TECHNICAL SHEET

B552	**NNN MODIFIED MEDIUM (TWIN PACK)**
Formula	
Ingredients :	**gms/lit.**
Part A	-
Meat extract	3.00
Peptone	5.00
Sodium chloride	8.00
Agar	15.00
Final pH (at 25°C)	7.3 ± 0.2
Part B	-
Sodium chloride	8.00
Potassium chloride	0.20
Calcium chloride	0.20
Monopotassium dihydrogen phosphate	0.30
Dextrose	2.50
Final pH (at 25°C)	7.0 ± 0.2

Directions :

Part A: Suspend 31 grams in 1000 ml distilled water. Heat to boiling to dissolve the medium completely. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes. Cool to 45-50°C and aseptically add 10% of sterile defibrinated rabbit or human blood after inactivation at 56°C for 30mins. Mix well and dispense in 5 ml amounts in test tubes or 25 ml amounts in flasks. Allow tubed media to cool in slanted position.
Part B: Suspend 11.2 grams of Part B in 1000 ml distilled water. Heat to boiling to dissolve the medium completely. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes. Cool and add approximately 2 ml in tubes or 10-15 ml in flasks over solidified Part A medium

Principle :

This medium consists of a blood agar base and an overlay medium. The blood agar base is a highly nutritious medium that supports the growth of fastidious organisms like Leishmania and Trypanosoma . The specimens are inoculated into the liquid phase of the diphasic medium and incubated. This favours the development of organisms in the insect vector. The amastigotes transform to promastigotes in about 24 hours.

QC Tests – (I)Dehydrated Medium		
	Colour :	Part A : Cream to tan Part B: White to cream
	Appearance :	Part A +Part B-Homogeneous Free Flowing powder
(II)Rehydrated medium		
	pH (post autoclaving/heating) :	Part A-7.3 ± 0.2, Part B-7.0 ± 0.2,
	Colour (post autoclaving/heating) :	Basal medium :Light amber After addition of sterile defibrinated rabbit or human blood : Red
	Clarity (post autoclaving/heating) :	Clear to slightly opalescent, opaque after addition of sterile defibrinated rabbit or human blood
(III)Q.C. Test Microbiological		
	Cultural characteristics observed after 48 – 72 hrs at 21 – 26°C.	
	MICROORGANISM	GROWTH
	Leishmania donovani	Luxuriant
	Trepanosoma cruzi	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.
Use :	It is used for cultivation of Leishmaniae and Trypanosomes.
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
B552	31.0 +11.2 g/l	11.84L(Part A+B)	Part A 7.3 ± 0.2 Part B-7.0 ± 0.2	10% of sterile defibrinated rabbit or human blood	121^0C / 15 minutes

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