

PRODUCT SPECIFICATION SHEET

Wilson Blair Agar Base (DM294)

Intended Use

Wilson Blair Agar Base is recommended with the addition of selective reagent for the isolation of Salmonellae particularly *Salmonella* Typhi.

Product Summary and Explanation

Salmonella is a genus of gram-negative Enterobacteriaceae - commonly implicated in food borne illness and the causative agent of typhoid and paratyphoid fever. Salmonella species have been isolated from humans and animals. More than 2000 serovars of Salmonella exists with each showing different host specificities. For example, human sera the only known natural reservoir for serotype Salmonella Typhi and serotypes Salmonella Paratyphi A, B and C ⁽¹⁾. The organism can be transmitted by the faecal-oral route. It is excreted by humans in faeces and may be transmitted by contaminated water, food, or by person-to-person contact (with inadequate attention to personal hygiene).

Principles of the Procedure

Wilson and Blair Agar, formulated by Wilson and Blair ⁽²⁾ is recommended for isolating *Salmonella* species especially *Salmonella Typhi* from clinical specimens. The selective reagent formulation is a modification of the bismuth sulphite reagent described by Hajna and Perry ⁽³⁾. This medium is particularly valuable for the isolation of *S. Typhi*. The medium is highly selective for Salmonellae, being inhibitory to coliforms, *Proteus* and *Shigellae*; occasional strains of coliforms grow to form dull green or brown colonies, but without a surrounding metallic sheen. The medium is also suitable for the isolation of lactose-fermenting strains of *Salmonellae* (which can not be differentiated on lactose containing differential media) since lactose is not the fermentable substrate used in this medium ⁽⁴⁾.

Peptone special and beef extract provide nitrogenous, carbonaceous compounds and other growth nutrients. Brilliant green dye inhibits all gram-positive bacteria. Dextrose is the fermentable carbohydrate. Ferrous sulphate aids in H₂S production. Bismuth is a heavy metal, which is inhibitory to most gram-negative enteric bacilli other than *Salmonella*. Ferrous sulphate is reduced by *Salmonella* species in presence of bismuth sulphite and dextrose to form iron sulphide, indicated by black coloured colonies. Disodium hydrogen phosphate buffers the medium well. Sodium chloride balances the osmotic equilibrium.

Ingredients	Gms / Litre				
Peptone, special	10.00				
Beef extract	5.00				
Dextrose	10.00				
Sodium chloride	5.00				
Agar	30.00				
Final pH: 7.3 ± 0.2 at 25°C					
Formula may be adjusted and/or supplemented as required to meet performance specifications					

Precautions

1. For Laboratory Use only.

2. IRRITANT. Irritating to eyes, respiratory system, and skin.

3. Do not store the medium in refrigerator $(4^{\circ}C)$ for longer than 2 days, as the medium changes to green colour and reduces its selectivity.





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Directions

- 1. Suspend 60 grams in 1000 ml distilled water.
- 2. Heat to boiling to dissolve the medium completely.
- 3. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes.
- 4. To sterile melted base, aseptically add 4 ml of 1% brilliant green solution and 70 ml of selective reagent.

Selective Reagent

Solution 1 : 40 gm sodium sulphite in 100 ml distilled water.

- Solution 2 : 21 gm dibasic sodium phosphate in 100 ml distilled water.
- Solution 3 : 12.5 gm bismuth ammonium citrate in 100 ml distilled water.
- Solution 4 : 0.96 gm ferrous sulphate in 20 ml distilled water with 2 drops of hydrochloric acid.

Prepare each solution separately and then combine. Boil the combined solution until a slate grey colour develops.

Quality Control Specifications

Dehydrated Appearance	Cream to yellow colored, homogeneous, free flowing powder	
Solution	6% Solution in Distilled or deionized water is soluble on boiling,	
Prepared Medium	 Basal Medium: Light yellow coloured clear to slightly opalescent gel. After addition of the selective reagent and 1% Brilliant green, greenish yellow coloured, opaque gel forms in Petri plates. 	
Reaction of 6.0% Solution	ction of 6.0% Solution pH 7.3 <u>+</u> 0.2 at 25°C	
Gel Strength	rength Firm, compared to 3.0% Agar Gel.	

Expected Cultural Response: Cultural characteristics observed with added 1% Brilliant green and selective reagents after incubation at 35-37°C for 24-48 hours.

Sr.	Organisms	Results to be achieved			
No.		Inoculum (CFU)	Growth	Recovery	Colour of Colony
1	Cash anishis asli ATCC 25022	>=10 ³	inhibited	0%	
1.	Escherichia coli ATCC 25922	>=10	Innibited	0%	
2.	Proteus mirabilis ATCC 25933	50-100	good-luxuriant	>=50%	Green
3.	Salmonella Typhi ATCC 6539	50-100	good-luxuriant	>=50%	black with sheen
4.	Salmonella Typhimurium ATCC 14028	50-100	good-luxuriant	>=50%	black with sheen

The organisms listed are the minimum that should be used for quality control testing.

Storage

Store the sealed bottle containing the dehydrated medium at 10- 30°C. Once opened and recapped, place container in a low humidity environment at the same storage temperature. Protect from moisture and light.

Expiration

Refer to the expiration date stamped on the container. The dehydrated medium should be discarded if not free flowing, or if the appearance has changed from the original color. Expiry applies to medium in its intact container when stored as directed.





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Packaging

Product Name : Wilson Blair Agar Base Product Code : DM294 Available Pack sizes : 100gm / 500gm

References

 Murray P. R., Baron J. H., Pfaller M. A., Jorgensen J. H. and Yolken R. H., (Ed.), 2003, Manual of Clinical Microbiology, 8th Ed., American Society for Microbiology, Washington, D.C.
 Wilson W. J. and Blair E. M., 1926, J. Pathol. Bacteriol., 29: 310.
 Hajna A. A. and Perry C. A., 1938, J. Lab. Clin. Med., 23:1185.

Further Information

For further information please contact your local MICROMASTER Representative.

MICROMASTER LABORATORIES PRIVATE LIMITED

Unit 38/39, Kalpataru Industrial Estate, Off G.B. Road, Near 'R-Mall', Thane (W) - 400607. M.S. INDIA. Ph: +91-22-25895505, 4760, 4681. Cell: 9320126789. Email: <u>micromaster@micromasterlab.com</u>

sales@micromasterlab.com

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Prepared By	Checked By	Approved By		
Fdelann 01.01.2018	Augdak 01.01.2018	(abole 0.01.2018		
Microbiologist	Head Quality Control	Head Quality Assurance		

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