IFU

Instructions for Use

CRITERION™ MIO (MOTILITY, INDOLE, ORNITHINE) MEDIUM

Cat. no. C6330	CRITERION™ MIO Medium	62gm
Cat. no. C6331	CRITERION™ MIO Medium	500gm
Cat. no. C6332	CRITERION™ MIO Medium	2kg
Cat. no. C6333	CRITERION™ MIO Medium	10kg
Cat. no. C6334	CRITERION™ MIO Medium	50kg

INTENDED USE

Hardy Diagnostics CRITERION™ MIO (Motility, Indole, Ornithine) Medium is recommended for use in testing motility, indole production, and ornithine decarboxylase activity of enteric bacilli.

This dehydrated culture medium is a raw material intended to be used in the making of prepared media products, which will require further processing, additional ingredients, or supplements.

SUMMARY

Ederer and Clark, et al., developed Motility Indole Ornithine (MIO) Medium to be used as an aid in the identification of members of the Enterobacteriaceae family.[7] Motility, indole production, and ornithine decarboxylation are the three differentiating tests that are provided in the one culture tube.

The addition of agar to the medium allows for the detection of motility along the stab line of inoculation. Motile organisms extend from the stab line and produce turbidity or cloudiness throughout the medium. Non-motile organisms grow only along the stab line and leave the surrounding medium clear.

Tryptophan, present in the basal medium, is degraded by organisms that possess the enzyme tryptophanase. Degradation of tryptophan produces indole which is detected upon addition of Kovacs Reagent to the surface of the medium. Indole combines with p-dimethylaminobenzaldehyde and produces a red band at the top of the medium. A negative indole test results in no color change upon addition of Kovacs Reagent.

Bromcresol purple serves as the pH indicator which allows for detection of decarboxylase activity. Organisms that ferment dextrose will produce acids, thereby lowering the pH. A decreased pH results in the indicator changing from purple to yellow. The presence of acid also results in stimulation of enzyme activity. Once the enzyme has decarboxylated ornithine, the by-product diamine putrescine is produced. Production of putrescine causes an alkaline shift which turns the medium a dark purple. Organisms which do not produce decarboxylase remain acidic due to dextrose-fermentation, and the medium retains a yellow (acidic) color throughout or yellow with a purple band near the top of the tube.

FORMULA

Gram weight per liter:	31.0gm/L
Pancreatic Digest of Gelatin	10.0gm
Pancreatic Digest of Casein	10.0gm
L-Ornithine	5.0gm
Yeast Extract	3.0gm
Dextrose	1.0gm
Bromcresol Purple	0.02gm
Agar	2.0gm

Final pH 6.5 +/- 0.2 at 25ºC.

* Adjusted and/or supplemented as required to meet performance criteria.

STORAGE AND SHELF LIFE

Store the sealed bottle(s) containing dehydrated culture medium at 2-30ºC. Dehydrated culture medium is very hygroscopic. Keep lid tightly sealed. Protect dehydrated culture media from moisture and light. The dehydrated culture media should be discarded if it is not free-flowing or if the color has changed from its original beige.

Store the prepared culture media at 15-30ºC.

The expiration date on the product label applies to the product in its intact packaging when stored as directed. The product may be used and tested up to the expiration date on the product label and incubated for the recommended incubation times as stated below.

Refer to the document "Storage" for more information.

PRECAUTIONS

This product may contain components of animal origin. Certified knowledge of the origin and/or sanitary state of the animals does not guarantee the absence of transmissible pathogenic agents. Therefore, it is recommended that these products be treated as potentially infectious, and handle observing the usual universal blood precautions. Do not ingest, inhale, or allow to come into contact with skin.

This product is for laboratory use only. It is to be used only by adequately trained and qualified laboratory personnel. Observe approved biohazard precautions and aseptic techniques. All laboratory specimens should be considered infectious and handled according to "standard precautions." Refer to the document "Guidelines for Isolation Precautions" from the Centers for Disease Control and Prevention.

For additional information regarding specific precautions for the prevention of the transmission of all infectious agents from laboratory instruments and materials, and for recommendations for the management of exposure to infectious disease, refer to CLSI document M29: *Protection of Laboratory Workers from Occupationally Acquired Infections.*

Sterilize all biohazard waste before disposal.

Refer to the document "Precautions When Using Media" for more information.

METHOD OF PREPARATION FOR DEHYDRATED CULTURE MEDIA

1. Suspend 31.0gm of the dehydrated culture media in 1 liter of distilled or deionized water.

2. Heat to boiling and mix to dissolve completely.

3. Dispense into tubes.

4. Sterilize in the autoclave at 121ºC. for 15 minutes.

PROCEDURE AND INTERPRETATION OF RESULTS

For information on procedures and interpretation of results, consult listed references or refer to the prepared media Instructions for Use (IFU) for Cat. No. Q20.

LIMITATIONS

It is recommended that biochemical, immunological, molecular, or mass spectrometry testing be performed on colonies from pure culture for complete identification.

Some formulations may require a settling period before pH testing of the prepared medium. If the pH is tested immediately after preparation and is out of specification, retest the medium after 24 hours to obtain final pH results. Always take pH reading at room temperature.

Weak motile organisms or organisms that possess damaged flagella (due to heating, shaking, or other trauma) often result in false-negative motility tests. Motility results may be confirmed by performing a hanging drop motility test. Consult listed references for procedure.[2-4,6]

Motility and Ornithine results must be interpreted prior to addition of Kovacs Reagent.

A purple color near the top of the tube may result due to oxidation.

Erroneous results may occur if caps are not loose during incubation.

Refer to the document "Limitations of Procedures and Warranty" for more information.

MATERIALS REQUIRED BUT NOT PROVIDED

Standard microbiological supplies and equipment such as autoclaves, incinerators, and incubators, etc., are not provided.

QUALITY CONTROL

Hardy Diagnostics tests each lot of commercially manufactured media using appropriate quality control microorganisms and quality specifications as outlined on the Certificate of Analysis (CofA) and the CLSI document M22-A3 *Quality Assurance for Commercially Prepared Microbiological Culture Media*. The following microorganisms are routinely used for testing at Hardy Diagnostics:

Test Organisms	Inoculation Method*	Incubation			Results
		Time	Temperature	Atmosphere	
Escherichia coli ATCC® 25922	D	18-48hr	35°C	Aerobic	Growth; motility positive, indole positive (turns pink after adding Kovacs Reagent), ornithine positive (purple throughout tube)
Klebsiella pneumoniae ATCC® 13883	D	18-48hr	35°C	Aerobic	Growth; motility negative, indole negative, ornithine negative (purple top layer, rest

					of tube yellow)

* Refer to the document "Inoculation Procedures for Media QC" for more information.

USER QUALITY CONTROL

Users of dehydrated culture media should perform QC testing in accordance with applicable government regulatory agencies, and in compliance with accreditation requirements. Hardy Diagnostics recommends end users check for signs of contamination and deterioration and, if dictated by laboratory quality control procedures or regulation, perform quality control testing to demonstrate growth or a positive reaction and to demonstrate inhibition or a negative reaction, if applicable. Hardy Diagnostics quality control testing is documented on the certificate of analysis (CofA) available from Hardy Diagnostics Certificate of Analysis website. In addition, refer to the following document "Finished Product Quality Control Procedures," for more information on QC or see the reference(s) for more specific information.

PHYSICAL APPEARANCE

CRITERION™ MIO Medium powder should appear homogeneous, free-flowing, and beige in color. The prepared media should appear clear, semi-solid, and purple in color.

REFERENCES

1. Anderson, N.L., et al. *Cumitech 3B; Quality Systems in the Clinical Microbiology Laboratory*, Coordinating ed., A.S. Weissfeld. American Society for Microbiology, Washington, D.C.

2. Jorgensen., et al. *Manual of Clinical Microbiology*, American Society for Microbiology, Washington, D.C.

3. Tille, P., et al. *Bailey and Scott's Diagnostic Microbiology*, C.V. Mosby Company, St. Louis, MO.

4. Koneman, E.W., et al. *Color Atlas and Textbook of Diagnostic Microbiology*, J.B. Lippincott Company, Philadelphia, PA.

5. Isenberg, H.D. *Clinical Microbiology Procedures Handbook*, Vol. I, II & III. American Society for Microbiology, Washington, D.C.

6. MacFaddin, J.F. *Biochemical Tests for Identification of Medical Bacteria*,, Lipincott Williams & Wilkins, Philadelphia, PA.

7. Ederer and Clark. 1970. *Appl. Microbiol.*; 2:849.

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The Hardy Diagnostics manufacturing facility and quality management system is certified to ISO 13485.

HDQA 2207B [D]