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PROFICIENCY TEST « RAEMA »

SCHEME N° 71A
(23 NOVEMBER 2020)
GENERAL REPORT

ACCREDITATION N°1-1836 SCOPE AVAILABLE ON WWW.COFRAC.FR

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1. GENERAL DATA

1.1. PARTICIPATING LABORATORIES

141 laboratories participated to the 71Ath Gel scheme on 23th November 2020 (J0).
We received **141** answers.

1.2. DELIVERY TIME OF THE PARCEL

Reception	J0	J0+1	J0+2	J0+3	J0+4	J0+5	J0+7	J0+8	J0+14	J0+15
Nb of laboratories	4	106	14	6	3	1	2	1	2	1

1.3. INFORMATIONS ABOUT SAMPLE

1.3.1. NATURE

- one sample included a strain of *Lactobacillus plantarum* at a concentration level of 1.10^6 cfu/g ;
- one sample included a strain of *Pseudomonas sp.* at a concentration level of 2.10^4 cfu/g ;
- one sample included a strain of *Bacillus cereus* at a concentration level of 1.10^5 cfu/g ;
- one sample included a strain of *Penicillium* at a concentration level of 5.10^3 cfu/g and a strain of *Rhodotorula rubra* at a concentration level of 5.10^3 cfu/g ;

1.3.2. SIZE

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

[(a)] Coordinator of the proficiency test « RAEMA »

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 26 November (J0+3), 30 November (J0+7) and 7 December 2020 (J0+14).

These checks were realized by a subcontractor accredited by Cofrac for *Bacillus cereus,* lactic bacteria and Yeast/Mould. The check of *Pseudomonas* was realized by the same subcontractor but not covered by Cofrac accreditation.

Homogeneity and stability of samples have 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 RECEPTION TEMPERATURE OF SAMPLES

118 laboratories specified it.

The average temperature is **8.0°C** with a standard deviation of 6.1°C. The minimum temperature indicated is 2.0°C and the maximum one is 26.4°C.

1.4.2 PRESERVATION TEMPERATURE OF SAMPLES BEFORE ANALYSIS

141 laboratories specified it.

The average temperature is **3.7°C** with a standard deviation of 0.7°C. The minimum temperature indicated is 2.0°C and the maximum one is 6.0°C.

Remark: Please note that samples must be conserved at 4°C on receipt, before analysis.

2. EXPLOITATION OF ANALYSIS REPORT

2.1. SIZE OF TEST SAMPLE

141 laboratories specified it.

The average size is **14 g** with a standard deviation of 6.4 g. The minimum size indicated is 1 g and the maximum one is 25 g.

2.2. PREPARATION OF THE INITIAL SUSPENSION

140 laboratories specified it.

139 laboratories prepare the initial suspension with adding diluent to gel.

1 laboratory prepares the initial suspension in another way.

2.3. DILUENT USED FOR THE INITIAL SUSPENSION

140 laboratories specified it.
124 laboratories use Buffered Peptone Water for the initial suspension.
10 laboratories use Peptone salt solution for the initial suspension.
6 laboratories used another diluent for the initial suspension.

2.4. HOMOGENIZATION TECHNIQUE

141 laboratories specified it.
135 laboratories homogenize their sampling with a Stomacher[ND].
6 laboratories used another technique.

The average duration is **2.4 min** with a standard deviation of 1.0 min. The data 10, 15, 20, 30 and 60 min given by 10 laboratories were not taken into account for this calculation. The minimum duration indicated is 0.5 min and the maximum one is 5.0 min.

2.5. LACTIC ACID BACTERIA

105 laboratories performed the enumeration.

DELIVERY TIME OF SAMPLES / BEGINNING OF ANALYZES

105 laboratories specified it.

Analysis time	J0+1	J0+2	J0+3	J0+4	J0+7	J0+8	J0+9	J0+10	J0+14	J0+15
Nb of laboratories	17	27	15	7	21	12	3	1	1	1

RESUSCITATION'S CONDITIONS

13 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

92 laboratories specified it.

The average duration is **20.0 min** with a standard deviation of 11.1 min. The minimum duration indicated is 1 min and the maximum one is 60 min. The data 120 min given by 2 laboratories were not taken into account for this calculation.

- TEMPERATURE

92 laboratories specified it.

The average temperature is **21.0°C** with a standard deviation of 3.4°C. The minimum temperature indicated is 4°C and the maximum one is 37°C. The data 100 min given by 1 laboratory was not taken into account for this calculation.

Method	Nb laboratories
NF EN ISO 15214	79
AFNOR 3M 01/19-11/17	9
TEMPO LAB	7
NM ISO 15214	5
Other	3

Culture medium	Nb laboratories
MRS pH 5.7	87
Petrifilm	9
TEMPO LAB	7
Other	1

Preparation	Nb laboratories
Home made	20
Ready to use not pre-poured	64
Ready to use, plate, film, card	21

Plating method	Nb laboratories
Surface (agar plate, film)	19
Pour	77
Culture medium for card	6

Incubation temperature	Nb laboratories
30°C	102
37°C	2
20°C	1

Incubation duration	Nb laboratories
69 – 73.5 h	83
48 h	22

2.6. PSEUDOMONAS

71 laboratories performed the enumeration.

DELIVERY TIME OF SAMPLES / BEGINNING OF ANALYZES

71 laboratories specified it.

Analysis time	J0+1	J0+2	J0+3	J0+4	J0+7	J0+8	J0+9	J0+14	J0+15
Nb of laboratories	13	22	13	3	11	5	2	1	1

RESUSCITATION'S CONDITIONS

11 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 **19.8 min** with a standard deviation of 12.4 min. The minimum duration indicated is 1 min and the maximum one is 60 min.

- TEMPERATURE

60 laboratories specified it.

The average temperature is **21.0°C** with a standard deviation of 3.0°C. The minimum temperature indicated is 9.0°C and the maximum one is 37.0°C. The data 100 min given by 1 laboratory was not taken into account for this calculation.

Method	Nb laboratories
NF EN ISO 13720	48
AFNOR BKR 23/09-05/15	17
NM ISO 13720	2
Other	4

Culture medium	Nb laboratories
CFC	53
Rhapsody agar	18
Other	0

Preparation	Nb laboratories
Home made	16
Ready to use not pre-poured	31
Ready to use, plate, film, card	24

Incubation temperature	Nb laboratories
25°C	52
30°C	18
22°C	1

Incubation duration	Nb laboratories
44 - 48 h	67
41 – 43 h	2
20 - 24 h	2

Confirmation test	Nb laboratories
None	28
Oxydase	39
Other	1

2.7. BACILLUS CEREUS

113 laboratories performed the enumeration.

DELIVERY TIME OF SAMPLES / BEGINNING OF ANALYZES

113 laboratories specified it.

Analysis time	J0+1	J0+2	J0+3	J0+4	J0+7	J0+8	J0+9	J0+10	J0+14	J0+15
Nb of laboratories	17	37	16	5	22	9	2	2	1	2

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

97 laboratories specified it.

The average duration is **20.8 min** with a standard deviation of 12.0 min. The minimum duration indicated is 1 min and the maximum one is 60 min. The data 120 min given by 2 laboratories were not taken into account for this calculation.

- TEMPERATURE

97 laboratories specified it.

The average temperature is **21.2°C** with a standard deviation of 3.2°C. The minimum temperature indicated is 4°C and the maximum one is 37°C. The data 100 min given by 1 laboratory was not taken into account for this calculation.

Method	Nb laboratories
NF EN ISO 7932	62
AFNOR BKR 23/06-02/10	20
AFNOR AES 10/10-07/10	20
NM ISO 7932	5
Microval 2014LR47	4
Other	2

Culture medium	Nb laboratories
Mossel	67
COMPASS *Bacillus cereus* Agar	22
BACARA	19
TEMPO BC	4
Other	1

Preparation	Nb laboratories
Home made	20
Ready to use not pre-poured	12
Ready to use, plate, film, card	81

Plating method	Nb laboratories
Surface (agar plate, film)	99
Pour	9
Culture medium for card	4

Incubation temperature	Nb laboratories
30°C	112
37°C	1

Incubation duration	Nb laboratories
19 - 25 h	72
42 - 48 h	41

Confirmation test	Nb laboratories
None	54
Biochemical (including hemolysis)	55
Other	1

Heat traitement before enumaration	Nb laboratories
Yes	1
No	110

2.8. YEAST / MOULDS

55 laboratories performed the enumeration.

DELIVERY TIME OF SAMPLES / BEGINNING OF ANALYZES

55 laboratories specified it.

Analysis time	J0+1	J0+2	J0+3	J0+4	J0+5	J0+7	J0+8	J0+9	J0+14
Nb of laboratories	5	19	11	3	1	10	2	3	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

47 laboratories specified it.

The average duration is **19.8 min** with a standard deviation of 13.1 min. The minimum duration indicated is 1 min and the maximum one is 60 min.

- TEMPERATURE

47 laboratories specified it.

The average temperature is **21.1°C** with a standard deviation of 3.2°C. The minimum temperature indicated is 8.5°C and the maximum one 30°C. The data 100 min given by 2 laboratories were not taken into account for this calculation.

Method	Nb laboratories
NF V08-059	34
→ *NM 08.0.123* [(1)]	3
AFNOR 3M 01/13-07/14	6
AFNOR BKR 23/11-12/18	6
NF ISO 21527-1	2
NM ISO 21527-1	1
Other	3

Culture medium	Nb laboratories
YGC	28
Symphony	7
Petrifilm	6
OGA	6
Chloramphenicol glucose agar	4
DRBC	1
Other	3

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

Preparation	Nb laboratories
Home made	17
Ready to use not pre-poured	29
Ready to use, plate, film, card	9

Plating method	Nb laboratories
Surface (agar plate, film)	17
Pour	38
Culture medium for card	0

Incubation temperature	Nb laboratories
24 - 25°C	51
20 - 22°C	2
30°C	2

Incubation duration	Nb laboratories
115 - 120 h	39
69 - 72 h	12
96 h	3
144 h	1

2.9. YEAST

58 laboratories performed the enumeration.

DELIVERY TIME OF SAMPLES / BEGINNING OF ANALYZES

58 laboratories specified it.

Analysis time	J0+1	J0+2	J0+3	J0+4	J0+5	J0+7	J0+8	J0+9	J0+15
Nb of laboratories	7	17	9	6	1	11	5	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

46 laboratories specified it.

The average duration is **22.3 min** with a standard deviation of 13.5 min. The minimum duration indicated is 1 min and the maximum one is 60 min. The data 120 min given by 2 laboratories were not taken into account for this calculation.

- TEMPERATURE

46 laboratories specified it.

The average temperature is **21.6°C** with a standard deviation of 2.9°C. The minimum temperature indicated is 20°C and the maximum one is 37°C.

Method	Nb laboratories
NF V08-059	32
→ *NM 08.0.123* [(1)]	5
AFNOR BKR 23/11-12/18	8
NF EN ISO 21527-1	5
AFNOR 3M 01/13-07/14	4
Other	4

Culture medium	Nb laboratories
YGC	27
Symphony	8
Chloramphenicol glucose agar	6
OGA	6
Petrifilm	4
DRBC	3
Other	4

Preparation	Nb laboratories
Home made	12
Ready to use not pre-poured	36
Ready to use, plate, film, card	10

Plating method	Nb laboratories
Surface (agar plate, film)	21
Pour	35
Culture medium for card	0

Incubation temperature	Nb laboratories
24 - 25°C	53
20 - 23°C	3
30°C	1

Incubation duration	Nb laboratories
120 h	37
69 - 72 h	15
96 h	4
144 h	1

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

2.10. MOULDS

58 laboratories performed the enumeration.

DELIVERY TIME OF SAMPLES / BEGINNING OF ANALYZES

58 laboratories specified it.

Analysis time	J0+1	J0+2	J0+3	J0+4	J0+5	J0+7	J0+8	J0+9	J0+15
Nb of laboratories	7	17	9	6	1	11	5	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

46 laboratories specified it.

The average duration is **22.3 min** with a standard deviation of 13.5 min. The minimum duration indicated is 1 min and the maximum one is 60 min. The data 120 min given by 2 laboratories were not taken into account for this calculation.

- TEMPERATURE

46 laboratories specified it.

The average temperature is **21.6°C** with a standard deviation of 2.9°C. The minimum temperature indicated is 20°C and the maximum one is 37°C.

Method	Nb laboratories
NF V08-059	32
→ *NM 08.0.123* [(1)]	5
AFNOR BKR 23/11-12/18	8
NF EN ISO 21527-1	5
AFNOR 3M 01/13-07/14	4
Other	4

Culture medium	Nb laboratories
YGC	27
Symphony	8
Chloramphenicol glucose agar	6
OGA	6
Petrifilm	4
DRBC	3
Other	4

Preparation	Nb laboratories
Home made	12
Ready to use not pre-poured	36
Ready to use, plate, film, card	10

Plating method	Nb laboratories
Surface (agar plate, film)	21
Pour	35
Culture medium for card	0

Incubation temperature	Nb laboratories
24 - 25°C	53
20 - 23°C	3
30°C	1

Incubation duration	Nb laboratories
120 h	37
69 - 72 h	15
96 h	4
144 h	1

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

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 be done to highlight potential relations between techniques used (delay of analysis, preservation temperature, 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, X_{pt}, obtained with algorithm A from the standard ISO 13528 applied to all laboratories results included in the statistical analysis.

A z score is then calculated with the following formula : $z_i = \frac{m_i - X_{pt}}{\sigma_{pt}}$, where σ_{pt} is the standard deviation for proficiency assessment (robust estimation of the standard deviation obtained by participants). When groups are constituted, each one is characterized by its own contamination's assigned value.
The standard ISO 13528 specifies that z score included between -2 and +2 must be considered as satisfactory signal. A z score included between -2 and -3 or between +2 and +3 must be considered as a warning signal. A z score lower than -3 or higher than +3 must be considered as an action signal

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	
Assigned value of the contamination (log CFU/g)	5.828
Uncertainty of assigned value (log CFU/g)	0.0431
Standard deviation for proficiency assessment (log CFU/g)	0.3449

3.2. PSEUDOMONAS

A significant "effect" of the preparation mode 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	
Assigned value of the contamination (log CFU/g)	4.489
Uncertainty of assigned value (log CFU/g)	0.0481
Standard deviation for proficiency assessment (log CFU/g)	0.3195

3.3. BACILLUS CEREUS

None significant effect of the analysis technique has been highlighted.

Bacillus cereus	
Assigned value of the contamination (log CFU/g)	5.252
Uncertainty of assigned value (log CFU/g)	0.0370
Standard deviation for proficiency assessment (log CFU/g)	0.3048

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	
Assigned value of the contamination (log CFU/g)	4.185
Uncertainty of assigned value (log CFU/g)	0.0435
Standard deviation for proficiency assessment (log CFU/g)	0.2511

3.5. YEAST

A significant "effect" of the incubation 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:

Yeast	
Assigned value of the contamination (log CFU/g)	3.802
Uncertainty of assigned value (log CFU/g)	0.0567
Standard deviation for proficiency assessment (log CFU/g)	0.3302

3.6. MOULDS

None significant effect of the analysis technique has been highlighted.

Moulds	
Assigned value of the contamination (log CFU/g)	3.800
Uncertainty of assigned value (log CFU/g)	0.0370
Standard deviation for proficiency assessment (log CFU/g)	0.2135

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 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.