

Data Sheet

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BCRJ Code:	0159
Cell Line:	McA-RH7777
Species:	<i>Rattus norvegicus</i>
Vulgar Name:	Rat
Tissue:	Liver
Morphology:	Epithelial
Disease:	Hepatoma; Morris Hepatoma 7777
Growth Properties:	Loosely Adherent
Sex:	Female
Applications:	This line is suitable as a transfection host.
Tumor Formation::	HEPATOMA, MORRIS HEPATOMA
Products:	alpha-fetoprotein (AFP, alpha fetoprotein)
Biosafety:	1
Additional Info:	Addition of glucocorticoids (dexamethasone) to the medium accelerates cell proliferation and reduces alpha fetoprotein production. The cells tend to shed from the growth surface before becoming confluent.
Culture Medium:	Dulbecco's Modified Eagle's Medium (DMEM) modified to contain 4 mM L-glutamine, 4500 mg/L glucose and fetal bovine serum to a final concentration of 10%.

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Heavy monolayer sloughs off; subculture before 70% confluency. Volumes used in this protocol are for 75 cm² flask; proportionally reduce or increase amount of dissociation medium for culture vessels of other sizes. Remove culture medium with floating cells to a centrifuge tube. If any cells are attached, tap flask gently or if necessary add 2.0 to 3.0 mL of 0.25% Trypsin-0.53 mM EDTA solution to flask and observe cells under an inverted microscope until cell layer is dispersed. Add 2.0 to 3.0 mL of complete growth medium and aspirate cells by gently pipetting. To remove trypsin-EDTA solution, transfer cell suspension to the centrifuge tube with the medium and cells from step #1 and spin at approximately 125 x g for 5 to 10 minutes. Discard supernatant and resuspend cells in fresh growth medium. Add appropriate aliquots of cell suspension to new culture vessels. Place culture vessels in incubators at 37°C. NOTE: For more information on enzymatic dissociation and subculturing of cell lines consult Chapter 12 in Culture of Animal Cells, a manual of Basic Technique by R. Ian Freshney, 6th edition, published by Alan R. Liss, N.Y., 2010.

Subculturing:

Subculturing Medium Renewal:

Every 2 to 3 days

Subculturing Subcultivation Ratio:

1:4 to 1:6 weekly is recommended

Culture Conditions:

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

Cryopreservation:

95% FBS + 5% DMSO (Dimethyl sulfoxide)

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SAFETY PRECAUTION: It is strongly recommended to always wear protective gloves, clothing, and a full-face mask when handling frozen vials. Some vials may leak when submerged in liquid nitrogen, allowing nitrogen to slowly enter the vial. Upon thawing, the conversion of liquid nitrogen back to its gas phase may cause the vial to explode or eject its cap with significant force, creating flying debris.

1. Thaw the vial by gently agitating it in a 37°C water bath. To minimize 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 its contents are thawed and decontaminate it by dipping in or spraying with 70% ethanol. From this point, all operations must be performed under strict aseptic conditions.
3. For cells sensitive to DMSO, it is recommended to remove the cryoprotective agent immediately. Transfer the vial contents to a centrifuge tube containing 9.0 mL of complete culture medium and centrifuge at approximately 125 × g for 5 to 7 minutes.
4. Discard the supernatant and resuspend the cell pellet in the recommended complete medium (see specific batch information for the appropriate dilution ratio).
5. Incubate the culture under appropriate atmospheric and temperature conditions (see "Culture Conditions" for this cell line).

NOTE: It is important to avoid excessive alkalinity of the medium during cell recovery. To minimize this risk, it is recommended to place the culture vessel containing the growth medium in the incubator for at least 15 minutes before adding the vial contents. This allows the medium to stabilize at its normal pH (7.0 to 7.6).

Thawing Frozen Cells:

References:

Recent Results Cancer Res. 44: 103-114, 1974. Kulas DT , et al. The transmembrane protein-tyrosine phosphatase LAR modulates signaling by multiple receptor tyrosine kinases. J. Biol. Chem. 271: 748-754, 1996. Schock D , et al. An auxiliary factor containi

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