



Product Sheet

20H2 (ATCC® HB-323™)

Please read this FIRST



Intended Use

This product is intended for research use only. It is not intended for any animal or human therapeutic or diagnostic use.

Complete Growth Medium

RPMI 1640 medium with 0.05 mM 2-mercaptoethanol and 2 mM L-glutamine, 95%; fetal bovine serum, 5%

Citation of Strain

If use of this culture results in a scientific publication, it should be cited in that manuscript in the following manner: 20H2 (ATCC® HB-323™)

American Type Culture Collection
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Manassas, VA 20108 USA
www.atcc.org

800.638.6597 or 703.365.2700
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Or contact your local distributor

Description

Organism: *Rattus norvegicus* (B cell); *Mus musculus* (myeloma), rat (B cell); mouse (myeloma)

Strain:

Strain: Wistar (B cell); BALB/c (myeloma)

Isotype: IgG1; kappa light chain

Cell Type: hybridoma: B lymphocyte

Morphology: lymphoblast

Growth Properties: mixed: adherent and suspension, The cells are rather adherent in nature, and grow best if maintained in suspension culture plates (e.g., Corning Catalog number 430591).

Batch-Specific Information

Refer to the Certificate of Analysis for batch-specific test results.

SAFETY PRECAUTION

ATCC highly recommends that protective gloves and clothing always be used and a full face mask always be worn when handling frozen vials. It is important to note that some vials leak when submersed in liquid nitrogen and will slowly fill with liquid nitrogen. Upon thawing, the conversion of the liquid nitrogen back to its gas phase may result in the vessel exploding or blowing off its cap with dangerous force creating flying debris.

Unpacking & Storage Instructions

1. Check all containers for leakage or breakage.
2. Remove the frozen cells from the dry ice packaging and immediately place the cells at a temperature below -130°C, preferably in liquid nitrogen vapor, until ready for use.

Handling Procedure for Frozen Cells

Handling Procedure for Frozen Cells

To insure the highest level of viability, thaw the vial and initiate the culture as soon as possible upon receipt. If upon arrival, continued storage of the frozen culture is necessary, it should be stored in liquid nitrogen vapor phase and not at -70°C. Storage at -70°C will result in loss of viability.

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1. Thaw the vial by gentle agitation in a 37°C water bath. To reduce the possibility of contamination, keep the O-ring and cap out of the water. Thawing should be rapid (approximately 2 minutes).

2. Remove the vial from the water bath as soon as the contents are thawed, and decontaminate by dipping in or spraying with 70% ethanol. All of the operations from this point on should be carried out under strict aseptic conditions.

3. Transfer the vial contents to a 75 cm² tissue culture flask and dilute with the recommended complete culture medium (see the specific batch information for the recommended dilution ratio). It is important to avoid excessive alkalinity of the medium during recovery of the cells. It is suggested that, prior to the addition of the vial contents, the culture vessel containing the growth medium be placed into the incubator for at least 15 minutes to allow the medium to reach its normal pH (7.0 to 7.6).

4. Incubate the culture at 37°C in a suitable incubator. A 5% CO₂ in air atmosphere is recommended if using the medium described on this product sheet.

It is not necessary to remove the cryoprotective agent. If it is desired that the cryoprotective agent be removed immediately, or that a more concentrated cell suspension be obtained, centrifuge the cell suspension at approximately 125 xg for 5 to 10 minutes. Discard the supernatant and resuspend the cells with fresh growth medium at the dilution ratio recommended in the specific batch information.

Handling Procedure for Flask Cultures

Handling Procedure For Flask Cultures (Mixed: Adherent and Suspension)

The flask was seeded with cells (see specific batch information), grown, and completely filled with medium at ATCC to prevent loss of cells during shipping.

1. Upon receipt visually examine the culture for macroscopic evidence of any microbial contamination. Using an inverted microscope (preferably equipped with phase-contrast optics), carefully check for any



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evidence of microbial contamination

2. Incubate the flask in an upright position for several hours at 37°C. After the temperature has equilibrated, aseptically remove the entire contents of the flask and centrifuge at 125 xg for 5 to 10 minutes. Remove shipping medium and save for reuse. Resuspend the cell pellet in 10 mL of this medium and return the cells to the shipping flask
3. Incubate the culture, horizontally, at 37°C in a 5% CO₂ in air atmosphere. Maintain the cell density of the culture as suggested under the subculture procedure.



Subculturing Procedure

Medium Renewal: Every 2 to 3 days

Cultures can be maintained by addition of fresh medium or replacement of medium. Alternatively, cultures can be established by centrifugation of the suspension with subsequent resuspension in fresh medium. Add medium as the cell density increases.

Maintain cultures at cell concentrations between 4 X 10⁵ and 8 X 10⁵ viable cells/ml.



Cryopreservation Medium

Cryoprotectant Medium

Complete culture medium described above supplemented with an additional 5% fetal bovine serum and 5% (v/v) DMSO.

Cell culture tested DMSO is available as ATCC Catalog No. 4-X.



Comments

Animals were immunized with C57BL/6 thymocytes, boosted three times with the RBL-CD1.1 rat cell line (CD1.1-transfected), boosted once with BALB/cJ thymocytes and finally with the CR1-CD1.1 human cell line (CD1.1-transfected).

Spleen cells were fused with Sp2/0-Ag14 cells.

The antibody reacts with CD1.1, and cross-reacts to the same extent with CD1.2.

The antibody blocks recognition of CD1.1 by T cells in vitro and in vivo.

The antibody performs well in flow cytometry applications, and is also useful in immunoprecipitation applications.



References

References and other information relating to this product are available online at www.atcc.org.



Biosafety Level: 1

Appropriate safety procedures should always be used with this material. Laboratory safety is discussed in the current publication of the *Biosafety in Microbiological and Biomedical Laboratories* from the U.S. Department of Health and Human Services Centers for Disease Control and Prevention and National Institutes for Health.

ATCC Warranty

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Disclaimers

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the misidentification or misrepresentation of such materials.

Please see the enclosed Material Transfer Agreement (MTA) for further details regarding the use of this product. The MTA is also available on our Web site at www.atcc.org

Additional information on this culture is available on the ATCC web site at www.atcc.org.

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