Data Sheet

BCRJ Code:	0333
Cell Line:	32D clone 3
Species:	Mus muscullus
Vulgar Name:	Mouse
Tissue:	Bone Marrow
Morphology:	Lymphoblast
Growth Properties:	Mixed, Adherent And Suspension
Applications:	This cell line is a suitable transfection host.
Biosafety:	1
Addtional Info:	This line is IL-3 dependent.
Culture Medium:	RPMI 1640 medium with 2 mM L-glutamine adjusted to contain 4.5 g/L glucose, 10% of heat-inactivated fetal bovine serum and 10% mouse Interleukin-3 culture supplement.
Subculturing:	Cultures can be maintained by the addition of fresh medium or replacement of medium. Alternatively, cultures can be established by centrifugation with subsequent resuspension at 2 X 105 viable cells/mL. Maintain cell density between 2 X 105 and 1 X 106 viable cells/mL. Scrape off the attached cells and transfer along with the floating cells into new flasks. 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 Medium Renewal:	Every 2 to 3 days
Culture Conditions:	Atmosphere: air, 95%; carbon dioxide (CO2), 5% Temperature: 37°C

Cryopreservation:	95% FBS + 5% DMSO (Dimethyl sulfoxide)
Thawing Frozen Cells:	SAFETY PRECAUTION: Is highly recommend 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 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 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 Oring 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 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 a 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:	Ahmed N, Berridge MV. Regulation of glucose transport by interleukin-3 in growth factor-dependent and oncogene-transformed bone marrow-derived cell lines. Leuk. Res. 21: 609-618, 1997. PubMed: 9301681 Ahmed N, Berridge MV. Distinct regulation of glucose transport by interleukin-3 and oncogenes in a murine bone marrow-derived cell line. Biochem. Pharmacol. 57: 387-396, 1999. PubMed: 9933027
Depositors:	Fábio Santos - Hospital Israelita Albert Einstein
ATCC:	CRL-11346