PRODUCT SPECIFICATION SHEET

Peptone Iron Agar (DM515)

Intended Use
Peptone Iron Agar (DM515) is recommended for detection of hydrogen sulphide production by microorganisms.

Product Summary and Explanation
Certain bacterial species have the ability to liberate sulfur from sulfur-containing amino acids or other compounds in the form of hydrogen sulphide which forms an important characteristic for their identification. Hydrogen sulphide production can be detected by incorporating a sulfur source and an H_2S indicator system in the medium.[1] Levine and co-workers described a medium containing proteose peptone and ferric citrate for detection of hydrogen sulfide production by coliform bacteria. They demonstrated that such a medium served to differentiate strains that were Voges-Proskauer negative, methyl-red positive and citrate positive from other members of the *Enterobacteriaceae*.[2, 3] Levine reported that ferric citrate was a much more sensitive indicator of hydrogen sulfide production than lead acetate, producing a medium that gave definite reactions within 12 hours. Peptone Iron Agar is a modification of Levine's original formula in which peptone has been included with proteose peptone and the more soluble ferric ammonium citrate is used in place of ferric citrate. This medium utilizes sodium thiosulphate, an inorganic compound as a supplemental source of sulfur and ferric ammonium citrate as the H_2S indicator in the medium. Tittsler and Sandholzer[4] compared Peptone Iron Agar with lead acetate agar for the detection of hydrogen sulfide and found that Peptone Iron Agar had the advantage of giving earlier reactions and clearer results.

Principles of the Procedure
Peptone Iron Agar contains peptic digest of animal tissue and proteose peptone provide carbonaceous and nitrogenous compounds, including trace elements. Sodium thiosulphate and ferric ammonium citrate are use to detect H_2S production. Sulphide is released from thiosulphate due to the action of bacterial enzymes. This sulphide then couples with a hydrogen ion to form H_2S, which then reacts with the ferric ions from ferric ammonium citrate to produce insoluble heavy metal sulphides that appear as a black precipitate. Sodium glycerophosphate buffers the medium.

Formula / Liter

Ingredients	Gms / Liter
Peptic digest of animal tissue	15.00
Proteose peptone	5.00
Ferric ammonium citrate	0.50
Sodium glycerophosphate	1.00
Sodium thiosulphate	0.08
Agar	15.00
Final pH: 6.7 ± 0.2 at 25°C	
Formula may be adjusted and/or supplemented as required to meet performance specifications	

Precautions
1. For Laboratory Use only.
2. IRRITANT. Irritating to eyes, respiratory system, and skin.

PRODUCT SPECIFICATION SHEET

Directions

1. Suspend 36.58 grams of the medium in one liter of distilled water.
2. Heat to boiling, to dissolve the medium completely.
3. Autoclave at 121°C, 15 psi pressure, for 15 minutes / validated cycle.
4. Allow the tubed medium to cool in an upright position or in a slanting position to form slants.

Quality Control Specifications

Dehydrated Appearance	Cream to yellow homogeneous free flowing powder
Prepared Medium	Light amber coloured clear to slightly opalescent gel forms in tubes as slants
Reaction of 3.66% Solution	pH : 6.7 ± 0.2 at 25°C
Gel Strength	Firm, comparable with 1.5% Agar gel

Expected Cultural Response: Cultural characteristics observed after an incubation at 35-37°C for 18-24 hours.

Sr. No.	Organisms	Results to be achieved	
		Inoculum (CFU)	H_2S Production
1.	*Escherichia coli ATCC 25922*	50 – 100	negative reaction, no blackening of medium
2.	*Enterobacter aerogenes ATCC 13048*	50 – 100	negative reaction, no blackening of medium
3.	*Salmonella Typhi ATCC 6539*	50 – 100	positive reaction, blackening of medium
4.	*Salmonella Enteritidis ATCC 13076*	50 – 100	positive reaction, blackening of medium

The organisms listed are the minimum that should be used for quality control testing.

Test Procedure

1. Obtain a pure culture of a test organism. With help of an inoculating needle pick the center of a single colony.
2. Using stab method, inoculate a tube of Peptone Iron Agar. Stab the needle to within 1/4 to 1/2 inch of the bottom. Withdraw the needle following the initial line of inoculation.
3. Incubate tubes at 35-37°C for 18-24 hours.
4. Read tubes for growth and hydrogen sulfide production.

Results

1. Any blackening of the medium along the line of inoculation or throughout the butt indicates hydrogen sulfide production.
2. Refer to appropriate references and procedures for a complete discussion of the identification of coliform bacteria.

Storage

Store the sealed bottle containing the dehydrated medium at 10 - 30°C. Once opened and recapped, place container in a low humidity environment at the same storage temperature. Protect from moisture and light.

PRODUCT SPECIFICATION SHEET

Expiration

Refer to the expiration date stamped on the container. The dehydrated medium should be discarded if not free flowing, or if the appearance has changed from the original color. Expiry applies to medium in its intact container when stored as directed.

Limitations of the Procedure

1. For identification, organisms must be in pure culture. Morphological, biochemical and/or serological tests should be performed for final identification.
2. Consult appropriate texts for detailed information and recommended procedures.

Packaging

Product Name : Peptone Iron Agar
Product Code : DM515
Available Pack sizes : 100gm

References

1. Koneman E. W, Allen S. D., Janda W. M., Schreckenberger P. C., Winn W.C. Jr., 1992, Colour Atlas and Textbook of Diagnostic Microbiology, 4th Ed, J. B. Lippincott Company, Philadelphia.
2. Levine, Vaughn, Epstein and Anderson. 1932. Proc. Soc. Exp. Biol. Med. *29*:1022.
3. Levine, Epstein and Vaughn. 1934. Am. J. Public Health *24*:505.
4. Tittsler and Sandholzer. 1937. Am. J. Public Health *27*:1240.

Further Information

For further information please contact your local MICROMASTER Representative.

MICROMASTER LABORATORIES PRIVATE LIMITED
Unit 38/39, Kalpataru Industrial Estate,
Near Runwal Estate, Behind 'R-Mall' ,Ghodbunder Raod,
Thane (W) – 400607. M.S. INDIA.
Ph: +91-22-25895505, 4760, Cell: 9320126789.
Email: micromaster@micromasterlab.com
sales@micromasterlab.com

DM515PSS,QAD/FR/024,Rev.00/01.01.2018

Prepared By	Checked By	Approved By
01.01.2018	01.01.2018	01.01.2018
Microbiologist	Head Quality Control	Head Quality Assurance

PRODUCT SPECIFICATION SHEET

Disclaimer :

All Products conform exclusively to the information contained in this and other related Micromaster Publications. Users must ensure that the product(s) is appropriate for their application, prior to use. The information published in this publication is based on research and development work carried out in our laboratory and is to the best of our knowledge true and accurate. Micromaster Laboratories Pvt Ltd reserves the right to make changes to specifications and information related to the products at any time. Products are intended for laboratory, diagnostic, research or further manufacturing use only and not for human or animal or therapeutic use, unless otherwise specified. Statements included herein should not be considered as a warranty of any kind, expressed or implied, and no liability is accepted for infringement of any patents.