

MPC 862L

CRL-3540[™]

Description

MPC 862L is a cell line isolated from a mouse pheochromocytoma.

Organism: Mus musculus, mouse

Tissue: Adrenal gland

Morphology: Epithelial like and/or rounded

Disease: Pheochromocytoma

Cells per vial: 6.0×10^6

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.

BSL₂

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.

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 (35-37°C) **Atmosphere:** 95% Air, 5% CO₂

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:



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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%

Subculturing procedure: Before initiating subculture flasks must be coated with 50 μ g/ mL collagen I from rat tail (Thermofisher Scientific catalog #: A1048301 or equivalent). It is recommended to follow the vendor instructions for thin coating procedure.

It is recommended to subculture at or before 80% confluence. Cells may not reach 100% confluence and grow in clumps which will become necrotic if not passaged. Before beginning subculture procedure prepare a solution of 0.25% GibcoTM Trypsin (ThermoFisher Scientific catalog # 15050- 065) + 80 μ g/mL Deoxyribonuclease I. Deoxyribonuclease I (DNase I) should be added to an aliquot of 0.25% GibcoTM Trypsin fresh prior to use.

To prepare 10 mg/mL DNase I stock solution, aseptically combine: o 100 mg RocheTM DNase I (Sigma catalog # 11284932001) o 10 mL Molecular Grade Water (ATCC catalog # 60-2450) Add 8 μ L stock DNase I per 1 mL 0.25% Trypsin immediately prior to use.

Volumes used in this protocol are for 75 cm² flask; proportionally reduce or increase amount of dissociation medium for culture vessels of other sizes.

- 1. Remove and discard culture medium.
- 2. Briefly rinse the cell layer with PBS to remove all traces of serum that contains trypsin inhibitor.
- 3. Add 2.0 to 3.0 mL of 0.25% GibcoTM Trypsin (ThermoFisher Scientific catalog # 15050-065) + 80 μg/mL Deoxyribonuclease I) to flask and observe cells under an inverted microscope until the cell layer is dispersed (usually within 5 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. Centrifuge the cell suspension at approximately 150 to 400 x g for 8 to 12 minutes to remove dissociation agent.
- 6. Resuspend the cell pellet in an appropriate amount of complete culture medium
- 7. If cell clumps are present then clumps should be disrupted using a 5 mL serological pipette followed by a P1000.
- 8. Add appropriate aliquots of the cell suspension to new culture vessels. Cultures can be established between 1.0×10^{54} and $2.06.0 \times 10^{5}$ viable cells/cm².
- 9. Incubate cultures at 37°C.

Interval: Maintain cultures at a cell concentration between 1.0×10^{54} and 2.0×10^{5}

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cell/cm².

Subcultivation Ratio: A subcultivation ratio of 1:2 to 1:3 is recommended

Medium Renewal: Every 2 to 3 days

Reagents for cryopreservation:

Gibco™ Recovery™ Cell Culture Freezing Media (ThermoFisher Scientific catalog # 12648-010).

Material Citation

If use of this material results in a scientific publication, please cite the material in the following manner: MPC 862L (ATCC CRL-3540)

References

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

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