

HTB-171[™]

Description

NCI-H446 [H446] cells were isolated in 1982 from the pleural fluid of a 61-year-old,

White, male patient with small cell cancer of the lung.

Organism: Homo sapiens, human

Tissue: Lung
Age: 61 years
Gender: Male

Morphology: epithelial

Growth properties: Mixed: adherent and suspension

Disease: Carcinoma

Storage Conditions

Product format: Frozen

Storage conditions: Vapor phase of liquid nitrogen

Intended Use

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

BSL₁

ATCC determines the biosafety level of a material based on our risk assessment as guided by the current edition of *Biosafety in Microbiological and Biomedical Laboratories* (*BMBL*), U.S. Department of Health and Human Services. It is your responsibility to understand the hazards associated with the material per your organization's policies



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and procedures as well as any other applicable regulations as enforced by your local or national agencies.

ATCC highly recommends that appropriate personal protective equipment is always used when handling vials. For cultures that require storage in liquid nitrogen, it is important to note that some vials may 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 vial exploding or blowing off its cap with dangerous force creating flying debris. Unless necessary, ATCC recommends that these cultures be stored in the vapor phase of liquid nitrogen rather than submersed in liquid nitrogen.

Certificate of Analysis

For batch-specific test results, refer to the applicable certificate of analysis that can be found at www.atcc.org.

Handling Procedures

Unpacking and 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.

Complete medium: The base medium for this cell line is ATCC-formulated RPMI-1640 Medium, ATCC 30-2001. To make the complete growth medium, add the following components to the base medium: fetal bovine serum (ATCC 30-2020) to a final concentration of 10%.

Handling Procedure:



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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.

- 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 an appropriate size vessel. 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.

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 x g 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.

Subculturing procedure:

This is a cell line that grows as both attached and suspended cells. The suspended cells are viable and can be used for subculture.

To subculture the attached cells:

- 1. Remove and discard culture medium. Keep the floating cells.
- 2. Rinse cells with PBS (ATCC 30-2101) to remove serum that contains trypsin inhibitor.
- 3. Once PBS is removed, add 0.25 trypsin, 0.53 mM EDTA (ATCC 30-2101), and incubate for 2-3 min at 37C (recommend starting with 2mL in a T25 and 3 mL in a T75 or 0.5mL/cm²).
- 4. Neutralize with the same volume of culture medium used for the Trypsin-EDTA

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solution.

- 5. Transfer cell suspension to a centrifuge tube and spin at approximately 125xg for 5 to 10 minutes. Discard the supernatant.
- 6. Resuspend the cell pellet in fresh growth media. Add appropriate aliquots to new culture vessels.
- 7. Incubate cultures at 37C.

Note: Maintain the floating population, failure to do so may create selective pressure on the culture.

Subcultivation Ratio: A subcultivation ratio of 1:3 to 1:9 is recommended

Medium Renewal: 2 to 3 times per week

Reagents for cryopreservation: Complete growth medium supplemented with 5%

(v/v) DMSO (ATCC 4-X)

Material Citation

If use of this material results in a scientific publication, please cite the material in the following manner: NCI-H446 [H446] (ATCC HTB-171)

References

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

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