

BLOOD AGAR BASE

Medium with the addition of blood used for the isolation, cultivation and of the hemolytic activity of fastidious microorganisms

TYPICAL FORMULA (g/L)

Beef Extract.....	10.0
Tryptose.....	10.0
Sodium Chloride.....	5.0
Agar.....	14.0
Final pH	7.4 ± 0.2

DESCRIPTION

BLOOD AGAR BASE is a general purpose agar base with, with the addition of 5% sterile blood, is used to cultivate a wide range of micro-organisms of clinical significance. It is specially designed to detect beta hemolytic reactions of streptococci in Lancefield in groups A and B, and to favour the growth of particularly fastidious aerobic and anaerobic bacteria. It is also used in the Camp Test.

PRINCIPLE

Peptones provides nitrogen, carbon, aminoacids and vitamins. Sodium chloride maintains the osmotic balance of the medium. The addition of SHEEP BLOOD DEFIBRINATED (code 83296) supplies further growth factors for fastidious microorganisms and allows to show hemolytic reactions. Agar is the solidifying agent.

PREPARATION

Melt the content of one bottle in a boiling water-bath at 100°C (loosing the caps partially unscrewed) until completely dissolved. After molten, verify the proper homogeneity of medium, turning upside down the bottle after screwing cap. Cool down to 45-50°C and aseptically add SHEEP BLOOD DEFIBRINATED (code 83296) at the concentration of 5%. Mix well avoiding the formation of bubbles and aseptically distribute into Petri dishes. Allow the medium to solidify. Store the plates in tightly closed containers.

TECHNIQUE

Process each specimen as appropriate, and inoculate directly onto the surface of the surface of the medium. Streak for isolation with an inoculating loop, then stab the agar several times to deposit beta-hemolytic streptococci beneath the agar surface. Subsurface growth will display the most reliable hemolytic reactions demonstrating both oxygen-stable and oxygen-labile streptolysins (1). Incubate plates aerobically, anaerobically or under conditions of increased CO₂ in accordance with established laboratory procedures.

INTERPRETATION OF RESULTS

Examine plates for growth and hemolytic reactions after 18-24 and 40-48 hours of incubation. Four different types of hemolysis on blood agar media can be described (2):

- Alpha (α)-hemolysis is the reduction of hemoglobin to methemoglobin in the medium surrounding the colony, causing a greenish discolorization of the medium.
- Beta (β)-hemolysis is the lysis of red blood cells, resulting in a clear zone surrounding the colony.
- Gamma (γ)- hemolysis indicates no hemolysis. No destruction of red blood cells occurs, and there is no change in the medium.
- Alpha-prime (α')-hemolysis is a small zone of complete hemolysis that is surrounded by an area of partial lysis.

STORAGE

10-25°C away from light, until the expiry date indicated on the label or until signs of deterioration or contamination are evident.

WARNING and PRECAUTIONS

The product is not classified as hazardous by current legislation and does not contain harmful substances in concentrations of ≥1%. The product is designed for *In vitro* diagnostic use and must be used only by properly trained operators.

DISPOSAL of WASTE

Disposal of waste must be carried out according to national and local regulations in force.

REFERENCES

- Ruoff, Wiley and Beighton. (1999). In Murray, Baron, Pfaller, Tenover and Tenover (ed.), Manual of clinical microbiology, 7th ed. American Society for Microbiology, Washington, D.C.
- Isenberg (ed). 1992. Clinical microbiology procedures handbook, vol. 1. American Society for Microbiology, Washington, D.C.
- Brown, J.H.. 1919. The use of blood agar for the study of streptococci, NY Monograph No. 9. In Rockefeller Institute for Medical Research
- NCCLS document M22-A2, 1996. Approved Standard.



LIOFILCHEM Bacteriology Products

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PRODUCT SPECIFICATIONS

NAME

BLOOD AGAR BASE

PRESENTATION

Code 402140

Glass bottles containing 200 ml of medium.

Code 470130

Glass bottles containing 500 ml of medium.

STORAGE

10-25°C

PACKAGING

Code	Content	Packaging
402140	6 bottles x 100 ml	6 bottles in box
470130	6 bottles x 500 ml	6 bottles in box

pH OF THE MEDIUM

7.4 ± 0.2

USE

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TECHNIQUE

Refer to technical sheet of the product.

APPEARANCE of the MEDIUM

Amber medium, opalescent.

SHELF LIFE

2 years

QUALITY CONTROL

- Control of general characteristics, label and print
- Sterility control
7 days at 25 ± 1°C, in aerobiosis
7 days at 36 ± 1°C, in aerobiosis
- Microbiological control
Inoculum for productivity: 10-100 CFU/ml
Inoculum for specificity: ≤ 10⁴ CFU/ml
Incubation conditions: 18-48 hours at 35 ± 2°C, with added CO₂.

Microorganisms		Inoculum CFU	Recovery with Sheep Blood
<i>Haemophilus influenzae</i>	ATCC 19418	10 ² -3x10 ²	N/A
<i>Neisseria meningitidis</i>	ATCC 13090	10 ² -3x10 ²	Good, gamma hemolysis
<i>Streptococcus pneumoniae</i>	ATCC 6305	10 ² -3x10 ²	Good, alpha hemolysis
<i>Streptococcus pyogenes</i>	ATCC 19615	10 ² -3x10 ²	Good, beta hemolysis

TABLE of SYMBOLS

Symbol	Meanings
	In vitro Diagnostic Medical Device
	Manufacturer
	Catalogue number
	Use by
	Temperature limitation
	Do not reuse
	Contains sufficient for <n> tests
	Consult accompanying documents
	Batch code
	Fragile, handle with care



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