

39484 Methyl Red Voges Proskauer Broth (MR VP Broth; Buffered Glucose Broth; Glucose Phosphate Broth)

A classic liquid medium recommended for the Methyl-red and Voges-Proskauer tests for the differentiation of the coli-aerogenes group according ISO standards 6579 and 6585 and FIL - IDF 93 standard.

Composition:

Ingredients	Grams/Litre
Buffered Peptone	7.0
Dextrose	5.0
Dipotassium Phosphate	5.0
Final pH 6.9 +/- 0.2 at 25°C	

Store prepared media below 8°C, protected from direct light. Store dehydrated powder, in a dry place, in tightly-sealed containers at 2-25°C.

Appearance: Cream coloured, homogeneous, free flowing powder.
Colour and Clarity: Light yellow coloured, clear solution without any precipitate.

Directions:

Dissolve 17 g of in 1 litre of distilled water. Mix well, distribute into test tubes in amounts of 10 ml and sterilise by autoclaving at 121°C for 15 minutes.

Inoculate two tubes of Methyl Red Voges Proskauer Broth per organism. The culture must be pure. Incubate up to 5 days at 30-35°C.

Principle and Interpretation:

Methyl red test (MR Test):

Clark and Lumbs used found that *E. coli* ferments glucose by producing mixed acids (e.g lactic, acetic and formic acid) which can be made visible with the addition of methyl red. These acids give a pH below 4.4 which means methyl red turns to red (yellow when pH > 5.1).

Add about 5-6 drops of the Methyl Red Solution (Cat. No. 08714) per 5 ml culture. Incubate 24-48 hours at 37°C and observe the color of the medium - if the pH falls below 4.4 the indicator changes to red. In case the result is doubtful the assay must be repeated incubating at 30°C for 5 days.

Voges-Proskauer test (VP Test):

Voges and Proskauer found a test to detect acetoin and 2,3-butanediol produced due to the fermentation of glucose by *Klebsiella* and *Enterobacter*. They found that under alkaline conditions this two compounds oxidise themselves to diacetyl. Diacetyl reacts with creatine (a guanidine derivative) to a red or with α -naphthol to a violet compound.

Different Reagents can be taken:

- Add 1.0 ml O'Meara's reagent (Cat. No. 07689). Shake tubes gently for 30 seconds to 1 minute to expose the medium to atmospheric oxygen in order to oxidize the acetoin so as to obtain a color reaction. Allow tube to stand at 35°C or room temperature for 4 hours. If acetoin was produced the medium turns to pinkish red.



- Add 0.6 ml (6 drops) of Barritt's Reagent A (Cat. No. 29333) and 0.2 ml (2 drops) of Barritt's Reagent B (Cat. No. 39442) for 10 ml medium. Shake tubes gently for 30 seconds to 1 minute to expose the medium to atmospheric oxygen in order to oxidize the acetoin so as to obtain a color reaction. Allow tube to stand at least 10 to 15 minutes. If the medium change to violet pinked the test was positive. This reagent is more sensitive.

Buffered Peptone is a source for nitrogen and carbon used as base nutrition for the Enterobacteriaceae. Dextrose is the fermentable sugar and dipotassium phosphate is the buffering agent.

Cultural characteristics after 48 hours at 30°C.

Organisms (ATCC)	Growth	MR Test	VP Test
<i>Escherichia coli</i> (25922)	+++	+ (red)	- (no change)
<i>Enterobacter aerogenes</i> (13048)	+++	- (yellow)	+ (red)
<i>Klebsiella pneumoniae</i> (23357)	+++	- (yellow)	+ (red)

References:

1. T. Smith, Amer. J. Med. Sci., 110, 283 (1895)
2. W.M. Clark, H.A. Lubs, J. Inf. Dis., 17, 160-173 (1915)
3. O. Voges, B. Proskauer, Z. f. Hyg., 28, 20-22 (1898)
4. M.M. Barritt, J. Path. Bact. 42, 441-454 (1936)
5. R.A.Q. O'Meara, J. Path. Bact., 34, 401 (1931)
6. International Organization for Standardization (ISO), Microbiology of food and animal feeding stuffs - Horizontal method for the detection of Salmonella spp., ISO Draft 6579:2002
7. International Organization for Standardization (ISO), Milk and milk products - Detection of Salmonella spp., ISO 6785:2001
8. FIL-IDF International Provisional Standard, Dried Milk Products, : Enumeration of Bacillus cereus MPN Technique, 181 (1998)
9. FIL-IDF, Milk and milk products - Detection of Salmonella spp. (2001)

Precautions and Disclaimer

This product is for R&D use only, not for drug, household, or other uses. Please consult the Material Safety Data Sheet for information regarding hazards and safe handling practices.

The vibrant M, Millipore, and Sigma-Aldrich are trademarks of Merck KGaA, Darmstadt, Germany or its affiliates. Detailed information on trademarks is available via publicly accessible resources.
© 2018 Merck KGaA, Darmstadt, Germany and/or its affiliates. All Rights Reserved.

The life science business of Merck KGaA, Darmstadt, Germany operates as MilliporeSigma in the US and Canada.

