

PRODUCT INFORMATION

Product Type: Divided Petri Dishes 90mm (DD)

Cat No. DD004 - BLOOD AGAR (TSBA) / CHOCOL. BL. AGAR + SUPP

Intended Use:

Blood Agar (TSBA):

Tryptic Soy Blood Agar Base No. 2 with 100 mL/L donor sheep blood is a robust, nutrient-rich medium for growing and differentiating fastidious bacteria, especially for observing hemolysis patterns.

Chocolate Blood Agar + Supplements:

Chocolate Blood Agar with sheep blood is used for the isolation and cultivation of fastidious pathogens, particularly *Neisseria* and *Haemophilus* species, in clinical microbiology.

Principle and Uses:

Blood Agar (TSBA):

Tryptic Soy Blood Agar Base No. 2 (often abbreviated as TSBA or TSA II) with sheep blood is a microbiological medium used for the cultivation, isolation, and identification of a wide range of microorganisms, particularly bacteria. For the isolation and cultivation of fastidious bacteria (e.g., *Streptococcus*, *Staphylococcus*, *Haemophilus*) in Food, clinical, and environmental microbiology, and for general and specific pathogen detection.

The addition of sheep blood to the Tryptic Soy Agar Base enhances the medium by providing additional nutrients and allowing for the observation of hemolytic reactions.

Hemolysis Patterns:

Beta-hemolysis: Complete hemolysis, resulting in a clear zone around bacterial colonies.

Alpha-hemolysis: Incomplete hemolysis, leading to a greenish discoloration around colonies.

Gamma-hemolysis: No hemolysis, with no change in the appearance of the agar around colonies.

Certifications / Compliance: AOAC, BAM, CCAM, COMPF, EP, EPA, ISO, JP, SMD, SMWW, USDA, USP

The nutrient-rich base supports the growth of a wide range of organisms. The addition of blood provides additional growth factors and allows for the detection of hemolytic activity, which is useful for differentiating bacterial species.

See also **PD049 - TRYPTIC SOY AGAR + DEF. SHEEP BLOOD**

Chocolate Blood Agar + Supplements:

For the non-selective cultivation of fastidious bacteria. Especially useful for the cultivation of *Haemophilus* and *Neisseria* in a CO₂ enriched atmosphere.

Contains an improved GC Agar base, 8.5% blood and Enrichment. The GC base contains nitrogenous nutrients in the form of casein and meat peptones, phosphate buffer to maintain pH and corn starch, which neutralizes toxic fatty acids that may be present in the agar. Sheep blood is lysed (chocolated) to release necessary growth factors. Blood is heated (chocolated) after addition to lyse red cells, releasing NAD (factor V) and hemin (factor X), making these nutrients available for fastidious organisms. Enrichment Supplement is critical for optimal growth and recovery of *Neisseria* and *Haemophilus* spp. Enrichment is a defined supplement which provides V factor (nicotinamide adenine dinucleotide, NAD) for *Haemophilus* species and vitamins, amino acids, co-enzymes, dextrose, ferric ion and other factors which improve the growth of pathogenic *Neisseria*.

Limitations

1. It is recommended that biochemical, immunological, molecular, or mass spectrometry testing be performed on colonies from pure culture for complete identification.
2. Chocolate Agar is an enriched medium, thus non-pathogenic organisms may overgrow pathogenic bacteria
3. The presence or absence of *N. gonorrhoeae* in a specimen does not rule out the possible presence of other pathogenic organisms.

Composition

Blood Agar (TSBA):

Tryptone H - 15 g/L,
Soytone - 5 g/L,
Sodium Chloride - 5 g/L,
Agar - 15 g/L
Donor Sheep Blood – 50 ml/L

Chocolate Blood Agar + Supplements:

Casein Peptone - 7.5 g/L
Corn Starch - 1.0 g/L
Meat Peptone - 7.5 g/L
Monopotassium Phosphate - 1.0 g/L
Sodium Chloride - 5.0 g/L
Agar - 10.0 g/L
Dipotassium Phosphate - 4.0 g/L
Donor Sheep Blood – 93.75 ml/L

Supplements:

Glucose - 2.0 g/L
Vitamin B12 - 0.2 mg/L
Adenine - 20.0 mg/L
L-Glutamine - 200.0 mg/L
Guanine - 0.6 mg/L
p-Aminobenzoic acid - 0.26 mg/L
L-Cystine - 22.0 mg/L
NAD (Coenzyme 1) - 5.0 mg/L
Coccarboxylase - 2.0 mg/L
Iron (III) nitrate - 0.4 mg/L
Thiamine hydrochloride - 0.6 mg/L
Cysteine hydrochloride - 518.0 mg/L

Storage: 2-8°C

Appearance:

Blood Agar (TSBA): Opaque, bright red agar.

Chocolate Blood Agar + Supplements: chocolate - brown

pH Range:

Blood Agar (TSBA): 7.2 - 7.6

Chocolate Blood Agar + Supplements: 7.2 - 7.6

Package contents: 10 plates in a package

Exp. Date: Printed on label and on the item.

Required materials not supplied: Laboratory equipment as required.

Warning and Precautions - For professional use only. Follow good microbiological lab practices while handling specimens and culture. Do not use Petri dishes if they show evidence of microbial contamination, discoloration, drying, cracking, or other signs of deterioration. Avoid freezing and overheating. The Petri Dishes may be used / inoculated up to the expiration date and incubated for the recommended incubation times. After use and prior to discarding, specimen containers and all contaminated material, including the used culture media and contaminated culture containers, must be sterilized or incinerated by validated procedures. Since the nutritional requirements of organisms vary, some strains may be encountered that fail to grow or grow poorly on this medium.

If excessive moisture is observed, invert the bottom over an off-set lid and allow to air dry in order to prevent formation of a seal between the top and bottom of the plate during incubation. Storage Instructions: On receipt, store plates in the dark at 2–8 °C. Avoid freezing and overheating. Do not open until ready to use.

Waste Disposal

After interpretation all plates should be destroyed by standard incineration methods.

Performance Testing Results:

GPT: inoculum 10-100 cfu.

TEST	ATCC	Incubation Temp. (°C)	Incubation Cond.	Reaction 1 TSA+blood		Reaction 2 CHOCO	
<i>Staphylococcus aureus</i>	25923	33-37 °C	5% CO ₂ , 24-48 hours	Growth	Beta hemolytic reaction	/	
<i>Streptococcus pyogenes</i>	19615	33-37 °C	5% CO ₂ , 24-48 hours	Growth	Beta hemolytic reaction	/	
<i>Streptococcus pneumoniae</i>	49619	33-37 °C	5% CO ₂ , 24-48 hours	Growth	Alpha hemolytic reaction	Growth	Yellow capsulated
<i>Neisseria gonorrhoeae</i>	49226	33-37 °C	5% CO ₂ , 24-48 hours	/		Growth	
<i>Neisseria meningitidis</i>	13090	33-37 °C	5% CO ₂ , 24-48 hours	/		Growth	
<i>Haemophilus influenzae</i>	49766	33-37 °C	5% CO ₂ , 24-48 hours	/		Growth	
<i>Haemophilus influenzae</i>	49247	33-37 °C	5% CO ₂ , 24-48 hours	/		Growth	