

**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 07/07 – 12/04**  
**for the enumeration at 37°C of  $\beta$ -glucuronidase positive**  
***Escherichia coli* in human food products, environmental samples**  
**and animal feed.**

**Expert Laboratory:**      Laboratoire MICROSEPT  
ZA de la Sablonnière  
15 rue Denis Papin  
49220 LE LION D'ANGERS  
FRANCE

**For:**                      BIO-RAD  
3, boulevard Raymond Poincaré  
92430 MARNES-LA-COQUETTE  
FRANCE

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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  $\beta$ -glucuronidase positive *Escherichia coli*– Colony count technique at 44°C using 5-bromo-4-chloro-3-indolyl  $\beta$ -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,
  - environmental samples,
  - animal feed

- Certification body:

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

## 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  $\beta$ -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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## **Appendices**

Appendix A: Protocol of the alternative method

Appendix B: Protocol of the reference method

Appendix C: Artificial contaminations

Appendix D: Relative trueness study - Raw results

Appendix E: Relative trueness study - Statistical calculations

Appendix F: Accuracy profile study - Raw results

Appendix G: Inclusivity and exclusivity study – Raw results

## 1. Introduction

The RAPID'*E. coli* 2 method is validated by AFNOR Certification under the NF VALIDATION brand with the certification number BRD 07/01 – 07/93 according to the standard ISO 16140-2:2016 for the enumeration of  $\beta$ -glucuronidase *Escherichia coli* at 37°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	/
Extension study	December 2022	ISO 16140-2:2016 ISO 16649-2:2001	Microsept	Addition of environmental samples category
Extension study	April 2023	ISO 16140-2:2016 ISO 16649-2:2001	Microsept	Addition of Animal Feed category
Fifth renewal study	October 2024	ISO 16140-2:2016 ISO 16649-2:2001	Microsept	No additional tests

This document is renewal study of validation study of NF Validation certification of the RAPID'*E. coli* 2 method according to the ISO 16140-2:2016 standard.

During the Technical Board of December 2022, it was decided to reclassify some samples from previous validations. This new classification was applied, and some new samples had to be generated in 2023 during the extension of animal feed category.

A part of 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.

## 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  $\beta$ -D-glucuronidase (GLUC) and the  $\beta$ -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 37±1°C for 21±3 hours.

After incubation, the violet to pink colonies are enumerated as  $\beta$ -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  $\beta$ -glucuronidase positive *Escherichia coli*– Colony count technique at 44°C using 5 bromo-4-chloro-3-indolyl  $\beta$ -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.

### 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, 224 samples were analyzed giving 131 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*

Category	Type	Samples analyzed	Inter. results
<b>Meats products</b>	a Raw products	26	9
	b Ready-to-reheat products	7	6
	c Smoked and cured products	7	6
	<b>Total</b>	<b>40</b>	<b>21</b>
<b>Dairy &amp; egg products</b>	a Pasteurized and dehydrated products	8	5
	b Raw milk products	10	6
	c Desserts and egg products	21	12
	<b>Total</b>	<b>39</b>	<b>23</b>
<b>Seafood products</b>	a Raw products	15	5
	b Marinated and smoked products	14	11
	c Ready-to-reheat products	8	5
	<b>Total</b>	<b>37</b>	<b>21</b>
<b>Vegetal products</b>	a Raw products	6	5
	b Pre-cut and pre-cooked products	10	5
	c Processed products	8	5
	<b>Total</b>	<b>24</b>	<b>15</b>
<b>Ready-to-eat &amp; ready-to-reheat products</b>	a Ready-to-eat products	8	5
	b Ready-to-reheat products	8	5
	c Smoked and marinated products	6	5
	<b>Total</b>	<b>22</b>	<b>15</b>
<b>Environmental samples</b>	a Surface	15	6
	b Process water	7	6
	c Dusts and residues	11	6
	<b>Total</b>	<b>33</b>	<b>18</b>
<b>Animal Feed</b>	a Pet food	11	5
	b Animal feed (cereals and flour)	8	6
	c Ingredients	10	7
	<b>Total</b>	<b>29</b>	<b>18</b>
<b>Total</b>		<b>224</b>	<b>131</b>



### 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 ISO 16140-2:2016.

Among the interpretable results, 56 correspond to naturally contaminated samples and 75 to artificially contaminated samples. The artificial contaminations performed are presented in 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 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 in figure 1 and for all categories in figure 2.

Figure 1: Two-dimensional plots per category

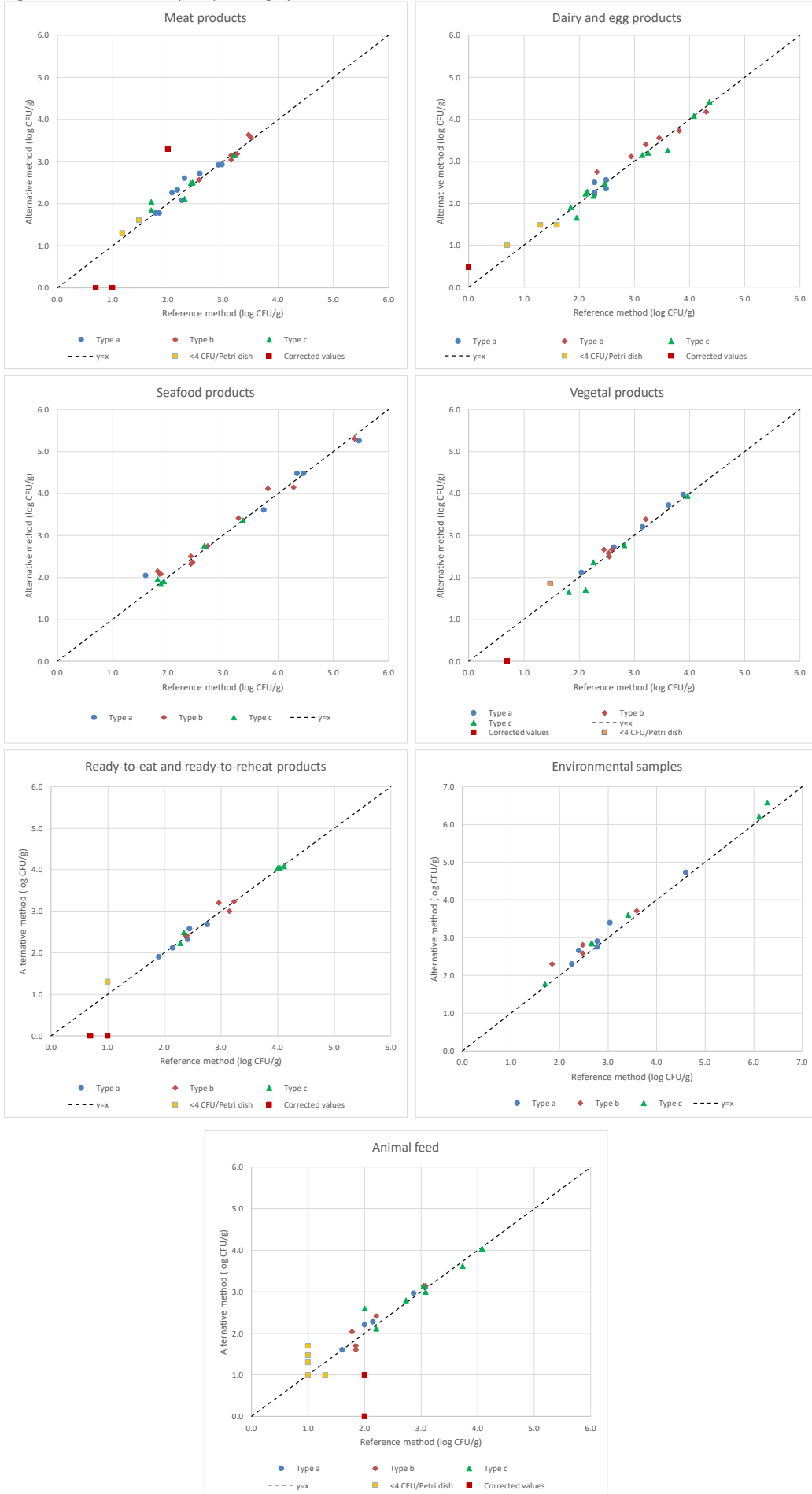
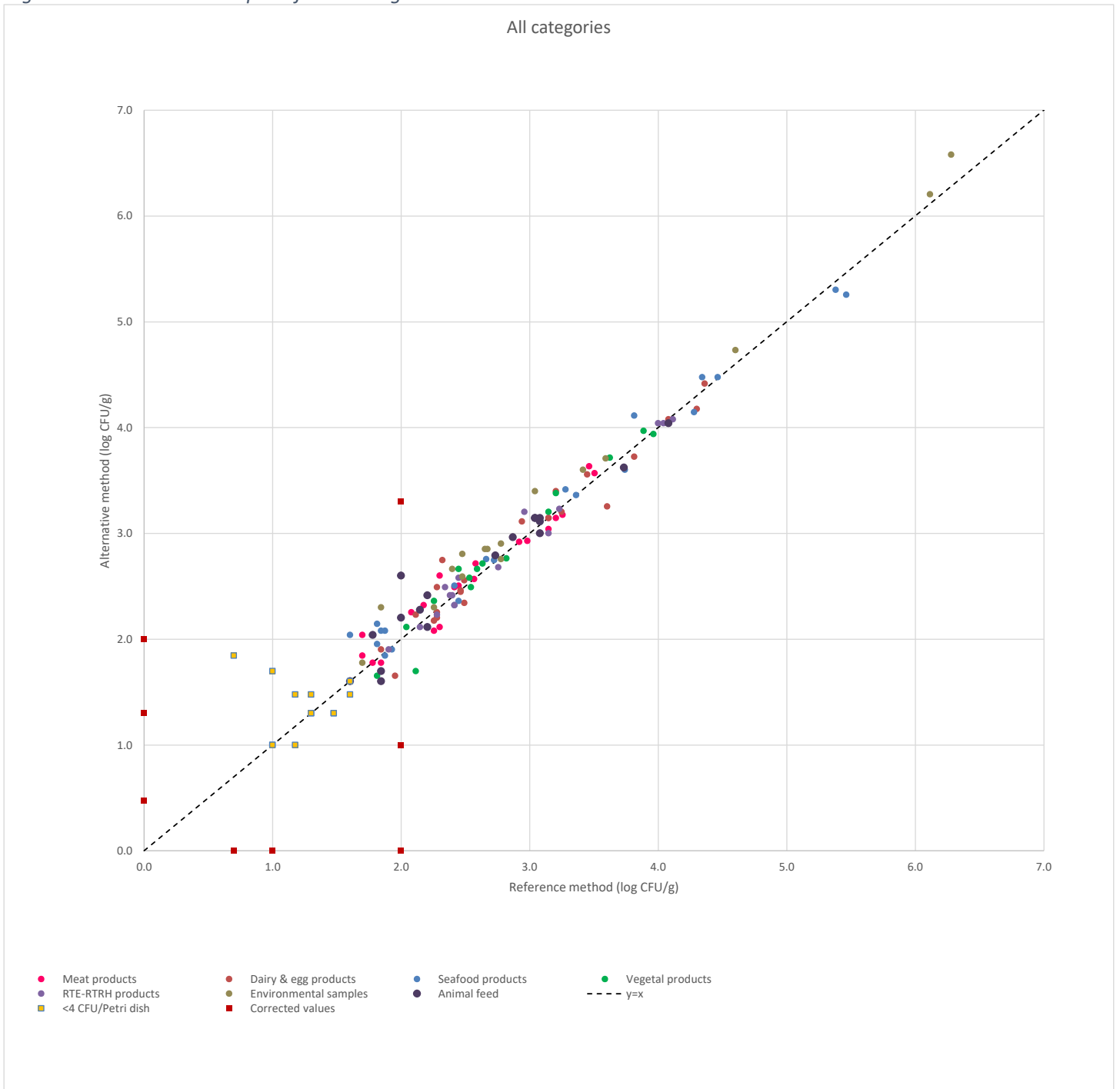


Figure 2: Two-dimensional plots for all categories



### 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	21	0.04	0.14	-0.26	0.34
Dairy products	23	0.01	0.17	-0.34	0.37
Seafood products	21	0.06	0.17	-0.30	0.31
Vegetal products	15	0.02	0.15	-0.32	0.36
Ready-to-eat & ready-to-reheat products	15	0.01	0.10	-0.21	0.07
Environmental samples	18	0.17	0.13	-0.11	0.45
Animal Feed	18	0.07	0.19	-0.34	0.47
<b>All categories</b>	<b>131</b>	<b>0.06</b>	<b>0.16</b>	<b>-0.26</b>	<b>0.37</b>

Overall, the average difference is equal to 0.06, showing a weak positive bias between the RAPID'E.coli 2 method and the reference method.

The average difference varies from 0.01 log CFU/g (dairy and egg products) to 0.17 CFU/g (environmental samples).

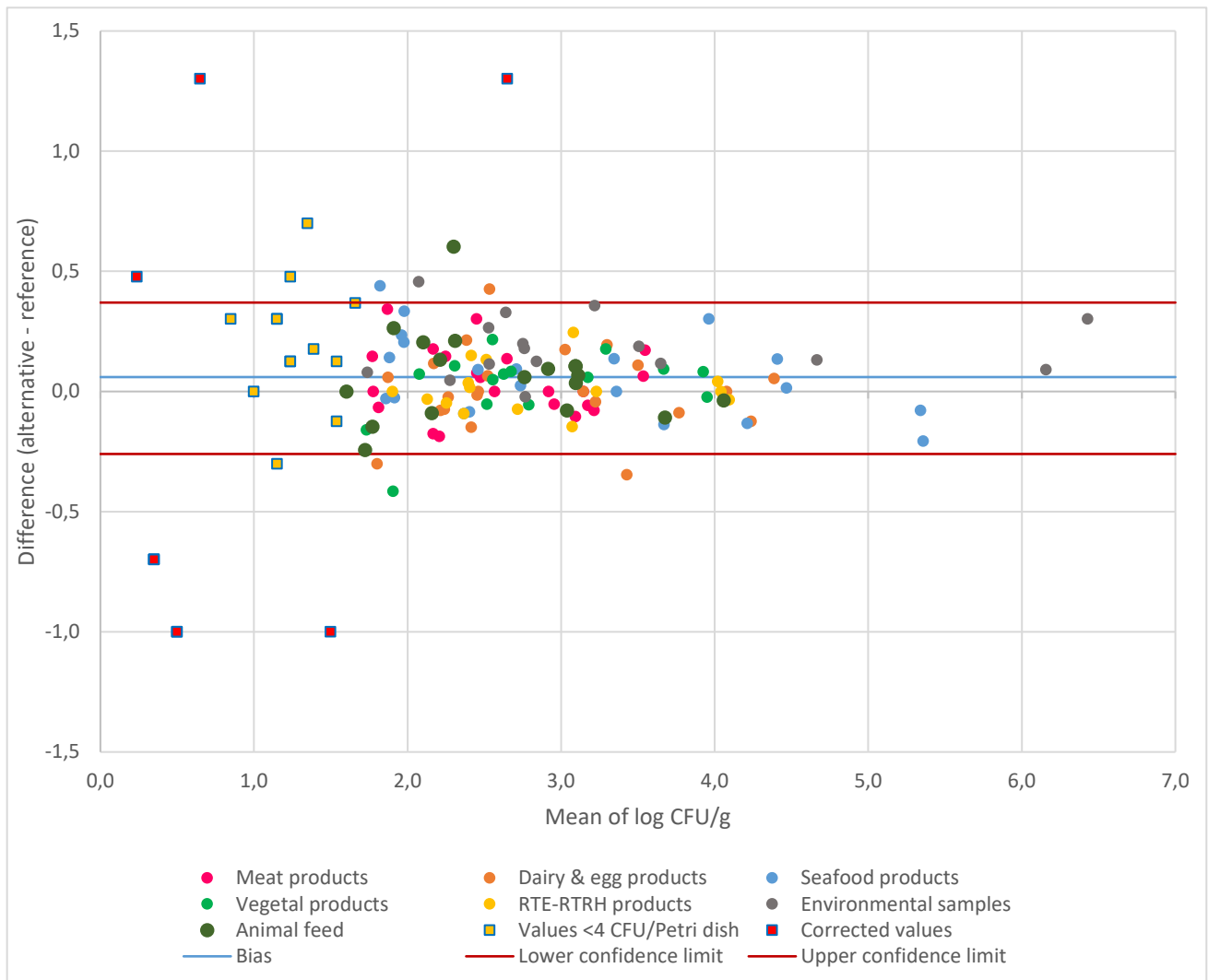
Upper and lower 95% confidence interval limits are lower than 0.5 log showing a good correlation between the two methods.

The Bland-Altman difference plots are presented for all categories in figure 3.

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.

Figure 3: Bland-Altman difference plot for all categories



- **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	AM	RM	AM	Mean	Difference
			CFU/g	CFU/g	log CFU/g	log CFU/g		
Meat products	a	26	10	<10	1.00	0.00	0.50	-1.00
	a	28	5	<10	0.70	0.00	0.35	-0.70
	c	ISHA 36	<1000	2000	2.00	3.30	2.65	1.30
Dairy and egg products	b	25	210	560	2.32	2.75	2.54	0.43
	b	5	<1	3	0.00	0.48	0.24	0.48
	c	20	90	45	1.95	1.65	1.80	-0.30
	c	72	4000	1800	3.60	3.26	3.43	-0.35
Vegetal products	b	ISHA 15	5	<10	0.70	0.00	0.35	-0.70
	c	ISHA 6	130	50	2.11	1.70	1.91	-0.41
Composite foods	a	19	5	<10	0.70	0.00	0.35	-0.70
	b	ISHA 14	10	<10	1.00	0.00	0.50	-1.00
Env. samples	b	2347756	70	200	1.85	2.30	2.07	0.46
Animal feed	a	2364270	100	<100	2.00	1.00	1.50	-1.00
	a	2364268	100	<100	2.00	0.00	1.00	-2.00
	a	2392518	10	50	1.00	1.70	1.35	0.70
	a	2392520	<10	20	0.00	1.30	0.65	1.30
	b	2392588	20	10	1.30	1.00	1.15	-0.30
	c	2392485	<10	100	0.00	2.00	1.00	2.00
	c	2392522	10	30	1.00	1.48	1.24	0.48
	c	2392525	100	400	2.00	2.60	2.30	0.60

Twenty samples are outside the confidence limits: 14 relate to corrected values or samples with less than 4 CFU/Petri dish, 3 are higher than the upper confidence limit and 3 are lower than the lower confidence limit.

### 3.1.6. Conservation of the incubated Petri dishes at 5±3°C for 3 days

During this extension study, evolutions were observed for 5 samples. Among these samples, 2 were not interpreted (2364266 and 2364270) and 3 were interpreted (2392525, 2319051 and 2392621).

A new statistical interpretation only for the category tested in this extension (i.e. Animal Feed) was realized.

	Category	n	Average difference = bias	Standard deviation differences	Lower Confidence Limit	Upper Confidence Limit
Before storage	Animal feed	18	0.07	0.19	-0.34	0.47
After storage	Animal feed	18	0.07	0.19	-0.34	0.47

No statistical difference was observed between the reading before and after the Petri dishes storage.

### 3.1.7. Conclusion

The relative trueness study of the alternative method is satisfactory.

## 3.2. Accuracy profile study

### 3.2.1. Protocols

Six matrix-strain pairs 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	<i>E. coli</i> ESC.1.15	Ground beef 20% fat	2.5
Dairy products	Raw milk cheese	<i>E. coli</i> ESC.1.5	Camembert	
Seafood products	Fish fillet	<i>E. coli</i> ESC.1.31	Scallop shell	
Vegetal products	Grated carrots	<i>E. coli</i> ESC.1.2	Grated carrots	3.5
Composite foods	Piemontese salad	<i>E. coli</i> ESC.1.28	Salad pasta, surimi, mayonnaise	
Environmental samples	Process water	<i>Escherichia coli</i> YMJ695	Tap water	5.0
Animal Feed	Dog pâté	<i>Escherichia coli</i> GQRP82	Raw turkey meat	

### 3.2.2. Results

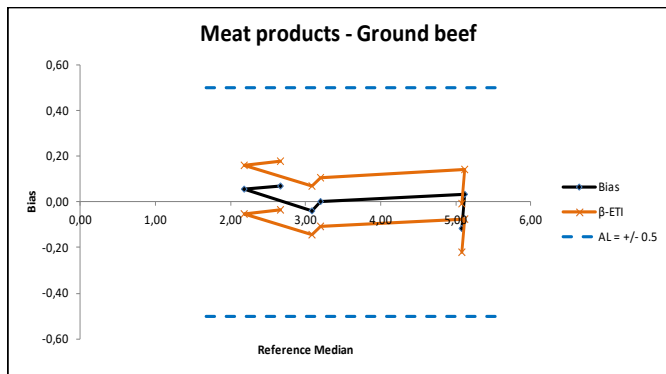
Raw data are provided in Appendix F.

The statistical data and the accuracy profiles are shown in figure 4.

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>.

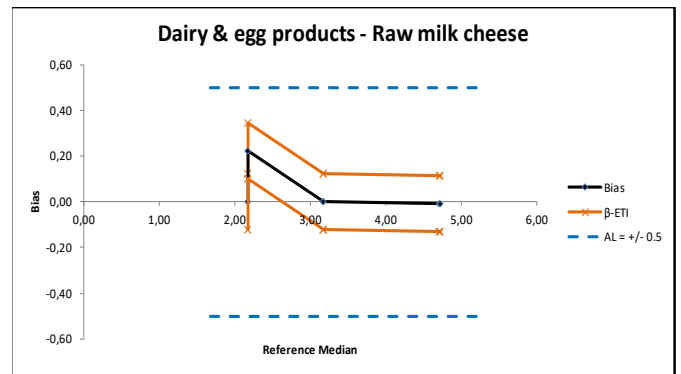
The probability for the tolerance interval is set at 80% and the central value is the median. The acceptability limit is set at AL = 0.5 log<sub>10</sub> CFU/g or ml.

Figure 4: Accuracy profiles per category



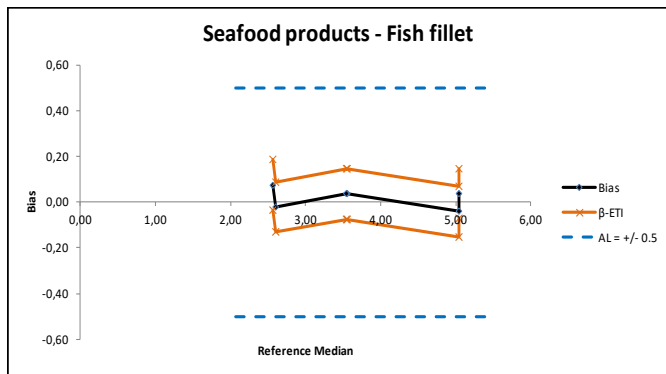
Sample Name	Reference central value	Bias	Lower $\beta$ -ETI	Upper $\beta$ -ETI	$\beta$ -ETI compared to AL=±0.5 Acceptable	$\beta$ -ETI compared to final AL Acceptable
2.66	0.070	-0.037	0.176		YES	YES
2.18	0.054	-0.052	0.161		YES	YES
3.08	-0.038	-0.145	0.069		YES	YES
3.20	0.000	-0.107	0.107		YES	YES
5.11	0.032	-0.075	0.139		YES	YES
5.08	-0.115	-0.222	-0.009		YES	YES

	Reference method	Alternative method	SD repeatability of reference method $\leq 0.125$	Final AL
SD Repeatability	0,085	0,074	YES	+/- 0,500



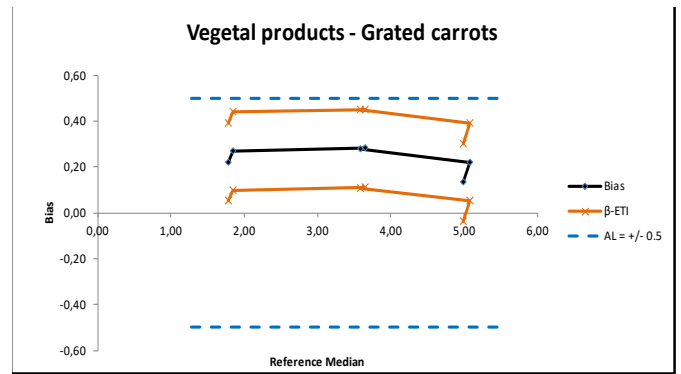
Sample Name	Reference Central value	Bias	Lower $\beta$ -ETI	Upper $\beta$ -ETI	$\beta$ -ETI compared to AL=±0.5 Acceptable	$\beta$ -ETI compared to final AL Acceptable
2.18	0.000	-0.121	0.121		YES	YES
2.18	0.222	0.101	0.343		YES	YES
3.18	0.000	-0.121	0.121		YES	YES
3.18	0.000	-0.121	0.121		YES	YES
4.71	-0.009	-0.130	0.113		YES	YES
4.72	-0.008	-0.130	0.113		YES	YES

	Reference method	Alternative method	SD repeatability of reference method $\leq 0.125$	Final AL
SD Repeatability	0,057	0,084	YES	+/- 0,500



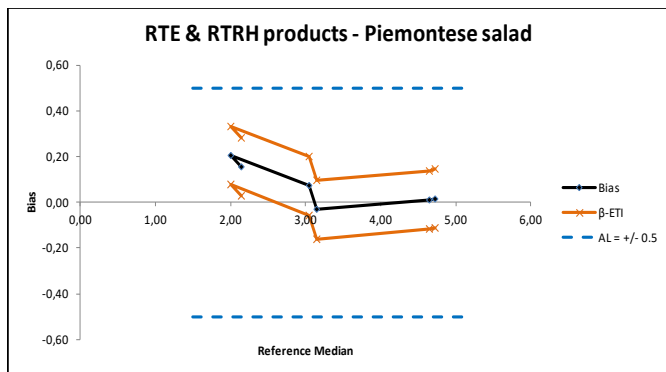
Sample Name	Reference Central value	Bias	Lower $\beta$ -ETI	Upper $\beta$ -ETI	$\beta$ -ETI compared to AL=±0.5 Acceptable	$\beta$ -ETI compared to final AL Acceptable
2.57	0.075	-0.034	0.185		YES	YES
2.60	-0.022	-0.132	0.087		YES	YES
3.56	0.035	-0.075	0.144		YES	YES
3.54	0.036	-0.074	0.145		YES	YES
5.04	-0.041	-0.151	0.068		YES	YES
5.04	0.038	-0.072	0.148		YES	YES

	Reference method	Alternative method	SD repeatability of reference method $\leq 0.125$	Final AL
SD Repeatability	0,072	0,076	YES	+/- 0,500



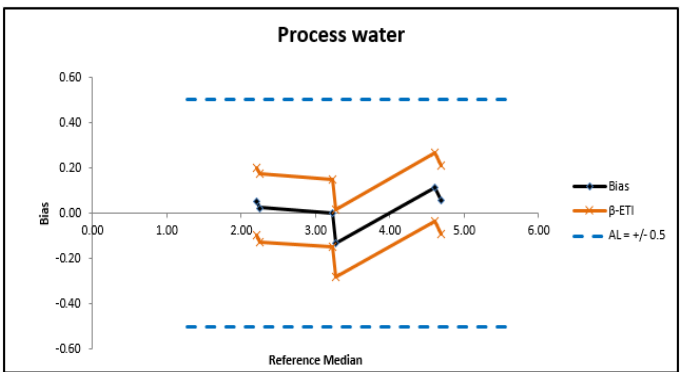
Sample Name	Reference Central value	Bias	Lower $\beta$ -ETI	Upper $\beta$ -ETI	$\beta$ -ETI compared to AL=±0.5 Acceptable	$\beta$ -ETI compared to final AL Acceptable
1.78	0.222	0.052	0.392		YES	YES
1.85	0.269	0.098	0.439		YES	YES
3.64	0.281	0.110	0.451		YES	YES
3.54	0.278	0.107	0.448		YES	YES
5.08	0.222	0.052	0.392		YES	YES
4.98	0.132	-0.039	0.302		YES	YES

	Reference method	Alternative method	SD repeatability of reference method $\leq 0.125$	Final AL
SD Repeatability	0,100	0,118	YES	+/- 0,500



Sample Name	Reference Central value	Bias	Lower $\beta$ -ETI	Upper $\beta$ -ETI	$\beta$ -ETI compared to AL=±0.5 Acceptable	$\beta$ -ETI compared to final AL Acceptable
2.15	0.155	0.026	0.283		YES	YES
2.00	0.204	0.076	0.333		YES	YES
3.04	0.073	-0.056	0.201		YES	YES
3.15	-0.032	-0.161	0.096		YES	YES
4.65	0.010	-0.119	0.138		YES	YES
4.72	0.016	-0.112	0.145		YES	YES

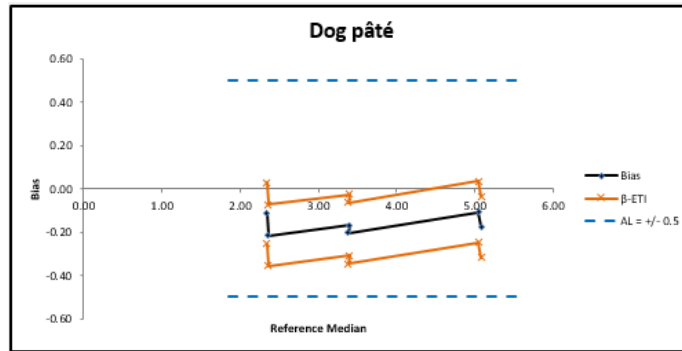
	Reference method	Alternative method	SD repeatability of reference method $\leq 0.125$	Final AL
SD Repeatability	0,080	0,089	YES	+/- 0,500



Sample Name	Reference Central value	Bias	Lower $\beta$ -ETI	Upper $\beta$ -ETI	$\beta$ -ETI compared to AL=±0.5 Acceptable	$\beta$ -ETI compared to final AL Acceptable
2.20	0.051	-0.099	0.201		YES	YES
2.26	0.023	-0.127	0.174		YES	YES
3.23	0.000	-0.150	0.150		YES	YES
3.28	-0.133	-0.283	0.018		YES	YES
4.60	0.114	-0.036	0.264		YES	YES
4.69	0.058	-0.092	0.208		YES	YES

	Reference method	Alternative method	SD repeatability of reference method $\leq 0.125$	Final AL
SD Repeatability	0,085	0,104	YES	+/- 0,500





Sample Name	Reference Central value	Bias	Lower $\beta$ -ETI	Upper $\beta$ -ETI	$\beta$ -ETI compared to AL $\leq \pm 0.5$ Acceptable	$\beta$ -ETI compared to final AL Acceptable
2.34		-0.112	-0.253	0.030	YES	YES
2.36		-0.216	-0.357	-0.074	YES	YES
3.40		-0.167	-0.309	-0.026	YES	YES
3.38		-0.204	-0.346	-0.063	YES	YES
5.04		-0.107	-0.248	0.035	YES	YES
5.08		-0.176	-0.318	-0.035	YES	YES

	Reference method	Alternative method	SD repeatability of reference method $\leq 0.125$	Final AL
SD Repeatability	0.093	0.098	YES	$\pm 0.500$

The tolerance intervals fall into the acceptability limits for all categories tested.

### 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  $\beta$ -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* ( $\beta$ -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.

- 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* 2 method.

- 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 for all food categories and environmental samples category.

The accuracy profile study illustrates that the performances of the alternative method are comparable to those of the reference method for all food categories and environmental samples category.

The alternative method is specific and selective.

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

## 4. Interlaboratory study

### 4.1. Study organization

The interlaboratory study has been realized in October 2004. 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 at 37°C.

The strain used for the contamination of the samples was a  $\beta$ -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	47
Level 2 ( $L_2$ )	500	460
Level 3 ( $L_3$ )	5000	5800

#### 4.2.2. Temperatures during shipping and at reception, delivery times

Among the 16 collaborators, 5 didn't realize the analyses (A, D, I, J, N).

The temperatures recorded at reception of the packages for the 11 remaining collaborators confirm that the samples were stored at correct temperatures (between 0.5°C and 5.4°C).

These 11 collaborators realized the analyses the day following the sending of the samples, except for collaborator G which realized the analyses 48 hours after reception.

#### 4.2.3. Conclusion

Among the 16 collaborators, only 11 realized the analyses.

The results obtained by the collaborator G were not taken into account because it realized the analyses 48 hours after reception of the samples.

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

### 4.3. Results

#### 4.3.1. Expert laboratory

The results obtained by the expert laboratory are shown in the table below.

Table 7: Results of the Expert Laboratory (in CFU/ml)

Level	Reference method		Alternative method	
	R1	R2	R1	R2
$L_0$	<1	<1	<1	<1
$L_1$	60	55	49	42
$L_2$	510	460	440	380
$L_3$	4800	5700	3500	3500

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

#### 4.3.2. Results obtained by the collaborators

The results of the 10 laboratories which realized the analysis are summarized in the tables 8 and 9 for the three levels of contamination.

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

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
B	<1	<1	<1	<1	48	51	49	40	440	460	530	540	4300	5000	5200	5200
C	<1	<1	<1	<1	58	49	53	59	720	710	600	650	9500	6800	5200	7200
E	<1	<1	<1	<1	52	65	63	55	520	530	640	560	6800	6400	5200	6000
F	<1	<1	<1	<1	45	43	55	66	430	440	470	580	3800	3400	5200	4800
H	<1	<1	<1	<1	26	48	52	55	560	480	610	790	6000	4100	7400	5200
K	<1	<1	<1	<1	48	55	55	55	310	420	560	500	3100	5500	5700	5500
L	<1	<1	<1	<1	41	20	42	90	360	470	390	420	4600	5300	5200	5500
M	<1	<1	<1	<1	22	30	43	50	290	250	480	470	6600	4300	6400	5300
O	<1	<1	<1	<1	32	20	41	65	440	360	440	520	4900	5200	5000	6800
P	<1	<1	<1	<1	54	45	49	43	670	610	420	390	5900	7800	5700	5800

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

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	R1	R1	R1	R1	R2	R1	R2	R1	R2	R1	R2	R1	R2	R1	R2
B	0	0	0	0	1,681	1,708	1,690	1,602	2,643	2,663	2,724	2,732	3,633	3,699	3,716	3,716
C	0	0	0	0	1,763	1,690	1,724	1,771	2,857	2,851	2,778	2,813	3,978	3,833	3,716	3,857
E	0	0	0	0	1,716	1,813	1,799	1,740	2,716	2,724	2,806	2,748	3,833	3,806	3,716	3,778
F	0	0	0	0	1,653	1,633	1,740	1,820	2,633	2,643	2,672	2,763	3,580	3,531	3,716	3,681
H	0	0	0	0	1,415	1,681	1,716	1,740	2,748	2,681	2,785	2,898	3,778	3,613	3,869	3,716
K	0	0	0	0	1,681	1,740	1,740	1,740	2,491	2,623	2,748	2,699	3,491	3,740	3,756	3,740
L	0	0	0	0	1,613	1,301	1,623	1,954	2,556	2,672	2,591	2,623	3,663	3,724	3,716	3,740
M	0	0	0	0	1,342	1,477	1,633	1,699	2,462	2,398	2,681	2,672	3,820	3,633	3,806	3,724
O	0	0	0	0	1,505	1,301	1,613	1,813	2,643	2,556	2,643	2,716	3,690	3,716	3,699	3,833
P	0	0	0	0	1,732	1,653	1,690	1,633	2,826	2,785	2,623	2,591	3,771	3,892	3,756	3,763

### 4.3.3. Conclusion

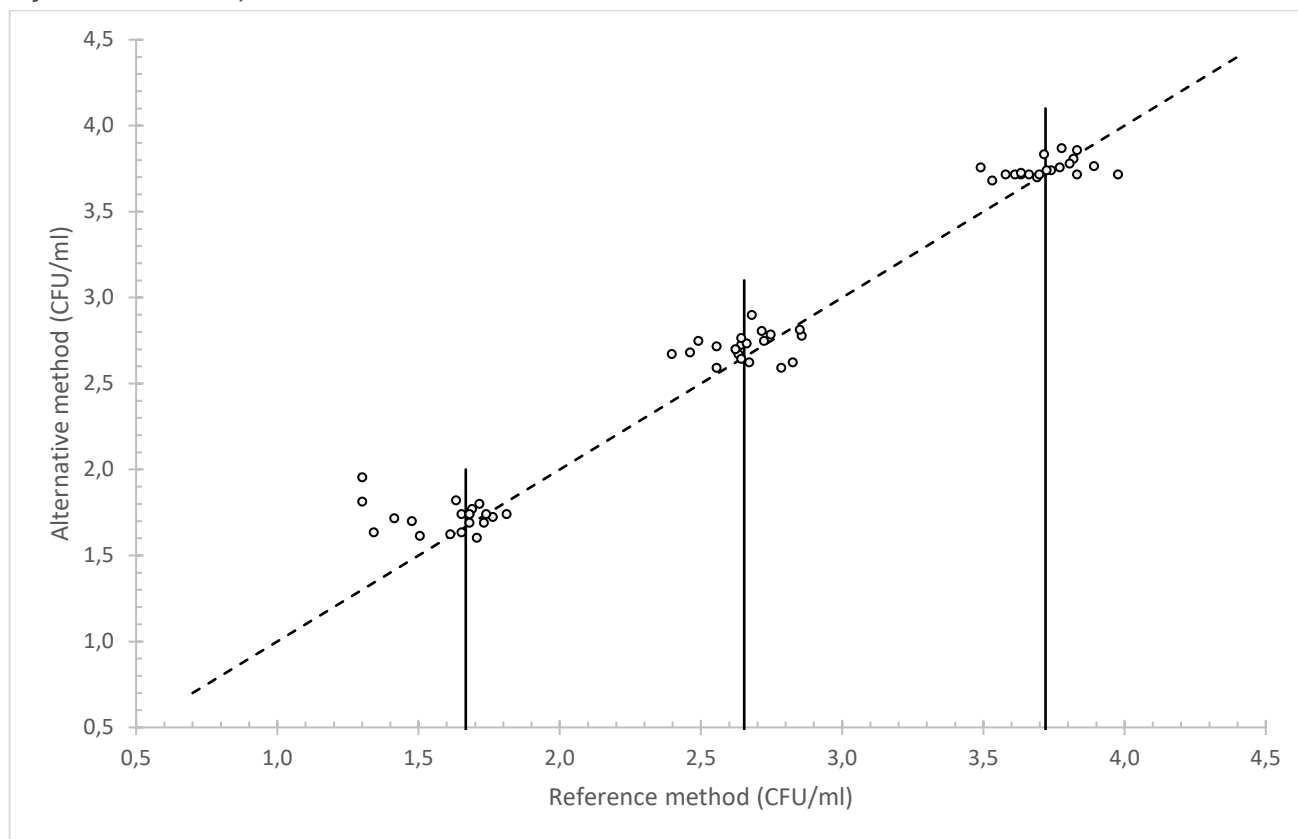
The results obtained by the collaborators were in agreement with those of the Expert Laboratory. The statistical interpretation has been realized with the results of 10 laboratories.

## 4.4. Statistical interpretations and calculations

### 4.4.1. Visual linearity checking

After the log<sub>10</sub> 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 5).

Figure 5: 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>.

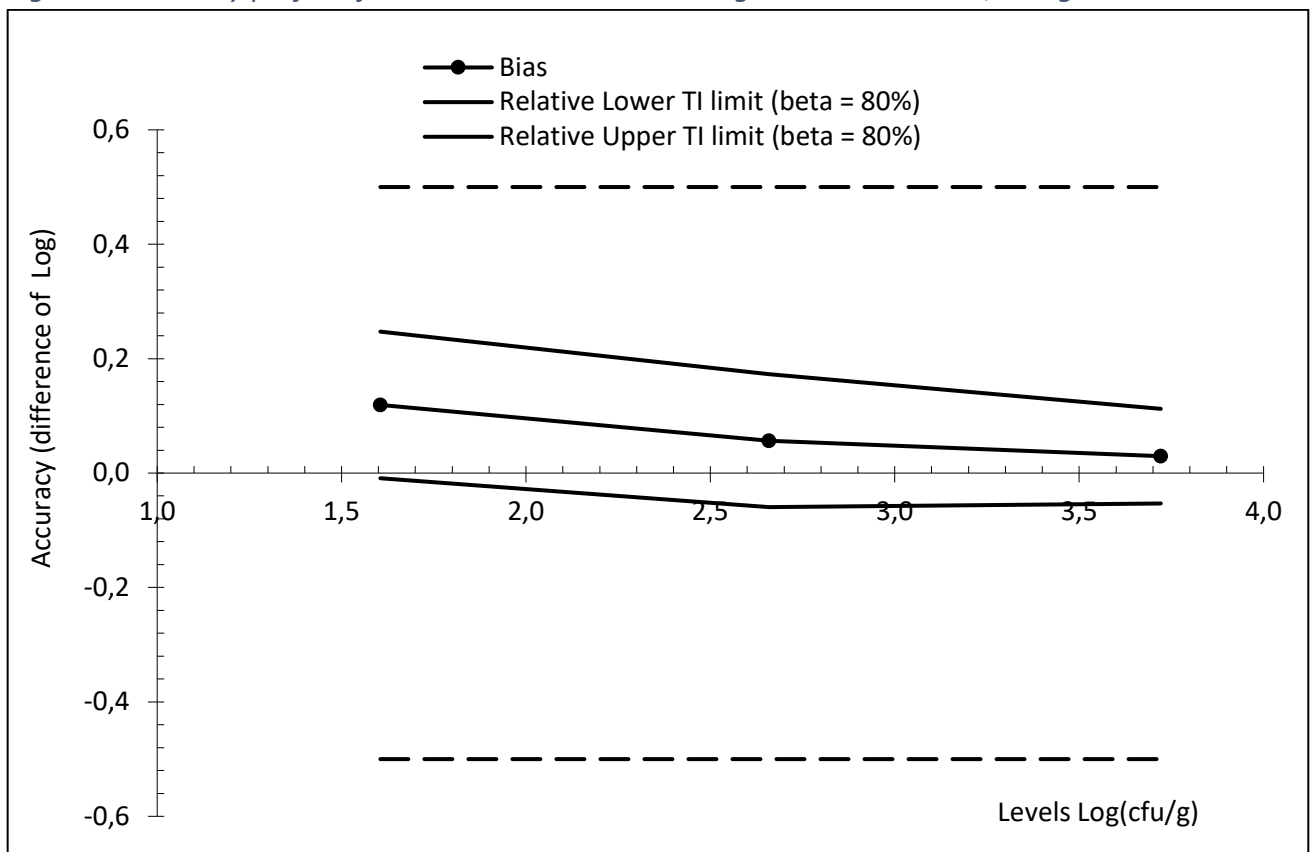
The results of the calculations are provided in table 10.

The graphical representation of the accuracy profile lies in figure 6.

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			
Levels	Alternative method			Reference method			
	Low	Medium	High	Low	Medium	High	
<b>Target value</b>	<b>1,605</b>	<b>2,659</b>	<b>3,721</b>				
Number of participants (K)	10	10	10	10	10	10	
Average for alternative method	1,724	2,715	3,751	1,605	2,659	3,721	
Repeatability standard deviation (sr)	0,094	0,042	0,061	0,113	0,050	0,093	
Between-labs standard deviation (sL)	0,000	0,070	0,000	0,113	0,117	0,082	
Reproducibility standard deviation (sR)	0,094	0,082	0,061	0,160	0,127	0,124	
Corrected number of dof	18,947	11,694	18,947	14,503	10,473	15,296	
Coverage factor	1,361	1,416	1,361				
Interpolated Student t	1,328	1,358	1,328				
Tolerance interval standard deviation	0,0965	0,0855	0,0623				
Lower TI limit	1,596	2,599	3,668				
Upper TI limit	1,852	2,832	3,834				
<b>Bias</b>	<b>0,119</b>	<b>0,057</b>	<b>0,030</b>				
<b>Relative Lower TI limit (beta = 80%)</b>	<b>-0,009</b>	<b>-0,059</b>	<b>-0,053</b>				
<b>Relative Upper TI limit (beta = 80%)</b>	<b>0,247</b>	<b>0,173</b>	<b>0,112</b>				
<b>Lower Acceptability Limit</b>	<b>-0,50</b>	<b>-0,50</b>	<b>-0,50</b>				
<b>Upper Acceptability Limit</b>	<b>0,50</b>	<b>0,50</b>	<b>0,50</b>				
Pooled repro standard dev of reference	0,138						

Figure 6: Accuracy profile of the alternative method using  $\beta = 80\%$  and  $\lambda = 0,50 \log_{10}$  unit



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 ( $\pm 0.5$  log CFU/ml).

The alternative method is regarded as being equivalent to the reference method.



## 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 for all food categories, environmental samples and animal feed.

Le Lion d'Angers, October 15, 2024.

Guillaume MESNARD  
Technical deputy manager



François Le Nestour  
Head of the Microbiology Department



## **APPENDICES**

## **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  $37\pm 1^{\circ}\text{C}$  for  $21\pm 3$  h



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

**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  $44\pm 1^\circ\text{C}$  for  $21\pm 3$  h  
If the presence of stressed cells is suspected, incubation for an initial period of 4 h at  $37^\circ\text{C}$   
and then incubation at  $44^\circ\text{C}$  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

**APPENDIX C - Artificial contaminations**

N° sample	Analysis date	Sample	Artificial contamination				
			Strain	Code	Origin	Injury protocol	Mesured stress
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)	<i>Escherichia coli</i>	ESC.1.33	Shrimp	Seeding : 4 °C/ 72h	/
ISHA 29-2	ISHA 2017	Cod Brandade (CA)	<i>Escherichia coli</i>	ESC.1.33	Shrimp	Seeding : 4 °C/ 72h	/
ISHA 31	ISHA 2017	Smoked Salmon (CA)	<i>Escherichia coli</i>	ESC.1.33	Shrimp	Seeding : 4 °C/ 72h	/
ISHA 32	ISHA 2017	Parmentier of duck (CA)	<i>Escherichia coli</i>	ESC.1.9	Ground beef 15 % fat	Seeding : 4 °C/ 72h	/
ISHA 33	ISHA 2017	"Croque monsieur" (CA)	<i>Escherichia coli</i>	ESC.1.6	Chicken ravioli	Seeding : 4 °C/ 72h	/
ISHA 34	ISHA 2017	Omelet (CA)	<i>Escherichia coli</i>	ESC.1.5	Camembert	Seeding : 4 °C/ 72h	/
ISHA 35	ISHA 2017	Smoked Chicken (CA)	<i>Escherichia coli</i>	ESC.1.9	Ground beef 15 % fat	Seeding : 4 °C/ 72h	/
ISHA 36	ISHA 2017	Smoked Veal (CA)	<i>Escherichia coli</i>	ESC.1.10	Marinated chicken	Seeding : 4 °C/ 72h	/
ISHA 37	ISHA 2017	Gratin (CA)	<i>Escherichia coli</i>	ESC.1.13	Stuffed croissant	Seeding : 4 °C/ 72h	/
ISHA 38	ISHA 2017	Smoked herring fillet (CA)	<i>Escherichia coli</i>	ESC.1.18	Smoked salmon rilette	Seeding : 4 °C/ 72h	/
ISHA 39	ISHA 2017	Smoked herring fillet (CA)	<i>Escherichia coli</i>	ESC.1.18	Smoked salmon rilette	Seeding : 4 °C/ 72h	/
ISHA 40	ISHA 2017	Egg Cream (CA)	<i>Escherichia coli</i>	ESC.1.5	Camembert	Seeding : 4 °C/ 72h	/
ISHA 42	ISHA 2017	Goat cheese (CA)	<i>Escherichia coli</i>	ESC.1.5	Camembert	Seeding : 4 °C/ 72h	/
ISHA 43	ISHA 2017	Emmental (CA)	<i>Escherichia coli</i>	ESC.1.5	Camembert	Seeding : 4 °C/ 72h	/
ISHA 44	ISHA 2017	Haddock fillet (CA)	<i>Escherichia coli</i>	ESC.1.18	Smoked salmon rilette	Seeding : 4 °C/ 72h	/
ISHA 45	ISHA 2017	Lingue fillet (CA)	<i>Escherichia coli</i>	ESC.1.18	Smoked salmon rilette	Seeding : 4 °C/ 72h	/
ISHA 46	ISHA 2017	Marinated mackerel (CA)	<i>Escherichia coli</i>	ESC.1.18	Smoked salmon rilette	Seeding : 4 °C/ 72h	/
ISHA 47	ISHA 2017	Smoked trout (CA)	<i>Escherichia coli</i>	ESC.1.18	Smoked salmon rilette	Seeding : 4 °C/ 72h	/
ISHA 48	ISHA 2017	Smoked Salmon (CA)	<i>Escherichia coli</i>	ESC.1.33	Shrimp	Seeding : 4 °C/ 72h	/
ISHA 49	ISHA 2017	Carpaccio (CA)	<i>Escherichia coli</i>	ESC.1.10	Marinated chicken	Seeding : 4 °C/ 72h	/
ISHA 50	ISHA 2017	Carpaccio (CA)	<i>Escherichia coli</i>	ESC.1.10	Marinated chicken	Seeding : 4 °C/ 72h	/
ISHA 51	ISHA 2017	Smoked mackerel (CA)	<i>Escherichia coli</i>	ESC.1.33	Shrimp	Seeding : 4 °C/ 72h	/
ISHA 52	ISHA 2017	Quiche Lorraine (CA)	<i>Escherichia coli</i>	ESC.1.6	Chicken ravioli	Seeding : 4 °C/ 72h	/
ISHA 54	ISHA 2017	Salad Mix (CA)	<i>Escherichia coli</i>	ESC.1.2	Grated carrot	Seeding : 4 °C/ 72h	/
ISHA 55	ISHA 2017	Grated carrots (CA)	<i>Escherichia coli</i>	ESC.1.2	Grated carrot	Seeding : 4 °C/ 72h	/
ISHA 56	ISHA 2017	Salad Bag (CA)	<i>Escherichia coli</i>	ESC.1.2	Grated carrot	Seeding : 4 °C/ 72h	/
ISHA 57	ISHA 2017	"Poêlée" of vegetables (CA)	<i>Escherichia coli</i>	ESC.1.2	Grated carrot	Seeding : 4 °C/ 72h	/
ISHA 58	ISHA 2017	Endive with Ham (CA)	<i>Escherichia coli</i>	ESC.1.6	Chicken ravioli	Seeding : 4 °C/ 72h	/
ISHA 59	ISHA 2017	Lasagna 3 meats (CA)	<i>Escherichia coli</i>	ESC.1.10	Marinated chicken	Seeding : 4 °C/ 72h	/
ISHA 60	ISHA 2017	Chicken Peas (CA)	<i>Escherichia coli</i>	ESC.1.13	Stuffed croissant	Seeding : 4 °C/ 72h	/
ISHA 61	ISHA 2017	Grilled Sausage (CA)	<i>Escherichia coli</i>	ESC.1.11	Chicken breast	Seeding : 4 °C/ 72h	/
ISHA 62	ISHA 2017	Parmentier of ham (CA)	<i>Escherichia coli</i>	ESC.1.11	Chicken breast	Seeding : 4 °C/ 72h	/
ISHA-63	ISHA 2017	Cod with sorrel sauce (CA)	<i>Escherichia coli</i>	ESC.1.31	Scallops	Seeding : 4 °C/ 72h	/
ISHA-64	ISHA 2017	Cod with tomato sauce (CA)	<i>Escherichia coli</i>	ESC.1.31	Scallops	Seeding : 4 °C/ 72h	/
ISHA-65	ISHA 2017	Salmon with sorrel sauce (CA)	<i>Escherichia coli</i>	ESC.1.31	Scallops	Seeding : 4 °C/ 72h	/
ISHA 66	ISHA 2017	Cucumber (CA)	<i>Escherichia coli</i>	ESC.1.2	Grated carrot	Seeding : 4 °C/ 72h	/
ISHA 67	ISHA 2017	Tomato (CA)	<i>Escherichia coli</i>	ESC.1.2	Grated carrot	Seeding : 4 °C/ 72h	/
ISHA 68	ISHA 2017	Apple (CA)	<i>Escherichia coli</i>	ESC.1.147	Cucumber	Seeding : 4 °C/ 72h	/
ISHA 69	ISHA 2017	Grape (CA)	<i>Escherichia coli</i>	ESC.1.147	Cucumber	Seeding : 4 °C/ 72h	/
ISHA 70	ISHA 2017	Red cabbage (CA)	<i>Escherichia coli</i>	ESC.1.147	Cucumber	Seeding : 4 °C/ 72h	/
ISHA 71	ISHA 2017	Basil Condiment (CA)	<i>Escherichia coli</i>	ESC.1.147	Cucumber	Seeding : 4 °C/ 72h	/
ISHA 71	ISHA 2017	Tabbouleh (CA)	<i>Escherichia coli</i>	ESC.1.6	Chicken ravioli	Seeding : 4 °C/ 72h	/
ISHA 72	ISHA 2017	Apple pie (CA)	<i>Escherichia coli</i>	ESC.1.17	Chicken ravioli	Seeding : 4 °C/ 72h	/
ISHA 73	ISHA 2017	Smoked breast of duck (CA)	<i>Escherichia coli</i>	ESC.1.17	Chicken ravioli	Seeding : 4 °C/ 72h	/
ISHA 74	ISHA 2017	Beef carpaccio (CA)	<i>Escherichia coli</i>	ESC.1.17	Chicken ravioli	Seeding : 4 °C/ 72h	/
ISHA 75	ISHA 2017	Beef marinated (CA)	<i>Escherichia coli</i>	ESC.1.17	Chicken ravioli	Seeding : 4 °C/ 72h	/
2347747	Ext 2022	Wipe cheese cold room shelf	<i>Enterobacter pyrinus</i> + <i>E. coli</i>	JLE589 + CJG405	Environment + Sage	Seeding : 4 °C/ 72h	/
2347748	Ext 2022	Wipe refrigerated delicatessen display shelf	<i>Klebsiella oxytoca</i> + <i>E. coli</i>	UNG158 + FRAU26	Environment + Organic thyme	Seeding : 4 °C/ 72h	/
2347750	Ext 2022	Wipe seafood working plan	<i>Escherichia coli</i>	FRAU26	Organic thyme	Seeding : 4 °C/ 72h	/
2347751	Ext 2022	Wipe refrigerated pastry display shelf	<i>Escherichia coli</i>	DMX101	Seafood galet	Seeding : 4 °C/ 72h	/
2347770	Ext 2022	Wipe on ventilation grid for refrigerated showcase butcher	<i>Klebsiella oxytoca</i> + <i>E. coli</i>	UNG158 + FRAU26	Environment + Organic thyme	Seeding : 4 °C/ 72h	/
2347771	Ext 2022	Wipe workplan delicatessen preparation	<i>Enterobacter pyrinus</i> + <i>E. coli</i>	JLE580 + CJG405	Environment + Sage	Seeding : 4 °C/ 72h	/
2347772	Ext 2022	Wipe workplan butcher	<i>Enterobacter pyrinus</i> + <i>E. coli</i>	JLE580 + DMX101	Environment + Seafood galet	Seeding : 4 °C/ 72h	/
2347753	Ext 2022	Process water cheese industry	<i>Hafnia alvei</i> + <i>E. coli</i>	SRZ676 + YMJ695	Main water + Main water	Seeding : 4 °C/ 72h	/
2347756	Ext 2022	Process water pastry industry	<i>Escherichia coli</i>	YMJ695	Main water	Seeding : 4 °C/ 72h	/
2347757	Ext 2022	Process water vegetables	<i>Escherichia coli</i>	DNS677	Main water	Seeding : 4 °C/ 72h	/
2347758	Ext 2022	Process water fish shop	<i>Escherichia coli</i>	DNS677	Main water	Seeding : 4 °C/ 72h	/
2347761	Ext 2022	Flour dust	<i>Escherichia coli</i>	FBV114	Dairy environment	Seeding : 4 °C/ 72h	/
2347762	Ext 2022	Dust milk industry	<i>Escherichia coli</i>	FQN709	Dairy powder environment	Seeding : 4 °C/ 72h	/
2347763	Ext 2022	Dust egg product industry	<i>Escherichia coli</i>	HMLZ85	Mill environment	Seeding : 4 °C/ 72h	/
2364266	Ext 2023	Salmon cat kibble	<i>E. coli</i> + <i>Crono Sakazakii</i>	AZD018 + AKUY55	Fish + Cat food	Spiking 10 minutes at 56°C + Spiking 10 minutes at 56°C	1.48 / 1.52
2364269	Ext 2023	Rabbit pellets	<i>E. coli</i>	HMLZ85	Mill environment	Spiking 15 minutes at 56°C	2.09
2364270	Ext 2023	Dog kibble	<i>E. coli</i>	BHXK78	Organic chicken liver	Spiking 15 minutes at 56°C	0.52
2364267	Ext 2023	Terrine dog poultry	<i>E. coli</i> + <i>Enterobacter cloacae</i>	GQRP82 + HWJ654	Turkey + Dairy environment	Seeding : 4 °C/ 72h	/
2364268	Ext 2023	Beef Cat Pate	<i>E. coli</i> + <i>Citrobacter youngae</i>	BHXK78 + RAX819A	Organic chicken liver + Minced beef	Seeding : 4 °C/ 72h	/
2392504	Ext 2023	Beef Cat Pate	<i>E. coli</i> + <i>Serratia marcescens</i>	GQRP82 + BJK3652	Turkey + Food	Seeding : 4 °C/ 72h	/
2392518	Ext 2023	Cat kibble with salmon	<i>Escherichia coli</i>	AZD018	Fish	Spiking 8 minutes at 56°C	0.84
2392519	Ext 2023	Beef dog kibble	<i>Escherichia coli</i>	BHXK78	Organic chicken liver	Spiking 10 minutes at 56°C	0.90

**APPENDIX C - Artificial contaminations**

N° sample	Analysis date	Sample	Artificial contamination				
			Strain	Code	Origin	Injury protocol	Mesured stress
2392520	Ext 2023	Rabbit pellets	<i>Escherichia coli</i>	HMLZ85	Mill environment	Spiking 5 minutes at 56°C	0.72
2392586	Ext 2023	Cat kibble with turkey	<i>Escherichia coli</i>	TZP821	Hummus + beets	Spiking 8 minutes at 56°C	1.31
2392587	Ext 2023	Cat kibble without cereals	<i>Escherichia coli</i>	FLEL39	Duck liver	Spiking 10 minutes at 56°C	2.38
2364274	Ext 2023	Ewes pellets	<i>Escherichia coli</i>	HMLZ85	Mill environment	Spiking 15 minutes at 56°C	1.08
2364277	Ext 2023	Soy Cattle cake	<i>Escherichia coli</i>	HMLZ85	Mill environment	Spiking 15 minutes at 56°C	1.08
2364279	Ext 2023	Mixed wheat	<i>Escherichia coli</i>	FRAU26	Plants	Spiking 10 minutes at 56°C	1.07
2392588	Ext 2023	Soy	<i>Escherichia coli</i>	TZP821	Hummus + beets	Spiking 8 minutes at 56°C	1.31
2392589	Ext 2023	Corn	<i>E.coli</i> + <i>Enterobacter cloacae</i>	TZP821 + EBJ453	Hummus + beets + Chicken	Spiking 8 minutes at 56°C + Spiking 10 minutes at 56°C	1.3 / 1.33
2392590	Ext 2023	Wheat	<i>E.coli</i> + <i>Enterobacter cloacae</i>	FLEL39 + EBJ453	Duck liver + Chicken	Spiking 10 minutes at 56°C + Spiking 10 minutes at 56°C	2.38 / 1.33
2392591	Ext 2023	Wheat	<i>E.coli</i> + <i>Enterobacter cloacae</i>	TZP821 + EBJ453	Hummus + beets + Chicken	Spiking 8 minutes at 56°C + Spiking 10 minutes at 56°C	1.31 / 1.33
2392481	Ext 2023	Chicken flour batch 1	<i>E.coli</i>	BHXK78	Organic chicken liver	Spiking 15 minutes at 56°C	1.21
2392485	Ext 2023	Salmon flour	<i>E.coli</i> + <i>Hafnia alvei</i>	AZD018 + HVC355	Fish + Environment	Spiking 15 minutes at 56°C + Spiking 10 minutes at 56°C	0.73 / 0.43
2392488	Ext 2023	Soy protein	<i>Escherichia coli</i>	FRAU26	Plants	Spiking 15 minutes at 56°C	1.07
2392522	Ext 2023	Chicken flour batch 2	<i>Escherichia coli</i>	BHXK78	Organic chicken liver	Spiking 10 minutes at 56°C	0.90
2392523	Ext 2023	Wheat fiber	<i>Escherichia coli</i>	HMLZ85	Mill environment	Spiking 5 minutes at 56°C	0.72
2392525	Ext 2023	Salmon flour	<i>Escherichia coli</i>	AZD018	Fish	Spiking 8 minutes at 56°C	0.84
2319051	Ext 2023	Beef flour	<i>Escherichia coli</i>	GQRP82	Turkey	Spiking 8 minutes at 56°C	0.63
2392603	Ext.2023	Iberian cured ham	<i>Escherichia coli</i>	GAR051	Delicatessen	Seeding : 4 °C/ 72h	/
2392604	Ext.2023	Smoked bacon	<i>Escherichia coli</i>	GAR051	Delicatessen	Seeding : 4 °C/ 72h	/
2392605	Ext.2023	Smoked ham	<i>Escherichia coli</i>	WTG304	Delicatessen	Seeding : 4 °C/ 72h	/
2392606	Ext.2023	Dry ham	<i>Escherichia coli</i>	WTG304	Delicatessen	Seeding : 4 °C/ 72h	/
2392607	Ext.2023	Smoked bacon	<i>Escherichia coli</i>	WTG304	Delicatessen	Seeding : 4 °C/ 72h	/
2392602	Ext.2023	Texas sandwich	<i>Escherichia coli</i>	UBS981	Croissant with ham	Seeding : 4 °C/ 72h	/

/: no information available

## **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

**Meat products**

Type	#	Study	Sample	Reference method: NF ISO 16649-2			Alternative method: REC2 37°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	/	/	ND
a	2	IVS	Ground beef	/	/	/	<10	/	/	<10
a	11	IVS	Leg of lamb	/	/	/	<10	/	/	<10
a	14	IVS	Turkey cutlet	/	/	/	130	/	/	ND
a	26	IVS	Turkey steak	/	/	/	10	/	/	<10
a	27	IVS	Chicken fillet	/	/	/	10	/	/	ND
a	28	IVS	Chicken fillet	/	/	/	5	/	/	<10
a	29	IVS	Beef kidneys	/	/	/	70	/	/	60
a	31	IVS	Sausage	/	/	/	30	/	/	40
a	32	IVS	Turkey cutlet	/	/	/	830	/	/	830
a	33	IVS	Chopped steak	/	/	/	60	/	/	60
a	35	IVS	Veal cutlet	/	/	/	200	/	/	400
a	38	IVS	Veal cutlet	/	/	/	<10	/	/	<10
a	39	IVS	Turkey steak	/	/	/	15	/	/	20
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	Ground beef	10	23	13	180	10	12	120
				100	3	0		100	1	
a	ISHA 25	R2017	Meatball	10	0	0	<10	10	0	<10
				100	0	0		100	0	
a	34	IVS	Chipolata	/	/	/	960	/	/	850
a	3	IVS	Merguez	/	/	/	120	/	/	180
a	4	IVS	Veal Sausage	/	/	/	150	/	/	210
a	6	IVS	Herb Sausage	/	/	/	380	/	/	520
a	13	IVS	Merguez	/	/	/	15	/	/	ND
b	12	IVS	Blanquette of turkey	/	/	/	<10	/	/	<10
b	ISHA 2	R2017	Poultry bites	100	30	29	2900	100	40	4300
				1000	3	1		1000	7	
b	ISHA 30	R2017	Ravioli of poultry	100	30	38	3200	100	37	3700
				1000	2	1		1000	4	
b	ISHA 32	R2017	Parmentier of duck (CA)	10	34	38	370	10	39	370
				100	5	5		100	2	
b	ISHA 59	R2017	Lasagna 3 meats (CA)	100	19	19	1800	100	16	1500
				1000	1	1		1000	1	
b	ISHA 61	R2017	Grilled Sausage (CA)	100	14	12	1400	100	15	1400
				1000	2	2		1000	0	
b	ISHA 62	R2017	Parmentier of ham (CA)	100	17	11	1400	100	10	1100
				1000	1	1		1000	2	
c	ISHA 35	R2017	Smoked chicken (CA)	100	16	16	1600	100	14	1400
				1000	3	0		1000	1	
c	ISHA 36	R2017	Smoked veal (CA)	1000	0	0	<1000	1000	2	2000
				10000	0	0		10000	0	



**Meat products**

Type	#	Study	Sample	Reference method: NF ISO 16649-2*			Alternative method: REC2 37°C			Alternative method: REC2 after storage 72h at 2-8°C		
				Dilution	CFU/ plate	Result (CFU/g)	Dilution	CFU/plate	Result (CFU/g)	Dilution	CFU/plate	Result (CFU/g)
c	2392603	Ext.2023	Iberian cured ham	-1	5	50	-1	7	70	-1	7	70
				-2	0		-2	1		-2	1	
c	2392604	Ext.2023	Smoked bacon	-1	5	50	-1	11	110	-1	11	110
				-2	0		-2	1		-2	1	
c	2392605	Ext.2023	Smoked ham	-1	18	200	-1	13	130	-1	13	130
				-2	4		-2	0		-2	0	
c	2392606	Ext.2023	Dry ham	-1	27	260	-1	31	310	-1	31	310
				-2	2		-2	3		-2	3	
c	2392607	Ext.2023	Smoked bacon	-1	30	280	-1	34	320	-1	34	320
				-2	1		-2	1		-2	1	

**Dairy and egg products**

Type	#	Study	Sample	Reference method: NF ISO 16649-2			Alternative method: REC2 37°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	23	40	310	10	22	220
a	ISHA 19	R2017	Pasteurized milk brie	10	23	17	190	10	15	160
a	ISHA 20	R2017	Pasteurized milk brie	10	25	31	310	10	36	360
a	ISHA 42	R2017	Goat cheese (CA)	10	18	20	190	10	32	310
a	ISHA 43	R2017	Emmental (CA)	10	17	22	190	10	19	180
a	ISHA 41	R2017	Fresh cream	10	0	0	<10	10	0	<10
b	L1	IVS	Raw milk	/	/	/	<1	/	/	<1
b	L2	IVS	Raw milk	/	/	/	<1	/	/	<1
b	5	IVS	Raw milk	/	/	/	<1	/	/	3
b	10	IVS	Reblochon	/	/	/	870	/	/	1300
b	24	IVS	Reblochon	/	/	/	40	/	/	30
b	25	IVS	Roquefort	/	/	/	210	/	/	560
b	36	IVS	Raw milk cheese	/	/	/	20000	/	/	15000
b	59	IVS	Raw milk cheese	/	/	/	2800	/	/	3600
b	78	IVS	Reblochon	/	/	/	6500	/	/	5300
b	90	IVS	Raw milk cheese (CA)	/	/	/	1600	/	/	2500
c	44	IVS	Strawberry cream	/	/	/	<10	/	/	<10
c	96	IVS	Chocolate cake	/	/	/	12000	/	/	12000
c	95	IVS	Versailles	/	/	/	23000	/	/	26000
c	93	IVS	Cream puffs	/	/	/	180	/	/	150
c	70	IVS	Coffee éclair	/	/	/	140	/	/	190
c	40	IVS	Banana cream	/	/	/	70	/	/	80
c	20	IVS	Coffee éclair	/	/	/	90	/	/	45
c	72	IVS	Peach cake	/	/	/	4000	/	/	1800
c	94	IVS	Apple pie	/	/	/	1770	/	/	1600
c	60	IVS	Apricot cake	/	/	/	290	/	/	280
c	ISHA 21	R2017	Almond cake	10	158	126	1400	10	137	1400
c	ISHA 40	R2017	Egg cream (CA)	10	10	15	130	10	18	170
c	17	IVS	Savarin with chocolate	/	/	/	5	/	/	10
c	18	IVS	"Eclair" with pastry cream	/	/	/	<10	/	/	<10
c	23	IVS	Chocolate Cream	/	/	/	<10	/	/	<10

**Dairy and egg products**

Type	#	Study	Sample	Reference method: NF ISO 16649-2			Alternative method: REC2 37°C			
				Dilution	CFU/ plate	CFU/ plate	Result (CFU/g or mL)	Dilution	CFU/plate	Result (CFU/g or mL)
c	46	IVS	Cream puffs	/	/	/	<10	/	/	<10
				/	/	/		/	/	
c	75	IVS	Cream puffs	/	/	/	<10	/	/	<10
				/	/	/		/	/	
c	76	IVS	Cream puffs	/	/	/	<10	/	/	<10
				/	/	/		/	/	
c	51	IVS	Apple pie	/	/	/	<10	/	/	<10
				/	/	/		/	/	
c	ISHA 72	R2017	Apple pie (CA)	10	26	31	290	10	29	290
				100	4	2		100	3	
c	ISHA 34	R2017	Omelet (CA)	10	2	1	20	10	3	30
				100	0	0		100	0	

**Seafood products**

Type	#	Study	Sample	Reference method: NF ISO 16649-2			Alternative method: REC2 37°C			
				Dilution	CFU/ plate	CFU/ plate	Result (CFU/g or mL)	Dilution	CFU/plate	Result (CFU/g or mL)
a	7	IVS	Fish	/	/	/	<10	/	/	<10
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	/	/	30000
a	53	IVS	Salmon Steak	/	/	/	29000	/	/	30000
a	54	IVS	Fish	/	/	/	290000	/	/	180000
a	80	IVS	Cod fillet	/	/	/	ND	/	/	ND
a	ISHA 45	R2017	Lingue fillet (CA)	1000	6	5	5500	1000	4	4000
				10000	1	2		10000	3	
a	ISHA 7	R2017	Julienne fillet	10	0	0	<10	10	0	<10
				100	0	0		100	0	
a	ISHA 10	R2017	Raw Salmon	10	0	0	<10	10	0	<10
				100	0	0		100	0	
a	ISHA 11	R2017	Raw Salmon	10	0	0	<10	10	0	<10
				100	0	0		100	0	
a	ISHA 27	R2017	Petoncle Nuts	10	0	0	<10	10	0	<10
				100	0	0		100	0	
a	ISHA 29-1	R2017	Sole fillet	10	0	0	<10	10	0	<10
				100	0	0		100	0	
b	81	IVS	Smoked Marlin Fillet	/	/	/	<10	/	/	<10
b	55	IVS	Smoked salmon	/	/	/	240000	/	/	200000
b	91	IVS	Smoked Salmon (CA)	/	/	/	260	/	/	210
b	92	IVS	Smoked Salmon (CA)	/	/	/	1900	/	/	2600
b	ISHA 17	R2017	Marinated salmon	10	0	0	<10	10	0	<10
				100	0	0		100	0	
b	ISHA 31	R2017	Smoked Salmon (CA)	10	8	5	65	10	14	140
				100	1	2		100	1	
b	ISHA 38	R2017	Smoked herring fillet (CA)	10	9	6	75	10	12	120
				100	0	0		100	1	
b	ISHA 39	R2017	Smoked herring fillet (CA)	10	10	5	70	10	10	120
				100	1	0		100	3	
b	ISHA 47	R2017	Smoked trout (CA)	10	23	35	280	10	23	230
				100	2	1		100	2	
b	ISHA 48	R2017	Smoked Salmon (CA)	10	26	24	260	10	28	320
				100	4	2		100	7	
b	ISHA 51	R2017	Smoked mackerel (CA)	10	46	61	530	10	57	560
				100	6	4		100	4	
b	ISHA 46	R2017	Marinated mackerel (CA)	1000	21	18	19000	1000	14	14000
				10000	2	1		10000	1	
b	79	IVS	Smoked salmon	/	/	/	ND	/	/	ND
b	ISHA 44	R2017	Haddock fillet (CA)	1000	7	6	6500	1000	14	13000
				10000	1	1		10000	0	
c	43	IVS	Cod fillet	/	/	/	<10	/	/	<10
c	61	IVS	Shrimp (CA)	/	/	/	2300	/	/	2300
c	63	IVS	Shrimp	/	/	/	ND	/	/	ND
				/	/	/		/	/	
c	ISHA 29-2	R2017	Cod Brandade (CA)	10	38	56	460	10	55	570
				100	3	5		100	8	
c	ISHA-63	R2017	Cod with sorrel sauce (CA)	10	8	5	65	10	9	90
				100	0	0		100	0	
c	ISHA-64	R2017	Cod with tomato sauce (CA)	10	7	8	75	10	7	70
				100	0	0		100	0	
c	ISHA-65	R2017	Salmon with sorrel sauce (CA)	10	6	11	85	10	8	80
				100	0	0		100	0	
c	ISHA 28	IVS	Cod Accras	10	0	0	<10	10	0	<10
				100	0	0		100	0	

**Seafood products**

Type	#	Study	Sample	Reference method: NF ISO 16649-2*			Alternative method: REC2 37°C			Alternative method: REC2 after storage 72h at 2-8°C		
				Dilution	CFU/ plate	Result (CFU/g)	Dilution	CFU/plate	Result (CFU/g)	Dilution	CFU/plate	Result (CFU/g)
a	2392601	Ext.2023	Whiting fillet	-1	4	40	-1	11	110	-1	11	110
				-2	0		-2	1		-2	1	

### Vegetal products

Type	#	Study	Sample	Reference method: NF ISO 16649-2			Alternative method: REC2 37°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	53	5200
				1000	5	7		1000	4	
a	ISHA 67	R2017	Tomato (CA)	100	87	64	7700	100	89	9300
				1000	9	8		1000	13	
a	ISHA 68	R2017	Apple (CA)	100	18	13	1400	100	16	1600
				1000	0	0		1000	1	
a	ISHA 69	R2017	Grape (CA)	10	11	12	110	10	11	130
				100	0	1		100	3	
a	ISHA 70	R2017	Red cabbage (CA)	10	39	45	430	10	51	520
				100	5	6		100	6	
a	57	IVS	Strawberry	/	/	/	30	/	/	70
				/	/	/		/	/	
b	89	IVS	Grated carrots	/	/	/	1600	/	/	2400
				/	/	/		/	/	
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	<10
				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	39	380
				100	6	7		100	3	
b	ISHA 55	R2017	Grated carrots (CA)	10	41	28	350	10	32	310
				100	4	4		100	2	
b	ISHA 56	R2017	Salad Bag (CA)	10	26	31	280	10	45	460
				100	2	2		100	5	
b	ISHA 57	R2017	"Poêlée" of vegetables (CA)	10	36	43	390	10	45	460
				100	4	3		100	6	
c	50	IVS	Macedonia	/	/	/	65	/	/	45
				/	/	/		/	/	
c	56	IVS	Fruit Compote	/	/	/	660	/	/	580
				/	/	/		/	/	
c	ISHA 12	R2017	Basil Condiment	10	26	11	180	10	23	230
				100	1	1		100	2	
c	ISHA 71	R2017	Basil Condiment (CA)	100	93	89	9200	100	87	8700
				1000	11	10		1000	9	
c	ISHA 6	R2017	Parsnip Purée	10	15	13	130	10	5	50
				100	1	0		100	1	
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	

**RTE-RTRH products**

Type	#	Study	Sample	Reference method: NF ISO 16649-2			Alternative method: REC2 37°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	19	IVS	Pastas salad	/	/	/	5	/	/	<10		
a	71	IVS	Tuna tomatoes	/	/	/	140	/	/	130		
a	ISHA 3	R2017	Gabonese salad	10	6	10	80	10	8	80		
a	ISHA 71	R2017	Tabbouleh (CA)	100	0	1		100	0			
a	ISHA 13	R2017	Tomato salad dressing	10	29	30	280	10	39	380		
a	21	IVS	Tomato salad	100	2	1		100	3			
b	ISHA 33	R2017	"Croque monsieur" (CA)	/	/	/	<10	/	/	<10		
b	ISHA 52	R2017	Quiche Lorraine (CA)	100	11	8	910	100	16	1600		
b	ISHA 53	R2017	Beef vegetables	1000	1	0		1000	2			
b	ISHA 37	R2017	Gratin (CA)	10	17	29	240	10	23	260		
b	ISHA 14	R2017	Salmon Tagliatelle (CA)	100	5	2		100	6			
b	ISHA 24	R2017	Royal noodles	10	24	27	250	10	24	260		
b	ISHA 60	R2017	Chicken Peas (CA)	100	1	3		100	5			
b	ISHA 58	R2017	Endive with Ham (CA)	10	1	0	10	10	2	20		
b	ISHA 74	R2017	Beef carpaccio (CA)	100	1	0	10	100	1	<10		
b	ISHA 75	R2017	Beef marinated (CA)	100	0	0	<10	10	0	<10		
c	ISHA 49	R2017	Carpaccio (CA)	100	0	0	10	100	0	<10		
c	ISHA 50	R2017	Carpaccio (CA)	10	16	19	190	10	28	170		
c	ISHA 26	R2017	Carpaccio	100	4	2		100	2			
c	ISHA 73	R2017	Smoked breast of duck (CA)	10	22	23	220	10	33	310		
c	ISHA 74	R2017	Beef carpaccio (CA)	100	1	2		100	1			
c	ISHA 75	R2017	Beef marinated (CA)	10	0	0	<10	10	0	<10		
c	ISHA 73	R2017	Smoked breast of duck (CA)	100	123	118	13000	100	115	12000		
c	ISHA 74	R2017	Beef carpaccio (CA)	1000	18	21		1000	16			
c	ISHA 75	R2017	Beef marinated (CA)	100	97	104	10000	100	107	11000		
c	ISHA 75	R2017	Beef marinated (CA)	1000	12	9		1000	13			
c	ISHA 75	R2017	Beef marinated (CA)	100	110	103	11000	100	108	11000		
c	ISHA 75	R2017	Beef marinated (CA)	1000	9	9		1000	11			
Type	#	Study	Sample	Reference method: NF ISO 16649-2*			Alternative method: REC2 37°C			Alternative method: REC2 after storage 72h at 2-8°C		
				Dilution	CFU/ plate	Result (CFU/g)	Dilution	CFU/plate	Result (CFU/g)	Dilution	CFU/plate	Result (CFU/g)
a	2392602	Ext.2023	Texas sandwich	-1	28	260	-1	22	210	-1	22	210
				-2	1		-2	1		-2	1	

Env. samples

Type	#	Study	Sample	Conta	Reference method: NF ISO 16649-2*			Alternative method: REC2 37°C			Alternative method: REC2 after storage 72h at 2-8°C		
					Dilution	CFU/ plate	Result (CFU/g or mL)	Dilution	CFU/plate	Result (CFU/g or mL)	Dilution	CFU/plate	Result (CFU/g or mL)
a	2333887	Ext 2022	Swab environment egg product	nc	-1 -2	0 0	<10	-1 -2	0 0	<10	-1 -2	0 0	<10
a	2333888	Ext 2022	Swab environment egg product	nc	-1 -2	0 0	<10	-1 -2	0 0	<10	-1 -2	0 0	<10
a	2347710	Ext 2022	Wipe pastry industry environment	nc	-1 -2	0 0	<10	-1 -2	0 0	<10	-1 -2	0 0	<10
a	2347711	Ext 2022	Wipe butcher environment	nc	-1 -2	0 0	<10	-1 -2	0 0	<10	-1 -2	0 0	<10
a	2347712	Ext 2022	Wipe seafood environment	nc	-1 -2	0 0	<10	-1 -2	0 0	<10	-1 -2	0 0	<10
a	2347713	Ext 2022	Wipe seafood environment	nc	-1 -2	0 0	<10	-1 -2	0 0	<10	-1 -2	0 0	<10
a	2347714	Ext 2022	Wipe butcher environment	nc	-1 -2	0 0	<10	-1 -2	0 0	<10	-1 -2	0 0	<10
a	2347715	Ext 2022	Wipe pastry & bakery	nc	-1 -2	0 0	<10	-1 -2	0 0	<10	-1 -2	0 0	<10
a	2347747	Ext 2022	Wipe cheese cold room shelf	ac/cc	-1 -2	57 9	600	-1 -2	55 8	570	-1 -2	55 8	570
a	2347748	Ext 2022	Wipe refrigerated delicatessen display shelf	ac/cc	-2 -3	6 0	600	-2 -3	8 0	800	-2 -3	8 0	800
a	2347750	Ext 2022	Wipe seafood working plan	ac	-3 -4	41 3	40 000	-3 -4	50 9	54 000	-3 -4	50 9	54 000
a	2347751	Ext 2022	Wipe refrigerated pastry display shelf	ac	-1 -2	0 0	<10	-1 -2	0 0	<10	-1 -2	0 0	<10
a	2347770	Ext 2022	Wipe on ventilation grid for refrigerated showcase butcher	ac	-2 -3	11 1	1100	-2 -3	25 2	2500	-2 -3	25 2	2500
a	2347771	Ext 2022	Wipe workplan delicatessen preparation	ac	-1 -2	26 1	250	-1 -2	43 8	460	-1 -2	43 8	460
a	2347772	Ext 2022	Wipe workplan butcher	ac	-1 -2	19 1	180	-1 -2	21 1	200	-1 -2	21 1	200
b	2333889	Ext 2022	Process water vegetables	nc	-2 -3	0 0	<100	-2 -3	0 0	<100	-2 -3	0 0	<100
b	2333890	Ext 2022	Process water poultry industry	nc	-5 -6	117 13	12 000 000	-5 -6	111 8	11 000 000	-5 -6	111 8	11 000 000
b	2333891	Ext 2022	Process water poultry industry	nc	-5 -6	87 12	9 000 000	-5 -6	118 11	12 000 000	-5 -6	118 11	12 000 000
b	2347753	Ext 2022	Process water cheese industry	ac/cc	-1 -2	32 1	300	-1 -2	37 6	390	-1 -2	37 6	390
b	2347756	Ext 2022	Process water pastry industry	ac	-1 -2	7 0	70	-1 -2	21 1	200	-1 -2	21 1	200
b	2347757	Ext 2022	Process water vegetables	ac	-1 -2	29 4	300	-1 -2	59 11	640	-1 -2	59 11	640
b	2347758	Ext 2022	Process water fish shop	ac	-2 -3	41 2	3900	-2 -3	53 3	5100	-2 -3	53 3	5100
c	2333892	Ext 2022	Pastry residue	nc	-1 -2	0 0	<10	-1 -2	0 0	<10	-1 -2	0 0	<10
c	2333893	Ext 2022	Egg product residue	nc	-1 -2	0 0	<10	-1 -2	0 0	<10	-1 -2	0 0	<10
c	2333894	Ext 2022	Poultry industry residue	nc	-5 -6	19 2	1 900 000	-5 -6	35 7	3 800 000	-5 -6	35 7	3 800 000
c	2333895	Ext 2022	Poultry industry residue	nc	-5 -6	13 1	1 300 000	-5 -6	15 3	1 600 000	-5 -6	15 3	1 600 000
c	2347708	Ext 2022	Cheese industry residue area 2	nc	-1 -2	49 3	470	-1 -2	61 17	710	-1 -2	61 17	710
c	2347709	Ext 2022	Fish industry residue	nc	-1 -2	0 0	<10	-1 -2	0 0	<10	-1 -2	0 0	<10
c	2347761	Ext 2022	Flour dust	nc+ac	-1 -2	5 0	50	-1 -2	6 0	60	-1 -2	6 0	60
c	2347762	Ext 2022	Dust milk industry	ac	-1 -2	48 2	450	-1 -2	65 13	710	-1 -2	65 13	710
c	2347763	Ext 2022	Dust egg product industry	ac	-2 -3	27 2	2600	-2 -3	42 2	4000	-2 -3	42 2	4000
c	2347764	Ext 2022	Residue animal feed	nc	-1 -2	0 0	<10	-1 -2	0 0	<10	-1 -2	0 0	<10
c	2347765	Ext 2022	Residue animal feed	nc	-1 -2	0 0	<10	-1 -2	0 0	<10	-1 -2	0 0	<10



**Animal feed**

Type	#	Study	Sample	Conta	Reference method: NF ISO 16649-2*			Alternative method: REC2 37°C			Alternative method: REC2 after storage 72h at 2-8°C		
					Dilution	CFU/ plate	Result (CFU/g)	Dilution	CFU/plate	Result (CFU/g)	Dilution	CFU/plate	Result (CFU/g)
a	2364266	Ext 2023	Salmon cat kibble	ac	-1 -2	1 0	10	-1 -2	2 0	20	-1 -2	3 0	30
a	2364269	Ext 2023	Rabbit pellets	ac	-3 -4	0 0	<1000	-3 -4	0 0	<1000	-3 -4	0 0	<1000
a	2364270	Ext 2023	Dog kibble	ac	-2 -3	1 0	100	-2 -3	0 0	<100	-2 -3	1 0	100
a	2364267	Ext 2023	Terrine dog poultry	ac	-2 -3	12 1	1200	-2 -3	13 0	1300	-2 -3	13 0	1300
a	2364268	Ext 2023	Beef Cat Pate	ac	-2 -3	1 0	100	-2 -3	0 0	<100	-2 -3	0 0	<100
a	2392504	Ext 2023	Beef Cat Pate	ac	-1 -2	10 1	100	-1 -2	16 2	160	-1 -2	16 2	160
a	2392518	Ext 2023	Cat kibble with salmon	ac	-1 -2	1 0	10	-1 -2	5 0	50	-1 -2	5 0	50
a	2392519	Ext 2023	Beef dog kibble	ac	-1 -2	14 0	140	-1 -2	20 1	190	-1 -2	19 1	190
a	2392520	Ext 2023	Rabbit pellets	ac	-1 -2	0 0	<10	-1 -2	2 0	20	-1 -2	2 0	20
a	2392586	Ext 2023	Cat kibble with turkey	ac	-1 -2	73 8	740	-1 -2	91 10	920	-1 -2	91 10	920
a	2392587	Ext 2023	Cat kibble without cereals	ac	-1 -2	4 0	40	-1 -2	4 0	40	-1 -2	4 0	40
b	2364274	Ext 2023	Ewes pellets	ac	-1 -2	7 1	70	-1 -2	4 1	40	-1 -2	4 1	40
b	2364277	Ext 2023	Soy Cattle cake	ac	-2 -3	12 1	1200	-2 -3	14 1	1400	-2 -3	14 1	1400
b	2364279	Ext 2023	Mixed wheat	ac	-1 -2	16 1	160	-1 -2	27 2	260	-1 -2	27 2	260
b	2364276	Ext 2023	Lamb pellets	nc	-1 -2	0 0	<10	-1 -2	0 0	<10	-1 -2	0 0	<10
b	2392588	Ext 2023	Soy	ac	-1 -2	2 0	20	-1 -2	1 0	10	-1 -2	1 0	10
b	2392589	Ext 2023	Corn	ac	-1 -2	114 9	1 100	-1 -2	144 12	1 400	-1 -2	144 12	1 400
b	2392590	Ext 2023	Wheat	ac	-1 -2	6 0	60	-1 -2	11 1	110	-1 -2	11 1	110
b	2392591	Ext 2023	Wheat	ac	-1 -2	7 0	70	-1 -2	5 0	50	-1 -2	5 0	50
c	2392481	Ext 2023	Chicken flour batch 1	ac	-1 -2	16 1	160	-1 -2	13 1	130	-1 -2	13 1	130
c	2392485	Ext 2023	Salmon flour	ac	-2 -3	0 0	<10	-2 -3	1 0	100	-2 -3	1 0	100
c	2392488	Ext 2023	Soy protein	ac	-1 -2	1 0	10	-1 -2	1 0	10	-1 -2	1 0	10
c	2392522	Ext 2023	Chicken flour batch 2	ac	-1 -2	1 0	10	-1 -2	3 0	30	-1 -2	3 0	30
c	2392523	Ext 2023	Wheat fiber	ac	-2 -3	12 1	1200	-2 -3	10 0	1000	-2 -3	10 0	1000
c	2392525	Ext 2023	Salmon flour	ac	-1 -2	10 1	100	-1 -2	42 2	400	-1 -2	43 2	420
c	2319051	Ext 2023	Beef flour	ac	-2 -3	11 1	1100	-2 -3	14 1	1400	-2 -3	15 1	1500
c	2392565	Ext 2023	Raw animal by-product (offal)	nc	-2 -3	58 1	5400	-2 -3	42 0	4200	-2 -3	42 0	4200
c	2392621	Ext 2023	Fresh beef meat	nc	-1 -2	54 5	540	-1 -2	60 8	620	-1 -2	61 8	630
c	2392622	Ext 2023	Raw animal by-product (offal)	nc	-2 -3	115 12	12 000	-2 -3	101 15	11 000	-2 -3	103 15	11 000

## **Appendix E**

### **Relative trueness study – Statistical calculations**

## Appendix E

	Type	#	Study	Sample	RM	AM	RM	AM	Mean	Difference	
					CFU/g	CFU/g	log CFU/g	log CFU/g			
Meat products	a	29	IVS	Beef kidneys	70	60	1.85	1.78	1.81	-0.07	
	a	32	IVS	Turkey cutlet	830	830	2.92	2.92	2.92	0.00	
	a	33	IVS	Chopped steak	60	60	1.78	1.78	1.78	0.00	
	a	35	IVS	Veal cutlet	200	400	2.30	2.60	2.45	0.30	
	a	ISHA 1	R2017	Ground beef	180	120	2.26	2.08	2.17	-0.18	
	a	34	IVS	Chipolata	960	850	2.98	2.93	2.96	-0.05	
	a	3	IVS	Merguez	120	180	2.08	2.26	2.17	0.18	
	a	4	IVS	Veal Sausage	150	210	2.18	2.32	2.25	0.15	
	a	6	IVS	Herb Sausage	380	520	2.58	2.72	2.65	0.14	
	b	ISHA 2	R2017	Poultry bites	2900	4300	3.46	3.63	3.55	0.17	
	b	ISHA 30	R2017	Ravioli of poultry	3200	3700	3.51	3.57	3.54	0.06	
	b	ISHA 32	R2017	Parmentier of duck (CA)	370	370	2.57	2.57	2.57	0.00	
	b	ISHA 59	R2017	Lasagna 3 meats (CA)	1800	1500	3.26	3.18	3.22	-0.08	
	b	ISHA 61	R2017	Grilled Sausage (CA)	1400	1400	3.15	3.15	3.15	0.00	
	b	ISHA 62	R2017	Parmentier of ham (CA)	1400	1100	3.15	3.04	3.09	-0.10	
	c	2392603	Ext. 2023	Iberian cured ham	50	70	1.70	1.85	1.77	0.15	
	c	2392604	Ext. 2023	Smoked bacon	50	110	1.70	2.04	1.87	0.34	
	c	2392605	Ext. 2023	Smoked ham	200	130	2.30	2.11	2.21	-0.19	
	c	2392606	Ext. 2023	Dry ham	260	310	2.41	2.49	2.45	0.08	
	c	2392607	Ext. 2023	Smoked bacon	280	320	2.45	2.51	2.48	0.06	
	c	ISHA 35	R2017	Smoked chicken (CA)	1600	1400	3.20	3.15	3.18	-0.06	
	<b>Average difference of the category</b>										0.04
	<b>Standard deviation of differences</b>										0.14

## Appendix E

	Type	#	Study	Sample	RM	AM	RM	AM	Mean	Difference	
					CFU/g	CFU/g	log CFU/g	log CFU/g			
Dairy and egg products	a	ISHA 18	R2017	Pasteurized milk brie	310	220	2.49	2.34	2.42	-0.15	
	a	ISHA 19	R2017	Pasteurized milk brie	190	160	2.28	2.20	2.24	-0.07	
	a	ISHA 20	R2017	Pasteurized milk brie	310	360	2.49	2.56	2.52	0.06	
	a	ISHA 42	R2017	Goat cheese (CA)	190	310	2.28	2.49	2.39	0.21	
	a	ISHA 43	R2017	Emmental (CA)	190	180	2.28	2.26	2.27	-0.02	
	b	10	IVS	Reblochon	870	1300	2.94	3.11	3.03	0.17	
	b	25	IVS	Roquefort	210	560	2.32	2.75	2.54	0.43	
	b	36	IVS	Raw milk cheese	20000	15000	4.30	4.18	4.24	-0.12	
	b	59	IVS	Raw milk cheese	2800	3600	3.45	3.56	3.50	0.11	
	b	78	IVS	Reblochon	6500	5300	3.81	3.72	3.77	-0.09	
	b	90	IVS	Raw milk cheese (CA)	1600	2500	3.20	3.40	3.30	0.19	
	c	96	IVS	Chocolate cake	12000	12000	4.08	4.08	4.08	0.00	
	c	95	IVS	Versailles	23000	26000	4.36	4.41	4.39	0.05	
	c	93	IVS	Cream puffs	180	150	2.26	2.18	2.22	-0.08	
	c	70	IVS	Coffee éclair	140	190	2.15	2.28	2.21	0.13	
	c	40	IVS	Banana cream	70	80	1.85	1.90	1.87	0.06	
	c	20	IVS	Coffee éclair	90	45	1.95	1.65	1.80	-0.30	
	c	72	IVS	Peach cake	4000	1800	3.60	3.26	3.43	-0.35	
	c	94	IVS	Apple pie	1770	1600	3.25	3.20	3.23	-0.04	
	c	60	IVS	Apricot cake	290	280	2.46	2.45	2.45	-0.02	
	c	ISHA 21	R2017	Almond cake	1400	1400	3.15	3.15	3.15	0.00	
	c	ISHA 40	R2017	Egg cream (CA)	130	170	2.11	2.23	2.17	0.12	
	c	ISHA 72	R2017	Apple pie (CA)	290	290	2.46	2.46	2.46	0.00	
	<b>Average difference of the category</b>										0.01
	<b>Standard deviation of differences</b>										0.17

## Appendix E

	Type	#	Study	Sample	RM	AM	RM	AM	Mean	Difference	
					CFU/g	CFU/g	log CFU/g	log CFU/g			
Seafood products	a	52	IVS	Fillet of Red mullet	22000	30000	4.34	4.48	4.41	0.13	
	a	53	IVS	Salmon Steak	29000	30000	4.46	4.48	4.47	0.01	
	a	54	IVS	Fish	290000	180000	5.46	5.26	5.36	-0.21	
	a	ISHA 45	R2017	Lingue fillet (CA)	5500	4000	3.74	3.60	3.67	-0.14	
	a	2392601	Ext.2023	Whiting fillet	40	110	1.60	2.04	1.82	0.44	
	b	55	IVS	Smoked salmon	240000	200000	5.38	5.30	5.34	-0.08	
	b	91	IVS	Smoked Salmon (CA)	260	210	2.41	2.32	2.37	-0.09	
	b	92	IVS	Smoked Salmon (CA)	1900	2600	3.28	3.41	3.35	0.14	
	b	ISHA 31	R2017	Smoked Salmon (CA)	65	140	1.81	2.15	1.98	0.33	
	b	ISHA 38	R2017	Smoked herring fillet (CA)	75	120	1.88	2.08	1.98	0.20	
	b	ISHA 39	R2017	Smoked herring fillet (CA)	70	120	1.85	2.08	1.96	0.23	
	b	ISHA 47	R2017	Smoked trout (CA)	280	230	2.45	2.36	2.40	-0.09	
	b	ISHA 48	R2017	Smoked Salmon (CA)	260	320	2.41	2.51	2.46	0.09	
	b	ISHA 51	R2017	Smoked mackerel (CA)	530	560	2.72	2.75	2.74	0.02	
	b	ISHA 46	R2017	Marinated mackerel (CA)	19000	14000	4.28	4.15	4.21	-0.13	
	b	ISHA 44	R2017	Haddock fillet (CA)	6500	13000	3.81	4.11	3.96	0.30	
	c	61	IVS	Shrimp (CA)	2300	2300	3.36	3.36	3.36	0.00	
	c	ISHA 29-2	R2017	Cod Brandade (CA)	460	570	2.66	2.76	2.71	0.09	
	c	ISHA-63	R2017	Cod with sorrel sauce (CA)	65	90	1.81	1.95	1.88	0.14	
	c	ISHA-64	R2017	Cod with tomato sauce (CA)	75	70	1.88	1.85	1.86	-0.03	
	c	ISHA-65	R2017	Salmon with sorrel sauce (CA)	85	80	1.93	1.90	1.92	-0.03	
	<b>Average difference of the category</b>										0.06
	<b>Standard deviation of differences</b>										0.17

## Appendix E

	Type	#	Study	Sample	RM	AM	RM	AM	Mean	Difference	
					CFU/g	CFU/g	log CFU/g	log CFU/g			
Vegetal products	a	ISHA 66	R2017	Cucumber (CA)	4200	5200	3.62	3.72	3.67	0.09	
	a	ISHA 67	R2017	Tomato (CA)	7700	9300	3.89	3.97	3.93	0.08	
	a	ISHA 68	R2017	Apple (CA)	1400	1600	3.15	3.20	3.18	0.06	
	a	ISHA 69	R2017	Grape (CA)	110	130	2.04	2.11	2.08	0.07	
	a	ISHA 70	R2017	Red cabbage (CA)	430	520	2.63	2.72	2.67	0.08	
	b	89	IVS	Grated carrots	1600	2400	3.20	3.38	3.29	0.18	
	b	ISHA 54	R2017	Salad Mix (CA)	340	380	2.53	2.58	2.56	0.05	
	b	ISHA 55	R2017	Grated carrots (CA)	350	310	2.54	2.49	2.52	-0.05	
	b	ISHA 56	R2017	Salad Bag (CA)	280	460	2.45	2.66	2.55	0.22	
	b	ISHA 57	R2017	"Poêlée" of vegetables (CA)	390	460	2.59	2.66	2.63	0.07	
	c	50	IVS	Macedonia	65	45	1.81	1.65	1.73	-0.16	
	c	56	IVS	Fruit Compote	660	580	2.82	2.76	2.79	-0.06	
	c	ISHA 12	R2017	Basil Condiment	180	230	2.26	2.36	2.31	0.11	
	c	ISHA 71	R2017	Basil Condiment (CA)	9200	8700	3.96	3.94	3.95	-0.02	
	c	ISHA 6	R2017	Parsnip Purée	130	50	2.11	1.70	1.91	-0.41	
	<b>Average difference of the category</b>										0.02
	<b>Standard deviation of differences</b>										0.15

## 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	130	2.15	2.11	2.13	-0.03	
	a	ISHA 3	R2017	Gabonese salad	80	80	1.90	1.90	1.90	0.00	
	a	ISHA 71	R2017	Tabbouleh (CA)	570	480	2.76	2.68	2.72	-0.07	
	a	2392602	Ext.2023	Texas sandwich	260	210	2.41	2.32	2.37	-0.09	
	a	ISHA 13	R2017	Tomato salad dressing	280	380	2.45	2.58	2.51	0.13	
	b	ISHA 33	R2017	"Croque monsieur" (CA)	910	1600	2.96	3.20	3.08	0.25	
	b	ISHA 52	R2017	Quiche Lorraine (CA)	240	260	2.38	2.41	2.40	0.03	
	b	ISHA 53	R2017	Beef vegetables	250	260	2.40	2.41	2.41	0.02	
	b	ISHA 60	R2017	Chicken Peas (CA)	1400	1000	3.15	3.00	3.07	-0.15	
	b	ISHA 58	R2017	Endive with Ham (CA)	1700	1700	3.23	3.23	3.23	0.00	
	c	ISHA 49	R2017	Carpaccio (CA)	190	170	2.28	2.23	2.25	-0.05	
	c	ISHA 50	R2017	Carpaccio (CA)	220	310	2.34	2.49	2.42	0.15	
	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	11000	4.00	4.04	4.02	0.04	
	c	ISHA 75	R2017	Beef marinated (CA)	11000	11000	4.04	4.04	4.04	0.00	
	<b>Average difference of the category</b>										0.01
	<b>Standard deviation of differences</b>										0.10

## Appendix E

	Type	#	Study	Sample	RM	AM	RM	AM	Mean	Difference	
					CFU/g	CFU/g	log CFU/g	log CFU/g			
Environmental samples	a	2347747	Ext 2022	Wipe cheese cold room shelf	600	570	2.78	2.76	2.77	-0.02	
	a	2347748	Ext 2022	Wipe refrigerated delicatessen display shelf	600	800	2.78	2.90	2.84	0.12	
	a	2347750	Ext 2022	Wipe seafood working plan	40 000	54 000	4.60	4.73	4.67	0.13	
	a	2347770	Ext 2022	Wipe on ventilation grid for refrigerated showcase butcher	1100	2500	3.04	3.40	3.22	0.36	
	a	2347771	Ext 2022	Wipe workplan delicatessen preparation	250	460	2.40	2.66	2.53	0.26	
	a	2347772	Ext 2022	Wipe workplan butcher	180	200	2.26	2.30	2.28	0.05	
	b	2347756	Ext 2022	Process water pastry industry	70	200	1.85	2.30	2.07	0.46	
	b	2347753	Ext 2022	Process water cheese industry	300	390	2.48	2.59	2.53	0.11	
	b	2347757	Ext 2022	Process water vegetables	300	640	2.48	2.81	2.64	0.33	
	b	2347758	Ext 2022	Process water fish shop	3900	5100	3.59	3.71	3.65	0.12	
	b	2333891	Ext 2022	Process water poultry industry	9 000 000	12 000 000	6.95	7.08	7.02	0.12	
	b	2333890	Ext 2022	Process water poultry industry	12 000 000	11 000 000	7.08	7.04	7.06	-0.04	
	c	2347761	Ext 2022	Flour dust	50	60	1.70	1.78	1.74	0.08	
	c	2347762	Ext 2022	Dust milk industry	450	710	2.65	2.85	2.75	0.20	
	c	2347708	Ext 2022	Cheese industry residue area 2	470	710	2.67	2.85	2.76	0.18	
	c	2347763	Ext 2022	Dust egg product industry	2600	4000	3.41	3.60	3.51	0.19	
	c	2333895	Ext 2022	Poultry industry residue	1 300 000	1 600 000	6.11	6.20	6.16	0.09	
	c	2333894	Ext 2022	Poultry industry residue	1 900 000	3 800 000	6.28	6.58	6.43	0.30	
	<b>Average difference of the category</b>										0.17
	<b>Standard deviation of differences</b>										0.13



## Appendix E

	Type	#	Study	Sample	RM	AM	RM	AM	Mean	Difference	
					CFU/g	CFU/g	log CFU/g	log CFU/g			
Animal feed	a	2392587	Ext 2023	Cat kibble without cereals	40	40	1.60	1.60	1.60	0.00	
	a	2392504	Ext 2023	Beef Cat Pate	100	160	2.00	2.20	2.10	0.20	
	a	2392519	Ext 2023	Beef dog kibble	140	190	2.15	2.28	2.21	0.13	
	a	2392586	Ext 2023	Cat kibble with turkey	740	920	2.87	2.96	2.92	0.09	
	a	2364267	Ext 2023	Terrine dog poultry	1200	1300	3.08	3.11	3.10	0.03	
	b	2392590	Ext 2023	Wheat	60	110	1.78	2.04	1.91	0.26	
	b	2392591	Ext 2023	Wheat	70	50	1.85	1.70	1.77	-0.15	
	b	2364274	Ext 2023	Ewes pellets	70	40	1.85	1.60	1.72	-0.24	
	b	2364279	Ext 2023	Mixed wheat	160	260	2.20	2.41	2.31	0.21	
	b	2392589	Ext 2023	Corn	1 100	1 400	3.04	3.15	3.09	0.10	
	b	2364277	Ext 2023	Soy Cattle cake	1200	1400	3.08	3.15	3.11	0.07	
	c	2392525	Ext 2023	Salmon flour	100	400	2.00	2.60	2.30	0.60	
	c	2392481	Ext 2023	Chicken flour batch 1	160	130	2.20	2.11	2.16	-0.09	
	c	2392621	Ext 2023	Fresh beef meat	540	620	2.73	2.79	2.76	0.06	
	c	2319051	Ext 2023	Beef flour	1100	1400	3.04	3.15	3.09	0.10	
	c	2392523	Ext 2023	Wheat fiber	1200	1000	3.08	3.00	3.04	-0.08	
	c	2392565	Ext 2023	Raw animal by-product (offal)	5400	4200	3.73	3.62	3.68	-0.11	
	c	2392622	Ext 2023	Raw animal by-product (offal)	12 000	11 000	4.08	4.04	4.06	-0.04	
	<b>Average difference of the category</b>										0.07
	<b>Standard deviation of differences</b>										0.19
<b>Average difference of all categories</b>										0.06	
<b>Standard deviation of differences</b>										0.16	

		Lower confidence limit	Upper confidence limit	
n =	21 T(0.025;20) 2.086	MP	-0.26	0.34
n =	23 T(0.025;22) 2.074	D&EP	-0.34	0.37
n =	21 T(0.025;20) 2.086	SFP	-0.30	0.31
n =	15 T(0.025;14) 2.145	VP	-0.32	0.36
n =	15 T(0.025;14) 2.145	RTE,RTRH	-0.21	0.07
n =	18 T(0.025;17) 2.110	ES	-0.11	0.45
n =	18 T(0.025;17) 2.110	AF	-0.34	0.47
n =	131 T(0.025;13) 1.978	All	-0.26	0.37
β =	95%			

<b>n =</b>	<b>131</b>		
<b>β =</b>	<b>0.95</b>	<b>Lower confidence limit</b>	<b>Upper confidence limit</b>
<b>T(0.025;130)=</b>	<b>1.98</b>	<b>-0.26</b>	<b>0.37</b>

## Appendix E

### Results not used in the statistical interpretation

Category	Type	#	Study	Sample	RM	AM	RM	AM	Mean	Difference
					CFU/g	CFU/g	log CFU/g	log CFU/g		
Meat products	a	31	IVS	Sausage	30	40	1.48	1.60	1.54	0.12
	a	39	IVS	Turkey steak	15	20	1.18	1.30	1.24	0.12
	a	26	IVS	Turkey steak	10	<10	1.00	0.00	0.50	-1.00
	a	28	IVS	Chicken fillet	5	<10	0.70	0.00	0.35	-0.70
	a	39	IVS	Turkey steak	15	20	1.18	1.30	1.24	0.12
	c	ISHA 36	R2017	Smoked veal (CA)	<1000	2000	2.00	3.30	2.65	1.30
Dairy and egg products	b	5	IVS	Raw milk	<1	3	0.00	0.48	0.24	0.48
	b	24	IVS	Reblochon	40	30	1.60	1.48	1.54	-0.12
	c	17	IVS	Savarin with chocolate	5	10	0.70	1.00	0.85	0.30
	c	ISHA 34	R2017	Omelet (CA)	20	30	1.30	1.48	1.39	0.18
Vegetal products	b	ISHA 15	R2017	Carrots in bags	5	<10	0.70	0.00	0.35	-0.70
	a	57	IVS	Strawberry	30	70	1.48	1.85	1.66	0.37
Composite foods	a	19	IVS	Pastas salad	5	<10	0.70	0.00	0.35	-0.70
	b	ISHA 37	R2017	Gratin (CA)	10	20	1.00	1.30	1.15	0.30
	b	ISHA 14	R2017	Salmon Tagliatelle (CA)	10	<10	1.00	0.00	0.50	-1.00
Animal feed	a	2364266	Ext 2023	Salmon cat kibble	10	20	1.00	1.30	1.15	0.30
	a	2364270	Ext 2023	Dog kibble	100	<100	2.00	1.00	1.50	-1.00
	a	2364268	Ext 2023	Beef Cat Pate	100	<100	2.00	0.00	1.00	-2.00
	a	2392518	Ext 2023	Cat kibble with salmon	10	50	1.00	1.70	1.35	0.70
	a	2392520	Ext 2023	Rabbit pellets	<10	20	0.00	1.30	0.65	1.30
	b	2392588	Ext 2023	Soy	20	10	1.30	1.00	1.15	-0.30
	c	2392485	Ext 2023	Salmon flour	<10	100	0.00	2.00	1.00	2.00
	c	2392488	Ext 2023	Soy protein	10	10	1.00	1.00	1.00	0.00
	c	2392522	Ext 2023	Chicken flour batch 2	10	30	1.00	1.48	1.24	0.48

## **Appendix F**

### **Accuracy profiles – Raw results**

## Meat products

Matrix	Strain	Level	Sample number	Reference method 16649-2						Alternative method REC2 37°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 <sup>3</sup> CFU/g	ESC.1.15	300 CFU/g	L1T1R1	-1	83	-1	50	660	2.820	-1	54	540	2.732
				-2	9	-2	3			-2	5		
			L1T1R2	-1	41	-1	52	460	2.663	-1	64	670	2.826
				-2	3	-2	5			-2	10		
			L1T1R3	-1	44	-1	35	400	2.602	-1	58	580	2.763
				-2	4	-2	4			-2	6		
			L1T1R4	-1	43	-1	62	510	2.708	-1	41	420	2.623
				-2	4	-2	4			-2	5		
			L1T1R5	-1	45	-1	51	460	2.663	-1	44	460	2.663
				-2	3	-2	3			-2	6		
		3000 CFU/g	L1T2R1	-1	118	-1	96	1200	3.079	-1	110	1 100	3.041
				-2	20	-2	19			-2	16		
			L1T2R2	-1	90	-1	98	960	2.982	-1	94	1 000	3.000
				-2	11	-2	12			-2	15		
			L1T2R3	-1	110	-1	100	1100	3.041	-1	112	1 100	3.041
				-2	18	-2	17			-2	10		
			L1T2R4	-1	132	-1	100	1200	3.079	-1	108	1 100	3.041
				-2	14	-2	10			-2	10		
			L1T2R5	-1	114	-1	120	1200	3.079	-1	108	1 100	3.041
				-2	8	-2	18			-2	13		
		100000 CFU/g	L1T3R1	-3	110	-3	111	110000	5.041	-3	127	130000	5.114
				-4	10	-4	14			-4	21		
			L1T3R2	-3	102	-3	80	94000	4.973	-3	140	140000	5.146
				-4	16	-4	8			-4	12		
			L1T3R3	-3	125	-3	139	130000	5.114	-3	146	140000	5.146
				-4	11	-4	13			-4	12		
			L1T3R4	-3	156	-3	160	160000	5.204	-3	146	150000	5.176
				-4	18	-4	13			-4	16		
			L1T3R5	-3	145	-3	140	140000	5.146	-3	123	130000	5.114
				-4	15	-4	14			-4	23		

## Meat products

Matrix	Strain	Level	Sample number	Reference method 16649-2						Alternative method REC2 37°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: 2,0x10 <sup>3</sup> CFU/g	ESC.1.15	300 CFU/g	L1T1R1	-1	14	-1	16	150	2.176	-1	22	210	2.322
				-2	1	-2	1						
			L1T1R2	-1	17	-1	13	150	2.176	-1	17	170	2.230
				-2	1	-2	2						
			L1T1R3	-1	15	-1	15	160	2.204	-1	18	170	2.230
				-2	3	-2	2						
			L1T1R4	-1	15	-1	8	100	2.000	-1	18	190	2.279
				-2	0	-2	0						
			L1T1R5	-1	22	-1	17	200	2.301	-1	15	170	2.230
				-2	0	-2	4						
		3000 CFU/g	L1T2R1	-2	16	-2	18	1700	3.230	-2	26	2 500	3.398
				-3	2	-3	2						
			L1T2R2	-2	17	-2	17	1600	3.204	-2	23	2 300	3.362
				-3	0	-3	1						
			L1T2R3	-2	18	-2	19	1800	3.255	-2	18	1 600	3.204
				-3	1	-3	1						
			L1T2R4	-2	13	-2	14	1300	3.114	-2	15	1 500	3.176
				-3	2	-3	0						
			L1T2R5	-2	7	-2	14	1000	3.000	-2	14	1 400	3.146
				-3	0	-3	2						
		100000 CFU/g	L1T3R1	-3	143	-3	106	120000	5.08	-3	86	85000	4.929
				-4	14	-4	10						
			L1T3R2	-3	128	-3	101	110000	5.04	-3	146	150000	5.176
				-4	12	-4	5						
			L1T3R3	-3	95	-3	106	98000	4.99	-3	99	92000	4.964
				-4	9	-4	6						
			L1T3R4	-3	131	-3	138	140000	5.15	-3	106	100000	5.000
				-4	14	-4	14						
			L1T3R5	-3	125	-3	141	130000	5.11	-3	88	85000	4.929
				-4	12	-4	14						

## Dairy and egg products

Matrix	Strain	Level	Sample number	Reference method 16649-2						Alternative method REC2 37°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 <sup>7</sup> CFU/g	ESC.1.5	300 CFU/g	L1T1R1	-1	13	-1	18	150	2.176	-1	16	150	2.176
				-2	1	-2	2			-2	1		
			L1T1R2	-1	18	-1	19	170	2.230	-1	16	150	2.176
				-2	1	-2	0			-2	0		
			L1T1R3	-1	14	-1	18	160	2.204	-1	18	200	2.301
				-2	2	-2	1			-2	4		
		L1T1R4	-1	12	-1	12	120	2.079	-1	15	150	2.176	
			-2	3	-2	0			-2	2			
		L1T1R5	-1	10	-1	16	120	2.079	-1	10	110	2.041	
			-2	1	-2	0			-2	2			
		3000 CFU/g	L1T2R1	-1	143	-1	151	1500	3.176	-1	142	1 500	3.176
				-2	10	-2	15			-2	20		
			L1T2R2	-1	144	-1	148	1500	3.176	-1	142	1 500	3.176
				-2	25	-2	14			-2	18		
			L1T2R3	-1	122	-1	150	1400	3.146	-1	146	1 500	3.176
				-2	19	-2	15			-2	20		
		L1T2R4	-1	165	-1	142	1500	3.176	-1	130	1 400	3.146	
			-2	14	-2	16			-2	22			
		L1T2R5	-1	162	-1	165	1700	3.230	-1	151	1 600	3.204	
			-2	22	-2	26			-2	22			
		100000 CFU/g	L1T3R1	-3	42	-3	54	51000	4.708	-3	38	37 000	4.568
				-4	8	-4	7			-4	3		
			L1T3R2	-3	57	-3	44	52000	4.716	-3	56	56 000	4.748
				-4	8	-4	5			-4	6		
			L1T3R3	-3	56	-3	56	56000	4.748	-3	55	58 000	4.763
				-4	5	-4	5			-4	9		
			L1T3R4	-3	43	-3	44	42000	4.623	-3	50	50 000	4.699
-4	2			-4	4	-4	5						
L1T3R5	-3		48	-3	46	47000	4.672	-3	44	44 000	4.643		
	-4		4	-4	6			-4	4				

## Dairy and egg products

Matrix	Strain	Level	Sample number	Reference method 16649-2						Alternative method REC2 37°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	24	260	2.415
				-2	1	-2	1			-2	5		
			L1T1R2	-1	16	-1	16	150	2.176	-1	25	260	2.415
				-2	1	-2	1			-2	4		
			L1T1R3	-1	15	-1	14	150	2.176	-1	14	160	2.204
				-2	3	-2	0			-2	4		
			L1T1R4	-1	8	-1	23	150	2.176	-1	26	250	2.398
				-2	1	-2	1			-2	2		
			L1T1R5	-1	21	-1	27	230	2.362	-1	23	250	2.398
				-2	2	-2	0			-2	5		
		3000 CFU/g	L1T2R1	-1	151	-1	155	1500	3.176	-1	145	1 500	3.176
				-2	10	-2	11			-2	22		
			L1T2R2	-1	169	-1	143	1600	3.204	-1	168	1 700	3.230
				-2	18	-2	15			-2	19		
			L1T2R3	-1	141	-1	140	1400	3.146	-1	116	1 100	3.041
				-2	19	-2	18			-2	10		
			L1T2R4	-1	170	-1	160	1700	3.230	-1	196	1900	3.279
				-2	19	-2	20			-2	15		
			L1T2R5	-1	140	-1	134	1400	3.146	-1	98	950	2.978
				-2	21	-2	12			-2	7		
		100000 CFU/g	L1T3R1	-3	41	-3	58	48000	4.681	-3	60	57 000	4.756
				-4	2	-4	4			-4	3		
			L1T3R2	-3	52	-3	70	59000	4.771	-3	40	41 000	4.613
				-4	4	-4	4			-4	5		
			L1T3R3	-3	54	-3	54	56000	4.748	-3	49	53 000	4.724
				-4	8	-4	7			-4	9		
			L1T3R4	-3	50	-3	52	51000	4.708	-3	54	51 000	4.708
				-4	5	-4	5			-4	2		
			L1T3R5	-3	53	-3	49	52000	4.716	-3	46	48 000	4.681
				-4	7	-4	5			-4	7		

## Seafood products

Matrix	Strain	Level	Sample number	Reference method 16649-2					Alternative method REC2 37°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 <sup>7</sup> CFU/g	ESC.1.31	300 CFU/g	L1T1R1	-1	51	-1	36	440	2.643	-1	56	540	2.732
				-2	5	-2	4			-2	3		
			L1T1R2	-1	31	-1	43	370	2.568	-1	46	520	2.716
				-2	5	-2	3			-2	0		
			L1T1R3	-1	42	-1	35	370	2.568	-1	45	440	2.643
				-2	3	-2	1			-2	3		
			L1T1R4	-1	32	-1	33	320	2.505	-1	36	390	2.591
				-2	1	-2	4			-2	7		
			L1T1R5	-1	41	-1	25	320	2.505	-1	32	350	2.544
				-2	4	-2	1			-2	6		
		3000 CFU/g	L1T2R1	-2	35	-2	36	3300	3.519	-2	43	4500	3.653
				-3	0	-3	1			-3	6		
			L1T2R2	-2	28	-2	35	3300	3.519	-2	43	4100	3.613
				-3	4	-3	5			-3	2		
			L1T2R3	-2	38	-2	47	4300	3.633	-2	28	3000	3.477
				-3	2	-3	8			-3	4		
			L1T2R4	-2	46	-2	30	4000	3.602	-2	40	3900	3.591
				-3	8	-3	4			-3	3		
			L1T2R5	-2	33	-2	34	3600	3.556	-2	30	3200	3.505
				-3	6	-3	6			-3	5		
		100000 CFU/g	L1T3R1	-3	112	-3	126	120000	5.079	-3	158	150000	5.176
				-4	12	-4	19			-4	7		
			L1T3R2	-3	107	-3	108	110000	5.041	-3	116	120000	5.079
				-4	15	-4	10			-4	13		
			L1T3R3	-3	78	-3	104	93000	4.968	-3	85	84000	4.924
				-4	12	-4	11			-4	7		
			L1T3R4	-3	106	-3	105	110000	5.041	-3	100	100000	5.000
-4	10			-4	8	-4	10						
L1T3R5	-3		96	-3	119	110000	5.041	-3	97	91000	4.959		
	-4		9	-4	11			-4	3				



## Seafood products

Matrix	Strain	Level	Sample number	Reference method 16649-2					Alternative method REC2 37°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: 2,5x10 <sup>7</sup> CFU/g	ESC.1.31	300 CFU/g	L1T1R1	-1	71	-1	68	670	2.828	-1	53	530	2.724
				-2	5	-2	4						
			L1T1R2	-1	39	-1	35	380	2.577	-1	37	360	2.556
				-2	6	-2	3						
			L1T1R3	-1	41	-1	41	400	2.607	-1	40	380	2.580
				-2	2	-2	5						
			L1T1R4	-1	25	-1	35	320	2.503	-1	34	330	2.519
				-2	6	-2	4						
			L1T1R5	-1	44	-1	42	400	2.597	-1	38	380	2.580
				-2	0	-2	1						
		3000 CFU/g	L1T2R1	-2	35	-2	32	3500	3.544	-2	37	3 800	3.580
				-3	5	-3	5						
			L1T2R2	-2	23	-2	38	3100	3.490	-2	40	4 100	3.613
				-3	3	-3	4						
			L1T2R3	-2	26	-2	22	2600	3.421	-2	39	3 800	3.580
				-3	6	-3	4						
			L1T2R4	-2	48	-2	38	4200	3.631	-2	30	2 910	3.464
				-3	5	-3	3						
			L1T2R5	-2	36	-2	42	3900	3.587	-2	40	4 000	3.602
				-3	2	-3	5						
		100000 CFU/g	L1T3R1	-3	127	-3	141	130000	5.117	-3	106	110000	5.041
				-4	13	-4	7						
			L1T3R2	-3	110	-3	103	100000	5.017	-3	118	120000	5.079
				-4	5	-4	11						
			L1T3R3	-3	95	-3	103	100000	5.014	-3	123	120000	5.079
				-4	12	-4	17						
			L1T3R4	-3	108	-3	109	110000	5.043	-3	101	100000	5.000
-4	14			-4	12								
L1T3R5	-3		128	-3	107	110000	5.057	-3	134	140000	5.146		
	-4		6	-4	10								

## Vegetal products

Matrix	Strain	Level	Sample number	Reference method 16649-2						Alternative method REC2 37°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: $2,0 \times 10^2$ CFU/g	ESC.1.2	300 CFU/g	L1T1R1	-1	6	-1	2	40	1.602	-1	7	70	1.845
				-2	2	-2	0			-2	0		
			L1T1R2	-1	4	-1	5	45	1.653	-1	10	100	2.000
				-2	0	-2	0			-2	1		
			L1T1R3	-1	8	-1	4	60	1.778	-1	11	110	2.041
				-2	0	-2	0			-2	1		
			L1T1R4	-1	8	-1	6	70	1.845	-1	8	80	1.903
				-2	1	-2	1			-2	3		
			L1T1R5	-1	9	-1	8	85	1.929	-1	13	120	2.079
				-2	1	-2	0			-2	0		
		3000 CFU/g	L1T2R1	-2	34	-2	46	4 200	3.623	-2	88	8 400	3.924
				-3	7	-3	5			-3	4		
			L1T2R2	-2	49	-2	51	4 800	3.681	-2	71	7 100	3.851
				-3	3	-3	3			-3	7		
			L1T2R3	-2	38	-2	38	3 700	3.568	-2	89	9 100	3.959
				-3	4	-3	1			-3	11		
			L1T2R4	-2	48	-2	40	4 400	3.643	-2	80	7 700	3.886
				-3	5	-3	3			-3	5		
			L1T2R5	-2	50	-2	60	5 600	3.748	-2	85	8 500	3.929
				-3	7	-3	5			-3	8		
		100000 CFU/g	L1T3R1	-4	10	-4	13	120 000	5.079	-4	18	170 000	5.230
				-5	2	-5	2			-5	1		
			L1T3R2	-4	14	-4	12	120 000	5.079	-4	29	290 000	5.462
				-5	1	-5	0			-5	3		
			L1T3R3	-4	13	-4	10	110 000	5.041	-4	18	170 000	5.230
				-5	1	-5	0			-5	1		
			L1T3R4	-4	14	-4	11	170 000	5.230	-4	21	200 000	5.301
				-5	6	-5	7			-5	1		
			L1T3R5	-4	10	-4	13	110 000	5.041	-4	23	220 000	5.342
				-5	2	-5	0			-5	1		

## Vegetal products

Matrix	Strain	Level	Sample number	Reference method 16649-2				Alternative method REC2 37°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: 1,0x10 <sup>3</sup> CFU/g	ESC.1.2	300 CFU/g	L1T1R1	-1	6	-1	8	70	1.845	-1	16	150	2.176
				-2	0	-2	0						
			L1T1R2	-1	8	-1	4	60	1.778	-1	12	120	2.079
				-2	0	-2	0						
			L1T1R3	-1	6	-1	7	65	1.813	-1	6	60	1.778
				-2	0	-2	0						
			L1T1R4	-1	8	-1	7	75	1.875	-1	12	130	2.114
				-2	0	-2	0						
			L1T1R5	-1	9	-1	9	90	1.954	-1	18	190	2.279
				-2	1	-2	0						
		3000 CFU/g	L1T2R1	-2	34	-2	36	3 600	3.556	-2	62	6 200	3.792
				-3	5	-3	3						
			L1T2R2	-2	43	-2	40	4 300	3.633	-2	71	7 200	3.857
				-3	7	-3	4						
			L1T2R3	-2	34	-2	28	3 500	3.544	-2	78	8 400	3.924
				-3	9	-3	5						
			L1T2R4	-2	39	-2	40	3 800	3.580	-2	88	8 500	3.929
				-3	2	-3	2						
			L1T2R5	-2	45	-2	53	4 800	3.681	-2	60	6 000	3.778
				-3	3	-3	4						
		100000 CFU/g	L1T3R1	-4	58	-3	55	55 000	4.740	-4	19	200 000	5.301
				-5	3	-4	5						
			L1T3R2	-3	81	-3	112	96 000	4.982	-4	22	230 000	5.362
				-4	9	-4	10						
			L1T3R3	-3	108	-3	80	91 000	4.959	-4	13	130 000	5.114
				-4	8	-4	5						
			L1T3R4	-4	16	-4	9	130 000	5.114	-4	10	100 000	5.000
				-5	3	-5	1						
			L1T3R5	-4	15	-4	12	130 000	5.114	-4	12	130 000	5.114
				-5	1	-5	0						

## Composite foods

Matrix	Strain	Level	Sample number	Reference method 16649-2						Alternative method REC2 37°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: $1,0 \times 10^3$ CFU/g	ESC.1.28	3000 CFU/g	L1T1R1	-1	13	-1	11	110	2.041	-1	27	260	2.415
				-2	0	-2	0						
			L1T1R2	-1	15	-1	17	160	2.204	-1	17	150	2.176
				-2	1	-2	2						
			L1T1R3	-1	13	-1	15	140	2.146	-1	19	200	2.301
				-2	0	-2	2						
			L1T1R4	-1	15	-1	10	120	2.079	-1	24	240	2.380
				-2	2	-2	0						
			L1T1R5	-1	15	-1	13	150	2.176	-1	20	200	2.301
				-2	4	-2	1						
		3000 CFU/g	L1T2R1	-1	104	-1	113	1 100	3.041	-1	128	1 300	3.114
				-2	9	-2	11						
			L1T2R2	-1	113	-1	91	1 000	3.000	-1	115	1 200	3.079
				-2	13	-2	10						
			L1T2R3	-1	112	-1	108	1 100	3.041	-1	125	1 200	3.079
				-2	15	-2	9						
			L1T2R4	-1	148	-1	152	1 500	3.176	-1	135	1 400	3.146
				-2	13	-2	13						
			L1T2R5	-1	20	-1	18	1 900	3.279	-1	155	1 500	3.176
				-2	3	-2	1						
		100000 CFU/g	L1T3R1	-3	59	-3	55	55 000	4.740	-3	67	67 000	4.826
				-4	4	-4	3						
			L1T3R2	-3	49	-3	46	47 000	4.672	-3	39	40 000	4.602
				-4	6	-4	2						
			L1T3R3	-3	28	-3	38	34 000	4.531	-3	48	46 000	4.663
				-4	5	-4	3						
			L1T3R4	-3	44	-3	46	45 000	4.653	-3	38	37 000	4.568
-4	4			-4	5								
L1T3R5	-3		46	-3	48	45 000	4.653	-3	50	50 000	4.699		
	-4		1	-4	5								

## 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 <sup>2</sup> CFU/g	ESC.1.28	3000 CFU/g	L1T1R1	-1	7	-1	15	100	2.000	-1	22	240	2.380
				-2	0	-2	1			-2	4		
			L1T1R2	-1	7	-1	11	90	1.954	-1	20	190	2.279
				-2	0	-2	1			-2	1		
			L1T1R3	-1	8	-1	19	130	2.114	-1	15	150	2.176
				-2	0	-2	2			-2	1		
		L1T1R4	-1	10	-1	10	100	2.000	-1	15	150	2.176	
			-2	0	-2	3			-2	1			
		L1T1R5	-1	10	-1	16	120	2.079	-1	17	160	2.204	
			-2	0	-2	1			-2	1			
		3000 CFU/g	L1T2R1	-1	113	-1	106	1 100	3.041	-1	96	990	2.996
				-2	12	-2	9			-2	13		
			L1T2R2	-1	130	-1	142	1 400	3.146	-1	134	1 300	3.114
				-2	15	-2	19			-2	14		
			L1T2R3	-1	123	-1	167	1 500	3.176	-2	145	1 500	3.176
				-2	10	-2	19			-3	19		
		L1T2R4	-1	123	-1	119	1 200	3.079	-1	111	1 100	3.041	
			-2	14	-2	9			-2	14			
		L1T2R5	-2	14	-2	18	1 600	3.204	-2	20	1 900	3.279	
			-3	1	-3	2			-3	1			
		100000 CFU/g	L1T3R1	-3	53	-3	56	54 000	4.732	-3	56	55 000	4.740
				-4	5	-4	5			-4	5		
			L1T3R2	-3	52	-3	63	57 000	4.756	-3	61	61 000	4.785
				-4	5	-4	5			-4	6		
			L1T3R3	-3	38	-3	39	37 000	4.568	-3	40	39 000	4.591
				-4	2	-4	3			-4	3		
		L1T3R4	-3	44	-3	62	53 000	4.724	-3	62	63 000	4.799	
-4	5		-4	5	-4	7							
L1T3R5	-3	55	-3	42	49 000	4.690	-3	54	53 000	4.724			
	-4	5	-4	5			-4	4					

**Environmental samples**

Matrix	Strain	Level	Sample number	Reference method 16649-2 <sup>a</sup>				Alternative method REC2 37°C			
				Dilution	CFU / Petri dish	CFU/g	log CFU/g	Dilution	CFU / Petri dish	CFU/g	log CFU/g
Process water - Batch 1 - TVC result: 2.1 10. <sup>3</sup> CFU/g	YMJ695	300 CFU/g	2347716	-1	13	140	2.15	-1	17	180	2.26
				-2	2			-2	3		
			2347717	-1	26	270	2.43	-1	22	200	2.30
				-2	4			-2	0		
			2347718	-1	16	160	2.20	-1	26	250	2.40
				-2	1			-2	1		
		2347719	-1	15	190	2.28	-1	11	130	2.11	
			-2	6			-2	3			
		2347720	-1	17	160	2.20	-1	12	130	2.11	
			-2	1			-2	2			
		3000 CFU/g	2347721	-2	19	2000	3.30	-2	26	2600	3.41
				-3	3			-3	2		
			2347722	-2	18	1800	3.26	-2	14	1400	3.15
				-3	2			-3	1		
			2347723	-2	13	1400	3.15	-2	16	1700	3.23
				-3	2			-3	3		
		2347724	-2	19	1700	3.23	-2	20	1900	3.28	
			-3	0			-3	1			
		2347725	-2	18	1700	3.23	-2	14	1400	3.15	
			-3	2			-3	1			
		100000 CFU/g	2347726	-3	63	63000	4.80	-3	71	68000	4.83
				-4	6			-4	4		
			2347727	-3	41	40000	4.60	-3	48	49000	4.69
				-4	3			-4	6		
2347728	-3		48	51000	4.71	-3	51	52000	4.72		
	-4		8			-4	6				
2347729	-3	36	38000	4.58	-3	51	52000	4.72			
	-4	6			-4	6					
2347730	-3	31	29000	4.46	-3	48	52000	4.72			
	-4	1			-4	9					

## Environmental samples

Matrix	Strain	Level	Sample number	Reference method 16649-2*				Alternative method REC2 37°C			
				Replicate 1		CFU/g	log CFU/g	Dilution	CFU / Petri dish	CFU/g	log CFU/g
				Dilution	CFU / Petri dish						
Process water - Batch 2 - TVC result: $9.5 \cdot 10^3$ CFU/g	YMJ695	300 CFU/g	2347731	-1	20	190	2.28	-1	24	250	2.40
				-2	1			-2	3		
			2347732	-1	19	190	2.28	-1	12	130	2.11
				-2	2			-2	2		
			2347733	-1	12	120	2.08	-1	15	140	2.15
				-2	1			-2	0		
		2347734	-1	16	160	2.20	-1	19	190	2.28	
			-2	1			-2	2			
		2347735	-1	19	180	2.26	-1	19	190	2.28	
			-2	1			-2	2			
		3000 CFU/g	2347736	-2	17	1600	3.20	-2	14	1500	3.18
				-3	1			-3	2		
			2347737	-2	22	2200	3.34	-2	26	2500	3.40
				-3	2			-3	1		
			2347738	-2	20	1900	3.28	-2	10	1100	3.04
				-3	1			-3	2		
		2347739	-2	19	1800	3.26	-2	15	1400	3.15	
			-3	1			-3	0			
		2347740	-2	22	2000	3.30	-2	12	1300	3.11	
			-3	0			-3	2			
		100000 CFU/g	2347741	-3	49	50000	4.70	-3	58	56000	4.75
				-4	6			-4	4		
			2347742	-3	39	42000	4.62	-3	61	64000	4.81
				-4	7			-4	9		
2347743	-3		42	41000	4.61	-3	60	61000	4.79		
	-4		3			-4	7				
2347744	-3	50	52000	4.72	-3	45	46000	4.66			
	-4	7			-4	6					
2347745	-3	49	49000	4.69	-3	50	50000	4.70			
	-4	5			-4	5					

## Animal feed

Matrix	Strain	Level	Sample number	Reference method 16649-2 <sup>*</sup>				Alternative method REC2 37°C			
				Dilution	CFU / Petri dish	CFU/g	log CFU/g	Dilution	CFU / Petri dish	CFU/g	log CFU/g
Dog pâté - Batch 1 - TVC result: <10 CFU/g	GQRP82	300 CFU/g	2392426	-1	22	220	2.34	-1	21	200	2.30
				-2	2			-2	1		
			2392427	-1	29	330	2.52	-1	21	240	2.38
				-2	7			-2	5		
			2392428	-1	20	190	2.28	-1	11	110	2.04
				-2	1			-2	1		
		2392429	-1	19	200	2.30	-1	14	140	2.15	
			-2	3			-2	1			
		2392430	-1	33	340	2.53	-1	18	170	2.23	
			-2	4			-2	1			
		3000 CFU/g	2392431	-2	27	3100	3.49	-2	23	2200	3.34
				-3	7			-3	1		
			2392432	-2	28	2800	3.45	-2	13	1400	3.15
				-3	3			-3	2		
			2392433	-2	13	1300	3.11	-2	23	2100	3.32
				-3	1			-3	0		
		2392434	-2	20	2000	3.30	-2	19	1700	3.23	
			-3	2			-3	0			
		2392435	-2	24	2500	3.40	-2	14	1500	3.18	
			-3	3			-3	2			
		100000 CFU/g	2392436	-3	135	130000	5.11	-3	94	89000	4.95
				-4	11			-4	4		
			2392437	-3	107	110000	5.04	-3	96	100000	5.00
				-4	11			-4	15		
2392438	-3		111	110000	5.04	-3	82	81000	4.91		
	-4		12			-4	7				
2392439	-3		109	120000	5.08	-3	75	80000	4.90		
	-4		20			-4	13				
2392440	-3		98	95000	4.98	-3	83	86000	4.93		
	-4		7			-4	12				



## Animal feed

Matrix	Strain	Level	Sample number	Reference method 16649-2*				Alternative method REC2 37°C			
				Replicate 1		CFU/g	log CFU/g	Dilution	CFU / Petri dish	CFU/g	log CFU/g
				Dilution	CFU / Petri dish						
Dog pâté - Batch 2 - TVC result: <10 CFU/g	GQRP82	300 CFU/g	2392441	-1	21	220	2.34	-1	9	90	1.95
				-2	3			-2	1		
			2392442	-1	23	230	2.36	-1	13	140	2.15
				-2	2			-2	3		
			2392443	-1	14	160	2.20	-1	11	110	2.04
				-2	3			-2	1		
		2392444	-1	28	270	2.43	-1	21	190	2.28	
			-2	2			-2	0			
		2392445	-1	27	260	2.41	-1	15	160	2.20	
			-2	2			-2	3			
		3000 CFU/g	2392446	-2	21	2200	3.34	-2	23	2500	3.40
				-3	3			-3	5		
			2392447	-2	28	2600	3.41	-2	15	1500	3.18
				-3	1			-3	2		
			2392448	-2	29	2900	3.46	-2	19	1800	3.26
				-3	3			-3	1		
		2392449	-2	19	2300	3.36	-2	17	1500	3.18	
			-3	6			-3	0			
		2392450	-2	22	2400	3.38	-2	13	1300	3.11	
			-3	4			-3	1			
		100000 CFU/g	2392451	-3	134	130000	5.11	-3	59	64000	4.81
				-4	9			-4	11		
			2392452	-3	103	110000	5.04	-3	83	84000	4.92
				-4	16			-4	9		
			2392453	-3	117	120000	5.08	-3	75	79000	4.90
				-4	12			-4	12		
		2392454	-3	111	110000	5.04	-3	81	80000	4.90	
-4	13		-4	7							
2392455	-3	120	130000	5.11	-3	80	84000	4.92			
	-4	20			-4	13					

## Appendix G

### Inclusivity/Exclusivity – Raw results

## SELECTIVITY - INITIAL VALIDATION

Souche	Origine	PCA à 30°C		REC2 à 37°C	
		Culture	Nombre de colonies	Couleur des colonies	Nombre de colonies
<i>Citrobacter diversus</i>	Aliments animaux	Positive	146	bleu	120
		Positive	132	bleu	112
<i>Citrobacter diversus</i>	Herbes séchées	Positive	136	bleu	152
		Positive	143	bleu	140
<i>Citrobacter diversus</i>	Levure	Positive	164	bleu	123
		Positive	112	bleu	99
<i>Citrobacter freundii</i>	Produit carné	Positive	39	bleu	40
		Positive	55	bleu	58
<i>Citrobacter freundii</i>	Végétaux	Positive	38	bleu	44
		Positive	59	bleu	56
<i>Citrobacter freundii</i>	Poisson	Positive	33	bleu	46
		Positive	51	bleu	50
<i>Citrobacter freundii</i>	Lait	Positive	66	bleu	67
		Positive	90	bleu	93
<i>Enterobacter amnigenus</i>	Brochette de poisson	Positive	96	bleu	87
		Positive	71	bleu	76
<i>Enterobacter amnigenus</i>	Jambon	Positive	60	bleu	70
		Positive	67	bleu	73
<i>Enterobacter cloacae</i>	Produit laitier	Positive	86	bleu	91
		Positive	81	bleu	75
<i>Enterobacter cloacae</i>	Produit laitier	Positive	48	bleu	50
		Positive	63	bleu	42
<i>Enterobacter cloacae</i>	Produit laitier	Positive	118	bleu	127
		Positive	110	bleu	124
<i>Enterobacter cloacae</i>	Produit laitier	Positive	88	bleu	105
		Positive	91	bleu	86
<i>Enterobacter sakazakii</i>	Aliments animaux	Positive	135	bleu	140
		Positive	113	bleu	104
<i>Enterobacter sakazakii</i>	Pâtisserie	Positive	75	bleu	85
		Positive	59	bleu	51
<i>Erwinia spp.</i>	Collection	Positive	23	blanc-gris	22
		Positive	112	blanc-gris	98
<i>Escherichia coli</i> O157	Collection	Positive	36	bleu	35
		Positive	103	bleu	89
<i>Escherichia hermanii</i>	Aliments animaux	Positive	56	bleu-gris	44
		Positive	102	bleu-gris	67
<i>Escherichia hermanii</i>	Produit carné	Positive	50	bleu	56
		Positive	143	bleu	124
<i>Escherichia hermanii</i>	Produit laitier	Positive	22	bleu	19
		Positive	89	bleu	78
<i>Escherichia vulneris</i>	Produit carné	Positive	31	bleu	14
		Positive	112	bleu	86
<i>Hafnia alvei</i>	Foie de porc	Positive	90	bleu très clair	90
		Positive	82	bleu-gris	69
<i>Hafnia alvei</i>	Reblochon	Positive	80	blanc	95
		Positive	75	blanc	71
<i>Hafnia alvei</i>	Persil	Positive	75	blanc	90
		Positive	76	bleu-gris	81
<i>Hafnia alvei</i>	Flétan	Positive	84	blanc	75
		Positive	50	bleu-gris	65
<i>Hafnia alvei</i>	Viande hachée	Positive	127	bleu-gris	134
		Positive	119	bleu-gris	124
<i>Hafnia alvei</i>	Lait cru	Positive	109	bleu	90
		Positive	134	bleu	132
<i>Hafnia alvei</i>	Echine de porc	Positive	126	bleu-gris	118
		Positive	97	bleu-gris	94
<i>Hafnia alvei</i>	Rognons de porc	Positive	150	blanc	132
		Positive	101	blanc	99

**SELECTIVITY - INITIAL VALIDATION**

Souche	Origine	PCA à 30°C		REC2 à 37°C	
		Culture	Nombre de colonies	Couleur des colonies	Nombre de colonies
<i>Hafnia alvei</i>	Persil	Positive	123	bleu-gris	135
		Positive	59	bleu-gris	59
<i>Hafnia alvei</i>	Brochette de poisson	Positive	134	bleu-gris	115
		Positive	128	bleu-gris	109
<i>Hafnia alvei</i>	Concombre	Positive	121	bleu-gris	121
		Positive	107	bleu-gris	100
<i>Hafnia alvei</i>	Tomate	Positive	150	bleu-gris	147
		Positive	89	bleu-gris	82
<i>Klebsiella oxytoca</i>	Aliments animaux	Positive	40	bleu	54
		Positive	134	bleu	133
<i>Klebsiella oxytoca</i>	Poisson	Positive	62	bleu	65
		Positive	36	bleu	41
<i>Klebsiella pneumoniae</i>	Poudre de lait	Positive	42	bleu	40
		Positive	36	bleu	42
<i>Klebsiella pneumoniae</i>	Macédoine	Positive	55	bleu	58
		Positive	38	bleu	37
<i>Moellerella wisconsensis</i>	Andouillette	Positive	45	bleu	80
		Positive	46	bleu	56
<i>Proteus mirabilis</i>	Produit carné	Positive	93	gris	72
		Positive	145	gris	132
<i>Proteus mirabilis</i>	Foies de volaille	Positive	115	blanc	93
		Positive	102	blanc	100
<i>Pseudomonas aeruginosa</i>	Filet de rouget	Positive	30	blanc	26
		Positive	87	blanc	77
<i>Salmonella arizonae</i> IIIb 61:--	Dinde	Positive	66	violet	72
		Positive	45	violet	32
		Positive	75	violet	63
		Positive	64	violet	46
<i>Salmonella arizonae</i> IIIa 48:24:223	Elevage d'oie	Positive	116	bleu	115
		Positive	76	bleu	79
<i>Salmonella arizonae</i> IIIb 61:i:z53	Cuisse de poulet	Positive	96	violet	90
		Positive	97	violet	90
		Positive	112	violet	96
		Positive	85	violet	84
<i>Salmonella</i> Enteritidis	Ovoproduit	Positive	121	blanc-gris	103
		Positive	105	blanc-gris	100
<i>Salmonella</i> Typhimurium	Foie de porc	Positive	83	blanc-gris	70
		Positive	103	blanc-gris	79
<i>Serratia marcescens</i>	Lait cru	Positive	45	bleu	33
		Positive	55	bleu	34
<i>Serratia liquefaciens</i>	Andouillette	Positive	63	bleu	51
		Positive	45	bleu-gris	35
<i>Shigella flexneri</i>	Collection	Positive	25	blanc-gris	22
		Positive	56	blanc-gris	44
<i>Shigella flexneri</i>	Collection	Positive	30	bleu-gris	41
		Positive	35	bleu-gris	40
<i>Shigella sonnei</i>	Collection	Positive	26	violet	30
		Positive	33	violet	27
		Positive	39	violet	33
		Positive	21	violet	19
<i>Yersinia kristensenii</i>	Collection	Positive	27	blanc	16
		Positive	67	blanc	46
<i>Yersinia enteritidis</i>	Ovoproduit	Positive	12	blanc	0
		Positive	34	blanc	0
<i>Yersinia enterocolitica</i>	Ovoproduit	Positive	28	blanc	20
		Positive	45	blanc	43

TBX à 44°C		
Couleur des colonies	des colonies	Nombre de colonies
bleu		56
bleu		31

TBX à 44°C		
Couleur des colonies	des colonies	Nombre de colonies
bleu		63
bleu		55

TBX à 44°C		
Couleur des colonies	des colonies	Nombre de colonies
bleu		23
bleu		11

## SELECTIVITY - INITIAL VALIDATION

Souche	Origine	PCA à 30°C		REC2 à 37°C	
		Culture	Nombre de colonies	Couleur des colonies	Nombre de colonies
<i>Escherichia coli</i>	Rognons de porc	Positive	60	violet	45
		Positive	74	violet	71
<i>Escherichia coli</i>	Chou rouge	Positive	47	violet	41
		Positive	70	violet	72
<i>Escherichia coli</i>	Persil	Positive	89	violet	88
		Positive	88	violet	78
<i>Escherichia coli</i>	Pâtisserie	Positive	123	violet	121
		Positive	122	violet	109
<i>Escherichia coli</i>	Crépinette	Positive	56	violet	53
		Positive	106	violet	98
<i>Escherichia coli</i>	Crépinette	Positive	111	violet	114
		Positive	145	violet	137
<i>Escherichia coli</i>	Chair à saucisse	Positive	134	violet	123
		Positive	98	violet	94
<i>Escherichia coli</i>	Tomate	Positive	87	violet	69
		Positive	84	violet	99
<i>Escherichia coli</i>	Céleri rémoulade	Positive	79	violet	78
		Positive	105	violet	111
<i>Escherichia coli</i>	Crème vanille	Positive	65	violet	60
		Positive	126	violet	120
<i>Escherichia coli</i>	Merguez	Positive	143	violet	141
		Positive	96	violet	98
<i>Escherichia coli</i>	Foie de porc	Positive	54	violet	56
		Positive	107	violet	110
<i>Escherichia coli</i>	Lait cru	Positive	67	violet	66
		Positive	122	violet	121
<i>Escherichia coli</i>	Fromage au lait cru	Positive	132	violet	142
		Positive	141	violet	129
<i>Escherichia coli</i>	Moules	Positive	128	violet	108
		Positive	109	violet	100
<i>Escherichia coli</i>	Chair à saucisse	Positive	76	violet	71
		Positive	56	violet	59
<i>Escherichia coli</i>	Carottes râpées	Positive	97	violet	89
		Positive	89	violet	81
<i>Escherichia coli</i>	Pâtisserie	Positive	104	violet	100
		Positive	76	violet	77
<i>Escherichia coli</i>	Chou à la crème	Positive	57	violet	61
		Positive	98	violet	91
<i>Escherichia coli</i>	Crème pâtissière	Positive	113	violet	101
		Positive	73	violet	77
<i>Escherichia coli</i>	Taboulé	Positive	93	violet	91
		Positive	91	violet	84
<i>Escherichia coli</i>	Crème chantilly	Positive	71	violet	67
		Positive	107	violet	100
<i>Escherichia coli</i>	Lardons	Positive	132	violet	129
		Positive	99	violet	94
<i>Escherichia coli</i>	Saumon fumé	Positive	96	violet	91
		Positive	141	violet	136
<i>Escherichia coli</i>	Filet de rouget	Positive	153	violet	152
		Positive	38	violet	30
<i>Escherichia coli</i>	Sandwich	Positive	49	violet	46
		Positive	99	violet	91
<i>Escherichia coli</i>	Pâtisserie	Positive	100	violet	99
		Positive	116	violet	109
<i>Escherichia coli</i>	Foie de porc	Positive	113	violet	111
		Positive	125	violet	123
<i>Escherichia coli</i>	Rognons de porc	Positive	88	violet	85
		Positive	94	violet	93
<i>Escherichia coli</i>	Lait cru	Positive	111	violet	100
		Positive	114	violet	109

## Inclusivity 2017

Strain	Reference	Origin	PCA (CFU/mL)		ISO 16649-2 ° (CFU/mL)		REC2 at 37°C ( CFU/mL)	
			R1	R2	R1	R2	R1	R2
<i>Escherichia coli</i>	ESC.1.11	Chicken fillet	97	95	98	100	101	99
<i>Escherichia coli</i>	ESC.1.111	Fountain water	89	93	78	82	82	89
<i>Escherichia coli</i>	ESC.1.12	Lamb necklance	54	60	49	49	52	54
<i>Escherichia coli</i>	ESC.1.13	Croissant stuffed	86	80	84	76	81	76
<i>Escherichia coli</i>	ESC.1.139	River water	85	85	83	80	80	91
<i>Escherichia coli</i>	ESC.1.140	River water	52	59	49	49	51	43
<i>Escherichia coli</i>	ESC.1.145	Crudeness	125	111	118	120	119	124
<i>Escherichia coli</i>	ESC.1.146	Cheese	47	47	39	44	41	46
<i>Escherichia coli</i>	ESC.1.15	Beef muscle	103	105	99	104	101	112
<i>Escherichia coli</i>	ESC.1.18	"Rillettes" of cooked and smoked salmon	98	94	89	78	86	91
<i>Escherichia coli</i>	ESC.1.19	Shrimp ravioli	49	48	48	40	49	53
<i>Escherichia coli</i>	ESC.1.2	Grated carrot	122	130	113	116	118	107
<i>Escherichia coli</i>	ESC.1.20	Meat for kebab	119	119	115	106	117	100
<i>Escherichia coli</i>	ESC.1.21	Beef granules with 20% of fat	38	45	40	40	36	35
<i>Escherichia coli</i>	ESC.1.26	Vegetable sauce	38	39	35	33	35	46
<i>Escherichia coli</i>	ESC.1.28	Mixed salad	123	126	118	113	123	129
<i>Escherichia coli</i>	ESC.1.3	Dairy industry	56	62	48	49	46	46
<i>Escherichia coli</i>	ESC.1.5	Cheese	56	53	51	56	52	55
<i>Escherichia coli</i>	ESC.1.50	Cheese	49	42	46	45	44	53
<i>Escherichia coli</i>	ESC.1.6	Chicken ravioli	112	108	101	104	98	98