

## Technical Information

### Charcoal Agar Base, MiVeg

#### Product Code : VM1344

**Application:-** Charcoal Agar Base, MiVeg with supplements is used for the cultivation of *Bordetella pertussis* for vaccine production and also for the maintenance of stock cultures.

#### Composition

Ingredients	Gms / Litre
MiVeg infusion	12.00
MiVeg peptone	10.00
Yeast extract	3.50
Sodium chloride	5.00
Starch, soluble	10.00
Charcoal	4.00
Agar	18.00
Final pH ( at 25°C)	7.3±0.2

\*\* Formula adjusted, standardized to suit performance parameters.

#### Principle & Interpretation

Charcoal Agar Base MiVeg is prepared by using vegetable peptones in place of animal based peptones which makes it BSE/TSE risk free. This medium is the modification of Charcoal Agar Base formulated according to the method devised by Mishulow et al (1) which is recommended for the cultivation of *Bordetella pertussis* and its vaccine production. Earlier medium viz. Bordet Gengou, can be replaced by this medium, as the conventional medium for the vaccine production of *Bordetella pertussis* as suggested by Ensminger et al (2) who added charcoal to the medium. It contains MiVeg infusion, MiVeg peptone and yeast extract which supplies essential nutrients to the organisms. Sodium chloride maintains osmotic equilibrium. Starch supports growth of organism as it is the carbohydrate source in the medium. With charcoal it neutralizes toxic substances like fatty acid, which can inhibit growth of *Bordetella*. The difficulty in the isolation of *Bordetella pertussis* from nasopharyngeal secretions is the repression of unwanted flora during the long incubation period on nutritious media. To resist other contaminants Penicillin can be added to the medium as an antimicrobial agent. However Penicillin resistant floras still cause the contamination which was observed by Lacey (3). Methicillin was found to be superior than Penicillin in suppressing unwanted nasopharyngeal flora as observed by Broome et al (4). Sutcliffe and Abbott found that Cephalexin was still better than Methicillin (5). Therefore this medium with added supplement, Cephalexin and blood is suitable for cultivation of *B. pertussis*.

The media can also be used for the maintenance of stock cultures of *Bordetella pertussis* on slants with weekly subcultures. This media can be converted to Chocolate Agar Base for isolation of *Haemophilus* species.

#### Methodology

Suspend 31.25 grams of powder media in 450 ml distilled water. Mix thoroughly. Boil to dissolve the medium with frequent stirring. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes. Cool to 45-50°C. Aseptically add sterile 50 ml of defibrinated blood and Bordetella Selective Supplement (MS2004).

#### Quality Control

##### Physical Appearance

Grey coloured, homogeneous, free flowing powder.

##### Gelling

Firm, comparable with 1.8% Agar gel

### Colour and Clarity of prepared medium

Black coloured, opaque gel forms in petri plates and contains undissolved black particles.

### Reaction

Reaction of 6.25 % w/v aqueous solution pH: 7.3  $\pm$  0.2 at 25°C

### pH range

7.1-7.5

### Cultural Response/Characteristics

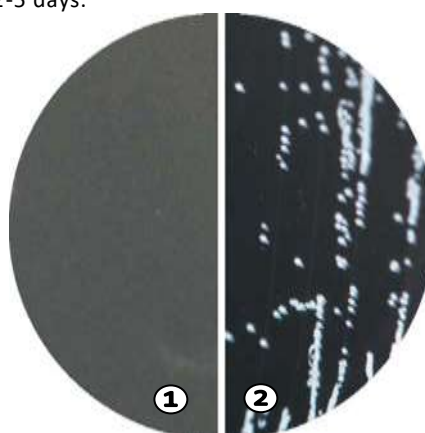
Cultural characteristics observed after an incubation at 35-37°C for 24-48 hours

Organisms (ATCC)	Inoculum (CFU)	Growth	Recovery
<i>Bordetella bronchiseptica</i> (4617)	$10^2$ - $10^3$	luxuriant	>50%
<i>Bordetella paraptussis</i> (15237)	$10^2$ - $10^3$	luxuriant	>50%
<i>Bordetella pertussis</i> (8467)	$10^2$ - $10^3$	luxuriant	>50%

## Storage and Shelf Life

**Dried Media:** Store below 30°C in tightly closed container and use before expiry date as mentioned on the label.

**Prepared Media:** 2-8° in sealable plastic bags for 2-5 days.



VM1344 Charcoal Agar Base MiVeg

1. Control
2. *Bordetella bronchiseptica*

## Further Reading

1. Mishulow L., Sharpe L. S. and Cohen L. L., 1953, J. Pub. Hlth., 43(II) : 1466.
2. Ensminger P. W., Gulberston C. G. and Powell H. M., 1953. J. Infect. Dis., 93(3):266
3. Lacey B.W., 1954, J. Hyg., 59:273.
4. Broome C.V., Fraser D.W. and English J.W., 1979, Internat. Symp. on Pertussis DHEW J., Washington D.C. pp 19-29.
5. Sutcliffe E.M. and Abbott J.D., 1979, B.M.J. II:732-733.

## Disclaimer :

- User must ensure suitability of the product(s) in their application prior to use.
- The product conform solely to the technical information provided in this booklet and to the best of knowledge research and development work carried at CDH is true and accurate
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