IFU

Instructions for Use

2x YT BROTH

Cat. no. C9160	CulGenex™ 2xYT Broth	62gm
Cat. no. C9161	CulGenex™ 2xYT Broth	500gm
Cat. no. C9162	CulGenex™ 2xYT Broth	2kg
Cat. no. C9163	CulGenex™ 2xYT Broth	10kg

INTENDED USE

Hardy Diagnostics CulGenex™ 2xYT Broth is recommended for use as a general purpose growth medium for the cultivation of recombinant strains of *Escherichia coli* and the propagation of M13 bacteriophage.

SUMMARY

2xYT Agar (2x Yeast Extract Tryptone Agar) was developed for the growth and maintenance of recombinant strains of *Escherichia coli* used for genetic research.[1,3,5,6] The medium is also ideal for the propagation of M13 bacteriophage for sequencing and antibody phage display research.[3-5]

Antibody-based immunologic research is useful for identifying, isolating or eliminating cells with particular characteristics related to various diseases, and phage display is a useful technique for antibody selection for this purpose.[4] From these technologies, large and diverse libraries of recombinant antibodies can be generated, typically for molecules associated with pathological conditions, and screening by antibody phage display may lead to new and more potent drug discoveries.[2,4]

Components of 2xYT Agar provide nitrogen and growth factors that allow bacteriophage to reproduce in large quantities, without exhausting resources from the host cell. *E. coli* also grows well in this medium due to readily available amino acids, nucleotide precursors, vitamins and other metabolites present for cell growth.[1,5]

FORMULA*

Gram weight per liter:	31 gm/L
Tryptone	16.0gm
Yeast Extract	10.0gm
Sodium Chloride	5.0gm

Final pH 6.8 +/- 0.2 at 25ºC.

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

STORAGE AND SHELF LIFE

Store the sealed container(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 light beige.

Store the prepared culture media at 2-8ºC.

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 PRODUCT(S)

1. Suspend 31gm of the dehydrated culture medium in one liter of distilled or deionized water.

2. Heat to boiling and mix to dissolve completely.

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

4. Allow media to cool to 50ºC before adding antibiotics or additional supplements.

PROCEDURE AND INTERPRETATION OF RESULTS

For information on procedures and interpretation of results, consult listed references.[1-6]

LIMITATIONS

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 Certificates of Analysis (CofA).

Test Organisms	Inoculation Method*	Incubation			Results
		Time	Temperature	Atmosphere	
Escherichia coli ATCC® 23724	A	24-48 hrs	35°C	Aerobic	Growth

* 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

CulGenex™ 2XYT Broth powder should appear homogeneous, free-flowing, and light beige in color. The prepared medium should appear translucent, slightly opalescent, and light to medium amber in color.

REFERENCES

1. Ausubel, F.M., R. Brent, R.E. Kingston, D.D. Moore, J.G. Seidman, J.A. Smith, K. Struhl, Editors. 2010. *Current Protocols in Molecular Biology* . John Wiley and Sons, Inc. Malden, MA.

2. Bullock, G.R. and Petrusz, P. 1989. *Techniques in Immunocytochemistry*, Volumes 1, 2, 3 and 4, Academic Press, London.m

3. Cseke, L.J., P.B. Kaufman, G.K. Podila, and C.J. Tsai. 2004. *Handbook of Molecular and Cellular Methods in Biology and Medicine* . CRC Press.Taylor & Francis LLC. Boca Raton, FL.

4. O'Brien, P.M and R. Aitken. 2002. *Antibody Phage Display* . The Humana Press, Inc. Clifton, NJ.

5. Sambrook and Russell. 2001. *Molecular Cloning: A Laboratory Manual*, 3rd ed. Cold Spring Harbor Laboratory Press. Woodbury, New York.

6. Walker, J.M. 1984. *Methods in Molecular Biology* . The Humana Press, Inc. Clifton, NJ.

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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 2207C [D]