

Also known as

Trypticase Glucose Yeast Agar; TGY; TGY Agar; Standard Methods Agar, SMA; SM Agar

Specification

Medium for aerobic plate counts by the surface inoculation method, according to ISO standards 4833, 8552, 17410 and IFU no. 6.

Formula * in g/L

Casein peptone.....	5.0
Yeast extract.....	2.5
Dextrose.....	1.0
Agar.....	15.0

Final pH 7.0 ±0.2 at 25 °C

* Adjusted and /or supplemented as required to meet performance criteria

Directions

Suspend 23,5 g of powder in 1 L of distilled water. Dissolve by bringing to the boil with frequent stirring. Distribute into final containers and sterilize in the autoclave at 121°C for 15 minutes.

Description

The Plate Count Agar formulation is according to that of Buchbinder *et al.* as recommended in their study of media for the plate count of microorganisms.

The original formulation of the standardized agar for dairy microbiology has been modified in order to avoid the addition of milk. This new composition allows the growth of most microorganisms without any further additions.

This medium's formulation is equivalent to that described by the 'Standard Methods for the Examination of Dairy products', the USP's 'Tryptone Glucose Yeast Agar', the 'Deutsche Landwirtschaft' and to the APHA and AOAC's Plate Count Agar. This is the medium of choice for the plate count of any type of sample.

Technique

Prepare 10-fold serial dilutions of the sample and take 1 ml aliquots from each dilution (in duplicates) and put them into sterile Petri plates. Pour approx. 20 ml of sterile cooled medium (around 45 °C) in each of the plates. Mix gently by swirling the plate in the form of a figure 8. Leave the plates undisturbed to solidify and incubate in an inverted position. The incubation time and temperature depend on the type of microorganism under study. For a general aerobic count, incubate for 3 days at 30 °C. Taking readings after 24, 48 and 72 hours.

The plate count method proposed by the APHA consists of pouring the molten agar at 50 °C on plates containing the diluted samples (pour plate technique). The final count is carried out after 48 hours of incubation at 32-35 °C.

For microorganisms with other temperature requirements, the following incubations have been suggested: 2 days at 32-35 °C, 2-3 days at 45 °C, 2 days at 55 °C, 3-5 days at 20 °C, 10 days at 6.5 °C ± 1 °C.

Sample dilutions are prepared with 1/4 Ringer's solution, buffered Peptone Water, or Maximum Recovery Diluent depending on their nature.

The poured plate count method is preferred to the spread plate technique, since it gives higher counts. Nevertheless, the latter facilitates isolation and reseeded of the colonies.

Quality control

Incubation temperature: 30 ± 1 °C

Incubation time: 72 ± 3 h

Inoculum: Practical range 100±20 CFU. min. 50 CFU (productivity), according to ISO 11133:2014/Amd 1:2018. Spiral Plate Method.

Microorganism
Growth
Remarks

<i>Bacillus subtilis</i> ATCC® 6633	Productivity > 0.70	-
<i>Staphylococcus aureus</i> ATCC® 25923	Productivity > 0.70	-
<i>Listeria monocytogenes</i> ATCC® 35152	Productivity > 0.70	-
<i>Escherichia coli</i> ATCC® 8739	Productivity > 0.70	-
<i>Pseudomonas fluorescens</i> ATCC® 13525	Productivity > 0.70	10 days/ 6.5 °C ±1 °C (ISO 17410:2019)

References

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- HORWITZ, W. (2000) Official Methods of Analysis. AOAC International. Gaithersburg.
- IFU Method No 6 (1996) Mesophilic, thermophilic and thermophilic bacteria: Spores Count. D-1 Mesophilic Aerobic Sporeforming bacteria: Spores count.
- ISO 4833 (2003) Microbiology of food and animal feeding stuffs. Horizontal method for the enumeration of microorganisms. Colony count technique at 30°C.
- ISO 8552 (2004) Milk - Estimation of psychrotrophic microorganisms. Colony count technique at 21°C (Rapid method).
- ISO 11133:2014/ Adm 1:2018. Microbiology of food, animal feed and water. Preparation, production, storage and performance testing of culture media.
- ISO 17410 (2019) Horizontal method for the enumeration of psychrotrophic microorganisms.
- MARSHALL, R.T. (1992) Standard Methods for the Examination of Dairy Products. 16th ed. APHA. Washington.

Storage

For laboratory use only. Keep tightly closed, away from bright light, in a cool dry place (+4 °C to 30 °C).