MEF (C57BL/6) IRR

SCRC-1008.1™

Description

Organism: *Mus musculus*, mouse
Cell Type: fibroblast
Tissue: Embryo; Whole
Age: 14 days gestation
Gender: Male and female mixed
Morphology: Fibroblast
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 Dulbecco's Modified Eagle's Medium, Catalog No. 30-2002. To make the complete growth

medium, add the following components to the base medium:
fetal bovine serum to a final concentration of 15%

This medium is formulated for use with a 5% CO2 in air atmosphere. (Standard DMEM formulations contain 3.7 g/L sodium bicarbonate and a 10% CO2 in air atmosphere is then recommended).

Handling Procedure: To insure the highest level of viability, be sure to warm media to 37°C before using it on the cells.

Flasks do not need to be coated before plating MEFs.

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.
2. Remove the vial from the water bath as soon as the contents are half way thawed (approximately 90 seconds), 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's contents plus 5 mL of complete medium (see below for recipe) to a 15 mL centrifuge tube. Use an additional 1 mL of medium to rinse the vial and transfer the liquid to the 15 mL tube. Add 4 mL of complete medium to bring the total volume to 10 mL.
4. Gently mix and pellet the cells by centrifugation @ 270 x g for 5 minutes.
5. Discard the supernatant, resuspend the cells with fresh growth medium (warm), and transfer to the appropriate size flask (see batch specific information).
6. Add more fresh growth medium (warm) to obtain the total volume recommended for the flask.
7. Incubate 37°C in a 5% CO_2 in air atmosphere.
8. Fluid change twice a week or when pH decreases. 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 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).

Cells should be plated 24 hours before use as a feeder layer for ES cells and kept for no more than 7 days.

Subculturing procedure:

Once the feeder cells have attached, the culture medium can be changed to accommodate the cells to be supported. It is recommended that the feeder cells be

plated 24 hours before use at 1 X 10^4 cells/cm^2 in order to obtain a supportive monolayer for stem cell growth.

Medium Renewal: Twice a week or when pH decreases

Material Citation

If use of this material results in a scientific publication, please cite the material in the following manner: MEF (C57BL/6) IRR (ATCC SCRC-1008.1)

References

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

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Contact Information

ATCC
10801 University Boulevard
Manassas, VA 20110-2209
USA
US telephone: 800-638-6597
Worldwide telephone: +1-703-365-2700
Email: tech@atcc.org or contact your local distributor