



BEAS-2B

CRL-3588™

Description

BEAS-2B are epithelial cells isolated from normal human bronchial epithelium derived from autopsies of noncancerous individuals. The cells retain the ability to undergo squamous differentiation in response to serum and can be used to screen chemical and biological agents for the ability to induce or affect differentiation and/or carcinogenesis. The BEAS-2B cell line is a suitable transfection host. This product is an ATCC manufactured progeny of CRL-9609 cited in US Pat. No. 4,885,238.

Organism: *Homo sapiens*, human

Cell Type: epithelial cell

Tissue: Lung; Bronchus

Morphology: epithelial

Growth properties: Adherent

Disease: Normal

Volume: 1.0 mL

Patent number:

4,885,238

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.

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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

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Handling Procedures

Complete medium: The complete medium for this cell line is Airway Epithelial Cell Basal Medium (PCS-300-030) and Bronchial Epithelial Cell Growth Kit (PCS-300-040). The protocol for mixing the complete growth medium can be found on the respective product pages.

Handling Procedure:

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.

Note: The culture flasks used should be pre-coated 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 (see references : U.S. Pat. 4,885,238 and Lechner, J.F. and LaVeck, M.A. A serum-free method for culturing normal human bronchial epithelial cells at clonal density. J. Tissue Culture Methods 9: 43-48, 1985).

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 the cell suspension at approximately $125 \times g$ for 5 to 10 minutes. Discard the supernatant.
4. Resuspend the cell pellet in the complete culture medium at the dilution ratio recommended in the specific batch information and dispense into a pre-coated T-25 cm^2 culture flask.
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.

Subculturing procedure:

These cells should be **subcultured before reaching confluence** since confluent cultures rapidly undergo squamous terminal differentiation. Volumes used in this

protocol are for 75 cm² flask; proportionally reduce or increase amount of dissociation medium for culture vessels of other sizes. Corning[®] T-75 flasks (catalog #430641) are recommended for subculturing this product.

1. Remove and discard culture medium.
2. Add 2.0 to 3.0 mL of 0.25% Trypsin - 0.53mM EDTA solution containing 0.5% polyvinylpyrrolidone (PVP) to flask and observe cells under an inverted microscope until cell layer is dispersed (usually with 5 to 10 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.
3. Add 6.0 to 8.0 mL of complete growth medium and aspirate cells by gently pipetting.
4. Transfer cell suspension to centrifuge tube and spin at approximately 125 x g for 5 to 10 minutes
5. Discard supernatant and resuspend cells in fresh growth medium. Inoculate new flasks at 1500 to 3000 cells per cm². The culture flasks used should be pre-coated with a mixture of 0.01mg/ml fibronectin, 0.03 mg/ml bovine collagen type I and 0.01 mg/mL bovine serum albumin dissolved in BEBM.
6. Place culture flasks in incubators at 37°C.

Note: For more information on enzymatic dissociation and subculturing of cell lines consult Chapter 10 in *Culture of Animal Cells, a manual of Basic Technique* by R. Ian Freshney, 3rd edition, published by Alan R. Liss, N.Y., 1994.

Interval: Subcultured before reaching confluence.

Medium Renewal: Every 2 to 3 days

Flask Coating

1. Prepare a mixture of 0.01 mg/mL fibronectin, 0.03 mg/mL bovine collagen type I and 0.01 mg/mL bovine serum albumin (BSA) dissolved in culture medium. Store pre-prepared Coating Solution at 4°C in cold room for up to 3 months.
2. For a growth area of 75 cm², add 4.5 mL of the fibronectin/collagen/BSA solution and rock gently to coat the entire surface.
3. Incubate the freshly coated vessel(s) in a 37°C incubator overnight (it is preferable to use tissue culture vessels with tightened, plug-seal caps to prevent evaporation during the coating process).
4. Store coated flasks with solution at room temperature, light protected, up to 1

month. Suction off solution before plating cells.

Reagents for cryopreservation: Complete growth medium supplemented with 1% PVP and 7.5% DMSO

Material Citation

If use of this material results in a scientific publication, please cite the material in the following manner: BEAS-2B (ATCC CRL-3588)

References

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

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