



Product Sheet

NCI-H676B [H676B] (ATCC® HTB-179™)

Please read this FIRST



Storage Temp.
liquid nitrogen
vapor phase



Biosafety Level
1

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

ACL-4 medium (serum-free)

The base medium for this cell line is ATCC formulated DMEM: F12 Medium Catalog No. 30-2006. To make the complete growth medium, add the following components to the base medium:

- 0.02 mg/ml insulin
- 0.01 mg/ml transferrin
- 25 nM sodium selenite (final conc.)
- 50 nM Hydrocortisone (final conc.)
- 1 ng/ml Epidermal Growth Factor (do not filter)
- 0.01 mM ethanolamine (final conc.)
- 0.01 mM phosphorylethanolamine (final conc.)
- 100 pM triiodothyronine (final conc.)
- 0.5% (w/v) bovine serum albumin (final conc.)
- 10 mM HEPES
- 0.5 mM sodium pyruvate (final conc.)
- extra 2mM L-glutamine (for final conc. of 4.5mM)

Citation of Strain

If use of this culture results in a scientific publication, it should be cited in that manuscript in the following manner: NCI-H676B [H676B] (ATCC® HTB-179™)

Description

Organism: *Homo sapiens*, human
Tissue: lung; Derived from metastatic site: pleural effusion
Disease: adenocarcinoma
Age: 63 years
Gender: male
Morphology: epithelial
Growth Properties: suspension, multicell aggregates
Isoenzymes:
AK-1, 2
ES-D, 1
G6PD, B
GLO-I, 1-2
Me-2, 0
PGM1, 1-2
PGM3, 2
DNA Profile:
Amelogenin: X
CSF1PO: 9
D13S317: 10
D16S539: 11,12
D5S818: 11
D7S820: 12
THO1: 9.3
TPOX: 8,11
vWA: 16,19

Cytogenetic Analysis: This is a hypodiploid cell line; modal number = 43; range = 39 to 47. Marker chromosomes consisted of over 70% of the chromosomes in each cell complement. Among the identifiable markers were t(1q8q), der(3)t(3;?)(q29;?), t(2q7q), der(9)t(9;14)(p13;q11) and der(11)t(?q23;?). All markers had a single copy per cells. The N5, N9, N10, N17, N18, N20, N21 and the X were the only structurally normal chromosomes found. Except for the paired N5, all others had single copies per cell.

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



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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 centrifuge tube containing 9.0 mL complete culture medium, and spin at approximately 125 xg for 5 to 7 minutes.
4. Resuspend cell pellet with the recommended complete medium (see the specific batch information for the culture recommended dilution ratio), and dispense into a **25 cm²** culture flask. 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 complete 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).
5. 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.



Handling Procedure for Flask Cultures

Handling Procedure For Flask Cultures

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 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.
3. Incubate the culture, horizontally, at 37°C in a 5% CO₂ in air atmosphere..



Subculturing Procedure

Protocol: This line grows as aggregates of cells in suspension. Culture can be maintained by addition of medium or by replacement of medium. Alternatively, the cells may be collected by centrifugation and dispersed into fresh medium.

Subcultivation Ratio: A subcultivation ratio of 1:2 to 1:5 is recommended

Medium Renewal: 2 to 3 times per week



Cryopreservation Medium

Cryoprotectant Medium

Complete culture medium described above supplemented with 10% FBS and 7.5% (v/v) DMSO. Cell culture tested DMSO is available as ATCC Catalog No. 4-X.



Comments

The NCI-H676B cell line was derived by A.F. Gazdar and H. Oie in 1984 from the pleural fluid of a patient with adenocarcinoma of the lung taken prior to treatment.

The cells produce an abnormal size p53 mRNA (2.3 kb) as well as the normal size mRNA (2.8 kb).

The line does not exhibit any gross structural DNA abnormalities.

The cells produce mucin, and occasionally signet ring cells are formed.



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

ATCC® products are warranted for 30 days from the date of shipment, and this warranty is valid only if the product is stored and handled according to the information included on this product information sheet. If the ATCC® product is a living cell or microorganism, ATCC lists the media formulation that has been found to be effective for this product. While other, unspecified media may also produce satisfactory results, a change in media or the absence of an additive from the ATCC recommended media may affect recovery, growth and/or



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function of this product. If an alternative medium formulation is used, the ATCC warranty for viability is no longer valid.

Disclaimers

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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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