

## TRYPTICASEIN SOY AGAR (TSA) (EUROPEAN PHARMACOPOEIA)

**CAT Nº: 1068**

For general bacteriology methods and the determination of hemolytic reactions

### FORMULA IN g/l

Pancreatic Digest of Casein	15.00	Sodium Chloride	5.00
Papaic Digest of Soya Bean	5.00	Bacteriological Agar	15.00

**Final pH 7.3 ± 0.2 at 25°C**

### PREPARATION

Suspend 40 grams of the medium in one liter 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 hemolysis 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 cooled medium and rotate the flask or bottle slowly to create a homogeneous solution. The prepared medium should be stored at 8-15°C. The color with the blood added is cherry red.

The dehydrated medium should be homogeneous, free-flowing and beige in color. If there are any physical changes, discard the medium.

### 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.

Some of the microorganisms that grow on this medium are the following: *Streptococcus*, *Neisseria*, *Brucella*, *Corynebacteria*, *Listeria*, *Pasteurella*, *Vibrio*, *Haemophilus vaginalis*, *Candida*, etc.

Since it lacks carbohydrates it is very useful in the study of hemolytic reactions and also in the preparation of chocolate agar. If desired, antibiotics can easily be incorporated as well as other supplements or inhibitory agents. Inoculate and incubate at 30-35°C and observe after 18-24 hours.

The European Pharmacopoeia recommends this medium in the Paragraph 2.6.12 "Microbiological examination of non – sterile products: Microbial enumeration test" for the preparation of test strain.

Inoculate and incubate at 30-35°C for 18 - 24 hours for *Staphylococcus aureus* ATCC 6538, *Pseudomonas aeruginosa* ATCC 9027 and *Bacillus subtilis* ATCC 6633.

For total aerobic microbial count (growth promotion) of *Staphylococcus aureus* ATCC 6538, *Pseudomonas aeruginosa* ATCC 9027 and *Bacillus subtilis* ATCC 6633, inoculate ≤100 cfu/ml at 30-35°C for ≤ 3 days, of and for *Candida albicans* ATCC 10231 and *Aspergillus brasiliensis* ATCC 16404 inoculate ≤100 cfu/ml at 30-35 °C ≤ 5 days.

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

## MICROBIOLOGICAL TEST

The following results were obtained in the performance of the medium, with the blood and without blood added, from type cultures after incubation at a temperature of 30-35°C and observed after 18-24 hours

Microorganisms	Growth	Growth with 5% sheep's Blood	Hemolysis	Inoculum Cfu/ml	Recovery Rate %
<i>Staphylococcus aureus</i> ATCC 25923	Good	Good	Beta	$10^2-10^3$	$\geq 70$
* <i>Staphylococcus aureus</i> ATCC 6538	Good			$\leq 100$	
<i>Staphylococcus epidermidis</i> ATCC 12228	Good	Good		$10^2-10^3$	$\geq 70$
<i>Streptococcus pneumoniae</i> ATCC 6303	Good	Good	Alpha	$10^2-10^3$	$\geq 70$
<i>Streptococcus pyogenes</i> ATCC 19615	Good	Good	Beta	$10^2-10^3$	$\geq 70$
* <i>Pseudomonas aeruginosa</i> ATCC 9027	Good			$\leq 100$	
* <i>Bacillus subtilis</i> ATCC 6633	Good			$\leq 100$	
** <i>Candida albicans</i> ATCC 10231	Good			$\leq 100$	
** <i>Aspergillus brasiliensis</i> ATCC 16404	Good			$\leq 100$	

\*According to European Pharmacopoeia 7.0. Total recount of aerobics  $\leq 100$  cfu/ml to incubate at 30-35°C for  $\leq 3$  days and preparation of test strains for 30-35°C during 18-24 hours

\*\* Total recount of aerobics  $\leq 100$  cfu/ml to incubate at 30-35°C for  $\leq 5$  days and preparation of test strains 30-35°C during 18-24 hours

## BIBLIOGRAPHY

Altord, Wiese, and Cunter, J. Bact., 69:516. 1955. Ctapper and Parker, J. Bact. 70. 1955.

Standard Methods for the Examination of Dairy Products. 11th Edition. APHA., Inc. New York, 1960.

Curry, A.S., G. Joyce and G.N. Mcerven, Jr. 1993 CTFA Microbiology guideline. The Cosmetic To iletary and Fragrance Association, Inc. Washington D.C.

European Pharmacopoeia. 7.0



## STORAGE

Once opened keep powdered medium closed to avoid hydration.

