

Department	Micro Laboratory	Document no	MICLAB – METHOD 010		
Title	Preparation of Asparagine Broth				
Prepared by:		Date:		Supersedes:	
Checked by:		Date:		Date Issued:	
Approved by:		Date:		Review Date:	

1.0 SUMMARY OF CHANGES

Version #	Revision History
MICLAB – METHOD 010	New

2.0 PURPOSE

This document describes the method for preparation of Asparagine Broth.

3.0 SCOPE

Asparagine Broth is a medium used in the confirmation step for Pseudomonas aeruginosa.

4.0 RESPONSIBILITY \ BUSINESS RULES

All microbiology staff in the laboratory.

5.0 PROCEDURE

5.1 Materials and Reagents required

- 5.1.1 Plastic spoon
- 5.1.2 Measuring Cylinder
- 5.1.3 RO Water & RO water squeeze bottle
- 5.1.4 3 Glass beakers
- 5.1.5 Funnel
- 5.1.6 Flat medical bottles / Schott bottles
- 5.1.7 Automatic dispenser
- 5.1.8 DL-Asparagine Monohydrate
- 5.1.9 Di Potassium Hydrogen Phosphate K_2HPO_4
- 5.1.10 Magnesium Sulphate Heptahydrate $MgSO_4 \cdot 7H_2O$
- 5.1.11 pH meter
- 5.1.12 1N NaOH and 1N HCl

5.2 Method

- 5.2.1 Weigh out 3.0g per litre of Asparagine DL.
- 5.2.2 Weigh out 1.0g per litre of Di Potassium Hydrogen Phosphate K_2HPO_4 .
- 5.2.3 Weigh out 0.5g per litre of Magnesium Sulphate Heptahydrate $MgSO_4 \cdot 7H_2O$
- 5.2.4 Dissolve the above ingredients in approximately 10mL of RO water.
- 5.2.5 Combine all 3 ingredients. Use the squeeze bottle to wash out all particles.
- 5.2.6 Make up to required volume with RO water.

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- 5.2.7 Adjust pH to 7.1 using 1N NaOH or 1N HCl.
- 5.2.8 Dispense 95mL volumes into flat medical bottles / schott bottles.
- 5.2.9 Label all bottles with medium name, batch number and date of preparation.
- 5.2.10 Sterilise by autoclaving at 121°C for 15 minutes.
- 5.2.11 Final pH after sterilisation should be 7.1 ± 0.2 . If not within range, notify the microbiologist and discard the batch.
- 5.2.12 Record all details of media preparation on SF150712.

6.0 DEFINITIONS / ACRONYMS

NA