/	/
34	*Klebsiella oxytoca*	Aliments animaux	40	134	bleu clair	45	bleu clair	150	/	/	/	/
35	*Klebsiella oxytoca*	Poisson	62	36	gris	32	gris	18*	/	/	/	/

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APPENDIX F - Exclusivity

#	Souche	Origine	PCA à 30°C		REC2 à 44°C				TBX à 44°C			
			R1	R2	R1		R2		R1		R2	
			Nombre de colonies	Nombre de colonies	Couleur des colonies	Nombre de colonies	Couleur des colonies	Nombre de colonies	Couleur des colonies	Nombre de colonies	Couleur des colonies	Nombre de colonies
36	*Klebsiella pneumoniae*	Poudre de lait	42	36	bleu	44	bleu	51	/	/	/	/
37	*Klebsiella pneumoniae*	Macédoine	55	38	bleu	43	bleu	36	/	/	/	/
38	*Moellerella wisconsensis*	Andouillette	45	46	ng	0	ng	0	/	/	/	/
39	*Proteus mirabilis*	Produit carné	93	145	bleu-gris	81	bleu-gris	123	/	/	/	/
40	*Proteus mirabilis*	Foies de volaille	115	102	bleu-gris	91*	bleu-gris	88	/	/	/	/
41	*Pseudomonas aeruginosa*	Filet de rouget	30	87	blanc	24	blanc	78	/	/	/	/
42	*Salmonella arizonae IIIb 61:-:-*	Dinde	66	45	violet	60	violet	43	/	/	/	/
			75	64	violet	53	violet	39	bleu	56	bleu	31
43	*Salmonella arizonae IIIa 48:24:223*	Elevage d'oie	116	76	bleu	105	bleu	81	/	/	/	/
44	*Salmonella arizonae IIIb 61:i:z53*	Cuisse de poulet	96	97	violet	100	violet	86	/	/	/	/
			112	85	violet	74	violet	51	bleu	63	bleu	55
45	*Salmonella* Enteritidis	Ovoproduit	121	105	blanc-gris	93	blanc-gris	103	/	/	/	/
46	*Salmonella* Typhimurium	Foie de porc	83	103	blanc-gris	82	blanc-gris	99	/	/	/	/
47	*Serratia marcescens*	Lait cru	45	55	ng	0	ng	0	/	/	/	/
48	*Serratia liquefaciens*	Andouillette	63	45	ng	0	ng	0	/	/	/	/
49	*Shigella flexneri*	Collection	25	56	blanc-gris	19*	blanc-gris	46	/	/	/	/
50	*Shigella flexneri*	Collection	30	35	ng	0	ng	0	/	/	/	/
51	*Shigella sonnei*	Collection	26	33	violet	35	violet	32	/	/	/	/
			39	21	violet	25	violet	15	bleu	23	bleu	11
52	*Yersinia kristensenii*	Collection	27	67	ng	0	ng	0	/	/	/	/
53	*Yersinia enteritidis*	Ovoproduit	12	34	ng	0	ng	0	/	/	/	/
54	*Yersinia enterocolitica*	Ovoproduit	28	45	ng	0	ng	0	/	/	/	/

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