



BBM

CRL-9482™

Description

Organism: *Homo sapiens*, human

Cell Type: epithelial cell

Tissue: Lung; Bronchus

Morphology: epithelial

Growth properties: Adherent

Disease: Carcinogen

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Patent number:

4,885,238

Technical information: ATCC Product Experience does not have technical information on patent deposits that are not produced or characterized by ATCC. Additional information can be found in the corresponding patent available from the patent holder or with the U.S. and/or international patent office.

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 2

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.

Cells contain polyomaviral DNA sequences

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

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 (BEBM) along with all the additives can be obtained from Lonza/Clonetics Corporation as a kit: BEGM, Kit Catalog No. CC-3170. ATCC does not use the GA-1000 (gentamycin-amphotericin B mix) provided with the BEGM kit. Note: Do not filter complete medium.

Handling Procedure:

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.

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.

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 spin at approximately 125 xg for 5 to 7 minutes.
4. Re-suspend cell pellet with fresh medium at the dilution ratio recommended in the specific batch information.. The flasks used should be **precoated with a mixture of 0.01 mg/ml fibronectin, 0.03 mg/ml bovine collagen type I and 0.01 mg/ml bovine serum albumin dissolved in BEBM medium.**
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.

Subculturing procedure:**Protocol:**

- Remove and discard culture medium.
- Briefly rinse the cell layer with 0.25% (w/v) Trypsin- 0.53 mM EDTA solution containing 0.5%(w/v) polyvinylpyrrolidone (PVP).
- Add 2.0 to 3.0 ml of Trypsin-EDTA-PVP 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.

- Add 6.0 to 8.0 ml of complete growth medium and aspirate cells by gently pipetting.
- To remove trypsin-EDTA solution, transfer cell suspension to centrifuge tube and spin at approximately 125 xg for 5 to 10 minutes. Discard supernatant and resuspend cells in fresh growth medium. Add appropriate aliquots of cell suspension to new culture vessels.
- Incubate cultures at 37°C.

Subcultivation Ratio: Inoculate new flasks at 1500 to 3000 cells per sq. cm.

Medium Renewal: Two to three times weekly

Reagents for cryopreservation: Leibovitz's L-15 medium with 2 mM L-glutamine supplemented with 10 mM HEPES, 1% PVP, 10% (v/v) fetal bovine serum and 7.5% (v/v) DMSO

Material Citation

If use of this material results in a scientific publication, please cite the material in the following manner: BBM (ATCC CRL-9482)

References

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

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