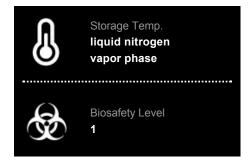


Product Sheet

PC-12 (ATCC® CRL-1721[™])

Please read this FIRST



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 ATCC-formulated RPMI-1640 Medium, Catalog No. 30-2001. To make the complete growth medium, add the following components to the base medium:

- heat-inactivated horse serum to a final concentration of 10%
- fetal bovine serum to a final concentration of 5%

Citation of Strain

If use of this culture results in a scientific publication, it should be cited in that manuscript in the following manner: PC-12 (ATCC® CRL-1721™)

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

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Description

Organism: Rattus norvegicus, rat

Tissue: adrenal gland Disease: pheochromocytoma

Gender: male

Morphology: small irregularly shaped cells

Growth Properties: floating clusters; few scattered lightly attached cells. Cytogenetic Analysis: 40 chromosomes; 38 autosomes plus XY



Batch-Specific Information

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



A 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.



Frozen Cells Handling Procedure for Frozen Cells

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.

- 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 50 ml tube containing 9 mL complete growth medium. Centrifuge cells at 180 - 225 x g for 8-15 minutes at room temperature. Remove and discard supernatant. Resuspend cells in 5 mL complete growth medium. Break up cell clusters by gently aspirating cells through a 22g needle 4 or 5 times. (see the specific batch information for the culture 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 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).
- 4. Incubate the culture at 37°C in a suitable incubator. A 5% CO2 in air atmosphere is recommended if using the medium described on this product sheet.



Handling Procedure for Flask Cultures

The flask was seeded with cells (see specific batch information) grown and completely filled with medium at ATCC to prevent loss of cells during shipping.

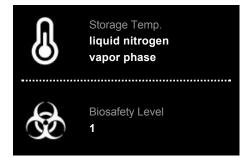
- 1. Upon receipt visually examine the culture for macroscopic evidence of any microbial contamination. Using an inverted microscope (preferably equipped with phase-contrast optics), carefully check for any evidence of microbial contamination
- 2. Incubate the flask in an upright position for several hours at 37°C. After the temperature has equilibrated, aseptically remove the entire contents of the flask and centrifuge at 125 xg for 5 to 10 minutes. Remove shipping medium and save for reuse. Resuspend the cell pellet in 10 ml of this medium.
- 3. From this cell suspension remove a sample for a cell count and viability. Adjust the cell density of the suspension to 5 x 10⁵ viable cells/ml in the shipping medium.
- 4. Incubate the culture, horizontally, at 37°C in a 5% CO₂ in air atmosphere. Maintain the cell density of the culture as suggested under the subculture procedure.



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Subculturing Procedure

Protocol: Volumes used for this protocol are for a 75cm2 flask; proportionally reduce or increase amount of dissociation medium for culture vessels of other sizes. Corning[®] T-75 flasks (catalog #431464) are recommended for subculturing this product.

- Transfer cell suspension to centrifuge tube. Centrifuge cells at 180 to 225 xg for 8-15 minutes at room temperature.
- Remove and discard supernatant leaving cell pellet.
- Resuspend the cell pellet with 5 mls of fresh medium (or use an appropriate volume of medium which is a multiple of 5 to facilitate the next step).
- Gently aspirate each 5 ml aliquot of cells 4 or 5 times with a new 20 ml syringe outfitted with a 22g (1½ in.) needle to break up cell clusters.
- Add appropriate aliquots of the cell suspension to new 75 cm2 flask with 10-15 ml fresh growth medium.
 Seed flask 5 x 10(5) to 1 x 10(6) viable cells/ml or use subcultivation ratio of 1:2 to 1:4.
- Place culture vessels in incubator at 37°C Subculture when cell density reaches between 2-4 x 10(6) viable cells/ml.

Medium Renewal: Every 2 to 3 days



Cryopreservation Medium

Complete growth medium described above supplemented with 10% (v/v) DMSO. Cell culture tested DMSO is available as ATCC Catalog No. 4-X.



Comments

The cells respond reversibly to NGF by induction of the neuronal phenotype when plated on Collagen IV coated culture flasks.

The cells do not synthesize epinephrine.



References

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



Biosafety Level: 1

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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