Colorcult®

Intended Use

Colorcult® culture media is used in qualitative procedures for the culture and recovery of microorganisms (bacteria and yeast) from blood.

Summary

Colorcult® culture vials comprise of highly nutritious media intended for the growth of significant pathogenic microorganisms including fastidious microorganisms present in blood. The samples to be tested are inoculated in Colorcult® culture vials. These vials are kept in incubator and monitored periodically for change in color of the chemical sensor for positive reporting.

Principle

Colorcult® culture media contains various types of proteins and other nutrients necessary to support the growth of fastidious and non-fastidious microorganisms present in blood. Sodium Polyanethol Sulfonate (SPS) is a polyanionic anticoagulant, which inhibits complements and lysozyme activity, interferes with phagocytosis, and inactivates many antibiotics. It is generally considered to enhance the rate and speed of bacterial isolation by counter-acting the bacterial inhibitors of human blood. After thorough and gentle mixing, the specimen is added to the Colorcult® culture vial. The vial is incubated and observed for color change of chemical sensor as evidence of microbial growth.

Each vial contains a chemical sensor at the bottom which can detect an increase in CO_2 produced by the growth of microorganisms. The sensor is monitored visually or by the instrument for color change which is proportional to the amount of CO_2 present. A positive color change indicates the presumptive presence of viable microorganisms in the vial.

Resins have been incorporated in the Colorcult® culture vials to enhance recovery of organisms without a need for special processing.

Formula*

Colorcult® culture media is formulated as:

Soybean Casein Digest Broth	3.0%
Yeast Extract	0.4%
Amino Acids	0.05%
Sugar	0.5%
Vitamins	0.025%
Antioxidants/ Reductants	0.005%
Sodium Polyanethol Sulfonate (SPS)	0.05%
Resin Mix	15.0%

*Adjusted to suit performance parameters.

pH:6.80-7.20

Additional Materials Required

Needle and syringe for blood collection, cotton or gauze, isopropyl alcohol (70%), incubator 35°C-37°C, sterile venting units.

Specimen Collection and Preparation

1. The specimen must be collected using sterile techniques to reduce the chances of contamination.
2. Collect approximately 8-10 mL of blood from adult patients and approximately 1-3 mL from paediatric patients.
3. Sample volumes as low as 3 mL can be used, however, recovery will not be as great as with larger volumes.

4. The inoculated Colorcult® culture vials should be transported to the laboratory as quickly as possible.
5. Samples should be stored at 2°C-8°C if not tested immediately.
6. Avoid using haemolyzed samples for testing.

Directions

Inoculation of the Colorcult® culture vials:

1. Bring the appropriate number of Colorcult® culture vials to R.T. (22°C-30°C) before testing.
2. Check the bottom of each vial, it should be purple in color. If the color of the chemical sensor is yellow, discard those vials.

Note: Any colour change other than yellow of the chemical sensor shouldn't be interpreted as contamination or a positive result. This may occur because of the media and resins' interaction with the chemical sensor.

3. Label the vials with the appropriate patient I.D. / Name / Time of sample collection and other relevant information.
4. Remove the dust cap from Colorcult® culture vials; swab the septum with isopropyl alcohol (70%) before inoculating.
5. Withdraw blood from the patient using a sterile needle and syringe.
6. Transfer the sample withdrawn immediately into the Colorcult® culture vials under aseptic conditions.
7. Incubate the vials at 35°C-37°C for 5-7 days for bacteria and 7-14 days for yeast and fungi or until growth appears before reporting negative.
8. Lower temperatures may be preferable for detection of fungi, *Listeria,* and some other pathogens.
9. Observe the bottles at an interval of 24 hours for changes in the color of the chemical sensor.
10. Positive Colorcult® culture vials should be subcultured on solid media or Gram-stained.

Quality Control

Media Appearance: Light amber colored, clear solution without any precipitate.
Appearance on Addition of Blood: Cherry red colored opaque solution without any blood clot.
Resin Mix Appearance: Colorless and Brown colored resin mix.
Bottom Dye Appearance: Purple
Cultural Response: Cultural response is observed after an incubation of 18-24 hours within 5 days at 35°C-37°C and sub-culturing is carried out on Blood Agar.

For Aerobic and Paediatric Testing

Organism (ATCC)	Growth	Subculturing on Blood Agar
Staphylococcus aureus subsp. *aureus* (25923)	Good	Beta haemolysis
Streptococcus pyogenes Strain Bruno (19615)	Good	Beta haemolysis
Streptococcus pneumoniae (6305)	Good	Alpha haemolysis
Escherichia coli (25922)	Good	-
Pseudomonas aeruginosa Strain Boston 41501 (27853)	Good	Beta haemolysis
Salmonella enterica subsp. *enterica* serovar *Typhimurium* (23564)	Good	-
Salmonella enterica subsp. *enterica* serovar *Typhimurium* (14028)	Good	-
Neisseria meningitidis (13090)	Good	-
Haemophilus influenzae (19418)	Fair	-
Candida glabrata (66032)	Good	-
Alcaligenes faecalis (8750)	Good	Alpha haemolysis

For Anaerobic Testing

Escherichia coli (25922)	Good	-
Staphylococcus aureus subsp. *aureus* (25923)	Good	Beta haemolysis
Streptococcus pneumoniae (6305)	Good	Alpha haemolysis
Clostridium perfringens (13124)	Good	Beta haemolysis
Clostridium sporogenes (19404)	Good	Beta haemolysis
Bacteroides fragilis (25285)	Good	Non-haemolytic
Bacteroides vulgatus (8482)	Good	Non-haemolytic

Note:

1. Formula adjusted (w/v) to suit performance standards.
2. Inoculum cfu for good growth is 10-100.

Interpretation of Results

Growth in the Colorcult® culture vials is indicated by the color change of the chemical sensor at the bottom from purple to bright yellow. For vials with no growth show no color change of the chemical sensor. They remain purple in color. The vials should be observed for 5-7 days for positive color change for bacteria, for yeast and fungi 14 days before reporting it negative.

Warning and Precautions

1. Colorcult® culture vials are for laboratory and professional use only.
2. Do not use vials that have cracks or defects.
3. Do not use vials that have contamination.
4. Do not use vials where the color of chemical sensor is already yellow before sample inoculation.
5. The anticoagulant SPS exhibits less toxicity and enables bacteria to survive for a much longer period.
6. Inoculated vials should be decontaminated prior to discarding. Clinical samples and microbial cultures should be considered as pathogenic biohazard and handled accordingly. Good laboratory practices and hazard precautions must always be observed.
7. Ensure that the blood is dispersed evenly throughout the medium.
8. If growth is detected in either aerobic or anaerobic vial, a Gram-stained smear should be prepared, and appropriate subculture methods and biochemical identification tests be carried for further identification.
9. Blood drawn for culture must not be allowed to clot. If bacteria are entrapped within a clot, their presence may go undetected.

Limitations

1. Various factors like antimicrobial therapy prior to sample collection; transitory bacteremia or contamination of the patient's blood/other body fluid by exogenous flora and volume of blood drawn may affect the recovery of clinically significant microorganisms. Ideally, blood cultures should be done before initiation of antimicrobial therapy.
2. SPS inhibits the growth of certain strains of *Neisseria* species, *Gardnerellla vaginalis*, *Streptobacillus moniliformis* and all strains of *Peptostreptococcus anaerobius*. Since blood can neutralize the toxicity of SPS towards organisms sensitive to SPS, the presence of maximum volumes of blood (8-10 mL) can help to optimize recovery of these organisms.
3. Premature discarding of apparently negative blood cultures or infrequent observations may result in failure to detect the presence of pathogenic microorganisms or in loss of viability.
4. A 14-day incubation period is adequate for the recovery of yeast. However, if dimorphic fungi are suspected to be present, blood cultures may require incubation for an additional two weeks.
5. False negative readings may result when certain organisms are present which do not produce enough CO_2 to be detected by the system.

6. False positive results may occur when white blood cell count is high.

Storage and Stability

1. Store the blood culture media in cool, dry place at 15°C-25°C away from direct light.
2. Stability of the blood culture media is as per the expiry date mentioned on the label.

Warranty

This product is designed to perform as described on the label and package insert. The manufacturer disclaims any implied warranty of use and sale for any other purpose.

References

1. Clinical Diagnosis & Management by Laboratory Methods, Todd, Sanford and Davidsohn, 17th edition 1998, Edited by John Bernard Henry.
2. Diagnostic Microbiology, Bailey & Scott, 9th Edition,Ellen Jo Baron, *et al.,* Mosby 1994.
3. Blood Cultures: Where do we stand?, Laboratory Techniques, R.C. Spencer, J. Clin. Pathol 1988; 41:668-670.
4. Effect of Sodium Polyanethol Sulfonate in Blood culture, Jan Eng, Journal of Clinical Microbiology, Feb. 1975, Vol. 1, No. 2, p:119-123.
5. Effect of Volume of Blood Cultured on Detection of Bacteremia, M. Marsha Hall, *et al.,* Journal of Clinical Microbiology, June 1976, Vol. 3, No. 6, p:643-645.
6. Evaluation of the Necessity for Routine Terminal Subcultures of Previously Negative Blood Cultures, Jolynne Campbell, *et al.,* Journal of Clinical Microbiology, Oct. 1990, Vol. 12, No. 4, p: 576-578.
7. Effect of Aerobic and Anaerobic Atmospheres on Isolation of Organisms from Blood Cultures, Donna J. Blazevic, *et al.,* Journal of Clinical Microbiology, Feb. 1975, Vol. 1, No. 2, p:154-156.
8. Effects of Volume and Periodicity on Blood Cultures, James Li, *et al.,* Journal of Clinical Microbiology, Nov. 1994, Vol. 32, No. 11, p: 2829-2831.
9. Data on file: Microxpress®, A Division of Tulip Diagnostics (P) Ltd.

Product Presentation:

Cat No.	Product description	Pack Size
203031220030	Colorcult® Aerobic Culture Vial	25 x 30 mL
203031060030	Colorcult® Anaerobic Culture Vial	1 x 30 mL
203031240030	Colorcult® Paediatric Culture Vial	25 x 40 mL