BIOMARK Laboratories-INDIA

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TECHNICAL SHEET

B994	**COLUMBIA BLOOD AGAR BASE**

Formula

Ingredients :	**gms/lit.**
Peptone, special	23.00
Corn starch	1.00
Sodium chloride	5.00
Agar	15.00

Final pH (at 25°C) : 7.3 ± 0.2

Directions :

Suspend 44 gms in 1000 ml. distilled water. Boil to dissolve the medium completely. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes Cool to 45-50°C before adding heat sensitive compounds.

For Blood Agar: Add 5% v/v sterile defibrinated sheep blood to sterile cool base.

For Chocolate Agar: Add 10% v/v sterile defibrinated sheep blood to sterile cool base. Heat to 80°C for 10 minutes with constant agitation.

The medium can be made selective by adding different antimicrobials to sterile base.

For Brucella species: Add rehydrated contents of 1 vial of Brucella Selective Supplement, Modified (BF012) to 500 ml sterile molten base.

For Campylobacter species: Add rehydrated contents of 1 vial of Campylobacter Supplement- I (Blaser-Wang) (BF013) or Campylobacter Supplement- II, Modified (BF014) or Campylobacter Supplement- III (Skirrow) (BF015) or Campylobacter Selective Supplement (BF041) or Campylobacter Supplement- VI (Butzler) (BF042) to 500 ml sterile molten base along with rehydrated contents of 1 vial of Campylobacter Growth Supplement (BF016) and 5-7% v/v horse or sheep blood.

For Gardnerella species: Add rehydrated contents of 1 vial of G.Vaginalis Selective Supplement (BF040) to 500 ml sterile molten base.

For Cocci: Add rehydrated contents of 1 vial of Staph-Strepto Supplement (BF148) or Strepto Supplement (BF017) or Streptococcus Selective Supplement (BF043) to 500 ml sterile molten base.

Principle:

Columbia Blood Agar Base uses specially selected raw materials to support good growth of fastidious microorganisms. Peptone provides nitrogen, carbon, amino acids and vitamins. Corn starch, increases growth of Neisseria and enhances the hemolytic reactions of some streptococci. Agar is a solidifying agent. Sodium Chloride maintains the osmotic balance of the medium.

Blood agar bases are relatively free of reducing sugars, which have been reported to adversely influence the hemolytic reactions of B-hemolytic streptococci. Supplementation with blood (5-10%) provides additional growth factors for fastidious microorganisms and aids in determining hemolytic reactions. Hemolytic patterns may vary with the source of animal blood and the type of basal medium used.

QC Tests – (I)Dehydrated Medium	
Colour :	Cream to light yellow
Appearance :	Homogeneous Free Flowing powder
(II)Rehydrated medium	
pH (post autoclaving/heating) :	7.3 ± 0.2
Colour (post autoclaving/heating) :	A) Basal medium : light yellow to light amber B) (After addition of 5% sterile defibrinated blood): Cherry red
Clarity (post autoclaving/heating) :	A) Clear to slightly opalescent gel B) Opaque
(III)Q.C. Test Microbiological	
Cultural characteristics observed after 24-48 hours at 35-37°C with added 5% w/v sterile defibrinated blood. Clostridium species incubated under anaerobic conditions	

MICROORGANISM (ATCC)	GROWTH w/5% BLOOD	HAEMOLYSIS
Neisseria meningitidis (13090)	Luxuriant	None
Staphylococcus aureus (25923)	Luxuriant	Beta or gamma
Staphylococcus epidermidis (12228)	Luxuriant	Gamma

	Streptococcus pneumoniae (6303)	Luxuriant	Alpha	
	Streptococcus pyogenes (19615)	Luxuriant	Beta	
	Staphylococcus aureus (6538)	Luxuriant	Beta or gamma	
	Clostridium sporogenes (19404)	Luxuriant	-	
	Clostridium sporogenes (11437)	good-luxuriant	-	
	Clostridium perfringens (13124)	Luxuriant	-	
	Clostridium perfringens (12934)	Luxuriant	-	

Precautions :	1. For Laboratory Use.
	2. Follow proper, established laboratory procedures in handling and disposing of infectious materials.
Limitations :	1. Since the nutritional requirements of organisms vary, some strains may be encountered that fail to grow or grow poorly on this medium.
	2. Blood agar base media are intended for use with blood supplements. Although certain diagnostic tests may be performed directly on these media, biochemical and, if indicated, immunological testing using pure cultures is recommended for complete identification. Consult appropriate references for further information.
	3. Haemolytic reactions of some strains of group D streptococci have been shown to be affected by differences in animal blood. Such strains are β – hemolytic on horse, human and rabbit blood agar and α- hemolytic on sheep blood agar.
	4. Colonies of Haemophilus haemolyticus are β –hemolytic on horse and rabbit blood agar and must be distinguished from colonies of β-hemolytic streptococci using other criteria. The use of sheep blood has been suggested to obviate this problem since sheep blood is deficient in pyridine nucleotides and does not support growth of H. haemolyticus.
	5. Atmosphere of incubation has been shown to influence hemolytic reactions of β-hemolytic streptococci. For optimal performance, incubate blood agar media under increased CO_2 or anaerobic conditions.
Use :	It is used as an efficient base for preparation of blood agar, chocolate agar and for preparation of various selective and identification media and isolation of organisms from clinical and non-clinical samples.
Storage :	Dehydrated medium- below 30°C Prepared medium–Between 2 to 8°C.
Packing :	500 gm bottle

Product profile:	Reconstitution	Quantity on Preparation (500g)	pH (25°C)	Supplement	Sterilization
B994	44g/l	11.36L	7.3 ± 0.2	5% v/v sterile defibrinated sheep blood, BF's as desired	121^0C / 15 minutes