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BCRJ Code: 0405**Cell Line:** MEF-1**Species:** Mus musculus**Vulgar Name:** Mouse**Tissue:** Embryo**Cell Type:** Sv40 Transformed**Morphology:** Fibroblast**Growth Properties:** Adherent**Age/Ethnicity:** Embryo /**Derivation:** The cell line MEF-1 was established by transfection of mouse embryo fibroblasts ((C57 BL/6 x 129) F1) with a SV40 coding plasmid. MEF-1 express the wild-type for low density lipoprotein receptor related protein (LRP). Together with the cell lines PEA10 and PEA13, which express heterozygous and homozygous forms of LPR, an experimental system is available for the analysis of cellular uptake of functionally diverse ligands and the effect of LPR - deficiency. This is of importance since LRP - deficient mouse embryos die early during gestation.**Biosafety:** 2**Culture Medium:** DMEM High glucose with 2 mM Glutamine and 10% of fetal Bovine Serum (FBS).**Subculturing:** Split sub-confluent cultures (70-80%) using 0.05% trypsin or trypsin/EDTA; 5% CO₂; 37°C. Note: For more information on enzymatic dissociation and subculturing of cell lines consult Chapter 10 in Culture of Animal Cells, a manual of Basic Technique by R. Ian Freshney, 3rd edition, published by Alan R. Liss, N.Y., 1994.**Subculturing Medium Renewal:** 2 to 3 times a week

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**Subculturing
Subcultivation Ratio:**

1:6 to 1:10

Culture Conditions:Atmosphere: air, 95%; carbon dioxide (CO₂), 5% Temperature: 37°C**Cryopreservation:**

95% FBS + 5% DMSO (Dimethyl sulfoxide)

Thawing Frozen Cells:

SAFETY PRECAUTION: It is highly recommended 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 submerged 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. 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. For cells that are sensitive to DMSO it is recommended that the cryoprotective agent be removed immediately. 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. Discard the supernatant and Resuspend cell pellet with the recommended complete medium (see the specific batch information for the culture recommended dilution ratio). 5. Incubate the culture in an appropriate atmosphere and temperature (see "Culture Conditions" for this cell line). **NOTE:** 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).

References:

Cell 1995; 82:331, J Cell Sci 1994; 107:719, J. Biol Chem 1994; 269:21117. Kounnas MZ, et al. LDL receptor-related protein, a multifunctional ApoE receptor, binds secreted beta-amyloid precursor protein and mediates its degradation. Cell 82: 331-340, 1995. PubMed: 7543026 Willnow TE, Herz J. Genetic deficiency in low density lipoprotein receptor-related protein confers cellular resistance to Pseudomonas exotoxin A. Evidence that this protein is required for uptake and degradation of multiple ligands. J. Cell Sci. 107: 719-726, 1994. PubMed: 8006085 Orth K, et al. Low density lipoprotein receptor-related protein is necessary for the internalization of both tissue-type plasminogen activator-inhibitor complexes and free tissue-type plasminogen activator. J. Biol. Chem. 269: 21117-21122, 1994. PubMed: 8063731

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