ATCC

Product Sheet

RTG-P1 (ATCC® CRL-2829™)

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

Leibovitz's L-15 medium with 2 mM L-glutamine supplemented with 0.2 mg/ml G418 and 10% fetal bovine serum.
(Note: The L-15 medium formulation was devised for use in a free gas exchange with atmospheric air. A CO2 and air mixture is detrimental to cells when using this medium for cultivation)

Citation of Strain

If use of this culture results in a scientific publication, it should be cited in that manuscript in the following manner: RTG-P1 (ATCC® CRL-2829™)

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: *Oncorhynchus mykiss*, trout, rainbow
Tissue: mixed; testis; ovary
Gender: male and female mixed
Morphology: fibroblast
Growth Properties: adherent

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

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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.

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1. Thaw the vial by gentle agitation in a **22°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 (see the specific batch information for the culture recommended dilution ratio) and dispense into a 25 cm^2 or a 75 cm^2 culture flask. 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).
5. Incubate the culture at **22°C** in a suitable incubator. A free gas exchange with **atmospheric air** is recommended if using the medium described on this product sheet.

Subculturing Procedure

Protocol:

1. Remove and discard culture medium.
2. Briefly rinse the cell layer with 0.25% (w/v) Trypsin - 0.53 mM EDTA solution to remove all traces of serum that contains trypsin inhibitor.
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.
4. Add 6.0 to 8.0 ml of complete growth medium and aspirate cells by gently pipetting.
5. Add appropriate aliquots of the cell suspension to new culture vessels.
   An inoculum of 6 X 10(3) to 1 X 10(4) viable cells/sq. cm is recommended.

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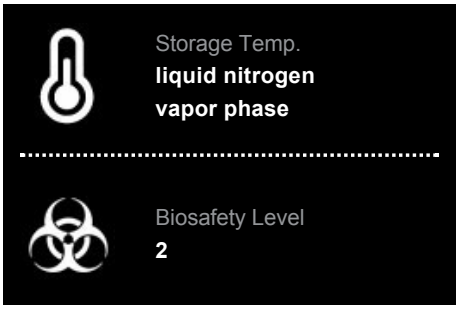

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6. Incubate cultures at 22C.

Interval: Maintain cultures at a cell concentration between 1 X 10(4) and 1 X 10(5) cells/sq. cm
Subcultivation Ratio: A subcultivation ratio of 1:3 to 1:4 is recommended
Medium Renewal: Two to three times weekly

Cryopreservation Medium

Cryoprotectant Medium
Complete growth medium described above supplemented with 5% (v/v) DMSO.
Cell culture tested DMSO is available as ATCC® Catalog No. 4-X.

Comments

The RTG-2 cell line (ATCC CCL-55) was transfected with a trout Mx1 promoter-luciferase construct and selected to produce a stable cell line, RTG-P1 [PubMed: 15043947]. The vector, pGL3-prMx1-Basic-Neo, expresses the firefly luciferase gene under the control of the promoter for the IFN-induced gene Mx1. The vector contains SV40 viral DNA sequences and the neomycin resistance gene. Luciferase expression is stable and highly induced by polyinosinic:polycytidylic acid (poly I:C) in RTG-P1 cells [PubMed: 15043947]. This cell line can be used to study the activation of the IFN pathway. The RTG-P1 reporter system constitutes an interesting tool to study the induction and regulation of IFN signaling in teleost fish.

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

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Disclaimers

This product is intended for laboratory research purposes only. It is not intended for use in humans.
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Additional information on this culture is available on the ATCC web site at www.atcc.org.