

SUGAR-FREE AGAR (SFA)

ENUMERATION OF CONTAMINANTS IN DAIRY PRODUCTS

1 INTENDED USE

Sugar-Free Agar for the enumeration of contaminants in dairy products is a carbohydrate-free medium used for the detection and enumeration of microorganisms that do not undergo specific fermentation processes during the preparation of dairy products. The result of the enumeration furnishes an indication of the level of contamination of the sample tested.

This media is recommended for the enumeration of contaminating microorganisms in butter, fermented milk and fresh cheeses.

The typical composition responds to that defined in the standard ISO 13559.

2 PRINCIPLES

The presence of the relatively non-nutritive substances supplied by peptones (Tryptone and pancreatic digest of gelatin), as well as the absence of fermentable carbohydrates, leads to a substantial reduction in the growth of specific microorganisms, in particular lactobacilli. This favors the growth of microorganisms that alter the quality of the product.

Sodium chloride maintains the osmotic balance..

3 TYPICAL COMPOSITION

The composition can be adjusted in order to obtain optimal performance.

For 1 liter of media :

- Tryptone 7,5 g
- Pancreatic digest of Gelatin 7,5 g
- Sodium chloride 5,0 g
- Bacteriological agar..... 14,0 g

pH of the ready-to-use media at 25 °C : 7,5 ± 0,2.

4 PREPARATION

Preparation from dehydrated media :

- Dissolve 34,0 g of dehydrated media (BK126) IN 1 liter of distilled or demineralized water.
- Slowly bring to boiling, stirring with constant agitation until complete dissolution.
- Dispense in tubes or vials.
- Sterilize in an autoclave at 121°C for 15 minutes.
- Cool and maintain in a molten state at 44-47 °C.
- Pour into Petri plates and let solidify on a cold, flat surface.

✓ **Reconstitution :**
34,0 g/L

✓ **Sterilization :**
15 min at 121 °C

Use of ready-to-melt media :

- If prepared in advance as above or when using ready-to-melt media (BM122), melt the medium using the least amount of time necessary to achieve total liquefaction.
- Pour into sterile Petri plates and let solidify on a cold, flat surface. .

NOTE : For fermented milk, if necessary, adjust the pH of the media so that after sterilization and cooling, the pH of the media taken at that moment and that of the initial media opening always is equal to 8,0 ± 0,1 at 25 °C.

5 INSTRUCTIONS FOR USE

- Dry the plates in an incubator, with the covers partially removed.
- On the surface of plates prepared as above, transfer 0,1 mL of the sample to test and its serial dilutions.
- Spread the inoculum over the surface of the agar with a sterile triangle or « hockey stick ».
- Incubate at 30 ± 1 °C for 72 ± 2 hours.

✓ **Inoculation :**
0,1 mL on surface

✓ **Incubation :**
72 ± 2 h at 30 °C

6 RESULTS

Retain only plates containing less than 150 colonies.

Small "pinhead" colonies are not counted, since they represent normal lactic acid flora. In case of doubt, carry out a catalase test with a significant number of colonies.

The production of gas bubbles confirms the presence of a contaminant.

7 QUALITY CONTROL

Dehydrated media : off-white powder, free-flowing and homogeneous.

Prepared media : light amber agar.

Typical culture response after 72 hours of incubation at 30 °C

Microorganisms		Growth (Productivity Ratio : P_R)
<i>Escherichia coli</i>	WDCM 00013	$P_R \geq 50$ %
<i>Staphylococcus aureus</i>	WDCM 00034	$P_R \geq 50$ %
<i>Lactobacillus casei</i> subsp. <i>rhamnosus</i>	WDCM 00101	Slowed
<i>Lactococcus lactis</i> subsp. <i>lactis</i>	ATCC® 11454	Slowed

8 STORAGE / SHELF LIFE

Dehydrated media : 2-30 °C.

Ready-to-melt media in vials : 2-25 °C

The expiration dates are indicated on the labels.

Prepared media in plates(*) : 30 days at 2-8 °C.

Prepared media in vials(*) : 180 days at 2-25 °C.

(*) Benchmark value determined under standard preparation conditions, following manufacturer's instructions.

9 PACKAGING

Dehydrated media :

500 g bottle BK126HA

Ready-to-melt media :

10 x 100 mL vials BM12208

10 BIBLIOGRAPHY

Circulaire DQ/SVHA/N86/N° 8163 du 25 Novembre 1986. Méthodes d'analyse des beurres.

ISO 13559 / IDF 153. Novembre 2002. Beurre, laits fermentés et fromage frais. Dénombrement des microorganismes contaminants. Méthode par comptage des colonies à 30 °C.

11 ADDITIONAL INFORMATION

The information provided on the labels take precedence over the formulations or instructions described in this document and are susceptible to modification at any time, without warning.

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