SHEEP BLOOD AGAR

- For in vitro use only - Catalogue No. PS58

Our Sheep Blood Agar is a highly nutritious medium used for the cultivation and isolation of a variety of microorganisms.

Our Sheep Blood Agar is based on the Oxoid formulation; the prepared medium is said to offer improved nutritional value resulting in better growth and larger colony size as well giving more consistent hemolytic reactions especially among the streptococci. The base is specifically designed for use with sheep blood as horse blood has shown to give different and conflicting hemolytic reactions when incorporated into other blood agars.

The nutritional components include pancreatic digest of casein, neutralized peptone, and yeast extract, and the addition of sodium chloride provides an osmotically balanced medium for bacterial cells. The addition of 5% defibrinated sheep blood allows for the determination of hemolytic reactions, an important differential characteristic.

Formula per Litre of Medium

Pancreatic Digest of Casein	14.0 g
Neutralized Peptone	4.5 g
Yeast Extract	4.5 g
Sodium Chloride	6.0 g
Agar	12.5 g
Defibrinated Sheep Blood	50.0 mL

pH 7.3 ± 0.2

Recommended Procedure

1. Allow medium to reach room temperature.
2. Using an inoculum from the specimen, perform a four-quadrant streak to obtain well-isolated colonies.
3. Incubate in an aerobic or CO_2-supplemented atmosphere at 35°C.
4. Examine after 18-24 hours.

Interpretation of Results

Sheep Blood Agar can be used as a primary-plating medium. Primary isolation is performed to separate and isolate organisms present in a sample. This separation allows for characterization of colony types and may indicate the presence of clinically significant bacteria. When examining plates a hand lens or stereoscopic microscope should be available for examining very small colonies. The different types of colonial morphology appearing on the agar plate should be noted as well as the number of each morphotype present. Hemolysis is also a very useful differential characteristic that is best viewed when a bright light is transmitted from behind the plate. Four different types of hemolysis can be described:

1. Alpha-hemolysis (α) – Partial hemolysis that results in a greenish discoloration around the colony
2. Beta-hemolysis (β) – Complete lysis of red blood cells resulting in a clear zone around the colony
3. Gamma-hemolysis (γ) – No hemolysis resulting in no change in the medium
4. Alpha-prime-hemolysis (α′) – A small zone of complete hydrolysis that is surrounded by an area of partial hemolysis

Additional tests should be performed on isolated colonies from pure culture in order to complete identification.

Quality Control

After checking for correct pH, colour, depth, and sterility, the following organisms are used to determine the growth performance of the completed medium.

Organism	Expected Result
Streptococcus pneumoniae ATCC 6305	Growth, α-hemolysis
Streptococcus pyogenes ATCC 19615	Growth, β-hemolysis
Escherichia coli ATCC 25922	Growth, γ-hemolysis
Staphylococcus aureus ATCC 25923	Growth, γ-hemolysis

Storage and Shelf Life

Our Sheep Blood Agar should be stored away from direct light at 4°C to 8°C. The medium side should be uppermost to prevent excessive accumulation of moisture on the agar surface. Under these conditions this medium has a shelf life of 12 weeks from the date of manufacture.

References

1. MacFaddin JF. Media for isolation-cultivation-maintenance of medical bacteria, Vol I. Baltimore: Williams & Wilkins, 1985.
2. The Oxoid Manual. 6th ed. Basingstoke, UK: Unipath, 1990.
3. Isenberg HD, Ed. Clinical microbiology procedures handbook. Washington, DC: ASM, 1992.
4. NCCLS. Quality assurance of commercially prepared microbiological culture media. 2nd ed. NCCLS document M22-A2. Wayne, PA: NCCLS, 1996.
5. Forbes BA, Sahm DF, Weissfeld AS. Bailey and Scott's diagnostic microbiology. 10th ed. St. Louis: Mosby, 1998.

Original: February 2003
Revised / Reviewed: October 2014