ATCC

Product Sheet

# RPTEC/TERT1 OAT3 (ATCC® CRL-4031-OAT3™)

**Please read this FIRST**

Storage Temp.
**liquid nitrogen vapor phase**

Biosafety Level
**2**

## Intended Use

This product is intended for research use only. It is not intended for any animal or human therapeutic or diagnostic use.

## Complete Growth Medium

The base medium for this cell line is DMEM/F-12 (ATCC 30-2006). To make the complete medium add the following components:

- hTERT Immortalized RPTEC Growth Kit (ATCC ACS-4007) with Supplement A (5 ml) and Supplement B (8ml)
- 0.3 µg/mL puromycin

## Citation of Strain

If use of this culture results in a scientific publication, it should be cited in that manuscript in the following manner: RPTEC/TERT1 OAT3 (ATCC® CRL-4031-OAT3™)

American Type Culture Collection
PO Box 1549
Manassas, VA 20108 USA
www.atcc.org

800.638.6597 or 703.365.2700
Fax: 703.365.2750
Email: Tech@atcc.org

*Or contact your local distributor*

## Description

**Organism:** *Homo sapiens*, human
**Tissue:** kidney: renal cortex; proximal tubules, epithelium
**Disease:** normal
**Morphology:** epithelial
**Growth Properties:** adherent
**DNA Profile:**
Amelogenin: X,Y
CSF1PO: 11
D13S317: 11,13
D16S539:11,12
D5S818: 9,11
D7S820: 10
THO1: 9,9.3
TPOX: 8,11
vWA: 16,18
**Cytogenetic Analysis:** normal diploid; abnormal pseudodiploid; abnormal near-diploid

## Batch-Specific Information

Refer to the Certificate of Analysis for batch-specific test results.

## 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.

## Unpacking & 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.

## Handling Procedure for Frozen Cells

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. Initial seeding density is 1x $10^4$ and 2x$10^4$ viable cells/$cm^2$. 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).
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 x *g* for 5 to 7 minutes.
4. Resuspend cell pellet with the recommended complete medium. 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 complete 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). pH (7.0 to 7.6).
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

Volumes used in this protocol are for 75 $cm^2$ 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. Briefly rinse the cell layer with Phosphate Buffered Saline (PBS; ATCC 30-2200) solution to remove all

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traces of serum that contains trypsin inhibitor.

3. Add 2.0 to 3.0 mL of Trypsin-EDTA for Primary Cells (ATCC PCS-999-003) 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. 0.1% Soybean Trypsin Inhibitor (ATCC 30-2104) is then used to neutralize the trypsin. After neutralization, aspirate cells by gently pipetting.
5. Remove Trypsin Inhibitor via centrifugation.
6. Resuspend the cells with complete growth medium.
7. Add appropriate aliquots of the cell suspension to new culture vessels.
   Cultures can be established between 1.0 x $10^4$ and 2.0 x $10^4$ viable cells/$cm^2$.
8. Incubate cultures at 37°C.

**Interval:** Maintain cultures at a cell concentration between 1.1X $10^4$ and 1.5 X $10^5$ cell/$cm^2$.
**Subcultivation Ratio:** A subcultivation ratio of 1:3 to 1:8 is recommended
**Medium Renewal:** 2 to 3 times per week

## Cryopreservation Medium

complete growth media + 10% DMSO (ATCC 4-X)

## Comments

RPTEC/TERT1 cell line stably expressing SLC transporter hOAT3(SLC22A8)

- Marker Testing (ICC) - > 75% positive for OAT3, E-Cadherin, CD13
- Dome formation – positive, comparable to parental line
- RT PCR OAT3 Expression Assay – one band at 1.6 kb
- Telomerase Activities (TRAP Assay) – Positive, ≥ 4 repeats
- OAT3 copy number confirmed via ddPCR – 7 copies (parental line has 2 copies)

## References

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

## Biosafety Level: 2

Appropriate safety procedures should always be used with this material. Laboratory safety is discussed in the current publication of the *Biosafety in Microbiological and Biomedical Laboratories* from the U.S. Department of Health and Human Services Centers for Disease Control and Prevention and National Institutes for Health.

## ATCC Warranty

ATCC® products are warranted for 30 days from the date of shipment, and this warranty is valid only if the product is stored and handled according to the information included on this product information sheet. If the ATCC® product is a living cell or microorganism, ATCC lists the media formulation that has been found to be effective for this product. While other, unspecified media may also produce satisfactory results, a change in media or the absence of an additive from the ATCC recommended media may affect recovery, growth and/or function of this product. If an alternative medium formulation is used, the ATCC warranty for viability is no longer valid.

## Disclaimers

This product is intended for laboratory research purposes only. It is not intended for use in humans.
While ATCC uses reasonable efforts to include accurate and up-to-date information on this product sheet, ATCC makes no warranties or representations as to its accuracy. Citations from scientific literature and patents are provided for informational purposes only. ATCC does not warrant that such information has been confirmed to be accurate.
This product is sent with the condition that you are responsible for its safe storage, handling, and use. ATCC is not liable for any damages or injuries arising from receipt and/or use of this product. While reasonable effort is made to insure authenticity and reliability of materials on deposit, ATCC is not liable for damages arising from the misidentification or misrepresentation of such materials.
Please see the enclosed Material Transfer Agreement (MTA) for further details regarding the use of this product. The MTA is also available on our Web site at www.atcc.org

Additional information on this culture is available on the ATCC web site at www.atcc.org.

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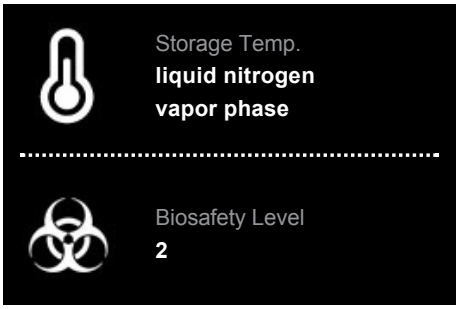


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