

Department	Micro Laboratory	Document no	MICLAB – METHOD 005		
Title	Preparation of Diluent plus 0.1% Tween 80				
Prepared by:		Date:		Supersedes:	
Checked by:		Date:		Date Issued:	
Approved by:		Date:		Review Date:	

1.0 REVIEW HISTORY

Version #	Revision History
MICLAB – METHOD 005	New

2.0 PURPOSE

This document describes the method for preparation of Diluent plus 0.1% Tween 80.

3.0 SCOPE

Diluent plus 0.1% Tween 80 is used as a high quality diluent for microbiology testing.

4.0 RESPONSIBILITY \ BUSINESS RULES

All microbiology staff at the GMP site.

5.0 PROCEDURE

5.1 Materials and Reagents required

- 5.1.1 Plastic spoon
- 5.1.2 Measuring Cylinder
- 5.1.3 RO Water
- 5.1.4 Beaker
- 5.1.5 Appropriate glassware
- 5.1.6 Automatic dispenser
- 5.1.7 pH meter
- 5.1.8 1N NaOH and 1N HCl
- 5.1.9 Bacteriological Peptone (Oxoid L34)
- 5.1.10 Tween 80. (Polysorbate BP 80 item 119A012)

5.2 Method

- 5.2.1 Weigh out 1g per litre of peptone into a beaker and add approximately 500mL of RO water. Mix well to dissolve.
- 5.2.2 Weigh out 1g per litre of Tween 80 into a beaker and add RO water.
- 5.2.3 Dissolve on high setting in the microwave with frequent stirring.
- 5.2.4 Combine dissolved peptone with dissolved Tween 80 and make up to required volume with RO water, mix well.
- 5.2.5 Adjust pH to 6.9.
- 5.2.6 Dispense into appropriate glassware.
- 5.2.7 Sterilise by autoclaving at 121°C for 15 minutes.
- 5.2.8 pH after autoclaving, 7.0 ± 0.2

Department	Micro Laboratory	Document no	MICLAB – METHOD 005
Title	Preparation of Diluent plus 0.1% Tween 80		

5.2.9 Record all details of media preparation into appropriate form.

QUALITY CONTROL REQUIREMENTS			
STORAGE:	BULK	-	6 months in dark cupboard
ECOMETRIC EVALUATION / FERTILITY			
CONTROL ORGANISMS:			
Positive:	P.aeruginosa A.T.C.C 9027	Negative:	S.aureus N.C.T.C 6571
Growth Index:	Growth	Growth Index:	Growth
INCUBATION CONDITIONS:			
Temperature:	30 ± 1°C	Time:	24 hours

For Sampling Plan and Acceptance Criteria for microbiological culture media refer to SF150154.

6.0 DEFINITIONS / ACRONYMS

NA