TECHNICAL DATA SHEET

TRYPTO-CASEIN SOY (TSA) AGAR

NUTRIENT MEDIA

1 INTENDED USE

Trypto Casein-Soy Agar is a universal nutrient medium suitable for a wide range of uses. In light of its excellent nutritive value, it can be used for the growth and isolation of both aerobic and anaerobic bacteria and to favor the development of the most fastidious microorganisms. Poured in plates or on strips, it is useful in rapid tests for examining the contamination of surfaces. In addition, under the denomination TSA (Tryptic Soy Agar), this formula corresponds to the reference medium used for the evaluation of productivity and selectivity criteria in the context of the ISO 11133 standard.

The typical composition corresponds to that defined in European (EP), United States (UP) and Japanese (JP) Pharmacopeias.

2 PRINCIPLES

The combination of Tryptone and Papaic digest of soybean meal leads to optimal growth, due to the synergy between the protein supply of casein and the carbohydrate supply of soybeans, favoring the growth of most fastidious or non-fastidious microorganisms.

Sodium chloride maintains osmotic balance.

3 TYPICAL COMPOSITION

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

For 1 liter of media:

- Tryptone ..... 15.0 g
- Papaic digest of soybean meal ..... 5.0 g
- Sodium chloride ..... 5.0 g
- Bacteriological agar ..... 15.0 g

pH of the ready-to-use media at 25 °C: 7.3 ± 0.2.

4 PREPARATION

Preparation from dehydrated medium:

- Dissolve 40.0 g of dehydrated medium (BK047) in 1 liter of distilled or demineralized water.
- Slowly bring to boiling, stirring with constant agitation until complete dissolution.
- Dispense in tubes or flasks.
- Sterilize in an autoclave at 121°C for 15 minutes.
- Cool and maintain the media in a molten state at 44-47 °C.

✓ **Reconstitution:**
40.0 g/L

✓ **Sterilization:**
15 min at 121 °C

Use of ready-to-melt media:

- Using the ready-to-melt media (BM017, BM049), or if the media was prepared in advance as above, melt the agar for the minimum amount of time necessary to achieve total liquefaction.
- Cool and maintain the media in a molten state at 44-47 °C.

5 INSTRUCTIONS FOR USE

Surface enumeration

✓ **Inoculation:**
0.1 mL on surface

✓ **Incubation:**
30-35 °C

- Pour the molten media held at 44-47 °C into sterile Petri plates.
- Let solidify on a cold, flat surface.
- Dry the plates in an incubator with the covers partially removed.
- To the surface of plates prepared as above or using pre-poured plates (BM050), inoculate 0.1 mL of the test sample and its serial with a sterile triangle or "hockey stock".
- Incubate according to the reference applied:
  - at 30-35 °C for 72 ± 6 hours (NF EN ISO 21149)
  - at 30-35 °C for 48 to 72 hours (NF EN ISO 18415)
  - at 30-35 °C for up to 3 days for bacteria and up to 5 days for yeasts and molds (Pharmacopeias)

NOTE:
The media can also be used after having filtered through a nitrocellulose membrane (Pharmacopeias, NF EN ISO 21149)

Enumeration in pour plates

✓ **Inoculation:**
1 mL in pour plates

✓ **Incubation:**
30-35 °C

- Transfer 1 mL of the initial suspension and its serial dilutions to successive sterile Petri plates.
- Pour roughly 15 mL of molten media per plate.
- Homogenize well and let solidify on a cold, flat surface.
- Incubate according to the reference applied:
  - at 30-35 °C for 72 ± 6 hours (NF EN ISO 21149)
  - at 30-35 °C for 48 to 72 hours (NF EN ISO 18415)
  - at 30-35 °C for up to 3 days for bacteria and up to 5 days for yeasts and molds (Pharmacopeias)

For other uses, refer to the references in vigor.

6 RESULTS

Following incubation, observe bacterial growth.
When performing enumeration, only retain the plates containing between 30 and 300 colonies.

7 QUALITY CONTROL

Dehydrated media: cream-white powder, free-flowing and homogeneous.
Prepared media: amber agar.

Typical culture response after incubation 48 h at 30-35 °C (NF EN ISO 11133):

Microorganisms		Growth (Productivity Ratio: P_R)
Staphylococcus aureus	WDCM 00032	$P_R \geq 50$ %
Pseudomonas aeruginosa	WDCM 00026	$P_R \geq 50$ %
Bacillus cereus	WDCM 00001	$P_R \geq 70$ %
Bacillus subtilis ssp. *spizizenii*	WDCM 00003	$P_R \geq 70$ %
Escherichia coli	WDCM 00012	$P_R \geq 70$ %
Listeria monocytogenes 4 b	WDCM 00021	$P_R \geq 70$ %
Staphylococcus aureus	WDCM 00034	$P_R \geq 70$ %

8 STORAGE / SHELF LIFE

Dehydrated medium: 2-30 °C.
Ready-to-melt media in vials: 2-25 °C
Pre-poured media in Petri plates: 2-8 °C
The expiration dates are indicated on the labels.

Prepared media in tubes or vials (*): 180 days at 2-25 °C.
Prepared media in plates (*): 30 days at 2-8 °C.
(*) Benchmark value determined under standard preparation conditions, following manufacturer's instructions.

9 PACKAGING

Dehydrated medium:
500 g bottle..... BK047HA

Pre-poured media in Petri plates (Ø 90 mm) :
20 plates..... BM05008
120 plates..... BM17808

Ready-to-melt media in vials:
10 x 100 mL vials BM01708
10 x 200 mL vials BM04908

10 BIBLIOGRAPHY

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The Japanese Pharmacopoeia. Chapter 4.05 Microbial Limit Test I. Microbiological examination of non-sterile products: Total viable aerobic count and II. Microbiological examination of non-sterile products: Test for specified products.

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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