MA-10

CRL-3050™

Description

MA-10 is a cell line derived from the testis of a transplantable Leydig cell tumor, M5480P, by alternately growing it in culture and in host animals. Cells obtained from culture-derived tumors were cloned. A clone, designated MA-10, was determined to have retained functional hCG receptors. These cells synthesize progesterone in response to human chorionic gonadotropin (hCG).

Organism: *Mus musculus*, mouse
Cell Type: Leydig cell
Tissue: Testis
Morphology: epithelial-like
Growth properties: Adherent

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 1

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
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, Catalog No. 30-2006. To make the complete growth medium, add the following components to the base medium:

- additional 20mM HEPES
- horse serum to a final concentration of 15%

Handling Procedure:

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.

Note: Corning or Costar tissue culture plasticware is recommended for culture, Nunc and Falcon plasticware should not be used

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 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 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). 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:

Note: Corning or Costar tissue culture plasticware is recommended for culture, Nunc and Falcon plasticware should not be used.

Note: These cells are cultured on vessels coated with 0.1% Gelatin (ATCC PCS-999-027).

Note: Use 0.025% Trypsin – 0.053mM EDTA. Dilute 0.25% Trypsin – 0.53mM EDTA (ATCC 30-2101) 1:10 with Ca^{2+}/Mg^{2+} free Dulbecco's phosphate-buffered saline (D-PBS) (ATCC 30-2200).

Note: If cell morphology changes from adherent and epithelial-like to being rounded up and forming loosely attached clusters, the serum may not be appropriate for these cells. Change serum lots and/or vendors or use charcoal-stripped horse serum to obtain correct morphology.

Volumes used in this protocol are for 75 cm^2 flasks; proportionally reduce or increase amount of solutions for culture vessels of other sizes.
1. Remove and discard culture medium.

2. Briefly rinse the cell layers 2 times with Ca^{2+}/Mg^{2+} free Dulbecco's phosphate-buffered saline (D-PBS).

3. Add 2.0 to 3.0 ml of 0.025% Trypsin-0.053mM 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. 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. Add appropriate aliquots of the cell suspension to new gelatin-coated culture vessels. An inoculum of 2 x 10^4 to 3 x 10^4 viable cells/cm^2 is recommended.
6. Incubate cultures at 37.0°C.
7. Subculture when cultures reach a cell concentration between 2 x 10^4 to 3 x 10^5 cells/cm2.

Subcultivation ratio: A subcultivation ratio of 1:3 to 1:8 is recommended.
Medium renewal: 2 to 3 times weekly

Reagents for cryopreservation: Complete growth medium supplemented with 10% (v/v) DMSO (ATCC 4-X)

Material Citation

If use of this material results in a scientific publication, please cite the material in the following manner: MA-10 (ATCC CRL-3050)

References

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

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Revision

This information on this document was last updated on 2026-08-27

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