Reinforced Clostridial Medium, RCM (i23158)

Reinforced Clostridial Medium is used for cultivating and enumerating *clostridia*, other anaerobes, and other species of bacteria from foods and clinical specimens.

Industry: Final product Quality Control/ Veterinary / Pharmaceutical

Principles & Uses

Reinforced Clostridial Medium, developed by Hirsch and Grinstead, is a semisolid formulation designed for cultivating and enumerating anaerobes, particularly *Clostridium*, in various samples, including food and clinical specimens. The medium's superiority in supporting *Clostridium* growth, especially from small inoculum sizes, is attributed to its well-balanced composition.

The components, including peptone, meat extract, yeast extract, glucose, sodium chloride, starch, L-Cysteine hydrochloride, and sodium acetate, play specific roles. They provide essential nutrients, act as fermentable carbohydrates, maintain osmotic balance, function as growth factors, and serve as reducing agents and buffers.

As a non-selective enrichment medium, it accommodates the growth of diverse anaerobic microorganisms and facultative bacteria under anaerobic conditions. Notably, it is recommended by the European Pharmacopoeia and USP for testing *Clostridia* in non-sterile products. Additionally, the medium can be modified by adding Polymyxin B to inhibit Gram-negative bacteria.

Incubation can be followed by colony counting and further testing if necessary. The addition of a layer of paraffin viscous or agar after inoculation is recommended for optimal results.

Composition (gr/L)

Peptone 5 g, Meat Extract 10 g, Yeast Extract 3 g, D (+) glucose 5 g, Sodium chloride 5 g, Starch 1 g, L-Cysteine HCl 0.5 g, Sodium Acetate 3 g, Agar 0.5 g.

Final pH at 25°C 6.8 ± 0.2

Preparation from dehydrated Powder

Dissolve 33 g / litre, dispense into test tubes. Autoclave for 15 min at 121 °C. Cool down. If required, add 0.02 g Polymyxin B / litre in form of an aqueous solution and mix.

Quality Control

Dehydrated Appearance: Cream to yellow, homogeneous, free flowing.

Prepared Appearance: Light yellow, clear to slightly opalescent.

Reaction of 3.3% Solution at 25°C: pH 6.8 ± 0.2

Cultural Response

Cultural response was observed in an anaerobic condition after incubation at 35 - 37 ˚C for 40 – 48 hours.

Organism (ATCC*)	Recovery
Clostridium sporogenes (11437)	good
Clostridium perfringens (10453)	good
Clostridium sporogenes (19404)	good
Clostridium novyi (17861)	good
Staphylococcus aureus (25923)	good
Escherichia coli (25922)	good

*ATCC is a registered trade mark of the American Type Culture Collection.

Storage

Keep the container at 15-30 °C. Store prepared medium at 2-8 °C.