

NF VALIDATION
Validation of alternative analytical methods
Application in food microbiology

Summary report

Validation study according to the ISO 16140-2

foodproof® Salmonella plus Cronobacter
Detection LyoKit for Cronobacter spp. detection

(Certificate number: QUA 18/13-12/24)

**in infant formula with or without probiotics, related ingredients, and
production environmental samples**

Qualitative method

> Expert Laboratory:	ADRIA ZA Creac'h Gwen 29196 Quimper Cedex (France)
> For:	HYGIENA LLC 941 Avenida Acaso Camarillo, CA 93012 (USA)

This report consists of 100 pages, including 7 appendices.

Only copies including the totality of this report are authorised.

Competencies of the laboratory are certified by COFRAC accreditation for the analyses marked with the symbol♦.

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Cancels and replaces the previous version



ADRIA > ZA Creac'h Gwen > 29000 Quimper > +33(0)2 98 10 18 18

adria.fr > adria-formationagroalimentaire.fr

Association loi de 1901 > N° existence 53290006329

N° Siret 306 964 271 00036 > N° TVA FR45306964271

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LIST OF ABBREVIATIONS

Method & protocol

BPW	Buffered Peptone Water
CFU	Colony Forming Units
ILS	Interlaboratory Study
MCS	Method Comparison Study
RLOD	Relative Level of Detection
RT	Relative Trueness
SE	Relative Sensitivity
SP	Relative Specificity
IC	Internal Control

Interpretation

AL	Acceptability Limit
alt	Alternative method
$\bar{D}$	Average difference
FN	False Negative results
FNR	False Negative Ratio
FP	False Positive results
FPR	False Positive Ratio
LOD	Limit of Detection
NA	Negative agreement
NA _{FN (alt)}	Negative Agreement due to false negative alternative-method results
ND	Negative Deviation
ND _{FN (alt)}	Negative Deviation due to false negative alternative-method results
PA	Positive Agreement
PA _{FP (alt)}	Positive Agreement due to false positive alternative-method results
PD	Positive deviation
PD _{FP (alt)}	Positive Deviation due to false positive alternative-method results
ref	Reference method
TNA	Total Negative Agreement
TND	Total Negative Deviation

Raw data

-	No typical colonies but presence of background microflora
-A	No typical colonies but presence of high level of background microflora
-B	No typical colonies but presence of medium level of background microflora
-C	No typical colonies but presence of low level of background microflora
(x)	Number of colonies in the plate
*	Dilution of the extract in case of inhibition according to the alternative protocol (1/3 or 1/5 depending on the extraction)
1/2	50% level of target analyte
d	Doubtful result
i	Inhibition
ne	New DNA extraction
NC	Non-characteristic colony
NI	No identification
ni	Not isolated colony
No amp	Non characteristic PCR curve
m	Minority level of target analyte
M	Majority level of target analyte
p	Pure culture level of target analyte
st	Plate without any colony
w	Weak reaction
Bold typing	artificially inoculated samples

Quality Assurance documents related to this study can be consulted upon request from **HYGIENA LLC**.

The technical protocol and the result interpretation were carried out according to the ISO 16140-2 (2016), ISO 16140-2/A1 (2024) and the AFNOR technical rules (Revision 12).

Validation protocols	- ISO 16140-1 (2016): Microbiology of the food chain - Method validation - <i>Part 1: Vocabulary</i> - ISO 16140-2 (2016) & ISO 16140-2/A1 (2024): Microbiology of the food chain - Method validation – <i>Part 2: Protocol for the validation of alternative (proprietary) methods against a reference method</i> - AFNOR Technical Rules (Revision 12)
Reference method[♦]	ISO 22964 (April 2017) - Microbiology of the food chain - Horizontal method for the detection of <i>Cronobacter</i> spp.
Alternative method	foodproof® <i>Salmonella</i> plus <i>Cronobacter</i> Detection LyoKit for <i>Cronobacter</i> spp. detection
Scope	☒ Infant formula with or without probiotics and related ingredients ☒ Production environmental samples
Certification organism	AFNOR certification

♦ Analyses performed according to the COFRAC accreditation (Accreditation Testing n°1-0144, scope available on www.cofrac.fr)

1 INTRODUCTION

The **foodproof®** *Salmonella* plus *Cronobacter* Detection LyoKit for *Cronobacter* spp. detection was validated in December 2024 (certificate number: QUA 18/13-12/24) in infant formula with or without probiotics, related ingredients and production environmental samples.

An extension was performed in February 2025 following a modification of the software to version 5.1. Data presented in this report were analyzed using this software version.

A new version of the report (version 1) was published in December 2025 with minor editorial corrections.

2 METHOD PROTOCOLS

2.1 Alternative method

The flow diagram of the alternative method is given in **Appendix 1**.

2.1.1 Principle

The **foodproof®** *Salmonella* plus *Cronobacter* Detection LyoKit is intended for the rapid detection of *Salmonella* spp. and *Cronobacter* spp. from Infant formula (IF) with or without probiotics and related ingredients and production environmental samples. The method is based on an enrichment step, DNA extraction and real-time PCR detection with PCR instruments with a FAM (*Salmonella* detection), HEX (*Cronobacter* detection) and a ROX (Internal Control) detection channel.

2.1.2 Protocols

Different enrichment protocols are available depending on the products tested. Detailed information is given in **Appendix 1** and an overview of the protocols is given in Table 1.

Table 1 - Overview of the enrichment protocols

Category	Type	Test portion size foodproof	Enrichment broth for the alternative method*	Dilution factor	Enrichment time	Enrichment temperature	Sample preparation
1- Infant formula with or without probiotics and related ingredients	a: Infant formula with probiotics	Up to 375 g	Pre-warmed BPW + vancomycin (10 mg/L)	1/10	16-24 h	37°C ± 1°C	ISO 6887-1 4-5 and ISO 27205
	c: Infant formula without probiotics	Up to 375 g	Pre-warmed BPW	1/10	16-24 h	37°C ± 1°C	ISO 6887-1 ISO 6887-5
	d: Related ingredients	Up to 375 g	Pre-warmed BPW	1/10	16-24 h	37°C ± 1°C	ISO 6887-1 ISO 6887-5
2- Production environmental samples	a: Surfaces	Swabs or sponges	BPW	10 ml or 100 ml	16-24 h	37°C ± 1°C	ISO 18593
	b: Vacuum dusts from non- probiotic production sites	Up to 200 g	Pre-warmed BPW	1/10	16-24 h	37°C ± 1°C	ISO 6887-1 ISO 6887-4

* Note: According to the ISO 6687 rules, for the usual 25 g test portion size for *Salmonella* screening and the usual 10 g test portion size for *Cronobacter* screening, no prewarming of the enrichment is required.

The extraction protocols are described in detail in **Appendix 1** and summarized in Table 2.

Table 2 - Extraction protocols

	StarPrep Three Kit (tube)	StarPrep Three 8-Strip Kit	BAX Lysis
Enriched sample	100 µL	100 µL	20 µL
Transfer in 8-tube strips	/		
Centrifuge	5 min at 8000 x g	10 min at 5400 x g or 25min at 2000 x g	/
Lysis buffer	200 µl	200 µl	150 µL protease + 12 mL lysis buffer: 200 µL/cluster tube
Heat treatment	10 min at 95-100°C	10 min at 95-100°C	20 minutes at 37°C 10 minutes at 95°C
Cooling	1 min at 15-25°C at room temperature	3-5 min at 15-25°C at room temperature	At least 5 minutes at 2-8°C
Mix vortex	2 sec	/	/
Centrifuge	2 min at 13000 x g	5 min at 5400 x g or 10 min at 2000 x g	/

The positive PCR results are confirmed by the tests described in the ISO method (ISO 22964 for *Cronobacter* spp. detection). In addition to using biochemical galleries, the observed characteristic colonies on the selective agar plates can be confirmed with any appropriate ISO 16140-6:2019 validated method; any alternative method based on a different measurement principle than the principle used in the detection or using different markers (e.g., different antibodies, primers) if properly validated (e.g., ISO 16140 validation) according to the ISO 7218:2024 standard.

Two thermocyclers and software are included the scope of the validation:

- The BAX Q7 from Hygiena with BAX software 5.1 or higher.
- The LightCycler 480 II (LC 480 II) from Roche Diagnostics with LightCycler 480 version: 1.5.1.62 SP3 or higher.

Results are interpreted following guidelines described in the kit IFU. While software automatically gives a Ct value in each channel, additional manual interpretation is required by the end users to finalize the analysis.

- For BAX Q7: The PCR results are provided by the BAX® software, version 5.1 or later.
- For LC 480II: if a Ct value is given by the software, it is recommended to look at the raw data and consider negative in case no exponential curve is observed (no amp).

The data should be then interpreted as follow by the end-user:

FAM	HEX	ROX	Result interpretation
Positive	Positive	Positive or Negative	Positive for <i>Salmonella</i> spp. and <i>Cronobacter</i> spp.
Positive	Negative	Positive or Negative	Positive for <i>Salmonella</i> spp. and Negative for <i>Cronobacter</i> spp.
Negative	Positive	Positive or Negative	Positive for <i>Cronobacter</i> spp. and Negative for <i>Salmonella</i> spp.
Negative	Negative	Positive	Negative for <i>Salmonella</i> spp. and <i>Cronobacter</i> spp;
Negative	Negative	Negative	Invalid

Each target is independently detected by PCR without interference from the other. In case of PCR inhibition, the DNA extract can be tested diluted 1/5 in sterile water for the StarPrep Three kits and 1/3 for BAX lysis.

It is possible to store the enriched samples for 72 h at 5°C ± 3°C.

2.1.3 Restriction

There is no restriction.

2.2 Reference method♦

The reference method used was the ISO 22964 (April 2017) - Microbiology of the food chain - Horizontal method for the detection of *Cronobacter* spp. (See **Appendix 2**):

A test portion of 10g was used during the study [(method comparison study (MCS) and inter-laboratory study (ILS)], as it has been proven than the reference method doesn't have equivalent performances with higher test portions.

For the preparation of initial suspensions, the guidelines specified in the EN ISO 6887-4:2017 standard and ISO 27205:2010 were followed for powdered infant formula containing probiotics. The following option were chosen during the different part of the study to mitigate pH decrease and competition from probiotic strains:

- During MCS, double-strength Buffered Peptone Water (BPW) was used.
- For ILS, BPW with antibiotic (vancomycin (10 mg/L)) was tested.

♦ Analyses performed according to the COFRAC accreditation (Accreditation Testing n°1-0144, scope available on www.cofrac.fr)

2.3 Study design

During the MCS, different test portions including large sample sizes were tested except for surface samples. The study was therefore performed using an unpaired study design for all samples except surfaces samples (paired).

During the ILS, the study follows a paired design, with both the reference and alternative methods tested using a 10 g test portion enriched in BPW supplemented with vancomycin (10 mg/L) to ensure consistency across testing conditions.

3 METHOD COMPARISON STUDY

The method comparison study is a study performed by the expert laboratory to compare the alternative method with the reference method.

The study was carried out on a diversity of samples and strains representative of agri-food products. This does not constitute an exhaustive list of the different matrices included in the scope.

For any comment on the alternative method, please contact AFNOR Certification at <http://nf-validation.afnor.org/contact-2/>.

3.1 Sensitivity study

The sensitivity (SE) is the ability of the method to detect the analyte by either the reference or alternative method.

3.1.1 Number and nature of samples

137 samples were analysed. The distribution per tested category and type is given in the following table. For all the tested extraction protocols (foodproof and BAX) and thermocycler (BAXQ7 and Lightcycler 480 II), the same repartition is obtained with 70 positive results and 67 negative results.

Table 3 – Distribution per tested category and type - All protocols

Category		Type		Positive samples	Negative samples	Total
1	Infant formula (IF) with or without probiotics and related ingredients	a	IF with probiotics	14	12	26
		b	IF without probiotics	14	8	22
		c	Related ingredients	12	10	22
		Total		40	30	70
2	Production environmental samples	a	Surfaces	15	15	30
		b	Vacuum dusts from non-probiotic production sites	15	22	37
		Total		30	37	67
All categories				70	67	137

3.1.2 Artificial contamination of samples

Due to the type of product evaluated, no natural contamination was found. Artificial contaminations were done by seeding with lyophilized strains or spiking protocol after heat treatment. The artificial contaminations are presented in **Appendix 3**.

78 samples were artificially contaminated, using 24 different strains. 70 gave a positive result. Among those positives, 24.3 % of samples were spiked after heat treatment at level ≤ 5 CFU and 71.4 % were inoculated by seeding at level ≤ 3 CFU. The repartition of the positive samples per inoculation protocol and inoculation level is given in Table 4.

Table 4 - Repartition of the positive samples per inoculation protocol and inoculation level

Category	Naturally contaminated	Spiking protocol		Seeding protocol		Total positive samples
		≤ 5 CFU	$5 < x \leq 10$ CFU	≤ 3 CFU	$3 < x \leq 5.6$ CFU	
1	0	5	0	32	3	40
2	0	12	0	18	0	30
Number of samples	0	17	0	50	3	70
%	0.0	24.3	0.0	71.4	4.3	100.0

Among those positive results, 42 positives results came from *Cronobacter* spp. strains inoculated alone and 28 for *Cronobacter* spp. strains co-inoculated with *Salmonella* spp. The repartition of the inoculation scheme per category is given in Table 5.

**Table 5 - Repartition of the positive samples
per inoculation protocol and inoculation level**

Category	<i>Cronobacter</i> spp	<i>Cronobacter</i> spp + <i>Salmonella</i> spp	Total positive samples
	Number of samples	Number of samples	
1	23	17	40
2	19	11	30
Total	42	28	70

0 % of the samples were naturally contaminated. This low percentage is linked to the categories tested with a low prevalence of *Cronobacter* spp. (heat treated products).

3.1.3 *Protocols applied during the validation study*

> Enrichment step

Higher test portion with the minimum incubation time was applied for the enrichment step: 16 h at 37°C.

> Lactic bacteria enumeration

For the category 1, for probiotics containing products, lactic bacteria enumeration was carried out using MRS pH 5.7 incubated in anaerobic conditions for 72 h at 30°C ± 1°C (ISO 15214).

> Extraction

Two types of extraction were evaluated on each sample:

- foodproof® StarPrep: Three Kit or Three 8-Strip Kit with each sample tested by one of the kit formats. The 10 min 5400 g centrifugation protocol was used during the validation.
- BAX Real-Time Lysis procedure for all sample tested.

The repartition of the samples tested with each protocol is shown in the table below.

Table 6 - Distribution of the number of samples per tested category and extraction protocols

Category	StarPrep Three Kit	StarPrep Three 8-Strip Kit	BAX Lysis	Total number of samples
	Number of samples	Number of samples	Number of samples	
1	36	34	70	70
2	34	33	67	67
Total	70 (51.1%)	67 (48.9%)	137 (100%)	137

> **PCR**

Two thermocyclers were included in the validation:

- The BAX Q7 from Hygiena with BAX software 5.1
- The LightCycler 480 II (LC 480 II) from Roche Diagnostics with Lightcycler 480 version: 1.5.1.62 SP3.

Analyses of the PCR results were carried out following guidelines given in the IFU.

> **Confirmations**

The positive PCR tests were confirmed by the tests described in the ISO methods.

- Subculture of the enriched sample into CSB broth (0.1 ml + 10 ml) incubated for 24 h $\pm$ 2 h at 41.5°C $\pm$ 1°C.
- Streaking (10µL) onto CCI agar incubated for 24 h $\pm$ 2 h at 41.5°C $\pm$ 1°C.
- Subculture of typical colonies onto one selective agar media incubated for 18-24 h at 37.0°C $\pm$ 1°C.
- Confirmation by oxidase test and biochemical galleries (API 32E).

> **Enrichment broths storage for 72 h at 5 °C $\pm$ 3 °C**

The enrichment broths from positive and discordant samples were stored for 72 h at 5°C $\pm$ 3 °C and were tested again (PCR and confirmatory tests).

> **Inhibition**

In case of inhibition, the DNA extracts were tested after 1/5 or 1/3 dilution in sterile water as described in the IFU.

3.1.4 Test results

Raw data per category are given in **Appendix 4**. The results are given in the following tables.

Table 7 – Summary of results obtained with the reference and alternative methods (after confirmation) of all samples for each category - foodproof® Extraction - BAX Q7

Categories		Portion	Types		Study Design	PA	PD	TND	TNA
1	Infant formula (IF) with or without probiotics and related ingredients	375g	a	IF with probiotics	Unpaired	8	6	1	11
		375g	b	IF without probiotics	Unpaired	14	0	0	8
		375g	c	Related ingredients	Unpaired	9	3	0	10
		Total				31	9	1	29
2	Production environmental samples	Swab/ sponge/ wipe	a	Surfaces	Paired	15	0	0	15
		200g	b	Vacuum dusts from non - probiotic production sites	Unpaired	10	3	1	23
		Total				25	3	1	38
All categories						56	12	2	67

Table 8 – Summary of results obtained with the reference and alternative methods (after confirmation) of all samples for each category - foodproof® Extraction - LightCycler 480 II

Categories		Portion	Types		Study Design	PA	PD	TND	TNA
1	Infant formula (IF) with or without probiotics and related ingredients	375g	a	IF with probiotics	Unpaired	8	6	1	11
		375g	b	IF without probiotics	Unpaired	14	0	0	8
		375g	c	Related ingredients	Unpaired	9	3	0	10
		Total				31	9	1	29
2	Production environmental samples	Swab/ sponge/ wipe	a	Surfaces	Paired	15	0	0	15
		200g	b	Vacuum dusts from non-probiotic production sites	Unpaired	8	3	3	23
		Total				23	3	3	38
All categories						54	12	4	67

Table 9 – Summary of results obtained with the reference and alternative methods (after confirmation) of all samples for each category - BAX lysis - BAX Q7

Categories		Portion	Types		Study Design	PA	PD	TND	TNA
1	Infant formula (IF) with or without probiotics and related ingredients	375g	a	IF with probiotics	Unpaired	8	6	1	11
		375g	b	IF without probiotics	Unpaired	14	0	0	8
		375g	c	Related ingredients	Unpaired	9	3	0	10
		Total				31	9	1	29
2	Production environmental samples	Swab/ sponge/ wipe	a	Surfaces	Paired	15	0	0	15
		200g	b	Vacuum dusts from non-probiotic production sites	Unpaired	8	3	3	23
		Total				23	3	3	38
All categories						54	12	4	67

Table 10 – Summary of results obtained with the reference and alternative methods (after confirmation) of all samples for each category - BAX lysis - LightCycler 480 II

Categories		Portion	Types		Study Design	PA	PD	TND	TNA
1	Infant formula (IF) with or without probiotics and related ingredients	375g	a	IF with probiotics	Unpaired	8	6	1	11
		375g	b	IF without probiotics	Unpaired	14	0	0	8
		375g	c	Related ingredients	Unpaired	9	3	0	10
		Total				31	9	1	29
2	Production environmental samples	Swab/ sponge/ wipe	a	Surfaces	Paired	15	0	0	15
		200g	b	Vacuum dusts from non-probiotic production sites	Unpaired	8	3	3	23
		Total				23	3	3	38
All categories						54	12	4	67

Paired evaluation:

$$TND = ND_{FN(alt)}$$

$$TNA = NA + PD_{FP(alt)}$$

Unpaired evaluation:

$$TND = ND + ND_{FN(alt)} + PA_{FP(alt)}$$

$$TNA = NA + NA_{FN(alt)} + PD_{FP(alt)}$$

3.1.5 Calculation of relative trueness (RT), sensitivity (SE) and false positive ratio (FPR) and false negative ratio (FNR) for the alternative method

The calculations are presented in the following tables.

A summary of the results is given in Table 15.

Table 11 – Calculation of relative trueness (RT), sensitivity (SE), false positive ratio (FPR) and false negative ratio (FNR) for the alternative method - foodproof® Extraction - BAX Q7

Categories		Portion	Types		Study Design	PA	PA _{FP(alt)}	NA	NA _{FN(alt)}	PD	ND	ND _{FN(alt)}	PD _{FP(alt)}	TND	TNA	SE _% ^{alt}	SE _% ^{ref}	RT _%	FPR _%	FNR _%
1	Infant formula (IF) with or without probiotics and related ingredients	375g	a	IF with probiotics	Unpaired	8	1	10	0	6	0	0	1	1	11	93.3	60.0	73.1	18.2	0.0
		375g	b	IF without probiotics	Unpaired	14	0	6	0	0	0	0	2	0	8	100.0	100.0	100.0	25.0	0.0
		375g	c	Related ingredients	Unpaired	9	0	10	0	3	0	0	0	0	10	100.0	75.0	86.4	0.0	0.0
		Total				31	1	26	0	9	0	0	3	1	29	97.6	78.0	85.7	13.8	0.0
2	Production environmental samples	Swab/ sponge/ wipe	a	Surfaces	Paired	15	0	15	0	0	0	0	0	0	15	100.0	100.0	100.0	0.0	0.0
		200g	b	Vacuum dusts from non-probiotic production sites	Unpaired	10	0	21	0	3	1	0	2	1	23	92.9	78.6	89.2	8.7	0.0
		Total				25	0	36	0	3	1	0	2	1	38	96.6	89.7	94.0	5.3	0.0
All categories						56	1	62	0	12	1	0	5	2	67	97.1	82.9	89.8	9.0	0.0

Table 12 – Calculation of relative trueness (RT), sensitivity (SE), false positive ratio (FPR) and false negative ratio (FNR) for the alternative method - foodproof® Extraction - LightCycler 480 II from Roche Diagnostics

Categories		Portion	Types		Study Design	PA	PA _{FP(alt)}	NA	NA _{FN(alt)}	PD	ND	ND _{FN(alt)}	PD _{FP(alt)}	TND	TNA	SE _{alt} %	SE _{ref} %	RT %	FPR %	FNR %
1	Infant formula (IF) with or without probiotics and related ingredients	375g	a	IF with probiotics	Unpaired	8	1	10	0	6	0	0	1	1	11	93.3	60.0	73.1	18.2	0.0
		375g	b	IF without probiotics	Unpaired	14	0	6	0	0	0	0	2	0	8	100.0	100.0	100.0	25.0	0.0
		375g	c	Related ingredients	Unpaired	9	0	10	0	3	0	0	0	0	10	100.0	75.0	86.4	0.0	0.0
		Total				31	1	26	0	9	0	0	3	1	29	97.6	78.0	85.7	13.8	0.0
2	Production environmental samples	Swab/ sponge/ wipe	a	Surfaces	Paired	15	0	15	0	0	0	0	0	0	15	100.0	100.0	100.0	0.0	0.0
		200g	b	Vacuum dusts from non-probiotic production sites	Unpaired	8	0	21	0	3	1	2	2	3	23	78.6	78.6	83.8	8.7	14.3
		Total				23	0	36	0	3	1	2	2	3	38	89.7	89.7	91.0	5.3	6.9
All categories						54	1	62	0	12	1	2	5	4	67	94.3	82.9	88.3	9.0	2.9

Table 13 – Calculation of relative trueness (RT), sensitivity (SE), false positive ratio (FPR) and false negative ratio (FNR) for the alternative method - BAX Real-Time lysis procedure - **BAX Q7**

Categories		Portion	Types		Study Design	PA	PA _{FP(alt)}	NA	NA _{FN(alt)}	PD	ND	ND _{FN(alt)}	PD _{FP(alt)}	TND	TNA	SE % ^{alt}	SE % ^{ref}	RT %	FPR %	FNR %
1	Infant formula (IF) with or without probiotics and related ingredients	375g	a	IF with probiotics	Unpaired	8	1	10	0	6	0	0	1	1	11	93.3	60.0	73.1	18.2	0.0
		375g	b	IF without probiotics	Unpaired	14	0	6	0	0	0	0	2	0	8	100.0	100.0	100.0	25.0	0.0
		375g	c	Related ingredients	Unpaired	9	0	10	0	3	0	0	0	0	10	100.0	75.0	86.4	0.0	0.0
		Total				31	1	26	0	9	0	0	3	1	29	97.1	78.0	85.7	13.8	0.0
2	Production environmental samples	Swab/ sponge/ wipe	a	Surfaces	Paired	15	0	15	0	0	0	0	0	0	15	100.0	100.0	100.0	0.0	0.0
		200g	b	Vacuum dusts from non-probiotic production sites	Unpaired	9	0	22	0	3	1	1	1	2	23	85.7	78.6	86.5	4.3	7.1
		Total				24	0	37	0	3	1	1	1	2	38	93.1	89.7	92.5	2.6	3.4
All categories						55	1	63	0	12	1	1	4	3	67	95.7	82.9	89.1	7.5	1.4

Table 14 – Calculation of relative trueness (RT), sensitivity (SE), false positive ratio (FPR) and false negative ratio (FNR) for the alternative method - BAX Real-Time lysis procedure - **LightCycler 480 II**

Categories		Portion	Types		Study Design	PA	PA _{FP(alt)}	NA	NA _{FN(alt)}	PD	ND	ND _{FN(alt)}	PD _{FP(alt)}	TND	TNA	SE _% ^{alt}	SE _% ^{ref}	RT %	FPR %	FNR
1	Infant formula (IF) with or without probiotics and related ingredients	375g	a	IF with probiotics	Unpaired	8	1	11	0	6	0	0	0	1	11	93.3	60.0	73.1	9.1	0.0
		375g	b	IF without probiotics	Unpaired	14	0	7	0	0	0	0	1	0	8	100.0	100.0	100.0	12.5	0.0
		375g	c	Related ingredients	Unpaired	9	0	10	0	3	0	0	0	0	10	100.0	75.0	86.4	0.0	0.0
		Total				31	1	28	0	9	0	0	1	1	29	97.6	78.0	85.7	6.9	0.0
2	Production environmental samples	Swab/ sponge/ wipe	a	Surfaces	Paired	15	0	15	0	0	0	0	0	0	15	100.0	100.0	100.0	0.0	0.0
		200g	b	Vacuum dusts from non-probiotic production sites	Unpaired	8	0	23	0	3	1	2	0	3	23	78.6	78.6	83.8	0.0	14.3
		Total				23	0	38	0	3	1	2	0	3	38	89.7	89.7	91.0	0.0	6.9
All categories						54	1	66	0	12	1	2	1	4	67	94.3	82.9	88.3	3.0	2.9

Table 15 - Summary of results

		foodproof® Extraction		BAX lysis	
		BAX Q7	LC 480 II	BAX Q7	LC 480 II
Sensitivity for the alternative method	$SE_{alt} = \frac{(PA + PD)}{(PA + TND + PD)} \times 100 \%$	97.1 %	94.3 %	95.7 %	94.3 %
Sensitivity for the reference method	$SE_{ref} = \frac{(PA + TND)}{(PA + TND + PD)} \times 100 \%$	82.9 %	82.9 %	82.9 %	82.9 %
Relative trueness	$RT = \frac{(PA + TNA)}{N} \times 100 \%$	89.8 %	88.3 %	89.1 %	88.3 %
False positive ratio for the alternative method	$FPR = \frac{PA_{FP(alt)} + PD_{FP(alt)}}{TNA} \times 100 \%$	9.0 %	9.0 %	7.5 %	3.0 %
False negative ratio for the alternative method	$FNR = \frac{NA_{FN(alt)} + ND_{FN(alt)}}{PA + TND + PD} \times 100 \%$	0.0 %	2.9 %	1.4 %	2.9 %

3.1.6 Analysis of discordant results

The negative deviations are given in Table 16 and the positive deviations in Table 17.

> **Negative deviations**

The number of negative deviations varies from 2 to 4 depending on the protocols evaluated and concerns 5 samples from the vacuum dust environmental samples type. The presence of *Cronobacter* spp was confirmed in the enrichment broth for 3 samples ($ND_{FN(alt)} = 3$): 3741; 561; 1364. For two false negative samples, the PCR were found positive with the BAX Q7 and negative in LightCycler 480 II. Several PCR tests were performed on negative deviation samples but only one showed positive PCR results on one of the replicates.

The presence of *Cronobacter* spp has not been confirmed in any of the samples in negative agreement (NA).

> **Positive deviations**

Twelve positive deviations were observed with similar results obtained for all protocols evaluated. Nine positive deviations concern category 1 while 3 concerns category 2.

The analyses of discordant results according to the ISO 16140-2 for all the categories is the following (See Tables 18 to 21).

Table 16 - Negative deviations

Sample N°	Product	Strain inoculated	Inoculation level CFU/test portion	ISO 22964*	foodproof® Salmonella Plus Cronobacter Detection LyoKit - Pre-warmed BPW - 16h-37°C																	Confirmation	Agreement				Category	Type
					PCR result																							
					foodproof® extraction						BAX lysis						Foodproof extraction		BAX Lysis									
					BAX Q7			LC480 II			BAX Q7			LC480 II			BAX Q7	LC 480 II	BAX Q7	LC 480 II								
					Ct HEX (target)	Ct ROX (IC)	Result	Ct HEX (target)	Ct ROX (IC)	Result	Ct HEX (target)	Ct ROX (IC)	Result	Ct HEX (target)	Ct ROX (IC)	Result												
3741	Vacuum cleaner dust P51 2889 A12 (dairy industry)	C. sakazakii Ad2407	1.5	+	36.0	36.7	+	33.21	32.77	+	0.0 0.0 0.0	37.9 38.5 38.2	-/-/-	35.49	32.69	+	+	PA	PA	ND _{FN(alt)}	PA	2	b					
561	Vacuum cleaner dust n°13-14 (non-probiotic dairy industry)	C. sakazakii Ad2386 (+ S. Livingstone A00E058)	1.4/1.0	+	39.3	37.6	+	0.0 0.0 0.0	0.0 0.0 33.32	-/-/-	37.0	36.8	+	0.0 0.0 0.0	32.2 32.27 33.34	-/-/-	+	PA	ND _{FN(alt)}	PA	ND _{FN(alt)}	2	b					
1363	Vacuum cleaner dust 24 (dairy industry)	C. sakazakii Ad2408 (+ S. Brandenburg Ad2151)	1.2/1.8	+	0.0	37.0	-	0	33.11	-	0.0	38.0	-	0	33.02	-	-	ND	ND	ND	ND	2	b					
1364	Vacuum cleaner dust 22 (dairy industry)	C. sakazakii Ad2408 (+ S. Brandenburg Ad2151)	1.2/1.8	+	37.2	37.6	+	0.0 0.0 0.0	31.22 30.44 29.65	-/-/-	36.9	37.7	+	0.0 35.45 0.0	35.65 33.92 0.0	-/+/-	+	PA	ND _{FN(alt)}	PA	ND _{FN(alt)}	2	b					
1371	Vacuum cleaner dust 27 (dairy industry)	C. sakazakii Ad2391	1.8	+	36.3 0.0 33.5	36.0 38.1 36.6	+/-/+	31.88 31.89 32.24	32.01 31.83 31.96	+/+/+	35.5 33.5 33.6	37.0 36.6 36.3	+/+/+	33.33 34.57 33.78	32.65 32.41 32.23	+/+/+	-	PA _{FP(alt)}	PA _{FP(alt)}	PA _{FP(alt)}	PA _{FP(alt)}	2	b					

♦ Analyses performed according to the COFRAC accreditation (Accreditation Testing n°1-0144, scope available on www.cofrac.fr)

Table 17 - Positive deviations

Sample N°	Product	Strain inoculated	Inoculation level CFU/test portion	ISO 22964♦	foodproof® Salmonella Plus Cronobacter Detection LyoKit - Pre-warmed BPW or Pre-warmed BPW + vancomycin (10mg/L) - 16h-37°C																Category	Type	
					PCR result												Confirmation	Agreement					
					foodproof® extraction						BAX lysis							Foodproof extraction		BAX Lysis			
					BAX Q7			LC480			BAX Q7			LC480 II				BAX Q7	LC 480 II	BAX Q7			LC 480 II
					Ct HEX (target)	Ct ROX (IC)	Result <i>Crono</i> spp	Ct HEX (target)	Ct ROX (IC)	Result <i>Crono</i> spp	Ct HEX (target)	Ct ROX (IC)	Result <i>Crono</i> spp	Ct HEX (target)	Ct ROX (IC)	Result <i>Crono</i> spp							
3567	Infant milk powder. 6-12 months. with probiotics (B. infantis 3.2.10 ⁷ CFU/g) (23%MG)	C. sakazakii 95	1.8	-	22.9	0.0	+	20.6	0	+	23.7	0.0	+	19.35	0	+	+	PD	PD	PD	PD	1	a
3797	Infant milk powder. 3rd age. with probiotics (B. lactis + S. thermophilus 1.5.10 ⁶ CFU/g) (30% fat)	C. sakazakii Ad935 (+ S. Ohio Ad2213)	0.3/2.4	-	22.0	0.0	+	18.31	0.0	+	26.1	0.0	+	21.72	0	+	+	PD	PD	PD	PD	1	a
763	Infant milk powder with probiotics. 3rd age (L. reuteuri DSM 17938 8.55.104 CFU/g) (26% fat)	C. malonaticus DSM18702	1.8	-	24.4	0.0	+	22.27	0.0	+	27.4	0.0	+	23.49	0	+	+	PD	PD	PD	PD	1	a
765	Infant milk powder with probiotics. after from 10 months (L. reuteuri DSM 17938 1.55.10 ⁵ CFU/g) (23% fat)	C. sakazakii Ad898	1.6	-	34.1	36.1	+	31.38	32.42	+	35.6	37.6	+	32.06	32.82	+	+	PD	PD	PD	PD	1	a
769	Infant milk powder with probiotics. organic from 6 months (L. reuteuri DSM 17938 2.45.10 ⁶ CFU/g) (26% fat)	C. malonaticus DSM18702	1.8	-	30.3	0.0	+	29.67	31.55	+	29.6	35.3	+	26.02	33.19	+	+	PD	PD	PD	PD	1	a
771	Infant milk powder with probiotics. 1st age for infant 0-6 months (L. fermentum hereditum 5.09.10 ⁶ CFU/g) (24% fat)	C. sakazakii Ad898	1.6	-	33.6	36.2	+	29.72	30.12	+	32.7	37.7	+	30.04	32.41	+	+	PD	PD	PD	PD	1	a
3089	Rice starch	C. sakazakii Ad2341	<0.5	-	23.0	0.0	+	19.02	0	+	22.8	0.0	+	21.13	0	+	+	PD	PD	PD	PD	1	c
3090	Corn starch	C. sakazakii Ad2341	<0.5	-	33.1	34.7	+	30.03	31.62	+	32.8	37.0	+	29.79	32.36	+	+	PD	PD	PD	PD	1	c
3094	Maltodextrin	C. sakazakii Ad2341	<0.5	-	21.1	0.0	+	18.87	19.00 (no amp)	+	16.8	0.0	+	14.99	0	+	-	PD	PD	PD	PD	1	c
3740	Vacuum cleaner dust A32 E553 A31 (dairy industry)	C. sakazakii Ad2407	1.5	-	38.4	37.1	+	33.01	32.79	+	37.1	37.3	+	33.79	32.36	+	+	PD	PD	PD	PD	2	b
3742	Vacuum cleaner dust P51 E2E2 (dairy industry)	C. sakazakii Ad2382	0.5	-	29.4	0.0	+	25.09	0	+	28.1	0.0	+	24.72	29.48 (no amp)	+	+	PD	PD	PD	PD	2	b
560	Vacuum cleaner dust n°11-12 (non-probiotic dairy industry)	C. sakazakii Ad2360 (+ S. Anatum A00E007)	1.6/0.2	-	18.1	0.0	+	16.54	0.0	+	22.0	0.0	+	18.71	0	+	+	PD	PD	PD	PD	2	b

♦ Analyses performed according to the COFRAC accreditation (Accreditation Testing n°1-0144, scope available on www.cofrac.fr)

Table 18 - Analyses of discordant results - foodproof® Extraction - BAX Q7

								Paired				Unpaired		Mixed		
Categories		Protocol	Types		Design	N+	TND	PD	TND-PD	AL	TND+PD	AL	TND-PD	AL	TND-PD	AL
1	Infant formula (IF) with or without probiotics and related ingredients	375g	a	IF with probiotics (L. johnsonii, S. thermophilus and each species of the genus Bifidobacterium)	Unpaired	15	1	6					-5		-5	
		375g	b	IF without probiotics	Unpaired	14	0	0					0		0	
		375g	c	Related ingredients	Unpaired	12	0	3					-3		-3	
		Total			/	41	1	9					-8	3	-8	3
2	Production environmental samples	Swab/sponge/wipe	a	Surfaces	Paired	15	0	0	0		0				0	
		200g	b	Vacuum dusts from non-probiotic production sites	Unpaired	14	1	3					-2		-2	
		Total			/	29	1	3	0	3	0	6	-2	3	-2	3
All categories					/	70	2	12	/	/	/	/	/	/	-10	4

Table 19 - Analyses of discordant results - foodproof® Extraction - LightCycler 480 II

								Paired				Unpaired		Mixed		
Categories		Protocol	Types		Design	N+	TND	PD	TND-PD	AL	TND+PD	AL	TND-PD	AL	TND-PD	AL
1	Infant formula (IF) with or without probiotics and related ingredients	375g	a	IF with probiotics (L. johnsonii, S. thermophilus and each species of the genus Bifidobacterium)	Unpaired	15	1	6					-5		-5	
		375g	b	IF without probiotics	Unpaired	14	0	0					0		0	
		375g	c	Related ingredients	Unpaired	12	0	3					-3		-3	
		Total			/	41	1	9					-8	3	-8	3
2	Production environmental samples	Swab/sponge/wipe	a	Surfaces	Paired	15	0	0	0		0				0	
		200g	b	Vacuum dusts from non-probiotic production sites	Unpaired	14	3	3					0		0	
		Total			/	29	3	3	0	3	0	6	0	3	0	3
All categories					/	70	4	12	/	/	/	/	/	/	-8	4

Table 20 - Analyses of discordant results - BAX lysis - BAX Q7

								Paired				Unpaired		Mixed		
Categories		Protocol	Types		Design	N+	TND	PD	TND-PD	AL	TND+PD	AL	TND-PD	AL	TND-PD	AL
1	Infant formula (IF) with or without probiotics and related ingredients	375g	a	IF with probiotics (L. johnsonii, S. thermophilus and each species of the genus Bifidobacterium)	Unpaired	15	1	6					-5		-5	
		375g	b	IF without probiotics	Unpaired	14	0	0					0		0	
		375g	c	Related ingredients	Unpaired	12	0	3					-3		-3	
		Total			/	41	1	9					-8	3	-8	3
2	Production environmental samples	Swab/sponge/wipe	a	Surfaces	Paired	15	0	0	0		0				0	
		200g	b	Vacuum dusts from no - probiotic production sites	Unpaired	14	2	3					-1		-1	
		Total			/	29	2	3	0	3	0	6	-1	3	-1	3
All categories					/	70	3	12	/	/	/	/	/	/	-9	4

Table 21 - Analyses of discordant results - BAX Real-Time lysis procedure - LightCycler 480 II

								Paired				Unpaired		Mixed		
Categories		Protocol	Types		Design	N+	TND	PD	TND-PD	AL	TND+PD	AL	TND-PD	AL	TND-PD	AL
1	Infant formula (IF) with or without probiotics and related ingredients	375g	a	IF with probiotics (L. johnsonii, S. thermophilus and each species of the genus Bifidobacterium)	Unpaired	15	1	6					-5		-5	
		375g	b	IF without probiotics	Unpaired	14	0	0					0		0	
		375g	c	Related ingredients	Unpaired	12	0	3					-3		-3	
		Total			/	41	1	9					-8	3	-8	3
2	Production environmental samples	Swab/sponge/wipe	a	Surfaces	Paired	15	0	0	0		0				0	
		200g	b	Vacuum dusts from non-probiotic production sites	Unpaired	14	3	3					0		0	
		Total			/	29	3	3	0	3	0	6	0	3	0	3
All categories					/	70	4	12	/	/	/	/	/	/	-8	4

The observed values for TND - PD meet the acceptability limit for each individual category and for the two combined categories (calculated values $\leq$ AL) whatever the extraction kits and thermocycler used.

3.1.7 Enrichment broth storage at 5 ± 3 °C for 72 h

Modification was observed only for one sample (561: vacuum cleaner dust).

Table 22 - Enrichment broth storage

Sample N°	Product	foodproof® Salmonella Plus Cronobacter Detection LyoKit								Category	Type
		Before storage				After 72 h storage					
		Agreement				Agreement					
		Foodproof extraction		BAX lysis		foodproof extraction		BAX lysis			
		BAX Q7	LC 480 II	BAX Q7	LC 480 II	BAX Q7	LC 480 II	BAX Q7	LC 480 II		
561	Vacuum cleaner dust	PA	ND _{FN(alt)}	PA	ND _{FN(alt)}	ND _{FN(alt)}	PA	ND _{FN(alt)}	ND _{FN(alt)}	2	b

The analyses of discordant results become (See Tables 23 to 26).

Table 23 - Analyses of discordant results after storage 72 h at 5 ± 3°C - foodproof® Extraction - BAX Q7

									Paired				Unpaired		Mixed	
Categories		Protocol	Types		Design	N+	TND	PD	TND-PD	AL	TND+PD	AL	TND-PD	AL	TND-PD	AL
1	Infant formula (IF) with or without probiotics and related ingredients	375g	a	IF with probiotics (L. johnsonii, S. thermophilus and each species of the genus Bifidobacterium)	Unpaired	15	1	6					-5		-5	
		375g	b	IF without probiotics	Unpaired	14	0	0					0		0	
		375g	c	Related ingredients	Unpaired	12	0	3					-3		-3	
		Total			/	41	1	9					-8	3	-8	3
2	Production environmental samples	Swab/sponge/wipe	a	Surfaces	Paired	15	0	0	0		0				0	
		200g	b	Vacuum dusts from non-probiotic production sites	Unpaired	14	2	3					-1		-1	
		Total			/	29	2	3	0	3	0	6	-1	3	-1	3
All categories					/	70	3	12	/	/	/	/	/	/	-9	4

Table 24 - Analyses of discordant results after storage 72 h at 5 ± 3°C - foodproof® Extraction - LightCycler 480 II

									Paired				Unpaired		Mixed	
Categories		Protocol	Types		Design	N+	TND	PD	TND-PD	AL	TND+PD	AL	TND-PD	AL	TND-PD	AL
1	Infant formula (IF) with or without probiotics and related ingredients	375g	a	IF with probiotics (L. johnsonii, S. thermophilus and each species of the genus Bifidobacterium)	Unpaired	15	1	6					-5		-5	
		375g	b	IF without probiotics	Unpaired	14	0	0					0		0	
		375g	c	Related ingredients	Unpaired	12	0	3					-3		-3	
		Total			/	41	1	9					-8	3	-8	3
2	Production environmental samples	Swab/sponge/wipe	a	Surfaces	Paired	15	0	0	0		0				0	
		200g	b	Vacuum dusts from non-probiotic production sites	Unpaired	14	2	3					-1		-1	
		Total			/	29	2	3	0	3	0	6	-1	3	-1	3
All categories					/	70	3	12	/	/	/	/	/	/	-9	4

Table 25 - Analyses of discordant results after storage 72 h at 5 ± 3°C - BAX lysis- **BAX Q7**

									Paired				Unpaired		Mixed	
Categories		Protocol	Types		Design	N+	TND	PD	TND-PD	AL	TND+PD	AL	TND-PD	AL	TND-PD	AL
1	Infant formula (IF) with or without probiotics and related ingredients	375g	a	IF with probiotics (L. johnsonii, S. thermophilus and each species of the genus Bifidobacterium)	Unpaired	15	1	6					-5		-5	
		375g	b	IF without probiotics	Unpaired	14	0	0					0		0	
		375g	c	Related ingredients	Unpaired	12	0	3					-3		-3	
		Total			/	41	1	9					-8	3	-8	3
2	Production environmental samples	Swab/sponge/wipe	a	Surfaces	Paired	15	0	0	0		0				0	
		200g	b	Vacuum dusts from non-probiotic production sites	Unpaired	14	3	3					0		0	
		Total			/	29	3	3	0	3	0	6	0	3	0	3
All categories					/	70	4	12	/	/	/	/	/	/	-8	4

Table 26 - Analyses of discordant results after storage 72 h at 5 ± 3°C - BAX lysis - **LightCycler 480 II**

									Paired				Unpaired		Mixed	
Categories		Protocol	Types		Design	N+	TND	PD	TND-PD	AL	TND+PD	AL	TND-PD	AL	TND-PD	AL
1	Infant formula (IF) with or without probiotics and related ingredients	375g	a	IF with probiotics (L. johnsonii, S. thermophilus and each species of the genus Bifidobacterium)	Unpaired	15	1	6					-5		-5	
		375g	b	IF without probiotics	Unpaired	14	0	0					0		0	
		375g	c	Related ingredients	Unpaired	12	0	3					-3		-3	
		Total			/	41	1	9					-8	3	-8	3
2	Production environmental samples	Swab/sponge/wipe	a	Surfaces	Paired	15	0	0	0		0				0	
		200g	b	Vacuum dusts from non-probiotic production sites	Unpaired	14	3	3					0		0	
		Total			/	29	3	3	0	3	0	6	0	3	0	3
All categories					/	70	4	12	/	/	/	/	/	/	-8	4

The observed values for TND - PD meet the acceptability limit for each individual category and for the two combined categories (calculated values $\leq$ AL) whatever the extraction kits and thermocycler used. It is possible to store the enrichment broth for 72h at $5 \pm 3^{\circ}\text{C}$.

3.1.8 Confirmation

Ten samples with positive PCR results in at least one of the protocols evaluated were not confirmed by cultural method. Seven samples came from infant formula and three from dust samples. For all these results, replicates of the PCR test were carried on.

For 9 samples, the final results were in accordance with the reference method (PPNA). For those, variation of the PCR results was observed between protocols and between the different PCR replicates, with in most cases a negative result in the HEX channel when the PCR was repeated.

5 replicates of the confirmation were carried on for two of the samples, but the results remained negative. It is possible that the presence of free DNA in the sample could be the cause of these false positive results.

One inoculated sample was in discordance with the reference method (PPND). For this sample, all PCR results were positive. This result could be explained by a difficulty to confirm the presence of the strain by cultural method or an important presence of free DNA in the sample.

Table 27 – Positive PCR results not confirmed by cultural methods

Sample N°	Product	Strain inoculated	Inoculation level CFU/test portion	ISO 22964*	foodproof® Salmonella Plus Cronobacter Detection LyoKit - Pre-warmed BPW or Prewarmed BPW + vancomycin (10mg/L) or BPW- 16h-37°C																Confirmed positive for Salmonella spp	Category	Type	
					PCR result												Confirmation	Agreement						
					foodproof® extraction						BAX lysis							foodproof extraction		BAX Lysis				
					BAX Q7			LC480 CC			BAX Q7			LC480 II				BAX Q7	LC 480 II	BAX Q7				LC 480 II
					Ct HEX (target)	Ct ROX (IC)	Result Crono spp	Ct HEX (target)	Ct ROX (IC)	Result Crono spp	Ct HEX (target)	Ct ROX (IC)	Result Crono spp	Ct HEX (target)	Ct ROX (IC)	Result Crono spp								
766	Infant milk powder with probiotics. 3rd age (L. reutei DSM 17938 8.24 x 10 ⁴ CFU/g) (23.4% fat)	/	/	-	36.8 0.0 0.0	37.3 38.1 38.5	+/-/-	33.66 0.0 32.25	31.82 32.03 32.02	+/-/+	0.0	38.1	-	0	32.73	-	-	PD _{FP(alt)}	PD _{FP(alt)}	NA	NA	+	1	a
3564	Infant milk powder. from 6 months. with probiotics (B. lactis 3.5 x 10 ⁷ CFU/g) (26% fat)	/	/	-	0.0	37.9	-	35.06 0.0 0.0 0.0*	33.20/ 0.0/ 0.0/ 0	+/-/i/-*	39.6 0.0 0.0	37.3 43.7 42.5	+/-/-	0.0	37.04	-	-	NA	PD _{FP(alt)}	PD _{FP(alt)}	NA	+	1	a
3568	Infant milk powder. 6-12 months. with probiotics (B. lactis 1.8. x 10 ⁶ CFU/g) (26%MG)	C. sakazakii 95	1.8	-	39.1 0.0 0.0	37.3 0.0 37.4	+/i/-	35.43 0.0 0.0	33.43 32.70 31.86	+/-/-	0.0	37.9	-	37.69 36.34 0.0	32.89 32.70 31.76	+/+/-	-	PD _{FP(alt)}	PD _{FP(alt)}	NA	PD _{FP(alt)}	-	1	a
2905	Organic infant formula. 6-12 months (23.1% FL)	/	/	-	39.2 0.0 0.0	37.3 37.4 0.0	+/-/-	0.0	33.17	-	0.0	37.2	-	0.0	39.27	-	-	PD _{FP(alt)}	NA	NA	NA	+	1	b
2906	Infant formula. stage 2. 6-12 months (26.0% FL)	/	/	-	39.9 0.0 0.0ne	37.8 40.2 40.8	+/-/-ne	38.5 (no amp)	33.02	-	0	37.6	-	0.0	34.5	-	-	PD _{FP(alt)}	NA	NA	NA	+	1	b
2907	Infant formula. stage 2. from 6 months (22.0% FL)	/	/	-	0.0	39.0	-	0.0	0.0	-	37.6 39.8 38.8 0.0	37.4 38.2 37.6 41.9	+/+ +/-	33.04 (no amp)	32.8	-	-	NA	NA	PD _{FP(alt)}	NA	+	1	b
2910	Infant formula. stage 2. 6-12 months (21.1% FL)	/	/	-	0.0	37.4	-	0.0	33.6	-	37.7 0.0 38.2	37.7 38.4 38.3	+/-/+	0.0	35.9	-	-	NA	NA	PD _{FP(alt)}	NA	+	1	b
3738	Vacuum cleaner dust P43 A32 (dairy industry)	/	/	-	0.0	36.7	-	32.74 0.0 0.0 ne	32.33 31.33 32.83	+/-/-ne	37.8 38.6 0.0	37.6/38.0/37.6	+/+/-	0	32.36	-	- (x5)	NA	PD _{FP(alt)}	PD _{FP(alt)}	NA	+	2	b
3739	Vacuum cleaner dust A22 E241 (dairy industry)	/	/	-	37.9 0.0 0.0	38.5 0.0 0.0	+/i/-	28.56 0.0 0.0 ne	40.61 0.0 0.0	+/-/-ne	0.0	38.6	-	0	37.66	-	- (x5)	PD _{FP(alt)}	PD _{FP(alt)}	NA	NA	+	2	b
1371	Vacuum cleaner dust 27 (dairy industry)	C. sakazakii Ad2391	1.8	+	36.3 0.0 33.5	36.0 38.1 36.6	+/-/+	31.88 31.89 32.24	32.01 31.83 31.96	+/+/+	35.5 33.5 33.6	37.0 36.6 36.3	+/+/+	33.33 34.57 33.78	32.65 32.41 32.23	+/+/+	-	PA _{FP(alt)}	PA _{FP(alt)}	PA _{FP(alt)}	PA _{FP(alt)}	-	2	b

ne: new DNA extraction i: inhibited result *: dilution 1:5

♦ Analyses performed according to the COFRAC accreditation (Accreditation Testing n°1-0144, scope available on www.cofrac.fr)

3.1.9 *PCR inhibition*

256 PCR was performed per extraction protocol and thermocycler. 5 PCR inhibitions were observed with the foodproof extraction and the BAX Q7 (2.3% of the PCR) and 1 inhibition with the foodproof extraction and the LC 480 II (0.4% of the PCR). A repetition of the testing or a 1:5 dilution of the DNA extracts was sufficient to obtain a result, and the result obtained where consistent with the other PCR condition and confirmation result (no false negative).

No inhibition was observed with the BAX lysis (0% of the PCR) and therefore, the 1/3 dilution was not evaluated.

Table 28 - Samples with PCR inhibitions

Sample N°	Product	Incubation time	foodproof extraction						BAX Lysis						Category	Type
			BAX Q7			LC480 II			BAX Q7			LC480 II				
			Ct HEX (target)	Ct ROX (IC)	Result	Ct HEX (target)	Ct ROX (IC)	Result	Ct HEX (target)	Ct ROX (IC)	Result	Ct HEX (target)	Ct ROX (IC)	Result		
3570	Infant milk powder. babies with probiotics (B. lactis 3.5 x 10 ⁷ CFU/g) (23.6% fat)	16h	0.0 20.0 21.3*	0.0 0.0 0.0*	i/+/*	17.1	0.0	+	19.6	0.0	+	14.65	0.0	+	1	a
3407	Floor surface swabs before cleaning (dairy industry)	16h	0.0 22.7 25.7*	0.0 0.0 0.0*	i/+/*	20.65	22.50 (no amp)	+	17.1	0.0	+	15.59	0.0	+	2	a
3638	Vacuum nozzle swab. before cleaning (dairy industry)	16 h + 72 h storage	0.0 0.0 0.0*	0.0 37.3 45.0*	i/-/*	0.0	33.18	-	0.0	38.4	-	0.0	37.11	-	2	a
3639	Swab before cleaning (dairy industry)	16 h + 72 h storage	0.0 18.2 21.9*	0.0 0.0 0.0*	i/+/*	16.03	0.0	+	23.9	0.0	+	19.86	0.0	+	2	a
3640	Swab after cleaning (dairy industry)	16 h + 72 h storage	0.0 21.8*	0.0 0.0*	i/+*	15.73	0.0	+	27.2	0.0	+	19.14	0.0	+	2	a
557	Vacuum cleaner dust n°8 (non-probiotic dairy industry)	16h	27.8	0.0	+	0.0 25.63*	0.0 26.43*	i/+*	29.9	0.0	+	25.91	35.25 (uncertain)	+	2	b

*: 1/5 dilution in water

3.2 Relative level of detection

The relative level of detection is the level of detection at $P = 0.50$ (LOD_{50}) of the alternative (proprietary) method divided by the level of detection at $P = 0.50$ (LOD_{50}) of the reference method.

The RLOD is defined as the ratio of the alternative and reference methods:

$$RLOD = \frac{LOD_{Alt.}}{LOD_{Ref.}}$$

The relative detection level is the smallest number of culturable micro-organisms that can be detected in the sample in 50% of occasions by the alternative and reference methods.

3.2.1 Experimental design

Two (matrix/strain) pairs were analyzed by the reference method and by the alternative method (See Table 29).

The following protocol was applied:

- A negative control: 5 samples,
- A low contamination level providing fractional recovery data, with 20 replicates,
- A high contamination level, with 5 replicates.

For these RLOD, the foodproof® extraction was performed using the StarPrep Three 8-strip kit.

For the category 1, for probiotics containing products, lactic bacteria enumeration was carried out using MRS pH 5.7 incubated in anaerobic conditions for 72 h at $30^{\circ}\text{C} \pm 1^{\circ}\text{C}$ (ISO 15214).

A total plate count determination was performed for category 2 to estimate the total microbial load on the day of analysis.

The inoculation level was determined by MPN and enumeration of the inoculum. Close results were obtained by both methods. LOD_{50} was determined using the MPN result.

Table 29 - Defined (matrix/strain) pairs for the RLOD determination

Category		Matrix	Inoculated strain	Test portion for the ISO 22964 method	Test portion for the alternative method	Origin	Inoculating cells	Stabilization conditions	Protocol applied for the ISO 22964 method	Protocol applied for Alt method	Study design
1	Infant formula with or without probiotics and related ingredients	Infant formula with probiotics (<i>Bifidobacterium lactis</i>)	<i>Cronobacter sakazakii</i> Ad2413	10 g	375 g	Infant formula	Lyophilized culture	Storage for 2 weeks at room temperature	BPW 2 T d 1/10 16 h at 37°C ± 1°C	Pre-warmed BPW + vancomycin d 1/10 16 h at 37°C ± 1°C	Unpaired
2	Production environmental samples	Dusts from infant formula production site	<i>Cronobacter muytjensii</i> E888	10 g	200 g	Infant formula	Lyophilized culture	Storage for 2 weeks at room temperature	BPW d 1/10 16 h at 37°C ± 1°C	Pre-warmed BPW + 16 h at 37 +/-1°C	Unpaired

3.2.2 Calculation and interpretation of the RLOD

The raw data are given in **Appendix 5**.

Several unconfirmed positive PCR results was obtained during the testing on infant formula with probiotics, even on uninoculated sample. Additional investigation showed positive results in the absence of enrichment and removal of the positive PCR results after application of a treatment for DNA removal. This confirms the presence of free DNA in the sample.

For the infant formula with probiotics, additional calculations were performed, according to the ISO 16140-2, ISO 16140-2/A1, the RLOD was determined using the alternative method results and the confirmed alternative method results (see Table below).

The RLOD calculations were performed using the Excel spreadsheet available at <http://standards.iso.org/iso/16140> - RLOD version 4 (2024-01-10).

Table 30 – RLOD before and after confirmation for infant formula with probiotics

Infant formula with probiotics / <i>Cronobacter sakazakii</i> Ad2413		
Protocol	RLOD using the alternative method results	RLOD using the confirmed alternative method results
foodproof® Extraction - BAX Q7	0.820	1.242
foodproof® Extraction – LightCycler 480 II	0.598	1.242
BAX Lysis - BAX Q7	N/A*	1.242
BAX Lysis - LightCycler 480 II	N/A*	1.242

* Impossible to calculate as all samples in level 1 and 2 are positives for PCR

Results after cultural confirmation were finally kept for RLOD determination and calculation of the LOD₅₀.

The RLOD are given in Table 31.

Table 31 – Presentation of RLOD after confirmation of the alternative method results

Category	Matrix / strain	foodproof® Extraction				BAX Lysis			
		BAX Q7		Lightcycler 480 II		BAX Q7		Lightcycler 480 II	
		AL	RLOD [95% confidence limit]	AL	RLOD [95% confidence limit]	AL	RLOD [95% confidence limit]	AL	RLOD [95% confidence limit]
1	Infant formula with probiotics / <i>Cronobacter sakazakii</i> Ad2413		1.242 [0.603-2.558]		1.242 [0.603-2.558]		1.242 [0.603-2.558]		1.242 [0.603-2.558]
2	Dusts from infant formula production site / <i>Cronobacter muytjensii</i> E888	2.5	1.543 [0.697-3.416]	2.5	1.375 [0.632-2.990]	2.5	1.375 [0.632-2.990]	2.5	1.232 [0.575-2.639]
Combined			1.372 [0.805-2.338]		1.301 [0.767-2.207]		1.301 [0.767-2.207]		1.235 [0.731-2.085]

The LOD₅₀ calculations were done using the Excel spreadsheet available at <http://standards.iso.org/iso/16140> POD-LOD calculation program - version 12, 2024-03-05. The tests are given in Table 32.

Table 32 - LOD₅₀ results

Category	(Strain / matrix) pair	Level of detection at 50% (CFU / test portion) according to Wilrich & Wilrich ¹				
		Reference method	Alternative method			
			foodproof extraction – BAX Q7	foodproof extraction – LC 480 II	BAX Lysis – BAX Q7	BAX Lysis – LC 480 II
1	Infant formula with probiotics (375 g) / <i>Cronobacter</i> <i>sakazakii</i> Ad2413	0.6 [0.3-1.0]	0.8 [0.5-1.5]	0.8 [0.5 – 1.5]	0.8 [0.5 – 1.5]	0.8 [0.5 – 1.5]
2	Dusts (200 g)/ <i>Cronobacter</i> <i>muytjensii</i> E888	1.0 [0.5-1.9]	1.8 [0.9-3.5]	1.5 [0.8 – 3.0]	1.5 [0.8 – 3.0]	1.3 [0.7 – 2.6]
Combined results		0.8 [0.5-1.1]	1.2 [0.7-1.8]	1.1 (0.7 – 1.7)	1.1 (0.7 – 1.7)	1.0 [0.7 – 1.6]

3.2.3 Conclusion

The RLOD values (using the confirmed alternative method results) meet the acceptability limit of 2.5 for unpaired studies, for the two matrix/strain pairs tested.

The LOD₅₀ varies from 0.7 to 1.6 CFU/test portion for the reference method and from 1.3 to 2.6 CFU/test portion for the alternative method.

3.3 Inclusivity / exclusivity

The inclusivity is the ability of the alternative method to detect the target analyte from a wide range of strains. The exclusivity is the lack of interference from a relevant range of non-target strains of the alternative method.

3.3.1 Test protocols

> Inclusivity

50 target strains were freshly cultured in BHI medium at 37°C. Dilutions were made in order to inoculate 10 -100 CFU/225 ml BPW supplemented with vancomycin

¹ Wilrich, C., and P.-Th. Wilrich: Estimation of the POD function and the LOD of a qualitative microbiological measurement method. AOAC International **92** (2009) 1763 - 1772.

(10 mg/L) as most stringent enrichment. The alternative method protocol was then performed.

The broths were incubated for 16 h at 37°C ± 1°C; the protocol of the alternative method was then applied using the foodproof® StarPrep (Three 8-Strip Kit or Three Kit) and BAX lysis. PCR were performed with the BAX® System Q7 and the LightCycler 480 II. Confirmatory tests were also carried on.

> **Exclusivity**

30 non-target strains were freshly cultured in BHI medium at 37°C. Dilutions were made in order to inoculate about 10⁵ CFU/ml BPW broth. The BPW broth was incubated for 24 h at 37°C ± 1°C. The alternative method was then performed using the foodproof® StarPrep (Three kit or Three 8-Strip Kit) and the BAX lysis protocol for extraction before performing the PCR using BAX® System Q7 and the LightCycler 480 II.

3.3.2 *Results*

Raw data are given in **Appendix 6**.

> **Inclusivity**

All the 50 targets strains gave positive results by PCR for all extraction protocols and thermocyclers. For one strain,

> **Exclusivity**

None of the 50 non-target strain was detected by the PCR in any of tested protocols (extraction protocol and thermocycler).

3.4 Practicability

The alternative method practicability was evaluated according to the AFNOR criteria relative to method comparison study.

<i>Storage conditions, shelf-life and modalities of utilisation after first use</i>	The storage temperature is 2 - 8°C. The shelf-life is given on the package. All the reagents shall be stored at the temperature mentioned on the package.		
<i>Time to result</i>	Steps	Reference method ISO 22964	Alternative method
	Negative samples		
	Sampling enrichment	Day 0	Day 0
	Subculture in CSB	Day 1	/
	Lysate	/	Day 1
	PCR test	/	Day 1
	Streaking onto plate (CCI)	Day 2	/
	Reading plate	Day 3	/
	Results	Day 3	Day 1
	Presumptive positive or positive results		
	Subculture in CSB	Day 1	Day 1
	Streaking onto plates	Day 2	Day 2
	Reading plate	Day 3	Day 3
	Confirmatory tests	Day 5	Day 5
	Results	Day 5	Day 5
<i>Common step with the reference method</i>	Sampling enrichment for BPW protocol		

The negative results are available in 1 day and the positive results in 5 days.

4 INTER-LABORATORY STUDY

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

4.1 Study Design

A first inter-laboratory study was performed in April 2024 with an unpaired study design on 25 g Infant formula with probiotics including a co-inoculation with *Salmonella* spp. Due to difficulties linked to the homogeneity of the inoculum, it was accepted by the technical committee to conduct a new inter-laboratory study with a paired study design to facilitate study organisation and limit any potential bias.

10 g test portion and BPW + vancomycin enrichment was therefore selected for both the reference and the alternative method. The study was conducted in October 2024.

4.2 Study organisation

> Collaborators number

Samples were sent to 16 participants from 12 laboratories and from 6 different countries.

The different extraction methods and instruments were tested by the participants as shown in the table below.

		Instrument		Total
		LC480	BAX Q7	
Extraction method	StarPrep Three	4	3	7
	BAX lysis	4	5	9
Total		8	8	16

> Matrix and strain used

Infant formula with probiotics (*Lactobacillus reuteri*) was inoculated with a lyophilized *Cronobacter sakazakii* Ad940 strain isolated from a milk powder.

> Samples

Samples were prepared and inoculated on Tuesday 1st October 2024, as described below:

- 24 blind coded samples (10 g) for the *Cronobacter spp.* detection by the **ISO 22964 reference method** and by the **foodproof® Salmonella plus Cronobacter Detection LyoKit**.
- 1 sample (labelled “Sample for lactic acid bacteria enumeration”) for mesophilic lactic acid bacteria enumeration in MRS agar (pH 5.7).
- 1 water flask labelled “Temperature Control”.
- 1 probe which follows the temperature during shipping and sample storage.

> Inoculation

The targeted inoculation levels were the following:

- Level: 0 CFU/25 g,
- Level 1: 1 CFU/25 g, inoculation level providing fractional positive recovery data,
- Level 2: 8 CFU/25 g.

Each level was inoculated in bulk and thoroughly mixed prior to repartition in sterile flasks (10g per samples). The samples were then stored at room temperature for two weeks prior to testing.

The contamination level was determined by enumeration of the inoculum the day of the analysis in VRBG and by MPN enumeration of the inoculated bulk using ISO 22964 as described in the table below.

Matrix	Inoculum level	Large test Portion size	Medium test Portion size	Low test Portion size
Infant formula with probiotics	Low	5 x 20 g	8 x 10 g	5 x 5 g
	High	8 x 10 g	5 x 5 g	5 x 2 g

> Labelling and shipping

Blind coded samples were placed in isothermal boxes and express-shipped to the different laboratories.

A temperature control flask and a sensor were added to the package to register the temperature profiles of the shipment during the transport, at reception and during the storage until analyses.

Samples were shipped to the involved laboratories. The temperature conditions had to stay between 18°C to 27°C (ambient temperature) during transport, and during the storage.

> Analyses

Collaborative study laboratories and the expert laboratory carried out the analyses on October 15th or 17th at the latest. The analyses by the reference method and the alternative method were performed on the same day.

4.3 Experimental parameters controls

4.3.1 Strain stability and background microflora stability

Strain stability was checked by evaluating the bulk inoculated with *Cronobacter* spp at the low level at different time points. Eight test portions per method (reference and alternative) were tested the day of inoculation, after 7 days and 14 days storage at ambient temperature. The lactic acid bacteria enumeration was also carried out; the results are given in Table 33.

Table 33 - Sample stability

Date of analysis	ISO 22964	foodproof® Sal/Cro (<i>Cronobacter</i> spp.)	Lactic flora enumeration
	Nb of positives	Nb of positives	CFU/g
D+0	4/8	4/8	4.4 x 10 ⁶
D+7	5/8	5/8	2.9 x 10 ⁶
D+14 (October 15 th)	5/8	5/8	5.6 x 10 ⁶

No evolution was observed during storage at 18 – 27°C for 2 weeks.

4.3.2 Contamination levels

The contamination levels and the sample codification were the following (see Table 34). The contamination was determined by MPN and enumeration of the inoculum (lyophilized strain) the day of the analysis.

Table 34 - Contamination levels

Level	Samples	Theoretical target level (CFU/ test portion)	MPN (CFU/test portion)	VRBG enumeration (CFU/test portion)
0	4, 5, 9, 11, 12, 18, 22, 23	0	0	0
	25, 26, 32, 35, 38, 43, 44, 47			
1	1, 7, 8, 10, 15, 17, 19, 24	1	0.5 [0.2; 0.9]	0.5
	27, 29, 30, 36, 37, 41, 45, 48			
2	2, 3, 6, 13, 14, 16, 20, 21	8	0.9 [0.4; 1.7]	1.3
	28, 31, 33, 34, 39, 40, 42, 46			

The obtained inoculation of this level is lower than expected for Level 2 (0.9 or 1.3 CFU/10 g compared to 8 CFU/10 g), probably due to the difficulty of the contamination with lyophilised strain.

Note that the observed inoculation levels comply with the ISO 16140-2 requirement.

4.3.3 Homogeneity of inoculation

Tests were conducted to verify the homogeneous inoculation with the lyophilized strain. The day of inoculation 2 sets of ten samples for the low level were analysed by the reference method and the alternative methods. The results are provided in Table 35.

Table 35 - Homogeneity test

Inoculation level	Sample replicate	Level 1 (target at 1 CFU/10 g) – Day of inoculation			
		Assay 1		Assay 2	
		ISO 22964	Alternative method	ISO 22964	Alternative method
Low	1	+	+	-	-
	2	-	-	+	+
	3	-	-	-	-
	4	-	-	+	+
	5	+	+	-	-
	6	-	-	+	+
	7	+	+	-	-
	8	+	+	-	-
	9	-	-	+	+
	10	-	-	-	-
	Total	4+/10	4+/10	4+/10	4+/10

The same number of positives was found between the two assays and the number of positives is comprised between 2 and 6, consistent with the target level of inoculation. The samples were sufficiently homogeneous and the inoculation process consistent.

4.3.4 Logistic conditions

Temperature conditions are given in Table 36.

Table 36 - Sample temperatures at receipt

Collaborator	Temperature measured by the probe (°C)	Temperature measured at receipt (°C)	Receipt date and time		Analysis date
A1	17.5	15.9	10/10/2024	9:30 am	16/10/2024
A2	17.5	15.9	10/10/2024	9:30 am	16/10/2024
B1	17.0	/	10/10/2024	11:00 am	15/10/2024
B2	17.0	/	10/10/2024	11:00 am	15/10/2024
C1	16.9	22.3	10/10/2024	1:00 pm	16/10/2024
C2	16.9	22.3	10/10/2024	1:00 pm	16/10/2024
D	14.3	17.3	10/10/2024	10:15 am	15/10/2024
E	20	18.9	10/10/2024	11:45 am	16/10/2024
F	19.9	18.6	10/10/2024	12:00AM	16/10/2024
G	18.5	21.8	10/10/2024	01:10 pm	16/10/2024
H	17.3	14	11/10/2024	11:45 am	15/10/2024
I1	18.1	20.6	10/10/2024	9:00 am	17/10/2024
I2	18.1	20.6	10/10/2024	9:00 am	17/10/2024
J	14.9	8.8	11/10/2024	4:00 pm	16/10/2024
K	21.6	22.3	11/10/2024	1:30 pm	15/10/2024
L	20	19.8	11/10/2024	12:00 am	15/10/2024
M (ADRIA)	20.2	18.4	11/10/2024	11:30 AM	15/10/2024

No problems were encountered during the transport or receipt of samples for 15 collaborators. All the samples were delivered on time and in appropriate conditions. All temperatures maintained during shipment and receipt were correct. For one collaborator (J), the temperature at reception is at 8.8°C but is not consistent with the probe measurement.

For two collaborators (lab J and L), while samples were delivered in good condition, the samples were stored at low temperatures instead of room temperature (17-27°C) before analysis:

- Collaborator J: samples storage temperature between 6 to 12°C for three days (11/10/2024 to 14/10/2024),
- Collaborator L: samples storage temperature at 5 ± 3°C for three days (11/10/2024 to 14/10/2024).

These two collaborators will be therefore removed from the analysis.

4.4 Results analysis

The raw data are given in **Appendix 7**.

4.4.1 Expert laboratory results

The results obtained by the expert laboratory are given in Table 37.

Table 37 – Results obtained by the expert Lab.

Level	Reference method	Alternative method			
		Foodproof StarPrep Three kit		BAX Lysis	
		BAX Q7	LC480	BAX Q7	LC480
L0	0/8	0/8	0/8	0/8	0/8
L1	4/8	4/8	4/8	4/8	4/8
L2	5/8	5/8	5/8	5/8	5/8

Results were consistent between sample preparation kits and instruments. With the Bax lysis and LC480 thermocycler, two PCR positive samples (1 for Level 1 and 1 for Level 2) were not confirmed by cultural method. Replicates of the PCR test gave negative PCR results. It is possible that the presence of free DNA in the sample could be the cause of these false positive results.

Fractional recovery is fulfilled at both inoculation levels, confirming the study was properly managed to comply with the ISO 16140-2: 2016 requirements.

4.4.2 Results observed by the collaborative laboratories

> **Lactic flora enumeration**

Depending on the Lab results, the enumeration levels varied from <10 to 8.2×10^6 CFU/g.

Table 38 – Lactic flora enumeration

Collaborator	Lactic flora (CFU/g)	Analysis date
A1	<10	16/10/2024
A2	<10	16/10/2024
B1	/	15/10/2024
B2	/	15/10/2024
C1	2.4×10^6	16/10/2024
C2	2.6×10^6	16/10/2024
D	1.5×10^2	15/10/2024
E	30	16/10/2024
F	8.2×10^6	16/10/2024
G	<10	16/10/2024
H	<10	15/10/2024
I1	<10	17/10/2024
I2	<10	17/10/2024
J	<10	16/10/2024
K	3.7×10^6	15/10/2024
L	5.0×10^6	15/10/2024
M (ADRIA)	7.7×10^6	15/10/2024

Some collaborators obtained a result <10 CFU/g for lactic flora enumeration. The incubation conditions were complying with the technical rules ($72 \pm 3\text{h}/30 \pm 1^\circ\text{C}$ in anaerobic conditions), except collaborators I1 and I2 who incubated the plates in aerobic conditions.

Due to delays in the delivery of MRS media, collaborators had to use their own media, which may have contributed to the variability in the results. Additionally, the colonies appeared very small, likely adding to the difficulty of accurate enumeration.

> *Cronobacter spp. detection*

14 onto 16 collaborators participated to the study. 2 collaborators (B1 and B2) have had major leaks during the incubation period. They were unable to carry out the analyses.

The results obtained for the 14 collaborators are provided in Table 39 (reference method) and Table 40 (alternative method).

Table 39 - Positive results by the reference method (ALL the collaborators)

Collaborator	Contamination level		
	L0	L1	L2
A1	0	6	4
A2	0	5	2
B1	/	/	/
B2	/	/	/
C1	0	5	4
C2	0	3	3
D	0	5	4
E	0	1	3
F	0	4	3
G	0	4	5
H	0	2	3
I1	0	3	4
I2	0	3	4
J	0	2	4
K	0	4	4
L	0	3	4
TOTAL	P₀ = 0	P₁ = 50	P₂ = 51

**Table 40 - Positive results (before and after confirmation)
by the alternative method (ALL the collaborators)**

Collaborators	Contamination level								
	L0			L1			L2		
	PCR result	Confirmation	Final result	PCR result	Confirmation	Final result	PCR result	Confirmation	Final result
A1	0	0	0	6	6	6	4	4	4
A2	2	0	0	5	5	5	3	2	2
B1	/	/	/	/	/	/	/	/	/
B2	/	/	/	/	/	/	/	/	/
C1	0	0	0	5	5	5	4	4	4
C2	0	0	0	3	3	3	3	3	3
D	0	0	0	5	5	5	4	4	4
E	0	0	0	1	1	1	3	3	3
F	0	0	0	3	4	3	3	3	3
G	0	0	0	4	4	4	5	5	5
H	0	0	0	2	2	2	3	3	3
I1	2	0	0	3	3	3	5	4	4
I2	1	0	0	3	3	3	5	4	4
J	0	0	0	2	2	2	4	4	4
K	0	0	0	4	4	4	4	4	4
L	0	0	0	3	3	3	4	4	4
TOTAL	P₀ = 5	C₀ = 0	CP₀ = 0	P₁ = 49	C₁ = 50	CP₁ = 49	P₂ = 54	C₂ = 51	CP₂ = 51

For three participants, positive PCR results with negative confirmation by cultural method were observed at Level L0 and Level L2:

- Collaborator A2 (BAX Lysis- BAX Q7 thermocycler):
 - Samples n°25 and 26 at Level 0 and n°34 at Level 2.
High Cq values were obtained for the three samples (Cq values 32.9, 36.9 and 36.9 respectively). A PCR replicate with the same DNA extract was carried out and negative results were obtained for the three samples.
- Collaborator I1 (StarPrep lysis - BAX Q7 thermocycler):
 - Samples n°5 et 23 at Level L0 and n°2 at Level L2
High Cq values were obtained for the three samples (Cq values 39.4, 39.9 and 37.5 respectively). A PCR replicate with the same DNA extract was carried out and positive results were obtained for the three samples. A new DNA extract showed a negative result for the samples n°5 and n°23 at Level 0, confirming a potential cross contamination of the original extract.
- Collaborator I2 (StarPrep lysis - BAX Q7 thermocycler):
 - Samples n°43 at Level 0 and n°34 at Level 2
High Cq values were obtained for the two samples (Cq values 36.0, 39.3 and 37.5 respectively). A PCR replicate with the same DNA extract was carried out and positive results were obtained for the two samples. A new DNA extract showed a negative result for both samples, confirming a potential cross contamination of the original extract.

According to the AFNOR technical rules, it is possible to include the results from a collaborator with maximum one cross contamination at Level L0 per method. For this study, this rule was applied and collaborator A2 and I1 were removed.

As described in paragraph 3.2.4, collaborators J and L were also removed due to wrong storage condition prior to analysis.

4.4.3 *Results of the collaborators retained for interpretation*

The results obtained with the 10 labs kept for interpretation are presented in Table 41 (reference method) and Table 42 (alternative method).

Table 41 - Positive results by the reference method
(Without Labs A2, B1, B2, I1, J and L)

Collaborators	Contamination level		
	L0	L1	L2
A1	0	6	4
C1	0	5	4
C2	0	3	3
D	0	5	4
E	0	1	3
F	0	4	3
G	0	4	5
I2	0	3	4
H	0	2	3
K	0	4	4
TOTAL	P₀ = 0	P₁ = 37	P₂ = 37

Table 42 - Positive results (before and after confirmation)
by the alternative method (Without Labs A2, B1, B2, I1, J and L)

Collabo- rators	Contamination level								
	L0			L1			L2		
	PCR result	Confirmation	Final result	PCR result	Confirmation	Final result	PCR result	Confirmation	Final result
A1	0	0	0	6	6	6	4	4	4
C1	0	0	0	5	5	5	4	4	4
C2	0	0	0	3	3	3	3	3	3
D	0	0	0	5	5	5	4	4	4
E	0	0	0	1	1	1	3	3	3
F	0	0	0	3	4	3	3	3	3
G	0	0	0	4	4	4	5	5	5
I2	1	0	0	3	3	3	5	4	4
H	0	0	0	2	2	2	3	3	3
K	0	0	0	4	4	4	4	4	4
TOTAL	P₀ = 1	C₀ = 0	CP₀ = 0	P₁ = 36	C₁ = 37	CP₁ = 36	P₂ = 38	C₂ = 37	CP₂ = 37

4.5 Calculation and interpretation

4.5.1 Calculation of the specificity percentage (SP)

The Specificity percentage (SP) of the reference method and of the alternative method, using the data after confirmation, based on the results of level L0 are presented in Table 43.

Table 43 - Specificity percentage (Without Labs A2, B1, B2, I1, J and L)

Specificity for the reference method	$SP_{ref} = \left(1 - \left(\frac{P_0}{N_-}\right)\right) \times 100 \% =$	100.0%
Specificity for the alternative method	$SP_{alt} = \left(1 - \left(\frac{CP_0}{N_-}\right)\right) \times 100 \% =$	98.8%

N: number of all L0 tests

P_0 = total number of false-positive results obtained with the blank samples before confirmation

CP_0 = total number of false-positive results obtained with blank samples

4.5.2 Calculation of sensitivity (SE_{alt}), sensitivity for the reference method (SE_{ref}), relative trueness (RT), false positive ratio for the alternative method (FPR) and false negative ratio for the alternative method (FNR)

Fractional positive results were obtained for both inoculation levels with respectively 46.3 at level 1 and 46.3% at level 2 for the reference method and 45 at level 1 and 46.3 % at level 2 for the alternative method. The data complies with the ISO16140-2 rules and both inoculation levels were retained for calculation.

A summary of the results of the collaborators retained for interpretation, obtained with the reference and the alternative methods for Level 1 and Level 2 is provided in Table 44.

Table 44 - Summary of results for all collaborators obtained with the reference and alternative methods for Level L1 and Level L2
(Without Labs A2, B1, B2, I1, J and L)

Level	Response	Reference method positive (R+)	Reference method negative (R-)
L1	Alternative method positive (A+)	Positive agreement (A+/R+) PA = 36	Positive deviation (R-/A+) PD = 0
	Alternative method negative (A-)	Total Negative deviation (A-/R+) TND = 1 (1 ND_{FN(alt)})	Total Negative agreement (A-/R-) TNA = 43 (0 NA_{FN(alt)})
L2	Alternative method positive (A+)	Positive agreement (A+/R+) PA = 37 (1 PD_{FP(alt)})	Positive deviation (R-/A+) PD = 0
	Alternative method negative (A-)	Total Negative deviation (A-/R+) TND = 0 (0 ND_{FN(alt)})	Total Negative agreement (A-/R-) TNA = 43 (0 NA_{FN(alt)})

Based on the data summarized in Table 44, the calculated values of the sensitivity of the alternative and reference methods, as well as the relative trueness and false

positive ratio for the alternative method (taking account the confirmations) are presented in Table 45.

Table 45 - Sensitivity, relative trueness, false positive and false negative ratio percentages

		Level L1	Level L2
Sensitivity for the alternative method	$SE_{alt} = \frac{(PA + PD)}{(PA + TND + PD)} \times 100 \%$	97.3 %	100.0 %
Sensitivity for the reference method	$SE_{ref} = \frac{(PA + TND)}{(PA + TND + PD)} \times 100 \%$	100.0 %	100.0 %
Relative trueness	$RT = \frac{(PA + TNA)}{N} \times 100 \%$	98.8 %	100.0 %
False positive ratio for the alternative method (paired evaluation)	$FPR = \frac{PD_{FP(alt)}}{TNA} \times 100 \%$	0.0 %	2.0 %
False negative ratio for the alternative method (paired evaluation)	$FNR = \frac{ND_{FN(alt)}}{PA + TND + PD} \times 100 \%$	2.7 %	0.0 %

4.5.3 Interpretation of data

No positive deviation was observed. Only negative deviation was obtained at Level L1.

The negative deviations are listed in Table 46.

**Table 46 - Negative deviations for Level L1
(Without Labs A2, B1, B2, I1, J and L)**

Lab	Sample N°	Results foodproof® Salmonella plus Cronobacter Detection LyoKit -Cronobacter spp -	Confirmation	Extraction kit	Thermocycler
F	10	-/+ / + / + (ne)	+ (C. sakazakii)	StarPrep Three	Lightcycler480 II

(ne): new DNA extract

One false negative result (negative PCR but positive by confirmation) was obtained for the collaborator F (samples n°10). Three PCR replicates with the same DNA extract and a new DNA extract allowed to obtain positive PCR results every time. Note the early Ct values were observed, respectively 13.0, 12.9 and 12.8: these results confirm a handling error during the first PCR test.

While most of the results were consistent between the PCR and confirmation, two positive PCR results were not confirmed for the alternative method for collaborator I2: sample n°43 at Level L0 and sample n°34 at Level L2. As explained in paragraph 3.3.2, new DNA extraction allowed to obtain a negative result for both samples,

confirming a potential cross contamination of the original extract while handling multiple contaminated samples. See Table 47.

Table 47 – PD_{FP(alt)} results

Lab	Level	Sample N°	Results foodproof® Salmonella plus Cronobacter Detection LyoKit -Cronobacter spp -	Confirmation	Extraction kit	Thermocycler
I2	1	43	+36.0/+34.3/-(ne)	-	StarPrep Three	BAX Q7
I2	2	34	+39.3/+36.0/-(ne)	-	StarPrep Three	BAX Q7

(ne): new DNA extract

For a **paired study design**, the difference between (TND – PD) and the sum of (TND + PD) are calculated for the level(s) where fractional recovery is obtained (so L_1 and L_2). The observed value found for (TND – PD) and (TND + PD) shall not be higher than the AL.

For 10 Labs, the limits are the following:

	Calculated values		AL	Conclusion	
	L1	L2		L1	L2
TND - PD	1	0	3	TND-PD < AL	TND-PD < AL
TND + PD	1	0	4	TND+PD < AL	TND+PD < AL

The ISO 16140-2:2016 & ISO 16140-2/A1: 2024 requirements are fulfilled as (TND - PD) and (TND + PD) meet the AL for Level L1 and Level L2.

4.5.4 Evaluation of the LOD_{50%}, LOD_{95%} and RLOD between laboratories

The LOD_{50%}, was calculated using the ISO 16140-2 Excel spreadsheet available at https://standards.iso.org/iso/16140/-2/ed-1/en/amd/1/PODLOD-interlab_ver2.xlsm.

The RLOD is defined as the ratio of the LODs of the alternative method and the reference method: **RLOD = LOD_{alt}/LOD_{ref}**.

The results are given in Table 48.

**Table 48 - LOD_{50%}, and RLOD
(Without Labs A2, B1, B2, I1, J and L)**

Method	LOD 50%	RLOD
Reference	0.780 [0.595 - 1.021]	1.017
Alternative	0.793 [0.604 - 1.041]	

5 CONCLUSION

The **method comparison study conclusions** are:

- The method comparison study scheme corresponds to a paired study design as the alternative and reference methods have a common enrichment procedure except for infant formula with probiotics for which an unpaired study design was used (addition of vancomycin in the alternative method).
- In the sensitivity study, 2 categories were tested on all alternative method protocols: Infant formula with or without probiotics and related ingredients and the production environmental samples.
- The alternative method shows 12 positive deviations (PD) and between 2 to 4 negative deviations (ND) for all combined categories and protocols. The TND - PD meet the acceptability limits (AL) for each individual categories as well as for the two combined categories.
- The Relative Levels of Detection (RLOD) meet the AL fixed at 2.5 for the unpaired data study.
- The inclusivity and exclusivity testing gave the expected results for the 50 target strains and the 30 non-target strains.
- It is possible to store the primary enrichment broth for 72 h at $5 \pm 3^{\circ}\text{C}$.
- The alternative method allows a one-day screening of the negative samples.
- The results of the method comparison study fulfil all the ISO 16140-2 (2016), ISO 16140-2/A1 (2024) and the AFNOR technical rules (revision 12).

The **inter-laboratory study conclusions** are:

- The first interlaboratory study was performed with an unpaired study design on 25 g Infant formula with probiotics including a co-inoculation with *Salmonella* spp. Due to difficulties encountered during the initial ILS conducted in April 2024 for *Cronobacter* spp. detection, a new inter-laboratory study was performed in paired study design with 10g test portion.
- 14 laboratories participated in the study. 4 of them were removed from the analysis due to cross-contamination at level 0 or wrong storage temperature of the samples.
- The TND - PD and the TND + PD results meet the AL calculated for the retained laboratories for Level L1 and the Level L2.

- These data and interpretations comply with the ISO 16140-2 requirements. **The foodproof® Salmonella plus Cronobacter Detection LyoKit is considered equivalent to the ISO standard method for *Cronobacter* spp detection.**

During method development, the manufacturer assessed whether a high level of one target would interfere with the accurate and reliable detection of the other, even at low levels, and confirmed that no interference occurred.

Quimper, 17 December 2025

Lizaïg GOUGUET

Study Engineer

Method performance in food microbiology

Astrid CARIOU

Manager

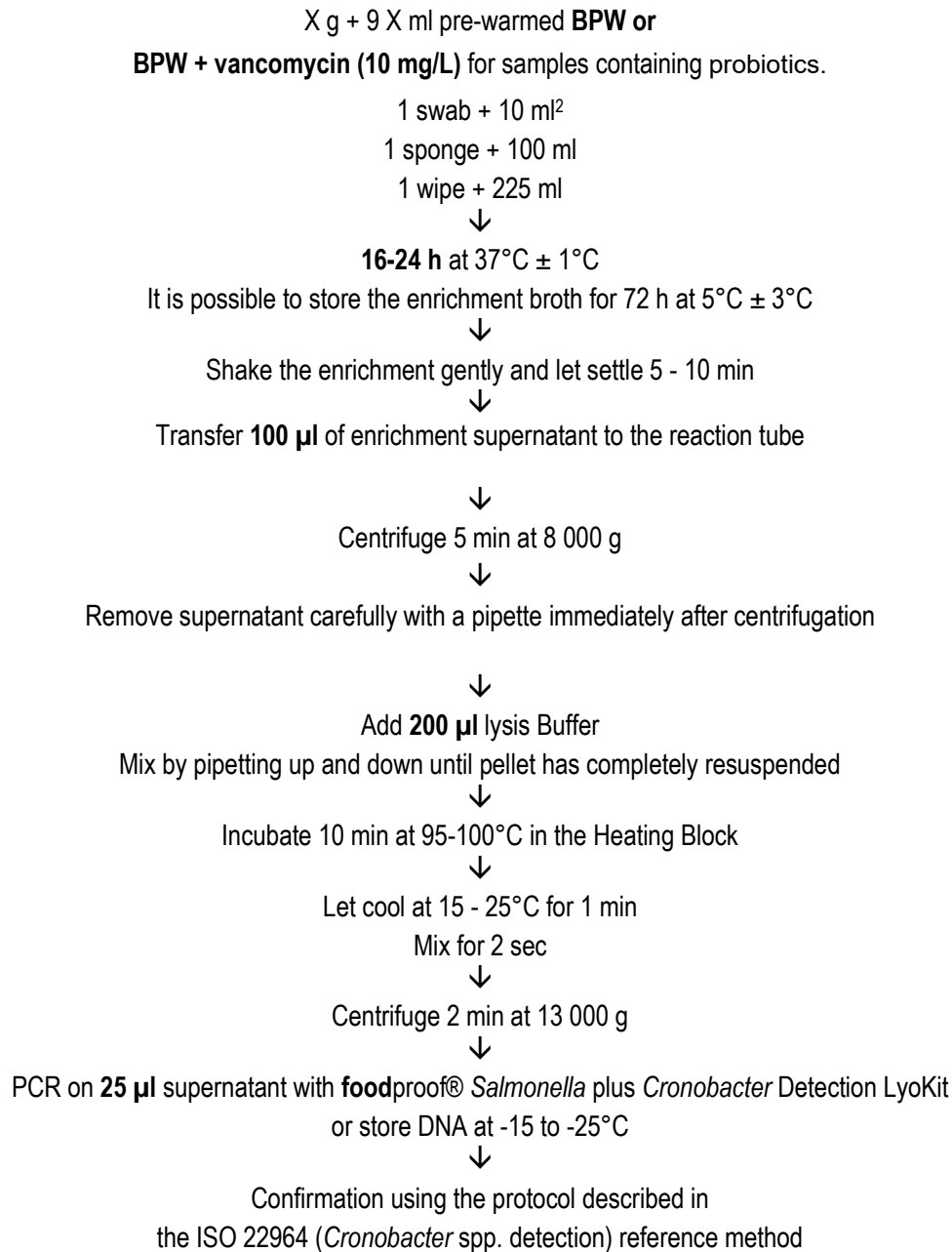
Method performance in food microbiology

I hereby attest to the validation of the results of the analyses carried out under the COFRAC accreditation.

I hereby attest to the validation of the verification of the conformity of the report (opinion and interpretation).

**Appendix 1 – Flow diagrams of the alternative method:
foodproof® *Salmonella* plus *Cronobacter* Detection LyoKit**

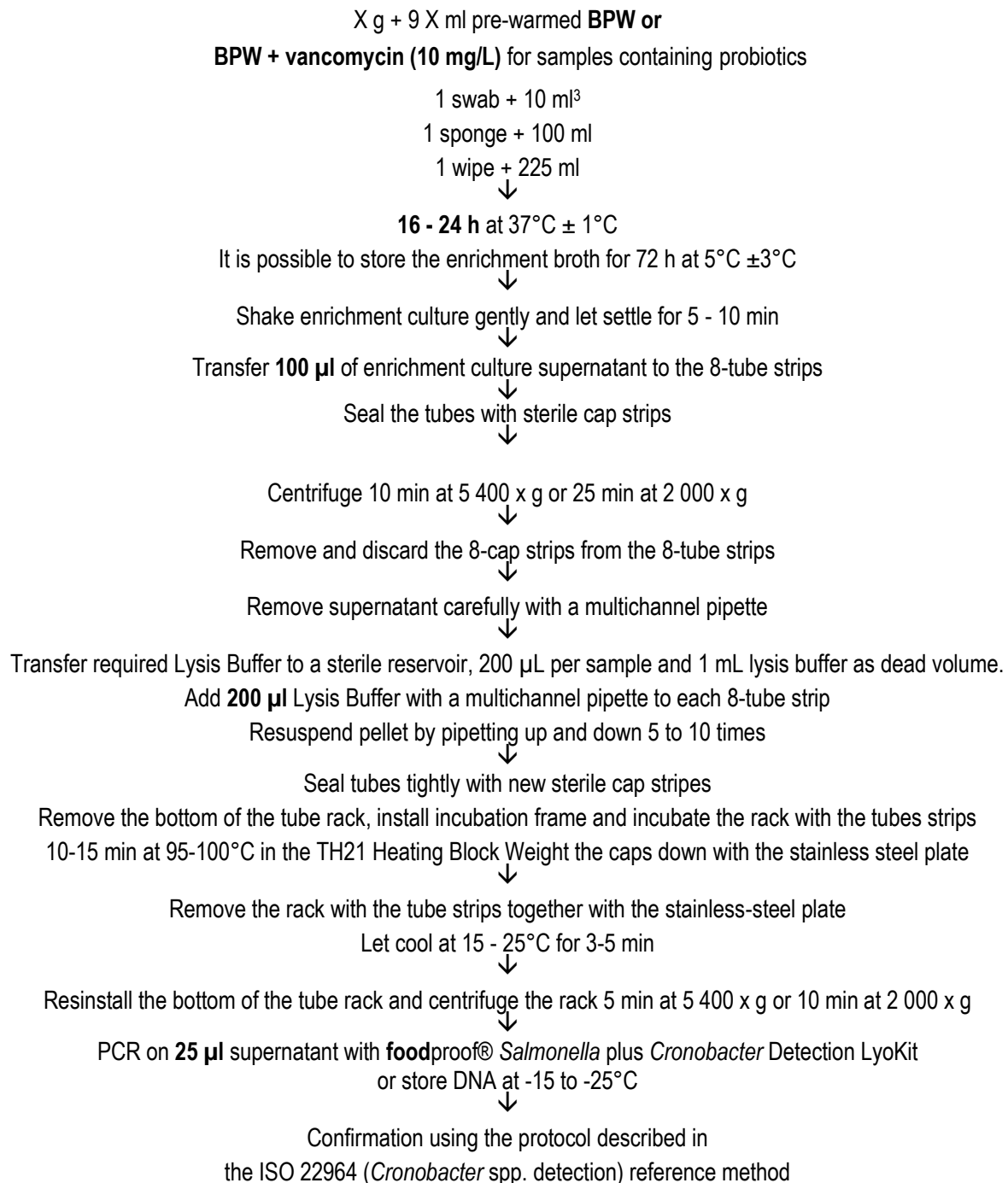
foodproof® StarPrep Three kit



² For sampling after cleaning process, premoisten:

- 1 swab + 1 ml broth universal neutralizing (+ 9 ml BPW broth)
- 1 sponge + 10 ml broth universal neutralizing (+ 90 ml BPW broth)
- 1 wipe + BPW + 10 % neutralizing agent (+ 225 ml BPW broth)

foodproof® StarPrep Three 8-Strip Kit



³ For sampling after cleaning process, premoisten:

- 1 swab + 1 ml broth universal neutralizing (+ 9 ml BPW broth)
- 1 sponge + 10 ml broth universal neutralizing (+ 90 ml BPW broth)
- 1 wipe + BPW + 10 % neutralizing agent (+ 225 ml BPW broth)

BAX Lysis

X g + 9 X ml pre-warmed **BPW** or
BPW + vancomycin (10 mg/L) for samples containing probiotics

1 swab + 10 ml⁴

1 sponge + 100 ml

1 wipe + 225 ml



16 -24 h at 37°C ± 1°C

It is possible to store the enrichment broth for 72 h at 5°C ±3°C



Shake enrichment culture gently and let settle for 5 - 10 min



Prepare lysis reagent by adding 150 µL protease to one 12 mL bottle of lysis buffer



Transfer **200 µL** lysis reagent to each cluster tube



Transfer **20 µL** enrichment culture supernatant to the corresponding cluster tube



Heat at 37°C for 20 minutes



Heat at 95°C for 10 minutes



Cool at 2-8°C at least 5 minutes



PCR on **30 µl** supernatant with **foodproof®** *Salmonella* plus *Cronobacter* Detection LyoKit
or store DNA at -15 to -25°C

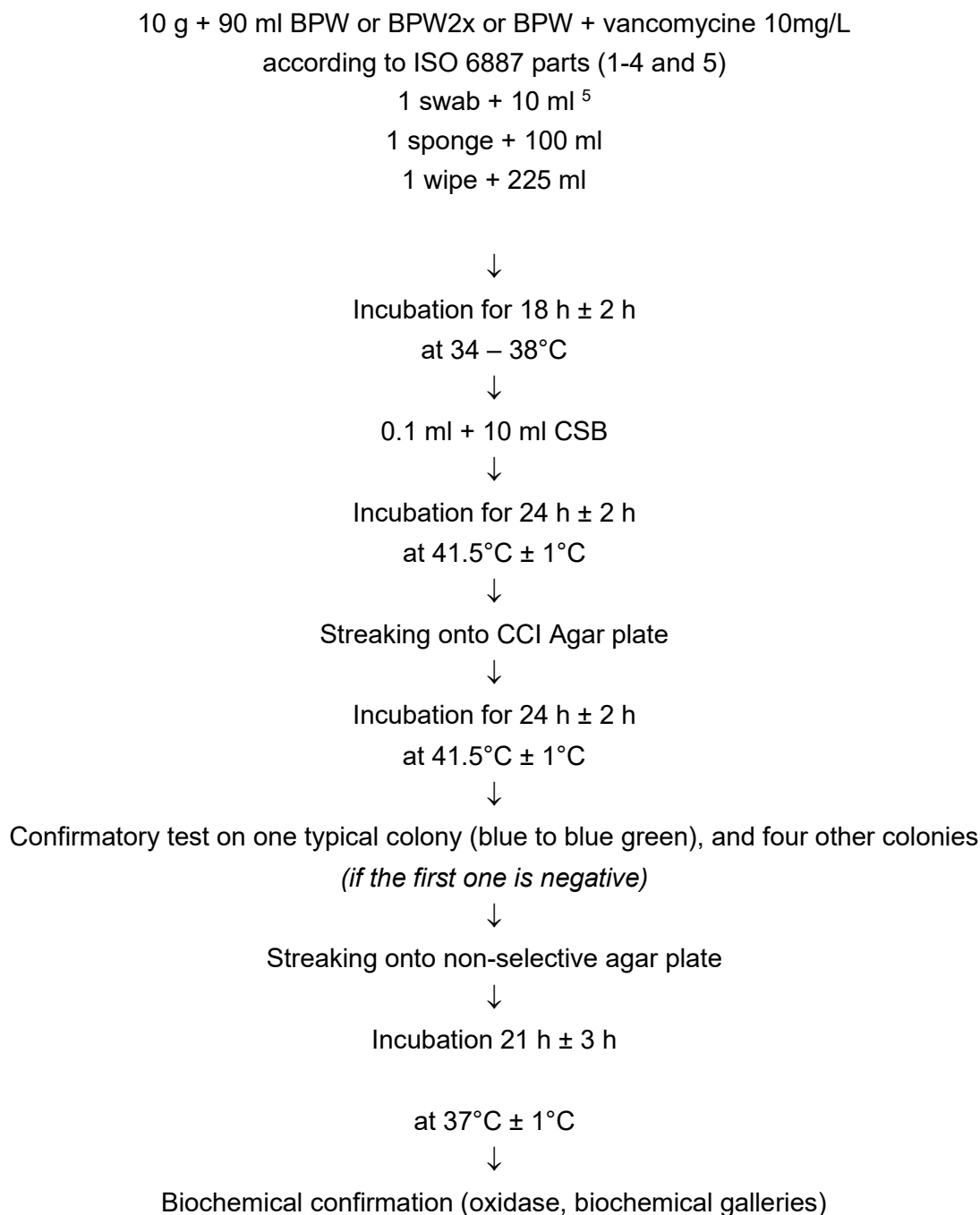


Confirmation using the protocol described
in the ISO 22964 (*Cronobacter* spp. detection) reference method

⁴ For sampling after cleaning process, premoisten:

- 1 swab + 1 ml broth universal neutralizing (+ 9 ml BPW broth)
- 1 sponge + 10 ml broth universal neutralizing (+ 90 ml BPW broth)
- 1 wipe + BPW + 10 % neutralizing agent (+ 225 ml BPW broth)

Appendix 2 – Flow diagram of the reference method: ISO 22964 (April 2017)
Microbiology of the food chain - Horizontal method for the detection
of *Cronobacter* spp.



⁵ For sampling after cleaning process, premoisten:

- 1 swab + 1 ml broth universal neutralizing (+ 9 ml BPW broth)
- 1 sponge + 10 ml broth universal neutralizing (+ 90 ml BPW broth)
- 1 wipe + BPW + 10 % neutralizing agent (+ 225 ml BPW broth)

Appendix 3 – Artificial contamination of samples

Year of analysis	Sample N°	Product	Product (French name)	Artificial contamination						Global result	Category	Type
				Strain	Origin	Injury protocol	Injury measure- ment	Inoculation level/ sample		Cronobacter spp		
								Enume- ration	Mean			
2022	3566	Poudre de lait infantile, 6-12 mois, lait de suite, avec probiotiques (<i>B. infantis</i> 2.5 x 10 ⁶ UFC/g) (26%MG)	Infant milk powder, 6-12 months, follow-on milk, with probiotics (<i>B. infantis</i> 2.5 x 10 ⁶ CFU/g) (26% fat)	<i>C. sakazakii</i> 95	Dairy product	Seeding protocol with lyophilised strain 2 weeks at ambient temperature	/	/	1.8	+	1	a
2022	3567	Poudre de lait infantile, 6-12 mois, avec probiotiques (<i>B. infantis</i> 3.2 x 10 ⁷ UFC/g) (23%MG)	Infant milk powder, 6-12 months, with probiotics (<i>B. infantis</i> 3.2 x 10 ⁷ CFU/g) (23%MG)	<i>C. sakazakii</i> 95	Dairy product	Seeding protocol with lyophilised strain 2 weeks at ambient temperature	/	/	1.8	+	1	a
2022	3568	Poudre de lait infantile, 6-12 mois, avec probiotiques (<i>B. lactis</i> 1.8 x 10 ⁶ UFC/g) (26%MG)	Infant milk powder, 6-12 months, with probiotics (<i>B. lactis</i> 1.8 x 10 ⁶ CFU/g) (26%MG)	<i>C. sakazakii</i> 95	Dairy product	Seeding protocol with lyophilised strain 2 weeks at ambient temperature	/	/	1.8	-	1	a
2022	3569	Poudre de lait infantile, dès 6 mois, avec probiotiques (<i>B. lactis</i> 3.5 x 10 ⁷ UFC/g) (26%MG)	Infant milk powder, from 6 months, with probiotics (<i>B. lactis</i> 3.5 x 10 ⁷ CFU/g) (26% fat)	<i>C. sakazakii</i> 95	Dairy product	Seeding protocol with lyophilised strain 2 weeks at ambient temperature	/	/	1.8	+	1	a
2022	3570	Poudre de lait infantile, bébés gourmands, avec probiotiques (<i>B. lactis</i> 3.5 x 10 ⁷ UFC/g) (23.1%MG)	Infant milk powder, babies, with probiotics (<i>B. lactis</i> 3.5 x 10 ⁷ CFU/g) (23.6% fat)	<i>C. sakazakii</i> 95	Dairy product	Seeding protocol with lyophilised strain 2 weeks at ambient temperature	/	/	1.8	+	1	a
2022	3797	Poudre de lait infantile, 3ème âge, avec probiotiques (<i>B. lactis</i> + <i>S. thermophilus</i> 1.5 x 10 ⁶ UFC/g) (30% MG)	Infant milk powder, 3rd age, with probiotics (<i>B. lactis</i> + <i>S. thermophilus</i> 1.5 x 10 ⁶ CFU/g) (30% fat)	<i>C. sakazakii</i> Ad935 (+ <i>S. Ohio</i> Ad2213)	Dairy product/ Dairy product	Seeding protocol with lyophilised strain 2 weeks at ambient temperature	/	/	0.3/2.4	+	1	a
2022	3798	Poudre de lait infantile, 3ème âge, avec probiotiques (<i>B. lactis</i> + <i>S. thermophilus</i> 1.5 x 10 ⁶ UFC/g) (30% MG)	Infant milk powder, 3rd age, with probiotics (<i>B. lactis</i> + <i>S. thermophilus</i> 1.5 x 10 ⁶ CFU/g) (30% fat)	<i>C. sakazakii</i> Ad2407 (+ <i>S. Duisburg</i> Ad1812)	Dairy product /Dairy product	Seeding protocol with lyophilised strain 2 weeks at ambient temperature	/	/	<0.5/3.0	+	1	a
2022	3799	Poudre de lait infantile, 2ème âge, avec probiotiques (<i>B. lactis</i> + <i>S. thermophilus</i> 3.1 x 10 ⁷ UFC/g) (23% MG)	Infant milk powder, 2nd age, with probiotics (<i>B. lactis</i> + <i>S. thermophilus</i> 3.1 x 10 ⁷ CFU/g) (23% fat)	<i>C. sakazakii</i> Ad935 (+ <i>S. Ohio</i> Ad2213)	Dairy product/ Dairy product	Seeding protocol with lyophilised strain 2 weeks at ambient temperature	/	/	0.3/2.4	+	1	a
2022	3800	Poudre de lait infantile, 2ème âge, avec probiotiques (<i>B. lactis</i> + <i>S. thermophilus</i> 3.1 x 10 ⁷ UFC/g) (23% MG)	Infant milk powder, 2nd age, with probiotics (<i>B. lactis</i> + <i>S. thermophilus</i> 3.1 x 10 ⁷ CFU/g) (23% fat)	<i>C. sakazakii</i> Ad2382 (+ <i>S. Duisburg</i> Ad1812)	Dairy product/ Dairy product	Seeding protocol with lyophilised strain 2 weeks at ambient temperature	/	/	<0.5/0.5	+	1	a
2022	3801	Poudre de lait infantile, 1er âge, avec probiotiques (<i>B. lactis</i> 8.6 x 10 ⁵ UFC/g) (26% MG)	Infant milk powder, 1st age, with probiotics (<i>B. lactis</i> 8.6 x 10 ⁵ CFU/g) (26% fat)	<i>C. sakazakii</i> Ad935 (+ <i>S. Ohio</i> Ad2213)	Dairy product/ Dairy product	Seeding protocol with lyophilised strain 2 weeks at ambient temperature	/	/	0.3/2.4	+	1	a
2023	763	Poudre de lait infantile avec probiotiques, croissance dès 1 an (<i>L. reuteuri</i> DSM 17938 8.55 x 10 ⁴ UFC/g) (26% MG)	Infant milk powder with probiotics, 3rd age (<i>L. reuteuri</i> DSM 17938 8.55 x 10 ⁴ CFU/g) (26% fat)	<i>C. malonaticus</i> DSM18702	Dairy product	Spiking protocol, heat treatment 56°C 8 min	1.13	1-1-3-2-2	1.8	+	1	a

Year of analysis	Sample N°	Product	Product (French name)	Artificial contamination						Global result	Category	Type
				Strain	Origin	Injury protocol	Injury measure- ment	Inoculation level/ sample		Cronobacter spp		
								Enume- ration	Mean			
2023	765	Poudre de lait infantile avec probiotiques, système immunitaire dès 10 mois (<i>L. reuteri</i> DSM 17938 1.55 x 10 ⁵ UFC/g) (23% MG)	Infant milk powder with probiotics, after from 10 months (<i>L. reuteri</i> DSM 17938 1.55 x 10 ⁵ CFU/g) (23% fat)	<i>C. sakazakii</i> Ad898	Dairy product	Spiking protocol, heat treatment 56°C 8 min	0.95	3-1-2-2-0	1.6	+	1	a
2023	767	Poudre de lait infantile avec probiotiques, 3ème âge dès 1 an (<i>L. reuteri</i> DSM 17938 8.24 x 10 ⁴ UFC/g) (23.1% MG)	Infant milk powder with probiotics, 3rd age from 1 year (<i>L. reuteri</i> DSM 17938 8.24 x 10 ⁴ CFU/g) (23.4% fat)	<i>C. sakazakii</i> Ad897	Dairy product	Spiking protocol, heat treatment 56°C 8 min	1.98	1-1-4-0-1	1.4	+	1	a
2023	769	Poudre de lait infantile avec probiotiques, bio dès 6 mois (<i>L. reuteri</i> DSM 17938 2.45 x 10 ⁶ UFC/g) (26% MG)	Infant milk powder with probiotics, organic from 6 months (<i>L. reuteri</i> DSM 17938 2.45 x 10 ⁶ CFU/g) (26% fat)	<i>C. malonaticus</i> DSM18702	Dairy product	Spiking protocol, heat treatment 56°C 8 min	1.13	1-1-3-2-2	1.8	+	1	a
2023	771	Poudre de lait infantile avec probiotiques.1er âge pour nourrisson 0-6 mois (<i>L. fermentum hereditum</i> 5.09 x 10 ⁶ UFC/g) (24% MG)	Infant milk powder with probiotics, 1st age for infant 0-6 months (<i>L. fermentum hereditum</i> 5.09 x 10 ⁶ CFU/g) (24% fat)	<i>C. sakazakii</i> Ad898	Dairy product	Spiking protocol, heat treatment 56°C 8 min	0.95	3-1-2-2-0	1.6	+	1	a
2022	2963	Poudre de lait infantile, 1er âge, 6-12 mois (22.0%MG)	Infant formula, stage 1, 6-12 months (22.0% FL)	<i>C. sakazakii</i> Ad2413	Milk powder	Seeding protocol with lyophilised strain 2 weeks at ambient temperature	/	/	2.1	+	1	b
2022	2964	Poudre de lait infantile bio, 0-6 mois (24.0%MG)	Organic infant formula, 0-6 months (24.0% FL)	<i>C. sakazakii</i> Ad2413	Milk powder	Seeding protocol with lyophilised strain 2 weeks at ambient temperature	/	/	2.1	+	1	b
2022	2965	Poudre de lait infantile bio, 0-6 mois (24.0%MG)	Organic infant formula, 0-6 months (24.0% FL)	<i>C. turicensis</i> Ad1445	Milk powder	Seeding protocol with lyophilised strain 2 weeks at ambient temperature	/	/	2.0	+	1	b
2022	2966	Poudre de lait infantile, 0-6 mois (22.0%MG)	Infant formula, 0-6 months (22.0% FL)	<i>C. turicensis</i> Ad1445	Milk powder	Seeding protocol with lyophilised strain 2 weeks at ambient temperature	/	/	2.0	+	1	b
2022	2967	Poudre de lait infantile, 0-6 mois (21.0%MG)	Infant formula, 0-6 months (21.0% FL)	<i>C. turicensis</i> Ad1445	Milk powder	Seeding protocol with lyophilised strain 2 weeks at ambient temperature	/	/	2.0	+	1	b
2022	2968	Poudre de lait infantile, 2ème âge, 6-12 mois (26.0%MG)	Infant formula, stage 2, 6-12 months (26.0% FL)	<i>C. sakazakii</i> Ad2413	Milk powder	Seeding protocol with lyophilised strain 2 weeks at ambient temperature	/	/	2.1	+	1	b
2022	2969	Poudre de lait infantile bio, après 6 mois (25.0%MG)	Organic infant formula, from 6 months (25.0%FL)	<i>C. sakazakii</i> Ad2413	Milk powder	Seeding protocol with lyophilised strain 2 weeks at ambient temperature	/	/	2.1	+	1	b
2022	2970	Poudre de lait infantile bio, 6-12mois (22.0%MG)	Organic infant formula, 6-12 months (22.0%FL)	<i>C. turicensis</i> Ad1445	Milk powder	Seeding protocol with lyophilised strain 2 weeks at ambient temperature	/	/	2.0	+	1	b
2022	3023	Poudre de lait infantile anti régurgitations, 0-6 mois (25% MG)	Infant formula anti-regurgitation, 0-6 months (25% FL)	<i>C. muytjensii</i> E888 (+ <i>S. Anatum</i> Ad2718)	Dairy product/ Milk powder	Seeding protocol with lyophilised strain 2 weeks at ambient temperature	/	/	<1.0/0.5	+	1	b

Year of analysis	Sample N°	Product	Product (French name)	Artificial contamination						Global result	Category	Type
				Strain	Origin	Injury protocol	Injury measure- ment	Inoculation level/ sample		Cronobacter spp		
								Enume- ration	Mean			
2022	3024	Poudre de lait infantile bio, 2ème âge, 6-12 mois (25%MG)	Organic infant formula, stage 2, 6-12 months (25%FL)	C. muytjensii E888 (+ S. Anatum Ad2718)	Dairy product/ Milk powder	Seeding protocol with lyophilised strain 2 weeks at ambient temperature	/	/	<1.0/0.5	+	1	b
2022	3025	Poudre de lait infantile, bébés gourmands, 6-12 mois (26%MG)	Infant milk powder, gourmet babies, 6-12 months (26%fat)	C. muytjensii E888 (+ S. Anatum Ad2718)	Dairy product/ Milk powder	Seeding protocol with lyophilised strain 2 weeks at ambient temperature	/	/	<1.0/0.5	+	1	b
2022	3026	Poudre de lait infantile, 6-12 mois (25.5%MG)	Infant milk powder, 6-12 months (25.5% fat)	C. sakazakii Ad935 (+ S. Cerro Ad2707)	Dairy product/ Milk powder	Seeding protocol with lyophilised strain 2 weeks at ambient temperature	/	/	3.6/3.0	+	1	b
2022	3027	Poudre de lait infantile bio, 2ème âge (25%MG)	Organic infant milk powder, 2nd age (25%fat)	C. sakazakii Ad935 (+ S. Cerro Ad2707)	Dairy product/ Milk powder	Seeding protocol with lyophilised strain 2 weeks at ambient temperature	/	/	3.6/3.0	+	1	b
2022	3028	Poudre de lait infantile premium, 0-6 mois (24.7%MG)	Premium infant milk powder, 0-6 months (24.7% fat)	C. sakazakii Ad935 (+ S. Cerro Ad2707)	Dairy product/ Milk powder	Seeding protocol with lyophilised strain 2 weeks at ambient temperature	/	/	3.6/3.0	+	1	b
2022	3089	Amidon de riz	Rice starch	C. sakazakii Ad2341	Vegetable product	Seeding protocol with lyophilised strain 2 weeks at ambient temperature	/	/	<0.5	+	1	c
2022	3090	Amidon de maïs	Corn starch	C. sakazakii Ad2341	Vegetable product	Seeding protocol with lyophilised strain 2 weeks at ambient temperature	/	/	<0.5	+	1	c
2022	3091	Maltodextrine	Maltodextrin	C. muytjensii E888	Milk powder	Seeding protocol with lyophilised strain 2 weeks at ambient temperature	/	/	1.2	+	1	c
2022	3092	Lactosérum	Whey	C. muytjensii E888	Milk powder	Seeding protocol with lyophilised strain 2 weeks at ambient temperature	/	/	1.2	+	1	c
2022	3093	Lactosérum	Whey	C. muytjensii E888	Milk powder	Seeding protocol with lyophilised strain 2 weeks at ambient temperature	/	/	1.2	+	1	c
2022	3094	Maltodextrine	Maltodextrin	C. sakazakii Ad2341	Vegetable product	Seeding protocol with lyophilised strain 2 weeks at ambient temperature	/	/	<0.5	+	1	c
2022	3175	Lactose	Lactose	C. sakazakii Ad2413 (+ S. Anatum Ad2718)	Dairy product/milk powder	Seeding protocol with lyophilised strain 2 weeks at ambient temperature	/	/	3.9	+	1	c
2022	3176	Lactose	Lactose	C. turicensis Ad1445 (+ S. Anatum Ad2718)	Dairy product/milk powder	Seeding protocol with lyophilised strain 2 weeks at ambient temperature	/	/	3.9	+	1	c
2022	3177	Lactosérum	Whey	C. sakazakii Ad2413 (+ S. Cerro Ad2707)	Dairy product/milk powder	Seeding protocol with lyophilised strain 2 weeks at ambient temperature	/	/	1.8	+	1	c
2022	3178	Amidon de blé	Wheat starch	C. sakazakii Ad2848 (+ S. Anatum Ad2718)	Dairy product / infant cereals	Seeding protocol with lyophilised strain 2 weeks at ambient temperature	/	/	3.9	+	1	c

Year of analysis	Sample N°	Product	Product (French name)	Artificial contamination						Global result	Category	Type
				Strain	Origin	Injury protocol	Injury measurement	Inoculation level/ sample		Cronobacter spp		
								Enumeration	Mean			
2022	3179	Maltodextrine	Maltodextrin	C. turicensis Ad1445 (+ S. Cerro Ad2707)	Dairy product/milk powder	Seeding protocol with lyophilised strain 2 weeks at ambient temperature	/	/	1.8	+	1	c
2022	3180	Protéines concentrées de lactosérum	Whey Protein Concentrate	C. turicensis Ad1445 (+ S. Cerro Ad2707)	Dairy product/milk powder	Seeding protocol with lyophilised strain 2 weeks at ambient temperature	/	/	1.8	+	1	c
2022	3399	Ecouvillons surface sol avant nettoyage (industrie de produits laitiers)	Floor surface swabs before cleaning (dairy industry)	C. sakazakii Ad2409	Environmental sample (dairy industry)	Seeding protocol 48h at 3±2°C	/	1-4-3-0-0	1.6	+	2	a
2022	3400	Ecouvillons surface sol avant nettoyage (industrie de produits laitiers)	Floor surface swabs before cleaning (dairy industry)	C. sakazakii Ad2409	Environmental sample (dairy industry)	Seeding protocol 48h at 3±2°C	/	1-4-3-0-0	1.6	+	2	a
2022	3401	Ecouvillons surface sol avant nettoyage (industrie de produits laitiers)	Floor surface swabs before cleaning (dairy industry)	C. sakazakii Ad2409	Environmental sample (dairy industry)	Seeding protocol 48h at 3±2°C	/	1-4-3-0-0	1.6	+	2	a
2022	3402	Ecouvillons surface sol avant nettoyage (industrie de produits laitiers)	Floor surface swabs before cleaning (dairy industry)	C. sakazakii Ad2409	Environmental sample (dairy industry)	Seeding protocol 48h at 3±2°C	/	1-4-3-0-0	1.6	+	2	a
2022	3407	Ecouvillons surface sol avant nettoyage (industrie de produits laitiers)	Floor surface swabs before cleaning (dairy industry)	C. sakazakii Ad2342	Environmental sample (dairy industry)	Seeding protocol 48h at 3±2°C	/	0-5-4-0-4	2.6	+	2	a
2022	3408	Ecouvillons surface sol avant nettoyage (industrie de produits laitiers)	Floor surface swabs before cleaning (dairy industry)	C. sakazakii Ad2342	Environmental sample (dairy industry)	Seeding protocol 48h at 3±2°C	/	0-5-4-0-4	2.6	+	2	a
2022	3409	Ecouvillons surface sol avant nettoyage (industrie de produits laitiers)	Floor surface swabs before cleaning (dairy industry)	C. sakazakii Ad2342	Environmental sample (dairy industry)	Seeding protocol 48h at 3±2°C	/	0-5-4-0-4	2.6	+	2	a
2022	3410	Ecouvillons surface sol avant nettoyage (industrie de produits laitiers)	Floor surface swabs before cleaning (dairy industry)	C. sakazakii Ad2342	Environmental sample (dairy industry)	Seeding protocol 48h at 3±2°C	/	0-5-4-0-4	2.6	+	2	a
2022	3475	Chiffonnette avant nettoyage sceau crème glacée (production crème glacée)	Swipe before cleaning (ice cream production)	C. dublinensis LMG23823T (+S. Braenderup Ad2151)	Environmental sample (dairy industry) / Environmental sample (dairy industry)	Seeding protocol 48h at 3±2°C	/	5-0-3-3-3/1-2-0-2-1	2.8/1.2	-	2	a
2022	3476	Chiffonnette avant nettoyage sceau crème glacée (production crème glacée)	Swipe before cleaning (ice cream production)	C. sakazakii Ad2343 (+ S. Senftenberg Ad2149)	Environmental sample (dairy industry) / Environmental sample (dairy industry)	Seeding protocol 48h at 3±2°C	/	1-3-2-2-3/2-0-2-1-1	2.2/1.2	+	2	a
2022	3477	Chiffonnette après nettoyage homo ADV crème glacée (production crème glacée)	Swipe after cleaning (ice cream production)	C. dublinensis LMG23823T (+ S. Braenderup Ad2151)	Environmental sample (dairy industry) / Environmental sample (dairy industry)	Seeding protocol 48h at 3±2°C	/	5-0-3-3-3/1-2-0-2-1	2.8/1.2	+	2	a
2022	3478	Chiffonnette avant nettoyage pompe crème glacée (production crème glacée)	Swipe before cleaning (Ice Cream Production)	C. sakazakii Ad2343 (+ S. Senftenberg Ad2149)	Environmental sample (dairy industry) / Environmental sample (dairy industry)	Seeding protocol 48h at 3±2°C	/	1-3-2-2-3/2-0-2-1-1	2.2/1.2	-	2	a
2022	3479	Chiffonnette après nettoyage ouverture tuyau machine (industrie de produits laitiers)	Swipe After cleaning (Dairy Industry)	C. dublinensis LMG23823T (+ S. Braenderup Ad2151)	Environmental sample (dairy industry) / Environmental sample (dairy industry)	Seeding protocol 48h at 3±2°C	/	5-0-3-3-3/1-2-0-2-1	2.8/1.2	-	2	a

Year of analysis	Sample N°	Product	Product (French name)	Artificial contamination						Global result	Category	Type
				Strain	Origin	Injury protocol	Injury measure- ment	Inoculation level/ sample		Cronobacter spp		
								Enume- ration	Mean			
2022	3480	Chiffonnette après nettoyage remplissage dessus container (industrie de produits laitiers)	Swipe After cleaning (Dairy Industry)	C. sakazakii Ad2343 (+ S. Senftenberg Ad2149)	Environmental sample (dairy industry) / Environmental sample (dairy industry)	Seeding protocol 48h at 3±2°C	/	1-3-2-2-3/ 2-0-2-1-1	2.2/1.2	+	2	a
2022	3481	Chiffonnette après nettoyage sol ascenseur tour 3 (industrie de produits laitiers)	Swipe After cleaning (Dairy Industry)	C. sakazakii Ad2343 (+ S. Senftenberg Ad2149)	Environmental sample (dairy industry) / Environmental sample (dairy industry)	Seeding protocol 48h at 3±2°C	/	1-3-2-2-3/ 2-0-2-1-1	2.2/1.2	+	2	a
2022	3639	Ecouvillon raclette plastique 1er étage, avant nettoyage (industrie de produits laitiers)	Swab before cleaning (dairy industry)	C. sakazakii Ad2364	Environmental sample (dairy product industry)	Seeding protocol 48h at 3±2°C	/	2-1-2-2-1	1.6	+	2	a
2022	3640	Ecouvillon main courante remplissage, après nettoyage (industrie de produits laitiers)	Swab after cleaning (dairy industry)	C. sakazakii Ad2364	Environmental sample (dairy product industry)	Seeding protocol 48h at 3±2°C	/	2-1-2-2-1	1.6	+	2	a
2022	3644	Chiffonnette pompe crème glacée, après nettoyage (production de crème glacée)	Swipe after cleaning (ice cream production)	C. sakazakii Ad2364	Environmental sample (dairy product industry)	Seeding protocol 48h at 3±2°C	/	2-1-2-2-1	1.6	+	2	a
2022	3740	Poussières d'aspirateur A32 E553 A31 (industrie de produits laitiers)	Vacuum cleaner dust A32 E553 A31 (dairy industry)	C. sakazakii Ad2407	Dairy product	Seeding protocol with lyophilised strain 2 weeks at ambient temperature	/	/	1.5	+	2	b
2022	3741	Poussières d'aspirateur P51 2889 A12 (industrie de produits laitiers)	Vacuum cleaner dust P51 2889 A12 (dairy industry)	C. sakazakii Ad2407	Dairy product	Seeding protocol with lyophilised strain 2 weeks at ambient temperature	/	/	1.5	+	2	b
2022	3742	Poussières d'aspirateur P51 E2E2 (industrie de produits laitiers)	Vacuum cleaner dust P51 E2E2 (dairy industry)	C. sakazakii Ad2382	Dairy product	Seeding protocol with lyophilised strain 2 weeks at ambient temperature	/	/	0.5	+	2	b
2022	3743	Poussières d'aspirateur P32 P41 (industrie de produits laitiers)	Vacuum cleaner dust P32 P41 (dairy industry)	C. sakazakii Ad2382	Dairy product	Seeding protocol with lyophilised strain 2 weeks at ambient temperature	/	/	0.5	-	2	b
2023	555	Poussières d'aspirateur n°6 (industrie de produits laitiers non probiotiques)	Vacuum cleaner dust n°6 (non-probiotic dairy industry)	C. sakazakii Ad2360	Environmental sample (dairy industry)	Spiking protocol, heat treatment 56°C 8 min	1.2	1-0-0-0-0	0.2	-	2	b
2023	556	Poussières d'aspirateur n°7 (industrie de produits laitiers non probiotiques)	Vacuum cleaner dust n°7 (non-probiotic dairy industry)	C. sakazakii Ad2360	Environmental sample (dairy industry)	Spiking protocol, heat treatment 56°C 8 min	1.2	1-0-0-0-0	0.2	-	2	b
2023	557	Poussières d'aspirateur n°8 (industrie de produits laitiers non probiotiques)	Vacuum cleaner dust n°8 (non-probiotic dairy industry)	C. sakazakii Ad2386	Environmental sample (dairy industry)	Spiking protocol, heat treatment 56°C 8 min	1.60	0-1-1-1-2	1.0	+	2	b
2023	558	Poussières d'aspirateur n°9 (industrie de produits laitiers non probiotiques)	Vacuum cleaner dust n°9 (non-probiotic dairy industry)	C. sakazakii Ad2386	Environmental sample (dairy industry)	Spiking protocol, heat treatment 56°C 8 min	160	0-1-1-1-2	1.0	+	2	b
2023	559	Poussières d'aspirateur n°10 (industrie de produits laitiers non probiotiques)	Vacuum cleaner dust n°10 (non-probiotic dairy industry)	C. sakazakii Ad2387	Environmental sample (dairy industry)	Spiking protocol, heat treatment 56°C 8 min	1.20	0-4-3-5-2	2.8	+	2	b
2023	560	Poussières d'aspirateur n°11-12 (industrie de produits laitiers non probiotiques)	Vacuum cleaner dust n°11-12 (non-probiotic dairy industry)	C. sakazakii Ad2360 (+ S. Anatum A00E007)	Environmental sample (dairy industry)/ Environmental sample (dairy industry)	Spiking protocol, heat treatment 56°C 8 min	0.9/1.2	4-1-2-0-1/ 1-0-0-0-0	1.6/0.2	+	2	b

Year of analysis	Sample N°	Product	Product (French name)	Artificial contamination						Global result	Category	Type
				Strain	Origin	Injury protocol	Injury measurement	Inoculation level/ sample		Cronobacter spp		
								Enumeration	Mean			
2023	561	Poussières d'aspirateur n°13-14 (industrie de produits laitiers non probiotiques)	Vacuum cleaner dust n°13-14 (non-probiotic dairy industry)	C. sakazakii Ad2386 (+ S. Livingstone A00E058)	Environmental sample (dairy industry)/ Environmental sample (dairy industry)	Spiking protocol, heat treatment 56°C 8 min	0.60/1.60	2-1-2-1-1/ 0-1-1-1-2	1.0/1.0	+	2	b
2023	562	Poussières d'aspirateur n°15-16 (industrie de produits laitiers non probiotiques)	Vacuum cleaner dust n°15-16 (non-probiotic dairy industry)	C. sakazakii Ad2387 (+ S. Derby A00E084)	Environmental sample (dairy industry)/ Environmental sample (dairy industry)	Spiking protocol, heat treatment 56°C 8 min	1.10/1.20	0-0-2-2-3/ 0-4-3-5-2	1.4/2.8	+	2	b
2023	1363	Poussières d'aspirateur 24 (industrie de produits laitiers)	Vacuum cleaner dust 24 (dairy industry)	C. sakazakii Ad2408 (+ S. Brandenburg Ad2151)	Environmental sample (dairy product industry)	Spiking protocol, heat treatment 56°C 8 min	1.50/2.5	2-2-1-0-1/ 2-0-3-1-3	1.2/1.8	+	2	b
2023	1364	Poussières d'aspirateur 22 (industrie de produits laitiers)	Vacuum cleaner dust 22 (dairy industry)	C. sakazakii Ad2408 (+ S. Brandenburg Ad2151)	Environmental sample (dairy product industry)	Spiking protocol, heat treatment 56°C 8 min	1.50/2.5	2-2-1-0-1/ 2-0-3-1-3	1.2/1.8	+	2	b
2023	1365	Poussières d'aspirateur A32 (industrie de produits laitiers)	A32 Vacuum Cleaner Dust (Dairy Industry)	C. sakazakii Ad2408 (+ S. Brandenburg Ad2151)	Environmental sample (dairy product industry)	Spiking protocol, heat treatment 56°C 8 min	1.50/2.5	2-2-1-0-1/ 2-0-3-1-3	1.2/1.8	-	2	b
2023	1366	Poussières d'aspirateur 29 (industrie de produits laitiers)	Vacuum cleaner dust 29 (dairy industry)	C. sakazakii Ad2401 (+ S. Manhattan 900	Environmental sample (dairy product industry)	Spiking protocol, heat treatment 56°C 8 min	1.20/1.30	2-1-3-1-1/ 4-2-4-2-1	1.6/2.6	+	2	b
2023	1367	Poussières d'aspirateur 12 (industrie de produits laitiers)	Vacuum cleaner dust 12 (dairy industry)	C. sakazakii Ad2401 (+ S. Manhattan 900)	Environmental sample (dairy product industry)	Spiking protocol, heat treatment 56°C 8 min	1.20/1.30	2-1-3-1-1/ 4-2-4-2-1	1.6/2.6	+	2	b
2023	1370	Poussières d'aspirateur P102+16 (industrie de produits laitiers)	Vacuum cleaner dust P102+16 (dairy industry)	C. sakazakii Ad2391	Environmental sample (dairy product industry)	Spiking protocol, heat treatment 56°C 8 min	1.80	0-3-1-1-4	1.8	+	2	b
2023	1371	Poussières d'aspirateur 27 (industrie de produits laitiers)	Vacuum cleaner dust 27 (dairy industry)	C. sakazakii Ad2391	Environmental sample (dairy product industry)	Spiking protocol, heat treatment 56°C 8 min	1.80	0-3-1-1-4	1.8	+	2	b

Appendix 4 – Sensitivity study: raw data

ne: new extraction

no amp : no amplification curves. To be considered as negative results

uncertain : interpretation given by the LC 480 II software. To be considered as negative results

BPW

BPW + vancomycin + Tween 80 (Alt) or prewarmed BPW 2T + Tween 80 (ref)

BPW (37°C) + Tween 80 (10g/L)

BPW (37°C) + α -amylase (1%)

Foodproof StarPrep Three Kit

Foodproof StarPrep Three 8-Strip Kit

INFANT FORMULA WITH OR WITHOUT PROBIOTICS AND RELATED INGREDIENTS																															
Year of analysis	Sample N°	Product (	ISO 22964*			Alternative method: foodproof® <i>Salmonella</i> Plus <i>Cronobacter</i> Detection LyoKit - 5'Nuclease-																						Category	Type		
						Pre-warmed BPW or Prewarmed BPW + vancomycin (10mg/L) or BPW- 16h-37°C																									
						PCR result												Confirmation			Final result <i>Cronobacter</i> spp.				Agreement						
			foodproof® extraction						BAX lysis						CSB	ISO 22964	All confirmation tests	foodproof® <i>Cronobacter</i> spp.													
			BAX Q7			LC480 II			BAX Q7			LC480 II			CCI			BAX Q7	LC 480 II	BAX Q7	LC 480 II	foodproof® extraction	BAX lysis								
			Ct HEX (target)	Ct ROX (IC)	Result	Ct HEX (target)	Ct ROX (IC)	Result	Ct HEX (target)	Ct ROX (IC)	Result.	Ct HEX (target)	Ct ROX (IC)	Result																	
2022	3568	Infant milk powder, 6-12 months, with probiotics (B. lactis 1.8 x10 ⁶ CFU/g) (26%MG)	st	/	-	39.1/ 0.0/ 0.0	37.3/ 0.0/ 37.4	+/-/-	35.43/ 0.0/ 0.0	33.43/ 32.70/ 31.86	+/-/-	0.0	37.9	-	37.69/ 36.34/ 0.0	32.89/ 32.70/ 31.76	+/-/-	st	/	-	-	-	-	-	PD _{FP(alt)}	PD _{FP(alt)}	NA	PD _{FP(alt)}	1	a	
2023	766	Infant milk powder with probiotics, 3rd age (L. reutei DSM 17938 8.24 x 10 ⁴ CFU/g) (23.4% fat)	-	/	-	36.8/ 0.0/ 0.0	37.3/ 38.1/ 38.5	+/-/-	33.66/ 0.0/ 32.25	31.82/ 32.03/ 32.02	+/-/+	0.0	38.0	-	0.0	32.73	-	-	/	-	-	-	-	-	PD _{FP(alt)}	PD _{FP(alt)}	NA	NA	1	a	
2022	3567	Infant milk powder, 6-12 months, with probiotics (B. infantis 3.2 x 10 ⁷ CFU/g) (23%MG)	st	/	-	22.9	0.0	+	20.6	0.0	+	23.7	0.0	+	19.35	0	+	+	+	+	+	+	+	+	PD	PD	PD	PD	1	a	
2022	3797	Infant milk powder, 3rd age, with probiotics (B. lactis + S. thermophilus 1.5 x 10 ⁶ CFU/g) (30% fat)	-	/	-	22.01	0.0	+	18.31	0.0	+	26.1	0.0	+	21.72	0	+	+1/2	+	+	+	+	+	+	PD	PD	PD	PD	1	a	
2023	763	Infant milk powder with probiotics, 3rd age (L. reutei DSM 17938 8.55 x10 ⁴ CFU/g) (26% fat)	st	/	-	24.4	0.0	+	22.27	0.0	+	27.4	0.0	+	23.49	0	+	+p	+	+	+	+	+	+	PD	PD	PD	PD	1	a	
2022	3561	Infant milk powder, 6-12 months, with probiotics (B. infantis 2.5 x 10 ⁶ CFU/g) (26% fat)	-	/	-	0.0	38.4	-	0.0	0.0	-	0.0	37.9	-	0.0	0	-	-	/	-	-	-	-	-	NA	NA	NA	NA	1	a	
2022	3562	Infant milk powder, 6-12 months, with probiotics (B. infantis 3.2 x 10 ⁷ CFU/g) (23%MG)	-	/	-	0.0	37.6	-	0.0	32.99	-	0.0	38.6	-	0.0	33.02	-	st	/	-	-	-	-	-	NA	NA	NA	NA	1	a	
2022	3563	Infant milk powder, 6-12 months, with probiotics (B. lactis 1.8 x 10 ⁶ CFU/g) (26%MG)	-	/	-	0.0	38.9	-	0.0	0	-	0.0	38.1	-	0.0	32.15	-	-	/	-	-	-	-	-	NA	NA	NA	NA	1	a	
2022	3564	Infant milk powder, from 6 months, with probiotics (B. lactis 3.5 x 10 ⁷ CFU/g) (26% fat)	-	/	-	0.0	37.9	-	35.06/ 0.0/ 0.0/ 0*	33.20/ 0.0/ 0.0/ 0	+/- /-/-*	39.6/ 0.0/ 0.0	37.3/ 43.7/ 42.5	+/-/-	0.0	37.04	-	-	/	-	-	-	-	-	NA	PD _{FP(alt)}	PD _{FP(alt)}	NA	1	a	
2022	3565	Infant milk powder, babies, with probiotics (B. lactis 3.5 x 10 ⁷ CFU/g) (23.6% fat)	-	/	-	0.0	37.7	-	0.0	33.53	-	0.0	38.4	-	0.0	33.13	-	-	/	-	-	-	-	-	NA	NA	NA	NA	1	a	
2022	3566	Infant milk powder, 6-12 months, follow-on milk, with probiotics (B. infantis 2.5 x 10 ⁶ CFU/g) (26% fat)	+	+	+	25.8	0.0	+	20.63	0	+	22.0	0.0	+	15.9	0	+	+	+	+	+	+	+	+	PA	PA	PA	PA	1	a	

* Analyses performed according to the COFRAC accreditation (Accreditation Testing n°1-0144, scope available on www.cofrac.fr)

INFANT FORMULA WITH OR WITHOUT PROBIOTICS AND RELATED INGREDIENTS																														
Year of analysis	Sample N°	Product (	ISO 22964*			Alternative method: foodproof® <i>Salmonella</i> Plus <i>Cronobacter</i> Detection LyoKit - 5'Nuclease-																							Category	Type
						Pre-warmed BPW or Prewarmed BPW + vancomycin (10mg/L) or BPW- 16h-37°C																								
						PCR result												Confirmation			Final result <i>Cronobacter</i> spp.				Agreement					
			CSB	Identifi- cation	Result	foodproof® extraction						BAX lysis						CSB	ISO 22964	All confirmation tests	foodproof® extraction		BAX lysis		foodproof® extraction		BAX lysis			
						BAX Q7			LC480 II			BAX Q7			LC480 II						CCI	BAX Q7	LC 480 II	BAX Q7	LC 480 II	BAX Q7	LC 480 II			
			Ct HEX (target)			Ct ROX (IC)	Result	Ct HEX (target)	Ct ROX (IC)	Result	Ct HEX (target)	Ct ROX (IC)	Result.	Ct HEX (target)	Ct ROX (IC)	Result	BAX Q7	LC 480 II										BAX Q7		
2023	765	Infant milk powder with probiotics, after from 10 months (L. reutei DSM 17938 1.55 x 10 ⁵ CFU/g) (23% fat)	st			/	-	34.1	36.1	+	31.38	32.42	+	35.6	37.6	+	32.06	32.82			+	+p	+	+	+	+	+	+	PD	PD
2022	3569	Infant milk powder, from 6 months, with probiotics (B. lactis 3.5 x 10 ⁷ CFU/g) (26% fat)	+	+	+	18.3/19.7	0.0/0.0	+	15.62	0	+	18.4	0.0	+	14.67	0	+	+	+	+	+	+	+	+	PA	PA	PA	PA	1	a
2022	3570	Infant milk powder, babies, with probiotics (B. lactis 3.5 x 10 ⁷ CFU/g) (23.6% fat)	+	+	+	0.0/20.0/21.3*	0.0/0.0*	i/+*	17.1	0	+	19.6	0.0	+	14.65	0	+	+	+	+	+	+	+	+	PA	PA	PA	PA	1	a
2023	769	Infant milk powder with probiotics, organic from 6 months (L. reutei DSM 17938 2.45 x 10 ⁶ CFU/g) (26% fat)	st	/	-	30.3	0.0	+	29.67	31.55	+	29.6	35.3	+	26.02	33.19	+	+p	+	+	+	+	+	+	PD	PD	PD	PD	1	a
2023	771	Infant milk powder with probiotics, 1st age for infant 0-6 months (L. fermentum hereditum 5.09 x 10 ⁶ CFU/g) (24% fat)	st	/	-	33.6	36.2	+	29.72	30.12	+	32.7	37.7	+	30.04	32.41	+	+p	+	+	+	+	+	+	PD	PD	PD	PD	1	a
2022	3798	Infant milk powder, 3rd age, with probiotics (B. lactis + S. thermophilus 1.5 x 10 ⁶ CFU/g) (30% fat)	+1/2	+	+	19.1	0.0	+	16.57	0.0	+	21.7	0.0	+	18.59	0	+	+1/2	+	+	+	+	+	+	PA	PA	PA	PA	1	a
2022	3799	Infant milk powder, 2nd age, with probiotics (B. lactis + S. thermophilus 3.1 x 10 ⁷ CFU/g) (23% fat)	+1/2	+	+	20.1	0.0	+	17.64	0.0	+	23.1	0.0	+	19.36	0	+	+1/2	+	+	+	+	+	+	PA	PA	PA	PA	1	a
2022	3800	Infant milk powder, 2nd age, with probiotics (B. lactis + S. thermophilus 3.1 x 10 ⁷ CFU/g) (23% fat)	+1/2	+	+	20.7	0.0	+	17	0.0	+	21.6	0.0	+	17.82	0	+	+1/2	+	+	+	+	+	+	PA	PA	PA	PA	1	a
2022	3801	Infant milk powder, 1st age, with probiotics (B. lactis 8.6 x 10 ⁵ CFU/g) (26% fat)	+1/2	+	+	25.7	0.0	+	21.36	0.0	+	27.5	0.0	+	23.89	0	+	+1/2	+	+	+	+	+	+	PA	PA	PA	PA	1	a
2023	762	Infant milk powder with probiotics, 3rd age (L. reutei DSM 17938 8.55 x 10 ⁴ CFU/g) (26% fat)	-	/	-	0.0	38.3	-	0.0	45	-	0.0	38.3	-	0.0	32.67	-	-	/	-	-	-	-	-	NA	NA	NA	NA	1	a
2023	764	Infant milk powder with probiotics, after 10 months (L. reutei DSM 17938 1.55 x 10 ⁵ CFU/g) (23% fat)	st	/	-	0.0	36.4	-	0.0	30.2	-	0.0	37.7	-	0.0	31.1	-	-	/	-	-	-	-	-	NA	NA	NA	NA	1	a

INFANT FORMULA WITH OR WITHOUT PROBIOTICS AND RELATED INGREDIENTS																														
Year of analysis	Sample N°	Product (	ISO 22964*			Alternative method: foodproof® <i>Salmonella</i> Plus <i>Cronobacter</i> Detection LyoKit - 5'Nuclease-																							Category	Type
						Pre-warmed BPW or Prewarmed BPW + vancomycin (10mg/L) or BPW- 16h-37°C																								
						PCR result												Confirmation			Final result <i>Cronobacter</i> spp.				Agreement					
			foodproof® extraction			BAX lysis						CSB	ISO 22964	All confirmation tests	foodproof® extraction		BAX lysis		foodproof® extraction		BAX lysis									
						BAX Q7			LC480 II			BAX Q7			LC480 II			CCI	BAX Q7	LC 480 II	BAX Q7	LC 480 II	BAX Q7	LC 480 II						
			Ct HEX (target)	Ct ROX (IC)	Result	Ct HEX (target)	Ct ROX (IC)	Result	Ct HEX (target)	Ct ROX (IC)	Result.	Ct HEX (target)			Ct ROX (IC)	Result	Ct HEX (target)								Ct ROX (IC)	Result	BAX Q7	LC 480 II		
2023	767	Infant milk powder with probiotics, 3rd age from 1 year (L. reutei DSM 17938 8.24 x 10 ⁴ CFU/g) (23.4% fat)	+	+	+	18.1	0.0	+	15.89	0.0	+	20.6	0.0	+	17.16	0	+	+p	+	+	+	+	+	+	PA	PA	PA	PA	1	a
2023	768	Infant milk powder with probiotics, organic from 6 months (L. reutei DSM 17938 2.45 x 10 ⁶ CFU/g) (26% fat)	-	/	-	0.0	37.1	-	0.0	33.05	-	0.0	37.7	-	0.0	33.08	-	st	/	-	-	-	-	-	NA	NA	NA	NA	1	a
2023	770	Infant milk powder with probiotics, 1st age for infant 0-6 months (L. fermentum hereditum 5.09 x 10 ⁶ CFU/g) (24% fat)	st	/	-	0.0	37.7	-	0.0	32.86	-	0.0	38.4	-	0.0	32.85	-	-	/	-	-	-	-	-	NA	NA	NA	NA	1	a
2023	772	Infant milk powder with probiotics, formula thickened from 6 months (L. fermentum hereditum 3.09 x 10 ⁶ CFU/g) (26% fat)	st	/	-	0.0	36.8	-	0.0	32.62	-	0.0	38.3	-	0.0	32.23	-	-	/	-	-	-	-	-	NA	NA	NA	NA	1	a
2022	2903	Infant formula, 0-6 months (23.5% FL)	-	/	-	0.0	37.2	-	0.0	33.11	-	00.0	38.3	-	0.0	35.35	-	-	/	-	-	-	-	-	NA	NA	NA	NA	1	b
2022	2904	Infant formula, 6-12 months (22.6% FL)	-	/	-	0.0	37.6	-	0.0	33.08	-	00.0	37.4	-	0.0	41.36	-	-	/	-	-	-	-	-	NA	NA	NA	NA	1	b
2022	2907	Infant formula, stage 2,from 6 months (22.0% FL)	-	/	-	0.0	39.0	-	0.0	0.0	-	37.6/ 39.8/ 38.8/ 0.0	37.4/ 38.2/ 37.6/ 41.9	+/-/-	33.04 (no amp)	32.8	-	-	/	-	-	-	-	-	NA	NA	PDF _{FP(alt)}	NA	1	b
2022	2908	Infant formula, stage 2, 6-12 months (22.0% FL)	-	/	-	0.0	38.0	-	0.0	32.91	-	0.0	37.6	-	0.0	32.68	-	-	/	-	-	-	-	-	NA	NA	NA	NA	1	b
2022	2909	Infant formula, stage 2, 6months-1year (23.6% FL)	-	/	-	0.0	37.3	-	0.0	33.12	-	0.0	37.9	-	0.0	39.02	-	-	/	-	-	-	-	-	NA	NA	NA	NA	1	b
2022	2910	Infant formula, stage 2, 6-12 months (21.1% FL)	-	/	-	0.0	37.4	-	0.0	33.6	-	37.7/ 0.0/ 38.2/ 0.0	37.7/ 38.4/ 38.3/ 41.5	+/- /+/-	0.0	35.9	-	-	/	-	-	-	-	-	NA	NA	PDF _{FP(alt)}	NA	1	b
2022	2963	Infant formula, stage 1, 6-12 months (22.0% FL)	+p	+	+	22.6	0.0	+	20.26	0	+	23.1	0.0	+	21.94	0	+	+p	+	+	+	+	+	+	PA	PA	PA	PA	1	b
2022	2964	Organic infant formula, 0-6 months (24.0% FL)	+p	+	+	26.5	0.0	+	24.09	24.21 (no amp)	+	26.3	0.0	+	24.92	26.36	+	+p	+	+	+	+	+	+	PA	PA	PA	PA	1	b
2022	2965	Organic infant formula, 0-6 months (24.0% FL)	+p	+	+	21.9	0.0	+	20.21	0	+	22.5	0.0	+	19.71	0	+	+p	+	+	+	+	+	+	PA	PA	PA	PA	1	b
2022	2966	Infant formula, 0-6 months (22.0% FL)	+p	+	+	22.5	0.0	+	20.23	21.5 (no amp)	+	23.0	0.0	+	19.66	0	+	+p	+	+	+	+	+	+	PA	PA	PA	PA	1	b
2022	2967	Infant formula, 0-6 months (21.0% FL)	+p	+	+	17.9	0.0	+	15.52	0	+	18.0	0.0	+	14.17	0	+	+p	+	+	+	+	+	+	PA	PA	PA	PA	1	b

INFANT FORMULA WITH OR WITHOUT PROBIOTICS AND RELATED INGREDIENTS																															
Year of analysis	Sample N°	Product (	ISO 22964*			Alternative method: foodproof® <i>Salmonella</i> Plus <i>Cronobacter</i> Detection LyoKit - 5'Nuclease-																								Category	Type
						Pre-warmed BPW or Prewarmed BPW + vancomycin (10mg/L) or BPW- 16h-37°C																									
						PCR result												Confirmation			Final result <i>Cronobacter</i> spp.				Agreement						
			foodproof® extraction			BAX lysis						CSB	ISO 22964	All confirmation tests	foodproof® <i>Cronobacter</i> spp.		Agreement														
			BAX Q7			LC480 II			BAX Q7			LC480 II			CCI	BAX Q7	LC 480 II	BAX Q7	LC 480 II	foodproof® extraction	BAX lysis										
			Ct HEX (target)	Ct ROX (IC)	Result	Ct HEX (target)	Ct ROX (IC)	Result	Ct HEX (target)	Ct ROX (IC)	Result.	Ct HEX (target)										Ct ROX (IC)	Result								
			2022	2968	Infant formula, stage 2, 6-12 months (26.0% FL)	+p	+	+	25.8	0.0	+	23.31	0	+	25.8	0.0	+	22.65	0	+	+p	+	+	+	+	+	+	PA	PA		
2022	2969	Organic infant formula, from 6 months (25.0%FL)	+p	+	+	27.9	0.0	+	25.26	27.34 (no amp)	+	27.7	0.0	+	24.62	25.85 (no amp)	+	+p	+	+	+	+	+	+	PA	PA	PA	PA	1	b	
2022	2970	Organic infant formula, 6-12 months (22.0%FL)	+p	+	+	21.8	0.0	+	20.24	0	+	20.4	0.0	+	17.1	0	+	+p	+	+	+	+	+	+	PA	PA	PA	PA	1	b	
2022	3023	Infant formula anti-regurgitation, 0-6 months (25.0% FL)	+m	+	+	28.7	0.0	+	24.09	28.71	+	30.0	35.0	+	32.51	43.02	+	+m	+	+	+	+	+	+	PA	PA	PA	PA	1	b	
2022	3024	Organic infant formula, stage 2, 6-12 months (25.0%FL)	+m	+	+	27.9	0.0	+	24.15	28.66	+	28.3	0.0	+	28.04	28.88	+	+mni/+	+	+	+	+	+	+	PA	PA	PA	PA	1	b	
2022	3025	Infant milk powder, gourmet babies, 6-12 months (26%fat)	+m	+	+	25.5	0.0	+	21.48	0	+	27.1	0.0	+	21.2	0	+	+m	+	+	+	+	+	+	PA	PA	PA	PA	1	b	
2022	3026	Infant milk powder, 6-12 months (25.5% fat)	+m	+	+	26.4	0.0	+	23.08	27.74 (no amp)	+	27.2	0.0	+	23.71	0	+	+1/2	+	+	+	+	+	+	PA	PA	PA	PA	1	b	
2022	3027	Organic infant milk powder, 2nd age (25%fat)	+m	+	+	28.4	0.0	+	24.65	24.45 (no amp)	+	29.6	0.0	+	26.55	27.21	+	+M	+	+	+	+	+	+	PA	PA	PA	PA	1	b	
2022	3028	Premium infant milk powder, 0-6 months (24.7% fat)	+m	+	+	35.0	37.9	+	30.69	31.96	+	33.0	37.0	+	33.69	31.46	+	+M	+	+	+	+	+	+	PA	PA	PA	PA	1	b	
2022	2905	Organic infant formula, 6-12 months (23.1% FL)	-	/	-	39.2/ 0.0/ 0.0	37.3/ 37.4/ 0.0	+/-/-	0.0	33.17	-	0.0	37.2	-	0.0	39.27	-	-	/	-	-	-	-	-	PDFP(alt)	NA	NA	NA	1	b	
2022	2906	Infant formula, stage 2, 6-12 months (26.0% FL)	-	/	-	39.9/ 0.0/ 0.0ne	37.8/ 40.2/ 40.8	+/-/- ne	38.5 (no amp)	33.02	-	0.0	37.6	-	0.0	34.5	-	-	/	-	-	-	-	-	PDFP(alt)	NA	NA	NA	1	b	
2022	3083	Rice starch	st	/	-	0.0	36.8	-	0.0	33.3	-	0.0	37.2	-	0.0	35.68	-	-	/	-	-	-	-	-	NA	NA	NA	NA	1	c	
2022	3084	Corn starch	st	/	-	0.0	42.8	-	0.0	33.03	-	0.0	38.8	-	0.0	32.4	-	-	/	-	-	-	-	-	NA	NA	NA	NA	1	c	
2022	3085	Maltodextrin	st	/	-	0.0	37.2	-	0.0	33.01	-	0.0	37.0	-	0.0	36.2	-	-	/	-	-	-	-	-	NA	NA	NA	NA	1	c	
2022	3086	Whey	-	/	-	0.0	37.6	-	0.0	32.82	-	0.0	37.4	-	0.0	32.81	-	-	/	-	-	-	-	-	NA	NA	NA	NA	1	c	
2022	3087	Whey	-	/	-	0.0	37.0	-	0.0	32.69	-	0.0	38.3	-	0.0	32.12	-	-	/	-	-	-	-	-	NA	NA	NA	NA	1	c	
2022	3088	Maltodextrin	-	/	-	0.0	37.3	-	0.0	32.95	-	0.0	37.1	-	0.0	38.66	-	-	/	-	-	-	-	-	NA	NA	NA	NA	1	c	
2022	3091	Maltodextrin	+p	+	+	16.5	0.0	+	13.98	0	+	16.9	0.0	+	15.27	0	+	+p	+	+	+	+	+	+	PA	PA	PA	PA	1	c	
2022	3092	Whey	+p	+	+	35.7	36.6	+	31.72	32.52	+	31.9	37.4	+	30.11	32.49	+	+p	+	+	+	+	+	+	PA	PA	PA	PA	1	c	
2022	3093	Whey	+p	+	+	35.7	36.3	+	30.79	32.33	+	33.6	37.2	+	31.61	32.78	+	+p	+	+	+	+	+	+	PA	PA	PA	PA	1	c	
2022	3175	Lactose	+M	+	+	23.2	0.0	+	20.13	0	+	28.8	0.0	+	21.89	21.09 (no amp)	+	+1/2	+	+	+	+	+	+	PA	PA	PA	PA	1	c	
2022	3176	Lactose	+M	+(API 20E)	+	19.1	0.0	+	16.68	0	+	21.4	0.0	+	17.85	18.66	+	+1/2	+(API 20E)	+	+	+	+	+	+	PA	PA	PA	PA	1	c
2022	3177	Whey	+1/2	+	+	19.7	0.0	+	17.48	0	+	21.6	0.0	+	18.75	19.55	+	+1/2	+	+	+	+	+	+	PA	PA	PA	PA	1	c	
2022	3178	Wheat starch	+M	+	+	20.5	0.0	+	18.09	0	+	20.2	0.0	+	17.92	0	+	+M	+	+	+	+	+	+	PA	PA	PA	PA	1	c	
2022	3179	Maltodextrin	+1/2	+(API 20E)	+	17.9	0.0	+	16.35	0	+	19.7	0.0	+	16.16	0	+	+1/2	+(API 20E)	+	+	+	+	+	+	PA	PA	PA	PA	1	c

INFANT FORMULA WITH OR WITHOUT PROBIOTICS AND RELATED INGREDIENTS																																		
Year of analysis	Sample N°	Product (	ISO 22964*			Alternative method: foodproof® <i>Salmonella</i> Plus <i>Cronobacter</i> Detection LyoKit - 5'Nuclease-																						Category	Type					
						Pre-warmed BPW or Prewarmed BPW + vancomycin (10mg/L) or BPW- 16h-37°C																												
						PCR result												Confirmation			Final result <i>Cronobacter</i> spp.				Agreement									
			CSB	Identifi- cation	Result	foodproof® extraction						BAX lysis						CSB	ISO 22964	All confirmation tests	foodproof® extraction		BAX lysis		foodproof® extraction		BAX lysis							
						BAX Q7			LC480 II			BAX Q7			LC480 II			CCI			BAX Q7	LC 480 II	BAX Q7	LC 480 II	BAX Q7	LC 480 II								
			Ct HEX (target)			Ct ROX (IC)	Result	Ct HEX (target)	Ct ROX (IC)	Result	Ct HEX (target)	Ct ROX (IC)	Result.	Ct HEX (target)	Ct ROX (IC)	Result	BAX Q7										LC 480 II			BAX Q7	LC 480 II	BAX Q7	LC 480 II	BAX Q7
			2022			3180	Whey Protein Concentrate	+m	+	+	20.8	0.0	+	18.6	0	+	23.0	0.0			+	19.00	0	+	+m	+	+			+	+	+	+	PA
2022	3732	Milk Protein	st			/	-	0.0	36.3	-	0.0	32.97	-	0.0	38.5	-	0.0	33.31			-	st	/	-	-	-	-	-	NA	NA	NA	NA	1	c
2022	3733	Lactose	st	/	-	0.0	36.6	-	0.0	32.92	-	0.0	38.4	-	0.0	32.85	-	st	/	-	-	-	-	-	NA	NA	NA	NA	1	c				
2022	3734	Maltodextrin	st	/	-	0.0	36.4	-	0.0	32.46	-	0.0	38.2	-	0.0	33.03	-	st	/	-	-	-	-	-	NA	NA	NA	NA	1	c				
2022	3735	Whey	st	/	-	0.0	36.5	-	0.0	33.08	-	0.0	38.5	-	0.0	33.15	-	st	/	-	-	-	-	-	NA	NA	NA	NA	1	c				
2022	3089	Rice starch	st	/	-	23.0	0.0	+	19.02	0	+	22.8	0.0	+	21.13	0	+	+p	+	+	+	+	+	+	PD	PD	PD	PD	1	c				
2022	3090	Corn starch	st	/	-	33.1	34.7	+	30.03	31.62	+	32.8	37.0	+	29.79	32.36	+	+p	+	+	+	+	+	+	PD	PD	PD	PD	1	c				
2022	3094	Maltodextrin	st	/	-	21.1	0.0	+	18.87	19.00 (no amp)	+	16.8	0.0	+	14.99	0	+	+p	+	+	+	+	+	+	PD	PD	PD	PD	1	c				

PRODUCTION ENVIRONMENTAL SAMPLES																														
Year of analysis	Sample N°	Product (French name)	ISO 22964*			Alternative method: foodproof® <i>Salmonella</i> Plus <i>Cronobacter</i> Detection LyoKit - 5'Nuclease-																							Category	Type
						Pre-warmed BPW or Prewarmed BPW + vancomycin (10mg/L) or BPW- 16h-37°C																								
						PCR result											Confirmation			Final result <i>Cronobacter</i> spp.				Agreement						
			foodproof® extraction						BAX lysis					CSB	ISO 22964	All confirmation tests	foodproof® <i>Cronobacter</i> spp.		Agreement											
			BAX Q7			LC480 II			BAX Q7			LC480 II		CCI			BAX Q7	LC 480 II	BAX Q7	LC 480 II										
			Ct HEX (target)	Ct ROX (IC)	Result	Ct HEX (target)	Ct ROX (IC)	Result	Ct HEX (target)	Ct ROX (IC)	Result <i>Crono</i> spp.	Ct HEX (target)	Ct ROX (IC)								Result									
2022	3396	Floor surface swabs before cleaning (dairy industry)	st	/	-	0.0	37.0	-	0.0	33.33	-	0.0	38.2	-	0.0	33.1	-	st (stx4 + - direct streaking)	/	-	-	-	-	-	NA	NA	NA	NA	2	a
2022	3397	Floor surface swabs before cleaning (dairy industry)	-	/	-	0.0	37.8	-	0.0	33.1	-	0.0	37.4	-	0.0	34.36	-	-	/	-	-	-	-	-	NA	NA	NA	NA	2	a
2022	3398	Floor surface swabs before cleaning (dairy industry)	-	/	-	0.0	37.7	-	0.0	32.91	-	0.0	38.1	-	0.0	31.92	-	-	/	-	-	-	-	-	NA	NA	NA	NA	2	a
2022	3399	Floor surface swabs before cleaning (dairy industry)	+p	+	+	16.5	0.0	+	11.98	0	+	17.7	0.0	+	16.55	0.0	+	+p	+	+	+	+	+	+	PA	PA	PA	PA	2	a
2022	3400	Floor surface swabs before cleaning (dairy industry)	+p	+	+	16.3	0.0	+	11.69	0	+	17.4	0.0	+	16.03	0	+	+p	+	+	+	+	+	+	PA	PA	PA	PA	2	a
2022	3401	Floor surface swabs before cleaning (dairy industry)	+p	+	+	16.1	0.0	+	13.47	0	+	18.0	0.0	+	16.3	0.0	+	+p	+	+	+	+	+	+	PA	PA	PA	PA	2	a
2022	3402	Floor surface swabs before cleaning (dairy industry)	+p	+	+	16.4	0.0	+	14.76	0	+	18.4	0.0	+	16.55	0	+	+p	+	+	+	+	+	+	PA	PA	PA	PA	2	a
2022	3403	Floor surface swabs before cleaning (dairy industry)	-	/	-	0.0	38.3	-	0.0	33.36	-	0.0	37.4	-	0.0	37.23	-	-	/	-	-	-	-	-	NA	NA	NA	NA	2	a
2022	3404	Floor surface swabs before cleaning (dairy industry)	st	/	-	0.0	36.6	-	0.0	33.1	-	0.0	38.5	-	0.0	33.2	-	st	/	-	-	-	-	-	NA	NA	NA	NA	2	a
2022	3405	Floor surface swabs before cleaning (dairy industry)	st	/	-	0.0	37.1	-	0.0	33.29	-	0.0	38.3	-	0.0	32.79	-	st	/	-	-	-	-	-	NA	NA	NA	NA	2	a
2022	3406	Floor surface swabs before cleaning (dairy industry)	st	/	-	0.0	36.0	-	0.0	33.37	-	0.0	38.1	-	0.0	32.95	-	st	/	-	-	-	-	-	NA	NA	NA	NA	2	a
2022	3407	Floor surface swabs before cleaning (dairy industry)	+p	+	+	0.0 / 22.7/ 25.7*	0.0/ 0.0/ 0.0	i/+/*	20.65	22.50 (no amp)	+	17.1	0.0	+	15.59	0	+	+p	+	+	+	+	+	+	PA	PA	PA	PA	2	a
2022	3408	Floor surface swabs before cleaning (dairy industry)	+p	+	+	16.1	0.0	+	15.19	0	+	20.1	0.0	+	16.86	0	+	+p	+	+	+	+	+	+	PA	PA	PA	PA	2	a
2022	3409	Floor surface swabs before cleaning (dairy industry)	+p	+	+	16.5	0.0	+	14.91	0	+	17.6	0.0	+	15.79	0	+	+p	+	+	+	+	+	+	PA	PA	PA	PA	2	a
2022	3410	Floor surface swabs before cleaning (dairy industry)	+p	+	+	16.2	0.0	+	14.88	15.19 (no amp)	+	18.2	0.0	+	13.99	0	+	+p	+	+	+	+	+	+	PA	PA	PA	PA	2	a
2022	3475	Swipe before cleaning (ice cream production)	-	/	-	0.0	37.5	-	0.0	39.83	-	0.0	38.1	-	0.0	31.99	-	-	/	-	-	-	-	-	NA	NA	NA	NA	2	a
2022	3476	Swipe before cleaning (ice cream production)	+1/2	+	+	20.5	0.0	+	17.99	0	+	21.9	0.0	+	18.4	0	+	+1/2	+	+	+	+	+	+	PA	PA	PA	PA	2	a
2022	3477	Swipe after cleaning (ice cream production)	+1/2	+	+	19.0	0.0	+	16.27	0	+	22.4	0.0	+	17.25	0	+	+1/2	+	+	+	+	+	+	PA	PA	PA	PA	2	a
2022	3478	Swipe before cleaning (Ice Cream Production)	-	/	-	0.0	37.5	-	0.0	40.75	-	0.0	37.3	-	0.0	33.53	-	-	/	-	-	-	-	-	NA	NA	NA	NA	2	a

* Analyses performed according to the COFRAC accreditation (Accreditation Testing n°1-0144, scope available on www.cofrac.fr)

PRODUCTION ENVIRONMENTAL SAMPLES																															
Year of analysis	Sample N°	Product (French name)	ISO 22964*			Alternative method: foodproof® <i>Salmonella</i> Plus <i>Cronobacter</i> Detection LyoKit - 5'Nuclease-																								Category	Type
						Pre-warmed BPW or Prewarmed BPW + vancomycin (10mg/L) or BPW- 16h-37°C																									
						PCR result												Confirmation			Final result <i>Cronobacter</i> spp.				Agreement						
			foodproof® extraction						BAX lysis						CSB	ISO 22964	All confirmation tests	foodproof® extraction		BAX lysis		foodproof® extraction		BAX lysis							
			BAX Q7			LC480 II			BAX Q7			LC480 II			CCI			BAX Q7	LC 480 II	BAX Q7	LC 480 II	BAX Q7	LC 480 II	BAX Q7	LC 480 II						
			Ct HEX (target)	Ct ROX (IC)	Result	Ct HEX (target)	Ct ROX (IC)	Result	Ct HEX (target)	Ct ROX (IC)	Result <i>Crono</i> spp.	Ct HEX (target)	Ct ROX (IC)	Result				BAX Q7	LC 480 II	BAX Q7	LC 480 II	BAX Q7	LC 480 II	BAX Q7	LC 480 II						
2022	3479	Swipe After Cleaning (Dairy Industry)	-	/	-	0.0	37.4	-	0.0	45.00	-	0.0	37.4	-	0.0	40.15	-	-	/	-	-	-	-	-	NA	NA	NA	NA	2	a	
2022	3480	Swipe After Cleaning (Dairy Industry)	+1/2	+	+	22.8	0.0	+	20.18	0	+	24.0	0.0	+	20.68	0	+	+1/2	+	+	+	+	+	+	PA	PA	PA	PA	2	a	
2022	3481	Swipe After Cleaning (Dairy Industry)	+M	+	+	20.8	0.0	+	18.5	19.46 (no amp)	+	29.2	0.0	+	21.64	0	+	+M	+	+	+	+	+	+	PA	PA	PA	PA	2	a	
2022	3637	Stainless steel bench swab, after cleaning (dairy industry)	-	/	-	0.0	37.2	-	0.0	32.92	-	0.0	37.3	-	0.0	41.67	-	-	/	-	-	-	-	-	NA	NA	NA	NA	2	a	
2022	3638	Vacuum nozzle swab, before cleaning (dairy industry)	-	/	-	0.0	36.5	-	0.0	33.01	-	0.0	37.2	-	0.0	37.07	-	-	/	-	-	-	-	-	NA	NA	NA	NA	2	a	
2022	3639	Swab before cleaning (dairy industry)	+	+	+	17.1	0.0	+	14.89	0	+	20.3	0.0	+	18.10	00.0	+	+	+	+	+	+	+	+	PA	PA	PA	PA	2	a	
2022	3640	Swab after cleaning (dairy industry)	+	+	+	17.0	0.0	+	14.21	0	+	19.6	0.0	+	16.63	0	+	+	+	+	+	+	+	+	PA	PA	PA	PA	2	a	
2022	3641	Swipe, before cleaning (dairy industry)	-	/	-	0.0	35.9	-	0.0	31.84	-	0.0	36.3	-	0.0	32.1	-	-	/	-	-	-	-	-	NA	NA	NA	NA	2	a	
2022	3642	Swipe, before cleaning (dairy industry)	-	/	-	0.0	37.2	-	0.0	33.08	-	0.0	37.4	-	0.0	38.72	-	-	/	-	-	-	-	-	NA	NA	NA	NA	2	a	
2022	3643	Swipe before cleaning (ice cream production)	-	/	-	0.0	37.1	-	0.0	32.97	-	0.0	37.5	-	0.0	33.67	-	-	/	-	-	-	-	-	NA	NA	NA	NA	2	a	
2022	3644	Swipe after cleaning (ice cream production)	+	+	+	16.9	0.0	+	14.13	0	+	19.3	0.0	+	16.58	0	+	+	+	+	+	+	+	+	PA	PA	PA	PA	2	a	
2022	3737	Vacuum cleaner dust P62 E1 E2 (dairy industry)	st	/	-	0.0	37.7	-	0.0	32.64	-	0.0	37.7	-	0.0	32.81	-	-	/	-	-	-	-	-	NA	NA	NA	NA	2	b	
2022	3738	Vacuum cleaner dust P43 A32 (dairy industry)	-	/	-	0.0	36.7	-	32.74/0.0/One	32.33/31.33/32.83	+/-ne	37.8/38.6/0.0	37.6/38.0/37.6	+/-+/-	0.0	32.36	-	-(-x5)	/	-	-	-	-	-	NA	PDFP(alt)	PDFP(alt)	NA	2	b	
2022	3741	Vacuum cleaner dust P51 2889 A12 (dairy industry)	+p	+	+	36.0	36.7	+	33.21	32.77	+	0.0/0.0/0.0	37.9/38.5/38.2	-/-/-	35.49	32.69	+	+1/2	+	+	+	+	-	+	PA	PA	NDFN(alt)	PA	2	b	
2022	3743	Vacuum cleaner dust P32 P41 (dairy industry)	st	/	-	0.0	36.3	-	0.0	32.66	-	0.0	36.6	-	0.0	32.25	-	-	/	-	-	-	-	-	NA	NA	NA	NA	2	b	
2022	3744	Vacuum cleaner dust E241 CE1 (dairy industry)	st	/	-	0.0	36.8	-	0.0	33.17	-	0.0	37.9	-	0.0	32.68	-	-	/	-	-	-	-	-	NA	NA	NA	NA	2	b	
2022	3745	Vacuum cleaner dust P62 (dairy industry)	st	/	-	0.0	37.5	-	0.0	33.5	-	0.0	38.7	-	0.0	32.66	-	-	/	-	-	-	-	-	NA	NA	NA	NA	2	b	
2022	3746	Vacuum cleaner dust P32 (dairy industry)	st	/	-	0.0	36.2	-	0.0	32.97	-	0.0	38.6	-	0.0	32.23	-	st	/	-	-	-	-	-	NA	NA	NA	NA	2	b	
2023	550	Vacuum cleaner dust n°1 (non-probiotic dairy industry)	-	/	-	0.0	37.4	-	0.0	42.17	-	0.0	37.1	-	0.0	35.68	-	-	/	-	-	-	-	-	NA	NA	NA	NA	2	b	
2023	551	Vacuum cleaner dust n°2 (non-probiotic dairy industry)	-	/	-	0.0	37.3	-	0.0	32.81	-	0.0	37.9	-	0.0	33.11	-	st	/	-	-	-	-	-	NA	NA	NA	NA	2	b	

PRODUCTION ENVIRONMENTAL SAMPLES																															
Year of analysis	Sample N°	Product (French name)	ISO 22964*			Alternative method: foodproof® <i>Salmonella</i> Plus <i>Cronobacter</i> Detection LyoKit - 5'Nuclease-																								Category	Type
						Pre-warmed BPW or Prewarmed BPW + vancomycin (10mg/L) or BPW- 16h-37°C																									
						PCR result												Confirmation			Final result <i>Cronobacter</i> spp.				Agreement						
			foodproof® extraction						BAX lysis						CSB	ISO 22964	All confirmation tests	foodproof® extraction		BAX lysis		foodproof® extraction		BAX lysis							
			BAX Q7			LC480 II			BAX Q7			LC480 II			CCI			BAX Q7	LC 480 II	BAX Q7	LC 480 II	BAX Q7	LC 480 II	BAX Q7	LC 480 II						
			Ct HEX (target)	Ct ROX (IC)	Result	Ct HEX (target)	Ct ROX (IC)	Result	Ct HEX (target)	Ct ROX (IC)	Result <i>Crono</i> spp.	Ct HEX (target)	Ct ROX (IC)	Result				BAX Q7	LC 480 II	BAX Q7	LC 480 II	BAX Q7	LC 480 II	BAX Q7	LC 480 II						
			2023	552	Vacuum cleaner dust n°3 (non-probiotic dairy industry)	-	/	-	0.0	38.3	-	0.0	31.96	-	0.0	38.5	-	0.0	32.39	-	-	/	-	-	-	-	-	NA	NA		
2023	553	Vacuum cleaner dust n°4 (non-probiotic dairy industry)	-	/	-	0.0	38.1	-	0.0	0.0	-	0.0	36.9	-	0.0	33.17	-	-	/	-	-	-	-	-	NA	NA	NA	NA	2	b	
2023	554	Vacuum cleaner dust n°5 (non-probiotic dairy industry)	-	/	-	0.0	37.7	-	0.0	45	-	0.0	38.0	-	0.0	32.41	-	-	/	-	-	-	-	-	NA	NA	NA	NA	2	b	
2023	555	Vacuum cleaner dust n°6 (non-probiotic dairy industry)	st	/	-	0.0	36.9	-	0.0	32.71	-	0.0	38.5	-	0.0	33.48	-	st	/	-	-	-	-	-	NA	NA	NA	NA	2	b	
2023	556	Vacuum cleaner dust n°7 (non-probiotic dairy industry)	st	/	-	0.0	36.4	-	0.0	30.71	-	0.0	37.6	-	0.0	32.94	-	st	/	-	-	-	-	-	NA	NA	NA	NA	2	b	
2023	557	Vacuum cleaner dust n°8 (non-probiotic dairy industry)	+	+	+	27.8	0.0	+	0.0/25.63*	0.0/26.43*	i/+*	29.9	0.0	+	25.91	35.25 (uncertain)	+	+p	+	+	+	+	+	+	PA	PA	PA	PA	2	b	
2023	558	Vacuum cleaner dust n°9 (non-probiotic dairy industry)	+	+	+	25.8	0.0	+	22.88	22.93 (no amp)	+	25.5	0.0	+	23.75	0	+	+p	+	+	+	+	+	+	PA	PA	PA	PA	2	b	
2023	559	Vacuum cleaner dust n°10 (non-probiotic dairy industry)	+	+	+	26.7	0.0	+	23.67	0.0	+	27.6	0.0	+	25.53	26.23 (no amp)	+	+p	+	+	+	+	+	+	PA	PA	PA	PA	2	b	
2023	561	Vacuum cleaner dust n°13-14 (non-probiotic dairy industry)	+	+	+	39.3	37.6	+	0.0/0.0/0.0	0.0/0.0/33.32	-/-/-	37.0	36.8	+	0.0/0.0/0.0	32.2/32.27/33.34	-/-/-	+M	+	+	+	-	+	-	PA	ND _{FN(alt)}	PA	ND _{FN(alt)}	2	b	
2023	562	Vacuum cleaner dust n°15-16 (non-probiotic dairy industry)	+	+	+	29.2	0.0	+	24.67	0.0	+	29.0	0.0	+	27.16	29.23	+	+m	+	+	+	+	+	+	PA	PA	PA	PA	2	b	
2023	563	Vacuum cleaner dust n°17-18 (non-probiotic dairy industry)	st	/	-	0.0	37.2	-	0.0	33.13	-	0.0	37.9	-	0.0	32.71	-	st	/	-	-	-	-	-	NA	NA	NA	NA	2	b	
2023	564	Vacuum cleaner dust n°19-20 (non-probiotic dairy industry)	st	/	-	0.0	37.4	-	0.0	32.93	-	0.0	37.4	-	0.0	32.95	-	st	/	-	-	-	-	-	NA	NA	NA	NA	2	b	
2023	565	Vacuum cleaner dust n°21-22 (non-probiotic dairy industry)	st	/	-	0.0	37.6	-	0.0	33.64	-	0.0	37.9	-	0.0	32.86	-	st	/	-	-	-	-	-	NA	NA	NA	NA	2	b	
2023	566	Vacuum cleaner dust n°23-24 (non-probiotic dairy industry)	st	/	-	0.0	37.8	-	0.0	33.28	-	0.0	38.2	-	0.0	32.94	-	st	/	-	-	-	-	-	NA	NA	NA	NA	2	b	
2023	567	Vacuum cleaner dust n°25-26 (non-probiotic dairy industry)	st	/	-	0.0	37.1	-	0.0	35.68	-	0.0	38.2	-	0.0	32.63	-	st	/	-	-	-	-	-	NA	NA	NA	NA	2	b	
2022	3740	Vacuum cleaner dust A32 E553 A31 (dairy industry)	-	/	-	38.4	37.0	+	33.01	32.79	+	37.1	37.3	+	33.79	32.36	+	+m	+	+	+	+	+	+	PD	PD	PD	PD	2	b	
2023	1371	Vacuum cleaner dust 27 (dairy industry)	+p	+	+	36.3/0.0/33.5	36.0/38.1/36.6	+/-/+	31.88/31.89/32.24	32.01/31.83/31.96	+/+/ +	35.5/33.5/33.6	37.0/36.6/36.3	+/+/ +	33.33/34.57/33.78	32.65/32.41/32.23	+/+/ +	-	/	-	-	-	-	-	PA _{FP(alt)}	PA _{FP(alt)}	PA _{FP(alt)}	PA _{FP(alt)}	2	b	
2022	3742	Vacuum cleaner dust P51 E2E2 (dairy industry)	-	/	-	29.4	0.0	+	25.09	0	+	28.1	0.0	+	24.72	29.48 (no amp)	+	+m	+	+	+	+	+	+	PD	PD	PD	PD	2	b	
2023	560	Vacuum cleaner dust n°11-12 (non-probiotic dairy industry)	st	/	-	18.1	0.0	+	16.54	0.0	+	22.0	0.0	+	18.71	0	+	+M	+	+	+	+	+	+	PD	PD	PD	PD	2	b	
2023	1363	Vacuum cleaner dust 24 (dairy industry)	+M	+	+	0.0	37.0	-	0.0	33.11	-	0.0	38.0	-	0.0	33.02	-	st	/	-	-	-	-	-	ND	ND	ND	ND	2	b	

PRODUCTION ENVIRONMENTAL SAMPLES																														
Year of analysis	Sample N°	Product (French name)	ISO 22964*			Alternative method: foodproof® <i>Salmonella</i> Plus <i>Cronobacter</i> Detection LyoKit - 5'Nuclease-																						Category	Type	
						Pre-warmed BPW or Prewarmed BPW + vancomycin (10mg/L) or BPW- 16h-37°C																								
						PCR result												Confirmation			Final result <i>Cronobacter</i> spp.				Agreement					
			foodproof® extraction			BAX lysis						CSB	ISO 22964	All confirmation tests	foodproof® extraction		BAX lysis		foodproof® extraction		BAX lysis									
						BAX Q7			LC480 II			BAX Q7			LC480 II			CCI	BAX Q7	LC 480 II	BAX Q7	LC 480 II	BAX Q7	LC 480 II						
			Ct HEX (target)	Ct ROX (IC)	Result	Ct HEX (target)	Ct ROX (IC)	Result	Ct HEX (target)	Ct ROX (IC)	Result <i>Crono</i> spp.	Ct HEX (target)			Ct ROX (IC)	Result	Ct HEX (target)								Ct ROX (IC)	Result	BAX Q7			LC 480 II
2023	1364	Vacuum cleaner dust 22 (dairy industry)	+1/2	+	+	37.2	37.6	+	0/0/0	31.22/30.44/29.65	-/-/-	36.9	37.7	+	0.0/35.45/0.0	35.65/33.92/0.0	-/+/-	+1/2	+	+	+	-	+	-	PA	ND _{FN(alt)}	PA	ND _{FN(alt)}	2	b
2023	1365	A32 Vacuum Cleaner Dust (Dairy Industry)	-	/	-	0.0	37.3	-	0.0	32.64	-	0.0	37.9	-	0.0	33.15	-	-	/	-	-	-	-	-	NA	NA	NA	NA	2	b
2023	1366	Vacuum cleaner dust 29 (dairy industry)	+1/2	+	+	31.8	34.5	+	27.74	29.56	+	31.5	35.4	+	27.29	29.44 (uncertain)	+	+1/2	+	+	+	+	+	+	PA	PA	PA	PA	2	b
2023	1367	Vacuum cleaner dust 12 (dairy industry)	+1/2	+	+	31.1	33.6	+	28.26	29.76	+	30.0	0.0	+	26.4	0	+	+M	+	+	+	+	+	+	PA	PA	PA	PA	2	b
2023	1368	Vacuum cleaner dust 20 (dairy industry)	-	/	-	0.0	35.7	-	0.0	33.03	-	0.0	38.4	-	0.0	33.51	-	st	/	-	-	-	-	-	NA	NA	NA	NA	2	b
2023	1369	Vacuum cleaner dust 26+P41 (dairy industry)	-	/	-	0.0	37.2	-	0.0	33.01	-	0.0	38.3	-	0.0	33.55	-	-	/	-	-	-	-	-	NA	NA	NA	NA	2	b
2023	1370	Vacuum cleaner dust P102+16 (dairy industry)	+M	+	+	24.6	0.0	+	22.56	0.0	+	28.3	0.0	+	24.09	0	+	+p	+	+	+	+	+	+	PA	PA	PA	PA	2	b
2022	3739	Vacuum cleaner dust A22 E241 (dairy industry)	-	/	-	37.9/0.0/0.0	38.5/0.0/0.0	+i/i/-	28.56/0.0/0ne	40.61/0.0/0	+/-/-ne	0.0	38.6	-	0.0	37.66	-	-(-x5)	/	-	-	-	-	-	PD _{FP(alt)}	PD _{FP(alt)}	NA	NA	2	b

INFANT FORMULA WITH OR WITHOUT PROBIOTICS AND RELATED INGREDIENTS																										
Year of analysis	Sample N°	Product (French name)	ISO 22964*	Alternative method: foodproof® Salmonella Plus Cronobacter Detection LyoKit - 5'Nuclease-																				Category	Type	
				Pre-warmed BPW or Prewarmed BPW + vancomycin (10mg/L) or BPW- 16h-37°C + 72h at 3±2°C																						
			Result	PCR result									All confirmation tests	Final result Cronobacter spp.				Agreement + 72h at 3±2°C								
				foodproof® extraction						BAX Real-Time lysis procedure						foodproof® extraction		BAX lysis		foodproof® extraction		BAX lysis				
				BAX Q7			LC480 II			BAX Q7				LC480 II			foodproof® extraction		BAX lysis		foodproof® extraction		BAX lysis			
Ct HEX (target)	Ct ROX (IC)	Result	Ct HEX (target)	Ct ROX (IC)	Result	Ct HEX (target)	Ct ROX (IC)	Result Crono spp.	Ct HEX (target)	Ct ROX (IC)	Result	Ct HEX (target)	Ct ROX (IC)	Result	BAX Q7	LC 480 II	BAX Q7	LC 480 II	BAX Q7	LC 480 II	BAX Q7	LC 480 II	BAX Q7	LC 480 II		
2022	3568	Infant milk powder, 6-12 months, with probiotics (B. lactis 1.8 x 10 ⁶ CFU/g) (26%MG)	-	0.0	37.0	-	35.03**/0.0/0.0/0*	33.11/0.0/0.0/33.15	+/i/i/-*	0.0/39.9/0.0	38.1/37.4/38.0	-/+/-	0.0	32.66	-	-	-	-	-	-	NA	PDF _{FP(alt)}	NA	NA	1	a
2023	766	Infant milk powder with probiotics, 3rd age (L. reutei DSM 17938 8.24 x 10 ⁴ CFU/g) (23.4% fat)	-	0.0	37.2	-	0.0	31.79	-	0.0	38.5	-	0.0	32.72	-	-	-	-	-	NA	NA	NA	NA	1	a	
2022	3567	Infant milk powder, 6-12 months, with probiotics (B. infantis 3.2 x 10 ⁷ CFU/g) (23%MG)	-	18.0	0.0	+	13.98	0.0	+	19.0	0.0	+	13.79	0.0	+	+	+	+	+	PD	PD	PD	PD	1	a	
2022	3797	Infant milk powder, 3rd age, with probiotics (B. lactis + S. thermophilus 1.5 x 10 ⁶ CFU/g) (30% fat)	-	19.7	0.0	+	16.29	0.0	+	21.6	0.0	+	18.23	0.0	+	+	+	+	+	PD	PD	PD	PD	1	a	
2023	763	Infant milk powder with probiotics, 3rd age (L. reutei DSM 17938 8.55 x 10 ⁴ CFU/g) (26% fat)	-	22.4	0.0	+	20.45	0.0	+	24.1	0.0	+	19.98	0.0	+	+	+	+	+	PD	PD	PD	PD	1	a	
2022	3561	Infant milk powder, 6-12 months, with probiotics (B. infantis 2.5 x 10 ⁶ CFU/g) (26% fat)	-	0.0	38.3	-	0.0	32.92	-	0.0	37.8	-	0.0	36.7	-	-	-	-	-	NA	NA	NA	NA	1	a	
2022	3562	Infant milk powder, 6-12 months, with probiotics (B. infantis 3.2 x 10 ⁷ CFU/g) (23%MG)	-	0.0	37.1	-	0.0	33.22	-	0.0	38.0	-	0.0	32.65	-	-	-	-	-	NA	NA	NA	NA	1	a	
2022	3563	Infant milk powder, 6-12 months, with probiotics (B. lactis 1.8 x 10 ⁶ CFU/g) (26%MG)	-	0.0	38.4	-	0.0	0.0	-	0.0	38.3	-	0.0	35.25	-	-	-	-	-	NA	NA	NA	NA	1	a	
2022	3564	Infant milk powder, from 6 months, with probiotics (B. lactis 3.5 x 10 ⁷ CFU/g) (26% fat)	-	0.0	37.5	-	35.61**/0.0/0.0	33.14/0.0/0.0	+/-/-	0.0	37.5	-	0.0	37.71	-	-	-	-	-	NA	PDF _{FP(alt)}	NA	NA	1	a	
2022	3565	Infant milk powder, babies, with probiotics (B. lactis 3.5 x 10 ⁷ CFU/g) (23.6% fat)	-	0.0	37.3	-	0.0	33.54	-	0.0	37.8	-	0.0	32.83	-	-	-	-	-	NA	NA	NA	NA	1	a	
2022	3566	Infant milk powder, 6-12 months, follow-on milk, with probiotics (B. infantis 2.5 x 10 ⁶ CFU/g) (26% fat)	+	21.1	0.0	+	13.95	0.0	+	18.4	0.0	+	14.80	0.0	+	+	+	+	+	PA	PA	PA	PA	1	a	
2023	765	Infant milk powder with probiotics, after from 10 months (L. reutei DSM 17938 1.55 x 10 ⁵ CFU/g) (23% fat)	-	32.3	35.0	+	27.34	31.04	+	32.5	36.5	+	29.31	34.06	+	+	+	+	+	PD	PD	PD	PD	1	a	
2022	3569	Infant milk powder, from 6 months, with probiotics (B. lactis 3.5 x 10 ⁷ CFU/g) (26% fat)	+	19.6	0.0	+	14.17	0.0	+	18.9	0.0	+	15.13	0.0	+	+	+	+	+	PA	PA	PA	PA	1	a	
2022	3570	Infant milk powder, babies, with probiotics (B. lactis 3.5 x 10 ⁷ CFU/g) (23.6% fat)	+	19.1	0.0	+	15.8	0.0	+	19.0	0.0	+	14.52	0.0	+	+	+	+	+	PA	PA	PA	PA	1	a	

* Analyses performed according to the COFRAC accreditation (Accreditation Testing n°1-0144, scope available on www.cofrac.fr)

INFANT FORMULA WITH OR WITHOUT PROBIOTICS AND RELATED INGREDIENTS																										
Year of analysis	Sample N°	Product (French name)	ISO 22964*	Alternative method: foodproof® <i>Salmonella</i> Plus <i>Cronobacter</i> Detection LyoKit - 5'Nuclease- Pre-warmed BPW or Prewarmed BPW + vancomycin (10mg/L) or BPW- 16h-37°C + 72h at 3±2°C																				Category	Type	
				PCR result												All confirmation tests	Final result <i>Cronobacter</i> spp.				Agreement + 72h at 3±2°C					
			foodproof® extraction						BAX Real-Time lysis procedure						foodproof® extraction		BAX lysis		foodproof® extraction		BAX lysis					
			BAX Q7			LC480 II			BAX Q7			LC480 II			BAX Q7		LC 480 II	BAX Q7	LC 480 II	BAX Q7	LC 480 II	BAX Q7	LC 480 II			
			Ct HEX (target)	Ct ROX (IC)	Result	Ct HEX (target)	Ct ROX (IC)	Result	Ct HEX (target)	Ct ROX (IC)	Result <i>Crono</i> spp.	Ct HEX (target)	Ct ROX (IC)	Result	BAX Q7		LC 480 II	BAX Q7	LC 480 II	BAX Q7	LC 480 II	BAX Q7	LC 480 II			
2023	769	Infant milk powder with probiotics, organic from 6 months (L. reuteuri DSM 17938 2.45 x 10 ⁶ CFU/g) (26% fat)	-	29.9	33.0	+	28.03	30.07	+	28.8	0.0	+	25.01	0.0	+	+	+	+	+	+	PD	PD	PD	PD	1	a
2023	771	Infant milk powder with probiotics, 1st age for infant 0-6 months (L. fermentum hereditum 5.09 x 10 ⁶ CFU/g) (24% fat)	-	32.5	35.1	+	29.84	31.78	+	32.4	37.0	+	29.26	33.19	+	+	+	+	+	+	PD	PD	PD	PD	1	a
2022	3798	Infant milk powder, 3rd age, with probiotics (B. lactis + S. thermophilus 1.5 x 10 ⁶ CFU/g) (30% fat)	+	16.7	0.0	+	14.01	0.0	+	18.0	0.0	+	14.87	0.0	+	+	+	+	+	+	PA	PA	PA	PA	1	a
2022	3799	Infant milk powder, 2nd age, with probiotics (B. lactis + S. thermophilus 3.1 x 10 ⁷ CFU/g) (23% fat)	+	17.7	0.0	+	14.52	0.0	+	19.1	0.0	+	15.39	0.0	+	+	+	+	+	+	PA	PA	PA	PA	1	a
2022	3800	Infant milk powder, 2nd age, with probiotics (B. lactis + S. thermophilus 3.1 x 10 ⁷ CFU/g) (23% fat)	+	19.4	0.0	+	16.26	0.0	+	21.0	0.0	+	17.76	0.0	+	+	+	+	+	+	PA	PA	PA	PA	1	a
2022	3801	Infant milk powder, 1st age, with probiotics (B. lactis 8.6 x 10 ⁵ CFU/g) (26% fat)	+	24.4	0.0	+	19.91	0.0	+	26.5	0.0	+	24.56	0.0	+	+	+	+	+	+	PA	PA	PA	PA	1	a
2023	762	Infant milk powder with probiotics, 3rd age (L. reuteuri DSM 17938 8.55 x 10 ⁴ CFU/g) (26% fat)	-	0.0	38.1	-	0.0	0.0	-	0.0	38.4	-	0.0	39.82	-	-	-	-	-	-	NA	NA	NA	NA	1	a
2023	764	Infant milk powder with probiotics, after 10 months (L. reuteuri DSM 17938 1.55 x 10 ⁵ CFU/g) (23% fat)	-	0.0	37.8	-	0.0	30.66	-	0.0	38.3	-	0.0	31.47	-	-	-	-	-	-	NA	NA	NA	NA	1	a
2023	767	Infant milk powder with probiotics, 3rd age from 1 year (L. reuteuri DSM 17938 8.24 x 10 ⁴ CFU/g) (23.4% fat)	+	18.5	0.0	+	15.24	0.0	+	19.4	0.0	+	16.62	0.0	+	+	+	+	+	+	PA	PA	PA	PA	1	a
2023	768	Infant milk powder with probiotics, organic from 6 months (L. reuteuri DSM 17938 2.45 x 10 ⁶ CFU/g) (26% fat)	-	0.0	37.1	-	0.0	33.26	-	0.0	38.5	-	38.95/ 39.01/ 39.71	33.20/ 32.91/ 33.17	+ / + / +	-	-	-	-	-	NA	NA	NA	PD _{FP(alt)}	1	a
2023	770	Infant milk powder with probiotics, 1st age for infant 0-6 months (L. fermentum hereditum 5.09 x 10 ⁶ CFU/g) (24% fat)	-	0.0	37.5	-	0.0	32.51	-	0.0	38.7	-	0.0	32.98	-	-	-	-	-	-	NA	NA	NA	NA	1	a
2023	772	Infant milk powder with probiotics, formula thickened from 6 months (L. fermentum hereditum 3.09 x10 ⁶ CFU/g) (26% fat)	-	0.0	37.9	-	0.0	32.54	-	0.0	38.1	-	0.0	32.8	-	-	-	-	-	-	NA	NA	NA	NA	1	a
2022	2903	Infant formula, 0-6 months (23.5% FL)	-	0.0	37.7	-	0.0	33.28	-	0.0	37.6	-	0.0	33.56	-	-	-	-	-	-	NA	NA	NA	NA	1	b
2022	2904	Infant formula, 6-12 months (22.6% FL)	-	0.0	38.5	-	0.0	33.57	-	0.0	37.1	-	0.0	37.47	-	-	-	-	-	-	NA	NA	NA	NA	1	b

INFANT FORMULA WITH OR WITHOUT PROBIOTICS AND RELATED INGREDIENTS																										
Year of analysis	Sample N°	Product (French name)	ISO 22964*	Alternative method: foodproof® <i>Salmonella</i> Plus <i>Cronobacter</i> Detection LyoKit - 5'Nuclease- Pre-warmed BPW or Prewarmed BPW + vancomycin (10mg/L) or BPW- 16h-37°C + 72h at 3±2°C																				Category	Type	
				PCR result												All confirmation tests	Final result <i>Cronobacter</i> spp.				Agreement + 72h at 3±2°C					
			foodproof® extraction						BAX Real-Time lysis procedure						foodproof® extraction		BAX lysis		foodproof® extraction		BAX lysis					
			BAX Q7			LC480 II			BAX Q7			LC480 II			BAX Q7		LC 480 II	BAX Q7	LC 480 II	BAX Q7	LC 480 II	BAX Q7	LC 480 II			
			Ct HEX (target)	Ct ROX (IC)	Result	Ct HEX (target)	Ct ROX (IC)	Result	Ct HEX (target)	Ct ROX (IC)	Result <i>Crono</i> spp.	Ct HEX (target)	Ct ROX (IC)	Result	Ct HEX (target)		Ct ROX (IC)	Result	BAX Q7	LC 480 II	BAX Q7	LC 480 II	BAX Q7			LC 480 II
2022	2907	Infant formula, stage 2, from 6 months (22.0% FL)	-	0.0	38.4	-	37.78/0.0/0.0	32.91/0.0/0.0/39.84/45.0	+/-/-	37.1/37.9/38.1	37.8/37.7/37.5	+/+/+	0.0	0.0	-	-	-	-	-	-	NA	PD _{FP(alt)}	PD _{FP(alt)}	NA	1	b
2022	2909	Infant formula, stage 2, 6months-1year (23.6% FL)	-	0.0	38.2	-	0.0	33.34	-	0.0	38.1	-	0.0	45.00	-	-	-	-	-	-	NA	NA	NA	NA	1	b
2022	2910	Infant formula, stage 2, 6-12 months (21.1% FL)	-	0.0	37.6	-	38.40 (uncertain)/0.0/0.0	33.61/45/0.0	+/-/-	38.7/0.0/0.0	38.2/37.5/37.7	+/-/-	0.0	0.0	-	-	-	-	-	-	NA	PD _{FP(alt)}	PD _{FP(alt)}	NA	1	b
2022	2963	Infant formula, stage 1, 6-12 months (22.0% FL)	+	21.3	0.0	+	18.49	0.0	+	21.5	0.0	+	19.99	0.0	+	+	+	+	+	+	PA	PA	PA	PA	1	b
2022	2964	Organic infant formula, 0-6 months (24.0% FL)	+	23.7	0.0	+	21.24	0.0	+	23.7	0.0	+	21.35	0.0	+	+	+	+	+	+	PA	PA	PA	PA	1	b
2022	2965	Organic infant formula, 0-6 months (24.0% FL)	+	21.9	0.0	+	19.78	0.0	+	22.9	0.0	+	20.66	0.0	+	+	+	+	+	+	PA	PA	PA	PA	1	b
2022	2966	Infant formula, 0-6 months (22.0% FL)	+	21.7	0.0	+	19.27	0.0	+	24.6	0.0	+	20.24	0.0	+	+	+	+	+	+	PA	PA	PA	PA	1	b
2022	2967	Infant formula, 0-6 months (21.0% FL)	+	19.3	0.0	+	15.9	0.0	+	20.4	0.0	+	16.27	0.0/	+	+	+	+	+	+	PA	PA	PA	PA	1	b
2022	2968	Infant formula, stage 2, 6-12 months (26.0% FL)	+	27.3	0.0	+	24.57	0.0	+	24.1	0.0	+	20.65	0.0	+	+	+	+	+	+	PA	PA	PA	PA	1	b
2022	2969	Organic infant formula, from 6 months (25.0%FL)	+	28.5	0.0	+	26.22	28.2 (no amp)	+	27.6	0.0	+	24.33	0.0	+	+	+	+	+	+	PA	PA	PA	PA	1	b
2022	2970	Organic infant formula, 6-12 months (22.0%FL)	+	21.4	0.0	+	18.94	0.0	+	21.0	0.0	+	16.29	0.0	+	+	+	+	+	+	PA	PA	PA	PA	1	b
2022	3023	Infant formula anti-regurgitation, 0-6 months (25.0% FL)	+	26.1	0.0	+	22.9	23.12 (no amp)	+	28.1	0.0	+	26.42	0.0	+	+	+	+	+	+	PA	PA	PA	PA	1	b
2022	3024	Organic infant formula, stage 2, 6-12 months (25.0%FL)	+	23.5	0.0	+	21.28	0.0	+	26.4	0.0	+	24.75	0.0	+	+	+	+	+	+	PA	PA	PA	PA	1	b
2022	3025	Infant milk powder, gourmet babies, 6-12 months (26%fat)	+	21.9	0.0	+	20.61	0.0	+	23.5	0.0	+	19.66	0.0	+	+	+	+	+	+	PA	PA	PA	PA	1	b
2022	3026	Infant milk powder, 6-12 months (25.5% fat)	+	24.0	0.0	+	22.14	0.0	+	24.7	0.0	+	22.01	0.0	+	+	+	+	+	+	PA	PA	PA	PA	1	b
2022	3027	Organic infant milk powder, 2nd age (25%fat)	+	24.4	0.0	+	22.73	0.0	+	26.6	0.0	+	23.63	0.0	+	+	+	+	+	+	PA	PA	PA	PA	1	b
2022	3028	Premium infant milk powder, 0-6 months (24.7% fat)	+	34.9	38.2	+	30.72	32.31	+	30.9	0.0	+	28.14	29.28	+	+	+	+	+	+	PA	PA	PA	PA	1	b
2022	2905	Organic infant formula, 6-12 months (23.1% FL)	-	39.8/0.0/0.0	37.5/37.0/0.0	+/-/i	42.05/0.0/0.0	40.69/0.0/0.0	+i/-	38.6/0.0/0.0	37.3/37.2/36.7	+/-/-	0.0	42.25	-	-	-	-	-	-	PD _{FP(alt)}	PD _{FP(alt)}	PD _{FP(alt)}	NA	1	b
2022	2906	Infant formula, stage 2, 6-12 months (26.0% FL)	-	38.6/0.0/0.0	37.6/37.0/37.5	+/-/-	34.5/0.0/0.0	33.00/0.0/40.44	+/-/-	39.9/0.0/0.0	38.2/37.8/37.7	+/-/-	0.0	40.03	-	-	-	-	-	-	PD _{FP(alt)}	PD _{FP(alt)}	PD _{FP(alt)}	NA	1	b
2022	3083	Rice starch	-	0.0	37.4	-	0.0	32.8	-	0.0	37.2	-	0.0	38.45	-	-	-	-	-	-	NA	NA	NA	NA	1	c
2022	3084	Corn starch	-	0.0	37.1	-	0.0	32.69	-	0.0	37.6	-	0.0	37.49	-	-	-	-	-	-	NA	NA	NA	NA	1	c
2022	3085	Maltodextrin	-	0.0	37.6	-	0.0	32.85	-	0.0	37.7	-	0.0	31.88	-	-	-	-	-	-	NA	NA	NA	NA	1	c
2022	3086	Whey	-	0.0	41.3	-	0.0	33.44	-	0.0	37.9	-	0.0	38.00	-	-	-	-	-	-	NA	NA	NA	NA	1	c

INFANT FORMULA WITH OR WITHOUT PROBIOTICS AND RELATED INGREDIENTS																										
Year of analysis	Sample N°	Product (French name)	ISO 22964*	Alternative method: foodproof® <i>Salmonella</i> Plus <i>Cronobacter</i> Detection LyoKit - 5'Nuclease- Pre-warmed BPW or Prewarmed BPW + vancomycin (10mg/L) or BPW- 16h-37°C + 72h at 3±2°C																				Category	Type	
				PCR result												All confirmation tests	Final result <i>Cronobacter</i> spp.				Agreement + 72h at 3±2°C					
				foodproof® extraction						BAX Real-Time lysis procedure							foodproof® extraction		BAX lysis		foodproof® extraction		BAX lysis			
				BAX Q7			LC480 II			BAX Q7			LC480 II				foodproof® extraction		BAX lysis		foodproof® extraction		BAX lysis			
				Ct HEX (target)	Ct ROX (IC)	Result	Ct HEX (target)	Ct ROX (IC)	Result	Ct HEX (target)	Ct ROX (IC)	Result <i>Crono</i> spp.	Ct HEX (target)	Ct ROX (IC)	Result		BAX Q7	LC 480 II	BAX Q7	LC 480 II	BAX Q7	LC 480 II	BAX Q7			LC 480 II
2022	3087	Whey	-	0.0	0.0	-	0.0	32.94	-	0.0	38.2	-	0.0	34.12	-	-	-	-	-	-	NA	NA	NA	NA	1	c
2022	3088	Maltodextrin	-	0.0	37.5	-	0.0	33.07	-	0.0	37.2	-	0.0	39.54	-	-	-	-	-	-	NA	NA	NA	NA	1	c
2022	3091	Maltodextrin	+	17.4	0.0	+	13.95	0.0	+	17.6	0.0	+	15.88	0.0	+	+	+	+	+	+	PA	PA	PA	PA	1	c
2022	3092	Whey	+	30.5	33.6	+	27.35	31.09	+	31.1	36.3	+	27.6	33.18	+	+	+	+	+	+	PA	PA	PA	PA	1	c
2022	3093	Whey	+	31.1	34.4	+	28.23	31.2	+	30.9	36.3	+	27.97	32.97	+	+	+	+	+	+	PA	PA	PA	PA	1	c
2022	3175	Lactose	+	19.3	0.0	+	16.99	0.0	+	21.6	0.0	+	18.34	0.0	+	+	+	+	+	+	PA	PA	PA	PA	1	c
2022	3176	Lactose	+	19.1	0.0	+	16.62	0.0	+	23.1	0.0	+	19.08	0.0	+	+	+	+	+	+	PA	PA	PA	PA	1	c
2022	3177	Whey	+	19.3	0.0	+	16.82	0.0	+	22.3	0.0	+	19.49	0.0	+	+	+	+	+	+	PA	PA	PA	PA	1	c
2022	3178	Wheat starch	+	17.9	0.0	+	16.27	0.0	+	19.9	0.0	+	17.74	0.0	+	+	+	+	+	+	PA	PA	PA	PA	1	c
2022	3179	Maltodextrin	+	21.1	0.0	+	17.94	0.0	+	22.5	0.0	+	19.09	0.0	+	+	+	+	+	+	PA	PA	PA	PA	1	c
2022	3180	Whey Protein Concentrate	+	21.8	0.0	+	18.38	0.0	+	23.3	0.0	+	19.65	0.0	+	+	+	+	+	+	PA	PA	PA	PA	1	c
2022	3732	Milk Protein	-	0.0	36.4	-	0.0	33.32	-	0.0	38.6	-	0.0	32.84	-	-	-	-	-	-	NA	NA	NA	NA	1	c
2022	3735	Whey	-	0.0	36.3	-	0.0	33.44	-	0.0	38.1	-	0.0	32.87	-	-	-	-	-	-	NA	NA	NA	NA	1	c
2022	3089	Rice starch	-	23.8	0.0	+	20.65	0.0	+	23.3	0.0	+	19.66	0.0	+	+	+	+	+	+	PD	PD	PD	PD	1	c
2022	3090	Corn starch	-	32.0	34.7	+	29.24	31.85	+	32.3	36.1	+	27.78	33.39	+	+	+	+	+	+	PD	PD	PD	PD	1	c
2022	3094	Maltodextrin	-	17.8	0.0	+	15.21	0.0	+	18.7	0.0	+	15.57	0.0	+	+	+	+	+	+	PD	PD	PD	PD	1	c

PRODUCTION ENVIRONMENTAL SAMPLES																											
Year of analysis	Sample N°	Product (French name)	ISO 22964*	Alternative method: foodproof® Salmonella Plus Cronobacter Detection LyoKit - 5'Nuclease-																				Category	Type		
				Pre-warmed BPW or Prewarmed BPW + vancomycin (10mg/L) or BPW- 16h-37°C + 72h at 3±2°C																							
				PCR result+ 72h at 3±2°C													All confirmation tests	Final result Cronobacter spp.				Agreement+ 72h at 3±2°C					
				foodproof® extraction						BAX Real-Time lysis procedure								foodproof® extraction		BAX lysis		foodproof® extraction				BAX lysis	
				BAX Q7			LC480 II			BAX Q7			LC480 II					foodproof® extraction		BAX lysis		foodproof® extraction				BAX lysis	
				Ct HEX (target)	Ct ROX (IC)	Result	Ct HEX (target)	Ct ROX (IC)	Result	Ct HEX (target)	Ct ROX (IC)	Result Crono spp.	Ct HEX (target)	Ct ROX (IC)	Result	BAX Q7		LC 480 II	BAX Q7	LC 480 II	BAX Q7	LC 480 II	BAX Q7			LC 480 II	BAX Q7
2022	3397	Floor surface swabs before cleaning (dairy industry)	-	0.0	37.4	-	0.0	33.09	-	0.0	37.6	-	0.0	35.87	-	-		-	-	-	-	NA	NA	NA	NA	2	a
2022	3398	Floor surface swabs before cleaning (dairy industry)	-	0.0	37.6	-	0.0	33.17	-	0.0	40.6	-	0.0	33.85	-	-	-	-	-	-	NA	NA	NA	NA	2	a	
2022	3399	Floor surface swabs before cleaning (dairy industry)	+	16.5	0.0	+	14.47	0.0	+	17.3	0.0	+	14.73	0.0	+	+	+	+	+	+	PA	PA	PA	PA	2	a	
2022	3400	Floor surface swabs before cleaning (dairy industry)	+	17.2	0.0	+	11.7	0.0	+	16.5	0.0	+	13.99	0.0	+	+	+	+	+	+	PA	PA	PA	PA	2	a	
2022	3401	Floor surface swabs before cleaning (dairy industry)	+	16.9	0.0	+	15.51	0.0	+	20.9	0.0	+	17.06	0.0	+	+	+	+	+	+	PA	PA	PA	PA	2	a	
2022	3402	Floor surface swabs before cleaning (dairy industry)	+	16.4	0.0	+	14.06	0.0	+	18.3	0.0	+	15.9	0.0	+	+	+	+	+	+	PA	PA	PA	PA	2	a	
2022	3403	Floor surface swabs before cleaning (dairy industry)	-	0.0	37.8	-	0.0	32.85	-	0.0	37.7	-	0.0	37.05	-	-	-	-	-	-	NA	NA	NA	NA	2	a	
2022	3407	Floor surface swabs before cleaning (dairy industry)	+	16.5	0.0	+	14.29	0.0	+	18.0	0.0	+	14.10	0.0	+	+	+	+	+	+	PA	PA	PA	PA	2	a	
2022	3408	Floor surface swabs before cleaning (dairy industry)	+	16.6	0.0	+	14.45	0.0	+	19.6	0.0	+	16.92	0.0	+	+	+	+	+	+	PA	PA	PA	PA	2	a	
2022	3409	Floor surface swabs before cleaning (dairy industry)	+	16.8	0.0	+	14.85	0.0	+	16.8	0.0	+	14.56	0.0	+	+	+	+	+	+	PA	PA	PA	PA	2	a	
2022	3410	Floor surface swabs before cleaning (dairy industry)	+	15.9	0.0	+	14.01	15.25 (no amp)	+	16.7	0.0	+	14.34	0.0	+	+	+	+	+	+	PA	PA	PA	PA	2	a	
2022	3475	Swipe before cleaning (ice cream production)	-	0.0	36.5	-	0.0	45	-	0.0	38.9	-	0.0	36.66	-	-	-	-	-	-	NA	NA	NA	NA	2	a	
2022	3476	Swipe before cleaning (ice cream production)	+	19.4	0.0	+	16.86	0.0	+	21.5	0.0	+	18.34	0.0	+	+	+	+	+	+	PA	PA	PA	PA	2	a	
2022	3477	Swipe after cleaning (ice cream production)	+	18.0	0.0	+	15.11	35.46 (no amp)	+	22.5	0.0	+	18.08	0.0	+	+	+	+	+	+	PA	PA	PA	PA	2	a	
2022	3478	Swipe before cleaning (Ice Cream Production)	-	0.0	36.9	-	0.0	0.0	-	0.0	37.6	-	0.0	33	-	-	-	-	-	-	NA	NA	NA	NA	2	a	
2022	3479	Swipe After Cleaning (Dairy Industry)	-	0.0	37.0	-	0.0	45	-	0.0	37.2	-	0.0	37.56	-	-	-	-	-	-	NA	NA	NA	NA	2	a	
2022	3480	Swipe After Cleaning (Dairy Industry)	+	23.4	0.0	+	19.83	0.0	+	24.2	0.0	+	21.68	0.0	+	+	+	+	+	+	PA	PA	PA	PA	2	a	
2022	3481	Swipe After Cleaning (Dairy Industry)	+	21.3	0.0	+	18.37	0.0	+	28.8	0.0	+	21.76	0.0	+	+	+	+	+	+	PA	PA	PA	PA	2	a	
2022	3637	Stainless steel bench swab, after cleaning (dairy industry)	-	0.0	0.0	-	0.0	33.32	-	0.0	37.9	-	0.0	35.98	-	-	-	-	-	-	NA	NA	NA	NA	2	a	
2022	3638	Vacuum nozzle swab, before cleaning (dairy industry)	-	0.0/0.0/0.0*	0.0/37.3/45.0*	i/-/*	0.0	33.18	-	0.0	38.4	-	0.0	37.11	-	-	-	-	-	-	NA	NA	NA	NA	2	a	
2022	3639	Swab before cleaning (dairy industry)	+	0.0/18.2/21.9*	0.0/0.0/0.0*	i/+/*	16.03	0.0	+	23.9	0.0	+	19.86	0.0	+	+	+	+	+	+	PA	PA	PA	PA	2	a	

* Analyses performed according to the COFRAC accreditation (Accreditation Testing n°1-0144, scope available on www.cofrac.fr)

PRODUCTION ENVIRONMENTAL SAMPLES																										
Year of analysis	Sample N°	Product (French name)	ISO 22964*	Alternative method: foodproof® <i>Salmonella</i> Plus <i>Cronobacter</i> Detection LyoKit - 5'Nuclease-																				Category	Type	
				Pre-warmed BPW or Prewarmed BPW + vancomycin (10mg/L) or BPW- 16h-37°C + 72h at 3±2°C																						
				PCR result+ 72h at 3±2°C												All confirmation tests	Final result <i>Cronobacter</i> spp.				Agreement+ 72h at 3±2°C					
			foodproof® extraction						BAX Real-Time lysis procedure						foodproof® extraction		BAX lysis		foodproof® extraction		BAX lysis					
			BAX Q7			LC480 II			BAX Q7			LC480 II			foodproof® extraction		BAX lysis		foodproof® extraction		BAX lysis					
			Ct HEX (target)	Ct ROX (IC)	Result	Ct HEX (target)	Ct ROX (IC)	Result	Ct HEX (target)	Ct ROX (IC)	Result <i>Crono</i> spp.	Ct HEX (target)	Ct ROX (IC)	Result	BAX Q7		LC 480 II	BAX Q7	LC 480 II	BAX Q7	LC 480 II	BAX Q7	LC 480 II			
2022	3640	Swab after cleaning (dairy industry)	+	0.0/18.2/21.8*	0.0/0.0/0.0*	i/+/*	15.73	0.0	+	27.2	0.0	+	19.14	0.0	+	+	+	+	+	+	PA	PA	PA	PA	2	a
2022	3641	Swipe, before cleaning (dairy industry)	-	0.0	36.4	-	0.0	32.1	-	0.0	36.5	-	36.90 (no amp)	31.89	-	-	-	-	-	-	NA	NA	NA	NA	2	a
2022	3642	Swipe, before cleaning (dairy industry)	-	0.0	36.7	-	0.0	32.86	-	0.0	37.2	-	0.0	38	-	-	-	-	-	-	NA	NA	NA	NA	2	a
2022	3643	Swipe before cleaning (ice cream production)	-	0.0	36.8	-	0.0	33.03	-	0.0	37.8	-	0.0	33.14	-	-	-	-	-	-	NA	NA	NA	NA	2	a
2022	3644	Swipe after cleaning (ice cream production)	+	19.7	0.0	+	17.17	0.0	+	21.5	0.0	+	17.22	0.0	+	+	+	+	+	+	PA	PA	PA	PA	2	a
2022	3738	Vacuum cleaner dust P43 A32 (dairy industry)	-	0.0	37.8	-	32.10/0.0/0.0/0.0ne	32.13/0.0/31.68/32.65	+i/-/-ne	38.2/37.4/37.7	37.2/36.9/37.4	+/*/*	0.0	31.87	-	-	-	-	-	-	NA	PDFP(alt)	PDFP(alt)	NA	2	b
2022	3741	Vacuum cleaner dust P51 2889 A12 (dairy industry)	+	36.0	37.1	+	32.4	32.05	+	0.0/0.0/37.2	37.9/38.3/37.9	-/-/*	33.01	32.37	+	+	+	+	-	+	PA	PA	NDFN(alt)	PA	2	b
2023	550	Vacuum cleaner dust n°1 (non-probiotic dairy industry)	-	0.0/0.0/0.0	39.1/0.0/0.0	-/-/-	0.0	34.26	-	0.0	37.4	-	0.0	36.38	-	-	-	-	-	-	NA	NA	NA	NA	2	b
2023	552	Vacuum cleaner dust n°3 (non-probiotic dairy industry)	-	0.0	37.3	-	0.0	33.48	-	0.0	38.6	-	0.0	32.03	-	-	-	-	-	-	NA	NA	NA	NA	2	b
2023	553	Vacuum cleaner dust n°4 (non-probiotic dairy industry)	-	0.0	36.9	-	0.0	33.14	-	0.0	36.9	-	0.0	37.07	-	-	-	-	-	-	NA	NA	NA	NA	2	b
2023	554	Vacuum cleaner dust n°5 (non-probiotic dairy industry)	-	0.0	38.0	-	0.0	33.3	-	0.0	39.0	-	0.0	32.14	-	-	-	-	-	-	NA	NA	NA	NA	2	b
2023	556	Vacuum cleaner dust n°7 (non-probiotic dairy industry)	-	0.0	36.9	-	0.0	32.89	-	0.0	38.1	-	0.0	32.84	-	-	-	-	-	-	NA	NA	NA	NA	2	b
2023	557	Vacuum cleaner dust n°8 (non-probiotic dairy industry)	+	29.6	32.8	+	25.9	0.0	+	28.4	0.0	+	24.7	0.0	+	+	+	+	+	+	PA	PA	PA	PA	2	b
2023	558	Vacuum cleaner dust n°9 (non-probiotic dairy industry)	+	25.9	0.0	+	23.0	0.0	+	26.2	0.0	+	21.91	0.0	+	+	+	+	+	+	PA	PA	PA	PA	2	b
2023	559	Vacuum cleaner dust n°10 (non-probiotic dairy industry)	+	27.7	0.0	+	24.48	0.0	+	27.8	0.0	+	22.81	0.0	+	+	+	+	+	+	PA	PA	PA	PA	2	b
2023	561	Vacuum cleaner dust n°13-14 (non-probiotic dairy industry)	+	0.0/0.0/0.0	37.1/36.8/36.8	-/-/-	33.88	33.08	+	0.0/0.0/0.0	37.3/37.6/36.8	-/-/-	0.0/0.0/0.0	33.31/33.75/32.22	-/-/-	+	-	+	-	-	NDFN(alt)	PA	NDFN(alt)	NDFN(alt)	2	b
2023	562	Vacuum cleaner dust n°15-16 (non-probiotic dairy industry)	+	27.8	0.0	+	24.18	0.0	+	30.1	0.0	+	26.05	0.0	+	+	+	+	+	+	PA	PA	PA	PA	2	b
2023	566	Vacuum cleaner dust n°23-24 (non-probiotic dairy industry)	-	0.0	37.4	-	0.0	33.34	-	0.0	38.2	-	0.0	32.87	-	-	-	-	-	-	NA	NA	NA	NA	2	b
2022	3740	Vacuum cleaner dust A32 E553 A31 (dairy industry)	-	35.5	36.7	+	31.92	32.28	+	39.2	37.7	+	34.65	33.19	+	+	+	+	+	+	PD	PD	PD	PD	2	b
2023	1371	Vacuum cleaner dust 27 (dairy industry)	+	35.9/33.9/34.1	35.1/36.2/36.5	+/*/*	31.32/0.0/0.0	31.47/0.0/0.0	+/*/*	37.4/0.0/0.0	36.9/38.0/37.8	+/-/-	34.78/31.63/32.57	32.77/31.92/32.1	+/*/*	-	-	-	-	-	PAFP(alt)	PAFP(alt)	PAFP(alt)	PAFP(alt)	2	b
2022	3742	Vacuum cleaner dust P51 E2E2 (dairy industry)	-	28.7	0.0	+	24.86	27.14 (no amp)	+	29.1	0.0	+	24.81	0.0	+	+	+	+	+	+	PD	PD	PD	PD	2	b

PRODUCTION ENVIRONMENTAL SAMPLES																										
Year of analysis	Sample N°	Product (French name)	ISO 22964♦	Alternative method: foodproof® <i>Salmonella</i> Plus <i>Cronobacter</i> Detection LyoKit - 5'Nuclease- Pre-warmed BPW or Prewarmed BPW + vancomycin (10mg/L) or BPW- 16h-37°C + 72h at 3±2°C																				Category	Type	
				PCR result+ 72h at 3±2°C												All confirmation tests	Final result <i>Cronobacter</i> spp.				Agreement+ 72h at 3±2°C					
			foodproof® extraction						BAX Real-Time lysis procedure						foodproof® extraction		BAX lysis		foodproof® extraction		BAX lysis					
			BAX Q7			LC480 II			BAX Q7			LC480 II			foodproof® extraction		BAX lysis		foodproof® extraction		BAX lysis					
			Ct HEX (target)	Ct ROX (IC)	Result	Ct HEX (target)	Ct ROX (IC)	Result	Ct HEX (target)	Ct ROX (IC)	Result <i>Crono</i> spp.	Ct HEX (target)	Ct ROX (IC)	Result	BAX Q7		LC 480 II	BAX Q7	LC 480 II	BAX Q7	LC 480 II	BAX Q7	LC 480 II			BAX Q7
2023	560	Vacuum cleaner dust n°11-12 (non-probiotic dairy industry)	-	19.0	0.0	+	15.56	0.0	+	20.2	0.0	+	15.79	0.0	+	+	+	+	+	+	PD	PD	PD	PD	2	b
2023	1363	Vacuum cleaner dust 24 (dairy industry)	+	0.0	35.8	-	0.0	32.7	-	0.0	38.1	-	0.0	32.99	-	-	-	-	-	-	ND	ND	ND	ND	2	b
2023	1364	Vacuum cleaner dust 22 (dairy industry)	+	34.3	36.5	+	0.0/ 0.0/ 0.0	45/ 42.2/ 37.66	-/- ne/- ne	36.7	37.3	+	0.0/ 0.0/ 29.78	33.89/ 33.72/ 31.96	-/-/+	+	+	-	+	-	PA	ND _{FN(alt)}	PA	ND _{FN(alt)}	2	b
2023	1365	A32 Vacuum Cleaner Dust (Dairy Industry)	-	0.0	37.3	-	0.0	32.72	-	0.0	38.1	-	0.0	32.76	-	-	-	-	-	-	NA	NA	NA	NA	2	b
2023	1366	Vacuum cleaner dust 29 (dairy industry)	+	28.7	0.0	+	24.8	0.0	+	30.3	0.0	+	26.16	0.0	+	+	+	+	+	+	PA	PA	PA	PA	2	b
2023	1367	Vacuum cleaner dust 12 (dairy industry)	+	28.7	0.0	+	25.48	26.01 (no amp)	+	30.4	0.0	+	26.05	0.0	+	+	+	+	+	+	PA	PA	PA	PA	2	b
2023	1368	Vacuum cleaner dust 20 (dairy industry)	-	0.0	37.0	-	0.0	32.73	-	0.0	38.5	-	0.0	33.04	-	-	-	-	-	-	NA	NA	NA	NA	2	b
2023	1369	Vacuum cleaner dust 26+P41 (dairy industry)	-	0.0	37.1	-	0.0	33.25	-	0.0	38.3	-	0.0	33.0	-	-	-	-	-	-	NA	NA	NA	NA	2	b
2023	1370	Vacuum cleaner dust P102+16 (dairy industry)	+	20.8	0.0	+	18.78	0.0	+	22.9	0.0	+	19.97	26.77	+	+	+	+	+	+	PA	PA	PA	PA	2	b
2022	3739	Vacuum cleaner dust A22 E241 (dairy industry)	-	38.3/ 0.0/ 37.8	38.9/ 0.0/ 38.0	+/-/+	38.11/ 0.0/ 0.0/ One	38.40/ 0.0/ 0.0/ 0	+/i/-/- ne	0.0/ 39.4/ 38.7	38.8/ 38.2/ 37.9	-/+/+	0.0	35.84	-	-	-	-	-	-	PD _{FP(alt)}	PD _{FP(alt)}	NA	NA	2	b

Appendix 5 – Relative level of detection study: raw data

Matrix: Infant formula with probiotics (*Bifidobacterium lactis*) (375g)
Strain: *Cronobacter sakazakii* Ad2413
Seeding protocol with lyophilized strain storage for 2 weeks at ambient temperature
Protocol: Pre-warmed BPW + vancomycin + Tween 80 - d 1/10 - 16 h at 37°C ± 1°C
UNPAIRED

Lactic flora: 6.8 .10⁴ CFU/g

N° sample	Level	Contamination level (CFU/sample) VRBG poor plate method	Contamination level (CFU/sample) MPN	ISO 22964* method (10g)			foodproof® Salmonella Plus Cronobacter Detection LyoKit - 5'Nuclease-																			
				CSB		Final result	Number positive samples/Total	PCR result										Confirmation			Number positive samples / Total					
								foodproof® extraction					BAX Real-Time lysis procedure					After 16h incubation	After 20h incubation							
								BAX Q7 (from Hygiena) – New software version			LC480 CC #17002		BAX Q7 (from Hygiena) – New software version			LC480 CC #17002		CSB	CSB	All confirmation tests (16h)	foodproof® extraction		BAX Real-Time lysis procedure			
								Ct HEX (<i>Cronobacter</i> spp.)	Ct ROX (Internal control)	Result	Ct HEX (<i>Cronobacter</i> spp.)	Ct ROX (Internal control)	Result	Ct HEX (<i>Cronobacter</i> spp.)	Ct ROX (Internal control)	Result <i>Cronobacter</i> spp.	Ct HEX (<i>Cronobacter</i> spp.)	Ct ROX (Internal control)	Result		CCI	CCI	BAX Q7 (from Hygiena) New software	LightCycler 480 II CC #17002	BAX Q7 (from Hygiena) New software	LightCycler 480 II CC #17002
852	0	/	/	st	/	-	0/5	0.0	37.3	-	35.98/0.0/0.0	33.61/30.92/0.0	+/-/i	0.0	38.2	-	0.0	32.98	-	st	st	-	0/5	0/5	0/5	0/5
864				st	/	-		0.0	37.7	-	35.87/0.0/0.0/37.42ne	33.34/0.0/0.0/33.47ne	+/i/i/+ ne	38.5	37.5	+	36.95/37.17/37.69	33.08/32.64/32.79	+/+/+	st	st	-				
866				st	/	-		0.0	36.3	-	36.45/35.12/0.0	33.73/33.02/0.0	+/+/i	39.5	37.9	+	37.06/36.71/35.37	32.69	+/+/+	st	st	-				
874				st	/	-		0.0	37.0	-	0.0	32.97	-	0.0	38.2	-	0.0	32.94	-	st	st	-				
879				st	/	-		0.0	36.1	-	0.0	33.66	-	0.0	38.3	-	0.0	32.88	-	st	st	-				
853	Low	0.2	0.8 [0.5-1.4]	st	/	-	12/20	17.5	0.0	+	14.91	0.0	+	19.1	0.0	+	15.72	0.0	+	+p	+p	+	11/20	11/20	11/20	11/20
854				+p	+	+		27.8	0.0	+	25.27	0.0	+	28.6	0.0	+	24.95	0.0	+	+p	+p	+				
857				+p	+	+		39.7	37.5	+	35.61/0.0/0.0/38.15ne	33.04/0/0/33.04ne	+/i/i/+ne	39.5/0.0/0.0	38/37.6/37.3	+/-/-	35.22/37.15/37.85	32.64/33.17/32.93	+/+/+	st	st	-				
858				st	/	-		37.8	37.4	+	34.83/0.0/0.0	33.24/32.17/0.0	+/-/i	39.8/0.0/0.0	37.7/37.1/37.7	+/-/-	37.06/36.95/36.78	32.90/32.79/33.07	+/+/+	st	st	-				
859				st	/	-		20.0	0.0	+	16.68	0.0	+	20.2	0.0	+	17.13	0.0	+	+p	+p	+				
860				+p	+	+		19.5	0.0	+	15.95	0.0	+	19.6	0.0	+	16.24	0.0	+	+p	+p	+				
861				+p	+	+		18.7	0.0	+	15.25	0.0	+	19.3	0.0	+	16.08	0.0	+	+p	+p	+				
862				+p	+	+		21.5	0.0	+	18.97	0.0	+	22.2	0.0	+	18.81	0.0	+	+p	+p	+				
863				+p	+	+		0.0	37.3	-	35.46/0.0/0.0/0.0ne	33.17/0.0/0.0/33.53ne	+/i/i/-ne	38.1	37.7	+	37.33/36.94/35.97	33.04/32.91/32.71	+/+/+	st	st	-				
865				st	/	-		38.5	37.3	+	34.79	33.05	+	39.4	37.9	+	37.09	33.01	+	+p	+p	+				
867				+p	+	+		0.0	37.1	-	35.06/0.0/38.18	33.1/32.63/32.59	+/-/+	39.2	37.7	+	38.09/36.93/35.01	32.81/32.67/32.71	+/+/+	st	st	-				
869				st	/	-		19.6	0.0	+	16.08	0.0	+	17.5	0.0	+	14.98	0.0	+	+p	+p	+				
870				+p	+	+		20.2	0.0	+	17.84	0.0	+	19.3	0.0	+	16.20	0.0	+	+p	+p	+				

♦ Analyses performed according to the COFRAC accreditation (Accreditation Testing n°1-0144, scope available on www.cofrac.fr)

N° sample	Level	Contamination level (CFU/sample) VRBG poor plate method	Contamination level (CFU/sample) MPN	ISO 22964* method (10g)			foodproof® Salmonella Plus Cronobacter Detection LyoKit - 5'Nuclease-																			
				CSB		Final result	Number positive samples/Total	PCR result										Confirmation			Number positive samples / Total					
								foodproof® extraction					BAX Real-Time lysis procedure					After 16h ncubation	After 20h incubation	All confirmation tests (16h)						
								BAX Q7 (from Hygiena) – New software version			LC480 CC #17002		BAX Q7 (from Hygiena) – New software version			LC480 CC #17002		CSB	CSB		foodproof® extraction		BAX Real-Time lysis procedure			
								Ct HEX (<i>Cronobacter</i> spp.)	Ct ROX (Internal control)	Result	Ct HEX (<i>Cronobacter</i> spp.)	Ct ROX (Internal control)	Result	Ct HEX (<i>Cronobacter</i> spp.)	Ct ROX (Internal control)	Result <i>Cronobacter</i> spp.	Ct HEX (<i>Cronobacter</i> spp.)	Ct ROX (Internal control)	Result		CCI	CCI	BAX Q7 (from Hygiena) New software	LightCycler 480 II CC #17002	BAX Q7 (from Hygiena) New software	LightCycler 480 II CC #17002
871	Low	0.2	0.8 [0.5-1.4]	st	/	-		0.0	37.2	-	0.0	33.02	-	37.9	37.3	+	35.74/ 38.70/ 35.71	32.88/ 32.89/ 32.72	+/+/+	st	st	-	11/20	11/20	11/20	11/20
873				st	/	-		0/0.0/0.0	0/36.1/37.3	i/-/-	0.0	33.29	-	38.3	37.5	+	35.68/ 37.17/ 37.12	32.99/ 32.72/ 32.65	+/+/+	st	st	-				
875				st	/	-		0.0	37.4	-	34.47/ 0.0/ 0.0	32.97/ 0.0/ 32.61	+/i/-	39.1	37.9	+	36.94/ 36.00/ 35.74	32.86/ 32.64/ 32.94	+/+/+	st	st	-				
877				+p	+	+		25.8	0.0	+	21.99	20.9	+	25.6	0.0	+	23.15	0.0	+	+p	+p	+				
878				+p	+	+		0.0	36.7	-	0.0	33.13	-	38.9	37.6	+	37.56/ 39.58/ 36.38	32.62/ 32.96/ 32.88	+/+/+	st	st	-				
880				+p	+	+		20.8	0.0	+	16.99	0.0	+	19.7	0.0	+	16.76	0.0	+	+p	+p	+				
881				+p	+	+		39.5	37.7	+	35.34/ 35.57/ 0.0	33.32/ 32.85/ 0.0	+/+/i	38.4/0.0/0.0	37.6/37.3/37.9	+/-/-	37.18/ 36.62/ 36.18	33.16	+/+/+	st	st	-				
855	High	2.2	3.0 [1.4-6.4]	+p	+	+	5/5	22.9	0.0	+	19.58	0.0	+	22.5	0	+	19.16	0.0	+	+p	+p	+	4/5	4/5	4/5	4/5
856				+p	+	+		39.9	37.3	+	35.60/ 0.0/ 0.0/ 37.0ne	33.54/ 0.0/ 0.0/ 33.1ne	+/i/i/+ ne	38.6/0.0/0.0	37.5/37.6/37.5	+/-/-	35.31/ 38.02/ 35.94	32.79/ 32.54/ 32.54	+/+/+	st	st	-				
868				+p	+	+		20.0	0.0	+	16.15	0.0	+	17.7	0	+	15.22	0.0	+	+p	+p	+				
872				+p	+	+		20.0	0.0	+	17.64	0.0	+	19.4	0	+	16.49	0.0	+	+p	+p	+				
876				+p	+	+		20.4	0.0	+	17.35	0.0	+	19.3	0	+	16.76	0.0	+	+p	+p	+				

* Analyses performed according to the COFRAC accreditation (Accreditation Testing n°1-0144, scope available on www.cofrac.fr)

Matrix: Dusts from infant formula production site (200g)

Strain: *Cronobacter muytjensii* E888

Seeding protocol with lyophilized strain storage for 2 weeks at ambient temperature

Protocol: Pre-warmed BPW - d 1/10 - 16 h at 37°C ± 1°C

Total viable count: 7.2.10³ CFU/g

UNPAIRED

N° sample	Level	Contamination level (CFU/sample) VRBG poor plate method	Contamination level (CFU/sample) MPN	ISO 22964* method (10g)			foodproof® Salmonella Plus Cronobacter Detection LyoKit - 5'Nuclease-																	
				CSB	Confirmation	Final result	Number positive samples/Total	PCR result												All confirmation	Number positive samples / Total			
								foodproof® extraction						BAX Real-Time lysis procedure							foodproof® extraction		BAX Real-Time lysis procedure	
								BAX Q7 (from Hygiena) – New software version			LC480 CC #17002 run number 20230711			BAX Q7 (from Hygiena) – New software version			LC480 CC #17002							
								Ct HEX (Cronobacter spp.)	Ct ROX (Internal control)	Result	Ct HEX (Cronobacter spp.)	Ct ROX (Internal control)	Result	Ct HEX (Cronobacter spp.)	Ct ROX (Internal control)	Result Cronobacter spp.	Ct HEX (Cronobacter spp.)	Ct ROX (Internal control)	Result.		BAX Q7 (from Hygiena) New software	LightCycler 480 II CC #17002	BAX Q7 (from Hygiena) New software	LightCycler 480 II CC #17002
1521	0	/	/	st	/	-	0/5	0.0	37.9	-	0.0	32.92	-	0	38.3	-	0.0	33.31	-	st	0/5	0/5	0/5	0/5
1524				st	/	-		0.0	37.6	-	0.0	32.69	-	0	38.1	-	0.0	33.10	-	-				
1534				st	/	-		0.0	38.1	-	0.0	32.19	-	0	38.4	-	0.0	33.61	-	st				
1540				st	/	-		0.0	37.8	-	0.0	32.69	-	0	38.2	-	0.0	33.15	-	st				
1541				st	/	-		0.0	37.3	-	0.0	32.88	-	0	38.1	-	0.0	33.18	-	-				
1522	Low	0.4	1.0 [0.6;1.6]	+p	+	+	10/20	0.0	38.0	-	0.0	33.09	-	0	38.4	-	0.0	33.27	-	st	6/20	7/20	7/20	8/20
1523				st	/	-		34.7	37	+	29.51	31.84	+	33.9	37.6	+	30.51	32.67	+	+p				
1525				st	/	-		0.0	37.9	-	0.0	32.60	-	0	38.6	-	0.0	33.36	-	st				
1527				st	/	-		0.0	37.6	-	0.0	32.35	-	0	38	-	0.0	33.05	-	st				
1528				st	/	-		35.3	36.9	+	27.84	32.04	+	33.7	38.2	+	29.20	32.07	+	+p				
1529				+p	+	+		36.1	37.4	+	31.58	32.19	+	36.2	37.9	+	31.58	32.56	+	+p				
1531				+p	+	+		0.0	37.9	-	0.0	32.88	-	0	38.7	-	0.0	33.33	-	st				
1532				st	/	-		0.0	37.6	-	0.0	32.91	-	0	38.3	-	0.0	33.19	-	st				
1533				+p	+	+		0.0	37.2	-	0.0	32.83	-	0	38.6	-	0.0	33.38	-	-				
1535				+p	+	+		0/ 0.0/ 0.0/ 37.0ne	36.8/ 0.0/ 0.0/ 37.4ne	-/i/- /+ne	36.46	32.90	+	38.1	37.7	+	33.51	32.64	+	+p				
1536				st	/	-		0.0/ 0.0/ 0.0	38.1/ 35.8/ 36.2	-/-/-	0/ 0/ 0	32.63/ 33.29/ 32.90	-/-/-	0/ 0/ 0	37.9/ 37.2/ 37.0	-/-/-	37.77	33.20	+	+p				
1537				st	/	-		34.4	37	+	29.39	31.60	+	32.2	36.9	+	28.32	32.08	+	+p				
1538				st	/	-		0.0	37.4	-	0.0	33.43	-	0	38.3	-	0.0	33.49	-	st				
1539				st	/	-		32.1	35.6	+	26.84	27.29	+	29.4	0	+	25.82	0.0	+	+p				
1543				+p	+	+		0.0	37.2	-	0.0	32.98	-	0	38	-	0.0	32.70	-	-				
1544				+p	+	+		35.8	36.7	+	29.81	31.61	+	34.8	38.2	+	30.39	32.14	+	+p				
1546				+p	+	+		0.0	37.3	-	0.0	32.55	-	0	38.1	-	0.0	33.15	-	st				
1547				+p	+	+		0.0	37.4	-	0.0	34.28	-	0	38.2	-	0.0	33.13	-	-				
1549				+p	+	+		0.0	37.1	-	0.0	32.82	-	0	38.3	-	0.0	33.18	-	st				
1550				st	/	-		0.0	37.9	-	0.0	33.51	-	0	38.1	-	0.0	33.22	-	st				
1526	High	1.6	8.8 [3.4;23]	+p	+	+	5/5	33.9	37.9	+	28.54	31.90	+	31.9	37.2	+	34.91	33.23	+	+p	5/5	5/5	5/5	5/5
1530				+p	+	+		33.5	36.8	+	28.91	33.41	+	32.9	37.8	+	28.20	32.11	+	+p				
1542				+p	+	+		33.1	36.7	+	27.61	32.61	+	31.2	36.8	+	27.27	31.88	+	+m				
1545				+p	+	+		32.1	36.0	+	27.56	30.95	+	31.5	37.7	+	28.39	32.11	+	+p				
1548				+p	+	+		33.5	35.9	+	30.08	32.27	+	31.7	37.8	+	28.72	32.12	+	+p				

* Analyses performed according to the COFRAC accreditation (Accreditation Testing n°1-0144, scope available on www.cofrac.fr)

Appendix 6 – Inclusivity and exclusivity study: raw data

INCLUSIVITY																		
No	Genus	Species	N°	Origin	Inoculation level (CFU/225mL)	foodproof® Salmonella Plus Cronobacter Detection LyoKit - 5'Nuclease-												
						BPW + vancomycin (10mg/L) - 16h at 37°C±1°C												
						foodproof® extraction						BAX lysis						Confirmation
						BAX Q7			LC480 II			BAX Q7			LC480 II			CSB
						Ct HEX (target)	Ct ROX (IC)	Result	Ct HEX (target)	Ct ROX (IC)	Result	Ct HEX (target)	Ct ROX (IC)	Result	Ct HEX (target)	Ct ROX (IC)	Result	CCI (10µl)
1	<i>Cronobacter</i>	<i>dublinensis</i>	DSM18705	Dairy Product	21	15.1	0.0	+	13.57	0	+	18.6	0.0	+	15.78	0.0	+	+
2	<i>Cronobacter</i>	<i>malonaticus</i>	DSM18702	Dairy Product	13	0.0 / 18.4	0.0 / 0.0	i / +	15.68	0	+	18.1	0.0	+	15.00	0.0	+	+(small colonies)
3	<i>Cronobacter</i>	<i>malonaticus</i>	Ad1708	Dairy Product	38	15.7	0.0	+	13.99	0	+	18.7	0.0	+	14.92	0.0	+	+
4	<i>Cronobacter</i>	<i>muytjensii</i>	CIP103581	/	21	16.3	0.0	+	13.84	0	+	18.0	0.0	+	15.14	0.0	+	+
5	<i>Cronobacter</i>	<i>sakazakii</i>	Ad939	Infant formula	8	18.1	0.0	+	15.57	16.39 (uncertain)	+	18.9	0.0	+	13.82	0.0	+	+
6	<i>Cronobacter</i>	<i>sakazakii</i>	Ad940	Infant formula	29	15.8	0.0	+	12.60	0	+	19.2	0.0	+	14.91	0.0	+	+
7	<i>Cronobacter</i>	<i>sakazakii</i>	Ad941	Infant formula	29	16.5	0.0	+	14.28	0	+	19.3	0.0	+	15.40	0.0	+	+
8	<i>Cronobacter</i>	<i>sakazakii</i>	Ad942	Infant formula	50	0.0 / 17.6	0.0 / 0.0	i / +*	13.66	0	+	19.0	0.0	+	14.62	0.0	+	+
9	<i>Cronobacter</i>	<i>sakazakii</i>	Ad943	Infant formula	13	16.2	0.0	+	13.6	0	+	26.0	0.0	+	14.81	0.0	+	+
10	<i>Cronobacter</i>	<i>sakazakii</i>	Ad944	Infant formula	16	15.9	0.0	+	13.91	0	+	18.5	0.0	+	15.58	0.0	+	+
11	<i>Cronobacter</i>	<i>sakazakii</i>	Ad945	Infant formula	17	15.7	0.0	+	13.94	0	+	19.7	0.0	+	15.22	0.0	+	+
12	<i>Cronobacter</i>	<i>sakazakii</i>	Ad946	Infant formula	6	0 / 21.3	0.0 / 0.0	i / +*	16.67	0	+	19.0	0.0	+	14.75	0.0	+	+
13	<i>Cronobacter</i>	<i>sakazakii</i>	Ad947	Infant formula	11	17.5	0.0	+	15.72	0	+	18.8	0.0	+	15.16	0.0	+	+
14	<i>Cronobacter</i>	<i>sakazakii</i>	Ad948	Infant formula	15	0.0 / 18.0	0.0 / 0.0	i / +*	13.66	0	+	17.9	0.0	+	14.47	0.0	+	+
15	<i>Cronobacter</i>	<i>sakazakii</i>	Ad949	Infant formula	18	16.5	0.0	+	13.66	0	+	18.8	0.0	+	14.32	0.0	+	+
16	<i>Cronobacter</i>	<i>sakazakii</i>	Ad950	Infant formula	45	14.5	0.0	+	13.44	0	+	17.6	0.0	+	14.65	0.0	+	+
17	<i>Cronobacter</i>	<i>sakazakii</i>	Ad951	Infant formula	12	16.4	0.0	+	13.90	0	+	20.3	0.0	+	15.04	0.0	+	+
18	<i>Cronobacter</i>	<i>sakazakii</i>	Ad952	Infant formula	51	15.6	0.0	+	12.86	0.0	+	18.6	0.0	+	15.01	0.0	+	+
19	<i>Cronobacter</i>	<i>sakazakii</i>	Ad953	Infant formula	26	16.5	0.0	+	13.92	0	+	19.0	0.0	+	15.20	0.0	+	+
20	<i>Cronobacter</i>	<i>sakazakii</i>	Ad963	Infant formula	15	16.6	0.0	+	13.83	0	+	19.9	0.0	+	15.63	0.0	+	+
21	<i>Cronobacter</i>	<i>sakazakii</i>	95	Infant formula	35	0.0 / 17.3	0.0 / 0.0	i / +*	13.00	0	+	18.4	0.0	+	14.81	0.0	+	+
22	<i>Cronobacter</i>	<i>sakazakii</i>	Ad704	Infant formula	27	14.8	0.0	+	12.99	0	+	19.0	0.0	+	15.03	0.0	+	+(pale colonies)
23	<i>Cronobacter</i>	<i>sakazakii</i>	Ad831	Infant formula	20	16.3	0.0	+	14.31	0	+	17.8	0.0	+	14.98	0.0	+	+
24	<i>Cronobacter</i>	<i>sakazakii</i>	Ad829	Infant formula	29	15.7	0.0	+	14.23	0	+	19.2	0.0	+	14.97	0.0/0.0/0.0	+	+
25	<i>Cronobacter</i>	<i>sakazakii</i>	Ad916	Infant formula	31	16.8	0.0	+	14.08	0	+	19.3	0.0	+	15.23	0.0	+	+
26	<i>Cronobacter</i>	<i>sakazakii</i>	Ad893	Infant formula	31	16.4	0	+	13.91	0	+	17	0.0	+	14.01	0.0	+	+
27	<i>Cronobacter</i>	<i>sakazakii</i>	Ad894	Infant formula	31	17.2	0	+	14.73	0	+	18.9	0.0	+	15.70	0.0/0.0/0.0	+	+
28	<i>Cronobacter</i>	<i>sakazakii</i>	Ad895	Infant formula	29	16.4	0	+	14.04	0	+	17.4	0.0	+	14.38	0.0	+	+
29	<i>Cronobacter</i>	<i>sakazakii</i>	Ad896	Infant formula	40	16.4	0	+	11.99	0	+	17.1	0.0	+	14.28	0.0	+	+
30	<i>Cronobacter</i>	<i>sakazakii</i>	Ad897	Infant formula	25	16.5	0	+	13.94	0	+	17.3	0.0	+	14.64	0.0	+	+
31	<i>Cronobacter</i>	<i>sakazakii</i>	Ad898	Infant formula	28	16.8	0	+	14.56	0	+	18.6	0.0	+	14.86	0.0	+	+
32	<i>Cronobacter</i>	<i>dublinensis lactaridi</i>	DSMZ18707 T	Dairy Product	27	17.4	0	+	14.33	0	+	17.3	0.0	+	14.05	0.0	+	+
33	<i>Cronobacter</i>	<i>dublinensis lausannensis</i>	DSMZ 18706 T	Dairy Product	38	17	0	+	14.49	0	+	18	0.0	+	15.05	0.0	+	+
34	<i>Cronobacter</i>	<i>sakazakii</i>	Ad1418	Infant formula	19	16.6	0	+	13.74	0	+	17.8	0.0	+	14.67	0.0	+	+
35	<i>Cronobacter</i>	<i>sakazakii</i>	Ad1419	Infant formula	34	17.8	0	+	11.75	0	+	17.3	0.0	+	14.98	0.0	+	+
36	<i>Cronobacter</i>	<i>sakazakii</i>	Ad1420	Infant formula	35	16.3	0	+	10.73	0	+	17.3	0.0	+	14.13	0.0	+	+
37	<i>Cronobacter</i>	<i>sakazakii</i>	Ad1421	Infant formula	13	16.6	0	+	14.17	0	+	17.3	0.0	+	14.7	0.0	+	+
38	<i>Cronobacter</i>	<i>sakazakii</i>	Ad1424	Infant formula	19	16.8	0	+	14.90	0	+	17.5	0.0	+	14.69	0.0	+	+
39	<i>Cronobacter</i>	<i>sakazakii</i>	Ad1433	Infant formula	51	17.1	0	+	14.88	0	+	17.9	0.0	+	14.95	0.0	+	+
40	<i>Cronobacter</i>	<i>sakazakii</i>	Ad1434	Infant formula	30	16.2	0	+	13.89	0	+	19.5	0.0	+	16.39	0.0	+	+
41	<i>Cronobacter</i>	<i>sakazakii</i>	Ad1435	Infant formula	9	16.6	0	+	0.0/16.84	0.0/0.0	i/+*	18.2	0.0	+	15.88	0.0	+	+
42	<i>Cronobacter</i>	<i>sakazakii</i>	Ad939	Infant formula	57	16.8	0	+	14.68	16.04 (uncertain)	+	17.5	0.0	+	14.45	0.0	+	+

INCLUSIVITY																		
No	Genus	Species	N°	Origin	Inoculation level (CFU/225mL)	foodproof® Salmonella Plus Cronobacter Detection LyoKit - 5'Nuclease-												
						BPW + vancomycin (10mg/L) - 16h at 37°C±1°C												
						foodproof® extraction						BAX lysis						Confirmation
						BAX Q7			LC480 II			BAX Q7			LC480 II			CSB
						Ct HEX (target)	Ct ROX (IC)	Result	Ct HEX (target)	Ct ROX (IC)	Result	Ct HEX (target)	Ct ROX (IC)	Result	Ct HEX (target)	Ct ROX (IC)	Result	CCI (10µl)
43	Cronobacter	malonaticus	E684	Food	11	17.1	0	+	13.02	0	+	18	0.0	+	14.94	0.0	+	+
44	Cronobacter	malonaticus	E752	Baby food	40	15.9	0	+	11.55	0	+	17	0.0	+	14.63	0.0	+	+
45	Cronobacter	muytjensii	E888	Milk powder	23	16.2	0	+	13.45	0	+	17.6	0.0	+	14.60	0.0	+	+
46	Cronobacter	muytjensii	E769	Milk powder	17	16.9	0	+	13.81	0	+	18.8	0.0	+	15.31	0.0	+	+
47	Cronobacter	dublinensis subsp dublinensis	LMG 23823T	Environmental sample	20	16.1	0	+	13.78	0	+	17.8	0.0	+	15.49	0.0	+	+
48	Cronobacter	dublinensis subsp lausanensis	E798	/	14	16.7	0	+	13.27	0	+	21.8	0.0	+	18.17	0.0	+	+
49	Cronobacter	universalis	NCTC 9529T	Water	46	16.7	0	+	14.05	0	+	18	0.0	+	15.18	0.0	+	+
50	Cronobacter	condimenti	LMG 26250T	Spiced meat	23	18.4/19.1	0	+	0/17.56	0/0	i/+	21.3	0.0	+	15.72	0.0	+	+

*: d1:5

EXCLUSIVITY																	
No	Genus	Species	N°	Origin	Inoculation level (CFU/mL)	foodproof® Salmonella Plus Cronobacter Detection LyoKit - 5'Nuclease-											
						BPW + vancomycin (10mg/L) - 16h at 37°C±1°C											
						foodproof® extraction						BAX lysis					
						BAX Q7			LC480 II			BAX Q7			LC480 II		
						Ct HEX (target)	Ct ROX (IC)	Result	Ct HEX (target)	Ct ROX (IC)	Result	Ct HEX (target)	Ct ROX (IC)	Result	Ct HEX (target)	Ct ROX (IC)	Result
1	Citrobacter	braakii	Ad833	Beef	3.3 x 10 ⁵	0.0	35.6	-	0.0	30.95	-	0.0	37.2	-	0.0	32.14	-
2	Citrobacter	diversus	Ad173	Dairy product	3.6 x 10 ⁵	0.0	36.1	-	0.0	30.71	-	0.0	36.2	-	0.0	32.36	-
3	Citrobacter	fameri	Ad116	Environmental sample	2.4 x 10 ⁵	0.0 / 0.0	0.0 / 37.3	i / -	0.0	31.87	-	0.0	36.7	-	0.0	33.29	-
4	Citrobacter	freundi	39	Environmental sample	2.5 x 10 ⁵	0.0	36.8	-	0.0	31.63	-	0.0	36.7	-	0.0	32.96	-
5	Citrobacter	koseri	CIP105177	/	2.4 x 10 ⁵	0.0	36.4	-	0.0	31.30	-	0.0	36.6	-	0.0	32.78	-
6	Enterobacter	aerogenes	Ad889	Meat flour	2.1 x 10 ⁵	0.0	36.2	-	0.0	31.16	-	0.0	37.3	-	0.0	33.04	-
7	Enterobacter	agglomerans	11	Dairy product	1.8 x 10 ⁵	0.0	35.7	-	0.0	31.73	-	0.0	36.8	-	0.0	32.84	-
8	Enterobacter	agglomerans	136	Dairy product	1.1 x 10 ⁵	0.0 / 0.0	0.0 / 37.1	i / -*	0.0	32.05	-	0.0	36.8	-	0.0	33.57	-
9	Enterobacter	amnigenus	52	Vegetables	8.6 x 10 ⁵	0.0	35.9/ 37.2	-	0.0	35.9	-	0.0	36.8	-	0.0	32.75	-
10	Enterobacter	amnigenus	129	Raw milk	9.4 x 10 ⁵	0.0	36.1	-	0.0	31.32	-	0.0	37.8	-	0.0	33.02	-
11	Enterobacter	amnigenus	A00C068	Poultry	2.6 x 10 ⁵	0.0	36.2	-	0.0	30.68	-	0.0	37.2	-	0.0	33.18	-
12	Enterobacter	cloacae	51	Vegetables	2.3 x 10 ⁵	0.0	36.3	-	0.0	31.20	-	0.0	36.7	-	0.0	32.66	-
13	Enterobacter	cloacae	10	Dairy product	8.4 x 10 ⁵	0.0	36.2	-	0.0	31.73	-	0.0	37.0	-	0.0	32.98	-
14	Enterobacter	fergusonii	2876	Environmental sample	3.3 x 10 ⁵	0.0	36.3	-	0.0	32.01	-	0.0	36.9	-	0.0	32.94	-
15	Enterobacter	gergoviae	CIP76.1	/	2.5 x 10 ⁵	0.0	35.9	-	0.0	32.01	-	0.0	36.5	-	0.0	32.94	-
16	Enterobacter	helveticus	DSM 18396 T	fruit powder	1.4 x 10 ⁵	0.0	37.4	-	0.0	32.24	-	0.0	37.3	-	0.0	32.57	-
17	Enterobacter	hormaechei	Ad990	Butter	2.2 x 10 ⁵	0.0	36.8	-	0.0	32.26	-	0.0	36.5	-	0.0	32.46	-
18	Enterobacter	intermedius	60	Vegetables	8.8 x 10 ⁴	0.0	36.6	-	0.0/0.0	0.0/32.89	i/-	0.0	36.5	-	0.0	32.99	-
19	Enterobacter	kobei	Ad706	Milk powder	2.2 x 10 ⁵	0.0	36.9	-	0.0	32.80	-	0.0	36.7	-	0.0	32.9	-
20	Escherichia	coli	16	Dairy product	3.2 x 10 ⁵	0.0	36.2	-	0.0	32.68	-	0.0	36.2	-	40.84 (no amp)	32.98	-
21	Escherichia	hermanii	Ad462	Dairy product	2.6 x 10 ⁵	0.0	36.5	-	0.0	32.11	-	0.0	36.3	-	39.56 (no amp)	32.48	-
22	Hafnia	alvei	Ad245	Dairy product	2.9 x 10 ⁵	0.0	36.9	-	0.0	33.08	-	0.0	35.9	-	0.0	32.85	-
23	Klebsiella	pneumoniae	122	Dairy product	2.5 x 10 ⁵	0.0	36.1	-	0.0	32.82	-	0.0	36.1	-	0	32.78	-
24	Kluyvera	oxytoca	MI030497b	Milk powder	2.6 x 10 ⁵	0.0	36.6	-	0.0	32.64	-	0.0	36.5	-	0	32.71	-
25	Leclercia	adecarboxylata	Ad707	Milk powder	6.0 x 10 ⁴	0.0	37.2	-	0.0	31.83	-	0.0	36.6	-	0.0	32.86	-
26	Salmonella	diarizonae SIIIb 65 :c :z	Ad1298	Dairy environmental sample	3.9 x 10 ⁵	0.0	36.5	-	0.0	0.0	-	0.0	36.8	-	0.0	0	-
27	Salmonella	Typhimurium	Ad1333	Dairy product	5.3 x 10 ⁵	0.0	37.2	-	0.0	33.12	-	0.0	34.6	-	0.0	38.59	-
28	Serratia	ficaria	113	Vegetables	2.4 x 10 ⁵	0.0	36.9	-	0.0/0.0	0.0/32.75	i/-	0.0	36	-	0.0	33.03	-
29	Serratia	fonticola	Ad1696	Raw milk	2.6 x 10 ⁵	0.0	36.4	-	0.0	32.68	-	0.0	36.6	-	0.0	33.09	-
30	Yersinia	intermedia	Ad133	Dairy product	3.2 x 10 ⁵	0.0	36.8	-	0.0	32.55	-	0.0	36.9	-	0.0	32.22	-

Appendix 7 - Results obtained by the collaborative laboratories and the expert laboratory

Laboratory
Lactic acid bacteria:

A1
<10 CFU/g

DNA extraction:
Thermocycler:

BAX Lysis
Q7-2481

N°Sample	ISO 22964				foodproof® <i>Salmonella</i> plus <i>Cronobacter</i> Detection LyoKit							Agreement Ref/Alt
					PCR test			PCR final result <i>Cronobacter</i> spp.	Confirmation final result <i>Cronobacter</i> spp. (results of the ISO 22964)	Final result		
	<i>Cronobacter</i> spp. (HEX)		Internal control (ROX)									
	CSB/CCI	Confirmation tests after subculture onto TSA		Final result	Results (+/-)	Cq value	Result or Cq value					
Typical colonies	Oxidase test (+/-)	RAPID ID 32E										
4	-	/	/	-	-	0.0	+37.1	-	-	-	NA	
5	-	/	/	-	-	0.0	+36.0	-	-	-	NA	
9	-	/	/	-	-	0.0	+5.9	-	-	-	NA	
11	-	/	/	-	-	0.0	+36.6	-	-	-	NA	
12	-	/	/	-	-	0.0	+35.9	-	-	-	NA	
18	-	/	/	-	-	0.0	+37.4	-	-	-	NA	
22	-	/	/	-	-	0.0	+37.0	-	-	-	NA	
23	-	/	/	-	-	0.0	+36.2	-	-	-	NA	
1	+	-	<i>Cronobacter sakazakii</i>	+	+	17.4	-0.0	+	+	+	PA	
7	+	-	<i>Cronobacter sakazakii</i>	+	+	17.0	- 0.0	+	+	+	PA	
8	-	/	/	-	-	0.0	+35.5	-	-	-	NA	
10	+	-	<i>Cronobacter sakazakii</i>	+	+	16.8	-0.0	+	+	+	PA	
15	+	-	<i>Cronobacter sakazakii</i>	+	+	16.2	-0.0	+	+	+	PA	
17	+	-	<i>Cronobacter sakazakii</i>	+	+	17.7	-0.0	+	+	+	PA	
19	+	-	<i>Cronobacter sakazakii</i>	+	+	17.1	-0.0	+	+	+	PA	
24	-	/	/	-	-	0.0	+37.3	-	-	-	NA	
2	-	/	/	-	-	0.0	+36.7	-	-	-	NA	
3	-	/	/	-	-	0.0	+35.6	-	-	-	NA	
6	-	/	/	-	-	0.0	+34.9	-	-	-	NA	
13	-	/	/	-	-	0.0	+38.0	-	-	-	NA	
14	+	-	<i>Cronobacter sakazakii</i>	+	+	17.0	-0.0	+	+	+	PA	
16	+	-	<i>Cronobacter sakazakii</i>	+	+	16.2	-0.0	+	+	+	PA	
20	+	-	<i>Cronobacter sakazakii</i>	+	+	17.6	-0.0	+	+	+	PA	
21	+	-	<i>Cronobacter sakazakii</i>	+	+	17.7	-0.0	+	+	+	PA	

Laboratory **A2**
Lactic acid bacteria: <10 CFU/g

DNA extraction:	BAX Lysis
Thermocycler:	Q7-2481

N°Sample	ISO 22964				foodproof® <i>Salmonella</i> plus <i>Cronobacter</i> Detection LyoKit						Agreement Ref/Alt
					PCR test			PCR final result <i>Cronobacter</i> spp.	Confirmation final result <i>Cronobacter</i> spp. (results of the ISO 22964)	Final result	
	CSB/CCI	Confirmation tests after subculture onto TSA		Final result	<i>Cronobacter</i> spp. (HEX)		Internal control (ROX)				
		Typical colonies	Oxidase test (+/-)		RAPID ID 32E	Results (+/-)					
25	-	/	/	-	+/-	32.9/0.0	+34.4/+38.7	+/-	-	-	PD _{FP(alt)}
26	-	/	/	-	+/-	34.7/0.0	+36.9/+38.2	+/-	-	-	PD _{FP(alt)}
32	-	/	/	-	-	0.0	+37.7	-	-	-	NA
35	-	/	/	-	-	0.0	+36.0	-	-	-	NA
38	-	/	/	-	-	0.0	+37.4	-	-	-	NA
43	-	/	/	-	-	0.0	+37.9	-	-	-	NA
44	-	/	/	-	-	0.0	+36.9	-	-	-	NA
47	-	/	/	-	-	0.0	+36.1	-	-	-	NA
27	+	-	<i>Cronobacter sakazakii</i>	+	+	16.1	-0.0	+	+	+	PA
29	+	-	<i>Cronobacter sakazakii</i>	+	+	16.6	-0.0	+	+	+	PA
30	-	/	/	-	-	0.0	+37.1	-	-	-	NA
36	-	/	/	-	-	0.0	+36.8	-	-	-	NA
37	+	-	<i>Cronobacter sakazakii</i>	+	+	17.1	-0.0	+	+	+	PA
41	+	-	<i>Cronobacter sakazakii</i>	+	+	16.4	-0.0	+	+	+	PA
45	+	-	<i>Cronobacter sakazakii</i>	+	+	16.4	-0.0	+	+	+	PA
48	-	/	/	-	-	0.0	+38.4	-	-	-	NA
28	+	-	<i>Cronobacter sakazakii</i>	+	+	16.3	-0.0	+	+	+	PA
31	-	/	/	-	-	0.0	+37.4	-	-	-	NA
33	+	-	<i>Cronobacter sakazakii</i>	+	+	17.3	-0.0	+	+	+	PA
34	-	/	/	-	+/-	34.7/0.0	+36.9 / +36.3	+/-	-	-	PD _{FP(alt)}
39	-	/	/	-	-	0.0	+38.9	-	-	-	NA
40	-	/	/	-	-	0.0	+37.7	-	-	-	NA
42	-	/	/	-	-	0.0	+36.6	-	-	-	NA
46	-	/	/	-	-	0.0	+ 36.4	-	-	-	NA

Laboratory C1
Lactic acid bacteria: 2.4.10⁶ CFU/g

DNA extraction: StarPrep Three
Thermocycler: BAX Q7

N°Sample	ISO 22964			foodproof® <i>Salmonella</i> plus <i>Cronobacter</i> Detection LyoKit							Agreement Ref/Alt
				PCR test			PCR final result <i>Cronobacter</i> spp.	Confirmation final result <i>Cronobacter</i> spp. (results of the ISO 22964)	Final result		
	CSB/CCI	Confirmation tests after subculture onto TSA		Final result	<i>Cronobacter</i> spp. (HEX)					Internal control (ROX)	
		Typical colonies	Oxidase test (+/-)		RAPID ID 32E	Results (+/-)					
4	-	/	/	-	-	0.0	36.8	-	-	-	NA
5	-	/	/	-	-	0.0	37.0	-	-	-	NA
9	-	/	/	-	-	0.0	37.3	-	-	-	NA
11	-	/	/	-	-	0.0	36.6	-	-	-	NA
12	-	/	/	-	-	0.0	36.8	-	-	-	NA
18	-	/	/	-	-	0.0	36.7	-	-	-	NA
22	-	/	/	-	-	0.0	36.7	-	-	-	NA
23	-	/	/	-	-	0.0	38.8	-	-	-	NA
1	-	/	/	-	-	0.0	37.2	-	-	-	NA
7	+	-	<i>Cronobacter sakazakii</i>	+	+	25.0	0.0	+	+	+	PA
8	+	-	<i>Cronobacter sakazakii</i>	+	+	18.7	0.0	+	+	+	PA
10	+	-	<i>Cronobacter sakazakii</i>	+	+	15.1	0.0	+	+	+	PA
15	-	/	/	-	-	0.0	36.5	-	-	-	NA
17	-	/	/	-	-	0.0	37.3	-	-	-	NA
19	+	-	<i>Cronobacter sakazakii</i>	+	+	15.6	0.0	+	+	+	PA
24	+	-	<i>Cronobacter sakazakii</i>	+	+	16.0	0.0	+	+	+	PA
2	+	-	<i>Cronobacter sakazakii</i>	+	+	18.1	0.0	+	+	+	PA
3	-	/	/	-	-	0.0	37.4	-	-	-	NA
6	+	-	<i>Cronobacter sakazakii</i>	+	+	15.8	0.0	+	+	+	PA
13	-	/	/	-	-	0.0	35.9	-	-	-	NA
14	-	/	/	-	-	0.0	37.3	-	-	-	NA
16	+	-	<i>Cronobacter sakazakii</i>	+	+	18.9	0.0	+	+	+	PA
20	+	-	<i>Cronobacter sakazakii</i>	+	+	22.4	0.0	+	+	+	PA
21	-	/	/	-	-	0.0	36.1	-	-	-	NA

Laboratory **C2**
Lactic acid bacteria: 2.6.10⁶CFU/g

DNA extraction:	BAX lysis
Thermocycler:	BAX Q7

N°Sample	ISO 22964				foodproof® <i>Salmonella</i> plus <i>Cronobacter</i> Detection LyoKit							Agreement Ref/Alt
					PCR test			PCR final result <i>Cronobacter</i> spp.	Confirmation final result <i>Cronobacter</i> spp. (results of the ISO 22964)	Final result		
	CSB/CCI	Confirmation tests after subculture onto TSA		Final result	<i>Cronobacter</i> spp. (HEX)		Internal control (ROX)					
		Typical colonies	Oxidase test (+/-)		RAPID ID 32E	Results (+/-)					Cq value	
25	-	/	/	-	-	0.0	38.3	-	-	-	NA	
26	-	/	/	-	-	0.0	38.2	-	-	-	NA	
32	-	/	/	-	-	0.0	37.9	-	-	-	NA	
35	-	/	/	-	-	0.0	38.1	-	-	-	NA	
38	-	/	/	-	-	0.0	38.0	-	-	-	NA	
43	-	/	/	-	-	0.0	37.8	-	-	-	NA	
44	-	/	/	-	-	0.0	37.8	-	-	-	NA	
47	-	/	/	-	-	0.0	37.8	-	-	-	NA	
27	-	/	/	-	-	0.0	39.0	-	-	-	NA	
29	-	/	/	-	-	0.0	38.5	-	-	-	NA	
30	+	-	<i>Cronobacter sakazakii</i>	+	+	24.6	0.0	+	+	+	PA	
36	-	/	/	-	-	0.0	38.0	-	-	-	NA	
37	-	/	/	-	-	0.0	38.3	-	-	-	NA	
41	-	/	/	-	-	0.0	38.4	-	-	-	NA	
45	+	-	<i>Cronobacter sakazakii</i>	+	+	17.9	0.0	+	+	+	PA	
48	+	-	<i>Cronobacter sakazakii</i>	+	+	18.6	0.0	+	+	+	PA	
28	-	/	/	-	-	0.0	38.6	-	-	-	NA	
31	+	-	<i>Cronobacter sakazakii</i>	+	+	19.9	0.0	+	+	+	PA	
33	+	-	<i>Cronobacter sakazakii</i>	+	+	27.2	0.0	+	+	+	PA	
34	-	/	/	-	-	0.0	38.1	-	-	-	NA	
39	-	/	/	-	-	0.0	37.3	-	-	-	NA	
40	+	-	<i>Cronobacter sakazakii</i>	+	+	19.7	0.0	+	+	+	PA	
42	-	/	/	-	-	0.0	38.3	-	-	-	NA	
46	-	/	/	-	-	0.0	37.7	-	-	-	NA	

Laboratory **D**
Lactic acid bacteria: 1.5.10²CFU/g

DNA extraction: BAX Lysis
Thermocycler: Q7

N°Sample	ISO 22964				foodproof® <i>Salmonella</i> plus <i>Cronobacter</i> Detection LyoKit						Agreement Ref/Alt
	CSB/CCI	Confirmation tests after subculture onto TSA		Final result	PCR test		PCR final result <i>Cronobacter</i> spp.	Confirmation final result <i>Cronobacter</i> spp. (results of the ISO 22964)	Final result		
		Oxidase test (+/-)	RAPID ID 32E		Cronobacter spp. (HEX)	Internal control (ROX)					
	Typical colonies				Results (+/-)	Cq value	Result or Cq value				
4	-	/	/	-	-	0.0	37.3	-	-	-	NA
5	-	/	/	-	-	0.0	38.1	-	-	-	NA
9	-	/	/	-	-	0.0	38.8	-	-	-	NA
11	-	/	/	-	-	0.0	38.7	-	-	-	NA
12	-	/	/	-	-	0.0	38.3	-	-	-	NA
18	-	/	/	-	-	0.0	38.6	-	-	-	NA
22	-	/	/	-	-	0.0	38.1	-	-	-	NA
23	-	/	/	-	-	0.0	38.4	-	-	-	NA
1	+	-	<i>Cronobacter sakazakii</i>	+	+	18.1	0.0	+	+	+	PA
7	-	/	/	-	-	0.0	38.9	-	-	-	NA
8	+	-	<i>Cronobacter sakazakii</i>	+	+	18.6	0.0	+	+	+	PA
10	-	/	/	-	-	0.0	38.7	-	-	-	NA
15	+	-	<i>Cronobacter sakazakii</i>	+	+	20.3	0.0	+	+	+	PA
17	-	/	/	-	-	0.0	38.9	-	-	-	NA
19	+	-	<i>Cronobacter sakazakii</i>	+	+	31.8	36.7	+	+	+	PA
24	+	-	<i>Cronobacter sakazakii</i>	+	+	18.8	0.0	+	+	+	PA
2	+	-	<i>Cronobacter sakazakii</i>	+	+	18.8	0.0	+	+	+	PA
3	-	/	/	-	-	0.0	38.2	-	-	-	NA
6	-	/	/	-	-	0.0	39.4	-	-	-	NA
13	+	-	<i>Cronobacter sakazakii</i>	+	+	32.2	36.8	+	+	+	PA
14	-	/	/	-	-	0.0	38.0	-	-	-	NA
16	+	-	<i>Cronobacter sakazakii</i>	+	+	33.4	36.5	+	+	+	PA
20	-	/	/	-	-	0.0	38.3	-	-	-	NA
21	+	-	<i>Cronobacter sakazakii</i>	+	+	22.0	0.0	+	+	+	PA

Laboratory **E**
Lactic acid bacteria: 30 CFU/g

DNA extraction: StarPrep Three Kit
Thermocycler: LC480 II

N°Sample	ISO 22964			foodproof® <i>Salmonella</i> plus <i>Cronobacter</i> Detection LyoKit							Agreement Ref/Alt
				PCR test			PCR final result <i>Cronobacter</i> spp.	Confirmation final result <i>Cronobacter</i> spp. (results of the ISO 22964)	Final result		
	CSB/CCI	Confirmation tests after subculture onto TSA		Final result	<i>Cronobacter</i> spp. (HEX)					Internal control (ROX)	
		Typical colonies	Oxidase test (+/-)		RAPID ID 32E	Results (+/-)					
4	-	/	/	-	-	0.0	34.47	-	-	-	NA
5	-	/	/	-	-	0.0	34.27	-	-	-	NA
9	-	/	/	-	-	0.0	34.03	-	-	-	NA
11	-	/	/	-	-	0.0	34.36	-	-	-	NA
12	-	/	/	-	-	0.0	34.35	-	-	-	NA
18	-	/	/	-	-	0.0	33.98	-	-	-	NA
22	-	/	/	-	-	0.0	34.01	-	-	-	NA
23	-	/	/	-	-	0.0	34.25	-	-	-	NA
1	-	/	/	-	-	0.0	33.96	-	-	-	NA
7	-	/	/	-	-	0.0	34.16	-	-	-	NA
8	-	/	/	-	-	0.0	34.26	-	-	-	NA
10	-	/	/	-	-	0.0	33.93	-	-	-	NA
15	-	/	/	-	-	0.0	34.15	-	-	-	NA
17	-	/	/	-	-	0.0	34.44	-	-	-	NA
19	-	/	/	-	-	0.0	34.11	-	-	-	NA
24	+	-	<i>Cronobacter sakazakii</i>	+	+	12.53	-	+	+	+	PA
2	-	/	/	-	-	0.0	33.97	-	-	-	NA
3	-	/	/	-	-	0.0	34.19	-	-	-	NA
6	-	/	/	-	-	0.0	34.11	-	-	-	NA
13	+	-	<i>Cronobacter sakazakii</i>	+	+	14.84	-	+	+	+	PA
14	+	-	<i>Cronobacter sakazakii</i>	+	+	12.91	-	+	+	+	PA
16	-	/	/	-	-	0.0	34.45	-	-	-	NA
20	-	/	/	-	-	0.0	34.04	-	-	-	NA
21	-	/	/	-	-	0.0	34.40	-	-	-	NA

Laboratory F
Lactic acid bacteria: 8.2.10⁶ CFU/g

DNA extraction: StarPrep Three Kit
Thermocycler: LC480 II

N°Sample	ISO 22964			foodproof® <i>Salmonella</i> plus <i>Cronobacter</i> Detection LyoKit							Agreement Ref/Alt
	CSB/CCI	Confirmation tests after subculture onto TSA		Final result	PCR test		Internal control (ROX)	PCR final result <i>Cronobacter</i> spp.	Confirmation final result <i>Cronobacter</i> spp. (results of the ISO 22964)	Final result	
		Typical colonies	Oxidase test (+/-)		RAPID ID 32E	Results (+/-)					
4	-	/	/	-	-	0.0	+	-	-	-	NA
5	-	/	/	-	-	0.0	+	-	-	-	NA
9	-	/	/	-	-	0.0	+	-	-	-	NA
11	-	/	/	-	-	0.0	+	-	-	-	NA
12	-	/	/	-	-	0.0	+	-	-	-	NA
18	-	/	/	-	-	0.0	+	-	-	-	NA
22	-	/	/	-	-	0.0	+	-	-	-	NA
23	-	/	/	-	-	0.0	+	-	-	-	NA
1	-	/	/	-	-	0.0	+	-	-	-	NA
7	-	/	/	-	-	0.0	+	-	-	-	NA
8	+	-	<i>Cronobacter sakazakii</i>	+	+	15.47	+	+	+	+	PA
10	+	-	<i>Cronobacter sakazakii</i>	+	-/+/(+)(ne)	0.0/13.02/12.92/12.76 (ne)	+/13.04/12.94/12.66(ne)	-/+/(+)(ne)	+	-	ND _{FN(alt)}
15	-	/	/	-	-	0.0	+	-	-	-	NA
17	+	-	<i>Cronobacter sakazakii</i>	+	+	22.93	+	+	+	+	PA
19	-	/	/	-	-	0.0	+	-	-	-	NA
24	+	-	<i>Cronobacter sakazakii</i>	+	+	14.89	+	+	+	+	PA
2	-	/	/	-	-	0.0	+	-	-	-	NA
3	-	/	/	-	-	0.0	+	-	-	-	NA
6	-	/	/	-	-	0.0	+	-	-	-	NA
13	+	-	<i>Cronobacter sakazakii</i>	+	+	15.24	+	+	+	+	PA
14	+	-	<i>Cronobacter sakazakii</i>	+	+	16.97	+	+	+	+	PA
16	+	-	<i>Cronobacter sakazakii</i>	+	+	18.8	+	+	+	+	PA
20	-	/	/	-	-	0.0	+	-	-	-	NA
21	-	/	/	-	-	0.0	+	-	-	-	NA

(ne): new DNA extract

Laboratory **G**
Lactic acid bacteria: <10 CFU/g

DNA extraction: StarPrep Three Kit
Thermocycler: LC480 II

N°Sample	ISO 22964			foodproof® <i>Salmonella</i> plus <i>Cronobacter</i> Detection LyoKit							Agreement Ref/Alt
	CSB/CCI	Confirmation tests after subculture onto TSA		Final result	PCR test		Internal control (ROX)	PCR final result <i>Cronobacter</i> spp.	Confirmation final result <i>Cronobacter</i> spp. (results of the ISO 22964)	Final result	
		Typical colonies	Oxidase test (+/-)		RAPID ID 32E	Results (+/-)					
4	-	/	/	-	-	0.0	33.05	-	-	-	NA
5	-	/	/	-	-	0.0	31.62	-	-	-	NA
9	-	/	/	-	-	0.0	32.95	-	-	-	NA
11	-	/	/	-	-	0.0	32.30	-	-	-	NA
12	-	/	/	-	-	0.0	32.96	-	-	-	NA
18	-	/	/	-	-	0.0	32.44	-	-	-	NA
22	-	/	/	-	-	0.0	32.97	-	-	-	NA
23	-	/	/	-	-	0.0	32.85	-	-	-	NA
1	-	/	/	-	-	0.0	33.16	-	-	-	NA
7	-	/	/	-	-	0.0	32.88	-	-	-	NA
8	+	-	<i>Cronobacter sakazakii</i>	+	+	12.66	0.0	+	+	+	PA
10	-	/	/	-	-	0.0	32.84	-	-	-	NA
15	+	-	<i>Cronobacter sakazakii</i>	+	+	14.04	0.0	+	+	+	PA
17	-	/	/	-	-	0.0	33.05	-	-	-	NA
19	+	-	<i>Cronobacter sakazakii</i>	+	+	13.01	0.0	+	+	+	PA
24	+	-	<i>Cronobacter sakazakii</i>	+	+	33.97	30.50	+	+	+	PA
2	-	/	/	-	-	0.0	33.06	-	-	-	NA
3	-	/	/	-	-	0.0	32.75	-	-	-	NA
6	+	-	<i>Cronobacter sakazakii</i>	+	+	12.60	0.0	+	+	+	PA
13	+	-	<i>Cronobacter sakazakii</i>	+	+	13.52	0.0	+	+	+	PA
14	-	/	/	-	-	0.0	0.0	-	-	-	NA
16	+	-	<i>Cronobacter sakazakii</i>	+	+	15.35	0.0	+	+	+	PA
20	+	-	<i>Cronobacter sakazakii</i>	+	+	12.12	0.0	+	+	+	PA
21	+	-	<i>Cronobacter sakazakii</i>	+	+	17.54	0.0	+	+	+	PA

Laboratory H
Lactic acid bacteria: <10 CFU/g

DNA extraction:	StarPrep Three kit
Thermocycler:	LC480

N°Sample	ISO 22964			foodproof® <i>Salmonella</i> plus <i>Cronobacter</i> Detection LyoKit							Agreement Ref/Alt
				PCR test			PCR final result <i>Cronobacter</i> spp.	Confirmation final result <i>Cronobacter</i> spp. (results of the ISO 22964)	Final result		
	CSB/CCI	Confirmation tests after subculture onto TSA		Final result	<i>Cronobacter</i> spp. (HEX)					Internal control (ROX)	
		Typical colonies	Oxidase test (+/-)		RAPID ID 32E	Results (+/-)				Cq value	
4	-	/	/	-	-	NA	32.85	-	-	-	NA
5	-	/	/	-	-	NA	33.02	-	-	-	NA
9	-	/	/	-	-	NA	32.80	-	-	-	NA
11	-	/	/	-	-	NA	32.97	-	-	-	NA
12	-	/	/	-	-	NA	32.60	-	-	-	NA
18	-	/	/	-	-	NA	33.00	-	-	-	NA
22	-	/	/	-	-	NA	32.78	-	-	-	NA
23	-	/	/	-	-	NA	32.59	-	-	-	NA
1	-	/	/	-	-	NA	32.88	-	-	-	NA
7	-	/	/	-	-	NA	32.77	-	-	-	NA
8	-	/	/	-	-	NA	32.85	-	-	-	NA
10	-	/	/	-	-	NA	32.84	-	-	-	NA
15	-	/	/	-	-	NA	32.81	-	-	-	NA
17	+	-	<i>Cronobacter sakazakii</i>	+	+	13.55	NA	+	+	+	PA
19	+	-	<i>Cronobacter sakazakii</i>	+	+	12.92	NA	+	+	+	PA
24	-	/	/	-	-	NA	32.82	-	-	-	NA
2	+	-	<i>Cronobacter sakazakii</i>	+	+	11.79	NA	+	+	+	PA
3	-	/	/	-	-	NA	32.89	-	-	-	NA
6	+	-	<i>Cronobacter sakazakii</i>	+	+	12.79	NA	+	+	+	PA
13	-	/	/	-	-	NA	33.15	-	-	-	NA
14	+	-	<i>Cronobacter sakazakii</i>	+	+	12.51	NA	+	+	+	PA
16	-	/	/	-	-	NA	32.64	-	-	-	NA
20	-	/	/	-	-	NA	33.01	-	-	-	NA
21	-	/	/	-	-	NA	32.81	-	-	-	NA

Laboratory I1
Lactic acid bacteria: <10 CFU/g

DNA extraction: Star Prep Three kit
Thermocycler: Bax Q7

N°Sample	ISO 22964				foodproof® Salmonella plus Cronobacter Detection LyoKit						Agreement Ref/Alt
					PCR test			PCR final result Cronobacter spp.	Confirmation final result Cronobacter spp. (results of the ISO 22964)	Final result	
	Cronobacter spp. (HEX)		Internal control (ROX)								
	CSB/CCI	Confirmation tests after subculture onto TSA		Final result	Results (+/-)	Cq value	Result or Cq value				
Typical colonies	Oxidase test (+/-)	RAPID ID 32E									
4	-	/	/	-	-	0.0	36.9	-	-	-	NA
5	-	/	/	-	+/-/-(ne)	39.4/35.3/0.0(ne)	36.5/36.5/38.3(ne)	+/-/-(ne)	-	-	PD _{FP(alt)}
9	-	/	/	-	-	-	37.2	-	-	-	NA
11	-	/	/	-	-	-	36.3	-	-	-	NA
12	-	/	/	-	-	-	36.6	-	-	-	NA
18	-	/	/	-	-	-	36.7	-	-	-	NA
22	-	/	/	-	+	31.7	34.4	+	-	-	NA
23	-	/	/	-	+/-/-(ne)	39.9/35.9/0.0(ne)	36.9/36.6/36.9(ne)	+/-/-(ne)	-	-	PD _{FP(alt)}
1	+	-	Cronobacter sakazakii	+	+	16.2	0.0	+	+	+	PA
7	-	/	/	-	-	-	37.3	-	-	-	NA
8	-	/	/	-	-	-	36.3	-	-	-	NA
10	-	/	/	-	-	-	37.3	-	-	-	NA
15	+	-	Cronobacter sakazakii	+	+	16.0	0.0	+	+	+	PA
17	+	-	Cronobacter sakazakii	+	+	15.7	0.0	+	+	+	PA
19	-	/	/	-	-	-	36.4	-	-	-	NA
24	-	/	/	-	-	-	37.5	-	-	-	NA
2	-	/	/	-	+/+/(ne)	37.5/34.3/39.4(ne)	36.8/36.2/37.2(ne)	+/+/(ne)	-	-	PD _{FP(alt)}
3	-	/	/	-	-	0.0	36.9	-	-	-	NA
6	-	/	/	-	-	-	36.6	-	-	-	NA
13	+	-	Cronobacter sakazakii	+	+	16.1	0.0	+	+	+	PA
14	-	/	/	-	-	-	36.9	-	-	-	NA
16	+	-	Cronobacter sakazakii	+	+	15.7	0.0	+	+	+	PA
20	+	-	Cronobacter sakazakii	+	+	16.7	0.0	+	+	+	PA
21	+	-	Cronobacter sakazakii	+	+	16.0	0.0	+	+	+	PA

(ne): new extraction

Laboratory I2
Lactic acid bacteria: <10 CFU/g

DNA extraction: Star Prep Three kit
Thermocycler: Bax Q7

N°Sample	ISO 22964				foodproof® Salmonella plus Cronobacter Detection LyoKit						Agreement Ref/Alt
	CSB/CCI	Confirmation tests after subculture onto TSA		Final result	PCR test		Internal control (ROX)	PCR final result Cronobacter spp.	Confirmation final result Cronobacter spp. (results of the ISO 22964)	Final result	
		Typical colonies	Oxidase test (+/-)		RAPID ID 32E	Results (+/-)					
25	-	/	/	-	-	-	37.4	-	-	-	NA
26	-	/	/	-	-	-	37.0	-	-	-	NA
32	-	/	/	-	-	-	37.3	-	-	-	NA
35	-	/	/	-	-	-	37.1	-	-	-	NA
38	-	/	/	-	-	-	36.6	-	-	-	NA
43	-	/	/	-	+/-/-(ne)	36.0/34.3/0.0(ne)	36.5/36.0/38.4(ne)	+/-/-(ne)	-	-	PD _{FP(alt)}
44	-	/	/	-	-	-	36.5	-	-	-	NA
47	-	/	/	-	-	-	36.7	-	-	-	NA
27	+	-	Cronobacter sakazakii	+	+	21.7	0.0	+	+	+	PA
29	-	/	/	-	+	33.1	35.5	+	-	-	NA
30	+	-	Cronobacter sakazakii	+	+	15.3	0.0	+	+	+	PA
36	+	-	Cronobacter sakazakii	+	+	14.8	0.0	+	+	+	PA
37	-	/	/	-	-	-	36.8	-	-	-	NA
41	-	/	/	-	-	-	37.1	-	-	-	NA
45	-	/	/	-	-	-	36.9	-	-	-	NA
48	-	/	/	-	-	-	36.7	-	-	-	NA
28	+	-	Cronobacter sakazakii	+	+	15.1	0.0	+	+	+	PA
31	+	-	Cronobacter sakazakii	+	+	20.5	0.0	+	+	+	PA
33	+	-	Cronobacter sakazakii	+	+	15.8	0.0	+	+	+	PA
34	-	/	/	-	+/-/-(ne)	39.3/36.0/36.3(ne)	37.1/36.8/36.2(ne)	+/-/-(ne)	-	-	PD _{FP(alt)}
39	-	/	/	-	-	-	36.9	-	-	-	NA
40	-	/	/	-	-	-	36.8	-	-	-	NA
42	-	/	/	-	-	-	36.9	-	-	-	NA
46	+	-	Cronobacter sakazakii	+	+	19.0	0.0	+	+	+	PA

Laboratory J
Lactic acid bacteria: <10 CFU/g

DNA extraction: Bax lysis
Thermocycler: LC480

N°Sample	ISO 22964				foodproof® <i>Salmonella</i> plus <i>Cronobacter</i> Detection LyoKit						Agreement Ref/Alt
	CSB/CCI	Confirmation tests after subculture onto TSA		Final result	PCR test			PCR final result <i>Cronobacter</i> spp.	Confirmation final result <i>Cronobacter</i> spp. (results of the ISO 22964)	Final result	
					<i>Cronobacter</i> spp. (HEX)		Internal control (ROX)				
					Results (+/-)	Cq value	Result or Cq value				
4	-	/	/	-	-	0.0	34.32	-	-	-	NA
5	-	/	/	-	-	0.0	35.30	-	-	-	NA
9	-	/	/	-	-	0.0	33.94	-	-	-	NA
11	-	/	/	-	-	0.0	33.92	-	-	-	NA
12	-	/	/	-	-	0.0	33.92	-	-	-	NA
18	-	/	/	-	-	0.0	34.05	-	-	-	NA
22	-	/	/	-	-	0.0	34.24	-	-	-	NA
23	-	/	/	-	-	0.0	34.54	-	-	-	NA
1	-	/	/	-	-	0.0	33.31	-	-	-	NA
7	-	/	/	-	-	0.0	34.51	-	-	-	NA
8	+	-	+	+	+	15.32	-	+	+	+	PA
10	-	/	/	-	-	0.0	34.28	-	-	-	NA
15	+	-	+	+	+	15.57	-	+	+	+	PA
17	-	/	/	-	-	0.0	34.23	-	-	-	NA
19	-	/	/	-	-	0.0	34.27	-	-	-	NA
24	-	/	/	-	-	0.0	34.67	-	-	-	NA
2	+	-	+	+	+	13.88	-	+	+	+	PA
3	-	/	/	-	-	0.0	34.50	-	-	-	NA
6	+	-	+	+	+	15.84	-	+	+	+	PA
13	-	/	/	-	-	0.0	34.82	-	-	-	NA
14	-	/	/	-	-	0.0	34.18	-	-	-	NA
16	+	-	+	+	+	14.36	-	+	+	+	PA
20	-	/	/	-	-	0.0	34.28	-	-	-	NA
21	+	-	+	+	+	17.78	-	+	+	+	PA

Laboratory **K**
Lactic acid bacteria: 3.7.10⁶ CFU/g

DNA extraction:	Bax lysis
Thermocycler:	LC480 II

N°Sample	ISO 22964				foodproof® <i>Salmonella</i> plus <i>Cronobacter</i> Detection LyoKit							Agreement Ref/Alt
	CSB/CCI	Confirmation tests after subculture onto TSA		Final result	PCR test			PCR final result <i>Cronobacter</i> spp.	Confirmation final result <i>Cronobacter</i> spp. (results of the ISO 22964)	Final result		
					<i>Cronobacter</i> spp. (HEX)		Internal control (ROX)					
					Results (+/-)	Cq value	Result or Cq value					
4	-	/	/	-	-	0.0	34.97	-	-	-	NA	
5	-	/	/	-	-	0.0	34.83	-	-	-	NA	
9	-	/	/	-	-	0.0	35.05	-	-	-	NA	
11	-	/	/	-	-	0.0	33.98	-	-	-	NA	
12	-	/	/	-	-	0.0	34.13	-	-	-	NA	
18	-	/	/	-	-	0.0	34.50	-	-	-	NA	
22	-	/	/	-	-	0.0	34.49	-	-	-	NA	
23	-	/	/	-	-	0.0	34.39	-	-	-	NA	
1	-	/	/	-	-	0.0	35.59	-	-	-	NA	
7	+	-	<i>Cronobacter sakazakii</i>	+	+	12.49	0.0	+	+	+	PA	
8	-	/	/	-	-	0.0	34.94	-	-	-	NA	
10	+	-	<i>Cronobacter sakazakii</i>	+	+	12.78	0.0	+	+	+	PA	
15	+	-	<i>Cronobacter sakazakii</i>	+	+	12.75	0.0	+	+	+	PA	
17	-	/	/	-	-	0.0	35.08	-	-	-	NA	
19	-	/	/	-	-	0.0	34.34	-	-	-	NA	
24	+	-	<i>Cronobacter sakazakii</i>	+	+	12.46	0.0	+	+	+	PA	
2	+	-	<i>Cronobacter sakazakii</i>	+	+	13.02	0.0	+	+	+	PA	
3	-	/	/	-	-	0.0	33.53	-	-	-	NA	
6	-	/	/	-	-	0.0	34.78	-	-	-	NA	
13	-	/	/	-	-	0.0	34.77	-	-	-	NA	
14	+	-	<i>Cronobacter sakazakii</i>	+	+	12.86	0.0	+	+	+	PA	
16	+	-	<i>Cronobacter sakazakii</i>	+	+	12.85	0.0	+	+	+	PA	
20	+	-	<i>Cronobacter sakazakii</i>	+	+	12.78	0.0	+	+	+	PA	
21	-	/	/	-	-	0.0	34.66	-	-	-	NA	

Laboratory L
Lactic acid bacteria: 5.0.10⁶CFU/g

DNA extraction: Bax Lysis
Thermocycler: Bax Q7

N°Sample	ISO 22964			foodproof® Salmonella plus Cronobacter Detection LyoKit							Agreement Ref/Alt
				PCR test			PCR final result <i>Cronobacter</i> spp.	Confirmation final result Cronobacter spp. (results of the ISO 22964)	Final result		
	CSB/CCI	Confirmation tests after subculture onto TSA		Final result	Cronobacter spp. (HEX)					Internal control (ROX)	
		Typical colonies	Oxidase test (+/-)		RAPID ID 32E	Results (+/-)				Cq value	
4	-	/	/	-	-	0	39.0	-	-	-	NA
5	-	/	/	-	-	0	38.9	-	-	-	NA
9	-	/	/	-	-	0	39.0	-	-	-	NA
11	-	/	/	-	-	0	38.7	-	-	-	NA
12	-	/	/	-	-	0	39.1	-	-	-	NA
18	-	/	/	-	-	0	38.8	-	-	-	NA
22	-	/	/	-	-	0	38.8	-	-	-	NA
23	-	/	/	-	-	0	39.0	-	-	-	NA
1	-	/	/	-	-	0	39.3	-	-	-	NA
7	-	/	/	-	-	0	38.1	-	-	-	NA
8	+	-	<i>Cronobacter sakazakii</i>	+	+	16.9	0	+	+	+	PA
10	-	/	/	-	-	0	38.9	-	-	-	NA
15	-	/	/	-	-	0	39.2	-	-	-	NA
17	+	-	<i>Cronobacter sakazakii</i>	+	+	16.9	0	+	+	+	PA
19	-	/	/	-	-	0	38.7	-	-	-	NA
24	+	-	<i>Cronobacter sakazakii</i>	+	+	17.1	0	+	+	+	PA
2	+	-	<i>Cronobacter sakazakii</i>	+	+	16.6	0	+	+	+	PA
3	-	/	/	-	-	0	38.8	-	-	-	NA
6	+	-	<i>Cronobacter sakazakii</i>	+	+	16.4	0	+	+	+	PA
13	+	-	<i>Cronobacter sakazakii</i>	+	+	17.1	0	+	+	+	PA
14	-	/	/	-	-	0	37.5	-	-	-	NA
16	-	/	/	-	-	0	38.6	-	-	-	NA
20	-	/	/	-	-	0	38.5	-	-	-	NA
21	+	-	<i>Cronobacter sakazakii</i>	+	+	16.3	0	+	+	+	PA

Laboratory M ADRIA
Lactic acid bacteria: 7.7.10⁶ CFU/g

N° Sample	ISO 22964*				foodproof® Salmonella plus Cronobacter Detection LyoKit– Cronobacter spp																								
					DNA EXTRACTION : BAX lysis								DNA EXTRACTION : StarPrep Three kit								Confirmation final result (results of the ISO 22964)	Final result				Agreement Ref/Alt			
					Thermocycler: BAX Q7				Thermocycler: LC 480 II				Thermocycler: BAX Q7				Thermocycler: LC 480 II					DNA EXTRACTION: BAX lysis		DNA EXTRACTION: StarPrep Three kit		DNA EXTRACTION: BAX lysis		DNA EXTRACTION: StarPrep Three kit	
					PCR test		PCR final result	PCR test		PCR final result	PCR test		PCR final result	PCR test		PCR final result	PCR test		PCR final result	BAX Q7		LC 480 II	BAX Q7	LC 480 II	BAX Q7	LC 480 II	BAX Q7	LC 480 II	
					Cronobacter spp. (HEX)	I.C. (ROX))		Cronobacter spp. (HEX)	I.C. (ROX)		Cronobacter spp. (HEX)	I.C. (ROX)		Cronobacter spp. (HEX)	I.C. (ROX)														
	CSB/CCI	Confirmation tests after subculture onto TSA		Final result	Results (+/-)	Cq value	Result or Cq value	Results (+/-)	Cq value	Result or Cq value	Results (+/-)	Cq value	Result or Cq value	Results (+/-)	Cq value	Result or Cq value	Results (+/-)	Cq value	Result or Cq value	Results (+/-)	Cq value	Result or Cq value	Results (+/-)	Cq value	Result or Cq value	Results (+/-)	Cq value	Result or Cq value	
	Typical colonies	Oxidase test (+/-)	RAPID ID 32E		Results (+/-)	Cq value	Result or Cq value	Results (+/-)	Cq value	Result or Cq value	Results (+/-)	Cq value	Result or Cq value	Results (+/-)	Cq value	Result or Cq value	Results (+/-)	Cq value	Result or Cq value	Results (+/-)	Cq value	Result or Cq value	Results (+/-)	Cq value	Result or Cq value	Results (+/-)	Cq value	Result or Cq value	
4	st	/	/	-	-	/	39.0	-	-	/	35.00	-	-	/	37.1	-	Un/no amp	/	32.78	-	-	-	-	NA	NA	NA	NA		
5	st	/	/	-	-	/	38.4	-	+(no amp) -	41.88 0 0	34.75	-/-/-	-/-/-	/	31.2 36.7 36.3	-	-	/	32.55	-	-	-	-	NA	NA	NA	NA		
9	st	/	/	-	-	/	38.2	-	-	/	34.74	-	-	/	36.9	-	-	/	32.80	-	-	-	-	NA	NA	NA	NA		
11	st	/	/	-	-	/	37.6	-	-	/	34.05	-	-	/	36.6	-	+(no amp) +(no amp) -	35.94 36.76 0	32.62	-/-/-	-	-	NA	NA	NA	NA			
12	st	/	/	-	-	/	38.3	-	-	/	34.35	-	-	/	37.0	-	-	/	32.70	-	-	-	-	NA	NA	NA	NA		
18	st	/	/	-	-	/	38.6	-	-	/	35.12	-	-	/	36.4	-	-	0	32.46	-	-	-	-	NA	NA	NA	NA		
22	st	/	/	-	-	/	39.3	-	-	/	35.41	-	-	/	37.1	-	-	/	32.78	-	-	-	-	NA	NA	NA	NA		
23	st	/	/	-	-	/	38.4	-	-	/	34.29	-	-	/	37.0	-	-	/	32.10	-	-	-	-	NA	NA	NA	NA		
1	+	-	C. sakazakii	+	+	16.6	-	+	+	13.20	-	+	+	15.2	-	+	+	12.06	-	+	+	+	+	PA	PA	PA	PA		
7	+	-	C. sakazakii	+	+	17.1	-	+	+	12.90	-	+	+	15.0	-	+	+	11.96	-	+	+	+	+	PA	PA	PA	PA		
8	+	-	C. sakazakii	+	+	26.3	-	+	+	22.13	-	+	+	21.0	-	+	+	17.74	18.26	+	+	+	+	PA	PA	PA	PA		
10	st	/	/	-	-	/	39.5	-	+/-/-	41.76 0 0	35.66	+/-/-	-	/	36.7	-	-	/	32.93	-	-	-	-	NA	NA	PDFP(alt)	NA		
15	st	/	/	-	-	/	39.1	-	-	/	34.56	-	-	/	27.0	-	-	/	32.77	-	-	-	-	NA	NA	NA	NA		
17	+	-	C. sakazakii	+	+	18.2	-	+	+	14.57	-	+	+	15.6	-	+	+	12.33	-	+	+	+	+	PA	PA	PA	PA		
19	st	/	/	-	-	/	37.7	-	-	/	34.40	-	-	/	36.7	-	-	/	32.60	-	-	-	-	NA	NA	NA	NA		
24	st	/	/	-	-	/	38.8	-	-	/	34.49	-	-	/	36.8	-	-	/	32.62	-	-	-	-	NA	NA	NA	NA		
2	+	-	C. sakazakii	+	+	22.3	-	+	+	17.98	-	+	+	17.6	-	+	+	14.37	-	+	+	+	+	PA	PA	PA	PA		
3	st	/	/	-	-	/	38.5	-	+/-/-	36.02 0 0	35.0	+/-/-	-	/	36.7	-	-	/	32.65	-	-	-	-	NA	NA	PDFP(alt)	NA		
6	+	-	C. sakazakii	+	+	17.5	-	+	+	13.72	-	+	+	15.1	-	+	+	12.05	-	+	+	+	+	PA	PA	PA	PA		
13	+	-	C. sakazakii	+	+	17.2	-	+	+	13.31	-	+	+	15.0	-	+	+	11.96	-	+	+	+	+	PA	PA	PA	PA		
14	+	-	C. sakazakii	+	+	17.4	-	+	+	13.56	-	+	+	15.3	-	+	+	12.17	-	+	+	+	+	PA	PA	PA	PA		
16	st	/	/	-	-	/	40.3	-	-	/	33.5	-	-	/	37.4	-	-	/	32.81	-	-	-	-	NA	NA	NA	NA		
20	+	-	C. sakazakii	+	+	17.1	-	+	+	13.91	-	+	+	15.4	-	+	+	12.26	-	+	+	+	+	PA	PA	PA	PA		
21	st	/	/	-	-	/	39.1	-	-	/	34.3	-	-	/	36.9	-	-	/	32.30	-	-	-	-	NA	NA	NA	NA		

un: uncertain, interpretation given by the LC 480 II software. To be considered as negative result

no amp : no amplification curves. To be considered as negative results

* Analyses performed according to the COFRAC accreditation (Accreditation Testing n°1-0144, scope available on www.cofrac.fr)