Data Sheet

BCRJ Code:	0369
Cell Line:	NCCIT
Species:	Homo sapiens
Vulgar Name:	Human
Tissue:	Embryo, placenta
Morphology:	Epithelial
Disease:	Pluripotent embryonal carcinoma; teratocarcinoma.
Growth Properties:	Adherent
Sex:	Male
Age/Ethinicity:	Adult / Japanese
Tumor Formation::	Yes, Tumors developed within 21 days at 100% frequency (5/5) in nude mice inoculated subcutaneously with 107 cells.
Biosafety:	1
Addtional Info:	This pluripotent stem cell line is capable of somatic and extraembryonic differentiation. The undifferentiated cells are equivalent to a stage intermediate between seminoma and embryonal carcinoma. They will differentiate in response to retinoic acid. NCCIT cells are negative for keratin. They are positive for vimentin and placental alkaline phosphatase.
Culture Medium:	RPMI-1640 medium modified to contain 2 mM L-glutamine, 4500 mg/L glucose with fetal bovine serum to a final concentration of 10%.

Subculturing:	Remove spent medium, add fresh 0.25% trypsin, 0.03% EDTA solution, rinse and remove trypsin. Let the culture sit at room temperature (or at 37°C) for 2 to 5 minutes. Add fresh medium, aspirate and dispense into new flasks. Subculture two times weekly. 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:	Add fresh medium at the time of subculture
Subculturing Subcultivation Ratio:	1:4 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: 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:	Teshima S, et al. Four new human germ cell tumor cell lines. Lab. Invest. 59: 328-336, 1988. PubMed: 2842544 Damjanov I, et al. Retinoic acid-induced differentiation of the developmentally pluripotent human germ cell tumor-derived cell line, NCCIT. Lab. Invest. 68: 220-232, 1993. PubMed: 7680083
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