RPTEC/TERT1

CRL-4031™

Description

RPTEC/TERT1 is an hTERT-immortalized epithelial cell that was isolated from the proximal tubule of a male patient. This cell line was deposited by R Grillari-Voglauer in 2004 and can be used in toxicology research.

- **Organism** *Homo sapiens*, human
- **Cell Type** epithelial cell
- **Tissue** kidney; Cortex; Epithelium; Proximal tubule
- **Age** adult
- **Gender** Male
- **Morphology** Epithelial-like
- **Growth properties** Adherent
- **Volume** 1.0 mL

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 SV40 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% CO_2

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 DMEM:F12 Medium (ATCC® 30-2006™).

To make the complete growth medium, add hTERT RPTEC Growth Kit (ATCC® ACS-4007™) to the base medium. The final concentration for each growth kit component in the complete hTERT immortalized RPTEC growth medium is as follows:

- 5 pM triiodo-L-thyronine
- 10 ng/mL recombinant human EGF
- 3.5 µg/mL ascorbic acid
- 5.0 µg/mL human transferrin
- 5.0 µg/mL insulin
- 25 ng/mL prostaglandin E_1
- 25 ng/mL hydrocortisone
- 8.65 ng/mL sodium selenite
- 1.2 mg/mL sodium bicarbonate

Required but not supplied: G418 solution MUST be added to the above medium to a final concentration of 0.1 mg/mL G418 to maintain the selective pressure for immortalization

Note: Do not filter complete medium.

This medium is formulated for use with a 5% CO_2 in air atmosphere.

- **Handling Procedure**

To ensure the highest level of viability, thaw the vial and initiate the culture as soon as possible upon receipt. If storage of the frozen culture is necessary upon arrival, store the vial in liquid nitrogen vapor phase and **NOT** at -70°C. Storage at -70°C will result in loss of viability.

1. Prepare a 25 cm^2 or a 75 cm^2 culture flask containing the recommended complete culture medium. Prior to the addition of the vial contents, the vessel containing the growth medium should be placed in the incubator for at least 15 minutes to allow the medium to reach its normal pH (7.0 to 7.6) and to avoid excessive alkalinity of the medium during recovery of the cells.
2. 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).
3. 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 operations from this point on should be carried out under strict aseptic conditions.
4. Transfer the vial contents to a centrifuge tube containing 9.0 mL of complete culture medium and centrifuge the cell suspension at approximately 250 x *g* for 5 to 7 minutes.
5. Discard the supernatant and resuspend the cells in fresh growth medium (see the batch-specific information for the recommended dilution ratio). Add this suspension to the prepared culture vessel.
6. Incubate the culture at 37°C in a suitable incubator.
7. A 5% CO_2/95% air atmosphere is recommended if using the medium described on this product sheet.

- **Subculturing procedure**
  Volumes are given for a 75 cm^2 flask; proportionally reduce or increase amount of dissociation medium for culture vessels of other sizes.
  1. Subculture when the culture is about 90% confluence. Expected cell yield is between 1.5 x 10^5 and 2 x 10^5 viable cells/cm^2.
  2. Remove and discard culture medium.
  3. Add 2.0 to 3.0 mL of Trypsin-EDTA for Primary Cells (ATCC PCS-999-003) to the flask and observe cells under an inverted microscope until the cell layer is dispersed (usually within 3 to 8 minutes). **Note:** To avoid clumping do not agitate the cells by hitting or shaking the flask while waiting for the cells to detach. Place at 37°C to facilitate dispersal.
  4. To stop trypsinization, add 2.0 to 3.0 mL of 0.1% Soybean Trypsin Inhibitor and aspirate cells by gently pipetting.
  5. Transfer cell suspension to a 15-mL centrifuge tube and spin at approximately 250 x *g* for 5 to 10 minutes.
  6. Discard supernatant and resuspend cells in fresh growth medium. Add appropriate aliquots of the cell suspension to new culture vessels. An inoculum of 4 to 6 x 10^4 viable cells/cm^2 is recommended.
  7. Incubate cultures at 37°C.

  Subcultivation ratio: A subcultivation ratio of 1:3 to 1:4 is recommended.
  Medium renewal: 2 to 3 times weekly
  Note: For more information on enzymatic dissociation and subculturing of cell lines consult Chapter 13 in Culture Of Aminal Cells: A Manual of Basic Techniques by R. Ian Freshney, 5th edition, published by Wiley-Liss, N.Y., 2005.
- **Reagents for cryopreservation** Complete culture medium + 10% DMSO
  Storage temperature: liquid nitrogen vapor phase

Material Citation

If use of this material results in a scientific publication, please cite the material in the following manner: RPTEC/TERT1 (ATCC CRL-4031)

References

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

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