Method Comparison Study Report for the ISO 16140-2:2016 validation of Compact Dry EC, for the enumeration of coliforms in a broad range of foods, environmental samples and pet food and animal feed

MicroVal study number: 2008LR04

Method/Kit name: CompactDry EC

Report version: MCS/ILS extension summary report

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The report is prepared in accordance with ISO 16140-2:20016 and the most recent version of the MicroVal Technical Committee for interpretation on ISO 16140-2.

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Method/Kit name: CompactDry EC

Validation standard: Microbiology of the food chain— Method validation

Part 1: Vocabulary (ISO 16140-1:2016) and

Part 2: Protocol for the validation of alternative (proprietary) methods against a reference method (ISO 16140-2:2016)

Reference methods: ISO 16649-2:2001 Microbiology of food and animal feeding stuffs — Horizontal method for the enumeration of beta-glucuronidase-positive Escherichia coli — Part 2: Colony-count technique at 44 degrees C using 5-bromo-4-chloro-3-indolyl beta-D-glucuronide

ISO 4832:2006 Microbiology of food and animal feeding stuffs: Horizontal method for the enumeration of coliforms - Colony Count Method for coliforms**.**

Scope of validation: 2 additional categories (Environmental samples and pet food and animal feed**)**

Certification organization: LRQA

List of abbreviations

- AL Acceptability Limit
- AP Accuracy Profile
- Art. Cont. Artificial contamination
- CFU Colony Forming Units
- CL confidence limit (usually 95%)
- EL Expert Laboratory
- $\bar{D}$ Average difference
- g Gram
- h Hour
- ILS Interlaboratory Study
- Inc/Ex Inclusivity and Exclusivity
- LOQ Level of Quantification
- MCS Method Comparison Study
- min minute
- ml Millilitre
- MR (MicroVal) Method Reviewer
- MVTC MicroVal Technical Committee
- EL Expert Laboratory
- n number of samples
- na not applicable
- neg negative (target not detected)
- NG no growth
- nt not tested
- RT Relative Trueness
- SD standard deviation of differences
- 10^{-1} dilution 10-fold dilution of original food
- 10^{-2} dilution 100-fold dilution of original food
- BPW Buffered Peptone Water
- PSD Peptone salt diluent
- MRD Maximum Recovery Diluent
- NA Nutrient Agar
- PCA Plate count Agar

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1 Introduction

In this project a MicroVal validation study, based on ISO 16140-2:2016, of alternative method(s) for the enumeration of coliforms in broad range of foods was carried out by Campden BRI as the MicroVal Expert Laboratory.

The original study was conducted following ISO16140:2013 and on renewal, additional work on relative trueness and accuracy profile was done to fulfil the requirements of ISO 16140-2:2016. As the design of the ILS remained unchanged between the 2 versions of the 16140 protocol the orignal data was analysed according to the new statistical approach outlined in ISO16140-2:2016.

An extension study was carried out at the request of the manufacturer to include 2 further categories (Environmental samples and pet food and animal feed) to extend the scope of the validation for this media. This study was completed in June 2023 by Campden BRI as the MicroVal Expert Laboratory.

The alternative method used is:

Compact Dry (Shimadzu Diagnostics Corporation) are ready-to-use dry media sheets comprising culture medium and a cold-soluble gelling agent, rehydrated by inoculating 1 ml diluted sample into the centre of the self-diffusible medium. This is a ready to use, chromogenic plate for the enumeration of *E. coli* and coliforms. These organisms are differentiated by the colony morphology. *E. coli* colonies form blue colonies and other coliforms form red colonies after the required incubation period of 24h ±2h at 37°C ±1°C.

Reference method is:

ISO 4832:2006 Microbiology of food and animal feeding stuffs: Horizontal method for the enumeration of coliforms - Colony Count Method for coliforms.

Scope of the validation study is: broad range of foods, environmental samples and pet food

Categories included:

- Milk and dairy products
- Fresh produce and fruits
- Raw poultry and meats (Combined category raw/ RTC meats and poultry)
- Ready to eat foods (Combined category RTE/RTRH meats and poultry)
- Multi component foods or meal components
- Environmental samples
- Pet food and animal feed

Criteria to be evaluated during the study:

- Method Comparison Study (MCS)
  - Relative Trueness study
  - Accuracy profile study
  - Inclusivity and exclusivity study
  - Interlaboratory Study (ILS)

Conclusions for the study

Overall, the conclusions for the Method Comparison are:

- The alternative method Compact Dry EC for enumeration of coliforms shows satisfactory results for relative trueness;
- The alternative Compact Dry EC for the enumeration of coliforms shows satisfactory results for accuracy profile;

- The Compact Dry EC for the enumeration of coliforms in foods method was shown to be specific and selective.
- The alternative method Compact Dry EC for the enumeration of coliforms shows satisfactory results for the ILS

- The alternative method Compact Dry EC for the enumeration of coliforms shows comparable performance to the reference method ISO 4832:2006 Microbiology of food and animal feeding stuffs: Horizontal method for the enumeration of coliforms - Colony Count Method.

2 Method protocols

The Method Comparison Study was carried out using 10 gram portions of sample material.

2.1 Reference method

See the flow diagram in Annex A.

Sample preparations used in the reference method was done according to ISO 6887-series parts 1, 2, 3, 4 and 5. Plating was done according to ISO 7218:2007+A1:2013 section 10.2.2 which says at least one plate per dilution shall be used with at least two successive dilutions. If only one dilution was used, then two plates of this dilution were used to improve reliability of the results. Depending on the sample being tested and the expected contamination level, single or multiple dilutions were used with single or duplicate plates if considered necessary to improve the reliability of the calculated result and ensure at least two relevant plates were available for use in calculations.

All samples were incubated at 37°C ±1°C during the study for the reference method. In ISO 4832 there are 2 incubation temperatures listed that could be used for dairy samples (30°C and 37°C), and in this study, the plates were incubated at 37°C ±1°C.

2.2 Alternative method

See the flow diagram of the alternative method in Annex A.

See the kit insert in Annex B.

Compact Dry (Shimadzu Diagnostics Corporation) are ready-to-use dry media sheets comprising culture medium and a cold-soluble gelling agent, rehydrated by inoculating 1 ml diluted sample into the centre of the self-diffusible medium. This is a ready to use, chromogenic plate for the enumeration of *E. coli* and coliforms. These organisms are differentiated by the colony morphology. *E. coli* colonies form blue colonies and other coliforms form red colonies after the required incubation period of 24h ±2h at 37°C ±1°C

2.3 Study design

According to ISO 16140-2 the reference method and alternative methods were performed with, as far as possible, exactly the same sample

Samples of product containing the target organism were diluted 1 in 10 with an appropriate diluent according to ISO 6887 and homonegised in a stomacher. Appropriate serial dilutions were made and all relevant dilutions were analysed using the reference method and alternative method.

3 Method comparison study

3.1 Relative trueness study

The trueness study is a comparative study between the results obtained by the reference method and the results of the alternative method. This study was conducted using naturally or artificially contaminated samples. Different categories, types and items were tested for this.

A total of 2 categories were included in this validation study. A minimum of 15 items for each category were tested by both the reference method and the alternative method in the relative trueness study, with a minimum of 15 interpretable results per category.

Each category was made up of 3 types, with at least 5 items representative for each type.

3.1.1 Number of samples

The categories, the types and the number of samples analyzed are presented in Table 1.

Table 1 – Categories, types and number of samples analysed

Category	Types	Items	No of samples	ISO 6887
Milk and dairy products	Dry milk product	milk powder, powder for milk based desserts, dried infant formula	5	6887-5
	Dairy products	ice-cream, yogurts, cream, hard cheese, soft cheese, raw milk cheese	5	6887-5
	Pasteurised milk products	skimmed, semi-skimmed, full fat and flavoured milks	5	6887-5
Fresh produce and fruits	Cut ready to eat fruit	fruit mixes, fruit juices	5	6887-4
	Cut ready to eat vegetables	Bagged pre-cut salads and shredded carrot, cabbage, vegetable juices	5	6887-4
	Leafy greens/Sprouts	soy, mung, alfalfa,	5	6887-4
Raw poultry and meats (Combined category raw/ RTC meats and poultry)	Fresh poultry cuts	turkey breast, turkey fillet	5	6887-2
	Fresh mince	lamb, beef, pork	5	6887-2
	Processed ready to cook	frozen patties, marinated kebabs, seasoned chicken breasts	5	6887-2
Ready to eat foods (Combined category	Ready to eat poultry	turkey fillet, chicken sausage, pate	5	6887-2
	Cooked fish products	prawns, terrine, pate, smoked fish	5	6887-3

Category	Types	Items	No of samples	ISO 6887
RTE/RTRH meats and poultry)	Cooked meat	ham, salami, pate, corned beef	5	6887-2
Multi component foods or meal components	Ready to re-heat refrigerated food	cooked chilled foods, rice and pasta, products	5	6887-1, 6887-4
	Ready to re-heat food frozen	fries, pizza	5	6887-1, 6887-4
	Composite foods with substantial raw ingredients	pasta salads, sandwiches, deli-salads		6887-1, 6887-4
Environmental samples (food or feed production)	Surfaces (wipes, swabs)	Equipment, floors, walls	5	ISO 18593
	Process water	Wash water, cooling water	5	6887-6
	Dust	Bakery and food manufacturing environment	5	6887-1, 6887-4
Pet food and animal feed	Dry food	Pellets, kibbles, treats	5	6887-4
	Wet food (raw and canned)	Pates, sausages	5	6887-4
	Animal feeds (poultry and fish)	Cereals and flours	5	6887-4

105 samples were analyzed, leading to 105 exploitable results.

3.1.2 Test sample preparation- coliforms

Samples were screened for natural contamination to ensure suitable levels of contamination for the study. None of the samples tested during the screening phase were naturally contaminated with colifoms. It was therefore necessary to use artificial contamination procedures.

Further details of the artificial inoculation used in the studies is given in the table below

Sample type	Procedure for artificial contamination
Chilled dairy products, fresh produce and fruits, ready to eat foods, fresh meat, chilled multicomponent foods and process water	Seeding and storage of samples post inoculation for 48h ±2h at 2-8°C to chill stress the cells
Frozen meat products and multicomponent foods	Seeding and storage of samples post inoculation for 2 weeks at -18°C

Sample type	Procedure for artificial contamination
Surfaces and dust samples	Seeding and storage of samples post inoculation for 2 weeks at ambient
Dry milk powder and animal feed	Seeding with lyophilised cells and storage for 2 weeks at ambient
Pasteurised milk products, wet pet food	Spiked with heat stressed cultures with a minimum of 0.5log injury. Note: The level of injury was evaluated by plating on a non-selective agar e.g. Nutrient Agar and a selective agar e.g. Nutrient Agar plus 3% salt. The observed injury measurements varied from 0.60 to 1.02 log cfu/g difference between non-selective and selective plates.

Twenty two coliform isolates were used for the artificial contamination in a mixture of seeding and spiking protocols. Each isolate was used to contaminate no more than 5 items during the study.

Inoculation of samples was at the range usually associated with the test organisms and within the capabilities of the test methods, covering the range 10^2cfu/g to 10^7cfu/g for food samples and 10^2cfu/g to 10^7cfu/g for food samples and 10^1cfu/surface to 10^6cfu/surface for surface samples.

3.1.3 Protocols applied during the validation study

Where required presumptive coliform colonies were confirmed in the reference method following the protocol described in ISO 4832. Up to five atypical colonies were inoculated into tubes of brilliant green lactose bile broth which was then incubated at 37°C for 24 ±2h. Lactose fermentation was confirmed if gas formation has occurred in the Durham tube and the colour of the broth changed from green to yellow.

The Alternative method was incubated at 37±1°C for 22 hours.

Test results:

The samples were analysed by the reference and the alternative methods in order to have 15 interpretable results per incubation protocol, and 5 interpretable results per tested type.

The obtained data were analyzed using the scatter plot. The graphs are provided with the line of identity (y = x).

Figure 1 shows the scatter plot for coliforms in Dairy products

Figure 2 shows the scatter plot for coliforms in Fresh produce and fruits

Figure 3 shows the scatter plot for coliforms in Raw meat and poultry

Figure 4 shows the scatter plot for coliforms in Ready to eat foods

Figure 5 shows the scatter plot for coliforms in multicomponent foods

Figure 6 shows the scatter plot for coliforms in Environmental samples (food or feed production)

Figure 7 shows the scatter plot for coliforms in Pet food and animal feed.

The Figure 7 shows the scatter plot for all the categories.

Figure 1 - Scatter plot of the reference method versus alternative method results for coliforms in Dairy products

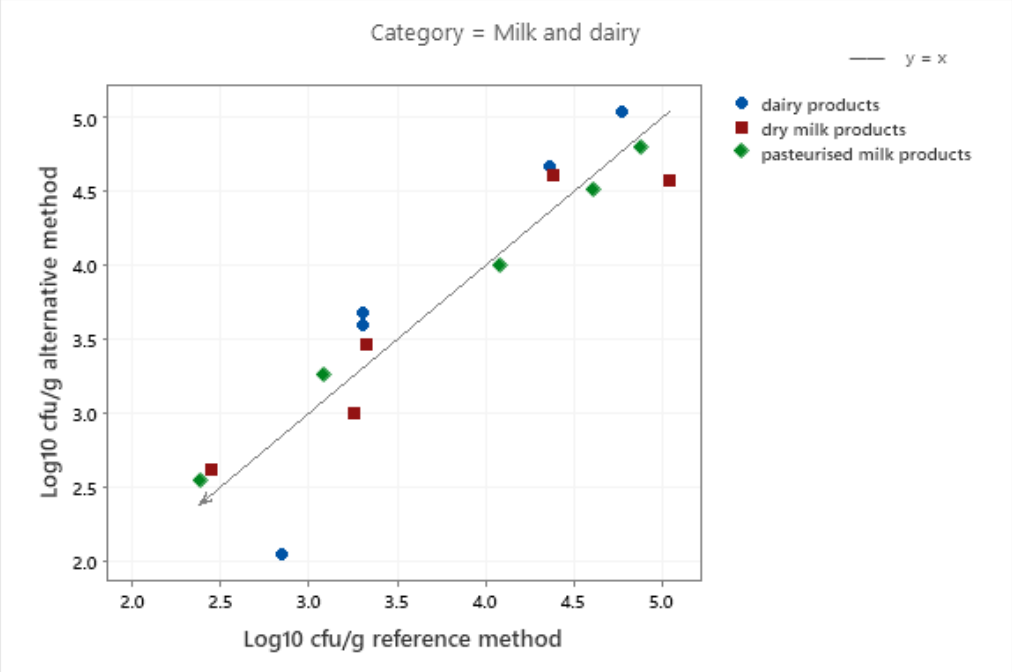


Figure 2 - Scatter plot of the reference method versus alternative method results for coliforms Fresh produce and fruits

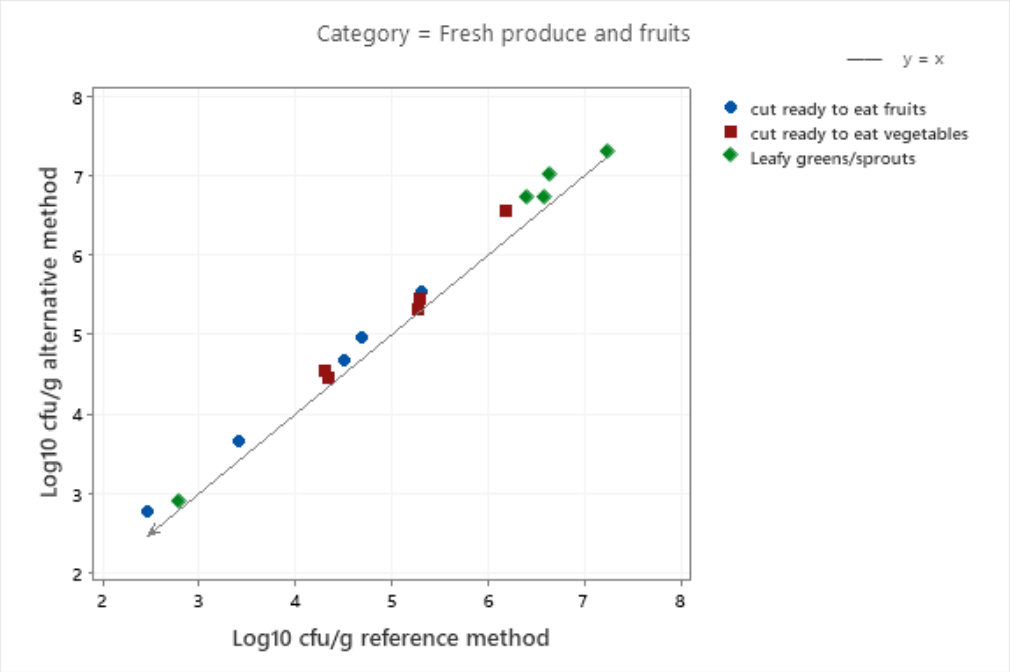


Figure 3 - Scatter plot of the reference method versus alternative method results for coliforms in Raw meat and Poultry

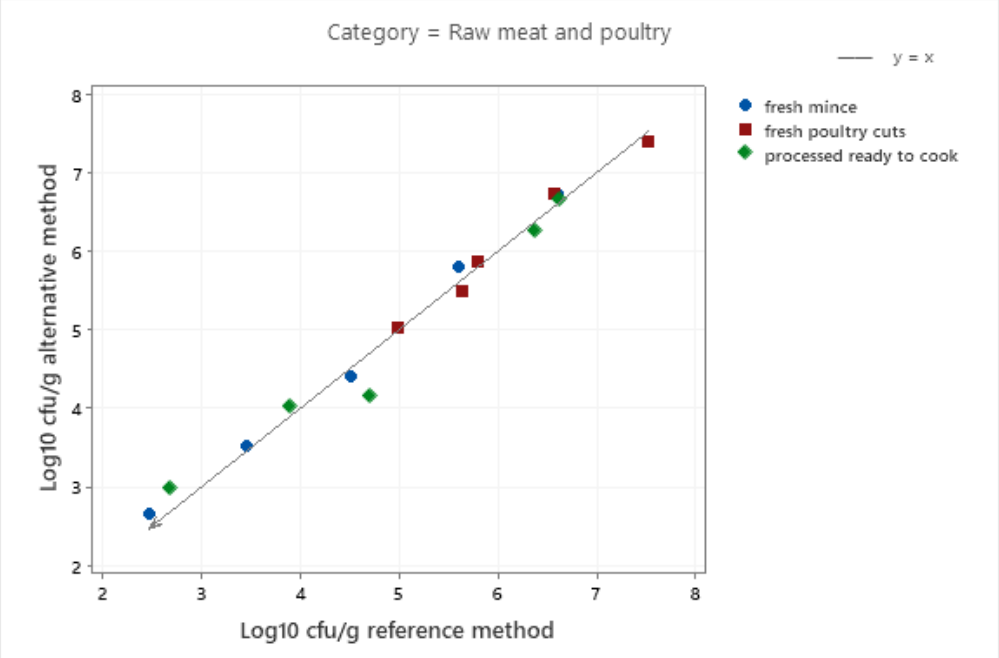


Figure 4 - Scatter plot of the reference method versus alternative method results for coliforms in Ready to Eat Foods

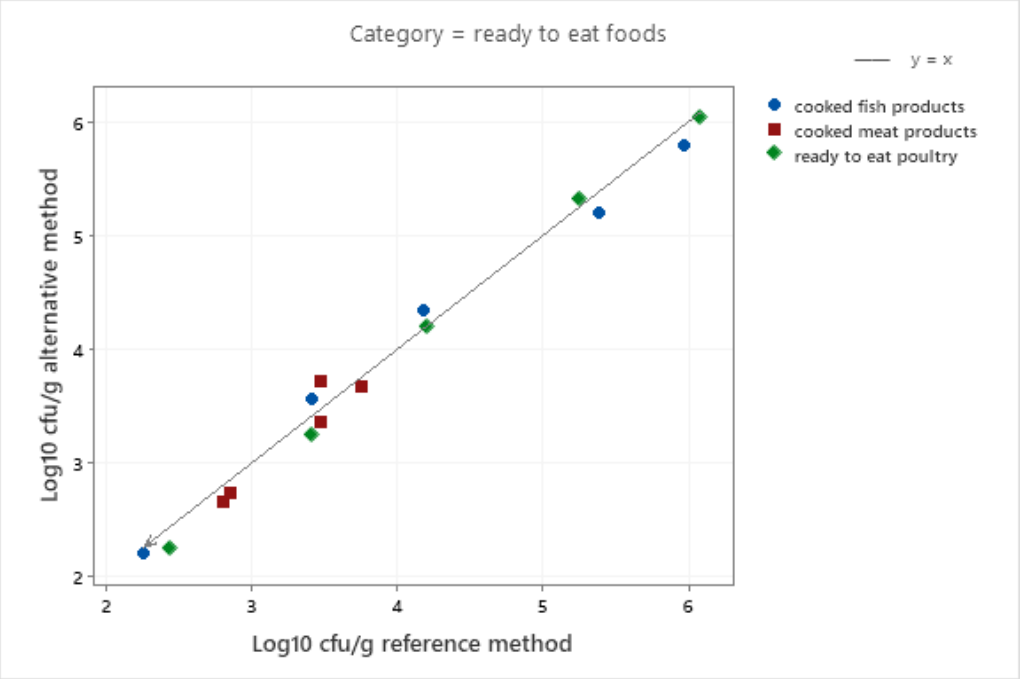


Figure 5 - Scatter plot of the reference method versus alternative method results for coliforms in Multicomponent foods

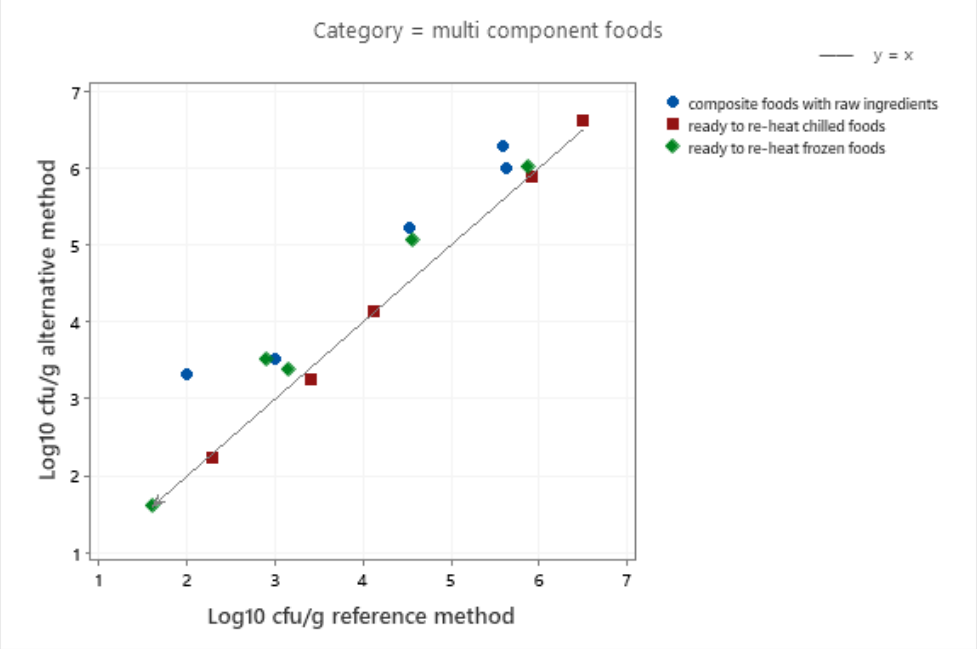


Figure 6 - Scatter plot of the reference method versus alternative method results for coliforms in Environmental samples (food or feed production)

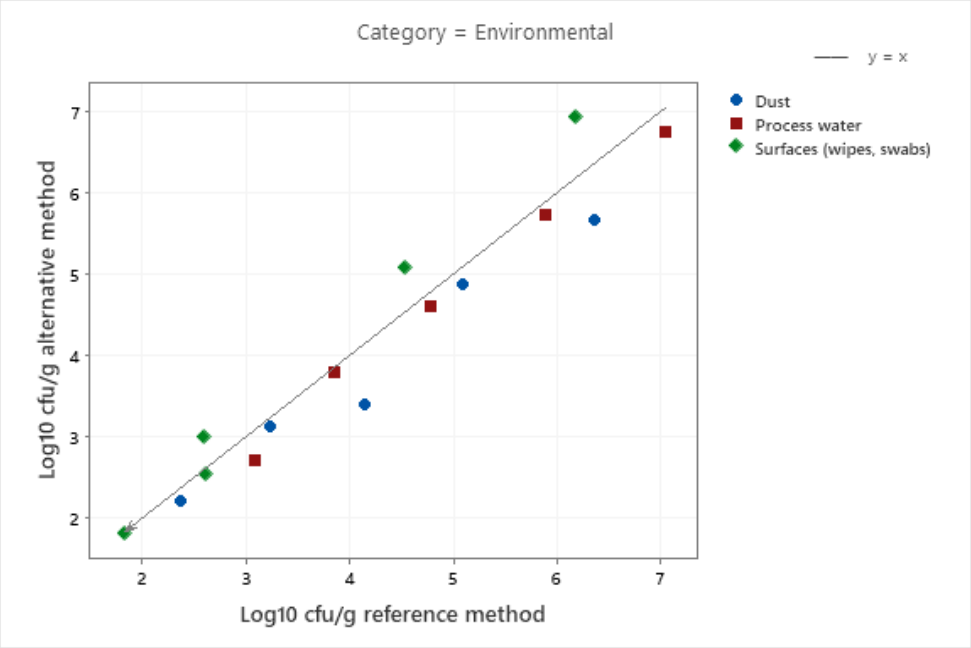


Figure 7 Scatter plot of the reference method versus alternative method results for the coliforms in Pet food and animal feed.

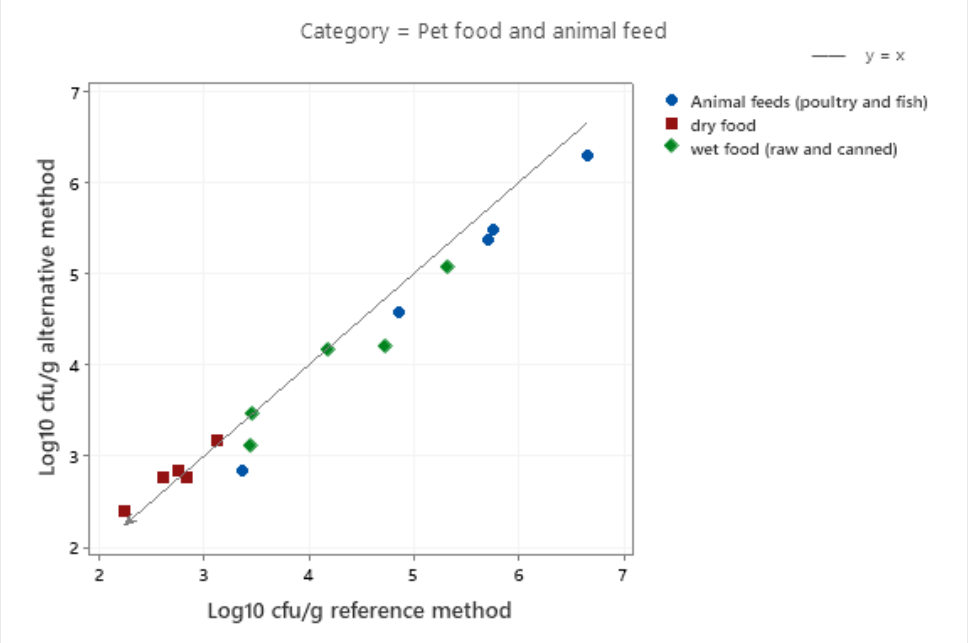
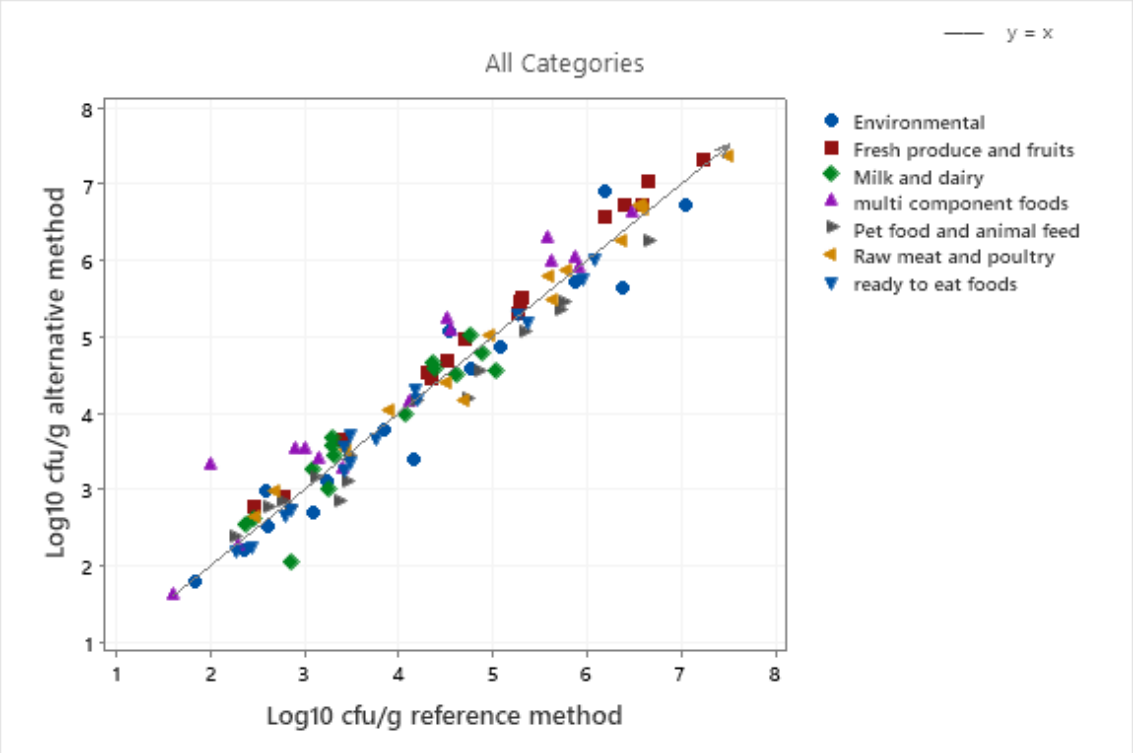


Figure 8 - Scatter plot of the reference method versus alternative method results for coiforms in all the categories



The data in the scatter plots show good agreement between the reference and alterntive methods with no obvious disagreement/ some signs of bias. A slight positive bias of 0.048 was noted in the 2 categories for this part of the study. A summary of the calculated values per category is provided in Table 4.

Table 4 - Summary of the calculated values per category

Category.	n	Dbar	sD	95% Lower limit	95% Upper limit
Environmental	15	-0.092	0.409	-0.999	0.815
Fresh produce and fruits	15	0.229	0.106	-0.005	0.463
Milk and dairy	15	0.023	0.327	-0.703	0.748
multi component foods	15	0.348	0.394	-0.525	1.221
Pet food and animal feed	15	-0.162	0.233	-0.678	0.354
Raw meat and poultry	15	0.032	0.201	-0.414	0.477

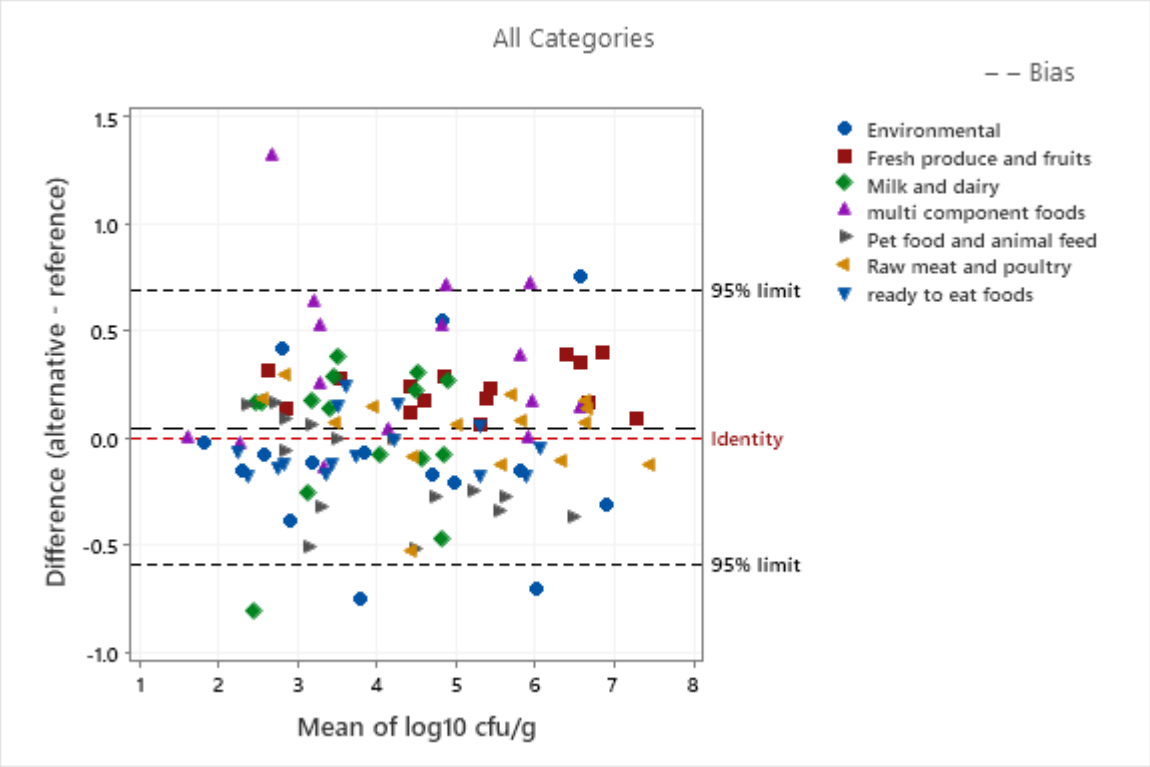
Category.	n	Dbar	sD	95% Lower limit	95% Upper limit
ready to eat foods	15	-0.039	0.138	-0.344	0.266
All Categories	105	0.048	0.321	-0.591	0.687

Key

$\overline{D}$: Average difference SD: standard deviation of differences n: number of samples

The Bland-Altman difference plot for all the samples is given Figure 9.

Figure 9 – Bland-Altman difference plot for all the samples



Samples for which the difference between the result observed with the reference and the alternative methods is above or lower than the limits are listed in the Table 5.

Table 5 - Data which are outside of the accepted limits

Category	**item**	Strain	**Code**	**Reference method Log cfu/g**	**Alternative method Log cfu/g**	**Mean Log cfu/g**	**Difference (Alternative-reference)**	**Lower/ upper limits**
dairy products	strawberry yogurt	*Citrobacter braakii* 16279	6	2.04	2.85	2.44	- 0.80	-0.591
dust	Oven top	*Escherichia hermanii* 7477	213	4.15	3.40	3.77	-0.75	-0.591
dust	Bakery sink corner		215	6.36	5.66	6.01	-0.70	-0.591
composite foods with raw ingredients	cheese and onion sandwich	*Klebsiella ozaenae* 4273	72	2.00	3.32	2.66	1.32	0.687
	bacon,lettuce, tomato sandwich		73	4.52	5.23	4.87	0.71	0.687
	minted bean salad		75	5.58	6.30	5.94	0.72	0.687
Surfaces (wipes, swabs)	MDF - lab bench	*Escherichia vulneris 16260*	95	6.18	6.93	6.55	0.75	0.687

It is expected that not more than one in 20 data values will lie outside the CLs. In this study there were 7 data points from a total of 105 data points which were outside of the accepted limits. This is higher than the expectation however, there are no trends to the outlying data which represented four of the seven categories. Four points were slightly higher than the upper limit of 0.687 and one point was slightly lower than the lower limit of -0.591. One point (sample 72) had a difference of 1.32 between the reference and alternative method.

The samples covered a diverse range of sample items with different isolates used to inoculate the samples used indicating that the bias seen was not strian dependant. Three out of the four samples with a positve discrepancy were inoculated with *Klebiella ozanae* in multi-component foods containing raw ingredients which were seeded and stored chilled. This strain gave a similar response on both the alternate and reference methods in the inclusivity studies (Annex H) but it appears that after chill storage in foods with raw ingredients. The alternate methods give a higher recovery than the reference method.

These results show good agreement between the two methods for enumeration of total coliforms with a slight positive bias for the alternate method with an overall bias from all the categories of 0.048.

Discordant results

It is commonly recognized that a bias higher than 0.5 Log CFU/g difference between the compared methods should be explained if possible. It is the case for 14 samples, 8 with a positive bias and 6 with negative bias.

There are more points with a positive bias than negative bias and samples inoculated with *Klebsiella* gave the majority of the discordant results. Six samples showing a positive bias were *Klebsiell*a strains which had been chilled or frozen prior to analysis. Other coliform strains which were chilled or frozen did not show the same behaviour. The remaning 2 environmental samples showing positve bias were inoculated with *Escherichia vulneris* and stored at ambient for 2 weeks. Other samples in the same type incoulated with the same strain did not give discordant results.

The results showing a HIGHER enumeration with the ALTERNATIVE method than with the REFERENCE method are shown below. (See Table 4).

Table 4 – Discordant results with a positive bias

Sample no.	**Product Category**	**Products**	**Bias log Alt - log Ref (log CFU/g)**	**Strain**	**Stress applied**
68	multi component foods	frozen chips	0.63	Klebsiella trevisanii NCIMB 8606	frozen 2 weeks
69	multi component foods	frozen cheese and tomato pizza	0.52	*Klebsiella trevisanii* NCIMB 8606	frozen 2 weeks
71	multi component foods	chicken salad mayo sandwich	0.52	*Klebsiella ozaenae* 4273	chill 2-3 days
72	multi component foods	cheese and onion sandwich	1.32	*Klebsiella ozaenae* 4273	chill 2-3 days
73B	multi component foods	bacon,lettuce , tomato sandwich	0.71	*Klebsiella ozaenae* 4273	chill 2-3 days
75B	multi component foods	minted bean salad	0.72	*Klebsiella ozaenae* 4273	chill 2-3 days

Sample no.	Product Category	Products	Bias log Alt - log Ref (log CFU/g)	Strain	Stress applied
204	Surfaces (wipes, swabs)	Plastic - equipment surface	0.55	*Escherichia vulneris 16260*	storage 2 weeks at ambient
205		MDF - lab bench	0.75		

The results showing a LOWER enumeration with the ALTERNATE method than with the REFERENCE method are shown below (See Table 5).

There were no trends observed with the samples showing negative bias in this study. All six samples were from different sample types and were artifcally contaminated with a different isolates and inoculation protocols.

Table 5 – Discordant results with a negative bias

Sample no.	Product Category	Products	Bias log Alt - log Ref (log CFU/g)	Strain	Stress applied
6B	Milk and dairy	Strawberry yogurt	-0.80	*Citrobacter braakii* 16279	Chill 2-3 days
43	Raw meat and poultry	southern fried chicken goujons	-0.52	*Escherchia fergusonii* CRA 7522	frozen 2 weeks
213	dust	Oven top	-0.75	*Escherichia hermanii* 7477	Lyophilisd cells stored 2 weeks at ambient
215		Bakery sink corner	-0.70		

Sample no.	Product Category	Products	Bias log Alt - log Ref (log CFU/g)	Strain	Stress applied
C14	Animal feeds (poultry and fish)	Bug bites insect lavae and salmon	-0.51	*Klebsiella oxytoca 15926*	lypholised culture - storage for 2 weeks at ambient
3B	wet food (raw and canned)	gourmet cat food - mouse with salmon and cascading gravy	-0.52	*Enterobacter aerogenes 4108*	heat stress 55°C for 10 minutes

3.1.4 Conclusion (RT study coliforms)

The relative trueness of the Alternative method is satisfied as the expectation of not more than 1 in 20 data points outside of the acceptability limits is met.

3.2 Accuracy profile study

The accuracy profile study is a comparative study between the results obtained by the reference and the results of the alternative method. This study is conducted using artificially contaminated samples, using one type per category.

3.2.1 Categories, sample types and strains

Two categories were tested with a single batch of two different sample types using 6 samples per type.

Two samples were contaminated at a low level, 2 at intermediate level, 2 at a high level. For each sample, 5 replicates (5 different test portions) were tested. A total of 30 samples were analysed per food type. The following food type/strain pairs were studied (See Table 6a and b)

Each sample was bulk inoculated and five replicate test portions examined from the bulk sample/ individually inoculated as a separate test portion, with the exception of salad where single test portions were inoculated. The tested categories, types and items are provided in Table 6a and b.

Preparation of samples was done as a bulk inoculation. A 100g sample was inoculated with 1ml of appropriate dilution of inoculating strain and homogenised by hand massaging or stomaching to evenly distribute the inoculum. For all matrices, the 100g samples were inoculated and stored at 2-8°C for 48-72h prior to analysis. The wet pet food samples were inoculated with heat stressed cultures with a minimum of 0.5log injury.

Five separate 10g test portions were removed from the bulk sample and mixed with 90ml PSD or appropriate diluent and enumerated on both methods as shown in Appendix A

Table 6. Categories, types, items, strains and inoculation levels for accuracy profile study (coliforms)

Category	Types	Inoculated Strain coliforms	Item	Inoculation levels	Test portions
Dairy products	Pasteurised dairy products	*E. coli* CRA 1476 from dried milk	Pasteurised cream	Low 10^2 cfu/g	5
				Medium : 10^4 cfu/g	5
				High : 10^6 cfu/g	5
		Enterobacter agglomerans CRA 5613 from milk powder	Cream cheese	Low 10^2 cfu/g	5
				Medium : 10^4 cfu/g	5
				High : 10^6 cfu/g	5
Fruits and vegetables	Fresh produce	*E.hermanii* CRA 7477 from sesame seeds	Ready to cook Vegetable preparation	Low 10^2 cfu/g	5
				Medium : 10^4 cfu/g	5
				High : 10^6 cfu/g	5
		Citrobacter amalonaticus CRA 7458 from beansprouts	Vegetable juice	Low 10^2 cfu/g	5
				Medium : 10^4 cfu/g	5
				High : 10^6 cfu/g	5
Raw poultry and meats (Combined category raw/ RTC meats and poultry)	Fresh meat	*Enterobacter aerogenes* NCTC 10006	Pork mince	Low 10^2 cfu/g	5
				Medium : 10^4 cfu/g	5
				High : 10^6 cfu/g	5
		Citrobacter freundii NCTC 9750	Raw bacon	Low 10^2 cfu/g	5
				Medium : 10^4 cfu/g	5
				High : 10^6 cfu/g	5
Ready to eat foods (Combined category RTE/RTRH meats and poultry)	Cooked fish products e.g. prawns	*E.coli* CRA 2003 from fish	Fresh prawns	Low 10^2 cfu/g	5
				Medium : 10^4 cfu/g	5
				High : 10^6 cfu/g	5
		Klebsiella oxytoca ATCC 15926	Fish pate	Low 10^2 cfu/g	5
				Medium : 10^4 cfu/g	5
				High : 10^6 cfu/g	5
Multi component foods	Composite foods with raw ingredients	*Enterobacter agglomerans* CRA 5513 from skimmed milk powder	Sandwiches	Low 10^2 cfu/g	5
				Medium : 10^4 cfu/g	5
				High : 10^6 cfu/g	5
		E. adecarboxylata CRA 5501 from skimmed milk powder	Cooked chilled rice	Low 10^2 cfu/g	5
				Medium : 10^4 cfu/g	5
				High : 10^6 cfu/g	5
			Wash water	Low 10^2 cfu/g	5
				Medium : 10^4 cfu/g	5
				High : 10^6 cfu/g	5
				Low 10^2 cfu/g	5

Category	Types	Inoculated Strain coliforms	Item	Inoculation levels	Test portions
Environmental samples	Process water	*Enterobacter intermedius* NCTC 12125 isolated from surface water	Cooling water	Medium : 10^4 cfu/g	5
				High : 10^6 cfu/g	5
Pet food and animal feed	Wet food (cooked)	*Citrobacter freundii* CRA 3163 isolated from sausage	dog food - pate	Low 10^2 cfu/g	5
				Medium : 10^4 cfu/g	5
				High : 10^6 cfu/g	5
			cat food with gravy	Low 10^2 cfu/g	5
				Medium : 10^4 cfu/g	5
				High : 10^6 cfu/g	5

Preparation of samples was done as a bulk inoculation. A 100g sample was inoculated with 1ml of appropriate dilution of inoculating strain and homogenised by hand massaging or stomaching to evenly distribute the inoculum. For all matrices, the 100g samples were inoculated and stored at 2-8°C for 48-72h prior to analysis. The wet pet food samples were inoculated with heat stressed cultures with a minimum of 0.5log injury.

Five separate 10g test portions were removed from the bulk sample and mixed with 90ml PSD or appropriate diluent and enumerated on both methods as shown in Appendix A

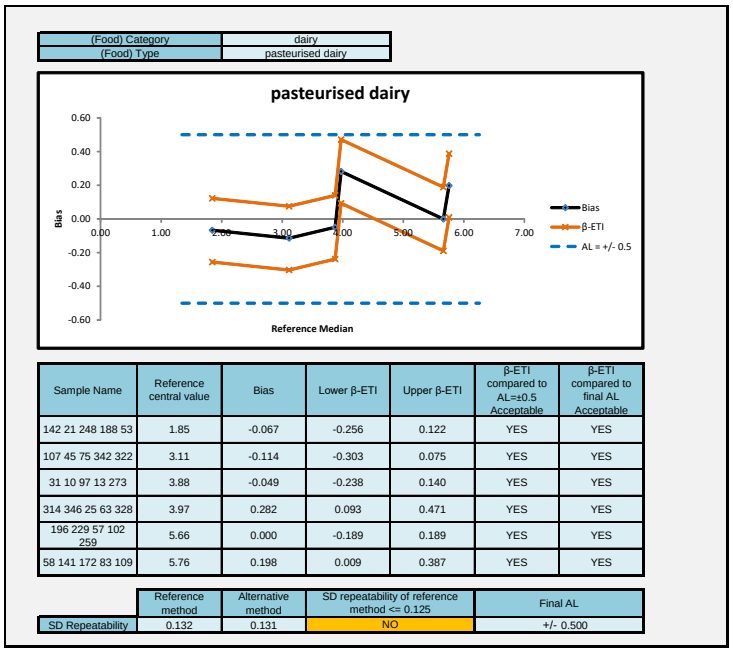
All results were tabulated, calculated and interpreted according to ISO 16140-2
Inclusivity and exclusivity study

3.2.2 Calculations and interpretation of accuracy profile study

All results are tabulated, calculated and interpreted according to ISO 16140-2. The statistical results are shown in Figures 9 to 15.

The calculations were done using the AP Calculation Tool MCS (Clause 6-1-3-3 calculation and interpretation of Accuracy profile study) available on http://standards.iso.org/iso/16140

Figure 9 – Accuracy profile for coliforms in Dairy products

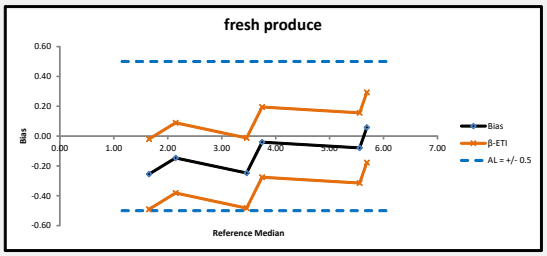


(Food) Category	dairy
(Food) Type	pasteurised dairy

Sample Name	Reference central value	Bias	Lower β-ETI	Upper β-ETI	β-ETI compared to AL=±0.5 Acceptable	β-ETI compared to final AL Acceptable
142 21 248 188 53	1.85	-0.067	-0.256	0.122	YES	YES
107 45 75 342 322	3.11	-0.114	-0.303	0.075	YES	YES
31 10 97 13 273	3.88	-0.049	-0.238	0.140	YES	YES
314 346 25 63 328	3.97	0.282	0.093	0.471	YES	YES
196 229 57 102 259	5.66	0.000	-0.189	0.189	YES	YES
58 141 172 83 109	5.76	0.198	0.009	0.387	YES	YES

	Reference method	Alternative method	SD repeatability of reference method <= 0.125	Final AL
SD Repeatability	0.132	0.131	NO	+/- 0.500

Figure 10 – Accuracy profile for coliforms in Fruit and vegetable products



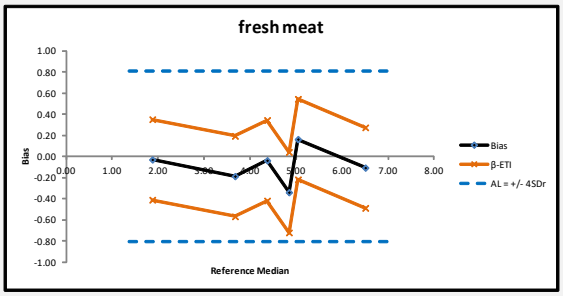
(Food) Category	fruit and veg
(Food) Type	fresh produce

Sample Name	Reference Central value	Bias	Lower β-ETI	Upper β-ETI	β-ETI compared to AL=±0.5 Acceptable	β-ETI compared to final AL Acceptable
8 17 327 246 37	1.65	-0.255	-0.491	-0.020	YES	YES
264 252 139 300 294	2.15	-0.146	-0.381	0.089	YES	YES
312 301 64 267 340	3.46	-0.248	-0.483	-0.012	YES	YES
236 92 223 9 158	3.75	-0.041	-0.276	0.195	YES	YES
316 86 323 117 203	5.56	-0.079	-0.314	0.156	YES	YES
74 234 272 34 144	5.69	0.058	-0.177	0.293	YES	YES

	Reference method	Alternative method	SD repeatability of reference method <= 0.125	Final AL
SD Repeatability	0.086	0.163	YES	+/- 0.500

Figure 11 – Accuracy profile for coliforms in Meat and poultry products

(Food) Category	raw poultry and meat
(Food) Type	fresh meat

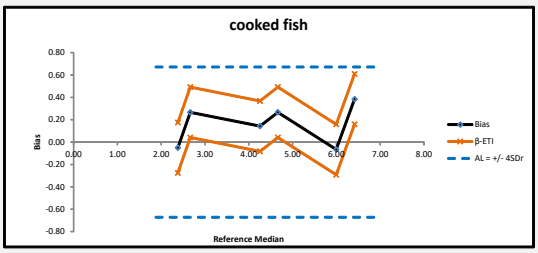


Sample Name	Reference Central value	Bias	Lower β-ETI	Upper β-ETI	β-ETI compared to AL=±0.5 Acceptable	β-ETI compared to final AL Acceptable
168 135 283 214 47	1.88	-0.030	-0.411	0.351	YES	YES
204 253 207 76 156	3.67	-0.188	-0.569	0.193	NO	YES
284 225 85 257 96	4.37	-0.038	-0.419	0.343	YES	YES
348 171 65 281 151	4.85	-0.340	-0.721	0.041	NO	YES
167 36 324 216 133	5.04	0.163	-0.218	0.544	NO	YES
165 69 310 118 349	6.51	-0.107	-0.488	0.274	YES	YES

	Reference method	Alternative method	SD repeatability of reference method <= 0.125	Final AL
SD Repeatability	0.202	0.264	NO	+/- 0.808

Figure 12 – Accuracy profile for coliforms in Ready to eat foods

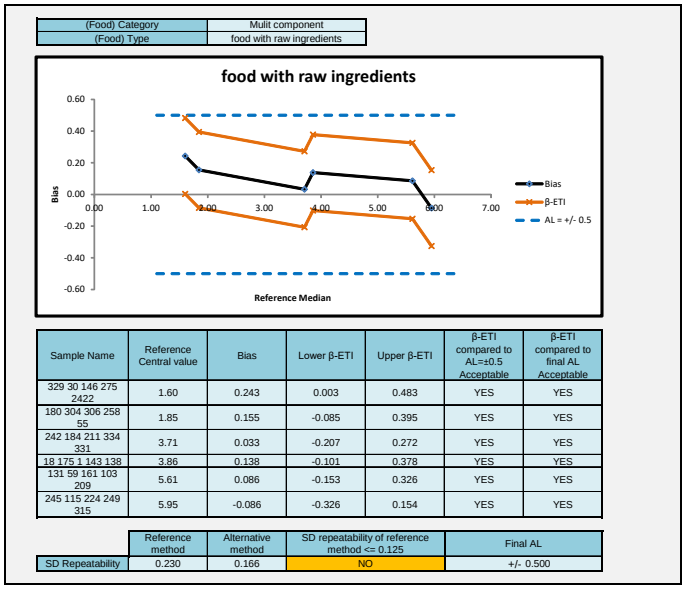
(Food) Category	RTE foods
(Food) Type	cooked fish



Sample Name	Reference Central value	Bias	Lower β-ETI	Upper β-ETI	β-ETI compared to AL=±0.5 Acceptable	β-ETI compared to final AL Acceptable
51 247 218 87 287	2.38	-0.050	-0.275	0.175	YES	YES
111 155 255 186 202	2.66	0.267	0.041	0.492	YES	YES
289 68 23 309 226	4.26	0.143	-0.083	0.368	YES	YES
256 192 295 16 298	4.66	0.267	0.041	0.492	YES	YES
195 261 42 61 320	6.00	-0.066	-0.291	0.159	YES	YES
319 189 78 82 182	6.41	0.384	0.159	0.610	NO	YES

	Reference method	Alternative method	SD repeatability of reference method <= 0.125	Final AL
SD Repeatability	0.168	0.156	NO	+/- 0.672

Figure 13 – Accuracy profile for coliforms in multicomponent foods

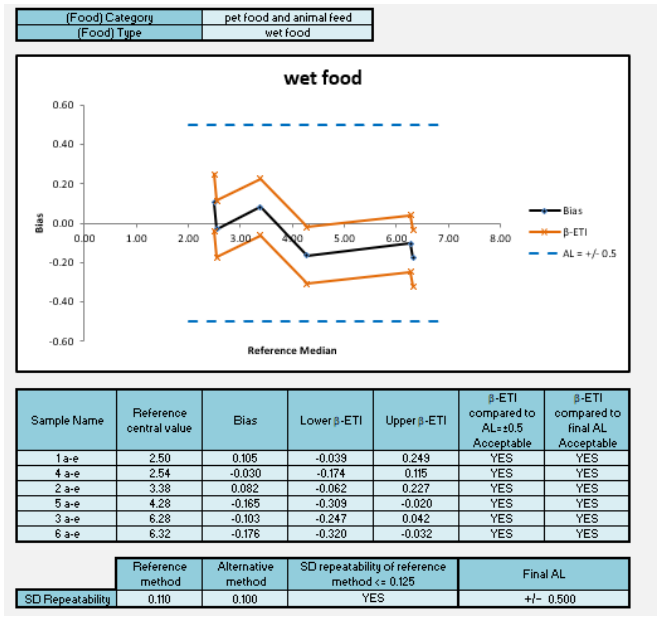


(Food) Category	Mulit component
(Food) Type	food with raw ingredients

Sample Name	Reference Central value	Bias	Lower β-ETI	Upper β-ETI	β-ETI compared to AL=±0.5 Acceptable	β-ETI compared to final AL Acceptable
329 30 146 275 2422	1.60	0.243	0.003	0.483	YES	YES
180 304 306 258 55	1.85	0.155	-0.085	0.395	YES	YES
242 184 211 334 331	3.71	0.033	-0.207	0.272	YES	YES
18 175 1 143 138	3.86	0.138	-0.101	0.378	YES	YES
131 59 161 103 209	5.61	0.086	-0.153	0.326	YES	YES
245 115 224 249 315	5.95	-0.086	-0.326	0.154	YES	YES

	Reference method	Alternative method	SD repeatability of reference method <= 0.125	Final AL
SD Repeatability	0.230	0.166	NO	+/- 0.500

Figure 14 – Accuracy profile for coliforms in Pet food and animal feed.

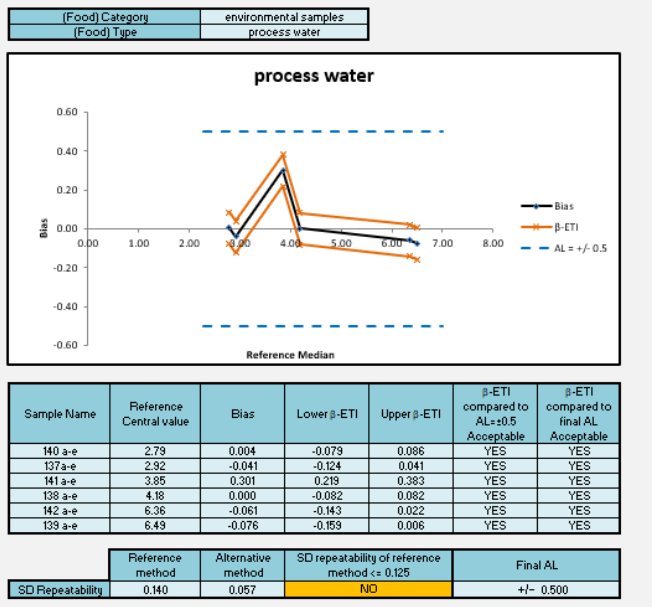


(Food) Category	pet food and animal feed
(Food) Type	wet food

Sample Name	Reference central value	Bias	Lower β-ETI	Upper β-ETI	β-ETI compared to AL=±0.5 Acceptable	β-ETI compared to final AL Acceptable
1 a-e	2.50	0.105	-0.039	0.249	YES	YES
4 a-e	2.54	-0.030	-0.174	0.115	YES	YES
2 a-e	3.38	0.082	-0.062	0.227	YES	YES
5 a-e	4.28	-0.165	-0.309	-0.020	YES	YES
3 a-e	6.28	-0.103	-0.247	0.042	YES	YES
6 a-e	6.32	-0.176	-0.320	-0.032	YES	YES

	Reference method	Alternative method	SD repeatability of reference method <= 0.125	Final AL
SD Repeatability	0.110	0.100	YES	+/- 0.500

Figure 15 – Accuracy profile ***for coliforms in*** *Environmental samples (food or feed production)*



(Food) Category	environmental samples
(Food) Type	process water

Sample Name	Reference Central value	Bias	Lower β-ETI	Upper β-ETI	β-ETI compared to AL=±0.5 Acceptable	β-ETI compared to final AL Acceptable
140 a-e	2.79	0.004	-0.079	0.086	YES	YES
137a-e	2.92	-0.041	-0.124	0.041	YES	YES
141 a-e	3.85	0.301	0.219	0.383	YES	YES
138 a-e	4.18	0.000	-0.082	0.082	YES	YES
142 a-e	6.36	-0.061	-0.143	0.022	YES	YES
139 a-e	6.49	-0.076	-0.159	0.006	YES	YES

	Reference method	Alternative method	SD repeatability of reference method <= 0.125	Final AL
SD Repeatability	0.140	0.057	NO	+/- 0.500

In this study the following 5 categories (Dairy products, fresh produce, multicomponent foods, Environmental samples and Pet food and animal feed) met the AL of 0.5log.

Two food categories (meat and RTE products) required additional AL calculation. For the meat category the lower level for ground beef and the medium level for pork mince showed a negative bias and the high level for pork mince showed positive bias. For RTE foods, the high level for fish pate showed a positive bias. Newly calculated AL's were 0.808 for the meat category and 0.672 for the RTE category.

Analysis of the data revealed that the accuracy of the Alternative method is satisfied as all the categories met the 0.5log AL or the re-calculated AL.

3.3 Inclusivity / exclusivity

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.

3.3.1 1 Protocol

After being grown according to appropriate conditions, decimal dilutions were made and the 20 target strains and 10 non-target strains were enumerated by the alternative method, the reference method and a non selective agar (PCA/MRSA).

3.3.2 2.4.2 Results

2.4.2.1 Original study

Thirty three inclusivity cultures and 20 exclusivity cultures were tested in the original study in 2007. Both methods were challenged with 2-3 $\log_{10}$ (100 times limit of detection) cfu/ml of each culture twice as required by EN ISO 16140. The inclusivity results (Appendix 8) revealed all 33 coliform strains produced typical colonies in VRBA (ISO 4832) and Compact Dry EC medium. The results from the 20 strains of non-target organisms used to determine the exclusivity of the EC method showed that 9 strains did not grow on either the EC medium or on VRBA. In addition, one strain of *Yersinia enterocolitica* did not grow on the Compact Dry EC medium but did grow in VRBA. For Compact Dry EC, there were 7 strains giving atypical growth and 3 giving typical growth. For VRBA there were 5 strains giving atypical growth and 6 giving typical growth (Appendix 8).

2.4.2.2 Additional Study

Of the 20 inclusivity strains tested 18 strains were detected using the alternative and reference methods. Those not detected by either method were *Shimwellia blattae* NCTC 12127 and *Klebsiella rhinoscleromatis* CRA 4272. Of the 10 exclusivity strains tested, three were detected by the alternate method and by the reference method these were *A.hydrophila* CRA 4111, *A.sobria* CRA 8390 and *S. fonticola* CRA 4613 (Appendix 8)

3.4 Limit of quantification (LOQ)

The LOQ applies only to instrumental methods. It does not apply to methods based on counting visible colonies. It may also not apply to instrumental methods where it is not possible to get blank samples e.g. instrumental methods for total plate counts.

The alternate method is based on visible colonies therefore the LOQ does not have to be calculated for the

3.4.1 Conclusion (MCS)

Overall, the conclusions for the Method Comparison are:

- The alternative method Compact Dry EC for enumeration of *E. coli* and coliforms shows satisfactory results for relative trueness;
- The alternative Compact Dry EC for enumeration of *E. coli* and coliforms shows satisfactory results for accuracy profile;

4 Interlaboratory study

The experimental design for the interlaboratory study is the same in ISO16140:2003 and ISO16140-2:2015. However, the statistical analysis of the data is different. It was proposed to use the existing ILS data to recalculate the new statistics using this data as shown below.

4.1 Organisation

There were 10 collaborative laboratories used in this study representing 5 different countries.

Pasteurised milk was used as the food matrix for the interlaboratory study and the samples of milk were artificially contaminated with a single strain of *E. coli* (CCFRA code 11017, NCTC 12241) and a single strain of *Enterobacter aerogenes* (CCFRA 15736, NCTC 10006). Each strain was cultured in 10 ml NB incubated overnight at 37 ± 1°C. Both cultures were serially diluted in MRD to give the desired levels of inoculum for the contamination of the samples and were mixed together in equal concentrations.

A set of 8 x 25 ml samples of pasteurised milk were prepared for each laboratory, including the organising laboratory. Two samples remained uninoculated, whereas the other six samples were inoculated at 3 different contamination levels (low, medium and high). Appropriate dilutions of the mixed culture cocktail were used to individually inoculate 2 x 25ml samples at the low (10^1 – 10^2 CFU/ml), medium (10^2 – 10^3 CFU/ml) and high (10^3 – 10^4 CFU/ml) contamination levels. The samples were blind coded and stored at 2 – 8°C prior to despatch to the collaborative laboratories. Before despatch, each set of eight samples was packed into a suitable container with cool packs. Also, an additional vial containing water was packed with each set of samples. This enabled the laboratories to take a temperature measurement upon receipt.

Upon receipt, each collaborative laboratory tested a 10 ml test portion from each of the eight milk samples by the ISO 4832:2006 method and the Compact Dry EC method. In addition, the organising laboratory tested a set of eight milk samples at the same time as the collaborative laboratories to confirm the presence of the target organism and the contamination levels. This data was not used in the analyses.

The study was done in November 2007

Table 7: Sample receipt data for ILS samples

Temperature of control sample upon receipt Laboratory	Date received	Temperature of control sample upon receipt (°C)
1	05/11/07	3.1
2	05/11/07	6
3	05/11/07	7
4	05/11/07	2.65
5	05/11/07	5.2
6	05/11/07	6.5
7	05/11/07	2.9
8	05/11/07	7
9	05/11/07	2.1
10	05/11/07	2.7

4.2 Calculations and interpretation of data

The data from the collaborative trial were calculated and interpreted according to section 6.2.3 of ISO 16140-2:2016 using the freely available Excel® spreadsheet (http://standards.iso.org/iso/16140).

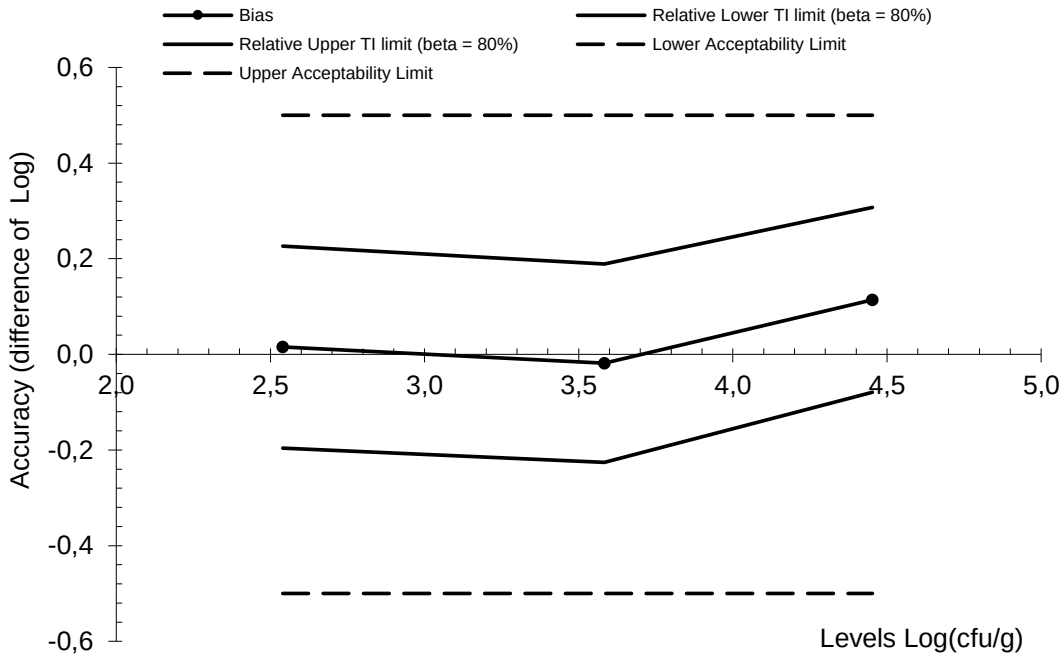
The log transformed data from the existing trial is shown in Table 8 below and the Accuracy profile graph is shown in Figure 4.

Table 8: Summary of the results of the interlaboratory study per analyte level (k)

		Reference method x_{ijk}		Alternative method k_{ijk}	
Collaborators (i)	**Level (k)**				
1	Blank	<10	<10	<10	<10
2	Blank	<10	<10	<10	<10
3	Blank	<10	<10	<10	<10
4	Blank	<10	<10	<10	<10
5	Blank	<10	<10	<10	<10
6	Blank	<10	<10	<10	<10
7	Blank	<10	<10	<10	<10
8	Blank	<10	<10	<10	<10
9	Blank	<10	<10	<10	<10
10	Blank	<10	<10	<10	<10
		Duplicate 1	Duplicate 2	Duplicate 1	Duplicate 2
1	Low	2.70	2.63	2.70	2.57
2	Low	2.74	2.63	2.54	2.53
3	Low	2.51	2.57	2.47	2.59
4	Low	2.46	2.53	2.30	2.54
5	Low	2.46	2.26	2.48	2.23
6	Low	2.50	2.44	2.70	2.63
7	Low	2.48	2.57	2.57	2.56
8	Low	2.65	2.61	2.65	2.61
9	Low	2.48	2.56	2.53	2.57
10	Low	2.49	2.86	2.59	3.02

		Reference method x $_{ijk}$		**Alternative method k $_{ijk}$**	
Collaborators (i)	**Level (k)**				
		Duplicate 1	Duplicate 2	Duplicate 1	Duplicate 2
1	Medium	3.65	3.58	3.62	3.76
2	Medium	3.67	3.71	3.75	3.71
3	Medium	3.69	3.55	3.60	3.55
4	Medium	3.47	3.55	3.41	3.41
5	Medium	3.62	3.53	3.53	3.47
6	Medium	3.49	3.68	3.64	3.82
7	Medium	3.81	3.66	3.75	3.60
8	Medium	3.78	3.60	3.63	3.62
9	Medium	3.57	3.58	3.49	3.47
10	Medium	3.60	3.50	3.61	3.39
		Duplicate 1	Duplicate 2	Duplicate 1	Duplicate 2
1	High	4.32	4.25	4.74	4.67
2	High	4.61	4.79	4.73	4.77
3	High	4.47	4.46	4.58	4.59
4	High	4.61	4.54	4.53	4.52
5	High	4.49	3.98	4.53	4.25
6	High	4.50	4.44	4.71	4.68
7	High	4.68	4.73	4.53	4.66
8	High	4.72	4.68	4.57	4.68
9	High	4.36	4.02	4.57	4.50
10	High	4.34	4.50	4.51	4.44

Figure 4: Accuracy profile of the alternative method (CD EC) in the Inter laboratory study



The statistical analysis of the existing ILS data is shown in Table 9 below. It can be seen that the repeatability standard deviation (S_r) was very similar for the alternate method and the reference method ranging from 0.080 to 0.132 for the compact dry EC and 0.084 to 0.150 for the reference method.

The between-labs standard deviation (S_L) was slightly better for the alternative method (0.086 to 0.101) and the reference method (0.074 to 0.164) as was the reproducibility standard deviation (S_R) with alternative method values of 0.125 to 0.157 and reference method values of (0.92 to 0.222).

According to the ISO 16140-2:2016 standard, if any of the values of the β-ETI fall outside of the ±0.5log AL then a further calculation is done to calculate the pooled average S_R of the reference method. This was not required as all values were within the required limits. The data are plotted in Figure 4 and it can be seen that no values lie outside of these AL_s values and therefore the alternative method is accepted as being equivalent to the reference method.

Table 9. Statistical analysis of the ILS data according to the ISO spreadsheet

Accuracy profile			
Study Name	Compact Dry EC coliuforms		
Date	Study done 11/2007 nd re-calculated 06/2017		
Coordinator	Campden BRI		
Tolerance probability (beta)	80%	80%	80%
Acceptability limit in log (lambda)	0.50	0.50	0.50

FALSE

Application of clause 6.2.3
Step 8: If any of the values for the β-ETI fall outside the acceptability limits, calculate the pooled average reproducibility standard deviation of the reference method.
Step 9: Calculate new acceptability limits as a function of this standard deviation.

	Alternative method			Reference method		
Levels	**Low**	**Medium**	**High**	**Low**	**Medium**	**High**
Target value	2.556	3.614	4.474			
Number of participants (K)	10	10	10	10	10	10
Average for alternative method	2.567	3.589	4.587	2.556	3.614	4.474
Repeatability standard deviation (sr)	0.132	0.080	0.080	0.106	0.084	0.150
Between-labs standard deviation (sL)	0.086	0.101	0.096	0.074	0.038	0.164
Reproducibility standard deviation (sR)	0.157	0.129	0.125	0.130	0.092	0.222
Corrected number of dof	16.928	13.124	13.429	16.593	18.104	14.013
Coverage factor	1.376	1.403	1.401			
Interpolated Student t	1.334	1.350	1.348			
Tolerance interval standard deviation	0.1622	0.1336	0.1294			
Lower TI limit	2.351	3.409	4.413			
Upper TI limit	2.783	3.770	4.762			
Bias	**0.011**	**-0.025**	**0.113**			
Relative Lower TI limit (beta = 80%)	**-0.205**	**-0.205**	**-0.061**			
Relative Upper TI limit (beta = 80%)	**0.228**	**0.156**	**0.288**			
Lower Acceptability Limit	**-0.50**	**-0.50**	**-0.50**			
Upper Acceptability Limit	**0.50**	**0.50**	**0.50**			
New acceptability limits may be based on reference method pooled variance						
Pooled repro standard dev of reference	0.158					

FALSE
FALSE

Select ALL blue lines to draw the accuracy profile as illustrated in the worksheet "Graph Profile"

5 Overall conclusions of the validation study

- The alternative method Compact Dry EC for enumeration of coliforms shows satisfactory results for relative trueness;
- The alternative Compact Dry EC for enumeration of coliforms shows satisfactory results for accuracy profile
- The alternative Compact Dry EC for enumeration of coliforms is selective and specific
- The alternative Compact Dry EC for enumeration of coliforms shows satisfactory results for the ILS
- The alternative method Compact Dry EC for the enumeration of coliforms shows comparable performance to the reference method ISO 4832:2006 Microbiology of food and animal feeding stuffs: Horizontal method for the enumeration of coliforms - Colony Count Method.

Date, 26/10/2023

Signature Suzanne Jordan

6 References

Shimadzu Diagnostics Corporation CompactDry EC Kit insert version March 2019

ISO 16649-2:2001 Microbiology of food and animal feeding stuffs — Horizontal method for the enumeration of beta-glucuronidase-positive Escherichia coli — Part 2: Colony-count technique at 44 degrees C using 5-bromo-4-chloro-3-indolyl beta-D-glucuronide

ISO 4832:2006 Microbiology of food and animal feeding stuffs: Horizontal method for the enumeration of coliforms - Colony Count Method for coliforms.

ISO 4833-1:2013; Microbiology of the food chain -- Horizontal method for the enumeration of microorganisms -- Part 1: Colony count at 30 degrees C by the pour plate technique

ISO 6887; Microbiology of the food chain -- Preparation of test samples, initial suspension and decimal dilutions for microbiological examination – All parts.

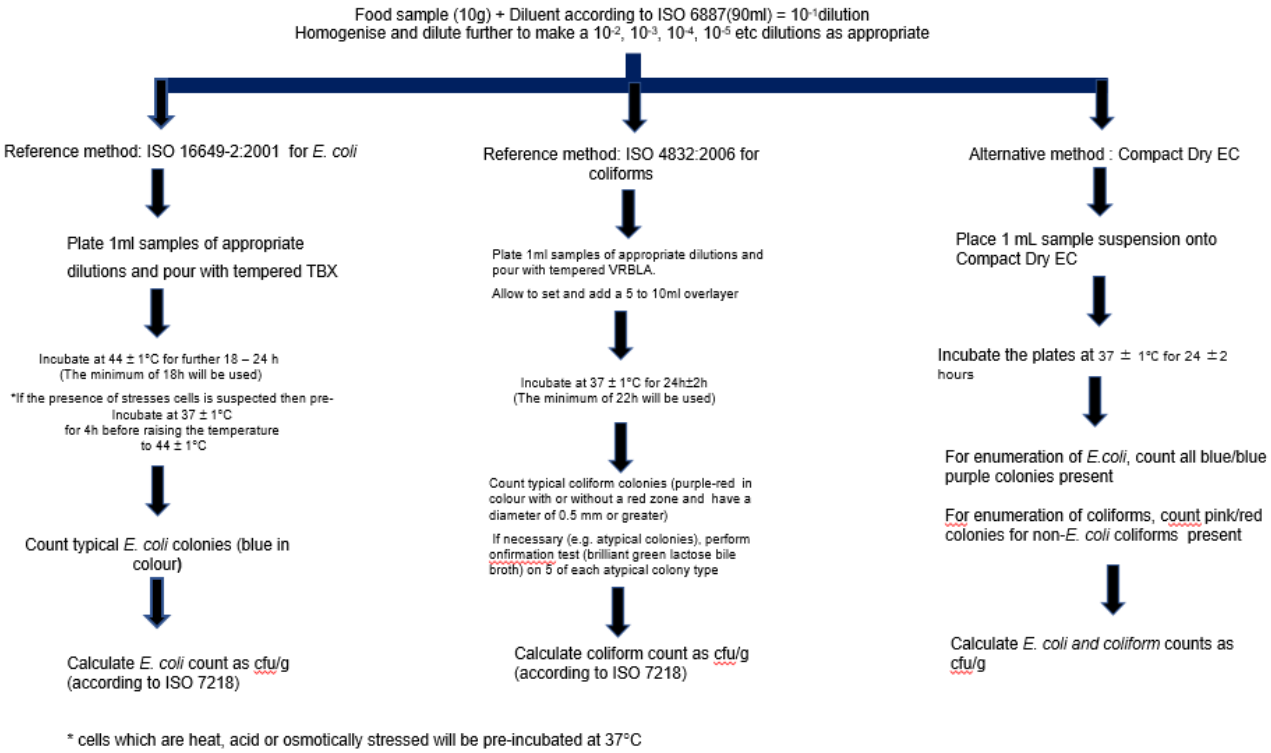
ISO 7218; Microbiology of food and animal feeding stuffs -- General requirements and guidance for microbiological examinations.

ISO 16140-2:2016; Microbiology of the food chain -- Method validation -- Part 2: Protocol for the validation of alternative (proprietary) methods against a reference method.

2016-028 (GC) Proposed MicroVal Technical committee interpretation of ISO 16140-2 and ISO

2017-063 (TC) The MicroVal Process

ANNEX A: Flow diagram of the reference method and alternative methods



* cells which are heat, acid or osmotically stressed will be pre-incubated at 37°C

ANNEX B: Kit insert(s)

Refer to separate pdf

ANNEX C: Raw data per category for the relative trueness study

Category	Types	codes	item	inoculum level	strain CF-EC (CF)	source CF-EC (CF)	inoculum level	spiking	seeding	Injury
Milk and dairy	dry milk products	1	dried skimmed milk	100	*E.coli* 1253	dried food	100		ambient 2 weeks	
Milk and dairy	dry milk products	2	strawberry whip	1000	*E.coli* 1253	dried food	1000		ambient 2 weeks	
Milk and dairy	dry milk products	3	blancmange	10000	*E.coli* 1253	dried food	10000		ambient 2 weeks	
Milk and dairy	dry milk products	4	instant custard mix	100000	E.coli 1253	dried food	100000		ambient 2 weeks	
Milk and dairy	dry milk products	5	baby mikk	1000000	*E.coli* 1253	dried food	1000000		ambient 2 weeks	
Milk and dairy	dairy products	6	strawberry yogurt	100	*Citrobacter braakii* 16279	industrial isolate	100		chill 2-3 days	
Milk and dairy	dairy products	7	single cream	1000	*Citrobacter braakii* 16279	industrial isolate	1000		chill 2-3 days	
Milk and dairy	dairy products	8	halloumi with chilli	10000	*Citrobacter braakii* 16279	industrial isolate	10000		chill 2-3 days	

Category	Types	codes	item	inoculum level	strain CF-EC (CF)	source CF-EC (CF)	inoculum level	spiking	seeding	Injury
Milk and dairy	dairy products	9	full fat soft cheese	100000	*Citrobacter braakii* 16279	industrial isolate	100000		chill 2-3 days	
Milk and dairy	dairy products	10	gran levato (raw cheese)	1000000	*Citrobacter braakii* 16279	industrial isolate	1000000		chill 2-3 days	
Milk and dairy	pasteurised milk products	11	full fat milk	100	*Citrobacter youngae* NCIMB 13435	industrial isolate	100	Heat 55°C for 5 mins		0.27
Milk and dairy	pasteurised milk products	12	semi skimmed milk	1000	*Citrobacter youngae* NCIMB 13435	industrial isolate	100	Heat 55°C for 5 mins		0.27
Milk and dairy	pasteurised milk products	13	skimmed milk	10000	Citrobacter youngae NCIMB 13435	industrial isolate	100	Heat 55°C for 5 mins		0.27
Milk and dairy	pasteurised milk products	14	chocolate milk	50000	*Citrobacter youngae* NCIMB 13435	industrial isolate	100	Heat 55°C for 5 mins		0.27
Milk and dairy	pasteurised milk products	15	strawberry milk	100000	*Citrobacter youngae* NCIMB 13435	industrial isolate	100	Heat 55°C for 5 mins		0.27
Fresh produce and fruits	cut ready to eat fruits	16	melon,pineapple,mango	100	*E.coli* 6121	Horlicks	100		chill 2-3 days	

Category	Types	codes	item	inoculum level	strain CF-EC (CF)	source CF-EC (CF)	inoculum level	spiking	seeding	Injury
Fresh produce and fruits	cut ready to eat fruits	17	garpe, strawberry,blackberry	1000	*E.coli* 6121	Horlicks	1000		chill 2-3 days	
Fresh produce and fruits	cut ready to eat fruits	18	pineapple	10000	*E.coli* 6121	Horlicks	10000		chill 2-3 days	
Fresh produce and fruits	cut ready to eat fruits	19	apple and mango juice	100000	*E.coli* 6121	Horlicks	100000		chill 2-3 days	
Fresh produce and fruits	cut ready to eat fruits	20	orange juice	1000000	*E.coli* 6121	Horlicks	1000000		chill 2-3 days	
Fresh produce and fruits	cut ready to eat vegetables	21	mixed leaf salad	100	*Escherchia vulneris* CRA 2005	vegetables	100		chill 2-3 days	
Fresh produce and fruits	cut ready to eat vegetables	22	carrot batons	1000000	*Escherchia vulneris* CRA 2005	vegetables	1000000		chill 2-3 days	
Fresh produce and fruits	cut ready to eat vegetables	23	brocoli	10000	*Escherchia vulneris* CRA 2005	vegetables	10000		chill 2-3 days	
Fresh produce and fruits	cut ready to eat vegetables	24	casserole veg selection	100000	*Escherchia vulneris* CRA 2005	vegetables	100000		chill 2-3 days	

Category	Types	codes	item	inoculum level	strain CF-EC (CF)	source CF-EC (CF)	inoculum level	spiking	seeding	Injury
Fresh produce and fruits	cut ready to eat vegetables	25	cucumber,kale and mint juice	1000	*Escherchia vulneris* CRA 2005	vegetables	1000		chill 2-3 days	
Fresh produce and fruits	Leafy greens/sprouts	26	beansprouts	1000000	*Enterobacter taylorae* 7530	seeds	1000000		chill 2-3 days	
Fresh produce and fruits	Leafy greens/sprouts	27	wild rocket	1000	*Enterobacter taylorae* 7530	seeds	1000		chill 2-3 days	
Fresh produce and fruits	Leafy greens/sprouts	28	pea shoots	100000	*Enterobacter taylorae* 7530	seeds	100000		chill 2-3 days	
Fresh produce and fruits	Leafy greens/sprouts	29	watercress,spinach and rocket	10000	*Enterobacter taylorae* 7530	seeds	10000		chill 2-3 days	
Fresh produce and fruits	Leafy greens/sprouts	30	flat leaf parsley	100000	*Enterobacter taylorae* 7530	seeds	100000		chill 2-3 days	
Raw meat and poultry	fresh poultry cuts	31	chicken thighs	100	*Enterobacter cloace* 6633	industrial isolate	100		chill 2-3 days	
Raw meat and poultry	fresh poultry cuts	32	chicken drumsticks	1000	*Enterobacter cloace* 6633	industrial isolate	1000		chill 2-3 days	

Category	Types	codes	item	inoculum level	strain CF-EC (CF)	source CF-EC (CF)	inoculum level	spiking	seeding	Injury
Raw meat and poultry	fresh poultry cuts	33	chicken breast fillets	10000	*Enterobacter cloace* 6633	industrial isolate	10000		chill 2-3 days	
Raw meat and poultry	fresh poultry cuts	34	diced turkey thigh	100000	*Enterobacter cloace* 6633	industrial isolate	100000		chill 2-3 days	
Raw meat and poultry	fresh poultry cuts	35	thin cut turkey breast steaks	1000000	*Enterobacter cloace* 6633	industrial isolate	1000000		chill 2-3 days	
Raw meat and poultry	fresh mince	36	lean beef steak mince	100	*Enterobacter agglomerans* 1488	raw mince	100		chill 2-3 days	
Raw meat and poultry	fresh mince	37	beef mince	1000	*Enterobacter agglomerans* 1488	raw mince	1000		chill 2-3 days	
Raw meat and poultry	fresh mince	38	lean pork mince	10000	*Enterobacter agglomerans* 1488	raw mince	10000		chill 2-3 days	
Raw meat and poultry	fresh mince	39	lamb mince	100000	*Enterobacter agglomerans* 1488	raw mince	100000		chill 2-3 days	
Raw meat and poultry	fresh mince	40	turkey breast mince	1000000	*Enterobacter agglomerans* 1488	raw mince	1000000		chill 2-3 days	

Category	Types	codes	item	inoculum level	strain CF-EC (CF)	source CF-EC (CF)	inoculum level	spiking	seeding	Injury
Raw meat and poultry	processed ready to cook	41	frozen beef burger	100	*Escherchia fergusonii* CRA 7522	sausages	100		frozen 2 weeks	
Raw meat and poultry	processed ready to cook	42	cheese and ham kievs	1000	*Escherchia fergusonii* CRA 7522	sausages	1000		frozen 2 weeks	
Raw meat and poultry	processed ready to cook	43	southern fried chicken goujons	10000	*Escherchia fergusonii* CRA 7522	sausages	10000		frozen 2 weeks	
Raw meat and poultry	processed ready to cook	44	breaded mini chicken fillets	100000	*Escherchia fergusonii* CRA 7522	sausages	100000		frozen 2 weeks	
Raw meat and poultry	processed ready to cook	45	garlic chicken kievs	1000000	*Escherchia fergusonii* CRA 7522	sausages	1000000		frozen 2 weeks	
ready to eat foods	ready to eat poultry	46	roast turkey breast slices	100	*Enterobacter arachdis* NCIMB 14469	industrial isolate	100		chill 2-3 days	
ready to eat foods	ready to eat poultry	47	roast chicken breast pieces	1000	*Enterobacter arachdis* NCIMB 14469	industrial isolate	1000		chill 2-3 days	
ready to eat foods	ready to eat poultry	48	chicken slices	10000	*Enterobacter arachdis* NCIMB 14469	industrial isolate	10000		chill 2-3 days	

Category	Types	codes	item	inoculum level	strain CF-EC (CF)	source CF-EC (CF)	inoculum level	spiking	seeding	Injury
ready to eat foods	ready to eat poultry	49	chicken drumsticks	100000	*Enterobacter arachdis* NCIMB 14469	industrial isolate	100000		chill 2-3 days	
ready to eat foods	ready to eat poultry	50	chicken thighs	1000000	*Enterobacter arachdis* NCIMB 14469	industrial isolate	1000000		chill 2-3 days	
ready to eat foods	cooked fish products	51	hot smoked mackerel	100	*Enterobacter amingenus* NCIMB 2118	sea water	100		chill 2-3 days	
ready to eat foods	cooked fish products	52	prawn cocktail	1000	*Enterobacter amingenus* NCIMB 2118	sea water	1000		chill 2-3 days	
ready to eat foods	cooked fish products	53	smoked salmon pate	10000	*Enterobacter amingenus* NCIMB 2118	sea water	10000		chill 2-3 days	
ready to eat foods	cooked fish products	54	cold water prawns	100000	*Enterobacter amingenus* NCIMB 2118	sea water	100000		chill 2-3 days	
ready to eat foods	cooked fish products	55	smoked salmon pieces	1000000	*Enterobacter amingenus* NCIMB 2118	sea water	1000000		chill 2-3 days	

Category	Types	codes	item	inoculum level	strain CF-EC (CF)	source CF-EC (CF)	inoculum level	spiking	seeding	Injury
ready to eat foods	cooked meat products	56	corned beef	100	Enterobacter gergoviae NCIMB 13304	industrial isolate	100	Heat 55°C for 5 mins		0.66
ready to eat foods	cooked meat products	57	cooked ham	300	Enterobacter gergoviae NCIMB 13304	industrial isolate	500	Heat 55°C for 5 mins		0.66
ready to eat foods	cooked meat products	58	pork luncheon meat	500	Enterobacter gergoviae NCIMB 13304	industrial isolate	1000	Heat 55°C for 5 mins		0.66
ready to eat foods	cooked meat products	59	German salami	1000	Enterobacter gergoviae MCIMB 13304	industrial isolate	5000	Heat 55oc for 5 mins		0.66
ready to eat foods	cooked meat products	60	Brussels pate	1000	Enterobacter gergoviae NCIMB 13304	industrial isolate	10000	Heat 55°C for 5 mins		0.66
multi component foods	ready to re-heat chilled foods	61	tagliatelle	100	Enterobacter xiangfangensis NCIMB 14836	industrial isolate	100		chill 2-3 days	
multi component foods	ready to re-heat chilled foods	62	spaghetti	1000	*Enterobacter xiangfangensis* NCIMB 14836	industrial isolate	1000		chill 2-3 days	

Category	Types	codes	item	inoculum level	strain CF-EC (CF)	source CF-EC (CF)	inoculum level	spiking	seeding	Injury
multi component foods	ready to re-heat chilled foods	63	chicken and bacon tortellini	10000	*Enterobacter xiangfangensis* NCIMB 14836	industrial isolate	10000		chill 2-3 days	
multi component foods	ready to re-heat chilled foods	64	egg fried rice	100000	*Enterobacter xiangfangensis* NCIMB 14836	industrial isolate	100000		chill 2-3 days	
multi component foods	ready to re-heat chilled foods	65	pilau rice	1000000	*Enterobacter xiangfangensis* NCIMB 14836	industrial isolate	1000000		chill 2-3 days	
multi component foods	ready to re-heat frozen foods	66	frozen fish cakes	100	*Klebsiella trevisanii* NCIMB 8606	NCIMB 8606	100		frozen 2 weeks	
multi component foods	ready to re-heat frozen foods	67	frozen chicken nuggets	1000	*Klebsiella trevisanii* NCIMB 8606	NCIMB 8606	1000		frozen 2 weeks	
multi component foods	ready to re-heat frozen foods	68	frozen chips	10000	*Klebsiella trevisanii* NCIMB 8606	NCIMB 8606	10000		frozen 2 weeks	

Category	Types	codes	item	inoculum level	strain CF-EC (CF)	source CF-EC (CF)	inoculum level	spiking	seeding	Injury
multi component foods	ready to re-heat frozen foods	69	frozen cheese and tomato pizza	100000	*Klebsiella trevisanii* NCIMB 8606	NCIMB 8606	100000		frozen 2 weeks	
multi component foods	ready to re-heat frozen foods	70	frozen pepperoni pizza	1000000	*Klebsiella trevisanii* NCIMB 8606	NCIMB 8606	1000000		frozen 2 weeks	
multi component foods	composite foods with raw ingredients	71	chicken salad mayo sandwich	100	*Klebsiella ozaenae* 4273	industrial isolate	100		chill 2-3 days	
multi component foods	composite foods with raw ingredients	72	cheese and onion sandwich	1000	*Klebsiella ozaenae* 4273	industrial isolate	1000		chill 2-3 days	
multi component foods	composite foods with raw ingredients	73	bacon,lettuce, tomato sandwich	10000	*Klebsiella ozaenae* 4273	industrial isolate	10000		chill 2-3 days	
multi component foods	composite foods with raw ingredients	74	rainbow salad	100000	*Klebsiella ozaenae* 4273	industrial isolate	100000		chill 2-3 days	

Category	Types	codes	item	inoculum level	strain CF-EC (CF)	source CF-EC (CF)	inoculum level	spiking	seeding	Injury
multi component foods	composite foods with raw ingredients	75	minted bean salad	1000000	*Klebsiella ozaenae* 4273	industrial isolate	1000000		chill 2-3 days	

Appendix 4 – Relative trueness study: raw data

codes	item	Reference										Alternative									
	dilutions	-1	-1	-2	-3	-4	-5	-6	-7	count cfu/g	Log cfu/g	-1	-1	-2	-3	-4	-5	-6	-7	count cfu/g	Log cfu/g
1C	dried skimmed milk	T	T	158	29	12				3.70E+04	4.57	T	T	109	9	3				1.10E+05	5.04
2	strawberry whip		48	10	1					1.00E+03	3.00		T	18	2					1.80E+03	3.26
3	blancmange		43	2						4.10E+02	2.61		30	1						2.80E+02	2.45
4	instant custard mix		T	30	2					2.90E+03	3.46	T		20	3					2.10E+03	3.32
5	baby mikk			T	41	3				4.00E+04	4.60			T	24	0				2.40E+04	4.38
6B	strawberry yogurt	11	7							1.10E+02	2.04	*	7							7.00E+02	2.85

codes	item	Reference										Alternative									
7	single cream		T	52	1					4.80E+03	3.68			20						2.00E+03	3.30
8	halloumi with chilli		T	37	5					3.90E+03	3.59			20	2					2.00E+03	3.30
9	full fat soft cheese		T	T	49	3				4.70E+04	4.67				22	4				2.30E+04	4.36
10	gran levato (raw cheese)				110	10				1.10E+05	5.04				55	10				5.90E+04	4.77
11	full fat milk	33	39	2						3.50E+02	2.54	30	18	2						2.40E+02	2.38
12	semi skimmed milk		T	18	2					1.80E+03	3.26		125	12						1.20E+03	3.08
13	skimmed milk		T	107	7					1.00E+04	4.00		T	115	10					1.20E+04	4.08
14	chocolate milk		T	T	31	5				3.30E+04	4.52		T	T	37	8				4.10E+04	4.61

codes	item	Reference										Alternative									
15	strawberry milk			T	65	5				6.40E+04	4.81			T	79	4				7.60E+04	4.88
16B	melon,pineapple,mango		T	T	51	2				4.80E+04	4.68		T	T	34	1				3.20E+04	4.51
17	garpe, strawberry,blackberry		59	7						6.00E+02	2.78		29							2.90E+02	2.46
18	pineapple		T	47	5					4.70E+03	3.67		T	25						2.50E+03	3.40
19	apple and mango juice		T	T	93	10				9.40E+04	4.97		T	T	44	10				4.90E+04	4.69
20	orange juice			T	T	35	2			3.40E+05	5.53			T	T	20	2			2.00E+05	5.30
21	mixed leaf salad				290					2.90E+05	5.46				192					1.90E+05	5.28
22	carrot batons				T	21				2.10E+05	5.32					18				1.80E+05	5.26

codes	item	Reference										Alternative									
23B	brocoli		T	T	30	2				2.90E+04	4.46		T	T	22	0				2.20E+04	4.34
24	casserole veg selection						36	5		3.70E+06	6.57					154	14			1.50E+06	6.18
25B	cucumber,kale and mint juice		T	T	34	4				3.50E+04	4.54		T	T	20	2				2.00E+04	4.30
26B	beansprouts					T	T	21		2.10E+07	7.32					T	T	17		1.70E+07	7.23
27	wild rocket		81	8						8.10E+02	2.91		54	11						5.90E+02	2.77
28	pea shoots						56			5.60E+06	6.75						25			2.50E+06	6.40
29	watercress,spinach and rocket						54	8		5.60E+06	6.75						37	5		3.80E+06	6.58
30	flat leaf parsley						111	9		1.10E+07	7.04						43	5		4.40E+06	6.64

codes	item	Reference										Alternative									
31B	chicken thighs					31	4			3.20E+05	5.51				42	5				4.30E+05	5.63
32	chicken drumsticks				110					1.10E+05	5.04				96	10				9.60E+04	4.98
33	chicken breast fillets					78	4			7.50E+05	5.88					62	6			6.20E+05	5.79
34	diced turkey thigh						53	6		5.40E+06	6.73						37	2		3.60E+06	6.56
35	thin cut turkey breast steaks							22	5	2.50E+07	7.40							31	5	3.30E+07	7.52
36	lean beef steak mince	54	41	2						4.50E+02	2.65	32	25	4						2.95E+02	2.47
37	beef mince		T	33	3					3.30E+03	3.52		T	24	7					2.80E+03	3.45
38	lean pork mince		T	T	26	0				2.60E+04	4.41		T	T	33	2				3.20E+04	4.51

codes	item	Reference										Alternative									
39	lamb mince					65	5			6.40E+05	5.81					39	5			4.00E+05	5.60
40	turkey breast mince						53	6		5.40E+06	6.73						41	2		3.90E+06	6.59
41	frozen beef burger	89	101	10						9.54E+02	2.98	49	46	5						4.77E+02	2.68
42	cheese and ham kievs		T	101	18					1.10E+04	4.04		T	75	11					7.80E+03	3.89
43	southern fried chicken goujons		T	156	14					1.50E+04	4.18				47	8				5.00E+04	4.70
44	breaded mini chicken fillets						19	2		1.90E+06	6.28						24			2.40E+06	6.38
45	garlic chicken kievs						51	2		4.80E+06	6.68						40	5		4.10E+06	6.61
46	roast turkey breast slices	20	16							1.80E+02	2.26	28	26	3						2.70E+02	2.43

codes	item	Reference										Alternative									
47	roast chicken breast pieces		T	18						1.80E+03	3.26		T	27	1					2.60E+03	3.41
48	chicken slices		T	T	16	1				1.60E+04	4.20		T	T	16	2				1.60E+04	4.20
49	chicken drumsticks		T	T	T	21				2.10E+05	5.32		T	T	T	17	3			1.80E+05	5.26
50	chicken thighs			T	T	110	6			1.10E+06	6.04			T	T	119	8			1.20E+06	6.08
51	hot smoked mackerel	18	14							1.60E+02	2.20	19	17	2						1.82E+02	2.26
52	prawn cocktail		T	38	3					3.70E+03	3.57		26	2						2.60E+03	3.41
53	smoked slamon pate		T	T	20	4				2.20E+04	4.34		T	150	10	4				1.50E+04	4.18

ANNEX D: Calculation and interpretation of relative trueness

Category	Type	Sample code	log(Ref)	log(Alt)	Mean	Difference
Environmental						
Dust	Industrial kitchen sink corner	211	2.36	2.20	2.28	-0.15
Dust	Radiator	212	3.23	3.11	3.17	-0.12
Dust	Oven top	213	4.15	3.40	3.77	-0.75
Dust	Base of equipment	214	5.08	4.87	4.97	-0.21
Dust	Bakery sink corner	215	6.36	5.66	6.01	-0.70
Process water	Cooling water	96	3.08	2.70	2.89	-0.38
Process water	Wash water	97	3.85	3.79	3.82	-0.07
Process water	Bakery prep area	98	4.77	4.60	4.69	-0.17
Process water	Surface run off - industri kitchen	99	5.88	5.72	5.80	-0.15
Process water	High pressure cooling water	210	7.04	6.73	6.89	-0.31
Surfaces (wipes, swabs)	Metal - industrial kitchen sink	91	1.83	1.81	1.82	-0.02
Surfaces (wipes, swabs)	Ceramic - sink splashback	92	2.58	3.00	2.79	0.42
Surfaces (wipes, swabs)	Rubber - equipment piping	93	2.60	2.53	2.56	-0.08
Surfaces (wipes, swabs)	Plastic - equipment surface	94	4.53	5.08	4.81	0.55
Surfaces (wipes, swabs)	MDF - lab bench	95	6.18	6.93	6.55	0.75
Fresh produce and fruits						

Category	Type	Sample code	log(Ref)	log(Alt)	Mean	Difference
cut ready to eat fruits	melon,pineapple,mango	16	4.51	4.68	4.59	0.18
cut ready to eat fruits	grape, strawberry,blackberry	17	2.46	2.78	2.62	0.32
cut ready to eat fruits	pineapple	18	3.40	3.67	3.54	0.27
cut ready to eat fruits	apple and mango juice	19	4.69	4.97	4.83	0.28
cut ready to eat fruits	orange juice	20	5.30	5.53	5.42	0.23
cut ready to eat vegetables	mixed leaf salad	21	5.28	5.46	5.37	0.18
cut ready to eat vegetables	carrot batons	22	5.26	5.32	5.29	0.07
cut ready to eat vegetables	brocoli	23	4.34	4.46	4.40	0.12
cut ready to eat vegetables	casserole veg selection	24	6.18	6.57	6.37	0.39
cut ready to eat vegetables	cucumber,kale and mint juice	25	4.30	4.54	4.42	0.24
Leafy greens/sprouts	beansprouts	26	7.23	7.32	7.28	0.09
Leafy greens/sprouts	wild rocket	27	2.77	2.91	2.84	0.14
Leafy greens/sprouts	pea shoots	28	6.40	6.75	6.57	0.35
Leafy greens/sprouts	watercress,spinach and rocke	29	6.58	6.75	6.66	0.17
Leafy greens/sprouts	flat leaf parsley	30	6.64	7.04	6.84	0.40
Milk and dairy						
dairy products	strawberry yogurt	6	2.85	2.04	2.44	-0.80
dairy products	single cream	7	3.30	3.68	3.49	0.38
dairy products	halloumi with chilli	8	3.30	3.59	3.45	0.29
dairy products	full fat soft cheese	9	4.36	4.67	4.52	0.31
dairy products	gran levato (raw cheese)	10	4.77	5.04	4.91	0.27
dry milk products	dried skimmed milk	1	5.04	4.57	4.80	-0.47
dry milk products	strawberry whip	2	3.26	3.00	3.13	-0.26
dry milk products	blancmange	3	2.45	2.61	2.53	0.17
dry milk products	instant custard mix	4	3.32	3.46	3.39	0.14
dry milk products	baby milk	5	4.38	4.60	4.49	0.22

Category	Type	Sample code	log(Ref)	log(Alt)	Mean	Difference
pasteurised milk products	full fat milk	11	2.38	2.54	2.46	0.16
pasteurised milk products	semi skimmed milk	12	3.08	3.26	3.17	0.18
pasteurised milk products	skimmed milk	13	4.08	4.00	4.04	-0.08
pasteurised milk products	chocolate milk	14	4.61	4.52	4.57	-0.09
pasteurised milk products	strawberry milk	15	4.88	4.81	4.84	-0.07
multi component foods						
composite foods with raw ingredients	chicken salad mayo sandwich	71	3.00	3.52	3.26	0.52
composite foods with raw ingredients	cheese and onion sandwich	72	2.00	3.32	2.66	1.32
composite foods with raw ingredients	bacon,lettuce, tomato sandwich	73	4.52	5.23	4.87	0.71
composite foods with raw ingredients	rainbow salad	74	5.62	6.00	5.81	0.38
composite foods with raw ingredients	minted bean salad	75	5.58	6.30	5.94	0.72
ready to re-heat chilled foods	tagliatelle	61	2.28	2.25	2.26	-0.03
ready to re-heat chilled foods	spaghetti	62	3.40	3.26	3.33	-0.14
ready to re-heat chilled foods	chicken and bacon tortellini	63	4.11	4.15	4.13	0.03
ready to re-heat chilled foods	egg fried rice	64	5.91	5.90	5.91	-0.01
ready to re-heat chilled foods	pilau rice	65	6.49	6.63	6.56	0.14
ready to re-heat frozen foods	frozen fish cakes	66	1.60	1.60	1.60	0.00
ready to re-heat frozen foods	frozen chicken nuggets	67	3.15	3.40	3.27	0.25
ready to re-heat frozen foods	frozen chips	68	2.89	3.52	3.20	0.63

Category	Type	Sample code	log(Ref)	log(Alt)	Mean	Difference
ready to re-heat frozen foods	frozen cheese and tomato pizza	69	4.56	5.08	4.82	0.52
ready to re-heat frozen foods	frozen pepperoni pizza	70	5.88	6.04	5.96	0.17
Pet food and animal feed						
Animal feeds (poultry and fish)	Organic mixed corn for chickens	86	6.64	6.28	6.46	-0.36
Animal feeds (poultry and fish)	Chicken oyster shell supplement	87	5.75	5.48	5.61	-0.27
Animal feeds (poultry and fish)	Aquacare goldfish flakes with mealworms	88	5.70	5.36	5.53	-0.34
Animal feeds (poultry and fish)	Bug bites insect lavae and salmon	89	3.36	2.85	3.11	-0.51
Animal feeds (poultry and fish)	Tetra goldfish flakes	90	4.85	4.57	4.71	-0.28
dry food	complete dry kitten food turkey and rice	76	3.11	3.18	3.15	0.06
dry food	complete adult dry cat food turkey and rice	77	2.76	2.85	2.80	0.09
dry food	Feline health dry kitten food	78	2.61	2.77	2.69	0.16
dry food	complete natural and small breed dry adult dog food lamb and rice	79	2.83	2.77	2.80	-0.06
dry food	complete nutrition dry puppy food chicken with vegetables	80	2.24	2.40	2.32	0.15
wet food (raw and canned)	Adult dog food - smooth pate with chicken	81	3.43	3.11	3.27	-0.32

Category	Type	Sample code	log(Ref)	log(Alt)	Mean	Difference
wet food (raw and canned)	gourmet cat food - mouse with salmon and cascading gravy	82	4.72	4.20	4.46	-0.52
wet food (raw and canned)	wet adult dog food lamb with brown rice	83	5.32	5.08	5.20	-0.24
wet food (raw and canned)	deliciously tasty luxury mousse wet adult cat food salmon	84	3.46	3.46	3.46	0.00
wet food (raw and canned)	deliviously tasty tender cuts in sauce wet adult cat food turkey	85	4.18	4.18	4.18	0.00
Raw meat and poultry						
fresh mince	lean beef steak mince	36	2.47	2.65	2.56	0.18
fresh mince	beef mince	37	3.45	3.52	3.48	0.07
fresh mince	lean pork mince	38	4.51	4.41	4.46	-0.09
fresh mince	lamb mince	39	5.60	5.81	5.70	0.20
fresh mince	turkey breast mince	40	6.59	6.73	6.66	0.14
fresh poultry cuts	frozen beef burger	31	5.63	5.51	5.57	-0.13
fresh poultry cuts	cheese and ham kievs	32	4.98	5.04	5.01	0.06
fresh poultry cuts	southern fried chicken goujon	33	5.79	5.88	5.83	0.08
fresh poultry cuts	breaded mini chicken fillets	34	6.56	6.73	6.64	0.18
fresh poultry cuts	garlic chicken kievs	35	7.52	7.40	7.46	-0.12
processed ready to cook	lean beef steak mince	41	2.68	2.98	2.83	0.30
processed ready to cook	beef mince	42	3.89	4.04	3.97	0.15
processed ready to cook	lean pork mince	43	4.70	4.18	4.44	-0.52
processed ready to cook	lamb mince	44	6.38	6.28	6.33	-0.10
processed ready to cook	turkey breast mince	45	6.61	6.68	6.65	0.07
ready to eat foods						

Category	Type	Sample code	log(Ref)	log(Alt)	Mean	Difference
cooked fish products	hot smoked mackerel	51	2.26	2.20	2.23	-0.06
cooked fish products	prawn cocktail	52	3.41	3.57	3.49	0.15
cooked fish products	smoked salmon pate	53	4.18	4.34	4.26	0.17
cooked fish products	cold water prawns	54	5.38	5.20	5.29	-0.18
cooked fish products	smoked salmon pieces	55	5.96	5.79	5.88	-0.17
cooked meat products	corned beef	56	2.80	2.67	2.73	-0.13
cooked meat products	cooked ham	57	2.86	2.74	2.80	-0.11
cooked meat products	pork luncheon meat	58	3.48	3.36	3.42	-0.12
cooked meat products	German salami	59	3.48	3.72	3.60	0.25
cooked meat products	Brussels pate	60	3.76	3.68	3.72	-0.07
ready to eat poultry	roast turkey breast slices	46	2.43	2.26	2.34	-0.18
ready to eat poultry	roast chicken breast pieces	47	3.41	3.26	3.34	-0.16
ready to eat poultry	chicken slices	48	4.20	4.20	4.20	0.00
ready to eat poultry	chicken drumsticks	49	5.26	5.32	5.29	0.07
ready to eat poultry	chicken thighs	50	6.08	6.04	6.06	-0.04

ANNEX F: Summary tables Accuracy profile study.

(Food) Category 1		dairy									
(Food) Type 1		pasteurised dairy									
			Reference method result					Alternative method result				
Sample Name	(Food) item	Level	rep 1	rep 2	rep 3	rep 4	rep 5	rep 1	rep 2	rep 3	rep 4	rep 5
142 21 248 188 53	CREAM CHEESE	Low	60	70	20	80	90	60	70	60	30	30
107 45 75 342 322	CREAM	Low	1700	1200	1300	1300	1500	1000	1300	800	1200	900
31 10 97 13 273	CREAM CHEESE	Med	6600	8000	5500	7600	7500	6200	6700	4500	7200	7100
314 346 25 63 328	CREAM	Med	11000	5100	10000	9400	7500	11000	16000	24000	21000	18000
196 229 57 102 259	CREAM CHEESE	High	530000	440000	460000	460000	310000	460000	550000	440000	360000	680000
58 141 172 83 109	CREAM	High	570000	540000	640000	480000	610000	900000	580000	910000	870000	1700000

(Food) Category 2		fruit and veg									
(Food) Type 2		fresh produce									
			Reference method result					Alternative method result				
Sample Name	(Food) item	Level	rep 1	rep 2	rep 3	rep 4	rep 5	rep 1	rep 2	rep 3	rep 4	rep 5
8 17 327 246 37	veg juice	Low	45	45	70	50	35	20	25	40	25	20
264 252 139 300 294	RTC	Low	140	190	130	160	120	100	110	20	90	100
312 301 64 267 340	RTC	Med	2800	3800	3100	2100	2900	1660	1750	1140	980	1640
236 92 223 9 158	veg juice	Med	5500	5000	5700	6900	5600	5400	5100	4800	4800	8100
316 86 323 117 203	RTC	High	380000	420000	320000	360000	320000	400000	300000	300000	500000	200000
74 234 272 34 144	veg juice	High	300000	490000	570000	560000	450000	560000	620000	500000	620000	510000

(Food) Category 4		RTE foods										
(Food) Type 4		cooked fish										
			Reference method result					Alternative method result				
Sample Name	(Food) item	Level	rep 1	rep 2	rep 3	rep 4	rep 5	rep 1	rep 2	rep 3	rep 4	rep 5
51 247 218 87 287	fresh prawns	Low	159	295	464	200	240	82	232	214	227	177
111 155 255 186 202	fish pate	Low	490	500	460	390	370	1200	850	750	880	760
289 68 23 309 226	fresh prawns	Med	18000	7600	35000	44000	18000	25000	8500	33000	40000	17000
256 192 295 16 298	fish pate	Med	46000	45000	35000	51000	58000	83000	56000	100000	85000	91000
195 261 42 61 320	fresh prawns	High	570000	1100000	800000	990000	1200000	660000	850000	610000	1000000	1300000
319 189 78 82 182	fish pate	High	2600000	3500000	2000000	2500000	4800000	6900000	6300000	4700000	6300000	7100000

(Food) Category 5		Mulit component										
(Food) Type 5		food with raw ingredients										
			Reference method result					Alternative method result				
Sample Name	(Food) item	Level	rep 1	rep 2	rep 3	rep 4	rep 5	rep 1	rep 2	rep 3	rep 4	rep 5
329 30 146 275 2422	cooked chilled rice	Low	35	45	45	35	40	65	70	95	75	55
180 304 306 258 55	sandwiches	Low	60	65	70	80	150	145	100	95	95	120
242 184 211 334 331	cooked chilled rice	Med	5800	6500	5100	4500	3900	4400	5500	6600	5800	4300
18 175 1 143 138	sandwiches	Med	7200	11000	14000	5300	7200	8100	13000	20000	9900	8700
131 59 161 103 209	cooked chilled rice	High	330000	410000	360000	530000	4200000	450000	420000	500000	1700000	1700000
245 115 224 249 315	sandwiches	High	1500000	1000000	760000	430000	890000	1300000	780000	600000	560000	730000

(Food) Category 1		pet food and										
(Food) Type 1		wet food										
			Reference method					Alternative method				
Sample Name	(Food) item	Level	rep 1	rep 2	rep 3	rep 4	rep 5	rep 1	rep 2	rep 3	rep 4	rep 5
1 a-e	dog pate	low	323	225	209	318	391	405	355	277	514	436
4 a-e	cat food with gravy	low	210	268	377	350	400	327	318	423	323	473
2 a-e	dog pate	interme diate	2500	1700	1200	2400	2800	3500	2900	1700	2500	3700
5 a-e	cat food with gravy	interme diate	27000	14000	20000	19000	19000	14000	9400	15000	9700	13000
3 a-e	dog pate	high	2300000	1900000	1700000	1800000	2000000	1500000	1100000	1500000	1300000	1700000
6 a-e	cat food with gravy	high	2600000	1500000	2100000	2100000	1500000	1800000	1000000	1700000	1400000	1200000

(Food) Category 4		environmental										
(Food) Type 4		process water										
			Reference method					Alternative method				
Sample Name	(Food) item	Level	rep 1	rep 2	rep 3	rep 4	rep 5	rep 1	rep 2	rep 3	rep 4	rep 5
140 a-e	cooling water	low	615	670	610	565	682	620	700	645	590	595
137a-e	wash water	low	825	695	545	830	825	770	750	770	730	715
141 a-e	cooling water	interme diate	7500	12000	5600	6200	7000	15000	13000	14000	14000	13000
138 a-e	wash water	interme diate	14000	15000	11000	15000	15000	16000	13000	15000	14000	17000
142 a-e	cooling water	high	2500000	2400000	2300000	2100000	2200000	1400000	2000000	1900000	2500000	2100000
139 a-e	wash water	high	1500000	7900000	3100000	1500000	3500000	1800000	2600000	2800000	2500000	3000000

7 Annex G Raw data accuracy profile study

Item	Target Level*	sample number	Alternative							cfu/g	log cfu/g	Reference							cfu/g	log cfu/g
			-1	-1	-2	-3	-4	-5	-6			-1	-1	-2	-3	-4	-5	-6		
cream	low	107		T	10					1.00E+03	3.00		T	17	0				1.70E+03	3.23
		45		T	13					1.30E+03	3.11		119	9	0				1.20E+03	3.08
		75		T	8					8.00E+02	2.90		141	5	1				1.30E+03	3.11
		342		T	12					1.20E+03	3.08		138	9	1				1.30E+03	3.11
		322		T	9					9.00E+02	2.95		T	15	0				1.50E+03	3.18
cream	medium	314		T	T	11				1.10E+04	4.04		T	116	7	1			1.10E+04	4.04
		346		T	T	16				1.60E+04	4.20		T	51	3				5.10E+03	3.71
		25		T	T	24				2.40E+04	4.38		T	106	5	0			1.00E+04	4.00
		63		T	T	21				2.10E+04	4.32		T	96	7	0			9.40E+03	3.97
		328		T	T	18				1.80E+04	4.26		T	75	7	0			7.50E+03	3.88
cream	high	58		T	T	T	T	9		9.00E+05	5.95			T	T	54	9		5.70E+05	5.76

Item	Target Level*	sample number	Alternative							cfu/g	log cfu/g	Reference							cfu/g	log cfu/g
			-1	-1	-2	-3	-4	-5	-6			-1	-1	-2	-3	-4	-5	-6		
		141		T	T	T	60	4		5.80E+05	5.76			T	T	54	5		5.40E+05	5.73
		172		T	T	T	91	9		9.10E+05	5.96			T	T	64	6		6.40E+05	5.81
		83		T	T	T	87	9		8.70E+05	5.94			T	T	44	9		4.80E+05	5.68
		109		T	T	T		16	3	1.70E+06	6.23			T	T	61	6		6.10E+05	5.79
cream cheese	low	142		6	2	0				60	1.78		6	0	0				6.00E+01	1.78
		21		7	0	0				70	1.85		7	0	0				7.00E+01	1.85
		248		6	0	0				60	1.78		2	0	0				2.00E+01	1.30
		188		3	0	0				30	1.48		8	0	0				8.00E+01	1.90
		53		3	0	0				30	1.48		9	0	0				9.00E+01	1.95
cream cheese	medium	31		T	63	5	0			6.20E+03	3.79		T	65	8				6.60E+03	3.82
		10		T	67	7	0			6.70E+03	3.83		T	76	12				8.00E+03	3.90
		97		T	42	7	0			4.50E+03	3.65		T	59	2				5.50E+03	3.74

Item	Target Level*	sample number	Alternative							cfu/g	log cfu/g	Reference							cfu/g	log cfu/g
			-1	-1	-2	-3	-4	-5	-6			-1	-1	-2	-3	-4	-5	-6		
		13		T	T	74	5			7.20E+03	3.86		T	75	9				7.60E+03	3.88
		273		T	T	72	6			7.10E+03	3.85		T	68	14				7.50E+03	3.88
cream cheese	high	196				T	49	1		4.60E+05	5.66			T	T	52	6		5.30E+05	5.72
		229				T	58	2		5.50E+05	5.74			T	T	46	2		4.40E+05	5.64
		57				T	43	5		4.40E+05	5.64			T	T	49	2		4.60E+05	5.66
		102				T	33	6		3.60E+05	5.56			T	T	47	4		4.60E+05	5.66
		259				T	68	7		6.80E+05	5.83			T	T	31	0		3.10E+05	5.49
RTC veg	low	264		10	3	0				1.00E+02	2.00		14	2	0				1.40E+02	2.15
		252		11	0	0				1.10E+02	2.04		16	5	0				1.90E+02	2.28
		139		2	0	0				2.00E+01	1.30		13	0	0				1.30E+02	2.11
		300		9	1	0				9.00E+01	1.95		16	0	0				1.60E+02	2.20
		294		10	0	0				1.00E+02	2.00		12	0	0				1.20E+02	2.08

Item	Target Level*	sample number	Alternative							cfu/g	log cfu/g	Reference							cfu/g	log cfu/g
			-1	-1	-2	-3	-4	-5	-6			-1	-1	-2	-3	-4	-5	-6		
RTC veg	medium	312		166	1	0				1.66E+03	3.22		T	29	2				2.80E+03	3.45
		301		175	0	0				1.75E+03	3.24		T	40	2				3.80E+03	3.58
		64		114	1	0				1.14E+03	3.06		T	30	4				3.10E+03	3.49
		267		98	0	0				9.80E+02	2.99		T	21	2				2.10E+03	3.32
		340		164	2	0				1.64E+03	3.21		T	30	2				2.90E+03	3.46
RTC veg	high	316						4		4.00E+05	5.60			T	T	39	3		3.80E+05	5.58
		86						3		3.00E+05	5.48			T	T	44	2		4.20E+05	5.62
		323						3		3.00E+05	5.48			T	T	31	4		3.20E+05	5.51
		117						5		5.00E+05	5.70			T	T	36	0		3.60E+05	5.56
		203						1		2.00E+05	5.30			T	T	31	4		3.20E+05	5.51
Vegetable juice	low	8	2	2						20	1.30	4	5						4.50E+01	1.65
		17	4	1						25	1.40	4	5						4.50E+01	1.65

Item	Target Level*	sample number	Alternative							cfu/g	log cfu/g	Reference							cfu/g	log cfu/g
			-1	-1	-2	-3	-4	-5	-6			-1	-1	-2	-3	-4	-5	-6		
		327	4	4						40	1.60	7	1						7.00E+01	1.85
		246	2	1						25	1.40	5	5						5.00E+01	1.70
		37	2	2						20	1.30	3	4						3.50E+01	1.54
Vegetable juice	medium	236		T	51	8	1			5.40E+03	3.73		T	56	4	0			5.50E+03	3.74
		92		T	50	6	0			5.10E+03	3.71		T	50	5	0			5.00E+03	3.70
		223		T	49	4	2			4.80E+03	3.68		T	54	9	3			5.70E+03	3.76
		9		T	49	4	1			4.80E+03	3.68		T	69	0	0			6.90E+03	3.84
		158		T	81	8	1			8.10E+03	3.91		T	53	8	1			5.60E+03	3.75
Vegetable juice	high	74			T	T	56	6		5.60E+05	5.75			T	T	28	5		3.00E+05	5.48
		234			T	T	59	9		6.20E+05	5.79			T	T	50	4		4.90E+05	5.69
		272			T	T	48	7		5.00E+05	5.70			T	T	59	4		5.70E+05	5.76
		34			T	T	60	8		6.20E+05	5.79			T	T	57	5		5.60E+05	5.75

Item	Target Level*	sample number	Alternative							cfu/g	log cfu/g	Reference							cfu/g	log cfu/g
			-1	-1	-2	-3	-4	-5	-6			-1	-1	-2	-3	-4	-5	-6		
		144			T	T	51	5		5.10E+05	5.71			T	T	46	3		4.50E+05	5.65
pork mince	low	168	6	8	0	0	0			7.00E+01	1.85	3	3	0	0	0	0		3.00E+01	1.48
		135	3	4	0	0	0			3.50E+01	1.74	5	6	0	0	0			5.50E+01	1.74
		283	23	13	0	0	0			1.80E+02	2.02	11	10	0	0	0			1.05E+02	2.02
		214	22	22	1	0	0			2.20E+02	2.10	12	13	0	0	0			1.25E+02	2.10
		47	3	3	0	0	0			3.00E+01	1.88	6	9	0	0	0			7.50E+01	1.88
pork mince	medium	348				49	5			4.90E+04	4.69		T	T	T	4			4.00E+04	4.60
		171				30	2			3.00E+04	4.48				T	8			8.00E+04	4.90
		65				30	7			3.00E+04	4.48				70	13			7.00E+04	4.85
		281				32	5			3.20E+04	4.51				89	8			8.00E+04	4.90
		151				32	3			3.20E+04	4.51				T	4			4.00E+04	4.60
pork mince	high	167				19	5			5.00E+04	4.70				T	6			6.40E+04	4.81

Item	Target Level*	sample number	Alternative							cfu/g	log cfu/g	Reference							cfu/g	log cfu/g
			-1	-1	-2	-3	-4	-5	-6			-1	-1	-2	-3	-4	-5	-6		
		36				T	30			3.00E+05	5.48				T	10			1.00E+05	5.00
		324				41	21			2.10E+05	5.32				T	15	0		1.50E+05	5.18
		216				51	8			8.00E+04	4.90				T	22	0		2.20E+05	5.34
		133				51	16			1.60E+05	5.20				T	11			1.10E+05	5.04
raw beef	low	204	T	T	37,24	1,4	0			3.05E+03	3.48	T	T	42,31	2,1	0			3.65E+03	3.56
		253	T	T	24.20	0,0	0			2.20E+03	3.34	T	T	48,46	5,2	0			4.70E+03	3.67
		207	T	T	17,25	1,0	0			2.10E+03	3.32	T	T	30,36	3,2	0			3.30E+03	3.52
		76	T	T	58,31	5,2	0			4.45E+03	3.65	T	T	44,29	1,1	0			7.30E+03	3.86
		156	T	T	31,31	3.4	0			3.10E+03	3.49	T	T	60,36	5,2	0			4.80E+03	3.68
raw beef	medium	284			T,T	23.23				2.30E+04	4.36			T,T	31,23				2.70E+04	4.43
		225			T,T	28,15				2.15E+04	4.33			T,T	23,14				1.85E+04	4.27
		85			216,215					2.16E+04	4.33			211,232					2.22E+04	4.35

Item	Target Level*	sample number	Alternative							cfu/g	log cfu/g	Reference							cfu/g	log cfu/g
			-1	-1	-2	-3	-4	-5	-6			-1	-1	-2	-3	-4	-5	-6		
		257			217,185					2.01E+04	4.30			264,206					2.35E+04	4.37
		96			237,217					2.27E+04	4.36			297,186					2.42E+04	4.38
raw beef	high	165						10,10		1.00E+06	6.00						15,15		1.50E+06	6.18
		69						25,25		2.50E+06	6.40						74,51		6.25E+06	6.80
		310						11,8		9.50E+06	6.98						15,19		1.70E+06	6.23
		118						27,25		2.60E+06	6.41						87,60		7.35E+06	6.87
		349						17,13		1.50E+06	6.18						28,36		3.20E+06	6.51
fresh prawns	low	51	7	9	1	0				8.20E+01	1.91	10	13	6	0				1.59E+02	2.20
		247	21	20	3	1				2.32E+02	2.37	23	36	0	1				2.95E+02	2.47
		218	17	26	5	1				2.14E+02	2.33	44	42	8	0				4.64E+02	2.67
		87	26	20	2	0				2.27E+02	2.36	19	21	2	0				2.00E+02	2.30
		287	22	15	1	0				1.77E+02	2.25	20	28	0					2.40E+02	2.38

Item	Target Level*	sample number	Alternative							cfu/g	log cfu/g	Reference							cfu/g	log cfu/g
			-1	-1	-2	-3	-4	-5	-6			-1	-1	-2	-3	-4	-5	-6		
fresh prawns	medium	289		T	T	26	1			2.50E+04	4.40		T	T	18	0			1.80E+04	4.26
		68		T	87	6				8.50E+03	3.93		T	76	7				7.60E+03	3.88
		23		T	T	36	1			3.30E+04	4.52		T	T	33	5			3.50E+04	4.54
		309		T	T	39	5			4.00E+04	4.60		T	T	44	4			4.40E+04	4.64
		226		T	T	17	2			1.70E+04	4.23		T	T	19	1			1.80E+04	4.26
fresh prawns	high	195			T	T	66	7		6.60E+05	5.82			T	T	59	4		5.70E+05	5.76
		261			T	T	86	7		8.50E+05	5.93			T	T	104	12		1.10E+06	6.04
		42			T	T	60	7		6.10E+05	5.79			T	T	78	10		8.00E+05	5.90
		61			T	T	104	9		1.00E+06	6.00			T	T	104	5		9.90E+05	6.00
		320			T	T	132	9		1.30E+06	6.11			T	T	T	12		1.20E+06	6.08
fish pate	low	111	94	136	14	0				1.20E+03	3.08	53	47	4	0				4.90E+02	2.69
		155	81	87	10	0				8.50E+02	2.93	45	52	7	0				5.00E+02	2.70

Item	Target Level*	sample number	Alternative							cfu/g	log cfu/g	Reference							cfu/g	log cfu/g
			-1	-1	-2	-3	-4	-5	-6			-1	-1	-2	-3	-4	-5	-6		
		255	83	63	9	0				7.50E+02	2.88	53	38	5	0				4.60E+02	2.66
		186	96	83	7	0				8.80E+02	2.94	40	31	7	0				3.90E+02	2.59
		202	79	65	11	1				7.60E+02	2.88	30	38	7	0				3.70E+02	2.57
fish pate	medium	256		T	T	83	9			8.30E+04	4.92		T	T	43	7			4.60E+04	4.66
		192		T	T	59	3			5.60E+04	4.75		T	T	45	4			4.50E+04	4.65
		295		T	T	104	9			1.00E+05	5.00		T	T	34	4			3.50E+04	4.54
		16		T	T	80	13			8.50E+04	4.93		T	T	51	5			5.10E+04	4.71
		298		T	T	95	5			9.10E+04	4.96		T	T	57	7			5.80E+04	4.76
fish pate	high	319			T	T	T	70	6	6.90E+06	6.84			T	T	T	20	8	2.60E+06	6.41
		189			T	T	T	63	6	6.30E+06	6.80			T	T	T	33	5	3.50E+06	6.54
		78			T	T	T	49	3	4.70E+06	6.67			T	T	T	21	1	2.00E+06	6.30
		82			T	T	T	65	4	6.30E+06	6.80			T	T	T	24	3	2.50E+06	6.40

Item	Target Level*	sample number	Alternative							cfu/g	log cfu/g	Reference							cfu/g	log cfu/g
			-1	-1	-2	-3	-4	-5	-6			-1	-1	-2	-3	-4	-5	-6		
		182			T	T	T	68	10	7.10E+06	6.85			T	T	T	48	4	4.80E+06	6.68
sandwiches	low	180	13	16	1	0				1.45E+02	2.16	5	7	2	0				6.00E+01	1.78
		304	10	10	1	1				1.00E+02	2.00	8	5	1	0				6.50E+01	1.81
		306	11	8	1	1				9.50E+01	1.98	8	6	1	1				7.00E+01	1.85
		258	10	9	0	0				9.50E+01	1.98	10	6	1	0				8.00E+01	1.90
		55	13	9	1	0				1.20E+02	2.08	16	14	1	0				1.50E+02	2.18
sandwiches	medium	18	T	80	9	1				8.10E+03	3.91		T	75	4				7.20E+03	3.86
		175		T	124	17	1			1.30E+04	4.11		T	116	9	0			1.10E+04	4.04
		1		T	T	21	1			2.00E+04	4.30		T	143	13	1			1.40E+04	4.15
		143		T	104	5	0			9.90E+03	4.00		T	55	3	0			5.30E+03	3.72
		138		T	85	11	1			8.70E+03	3.94		T	73	6	0			7.20E+03	3.86
sandwiches	high	245			T	T	130	13	1	1.30E+06	6.11			T	T	15	1		1.50E+06	6.18

Item	Target Level*	sample number	Alternative							cfu/g	log cfu/g	Reference							cfu/g	log cfu/g
			-1	-1	-2	-3	-4	-5	-6			-1	-1	-2	-3	-4	-5	-6		
		115			T	T	78	8	1	7.80E+05	5.89			T	T	100	10	1	1.00E+06	6.00

9 Annex H – Inclusivity / Exclusivity: raw data Inclusivity strains

				ALTERNATE				REFERENCE				NON SELECTIVE			
Number	Organism	Source (if known)	code	-5	-6	-7	cfu/g	-5	-6	-7	cfu/g	-5	-6	-7	cfu/g
1	Citrobacter amalonaticus	Beansprouts	7458	T	T	40	4.00E+08	T	T	44	4.40E+08	T	T	75	7.50E+08
2	Citrobacter braakii	Industrial isolate	16279	T	T	38	3.80E+08	T	T	23	2.30E+08	T	T	58	5.80E+08
3	Citrobacter youngae	Industrial isolate	ncimb 1345	T	T	20	2.00E+08	T	T	18	1.80E+08	T	T	49	4.90E+08
4	Enterobacter agglomerans	Skimmed milk powder	5513	T	T	40	4.00E+08	T	T	32	3.20E+08	T	T	44	4.40E+08
5	Enterobacter agglomerans	Raw mince	1488	T	45	4	4.50E+07	T	11	1	1.10E+07	T	50	3	4.80E+07
6	Enterobacter amingenus	Sea water	ncimb 1218	T	T	39	3.90E+08	T	T	40	4.00E+08	T	T	44	4.40E+08

7	Enterobacter arachdis	Industrial isolate	ncimb14469	T	T	13	1.30E+08	T	15	0	1.50E+07	T	T	43	4.30E+08
8	Enterobacter cloace	Industrial isolate	6633	T	T	28	2.80E+08	T	T	27	2.70E+08	T	T	39	3.90E+08
9	Enterobacter gergoviae	Industrial isolate	ncimb13304	T	T	83	8.30E+08	T	T	80	8.00E+08	T	T	81	8.10E+08
10	Enterobacter taylorae	seeds	7530	T	T	99	9.90E+08	T	T	120	1.20E+09	T	T	88	8.80E+08
11	Enterobacter xiangfangensis	Industrial isolate	ncimb 14836	T	T	95	9.50E+08	T	T	92	9.20E+08	T	T	89	8.90E+08
12	Escherichia adecarboxylata	Skimmed milk powder	5501	T	T	30	3.00E+08	T	T	26	2.60E+08	T	T	29	2.90E+08
13	Shimwellia blattae	NCTC 12127	nctc12127	0	0	0	<1.0E+05	0	0	0	<1.0E+05	T	T	29	2.90E+08
14	Escherichia fergusonii	Sausages	7522	T	T	56	5.60E+08	T	T	60	6.00E+08	T	T	103	1.00E+09
15	Escherichia hermanii	Sesame seeds	7477	46	6	2	4.70E+06	4	0	0	4.00E+05	T	T	28	2.80E+08
16	Escherichia vulneris	Industrial isolate	16260	T	T	16	1.60E+08	T	T	20	2.00E+08	T	T	27	2.70E+08

17	Escherichia vulneris	vegetables	2005	T	T	17	1.70E+08	T	T	23	2.30E+08	T	T	28	2.80E+08
18	Klebsiella ozaenae	Industrial isolate	4273	T	T	14	1.40E+08	T	T	14	1.40E+08	T	T	16	1.60E+08
19	Klebsiella rhinoscleromatis	Industrial isolate	4272	0	0	0	<1.0E+05	0	0	0	<1.0E+05	T	114	4	1.10E+08
20	Klebsiella trevisanii	NCIMB 8606	ncimb8606	T	34	5	3.60E+07	T	67	1	6.20E+07	T	T	43	4.30E+08

T=Too many to count

Shaded = no growth on alternate or reference method *Shaded cells represent non-detection of an inclusivity strain by one or both methods*

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Raw data Exclusivity strains

				ALTERNATE				REFERENCE				NON SELECTIVE			
Number	Organism	Source (if known)	code	-5	-6	-7	cfu/g	-5	-6	-7	cfu/g	-5	-6	-7	cfu/g
21	Aeromonas hydrophila	Not known	4111	T	12	0	1.20E+07	T	31	4	3.20E+07	T	T	32	3.20E+08
22	Aeromonas salmonicida	Not known	8388	0	0	0	<1.0E+05	0	0	0	<1.0E+05	t	39	3	3.80E+07
23	Aeromonas sobria	sewage	8390	T	T	17	1.70E+08	T	T	24	2.40E+08	T	T	105	1.10E+09
24	Flavobacterium resinovorum	soil	9000	0	0	0	<1.0E+05	0	0	0	<1.0E+05	T	T	48	4.80E+08
25	Lactobacillus acidophilus	rats	7570	0	0	0	<1.0E+05	0	0	0	<1.0E+05	T	41	0	4.10E+07
26	Pasteurella avium	Not known	8389	0	0	0	<1.0E+05	0	0	0	<1.0E+05	T	66	7	6.60E+07
27	Serratia fonticola	Chicken	4613	T	T	25	2.50E+08	T	44	5	4.50E+07	T	T	37	3.70E+08
28	Serratia liquefaciens	Industrial isolate	10670	0	0	0	<1.0E+05	0	0	0	<1.0E+05	T	T	42	4.20E+08
29	Serratia proteamaculans	Mushroom	1643	0	0	0	<1.0E+05	0	0	0	<1.0E+05	T	T	78	7.80E+08
30	Shewanella putrifaciens	Not known	16056	0	0	0	<1.0E+05	0	0	0	<1.0E+05	T	T	91	9.10E+08

T= Too many too count

Note: Shaded cells represent detection of an exclusivity strain by one or both methods

Inclusivity results obtained with 33 strains using the Compact Dry EC coliform method and ISO 4832 (data from old study)

ORGANISM	CCFRA CODE	COMPACT DRY EC	ISO 4832 (2006)
Citrobacter amalonaticus	6784	T	T
Citrobacter freundii	40	T	T
Citrobacter freundii	3163	T	T
Citrobacter freundii	6759	T	T
Citrobacter freundii	6763	T	T
Citrobacter koseri	7119	T	T
Citrobacter koseri	7126	T	T
Enterobacter aerogenes	15736	T	T
Enterobacter cloacae	1472	T	T
Enterobacter cloacae	6633	T	T
Escherichia coli	1476	T	T
Escherichia coli	1871	T	T
Escherichia coli	1872	T	T
Escherichia coli	1873	*T*	*T*
Escherichia coli	1967	T	T
Escherichia coli	1968	T	T
Escherichia coli	2003	T	T
Escherichia coli	2006	T	T

Escherichia coli	2091	T	T
Escherichia coli	2092	T	T
Escherichia coli	11017	T	T
Escherichia coli	11626	***9.1*** *T*	***9.2*** *T*
Escherichia coli	15943	T	T
Escherichia coli	16041	T	T
Escherichia coli	16042	T	T
Escherichia coli	16043	T	T
Escherichia coli	16044	T	T
Klebsiella oxytoca	6764	T	T
Klebsiella oxytoca	8387	T	T
Klebsiella oxytoca	15926	T	T
Klebsiella pneumoniae	6786	*T*	*T*
Klebsiella pneumoniae	6787	*T*	*T*
Serratia marcescens	1521	*T*	*T*

Exclusivity obtained with 20 strains using the Compact Dry EC coliform method and ISO 4832 (data from old study)

ORGANISM	CCFRA CODE	GROWTH OBSERVED	
		COMPACT DRY EC	ISO 4832 (2006)
Avibacterium avium	8389	NG	NG
Bacillus cereus	4110	NG	NG
Bacillus subtilis	4112	NG	NG
Edwardsiella tarda	8392	A	A
Pasteurella bettyae	8391	A	T
Proteus mirabilis	1588	A	A
Proteus vulgaris	1581	A	A
Pseudomonas aeruginosa	8299	NG	NG
Pseudomonas fluorescens	15937	NG	NG
Pseudomonas fragi	16050	NG	NG
Salmonella Typhimurium	11634	A	A
Shigella boydii	324	T	T
Shigella flexneri	325	A	T
Shigella sonnei	326	T	T
Shigella sonnei	4107	T	T
Staphylococcus aureus	4105	NG	NG
Staphylococcus aureus	1216	NG	NG
Vibrio mimicus	6351	NG	NG

ORGANISM	CCFRA CODE	GROWTH OBSERVED	
		COMPACT DRY EC	ISO 4832 (2006)
Vibrio parahaemolyticus	15737	A	A
Yersinia enterocolitica	4103	NG	T

Key

NG = No Growth

T = Typical Colonies

A = Atypical Growth

* = Very little or poor growth