

PRODUCT CODE: 413819

Tryptone Soy Agar (TSA) (Ph. Eur., USP, ISO) (Dehydrated Culture Media) for microbiology

Practical information

Applications: nonselective enumeration.

Categories: Water / Clinical / Food / Cosmetics

Industry: Pharmaceutical/Veterinary / Cosmetics / Clinical / Food

Regulations: USP / ISO 11133 / ISO 11930 / ISO 18415 / ISO 18416 / European Pharmacopoeia / ISO 21149 / ISO 21150 / ISO 22717 / ISO 22718 / ISO 9308

Preparation

Suspend 40 grams of the medium in one litre of distilled water. Mix well and dissolve by heating with frequent agitation. Boil for one minute until complete dissolution. Dispense into appropriate containers and sterilize in autoclave at 121 °C for 15 minutes. Large quantities may require a longer sterilization time, but the temperature should not be increased.

To prepare blood plates for haemolysis studies, add 5 - 10% of defibrinated sterile blood to the sterile medium, previously cooled to 45°C. Be careful to avoid bubble formation when adding the blood to the cooling medium and rotate the flask or bottle slowly to create a homogeneous solution.

Uses

Trypticasein Soy Agar (TSA) is a general-purpose medium very rich in nutrients for general use in microbiological laboratories and for the cultivation and isolation of fastidious or non-fastidious microorganisms, or for the maintenance of stock culture. It supports the abundant growth of fastidious organisms such as pneumococci, streptococci, Neisseria, etc. from clinical samples.

Containing two peptones as rich nitrogen sources, obtained by the enzymatic hydrolysis of casein and soy proteins, this medium supports the growth of a great variety of microorganisms, including fastidious aerobes and anaerobes. Soy peptone also contains natural sugars which promote bacterial growth.

Sodium chloride supplies essential electrolytes for transport and osmotic balance. Bacteriological agar is the solidifying agent.

Since it lacks carbohydrates it is very useful in the study of haemolytic reactions and also in the preparation of chocolate agar. If desired, antibiotics can easily be incorporated as well as other supplements or inhibitory agents. Some of the microorganisms that grow on this medium are the following: *Streptococcus*, *Neisseria*, *Brucella*, *Corynebacteria*, *Listeria*, *Pasteurella*, *Vibrio*, *Haemophilus vaginalis*, *Candida*, etc.

The European Pharmacopoeia, USP recommends this medium in the paragraph 2.6.12: "Microbiological examination of non – sterile products: Microbial enumeration test" for the examination of TAMC and TYMC in products.

The ISO 11133 Norm recommends the Trypticasein Soy Agar (TSA) as a reference medium.

For total aerobic microbial count (Suitability of counting method in the presence of the product) inoculate ≤100 cfu/ml of *Staphylococcus aureus*, *Pseudomonas aeruginosa* and *Bacillus subtilis* at 30-35°C for ≤ 3 days, and for *Candida albicans* ATCC 10231 and *Aspergillus brasiliensis* ATCC 16404 inoculate ≤100 cfu/ml at 30-35 °C ≤ 5 days.

For general uses:

Examination of TAMC and TYMC in products according to European Pharmacopoeia:

Membrane filtration:

- Prepare the product sample suspending, dissolving or diluting the product to be examined in the Trypticasein Soy Broth.
- Transfer the appropriate amount of the sample to a membrane filter.
- Place the membrane to the surface of Trypticasein Soy Agar (Ref. 413819) in case of TAMC or Sabouraud Glucose Agar (Ref. 413802) in case of TYMC.
- Incubate the plate of Trypticasein Soy Agar (Ref. 413819) at 30-35 °C for 3-5 days and the plate of Sabouraud Glucose Agar (Ref. 413802) at 20-25 °C for 5-7 days.

Plate-count methods:

- Prepare the product sample suspending, dissolving or diluting the product to be examined in the Trypticasein Soy Broth.
- Inoculate the plates of Trypticasein Soy Agar (Ref. 413819) in case of TAMC or Sabouraud Glucose Agar (Ref. 413802) in case of TYMC, conforming to the pour-plate method or the surface-spread method.
- Incubate the plates of Trypticasein Soy Agar (Ref. 413802) at 30-35 °C for 3-5 days and the plates of Sabouraud Glucose Agar (Ref. 413802) at 20-25 °C for 5-7 days.
- Select the plates corresponding to a given dilution and showing the highest number of colonies less than 250 (TAMC) or 50 (TYMC).

Composition

See in Data Sheet (TDS).

Quality control

Solubility	Appearance	Color of the dehydrated medium	Color of the prepared medium	Final pH (25°C)
w/o rests	Fine powder	Beige	Amber / Cherry-red with blood	7,3 ± 0,2

Microbiological Test

According to European Pharmacopoeia:

Total aerobic microbial count (TAMC); *Staphylococcus aureus* ATCC 6538, *Pseudomonas aeruginosa* ATCC9027, *Bacillus subtilis* ATCC 6633, *Candida albicans* ATCC 10231 and *Aspergillus brasiliensis* ATCC 16404:

Incubation conditions: (30-35 °C / <=3 days: bacteria /<=5 days: fungi).

Inoculation conditions: (10³-10⁴ CFU).

According to ISO 11133:

Incubation conditions: *Bacillus cereus* ATCC 11778 (30±1 °C / 24-48 h) / *Bacillus subtilis* ATCC 6633 (30±1 °C / 24-48 h) / *Escherichia coli* ATCC 8739 (44±1 °C / 21±3 h) / *Escherichia coli* O157 H7 ATCC 700728 (37±1 °C / 21±3 h) / *Listeria monocytogenes* ATCC 13932 (37±1 °C / 44±4 h) / *Staphylococcus aureus* ATCC 25923 (37±1 °C 24-48 h) / *Escherichia coli* ATCC 11775 (36±2 °C/20±2 h) / *Clostridium perfringens* ATCC 13124 (44±1 °C, anaerobic conditions / 21±3 h) / *Pseudomonas aeruginosa* ATCC 10145 (44±4 h / 36±2 °C) / *Enterococcus faecalis* ATCC 29212 (36±2 °C / 44±4 h).

Inoculation conditions: (100±20. Min. 50 CFU).

Reference media: Media batch TSA already validated.

Rest of strains; *Streptococcus pneumoniae* ATCC 6305, *Streptococcus pyogenes* ATCC 19615:

Incubation conditions: (30-35°C / 18-24 h).

Reference media: Media batch TSA already validated.

Microorganism	Specification	Characteristic reaction
<i>Pseudomonas aeruginosa</i> ATCC 10145	Good growth, >70%	
<i>Candida albicans</i> ATCC 10231	Good growth	
<i>Escherichia coli</i> ATCC 11775	Good growth, >70%	
<i>Bacillus cereus</i> ATCC 11778	Good growth, >70%	
<i>Clostridium perfringens</i> ATCC 13124	Good growth, >70%	
<i>Listeria monocytogenes 4b</i> ATCC 13932	Good growth, 70%	
<i>Aspergillus brasiliensis</i> ATCC 16404	Good growth	
<i>Streptococcus pyogenes</i> ATCC 19615	Good growth	Beta hemolysis
<i>Staphylococcus aureus</i> ATCC 25923	Good growth, >70%	Beta hemolysis
<i>Enterococcus faecalis</i> ATCC 29212	Good growth, >70%	
<i>Streptococcus pneumoniae</i> ATCC 6305	Good growth	Alpha hemolysis
<i>Staphylococcus aureus</i> ATCC 6538	Good growth	
<i>Bacillus subtilis</i> ATCC 6633	Good growth, >70%	
<i>Escherichia coli</i> O157:H7 ATCC 700728	Good growth, >70%	
<i>Escherichia coli</i> ATCC 8739	Good growth, >70%	
<i>Pseudomonas aeruginosa</i> ATCC 9027	Good growth	

Storage

Temp. Min.: 2 °C

Temp. Max.: 25 °C

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