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PROFICIENCY TEST « RAEMA »

réseau d'analyses et d'échanges en microbiologie des aliments

SCHEME N° 74
(7th MARCH 2022)
GENERAL REPORT

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1- GENERAL DATA

1.1. PARTICIPATING LABORATORIES

348 laboratories participated to the 74th scheme. The sending was made on Monday 7th March 2022. We received **342** answers (98.3%).

1.2. DELIVERY TIME OF THE PARCEL

Reception	J0	J0+1	J0+2	J0+3	J0+4	J0+5	J0+7	J0+8	J0+11
Nb of laboratories	7	214	70	20	14	1	9	5	1

1.3. INFORMATIONS ABOUT SAMPLE

1.3.1. NATURE

The sample included :

- one strain of *Enterococcus sp*. at a concentration level of 10^5 cfu/g in 5 units ;
- one strain of *Citrobacter sp.* at a concentration level of 10^3 cfu/g in 5 units ;
- one strain of *Serratia liquefaciens* at a concentration level of 10^3 cfu/g in 5 units ;
- one strain of *Escherichia coli* at a concentration level of 7.10^2 cfu/g in 5 units ;
- one strain of *Clostridium perfringens* at a concentration level of 10^2 cfu/g in 2 units ;
- one strain of *Staphylococcus aureus* at a concentration level of 5.10^3 cfu/g in 5 units ;
- one strain of *Salmonella* Anatum at a concentration level of 25 cfu/g in 3 units ;
- one strain of *Listeria monocytogenes* at a concentration level of 3.10^3 cfu/g in 3 units .

1.3.2. SIZE

180 kilogrammes of powder were produced and distributed after contamination in bottles containing 75 grammes at least. Bottles were covered by a label with a 6 digit identification number.

1.3.3. HOMOGENEITY AND STABILITY TEST OF THE CONTAMINATION

Homogeneity and stability of samples are checked during the statistical analysis of participants results.
A supplementary check of the contamination's homogeneity was realized on 10 samples for each unit by a double enumeration of aerobic microorganisms at 30°C.
The contamination's stability was also checked by enumeration / detection of all flora on 14, 21 and 28 March 2022. These checks were realized by a subcontractor accreditated by Cofrac.
Homogeneity and stability of samples have been validated.

1.3.4. FLORA FOR ENUMERATION / DETECTION

Enumeration of the following flora was proposed : microorganisms at 30°C, Enterobacteriaceae, total and thermotolerant coliforms, beta-glucuronidase positive *Escherichia coli*, anaerobic sulfite-reducing bacteria, *Clostridium perfringens*, coagulase positive staphylococci, *Listeria monocytogenes*, as well as detection of *Salmonella* and *Listeria monocytogenes*.

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1.4. EXECUTION OF ANALYZES

1.4.1. DELIVERY TIME OF SAMPLES / BEGINNING OF ANALYZES

342 laboratories (100%) specified it.

Analysis time	J0+1	J0+2	J0+3	J0+4	J0+5	J0+7	J0+8	J0+9	J0+10	J0+11	J0+14	J0+15	J0+18
Nb of laboratories	34	57	37	5	1	128	52	8	4	2	9	4	1

1.4.2. PRESERVATION TEMPERATURE OF SAMPLES BEFORE ANALYSIS

339 laboratories (99.1%) specified it. The average temperature is **3.9°C** with a standard deviation of 0.9°C. The given data 17, 20, 20.1, 22, 22.5, 24.1 and 25°C given by 11 laboratories were not taken into account for this calculation.

2. EXPLOITATION OF ANALYSIS REPORT

2.1. PREPARATION OF THE INITIAL SUSPENSION

For **342** answers (100%) :
217 laboratories (63.4%) prepare the initial suspension with adding diluent to powder.
124 laboratories (36.3%) prepare the initial suspension with adding powder to diluent.
1 laboratory (0.3%) used another technique to prepare the initial suspension.

2.2. DILUENT USED FOR THE INITIAL SUSPENSION

For **338** answers (98.8%) :
301 laboratories (88.0%) use Buffered Peptone Water (or equivalent) for the initial suspension.
35 laboratories (10.2%) use Peptone salt for the initial suspension.
2 laboratories (0.6%) used another diluent for the initial suspension.

2.3. HOMOGENEIZATION TECHNIQUE

For **339** answers (99.1%) :
314 laboratories (91.8%) homogeneize their sampling with a StomacherND.
18 laboratories (5.2%) used a manual homogenization.
3 laboratories (0.9%) used a Vortex mixer.
4 laboratories (1.2%) used another technique.

2.4. RESUSCITATION'S CONDITIONS

2.4.1. DURATION

330 laboratories (96.5%) specified it.
The average duration is **26.9 min** with a standard deviation of 15.2 min. The data 90, 120 and 1440 min given by 4 laboratories were not taken into account for this calculation.

2.4.2. TEMPERATURE

330 laboratories (96.5%) specified it.
The average temperature is **21.4°C** with a standard deviation of 3.4°C.

2.5. MICROORGANISM AT 30°C

328 laboratories performed the enumeration.

Parameters	Mode	Nb laboratories
Method	ISO/NF EN ISO 4833-1	211
	AFNOR 3M-01/1-09/89	52
	NM ISO 4833-1	22
	ISO/NF EN ISO 4833-2	11
	AFNOR BIO-12/35-05/13	9
	XP V08-034	8
	Internal method	6
	Other	8
	+ V08-100 (spiral)	19
Culture medium	Plate Count Agar	243
	Petrifilms	52
	Plate Count Agar + Milk	23
	Tempo AC	9
	Other	1
Preparation	Home made	112
	Ready to use not pre-poured	143
	Ready to use, plate, film, card	71
Plating method	Surface	67
	Pour	246
	Culture medium for card	10
1st dilution retained	- 1	11
	- 2	16
	- 3	175
	- 4	113
	- 5	3
	1/400	5
	1/4000	1
Incubation temperature	30°C	324
	33-35°C	2
	37°C	1
	25°C	1
Incubation duration	68-74 h	269
	40-48 h	56
	24-26 h	3

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2.6. ENTEROBACTERIACEA

288 laboratories performed the enumeration.

Parameters	Mode	Nb laboratories
Method	NF V08-054	109
	→ *NM 08.0.109*(1)	18
	ISO/NF EN ISO 21528-2	77
	AFNOR 3M-01/6-09/97	46
	AFNOR BIO-12/21-12/06	10
	AFNOR AES-10/07-01/08	9
	NM ISO 21528-2	8
	AFNOR BRD-07/24-11/13	7
	Internal method	2
	Other	2
Culture medium	VRBG	211
	Petrifilms	48
	Tempo EB	10
	Rebecca	10
	Rapid'Enterobacteriaceae	8
	Other	1
Preparation	Home made	93
	Ready to use not pre-poured	137
	Ready to use, plate, film, card	57
1st dilution retained	- 1	182
	- 2	95
	- 3	1
	1/40	2
	1/400	5
Incubation temperature	37±1°C	183
	30°C	95
	35°C	10
Incubation duration	20-26 h	281
	48 h	5
	30 h	1
	37 h	1
Confirmatory test	Yes	69
	No	215

(1) *Similar method to NF V08-054 according to ONSSA (Office National de Sécurité Sanitaire des produits Alimentaires).*

2.7. TOTAL COLIFORMS

236 laboratories performed the enumeration.

Parameters	Mode	Nb laboratories
Method	NF V08-050	119
	→ *NM 08.0.142*(2)	8
	ISO/NF ISO 4832	59
	NM ISO 4832	22
	AFNOR 3M	18
	AFNOR BRD-07/08-12/04	4
	AFNOR BIO-12/17-12/05	2
	Internal method	2
	Other	2
Culture medium	VRBL	207
	Petrifilms	19
	Rapid Ecoli	5
	Tempo TC	2
	Other	2
Preparation	Home made	90
	Ready to use not pre-poured	124
	Ready to use, plate, film, card	21
1st dilution retained	-1	179
	-2	54
	-3	1
	1/400	1
Incubation temperature	30°C	220
	37°C	16
Incubation duration	20-26 h	232
	48 h	3
	30 h	1

AFNOR 3M method including :
3 laboratories specified utilization of AFNOR 3M-01/02-09/89 A method.

(2) *Similar method to NF V 08-050 according to ONSSA.*

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2.8. THERMOTOLERANT COLIFORMS

217 laboratories performed the enumeration.

Parameters	Mode	Nb laboratories
Method	NF V08-060	145
	→ *NM 08.0.124*(3)	27
	AFNOR 3M	23
	ISO/NF ISO 4832	18
	Internal method	2
	Other	2
Culture medium	VRBL	190
	Petrifilms	24
	Other	3
Preparation	Home made	87
	Ready to use not pre-poured	106
	Ready to use, plate, film, card	23
1st dilution retained	-1	173
	-2	43
Incubation temperature	42-45°C	214
	37°C	2
	30°C	1
Incubation duration	21-25 h	214
	48 h	3

AFNOR 3M method including :

4 laboratories specified utilization of AFNOR 3M-01/02-09/89 C method.
1 laboratory specified utilization of AFNOR 3M-01/05-03/97 B method.
1 laboratory specified utilization of AFNOR 3M Petrifilm high sensitivity method.
1 laboratory specified utilization of AFNOR 3M Petrifilm EC method.

(3) *Similar method to NF V08-060 according to ONSSA.*

2.9. ESCHERICHIA COLI

308 laboratories performed the enumeration.

Parameters	Mode	Nb laboratories
Method	ISO/NF ISO 16649-2	182
	AFNOR 3M	47
	NM ISO 16649-2	18
	AFNOR BRD-07/01-07/93	17
	AFNOR BIO-12/13-02/05	10
	AFNOR AES-10/06-01/08	9
	NM 08.0.108	8
	AFNOR BIO-12/05-01/99	4
	ISO/NF EN ISO 16649-3	3
	Internal method	3
	Other	7
Culture medium	TBX	210
	Petrifilms	48
	Rapid E. coli	21
	Rebecca	11
	Tempo EC	10
	Coli ID	6
	Other	2
Preparation	Home made	89
	Ready to use not pre-poured	163
	Ready to use, plate, film, card	56
Plating method	Surface	46
	Pour	246
	Culture medium for card	12
1st dilution retained	-1	262
	-2	36
	1/40	2
	1/400	6
Incubation temperature	41-45°C	276
	37±1°C	31
	30°C	1
Incubation duration	18-25 h	304
	48 h	4

AFNOR 3M method including :

16 laboratories specified utilization of AFNOR 3M-01/08-06/01 (*SELECT'E. COLI*) method.

1 laboratory specified utilization of AFNOR 3M-01/06-09/97 method.

2 laboratories specified utilization of AFNOR 3M Petrifilm EC method.

2.10. ANAEROBIC SULFITE-REDUCING BACTERIA

244 laboratories performed the enumeration.

Parameters	Mode	Nb laboratories
Method	NF V08-061	155
	→ *NM 08.0.125*[4]	15
	ISO/NF ISO 15213	47
	NM ISO 15213	14
	Internal method	7
	Other	5
Culture medium	TSC	230
	TSN	6
	Iron Sulfite agar	6
	Other	2
Preparation	Home made	90
	Ready to use not pre-poured	125
	Ready to use, plate, film, card	29
Seeding way	Plates	164
	Tubes	80
1st dilution retained	-1	220
	-2	21
Incubation temperature	44-48°C	174
	37°C	70
Incubation duration	18-24 h	198
	44-48 h	40
	72 h	6

[4] *Similar method to NF V08-061 according to ONSSA.*

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2.11. CLOSTRIDIUM PERFRINGENS

188 laboratories performed the enumeration.

Parameters	Mode	Nb laboratories
Method	ISO/NF EN ISO 7937	149
	NM ISO 7937	29
	NM 08.0.111	2
	Internal method	1
	Other	6
Culture medium	TSC	187
	Other	1
Preparation	Home made	64
	Ready to use not pre-poured	119
	Ready to use, plate, film, card	5
1st dilution retained	-1	180
	-2	6
Incubation temperature	37°C	180
	44-46°C	7
	34°C	1
Incubation duration	18-24 h	181
	48 h	7
Confirmation test	None	29
	Lactose-sulfite	137
	Strip	9
	MALDI-TOF mass spectrometry	3
	Other	1

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2.12. COAGULASE POSITIVE STAPHYLOCOCCI

303 laboratories performed the enumeration.

Parameters	Mode	Nb laboratories
Method	ISO/NF EN ISO 6888-2	135
	ISO/NF EN ISO 6888-1	74
	AFNOR BKR-23/10-12/15	23
	NM ISO 6888-1	22
	AFNOR 3M-01/9-04/03	15
	AFNOR BIO-12/28-04/10	9
	NM ISO 6888-2	5
	Internal method	4
	NM 08.0.112	3
	NordVal No :049	3
	ISO/NF EN ISO 6888-3	3
	Other	5
Culture medium	RPF	131
	BP+egg yolk tellurite	94
	Easy Staph	28
	Petrifilm	16
	BP+egg yolk tellurite+ sulfamethazine	16
	Tempo STA	9
	Rapid Staph	5
	Other	2
Preparation	Home made	69
	Ready to use not pre-poured	129
	Ready to use, plate, film, cards	103
Plating method	Surface	155
	Pour	135
	Culture medium for card	10
1st dilution retained	-1	105
	-2	182
	-3	6
	1/40	4
	1/400	2
Incubation temperature	37±1°C	298
	27-30°C	4
	44°C	1
Incubation duration	40-49 h	208
	18-25 h	95
Confirmation test	None	185
	Staphylo-coagulase	81
	Clumping factor	16
	DNase	10
	MALDI-TOF mass spectrometry	3
	Other	2

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2.13. LISTERIA MONOCYTOGENES – ENUMERATION

249 laboratories performed the enumeration.

RESUSCITATION

92 laboratories announce the realization of a resuscitation step.

Details concerning temperature and average duration of this step are not required anymore in the input form.

Parameters	Mode	Nb laboratories
Method	ISO/NF EN ISO 11290-2	74
	AFNOR AES-10/05-09/06	59
	AFNOR BKR-23/05-12/07	53
	NM ISO 11290-2	26
	AFNOR BRD-07/05-09/01	25
	AFNOR BRD-07/17-01/09	10
	Other	2
Resuscitation medium	Buffered Peptone Water or equivalent	216
	Half-fraser	26
	Fraser base	3
	Other	6
Enumeration medium	ALOA Count	116
	Compass Listeria	84
	Rapid Lmono	26
	AL Agar	18
	OCLA	3
	Palcam	1
	Other	1
Preparation	Home made	38
	Ready to use not pre-poured	51
	Ready to use, plate, film, card	159
Plating method	Surface	207
	Pour	39

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Parameters	Mode	Nb laboratories
1st dilution retained	-1	158
	-2	88
Incubation temperature	37°C	243
	30°C	5
	20°C	1
Incubation duration	44-49 h	207
	24-27 h	41
	15 h	1
Confirmation test	None	47
	Biochemical	148
	Biochemical + CAMP	37
	MALDI-TOF mass spectrometry	3
	Other	10
Nb of colonies tested per plate	1	63
	2-4	14
	5	111
	6	2

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2.14. SALMONELLA – DETECTION

312 laboratories performed the detection.
Methods used by laboratories are clarified in the following table :

Parameter	Mode	Nb laboratories
Method	ISO/NF EN ISO 6579-1	81
	AFNOR BKR 23/07-10/11 (IRIS Salmonella)	77
	NM ISO 6579-1	36
	AFNOR BRD 07/11-12/05 (Rapid Salmonella)	31
	AFNOR BIO 12/32-10/11 (VIDAS SPT)	24
	AFNOR BIO 12/41-03/17 (SALMA One day)	24
	AFNOR BIO 12/16-09/05 (VIDAS Easy Salmonella)	16
	AFNOR BIO 12/01-04/94 (VIDAS SLM)	7
	AFNOR UNI 03/07-11/13 (PCR)	3
	AFNOR BRD 07/06-07/04 (PCR)	2
	AFNOR BIO 12/38-06/16 (GENE UP Salmonella)	2
	AFNOR TRA 02/08-03/01 (TRANSIA PLATE Salmonella GOLD)	2
	Internal method	1
	Other	6

No detail of methodology was asked to laboratories using other methods than ISO/NF EN ISO 6579-1 and NM ISO 6579-1 methods and proposed in the questionnaire.

You will find below a short description of these methods :

Method	Pre-enrichment	Enrichment	Isolation
AFNOR BKR 23/07-10/11 **IRIS Salmonella**		IRIS Salmonella Enrichment / 41,5°C - 18±2h	IRIS / 37°C - 24±3h
AFNOR BRD 07/11-12/05 **Rapid Salmonella**		BPW + Salmonella capsule / 41,5°C - 18±2h	Rapid Salmonella / 37°C - 24±2h
AFNOR BIO 12/32-10/11 **VIDAS SPT**		BPW + Salmonella supplement / 41,5°C - 18/24h	Chrom ID / 37°C - 24h
AFNOR BIO 12/41-03/17 **SALMA One day**		BPW + Salmonella supplement / 41.5°C – 16/24h	SALMA / 37°C - 24±3h
AFNOR BIO 12/16-09/05 **VIDAS Easy Salmonella**	BPW / 37°C - 16/20h	SX2 / 41,5°C - 22/26h	Chrom ID / 37°C - 24h
AFNOR BIO 12/01-04/94 **VIDAS SLM**	BPW / 35°C – 24±2h	Tetrathionate (42°C - 6/8h) – Selenite cystine (35-37°C – 6/8h) + M-Broth (42°C – 18h)	Vidas Heat & Go
AFNOR UNI 03/07-11/13 **PCR**		BPW + supplement / 34-38°C – 20/24h	Lysis + PCR
AFNOR BRD 07/06-07/04 **PCR**		BPW / 37°C – 18/21h	Lysis + PCR
AFNOR BIO 12/38-06/16 **GENE UP Salmonella**		BPW / 42°C – 18/24h	Lysis + PCR
AFNOR TRA 02/08-03/01 **TRANSIA PLATE Salmonella GOLD**	BPW / 37°C – 16/20h	RVS / 41.5°C – 18/24h	ELISA test

The detail of the methodology followed by 117 laboratories using ISO/NF EN ISO 6579-1 and NM ISO 6579-1 method, and the 7 laboratories using an internal or another method, is clarified in the following table :

Parameter	Mode	Nb laboratories
Method	ISO/NF EN ISO 6579-1	81
	NM ISO 6579-1	36
	Internal method	1
	Other	6
Pre-enrichment medium	None pre-enrichment	2
	Buffered Peptone Water	118
	Other	4
Pre-enrichment temperature	37±1°C	115
	42-42.5°C	4
	22°C	2
	30°C	1
Pre-enrichment duration	16-20 h	90
	22-24 h	32
Enrichment medium	None enrichment	5
	RVS	113
	MKTTn	108
	Selenite-cystine broth	26
	Other	3
Isolation medium	XLD	107
	Hektoen	30
	Bismuth Sulfate	25
	IRIS Salmonella agar	15
	ASAP	14
	GVB	12
	SS	10
	Brilliance Salmonella	7
	Rapid Salmonella	5
	Compass Salmonella	5
	Rambach	2
	Other	12
Confirmation test	Biochemical	46
	Biochemical + serological agglutination	67
	MALDI-TOF mass spectrometry	5
	Other	4

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2.15. LISTERIA MONOCYTOGENES – DETECTION

283 laboratories performed the detection.

Parameter	Mode	Nb laboratories
Method	AFNOR AES 10/03-09/00 (ALOA one day)	57
	ISO/NF EN ISO 11290-1	57
	AFNOR BKR 23/02-11/02 (Compass L. mono)	57
	NM ISO 11290-1	31
	AFNOR BRD 07/04-09/98 (Rapid' L. mono)	23
	AFNOR BRD 07/16-01/09 (Agar Listeria)	11
	AFNOR BIO 12/11-03/04 (VIDAS LMO2-37°C)	9
	AFNOR BIO 12/27-02/10 (VIDAS LMX)	8
	AFNOR BIO 12/02-06/94 (VIDAS Listeria)	7
	AFNOR BIO 12/18-03/06 (VIDAS LDUO)	4
	AFNOR BRD 07/10-04/05 (IQ Check Listeria)	4
	AFNOR UNI 03/08-11/13 (PCR)	3
	AFNOR BIO 12/40-11/16 (GENE UP LMO)	3
	AFNOR UNI 03/04-04/05 (Listeria Precis)	3
	Internal method	2
	Other	4

No detail of methodology was asked to laboratories using other method than ISO/NF EN ISO 11290-1 and NM ISO 11290-1 methods and proposed in the questionnaire.

You will find below a short description of these methods :

Méthod	Primary enrichment		Secondary enrichment		Isolation
	Medium	Incubation	Medium	Incubation	
AFNOR AES 10/03-09/00 **ALOA one day**	Half-Fraser	30°C - 24±2h			ALOA One Day 37°C – 24/48h
AFNOR BKR 23/02-11/02 **Compass L. mono**	Half-Fraser	30°C - 24±2h			Compass Listeria Agar 37°C – 24h
AFNOR BRD 07/04-09/98 **Rapid' L. mono**	Half-Fraser	30°C - 24±2h			Rapid L'mono 37°C – 24h
AFNOR BRD 07/16-01/09 **Agar Listeria**	Half-Fraser	30°C - 24±2h			Agar Listeria 37°C – 24h
AFNOR BIO 12/11-03/04 **VIDAS LMO2 (37°C)**	Half-Fraser	30°C - 24/26h	Fraser	37°C - 24/26h	Chromogenic medium / Palcam / Oxford
AFNOR BIO 12/27-02/10 **VIDAS LMX**	LMX	37°C - 26/30h			ChromID 37°C – 24h
AFNOR BIO 12/02-06/94 **VIDAS Listeria**	Half-Fraser	37°C - 26/30h	Fraser	30°C - 24/26h	Palcam et Oxford 37°C – 24h
AFNOR BIO 12/18-03/06 **VIDAS LDUO**	LX	30°C - 24±2h	LX	30°C - 24/26h	Chromogenic medium / Palcam / Oxford
AFNOR BRD 07/10-04/05 **IQ Check Listeria**	Half-Fraser / LSB	30°C – 23/25h			Lysis + PCR
AFNOR UNI 03/08-11/13 **PCR**	LEB	37°C - 24/28h			Lysis + PCR
AFNOR BIO 12/40-11/16 **GENE UP LMO**	LPT	35-37°C - 24±2h			ALOA 35-37°C – 24/48h
AFNOR UNI 03/04-04/05 **Listeria Precis**	One Broth Listeria	30°C - 24±2h			Brilliance Listeria 37°C – 24h

The detail of the methodology followed by 88 laboratories using NF ISO/EN ISO 11290-1 and NM ISO 11290-1 methods, and the 6 laboratories using an internal or another method, is clarified in the following table :

Parameter	Mode	Nb laboratories
Method	ISO/NF EN ISO 11290-1	57
	NM ISO 11290-1	31
	Internal method	2
	Other	4
Primary enrichment medium	None primary enrichment	0
	Half-Fraser	86
	One broth Listeria	1
	Other	7
Primary enrichment temperature	30°C	86
	37°C	6
	22°C	1
Primary enrichment duration	21-27 h	91
	28 h	1
	48 h	1
Secondary enrichment medium	None secondary enrichment	7
	Fraser	86
	Other	1
Secondary enrichment temperature	37°C	83
	30°C	2
	22°C	1
Secondary enrichment duration	23-27 h	72
	48 h	14
Isolation medium	Palcam	67
	Ottaviani et Agosti	52
	Compass Listeria	37
	Oxford	10
	Rapid L'mono	7
	Brilliance Listeria	1
	Other	0
Isolation temperature	37°C	90
	30°C	2
Isolation duration	44-48 h	54
	22-24 h	38
Confirmation test	None	3
	Biochemical	62
	Biochemical + CAMP	22
	MALDI-TOF mass spectrometry	3
	Other	3
Nb of colonies per plate	1	36
	2-3	7
	5	41

3. ASSESSMENT OF PERFORMANCE (INDIVIDUEL REPORTS)

3.1. PERFORMANCES IN ENUMERATION

Performance is assessed on two criteria : **precision and trueness**.
The assigned value of the contamination used to assess the trueness and the reference standard deviation for the assessment of the precision are consensual values obtained with the results of all the participants. These values are obtained by robust estimation methods in order to eliminate influence of aberrant results. However some results are excluded of the statistical analysis. That is the case when laboratories do not give results for all contaminated units, when results are “less than CFU/g“, when samples are analyzed after the deadline (time of receipt > 4 days after sending or time of analysis >15 days after sending) or when this information is not specified.

A statistical analysis has also be done to highlight potential relations between techniques used (delay of analysis, preservation temperature, preparation of the initial suspension, homogenization technique, resuscitation conditions, method used, media used, manufacturers of media, preparation mode, plating method, incubation conditions) and results obtained. We need to clarify that this statistical link is not involved in a cause - effect relationship. Indeed, this link may be due to a not documented factor.

When a significant statistical link is identified between use of a technique and the obtained results, the assessment of performance is done considering the influence of one or several factors involved if their effect translates into a contamination’s difference higher than 0.15 log CFU/g for non-selective media or higher than 0.30 log CFU/g for selective media (these limits match with productivity limits of culture media usually recommended in the standard NF EN ISO 11133).

PRECISION
The precision reflects the repeatability (or reproducibility intra-laboratory) of your work.

The standard deviation of your results, *s*, is compared to the robust estimation of the standard deviation (reference standard deviation of precision), *s**, obtained with algorithm S from the standard ISO 13528 applied to all standard deviations obtained by laboratories included in the statistical analysis.

An index score is then calculated using the following formula : $i = (k-1) \cdot \frac{s^2}{s^{*2}}$ (with k, number of contaminated units and retained in the statistical analysis, usually 5).

The standard ISO 13528 do not provide warning and action limits for this score, so its interpretation is left to your discretion.

As an indicator, we suggest following values by analogy with those indicated for the evaluation of trueness.
For k=5, a score lower than 0.1 or higher than 18 may be considered as an action signal and a score lower than 0.45 or higher than 11.5 may be considered as a warning signal.
For k=4, a score lower than 0.03 or higher than 15.5 may be considered as an action signal and a score lower than 0.2 or higher than 9.5 may be considered as a warning signal.
For k=3, a score lower than 0.003 or higher than 13.2 may be considered as an action signal and a score lower than 0.05 or higher than 7.5 may be considered as a warning signal.
For k=2, a score lower than 0.000002 or higher than 10.3 may be considered as an action signal and a score lower than 0.0008 or higher than 5.2 may be considered as a warning signal.

TRUENESS

The trueness reflects the closeness of the mean of your results to the contamination's assigned value of samples. It has been evaluated for all enumerated flora.

The mean of your results in log CFU/g, *m* (on contaminated units and included in the statistical analysis), is compared to the contamination's assigned value, m_{pt}, obtained with algorithm A from the standard ISO 13528 applied to all laboratories mean included in the statistical analysis. When groups are formed, each one is characterize by its own assigned value.

A z score is then calculated with the following formula : $z = \frac{m - m_{pt}}{\sigma_{pt}}$, where σ_{pt} is the standard deviation for proficiency assessment (robust estimation of the standard deviation obtained by participants).

The standard ISO 13528 specifies that *z* score lower than -3 or higher than +3 must be considered as an action signal and that a z score lower than -2 or higher than +2 must be considered as a warning signal.

In this report, we specify, estimations of interlaboratories standard deviation for enumerations proposed as well as reproducibility standard deviation or global standard deviation for the test (parameters including interlaboratories variability and the variability of the precision).

INDIVIDUAL REPORTS – FOR EACH CRITERIA YOU FIND THE FOLLOWING INFORMATIONS

- your results in logarithm base 10 (-1 when the answer is < limit and NaN when there is no answer). Comment : the presentation order of your results does not necessarily correspond to the order you sent them, this order is the same for all the flora.
- histogram for the studied parameter (laboratories standard deviations for the precision and laboratorie's means for the trueness) with an asterisk indicating the location of your result,
- standard deviation (precision) or mean (trueness) of your results (on contaminated units and retained in the statistical analysis),
- the method declared in your results input,
- when necessary, your group in relation to the technique used,
- precision score or z score,
- number of laboratories which made analysis (and belonging to your group),
- number of laboratories included in the statistical analysis,
- reference standard deviation for the precision or assigned value of the contamination and standard deviation aptitude assessment (trueness),
- number of laboratories with a satisfactory signal,
- number of laboratories with a warning signal,
- number of laboratories with an action signal.

3.1.1. MICROORGANISMS AT 30°C

None significant effect of the analysis technique has been highlighted.

Microorganisms at 30°C	
Assigned value of the contamination (log cfu/g)	5.469
Assigned value uncertainty (log cfu/g)	0.0060
Standard deviation for proficiency assessment (log cfu/g)	0.0843
Standard deviation for precision (log cfu/g)	0.0530
Interlaboratory's standard deviation (log cfu/g)	0.0809
Reproducibility standard deviation (log cfu/g)	0.0967

3.1.2. ENTEROBACTERIACEAE

A significant "effect" of the culture medium, manufacturer and the retained dilution has been highlighted. This effect results in a contamination's difference higher than 0.3 log cfu/g, then results have been gathered in three groups :

Enterobacteriaceae	Group 1	Group 2	Group 3
Assigned value of the contamination (log cfu/g)	3.003	3.241	3.429
Assigned value uncertainty (log cfu/g)	0.0186	0.0364	0.0229
Standard deviation for proficiency assessment (log cfu/g)	0.1948	0.2039	0.1307
Standard deviation for precision (log cfu/g)		0.0856	
Interlaboratory's standard deviation (log cfu/g)	0.1910	0.2002	0.1249
Reproducibility standard deviation (log cfu/g)	0.2093	0.2178	0.1514

3.1.3. TOTAL COLIFORMS

A significant "effect" of the culture medium, manufacturer, and the retained dilution has been highlighted. This effect results in a contamination's difference higher than 0.3 log cfu/g, then results have been gathered in three groups :

Total coliforms	Group 1	Group 2	Group 3
Assigned value of the contamination (log cfu/g)	2.915	3.078	3.366
Assigned value uncertainty (log cfu/g)	0.0211	0.0334	0.0410
Standard deviation for proficiency assessment (log cfu/g)	0.1999	0.1907	0.1672
Standard deviation for precision (log cfu/g)		0.0775	
Interlaboratory's standard deviation (log cfu/g)	0.1969	0.1875	0.1636
Reproducibility standard deviation (log cfu/g)	0.2147	0.2061	0.1846

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3.1.4. THERMOTOLERANT COLIFORMS

A significant “effect” of the retained dilution has been highlighted. This effect results in a contamination’s difference higher than 0.3 log cfu/g, then results have been gathered in two groups :

Thermotolerant coliforms	Group 1	Group 2
Assigned value of the contamination (log cfu/g)	2.922	3.288
Assigned value uncertainty (log cfu/g)	0.0177	0.0369
Standard deviation for proficiency assessment (log cfu/g)	0.1782	0.1890
Standard deviation for precision (log cfu/g)	0.0884	
Interlaboratory’s standard deviation (log cfu/g)	0.1737	0.1848
Reproducibility standard deviation (log cfu/g)	0.1937	0.2037

3.1.5. *ESCHERICHIA COLI*

A significant “effect” of the preparation mode of the culture medium and the retained dilution has been highlighted. This effect results in a contamination’s difference lower than 0.3 log cfu/g, then results have been gathered in one group :

Escherichia coli	
Assigned value of the contamination (log cfu/g)	2.899
Assigned value uncertainty (log cfu/g)	0.0150
Standard deviation for proficiency assessment (log cfu/g)	0.2046
Standard deviation for precision (log cfu/g)	0.0877
Interlaboratory’s standard deviation (log cfu/g)	0.2008
Reproducibility standard deviation (log cfu/g)	0.2192

3.1.6. ANAEROBIC SULFITE-REDUCING BACTERIA

Only units n°3 and 4 were artificially contaminated.

None significant effect of the analysis technique has been highlighted.

Anaerobic sulfite-reducing bacteria	
Assigned value of the contamination (log cfu/g)	2.112
Assigned value uncertainty (log cfu/g)	0.0171
Standard deviation for proficiency assessment (log cfu/g)	0.2022
Standard deviation for precision (log cfu/g)	0.1078
Interlaboratory’s standard deviation (log cfu/g)	0.1873
Reproducibility standard deviation (log cfu/g)	0.2161

Comment :
- 5 laboratories detected ASR in unit n°1 non-artificially contaminated by *C. perfringens* with a contamination level from 10 to 3800 cfu/g.
- 8 laboratories detected ASR in unit n°2 non-artificially contaminated by *C. perfringens* with a contamination level from 1 to 4300 cfu/g.
- 13 laboratories detected ASR in unit n°5 non-artificially contaminated by *C. perfringens* with a contamination level from 10 to 5200 cfu/g.

3.1.7. *CLOSTRIDIUM PERFRINGENS*

Only units n°3 and 4 were artificially contaminated.

None significant effect of the analysis technique has been highlighted.

Clostridium perfringens	
Assigned value of the contamination (log cfu/g)	2.105
Assigned value uncertainty (log cfu/g)	0.0158
Standard deviation for proficiency assessment (log cfu/g)	0.1660
Standard deviation for precision (log cfu/g)	0.0995
Interlaboratory's standard deviation (log cfu/g)	0.1504
Reproducibility standard deviation (log cfu/g)	0.1803

Comment :
- 3 laboratories detected *C. perfringens* in unit n°1 non-artificially contaminated by *C. perfringens* with a contamination level from 10 to 2400 cfu/g.
- 5 laboratories detected *C. perfringens* in unit n°2 non-artificially contaminated by *C. perfringens* with a contamination level from 10 to 4400 cfu/g.
- 5 laboratories detected *C. perfringens* in unit n°5 non-artificially contaminated by *C. perfringens* with a contamination level from 10 to 4000 cfu/g.

3.1.8. COAGULASE POSITIVE STAPHYLOCOCCI

A significant "effect" of the resuscitation's duration has been highlighted. This effect results in a contamination's difference lower than 0.3 log cfu/g, then results have been gathered in one group :

Coagulase positive Staphylococci	
Assigned value of the contamination (log cfu/g)	3.836
Assigned value uncertainty (log cfu/g)	0.0124
Standard deviation for proficiency assessment (log cfu/g)	0.1669
Standard deviation for precision (log cfu/g)	0.0657
Interlaboratory's standard deviation (log cfu/g)	0.1643
Reproducibility standard deviation (log cfu/g)	0.1769

3.1.9. *LISTERIA MONOCYTOGENES*

Only units n°3, 4 and 5 were artificially contaminated.

A significant "effect" of the culture medium has been highlighted. This effect results in a contamination's difference lower than 0.3 log cfu/g, then results have been gathered in one group :

Listeria monocytogenes	
Assigned value of the contamination (log cfu/g)	3.514
Assigned value uncertainty (log cfu/g)	0.0083
Standard deviation for proficiency assessment (log cfu/g)	0.1019
Standard deviation for precision (log cfu/g)	0.0724
Interlaboratory's standard deviation (log cfu/g)	0.0929
Reproducibility standard deviation (log cfu/g)	0.1178

3.2. PERFORMANCES IN DETECTION

The performance is assessed by the capacity to detect only samples contaminated by *Salmonella* and *Listeria monocytogenes* (no false positive or false negative results).

3.2.1. DETECTION – *SALMONELLA*

Only units n°3, 4 and 5 were artificially contaminated.
297 laboratories obtained correct results.
8 laboratories obtained false positive results (respectively 5 and 5 false-positive for units n° 1 and 2).
13 laboratories obtained false negative results (respectively 4, 3 and 8 false-negative for units n° 3, 4 and 5).

3.2.2. DETECTION – *LISTERIA MONOCYTOGENES*

Only units n°3, 4 and 5 were artificially contaminated.
274 laboratories obtained correct results.
6 laboratories obtained false positive results (respectively 2 and 4 false-positive for units n°1 and 2).
4 laboratories obtained false negative results (respectively 1, 0 and 3 false-negative for units n°3, 4 and 5).

3.3. EVOLUTION OF PERFORMANCE

You will find, on each page of your performance's assessment, a graph representing evolution of it on different tests since the 54th scheme.

In order to interpret your control card with z scores, you can refer to the standard ISO 13528 §10.8.2.2, explaining the 3 « out of control » situations:

- Just one overtaking of the action limit (z<-3 or z>3),
- 2 consecutives z scores out of 3 overtaking of the warning limit (2<z<3 or -3<z<-2),
- 6 consecutives z scores regularly increasing or decreasing.