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PROFICIENCY TEST « RAEMA »

SCHEME N° 80 A
(16th JUNE 2025)
GENERAL REPORT

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1. GENERAL DATA

1.1. PARTICIPATING LABORATORIES

157 laboratories participated to the 80Ath Gel scheme on 16th June 2025 (J0).
We received **157** answers (100%).

1.2. DELIVERY TIME OF THE PARCEL

Delivery time	J0	J0+1	J0+2	J0+3	J0+4	J0+7	J0+8	J0+10	J0+15
Nb of laboratories	4	116	20	7	5	2	1	1	1

1.3. INFORMATIONS ABOUT SAMPLE

1.3.1. NATURE

- one sample included a strain of *Lactobacillus plantarum* at a concentration level of 1,5.10^5 cfu/g ;
- one sample included a strain of *Pseudomonas sp.* at a concentration level of 8.10^3 cfu/g ;
- one sample included a strain of *Bacillus cereus* at a concentration level of 7,5.10^2 cfu/g ;
- one sample included a strain of *Penicillium* at a concentration level of 5,5.10^3 cfu/g and a strain of *Rhodotorula rubra* at a concentration level of 7.10^3 cfu/g ;

1.3.2. SIZE

Samples were composed of a gel and distributed in bottles containing 50 grammes.

1.3.3. HOMOGENEITY AND STABILITY TEST OF THE CONTAMINATION

A check of the contamination's homogeneity was realized on 10 samples per numeration in duplicate for all flora.
The contamination's stability was checked by enumeration of all flora on 19 June (J0+3), 23 June (J0+7) and 30 June 2025 (J0+14).
These checks were realized by an external provider accredited by Cofrac for *Bacillus cereus,* lactic bacteria and Yeast/Mould. The check of *Pseudomonas* was realized by the same provider but not covered by Cofrac accreditation.
Homogeneity of samples has been validated except for Moulds. For this parameter, intra-sample standard deviation has been included in the calculation of the standard deviation for proficiency assessment (NF ISO 13528 § B.2.5.a).
Stability of samples has been validated.

1.3.4 FLORA FOR ENUMERATION

Enumeration of the following flora was proposed:
- lactic acid bacteria
- *Pseudomonas*
- *Bacillus cereus*
- Yeast - Moulds analyzed together
- Yeast
- Moulds

1.4. EXECUTION OF ANALYZES

1.4.1 PRESERVATION TEMPERATURE OF SAMPLES BEFORE ANALYSIS

157 laboratories (100%) specified it.
The average temperature is **4.5°C** with a standard deviation of 2.5°C. The minimum temperature indicated is 2.0°C and the maximum one is 23.0°C.

Remark: Please note that samples must be conserved at 5±3°C on receipt, before analysis. They should not be frozen.

2. EXPLOITATION OF ANALYSIS REPORT

2.1. SIZE OF TEST SAMPLE

157 laboratories (100%) specified it.
The average size is **14.3 g** with a standard deviation of 6.4 g. The data 1.09 g given by one laboratory was not taken into account for this calculation. The minimum size indicated is 10.0 g and the maximum one is 26.7 g.

2.2. PREPARATION OF THE INITIAL SUSPENSION

156 laboratories (99.4%) specified it.
154 laboratories (98.1%) prepare the initial suspension with adding diluent to gel.
2 laboratories (1.3%) prepare the initial suspension in another way.

2.3. DILUENT USED FOR THE INITIAL SUSPENSION

157 laboratories (100%) specified it.
143 laboratories (91.1%) use Buffered Peptone Water for the initial suspension.
12 laboratories (7.6%) use Peptone salt solution for the initial suspension.
2 laboratories (1.3%) use another diluent for the initial suspension.

2.4. HOMOGENIZATION TECHNIQUE

157 laboratories (100%) specified it.
153 laboratories (97.4%) homogenize their sampling with a StomacherND.
2 laboratories (1.3%) used a manual homogenization.
2 laboratories (1.3%) used a Vortex mixer.

The average duration is **2.4 min** with a standard deviation of 1.0 min. The data 10, 15, 30 and 60 min given by 7 laboratories were not taken into account for this calculation. The minimum duration indicated is 0.5 min and the maximum one is 6.0 min.

2.5. LACTIC ACID BACTERIA

116 laboratories performed the enumeration.

TIME / SENDING SAMPLES - BEGINNING OF ANALYZES

116 laboratories specified it.

Analysis time	J0+1	J0+2	J0+3	J0+4	J0+7	J0+8	J0+9	J0+10	J0+16
Nb of laboratories	20	35	19	12	18	9	1	1	1

RESUSCITATION'S CONDITIONS

16 laboratories specified a duration of 0 min (or did not specify it) for the resuscitation step, they are not taken into account for the calculation.

- DURATION

100 laboratories specified it.

The average duration is **19.6 min** with a standard deviation of 13.9 min. The minimum duration indicated is 1.0 min and the maximum one is 60.0 min.

- TEMPERATURE

100 laboratories specified it.

The average temperature is **21.1°C** with a standard deviation of 4.0°C. The data 100°C given by one laboratory was not taken into account for this calculation. The minimum temperature indicated is 4.0°C and the maximum one is 37.0°C.

Method	Nb laboratories
ISO / NF EN ISO 15214	81
NM ISO 15214	12
AFNOR 3M 01/19-11/17	10
TEMPO LAB	6
Internal method	4
Other	3

Culture medium	Nb laboratories
MRS pH 5.7	91
Neogen® Petrifilm®	10
TEMPO LAB	6
MRS pH 6.4	8
Other	1

Preparation	Nb laboratories
Home made	28
Ready to use not pre-poured	64
Ready to use, plate, film, card	23

Plating method	Nb laboratories
Surface (agar plate, film)	22
Pour	87
Transfer Tempo filler ®	6

Incubation temperature	Nb laboratories
30°C	114
37°C	2

Incubation duration	Nb laboratories
70 – 72.4 h	97
42 – 48 h	19

2.6. PSEUDOMONAS

78 laboratories performed the enumeration.

TIME / SENDING SAMPLES - BEGINNING OF ANALYZES

78 laboratories specified it.

Analysis time	J0+1	J0+2	J0+3	J0+4	J0+6	J0+7	J0+8	J0+9	J0+16
Nb of laboratories	15	26	12	5	1	11	6	1	1

RESUSCITATION'S CONDITIONS

12 laboratories specified a duration of 0 min (or did not specify it) for the resuscitation step, they are not taken into account for the calculation.

- DURATION

66 laboratories specified it.

The average duration is **17.4 min** with a standard deviation of 11.4 min. The minimum duration indicated is 1.0 min and the maximum one is 60.0 min.

- TEMPERATURE

66 laboratories specified it.

The average temperature is **21.3°C** with a standard deviation of 3.1°C. The data 100°C given by one laboratory was not taken into account for this calculation. The minimum temperature indicated is 8.0°C and the maximum one is 37.0°C.

Method	Nb laboratories
ISO / NF EN ISO 13720	44
AFNOR BKR 23/09-05/15	23
NM ISO 13720	7
Internal method	2
Other	2

Culture medium	Nb laboratories
CFC	53
Rhapsody agar	24
Other	1

Preparation	Nb laboratories
Home made	22
Ready to use not pre-poured	30
Ready to use, plate, film, card	26

Incubation temperature	Nb laboratories
25°C	52
30°C	24
37±1°C	2

Incubation duration	Nb laboratories
41.5 - 48 h	77
24 h	1

Confirmation test	Nb laboratories
None	31
Oxydase	44
MALDI-TOF mass spectrometry	1

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2.7. BACILLUS CEREUS

129 laboratories performed the enumeration.

TIME / SENDING SAMPLES - BEGINNING OF ANALYZES

129 laboratories specified it.

Analysis time	J0+1	J0+2	J0+3	J0+4	J+6	J0+7	J0+8	J0+9	J0+10	J0+16
Nb of laboratories	17	43	24	6	1	22	10	4	1	1

RESUSCITATION'S CONDITIONS

21 laboratories specified a duration of 0 min (or did not specify it) for the resuscitation step, they are not taken into account for the calculation.

- DURATION

108 laboratories specified it.

The average duration is **21.0 min** with a standard deviation of 14.0 min. The minimum duration indicated is 1.0 min and the maximum one is 60.0 min.

- TEMPERATURE

108 laboratories specified it.

The average temperature is **21.6°C** with a standard deviation of 4.4°C. The minimum temperature indicated is 4.0°C and the maximum one is 47.0°C.

Method	Nb laboratories
ISO / NF EN ISO 7932 (+A1)	55
AFNOR BKR 23/06-02/10	32
AFNOR AES 10/10-07/10	20
NM ISO 7932 (+A1)	11
Microval 2014LR47	5
AFNOR BRD 07/26-03/19	4
Other	1

Culture medium	Nb laboratories
Mossel	64
COMPASS *Bacillus cereus* Agar	32
BACARA	24
TEMPO BC	5
RAPID'B. cereus	4

Preparation	Nb laboratories
Home made	25
Ready to use not pre-poured	17
Ready to use, plate, film, card	87

Plating method	Nb laboratories
Surface (agar plate, film)	107
Pour	15
Transfer Tempo filler ®	5

Incubation temperature	Nb laboratories
30°C	128
24°C	1

Incubation duration	Nb laboratories
20 – 24.5 h	84
42 - 48 h	45

Confirmation test	Nb laboratories
None	70
Biochemical (including hemolysis)	54
MALDI-TOF mass spectrometry	1

2.8. YEAST / MOULDS

70 laboratories performed the enumeration.

TIME / SENDING SAMPLES - BEGINNING OF ANALYZES

70 laboratories specified it.

Analysis time	J0+1	J0+2	J0+3	J0+4	J0+6	J0+7	J0+8	J0+9	J+16
Nb of laboratories	10	17	17	12	1	6	4	2	1

RESUSCITATION'S CONDITIONS

8 laboratories specified a duration of 0 min (or did not specify it) for the resuscitation step, they are not taken into account for the calculation.

- DURATION

62 laboratories specified it.

The average duration is **19.8 min** with a standard deviation of 12.9 min. The minimum duration indicated is 1.0 min and the maximum one is 60.0 min.

- TEMPERATURE

62 laboratories specified it.

The average temperature is **22.4°C** with a standard deviation of 4.9°C. The data 100°C given by one laboratory was not taken into account for this calculation. The minimum temperature indicated is 8.0°C and the maximum one 47.0°C.

Method	Nb laboratories
NF V08-059	39
→ *NM 08.0.123* [(1)]	7
AFNOR BKR 23/11-12/18	11
ISO / NF ISO 21527-1	5
AFNOR 3M 01/13-07/14	2
AOAC RI 041001	1
NM ISO 21527-1	1
Internal method	1
Other	3

Culture medium	Nb laboratories
YGC	36
Symphony	11
Chloramphenicol glucose agar	11
DRBC	4
Neogen® Petrifilm®	3
OGA	3
TEMPO YM	2

[(1)] *Similar method to NF V08-059 according to ONSSA (Office National de Sécurité Sanitaire des produits Alimentaires).*

Preparation	Nb laboratories
Home made	24
Ready to use not pre-poured	38
Ready to use, plate, film, card	7

Plating method	Nb laboratories
Surface (agar plate, film)	22
Pour	46
Transfer Tempo filler ®	1

Incubation temperature	Nb laboratories
25°C	66
22 – 22.5°C	2
30°C	2

Incubation duration	Nb laboratories
114 - 120 h	52
70 - 72 h	15
96 h	2
54 h	1

2.9. YEAST

69 laboratories performed the enumeration.

TIME / SENDING SAMPLES - BEGINNING OF ANALYZES

69 laboratories specified it.

Analysis time	J0+1	J0+2	J0+3	J0+4	J0+6	J0+7	J0+8	J0+9	J0+10
Nb of laboratories	8	16	13	13	1	13	3	1	1

RESUSCITATION'S CONDITIONS

9 laboratories specified a duration of 0 min (or did not specify it) for the resuscitation step, they are not taken into account for the calculation.

- DURATION

60 laboratories specified it.

The average duration is **21.5 min** with a standard deviation of 14.3 min. The minimum duration indicated is 1.0 min and the maximum one is 60.0 min.

- TEMPERATURE

60 laboratories specified it.

The average temperature is **21.8°C** with a standard deviation of 2.9°C. The minimum temperature indicated is 18.0°C and the maximum one is 37.0°C.

Method	Nb laboratories
NF V08-059	30
→ *NM 08.0.123* [(1)]	14
AFNOR BKR 23/11-12/18	10
ISO / NF EN ISO 21527-1	4
AFNOR 3M 01/13-07/14	4
NM ISO 21527-1	1
Internal method	1
Other	5

Culture medium	Nb laboratories
YGC	29
Chloramphenicol glucose agar	14
Symphony	10
DRBC	5
Neogen® Petrifilm®	4
OGA	2
Other	4

[(1)] *Similar method to NF V08-059 according to ONSSA (Office National de Sécurité Sanitaire des produits Alimentaires).*

Preparation	Nb laboratories
Home made	22
Ready to use not pre-poured	37
Ready to use, plate, film, card	10

Plating method	Nb laboratories
Surface (agar plate, film)	21
Pour	45

Incubation temperature	Nb laboratories
25°C	67
20°C	1
30°C	1

Incubation duration	Nb laboratories
120 h	50
70 - 72 h	17
96 h	2

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2.10. MOULDS

69 laboratories performed the enumeration.

TIME / SENDING SAMPLES - BEGINNING OF ANALYZES

69 laboratories specified it.

Analysis time	J0+1	J0+2	J0+3	J0+4	J0+6	J0+7	J0+8	J0+9	J0+10
Nb of laboratories	8	16	13	13	1	13	3	1	1

RESUSCITATION'S CONDITIONS

9 laboratories specified a duration of 0 min (or did not specify it) for the resuscitation step, they are not taken into account for the calculation.

- DURATION

60 laboratories specified it.

The average duration is **21.5 min** with a standard deviation of 14.3 min. The minimum duration indicated is 1.0 min and the maximum one is 60.0 min.

- TEMPERATURE

60 laboratories specified it.

The average temperature is **21.8°C** with a standard deviation of 2.9°C. The minimum temperature indicated is 18.0°C and the maximum one is 37.0°C.

Method	Nb laboratories
NF V08-059	30
→ *NM 08.0.123* [(1)]	14
AFNOR BKR 23/11-12/18	10
ISO / NF EN ISO 21527-1	4
AFNOR 3M 01/13-07/14	4
NM ISO 21527-1	1
Internal method	1
Other	5

Culture medium	Nb laboratories
YGC	29
Chloramphenicol glucose agar	14
Symphony	10
DRBC	5
Neogen® Petrifilm®	4
OGA	2
Other	4

[(1)] *Similar method to NF V08-059 according to ONSSA (Office National de Sécurité Sanitaire des produits Alimentaires).*

Preparation	Nb laboratories
Home made	22
Ready to use not pre-poured	37
Ready to use, plate, film, card	10

Plating method	Nb laboratories
Surface (agar plate, film)	21
Pour	45

Incubation temperature	Nb laboratories
25°C	67
20°C	1
30°C	1

Incubation duration	Nb laboratories
120 h	50
70 - 72 h	17
96 h	2

3. ASSESSMENT OF PERFORMANCE (INDIVIDUEL REPORTS)

Performance is assessed on **trueness**.
The assigned value of the contamination used to assess the trueness is the consensual value obtained with the results of all the participants. This value is obtained by a robust estimation method 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 result for the contaminated unit, 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 >10 days after sending) or when this information is not specified.

A statistical analysis has also been done to highlight potential relations between techniques used (preservation temperature, preparation of initial suspension and 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).

TRUENESS
The trueness reflects the closeness of your results to the contamination’s assigned value of samples. It has been evaluated for all enumerated flora.
Your result m_i is compared to the contamination’s assigned value, m_{pt}, obtained with algorithm A from the standard NF ISO 13528 applied to all laboratories results included in the statistical analysis.
When groups are constituted, each one is characterized by its own contamination’s assigned value.
The assigned value uncertainty is calculated with the following formula :

$$u(Xpt) = 1{,}25 \times \frac{\sigma pt}{\sqrt{p}}$$

with σ_{pt}, robust standard deviation (standard deviation for proficiency assessment) and p, number of laboratories.

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).

Z-score values are proposed with 3 significant figures.

The standard NF ISO 13528 specifies that:
- |z| ≤ 2,0 is considered as satisfactory (acceptable),
- 2,0 < |z| < 3,0 is considered as a warning signal (questionable),
- |z| ≥ 3,0 is considered as an action signal (or unacceptable).

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),
- histogram for the studied parameter (results of laboratories) with an asterisk indicating the location of your result,
- when necessary, your group in relation to the technique used,
- z score,
- number of laboratories which made analysis (and belonging to your group),
- number of laboratories included in the statistical analysis,
- assigned value of the contamination and standard deviation for proficiency assessment,
- number of laboratories with a satisfactory signal,
- number of laboratories with a warning signal,
- number of laboratories with an action signal.

3.1. LACTIC ACID BACTERIA

None significant effect of the analysis technique has been highlighted.

Lactic acid bacteria	
Number of laboratories included in the statistical analysis	112
Assigned value of the contamination (log cfu/g)	5.236
Uncertainty of assigned value (log cfu/g)	0.0170
Standard deviation for proficiency assessment (log cfu/g)	0.1437

3.2. PSEUDOMONAS

A significant “effect” of the method, culture medium and incubation temperature 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 :

Pseudomonas	
Number of laboratories included in the statistical analysis	74
Assigned value of the contamination (log cfu/g)	3.909
Uncertainty of assigned value (log cfu/g)	0.0300
Standard deviation for proficiency assessment (log cfu/g)	0.2064

3.3. BACILLUS CEREUS

A significant “effect” of the method, culture medium and plating method has been highlighted. Given the statistical data, results have been gathered in one group :

Bacillus cereus	
Number of laboratories included in the statistical analysis	114
Assigned value of the contamination (log cfu/g)	2.787
Uncertainty of assigned value (log cfu/g)	0.0253
Standard deviation for proficiency assessment (log cfu/g)	0.2162

3.4. YEAST / MOULDS

A significant “effect” of the plating method 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 :

Yeast - Moulds	
Number of laboratories included in the statistical analysis	67
Assigned value of the contamination (log cfu/g)	4.103
Uncertainty of assigned value (log cfu/g)	0.0433
Standard deviation for proficiency assessment (log cfu/g)	0.2837

3.5. YEAST

None significant effect of the analysis technique has been highlighted.

Yeast	
Number of laboratories included in the statistical analysis	66
Assigned value of the contamination (log cfu/g)	3.865
Uncertainty of assigned value (log cfu/g)	0.0578
Standard deviation for proficiency assessment (log cfu/g)	0.3759

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3.6. MOULDS

None significant effect of the analysis technique has been highlighted.

Moulds	
Number of laboratories included in the statistical analysis	66
Assigned value of the contamination (log cfu/g)	3.783
Uncertainty of assigned value (log cfu/g)	0.0330
Standard deviation for proficiency assessment (log cfu/g)	0.2145

Comment : We specify that the homogeneity criterion is unsatisfactory for Mould enumeration. Intra-sample standard deviation has then been included in the calculation of standard deviation for proficiency assessment (NF ISO 13528 §B.2.5.a).

3.7. EVOLUTION OF PERFORMANCE

You will find, at the end of the individual report, graphs representing evolution of your performance on different tests since the 61A scheme.

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

- Just one overtaking of the action limit ($z \leq -3.0$ or $z \geq 3.0$),
- 2 consecutives z scores out of 3 overtaking of the warning limit ($2.0 < z$ or $z < -2.0$),
- 6 consecutives z scores either positive or negative.