

INSTRUCTIONS FOR USE – READY-TO-USE BOTTLED MEDIA

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Rev.: Febr. 2008

BA-257107.03

BD™ Tryptic Soy Broth (TSB)

INTENDED USE

BD Tryptic Soy Broth (Soybean-Casein Digest Medium) is a general purpose liquid enrichment medium used in qualitative procedures for the sterility test and for the enrichment and cultivation of aerobic microorganisms that are not excessively fastidious. In clinical microbiology, it may be used for the suspension, enrichment and cultivation of strains isolated on other media.

PRINCIPLES AND EXPLANATION OF THE PROCEDURE

Microbiological method.

Tryptic Soy Broth (TSB) is a nutritious medium that will support the growth of a wide variety of microorganisms, especially common aerobic and facultatively anaerobic bacteria. Because of its capacity for growth promotion, this formulation was adopted by The United States Pharmacopeia (USP) and the European Pharmacopeia (EP) as a sterility test medium. 44

In clinical microbiology, the medium is used in a variety of procedures, e.g., for the preparation of the inoculum and for suspending strains for Kirby-Bauer disc diffusion susceptibility testing, and for the microbiological test procedure of culture media according to the NCCLS standards.^{5,6} However, unsupplemented Tryptic Soy Broth is not recommended as a primary enrichment medium directly inoculated with the clinical specimen.

In **BD Tryptic Soy Broth**, enzymatic digests of casein and soybean meal provide amino acids and other complex nitrogenous substances. Glucose (=dextrose) is an energy source. Sodium chloride maintains the osmotic equilibrium. Dibasic potassium phosphate acts as a buffer to control pH.

REAGENTS

BD Tryptic Soy Broth

Formula** Per Liter Purified Water

J	17.0 g
Bacto Soytone (Peptic Digest of Soybean Meal)	3.0
Glucose (=Dextrose)	2.5
Sodium Chloride	5.0
Dipotassium Hydrogen Phosphate	2.5

pH 7.3 ± 0.2

PRECAUTIONS

IVD . For professional use only.

Do not use containers if they show evidence of microbial contamination, discoloration, drying, cracking or other signs of deterioration.

Consult **GENERAL INSTRUCTIONS FOR USE** document for aseptic handling procedures, biohazards, and disposal of used product.

STORAGE AND SHELF LIFE

On receipt, store vials in the dark at 5 to 25° C until just prior to use. Avoid freezing and overheating. The vials may be inoculated up to the expiration date (see container or package label) and incubated for the recommended incubation times.

Vials from opened packages can be used up to the expiration date. Opened vials must be used immediately.

^{**}Adjusted and/or supplemented as required to meet performance criteria.

USER QUALITY CONTROL

Inoculate representative samples with the following strains (for details, see **GENERAL INSTRUCTIONS FOR USE** document). According to the European Pharmacopeia, incubate the containers inoculated with bacterial strains at 30 to 35°C for a maximum of 3 days and at 22.5 ± 2.5° C for the fungi, for a maximum of 5 days, in normal air. To provide sufficient aeration, containers should be vented during incubation (see **Test Procedure**).

Test Strain	Expected Growth Results (=Turbidity)*
Staphylococcus aureus ATCC™ 6538	+++ or higher
Bacillus subtilis ATCC 6633	+++ or higher
Pseudomonas aeruginosa ATCC 9027	+++ or higher
Candida albicans ATCC 10231	+++ or higher
Aspergillus niger ATCC 16404	+++** or higher
Uninoculated	Light to medium amber, clear, no precipitates

^{* ++++ =} opaque, heavy +++ = opalescent, heavy ++ = opalescent + = slight haze

PROCEDURE

Materials Provided

BD Tryptic Soy Broth (TSB), provided in vials (see PACKAGING/AVAILABILITY for details).

STERILE |

Materials Not Provided

Ancillary culture media, reagents and laboratory equipment as required.

Specimen Types

This medium is not intended to be used for inoculation with clinical specimens. It must only be used in selected procedures (see PRINCIPLES AND EXPLANATION OF THE PROCEDURE and PERFORMANCE CHARACTERISTICS AND LIMITATIONS OF THE PROCEDURE). Consult the references for the applications in industrial microbiology.^{1,3,4}

Test Procedure

For application in clinical microbiology, inoculate the medium with the strain and incubate as required. Note that specimens should also be inoculated directly onto solid media, such as **BD Columbia Agar with 5% Sheep Blood** or **BD Trypticase Soy Agar II with 5% Sheep Blood** and, eventually, on additional selective and nonselective media.

Usually, an incubation temperature of $35 \pm 2^{\circ}$ C is adequate. Incubate for 18 to 24 h or longer if required. For use as a suspension medium, inoculate the tube with a small amount of growth from an overnight culture on a solid medium.

For use in industrial microbiology, inoculate the sample or material to be tested into the medium. See the references for details.^{3,4} According to the European Pharmacopeia, incubate aerobically at 32.5 ± 2.5 °C for a maximum of 3 days (for the bacteria) and at 22.5 ± 2.5 °C for a maximum of 5 days (for the fungi).

For use in sterility testing, consult the USP or EP for procedural details and specifications for volume of medium relative to container size.^{3,4}

For all applications, it is important to provide sufficient aeration during incubation. Therefore, containers with this medium should be vented. Depending on the type of closure of the container, this can be achieved by slightly loosening the caps or by introducing a sterile injection needle plugged with sterile cotton wool into the septum of the cap. Alternatively, injection needles fitted with a membrane filter can be used.

Results

Growth in broth media is indicated by the presence of turbidity, specks, or flocculation in the medium while an uninoculated control remains clear and without turbidity after incubation. If the material tested causes turbidity of the medium, subcultures onto appropriate solid media must

^{**}Aspergillus niger and other filamentous fungi may produce a mycelium on top of the broth or specks in the broth rather than a homogenous turbidity.

be performed after incubation to decide if the turbidity is caused by the material only or by microorganisms that have multiplied in the broth.

Subcultures onto suitable solid media and biochemical and microscopic tests are necessary to determine the purity of the culture and for the identification of the isolated organisms. When used for the isolation of pathogens from clinical specimens, subculture a 10 to 50 μ l portion of the medium onto **BD Columbia Agar with 5% Sheep Blood** or **BD Trypticase Soy Agar II with 5% Sheep Blood**. For details, consult the references.^{5,6}

PERFORMANCE CHARACTERISTICS AND LIMITATIONS OF THE PROCEDURE

BD Tryptic Soy Broth is a universal enrichment and isolation medium for many nonclinical procedures. ^{1,3,4} In clinical microbiology, it is mainly used for suspending cultures for susceptibility tests and for the preparation of inocula in quality control test procedures. ^{5,6}

Growth obtained in this medium must be subcultured onto appropriate solid media to obtain pure cultures which afterwards can be identified with methods appropriate for the isolates.

Tryptic Soy Broth is not the appropriate medium for the cultivation of fastidious mircoorganisms (e.g., *Haemophilus* or *Neisseria* spp.) and for the detection and recovery of strict anaerobes. Fluid Thioglycollate Media should be used for the cultivation of strict anaerobes.

REFERENCES

- 1. Marshall, R.T. (ed.). 1993. Standard methods for the examination of dairy products, 16th ed. American Public Health Association, Washington, D.C.
- 2. MacFaddin, J.F. 1985. Media for the isolation cultivation maintenance of medical bacteria. Volume 1. Williams and Wilkins, Baltimore, London
- 3. U.S. Pharmacopeial Convention, Inc. 1999. The U.S. Pharmacopeia 24/The national formulary NF 19--2000. U.S. Pharmacopeial Convention, Inc., Rockville, Md
- 4. Council of Europe, 2002. European Pharmacopoeia, 4th edition. European Pharmacopoeia Secretariat. Strasbourg/France.
- 5. National Committee for Clinical Laboratory Standards. 2000. Approved standard: M2-A7. Performance standards for antimicrobial disk susceptibility tests, 7th ed. National Committee for Clinical Laboratory Standards, Wayne, Pa.
- National Committee for Clinical Laboratory Standards. 1996. M22-A2. Quality assurance for commercially prepared microbiological culture media – second edition; approved standard. NCCLS. Wayne, PA, USA.

PACKAGING/AVAILABILITY

BD Tryptic Soy Broth (TSB)

Cat. No. 257107 Ready-to-use Bottled Medium cpu 50; 20 ml in a 30 ml screw cap vial cpu 50; 9 ml in a 30 ml screw cap vial cpu 50; 9 ml in a 30 ml screw cap vial

FURTHER INFORMATION

For further information please contact your local BD representative.



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