IFU

HARDY DIAGNOSTICS
A Culture of Service™

Instructions for Use

OGYE (OXYTETRACYCLINE-GLUCOSE YEAST EXTRACT) AGAR

Cat. no. W19	OGYE (Oxytetracycline-Glucose Yeast Extract) Agar, 15x100mm Plate, 26ml	10 plates/bag

INTENDED USE

Hardy Diagnostics OGYE (Oxytetracycline-Glucose Yeast Extract) Agar is recommended for the detection and isolation of yeasts and molds from foods.

This product is not intended to be used for the diagnosis of human disease.

SUMMARY

OGYE Agar was formulated by Mossel et al. for the selective isolation and enumeration of yeast and mold from foods.[1,2,4] OGYE Agar was initially developed for the examination of meats, especially raw minced meat, which may contain high numbers of yeast.[1] The medium is also specified for use with dairy products, and is recommended for enumeration of yeast in non-carbonated, high-acid beverage products.[7,9]

Unlike many selective fungal media, OGYE has a neutral pH and contains an antibiotic to improve recovery of fungi from mixed samples. Koburger and Mace showed that physically stressed yeast cells respond better to media using broad-spectrum antibiotics and a neutral pH.[3] In addition, the medium has also been shown to suppress bacterial growth and provide improved recovery of fungi from a variety of foods over traditional acidified media.[1,2,8]

OGYE contains yeast extract to provide essential vitamins and nutrients to promote fungal growth. Glucose acts as a carbon and energy source. Oxytetracycline is a selective agent used to inhibit the growth of lactobacilli and other bacteria commonly found in foods. Agar is the solidifying agent.

FORMULA

Ingredients per liter of deionized water:*

Glucose	20.0g
Oxytetracycline solution (100mg/mL)	10.0mL
Yeast Extract	5.0g
Agar	12.0g

Final pH 7.0 +/- 0.2 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, moisture, 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 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.

PROCEDURE

Refer to appropriate references for specific procedures on yeast and mold testing in foods.[1-9]

For the isolation and enumeration of yeasts and/or molds from foods:

1. Analyze an appropriate quantity of food sample according to the reference method.

2. Add the food sample to an appropriate volume of 0.1% peptone water to achieve a 10^{-1} dilution.

3. Homogenize the sample solution in a stomacher for approximately 2 minutes.

4. If indicated, make a series of 1:10 dilutions of the sample using 0.1% peptone water by aliquoting 10mL to a dilution vial (Cat. no D290).

5. Inoculate 0.1mL of the sample onto the agar surface. NOTE: Plate a series of dilutions if colony counts are unknown to ensure accurate counting.

6. Spread the inoculum evenly over the entire surface of the agar using a sterile bent glass rod or disposable spreader (Cat. no. 174CS01).

7. Incubate plates aerobically at 22 to 25ºC. Count the numbers of colony forming units (CFU) on plates containing 50 to 100 colonies after 5 days, or in any countable plates when aerial mycelia threaten to obscure further readings after 2 days.[1,2] Calculate the number of CFU by multiplying the number of colonies by the dilution factor and the volume plated. Record results as yeast and mold CFU per gram or mL of food.

INTERPRETATION OF RESULTS

Consult listed references for appropriate interpretation of results.[1-9]

Colonies should be apparent within five days of incubation. However, it is recommended plates be inspected beginning at 2 days for aerial mycelia that may threaten to obscure readings. Report counts as colony forming units (CFU) per gram or mL of sample.

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.

Lactic acid bacteria are inhibited on this medium.

Due to varying nutritional requirements, some strains may grow poorly or fail to grow on this medium.

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

MATERIALS REQUIRED BUT NOT PROVIDED

Standard microbiological supplies such as pipets (Cat no. 3301200 or Cat. no. 152143R) and equipment such as loops, swabs, applicator sticks (Cat. no. 174CS01), other culture media (Cat. no D290), incinerators, 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	
Aspergillus brasiliensis ATCC®16404	G	1-5 days	20-25°C	Aerobic	Growth
Saccharomyces cerevisiae ATCC® 9763	A	1-5 days	20-25°C	Aerobic	Growth
Candida albicans ATCC® 10231	A	1-5 days	20-25°C	Aerobic	Growth
Escherichia coli ATCC® 25922	B	24 hrs	30-35°C	Aerobic	Partial to complete inhibition

* 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

OGYE Agar should appear slightly hazy and light amber in color.

REFERENCES

1. Mossel D. A. A., Kleynen-Semmeling H.M., Vincentie H., Beerens H. and Catsaras M., 1970, *J. Appl. Bacteriol*., 33:454.

2. Mossel D. A. A., Visser M. and Mengerink W.H.J., 1962, *Lab. Pract.* 11:109.

3. Koburger J. A. and Mace F. E. 1967. *Proc. W. Va. Acad. Sci*. 39. 102-106.

4. Mossel D. A. A., Vega Clara L. and Put H. M. C. 1975. *J. Appl. Bact*. 39. 15-22.

5. Dijkmann K.E., Koopmans M. and Mossel D.A.A. 1979. *J. Appl. Bact*. 47. ix.

6. U.S. Food and Drug Administration. *Bacteriological Analytical Manual*.Arlington, VA http://www.fda.gov/Food/FoodScienceResearch/LaboratoryMethods/

7. International Organization for Standardization. 2004. Milk and Milk Products-Enumeration of Colony Forming Units of Yeasts and/or Molds-Colony Count Technique at 25ºC. ISO 6611/IDF 94, 2004-10-15, 2nd Ed. ISO, Geneva, Switzerland.

8. Beuchat and Cousin, 2001. *In* Downes and Ito (Ed.), Compendium of Methods for the Microbiological Examination of Foods, 4th Ed. American Public Health Association, Washington, D.C.

9. King Jr., Pitt, Beuchat and Corry, Series A: Life Sciences Vol. 122, 1986, Methods for the Mycological Examination of Food, Comparison of DRBC, OGY and PDA Media for Enumeration of Yeasts in Beverage Products, p.169.

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

HDQA 2207F [D]