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Yogurt — Identification of characteristic microorganisms (*Lactobacillus delbrueckii* subsp. *bulgaricus* and *Streptococcus thermophilus*)

AMENDMENT 1: Inclusion of performance testing of culture media and reagents

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Forewords

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This document was prepared by Technical Committee ISO/TC 34, *Food products*, Subcommittee SC 5, *Milk and milk products*, and the International Dairy Federation (IDF). It is being published jointly by ISO and IDF.

Any feedback or questions on this document should be directed to the user's national standards body. A complete listing of these bodies can be found at www.iso.org/members.html.

IDF (the International Dairy Federation) is a non-profit private sector organization representing the interests of various stakeholders in dairying at the global level. IDF members are organized in National Committees, which are national associations composed of representatives of dairy-related national interest groups including dairy farmers, dairy processing industry, dairy suppliers, academics and governments/food control authorities.

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This document was prepared by the IDF *Standing Committee on Analytical Methods for Dairy Microorganisms* and ISO Technical Committee ISO/TC 34, *Food products*, Subcommittee SC 5, *Milk and milk products*. It is being published jointly by ISO and IDF.

The work was carried out by the *Standing Committee on Analytical Methods for Dairy Microorganisms* under the aegis of its project leader Mrs Barbara Gerten.

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Yogurt — Identification of characteristic microorganisms (*Lactobacillus delbrueckii* subsp. *bulgaricus* and *Streptococcus thermophilus*)

AMENDMENT 1: Inclusion of performance testing of culture media and reagents

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Clause 2

Replace the reference to ISO 8261|IDF 122 with the following:

ISO 6887 (all parts), *Microbiology of the food chain — Preparation of test samples, initial suspension and decimal dilutions for microbiological examination*

Add the following normative reference:

ISO 11133, *Microbiology of food, animal feed and water — Preparation, production, storage and performance testing of culture media*

Clause 5, first paragraph

Replace the reference “ISO 8261|IDF 122” with “ISO 6887 (all parts)”.

Clause 5, after the first paragraph

Add the following text:

For current laboratory practice, see ISO 7218 and ISO 11133.

The general specifications of ISO 11133 are applicable to the preparation and performance testing of the culture media described in this clause. If culture media or reagents are prepared from dehydrated complete media/reagents or if ready-to-use media/reagents are used, follow the manufacturer's instructions regarding preparation, storage conditions, expiry date and use.

The shelf life of the media and reagents indicated in this clause has been determined in some studies. The user shall verify these under their own storage conditions (in accordance with ISO 11133).

Performance testing of culture media and reagents is described in 5.5.

After 5.4

Add the following as 5.5.

5.5 Performance testing

The definition of productivity is specified in ISO 11133. In general, follow the procedures for performance testing described in ISO 11133. [Table 1](#) provides the performance testing for the quality assurance of the culture media and reagents.

Table 1 — Performance testing for the quality assurance of the culture media and reagents

Medium	Function	Incubation	Control strains ^a	WDCM numbers ^b	Method of control	Criteria ^c
Catalase reagent (3 % hydrogen peroxide solution)	Detection of catalase after adding hydrogen peroxide solution	Room temperature over 20 min	<i>Staphylococcus aureus</i> or <i>Campylobacter jejuni</i> or <i>Listeria monocytogenes</i> or <i>Listeria innocua</i> or <i>Listeria ivanovii</i>	00032 or 00034 00005 00020 or 00021 00017 00018	qualitative	Positive reaction: Formation of bubbles of oxygen
			<i>Lactobacillus delbrueckii</i> subsp. <i>bulgaricus</i> or <i>Streptococcus thermophilus</i> or <i>Enterococcus faecalis</i> or <i>Enterococcus faecium</i>	00102 00134 00009 or 00087 or 00176 00177 or 00178	qualitative	Negative reaction: No formation of bubbles of oxygen
Glucose MRS broth with overlay agar	Detection of CO ₂ production	18 h ± 3 h / 37 °C ± 1 °C	<i>Lactobacillus brevis</i>	00099	qualitative	Positive reaction: Agar layer detaches itself from the underlying contents
			<i>Lactobacillus delbrueckii</i> subsp. <i>bulgaricus</i>	00102	qualitative	Negative reaction: No gas production, agar layer not detached
Litmus milk	Growth at 10 °C	up to 7 d / 37 °C ± 1 °C	<i>Streptococcus thermophilus</i>	00134	qualitative	Negative reaction: No change
	Growth at 45 °C	up to 7 d / 37 °C ± 1 °C	<i>Streptococcus thermophilus</i>	00134	qualitative	Positive reaction: turn to pink and than coagulation After coagulation the colour remains pink due to very slow and often incomplete reduction of litmus, with a more intensely coloured upper ring.
M17 broth	Productivity	18 h ± 3 h / 37 °C ± 1 °C	<i>Streptococcus thermophilus</i>	00134	qualitative	Turbidity (1-2)

^a Strain free of choice; one of the strains has to be used as a minimum.

^b Refer to the reference strain catalogue on <http://www.wfcc.info> for information on culture collection strain numbers and contact details;

WDCM: World Data Centre for Microorganisms.

^c Growth is categorized as 0: no growth; 1: weak growth (partial inhibition); 2: good growth (see ISO 11133).

Table 1 (continued)

Medium	Function	Incubation	Control strains ^a	WDCM numbers ^b	Method of control	Criteria ^c
NaCl M17 broth	Growth in presence of 6,5 % NaCl	up to 7 d / 37 °C ± 1 °C	<i>Enterococcus faecalis</i>	00009 or 00087 or 00176	qualitative	Positive reaction: Turbidity
			<i>Streptococcus thermophilus</i>	00134	qualitative	Negative reaction: No turbidity
MRS broth	Productivity	18 h ± 3 h / 37 °C ± 1 °C anaerobic atmosphere	<i>Lactobacillus delbrueckii</i> subsp. <i>bulgaricus</i>	00102	qualitative	Turbidity (1-2)
	Growth at 15 °C	up to 7 d / 15 °C ± 1 °C anaerobic atmosphere	<i>Lactobacillus casei</i> or <i>Lactobacillus plantarum</i>	00100 00104	qualitative	Positive reaction: Turbidity
			<i>Lactobacillus delbrueckii</i> subsp. <i>bulgaricus</i>	00102	qualitative	Negative reaction: No turbidity
	Growth at 45 °C	up to 7 d / 45 °C ± 1 °C anaerobic atmosphere	<i>Lactobacillus delbrueckii</i> subsp. <i>bulgaricus</i>	00102	qualitative	Positive reaction: Turbidity
			<i>Lactobacillus plantarum</i>	00104	qualitative	Negative reaction: No turbidity
Skimmed milk	Productivity	18 h ± 3 h / 37 °C ± 1 °C anaerobic atmosphere	<i>Lactobacillus delbrueckii</i> subsp. <i>bulgaricus</i>	00102	qualitative	Growth (1-2)

^a Strain free of choice; one of the strains has to be used as a minimum.

^b Refer to the reference strain catalogue on <http://www.wfcc.info> for information on culture collection strain numbers and contact details;
WDCM: World Data Centre for Microorganisms.

^c Growth is categorized as 0: no growth; 1: weak growth (partial inhibition); 2: good growth (see ISO 11133).

Clause 6, first paragraph

Replace the reference “ISO 8261|IDF 122” with “ISO 6887 (all parts)”.

B.4, first paragraph

Replace the reference “ISO 8261|IDF 122” with “ISO 6887 (all parts)”.