## Milk Medium with Reducing Agent

#### Intended Use

Milk medium with reducing agent is used for determination of litmus reaction of Clostridium species.

## Summary

Milk is a complex nutritional source that contains proteins (mainly casein) in an aqueous solution of lactose and minerals. Bacterial enzymes alter the media and may bring about various changes. Litmus is added to the medium to detect pH changes that may occur as a result of these enzymatic reactions. Above pH 8.3, litmus is blue, while below pH 4.5 litmus is red. Fermentation of lactose results in the production of acid, which causes milk to curdle or form a clot at the bottom of the tube. Litmus may also act as an electron acceptor thus becoming reduced by bacterial metabolism. This reaction is observed as a white colour in the medium. Milk medium with reducing agent is used for determination of litmus reaction of *Clostridium* species.

#### **Principle**

Peptic digest of animal tissue and skim milk provide nitrogen, sulphur, vitamins and other growth nutrients. Sodium thioglycollate is a reducing agent, which absorbs oxygen and creates a reduced environment required by anaerobes. This medium has been found satisfactory for the cultivation of *Clostridium* species and allows observation of their reactions in litmus milk. In anaerobically grown Litmus Milk cultures, enzymes of *Clostridium* perfringens attack the proteins and carbohydrates of the milk producing a stormy fermentation with clotting and gas formation.

Formula*	
Ingredients	g/L
Skim Milk	100.0
Peptic Digest of Animal Tissue	10.0
Sodium Thioglycollate	0.5
Litmus	5.0
Final pH (at 25°C)	$7.0 \pm 0.2$
*Adjusted to suit performance pa	arameters

# Storage and Stability

Store dehydrated medium below 30°C in tightly closed container and the prepared medium at 2°C-8°C. Avoid freezing and overheating. Use before expiry date on the label. Once opened keep powdered medium closed to avoid hydration.

# **Specimen Collection and Handling**

Ensure that all samples are properly labelled. Follow appropriate techniques for handling samples as per established guidelines. Some samples may require special handling, such as immediate refrigeration or protection from light, follow the standard procedure. The samples must be stored and tested within the permissible time duration. After use, contaminated materials must be sterilized by autoclaving before discarding.

# Directions

- 1. Suspend 115.50 g of the powder in 1000 mL distilled water agitating.
- 2. Sterilize by autoclaving at 121°C (15 psi) for 5 min as per validated cycle.
- 3. Mix well and dispense as desired.

# **Quality Control**

**Dehydrated Appearance:** Light purple coloured, homogeneous, free flowing powder. **Prepared Appearance:** Purple coloured opalescent solution forms in tubes. **Cultural Response:** Cultural characteristics is observed after an incubation at 35°C-37°C for 5 days.

Organism (ATCC)	Observation	Cause
Clostridium perfringens (12924)	Stormy fermentation	Gas trapped in acid coagulated casein peptonization
Clostridium sporogenes (11437)	Acid with gas proteolysis	-

# Performance and Evaluation

Performance of the product is dependent on following parameters as per product label claim:

- 1. Directions
- 2. Storage
- 3. Expiry

## Warranty

This product is designed to perform as described on the label and package insert. The manufacturer disclaims any implied warranty of use and sale for any other purpose.

# Reference

- 1. Gainor C. and Wegemer D. E., Appl. Microbiol., 1954 March; 2(2): 9597.
- 2. Data on file: Microxpress®, A Division of Tulip Diagnostics (P) Ltd.

#### **Product Presentation:**

Cat No.	Product description	Pack Size
201130550500	Dehydrated Culture Media	500 g

#### Disclaimer

Information provided is based on our inhouse technical data on file, it is recommended that user should validate at his end for suitable use of the product.