

Fluid medium A

Trypto-Casein-Soy broth/TSB

(culture of aerobic bacteria, also suitable for fungi)

Fluid Soybean-Casein Digest medium (III)

355-3454

356-4144

DEFINITION

Medium used for the detection of aerobic-anaerobic bacteria in the analysis of food products, in sterility tests and in the monitoring of non-sterile Pharmacopeia products.

Equivalent USP 30/NF 25: Medium II

STANDARDS

- **European Pharmacopeia 6.0** - Biological methods - **2.6.13.**: Microbiological test of non-sterile products (Detection of specified micro-organisms).
- **USP 30/NF 25 U.S. Pharmacopeia and National Formulary (2007)**: Microbial Limit Tests (**61**) - Microbiological Tests.

PRINCIPLE

The nutrient substances provided by tryptic casein hydrolysate and soy peptone, and the glucose used as an energy source, favor the growth of most aerobic-anaerobic bacteria.

PRESENTATION

- **Ready to use**
10 ml x 25 tubes **code 355-3454**
- **Dehydrated**
500 g **code 356-4144**

TYPICAL FORMULA

Pancreatic casein hydrolysate	17 g
Soy peptone	3 g
Sodium chloride	5 g
Potassium phosphate	2.5 g
Glucose	2.5 g
Distilled water	1,000 ml
Final pH (25°C) = 7.3 ± 0.2	

Remark: the formula has been adapted to attain the required performance criteria.

STORAGE

- Ready to use: + 15° to 25°C
- Dehydrated: + 15° to 25°C, in carefully-sealed bottles in a cool, dry place.
- Expiration date and batch number are shown on the package.

OTHER PRODUCTS REQUIRED (NOT SUPPLIED)

- Diluent(s)
- Distilled water

EQUIPMENT REQUIRED (NOT SUPPLIED) (non-exhaustive)

- Scales
- Sterile weighing bags
- Grinder
- Hotplate
- Mixer-homogenizer
- Test tubes (16 x 160 mm) with autoclave-proof stoppers
- 125 ml Pyrex bottles with autoclave-proof stoppers
- Water-bath precise to ± 1°C
- Thermostatically-controlled incubator or incubation room, precise to ± 1°C
- Autoclave
- All usual laboratory equipment

PRECAUTIONS

- The time lapse between the end of preparation of the stock solution (or the 10⁻¹ dilution in the case of a solid product) and the moment when the dilutions come into contact with the culture medium must not exceed 15 minutes.
- Violent swirling of the regenerated medium should be avoided so as to prevent its re-oxygenation
- Comply with Good Laboratory Practice.

PREPARATION OF DEHYDRATED MEDIUM

Always shake well before use

Dissolve 30 g of powder in 1 liter of distilled water, mix until a homogenous suspension is obtained.

Heat gently, swirling frequently, then bring to boiling point until completely dissolved.

Dispense 10 ml per tube or 100 ml per bottle and sterilize in autoclave at 121°C ± 1°C for 15 minutes.

Reconstitution ratio: 30 g/l.
500 g of powder makes 16.6 liters of medium.

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V3 – 10/04/13

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PROTOCOL

• Preparation of samples

According to standards or recommendations applicable to the product concerned.

• Inoculation and incubation

Inoculate the recommended quantity of product to be analyzed and incubate at 37°C ± 1°C for 14 days.

The addition of agar (0.5-1 g/l) makes it possible to grow anaerobic bacteria such as *Clostridium perfringens* on this medium. In this case the medium must be regenerated prior to use.

This medium can also be used for hemoculture and for the study of the susceptibility of bacteria to antibiotics by the dilutions method.

READING AND INTERPRETATION

Tubes containing microbial turbidity are to be considered positive.

QUALITY CONTROL

STRAINS	Results of culture 5 days at 22.5°C
<i>Aspergillus niger</i> ATCC 16404	Good growth
<i>Candida albicans</i> ATCC 10231	Good growth
<i>Bacillus subtilis</i> ATCC 6633	Good growth

STRAINS	Results of culture 18-24H at 30-35°C
<i>Staphylococcus aureus</i> ATCC 6538	Good growth

Every product manufactured and marketed by Bio-Rad is subject to a quality-assurance procedure at all stages, from the reception of raw materials to the marketing of the end-product. Each batch of finished product undergoes quality control and is marketed only if it satisfies the acceptability criteria.

Documentation relative to the production and control of each batch is kept on file.

KEY WORDS

Trypto-Casein-Soy/Aerobic-anaerobic bacteria/
Food products/Sterility tests/Growth/ Medium