IFU

Instructions for Use

PSEUDOMONAS ISOLATION AGAR

Cat. no. G145	Pseudomonas Isolation Agar, 15x60mm Plate, 11ml	10 plates/bag
Cat. no. G219	Pseudomonas Isolation Agar, 15x100mm Plate, 18ml	10 plates/bag

INTENDED USE

Hardy Diagnostics Pseudomonas Isolation Agar is recommended for the selective isolation and differentiation of *Pseudomonas aeruginosa* .

SUMMARY

Non-fermentative gram-negative bacteria (NFB) can exploit the wet environments in hospitals and are often associated with nosocomial infections. For instance, non-fermentative bacteria have been implicated in the contamination of faucet aerators, respiratory therapy equipment, as well as sterile solutions and medications. Among the NFB, *Pseudomonas aeruginosa* is the most clinically significant bacteria.(8)

Hardy Diagnostics Pseudomonas Isolation Agar is a modification of the formula developed by King, et al.(1) The medium is designed to enhance pyocyanin production, thereby improving the differentiation of pseudomonads. The modified formula contains a low phosphorous content, glycerol, magnesium chloride and potassium sulfate, all of which promote pyocyanin production.(1) Pyocyanin production is unique to *Pseudomonas aeruginosa* and is noted as a blue-green water soluble pigment that imparts a greenish color into the media.(8)

Pseudomonas Isolation Agar also includes glycerol as an energy source, while peptone provides nutrients necessary for bacterial growth. Irgasan® is incorporated into the medium to inhibit the growth of many gram-positive and gram-negative microorganisms other than *Pseudomonas* spp.(2)

FORMULA

Ingredients per 980ml of deionized water:*

Pancreatic Digest of Gelatin	20.0gm
Potassium Sulfate	10.0gm
Magnesium Chloride	1.4gm
Irgasan ®	25.0mg
Glycerol	20.0ml
Agar	13.6gm

Final pH 7.0 +/- 0.3 at 25ºC.

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

STORAGE AND SHELF LIFE

Storage: Upon receipt store at 2-8ºC. away from direct light. Media should not be used if there are any signs of deterioration (shrinking, cracking, or discoloration), contamination, or if the expiration date has passed. Product is light and temperature sensitive; protect from light, excessive heat and freezing.

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 Precautions for blood. Do not ingest, inhale, or allow to come into contact with skin.

This product is for *in vitro* diagnostic 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.

PROCEDURE

Specimen Collection: Consult listed references for information on specimen collection.[4,5] Infectious material should be submitted directly to the laboratory without delay and protected from excessive heat and cold.

Method of Use:

1. Allow the plates to warm to room temperature and dry the agar surface before inoculating.

2. Inoculate and streak the specimen as soon as possible after collection. If the specimen to be cultured is on a swab, roll the swab over one third of the agar surface.

3. Streak for isolation with a sterile loop.

4. Incubate plates aerobically at 35ºC. for 18-48 hours. Some pseudomonads do not grow well at 35ºC. If after 24 hours no growth appears, reincubate the plate(s) at 25ºC. for an additional 24 hours.

5. Examine plates daily for colony morphology and growth characteristics.

INTERPRETATION OF RESULTS

Pseudomonas aeruginosa will produce blue to blue-green pigmented colonies.

Pseudomonas fluorescens and other non-fermenting bacteria will produce colonies that are not blue-green in color.

LIMITATIONS

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

Some pseudomonads do not grow well at 35ºC. If no growth appears after 24 hours of incubation at 35ºC., the plates should be reincubated at 25ºC. for 24 hours.

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

MATERIALS REQUIRED BUT NOT PROVIDED

Standard microbiological supplies and equipment such as loops, slides, staining supplies, other culture media, microscopes, incinerators, and incubators, etc., as well as serological and biochemical reagents, 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	
Pseudomonas aeruginosa ATCC® 27853	A	18-48hr	35°C	Aerobic	Growth; blue-green colonies
Pseudomonas aeruginosa ATCC® 9027	A	18-48hr	35°C	Aerobic	Growth; blue-green colonies
Burkholderia (*Pseudomonas*) *cepacia* ATCC® 25416	A	18-48hr	35°C	Aerobic	Growth; no pigment
Escherichia coli ATCC® 25922	B	18-48hr	35°C	Aerobic	Inhibited
Staphylococcus aureus ATCC® 25923	B	18-48hr	35°C	Aerobic	Inhibited

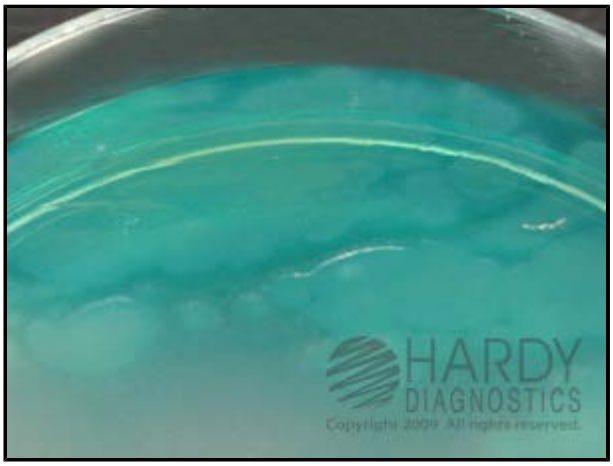
* Refer to the document "Inoculation Procedures for Media QC" for more information.

USER QUALITY CONTROL

End users of commercially prepared 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. Also refer to the document "Finished Product Quality Control Procedures," and the CLSI document M22-A3 *Quality Assurance for Commercially Prepared Microbiological Culture Media* for more information on the appropriate QC procedures. See the references below.

PHYSICAL APPEARANCE

Pseudomonas Isolation Agar should appear slightly opalescent, and light amber in color.



Pseudomonas aeruginosa (ATCC® 27853) growing on Pseudomonas Isolation Agar (Cat. no. G145). Incubated aerobically for 24 hours at 35ºC.

Burkholderia (*Pseudomonas*) *cepacia* (ATCC® 25416) growing on Pseudomonas Isolation Agar (Cat. no. G145). Incubated aerobically for 24 hours at 35ºC.

Uninoculated plate of Pesudomonas Isolation Agar (Cat. no. G145).

REFERENCES

1. King, E.O., et al. 1954. *J. Lab. Clin. Med.*; 44:301.

2. Soap and Chemical Specialties, January 1968.

3. 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.

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

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

6. MacFaddin, J.F. 1985. *Media for Isolation, Cultivation, Identification, Maintenance of Bacteria*, Vol. I. Williams & Wilkins, Baltimore, MD.

7. *Quality Assurance for Commercially Prepared Microbiological Culture Media*, M22. Clinical and Laboratory

Standards Institute (CLSI - formerly NCCLS), Wayne, PA.

8. Bodey, G.D., et al. 1989. "Infections caused by *P. aeruginosa*". *Rev. Infect. Dis.*; 5:279-313.

IFU-10694[A]

1430 West McCoy Lane, Santa Maria, CA 93455, USA
Phone: (805) 346-2766 ext. 5658
Fax: (805) 346-2760
Website: HardyDiagnostics.com
Email: TechnicalServices@HardyDiagnostics.com
Ordering Information

Distribution Centers:
California · Washington · Utah · Arizona · Texas · Ohio · New York · Florida · North Carolina

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

HDQA 2207F [D]