



# 184B5

CRL-8799™

## Description

**Tissue:** Breast; Mammary gland

**Age:** 21 years

**Gender:** Female

**Morphology:** epithelial

**Growth properties:** Adherent

**Disease:** Normal

**Patent depository:** This material was deposited with the ATCC Patent Depository to fulfill U.S. or international patent requirements. This material may not have been produced or characterized by ATCC. As an International Depository Authority (IDA) for patent deposits, ATCC is required to complete viability testing only at time of initial deposit of patent material. Patent deposits are made available on behalf of the Depositor when the pertinent U.S. or international patent is issued, but material may not be used to infringe the patent claims.

**Patent number:**

4,808,532

**Technical information:** ATCC Technical Services 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.

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## Storage Conditions

**Product format:** Frozen

**Storage conditions:** Vapor phase of liquid nitrogen

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## 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

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diagnostic use.

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### BSL 1

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.

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### Certificate of Analysis

For batch-specific test results, refer to the applicable certificate of analysis that can be found at [www.atcc.org](http://www.atcc.org).

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### Growth Conditions

**Temperature:** 37°C

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## 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  $-130^{\circ}\text{C}$ , preferably in liquid nitrogen vapor, until ready for use.

**Complete medium:** The base medium for this cell line (MEBM) along with the additives can be obtained from Lonza/Clonetics Corporation as a kit: MEGM, Kit Catalog No. CC-3150. ATCC does not use the GA-1000 (gentamycin-amphotericin B mix) provided with kit. To make the complete growth medium, you will need to add the following components to the kit (sold separately):

1 ng/ml cholera toxin

Note: Do not filter complete medium

### 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  $-70^{\circ}\text{C}$ . Storage at  $-70^{\circ}\text{C}$  will result in loss of viability.

1. Thaw the vial by gentle agitation in a  **$37^{\circ}\text{C}$**  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 Transfer to a  $75 \text{ cm}^2$  tissue culture flask and dilute with the recommended complete culture medium (see the specific batch information for the recommended dilution ratio). 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).

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5. Incubate the culture at **37°C** in a suitable incubator. A 5% CO<sub>2</sub> in air atmosphere is recommended if using the medium described on this product sheet.

**Subculturing procedure:** Volumes are given for a 75 cm<sup>2</sup> flask. Increase or decrease the amount of dissociation medium needed proportionally for culture vessels of other sizes.

1. Remove and discard culture medium.
2. Briefly rinse the cell layer with 0.25% (w/v) Trypsin- 0.53 mM EDTA solution.
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.

4. Add 6.0 to 8.0 mL of complete growth medium and aspirate cells by gently pipetting.
5. To remove trypsin-EDTA solution, transfer cell suspension to centrifuge tube and spin at approximately 125 x g for 5 to 10 minutes. Discard supernatant and resuspend cells in fresh serum-free growth medium. Add appropriate aliquots of cell suspension to new culture vessels.
6. Place culture vessels in incubators at 37°C.

**Subcultivation Ratio:** A subcultivation ratio of 1:2 to 1:6 is recommended

**Medium Renewal:** Every 2 to 3 days

**Reagents for cryopreservation:** Complete growth medium described above supplemented with 10% FBS and 5% (v/v) DMSO.

Cell culture tested DMSO is available as ATCC Catalog No. 4-X.

**Note:** Lots manufactured prior to 05/20/2020 may have used a different cryopreservative, contact technical service if needed.

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### Material Citation

If use of this material results in a scientific publication, please cite the material in the following manner: 184B5 (ATCC CRL-8799)

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### References

References and other information relating to this material are available at [www.atcc.org](http://www.atcc.org).

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**Revision**

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**Contact Information**

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