



Calu-3

HTB-55™

Description

Calu-3 epithelial cells are isolated from lung tissue derived from a 25-year-old, White, male patient with lung adenocarcinoma who received prior therapy with cytoxan, bleomycin, and adriamycin. This cell line is valuable for SARS-CoV-2 propagation in vitro, is a suitable transfection host, and has applications in cancer and toxicology research.

Organism: *Homo sapiens*, human

Cell Type: epithelial cell

Tissue: Lung

Age: 25 years

Gender: Male

Morphology: epithelial

Growth properties: Adherent

Disease: Adenocarcinoma

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.

BSL 1

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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 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.

Growth Conditions

Temperature: 37°C

Atmosphere: 95% Air, 5% CO₂

Handling Procedures

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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 Eagle's Minimum Essential Medium, (ATCC 30-2003). To make the complete growth medium, add the following components to the base medium: fetal bovine serum to a final concentration of 10%.

Media preparation:

- 500 mL EMEM (ATCC 30-2003)
- 56 mL FBS (ATCC 30-2020) – not heat-inactivated

Handling Procedure:

To ensure 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 a centrifuge tube containing 9.0 mL complete culture medium and centrifuge at approximately $250 \times g$ for 10 minutes.
4. Resuspend cell pellet with the recommended complete growth medium (see the lot information on Certificate of Analysis (COA) for the culture recommended dilution ratio) and dispense into a 25 cm^2 or a 75 cm^2 culture flask as recommended on the COA. The recommended seeding density for HTB-55 is 2.0×10^4 to 4.0×10^4 viable cells/ cm^2 .
5. Incubate the culture at **37°C** in a suitable incubator. A 5% CO_2 in air atmosphere is recommended if using the medium described on this product sheet.

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Comment: HTB-55 cells may grow in patches that eventually spread out to form a monolayer.

Subculturing procedure:

1. Remove and discard culture medium.
2. Briefly rinse the cell layer with 0.25% (w/v) Trypsin- 0.53 mM EDTA solution to remove all traces of serum that contains trypsin inhibitor.
3. Add 2.0 to 3.0 mL of Trypsin-EDTA solution to flask and observe cells under an inverted microscope until cell layer is dispersed (usually within 5 to 15 minutes).

Note: To avoid clumping do not agitate the cells by hitting or shaking the flask while waiting for the cells to detach. Cells that are difficult to detach may be placed at 37°C to facilitate dispersal.

Note: HTB-55 cells may take as long as 30 to 45 minutes to dissociate.

Dissociation is enhanced by subculturing prior to 100% confluence.

4. Add 6.0 to 8.0 mL of complete growth medium and aspirate cells by gently pipetting.
5. Add appropriate aliquots of the cell suspension to new culture vessels. Corning[®] T-75 flasks (catalog #430641) are recommended for subculturing this product.
6. Incubate cultures at 37°C.

Subcultivation Ratio/ Subculture seeding density: A subcultivation ratio of 1:3 to 1:6 is recommended or a subculture seeding density 1.0×10^4 to 4.0×10^4 viable cells/cm².

Note: Unless floating cells have been verified to be non-viable via a trypan blue or some other sort of viability assay, floating cells should be kept with the main culture.

Medium Renewal: 2 to 3 times per week

Material Citation

If use of this material results in a scientific publication, please cite the material in the following manner: Calu-3 (ATCC HTB-55)

References

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

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