BCRJ Code:	0273
Cell Line:	J774 1.6
Species:	Mus musculus
Vulgar Name:	Mouse
Tissue:	Reticulum
Cell Type:	Macrophage-Like
Morphology:	Macrophage
Disease:	Sarcoma
Growth Properties:	Adherent
Products:	nitric oxide
Biosafety:	1
Culture Medium:	Dulbecco's Modified Eagle's Medium (DMEM) with 1% non-essential amino acids, 2 mM L-glutamine, 1.0 g/L glucose and 10% of fetal bovine serum.
Subculturing:	Subcultures are prepared by scraping. For a 75 cm2 flask, remove all but 10 mL culture medium (adjust amount accordingly for other culture vessels). Dislodge cells from the flask substrate with a cell scraper; aspirate and add appropriate aliquots of the cell suspension into new culture vessels.
Subculturing Medium Renewal:	Twice per week
Subculturing Subcultivation Ratio:	1:6 to 1:8

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 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).
References:	Guido Damiani et al. J. Exp. MEn. © The Rockefeller University Press. Volume 152:808-822, October 1980.
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