

Data Sheet

BCRJ Code:	0303
Cell Line:	NTERA-2 cl.D1 [NT2/D1]
Species:	Homo sapiens
Vulgar Name:	Human
Tissue:	Testis; Derived From Metastatic Site: Lung
Morphology:	Epithelial-Like, Differentiation Changes Phenotype
Disease:	Malignant Pluripotent Embryonal Carcinoma
Growth Properties:	Adherent
Sex:	Male
Age/Ethnicity:	22 Year / Caucasian
Derivation:	The parental NTERA-2 lines was established in 1980 from a nude mouse xenograft of the Tera-2 cell line. The NTERA-2 cl.D1 cell line is a pluripotent human testicular embryonal carcinoma cell line derived by cloning the NTERA-2 cell line.
Applications:	This cell line is a suitable transfection host.
DNA Profile:	Amelogenin: X,Y CSF1PO: 10,12 D13S317: 13 D16S539: 11,12,13 D5S818: 9,12 D7S820: 10,12 THO1: 9.3 TPOX: 8 vWA: 18,19
Virus Resistance::	UNTREATED CELLS: human cytomegalovirus (HCMV); human immunodeficiency virus (HIV-1, HTLV-III)
Biosafety:	1

Data Sheet

PAGE 2/4

Additional Info:	<p>This clone differentiates along neuroectodermal lineages after exposure to retinoic acid (RA) or hexamethylene bisacetamide (HMBA). The RA induced differentiation is characterized by glycolipid changes, appearance of neurons, and induction of homeobox (HOX) gene clusters. The cells exhibit high expression of N-myc oncogene activity. To induce differentiation, the cells should be trypsinized and seeded at a density 1×10^6 cells per 75 sq. cm. in medium containing 0.01 mM trans-retinoic acid. Stock solutions of trans-retinoic acid (10 mM, dissolved in DMSO) should be stored frozen (preferably under a nitrogen atmosphere).</p>
Culture Medium:	<p>Dulbecco's Modified Eagle's Medium (DMEM) modified to contain 2 mM L-glutamine, 4500 mg/L glucose and 10% of fetal bovine serum.</p>
Subculturing:	<p>Subcultures are prepared by scraping. Cells from confluent cultures (approximately 20 million cells per 75 cm²) are dislodged from the flask surface, aspirated and dispensed into new flasks. Cultures should be maintained at high density. Seed new flasks at a density of at least 5×10^6 viable cells per 75 cm² flask.</p>
Subculturing Medium Renewal:	<p>Every 2 to 3 days</p>
Culture Conditions:	<p>Atmosphere: air, 95%; carbon dioxide (CO₂), 5% Temperature: 37°C</p>
Cryopreservation:	<p>95% FBS + 5% DMSO (Dimethyl sulfoxide)</p>

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 vial 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:

Andrews PW. Human teratocarcinomas. *Biochim. Biophys. Acta* 948: 17-36, 1988. PubMed: 3293662 Mavilio F, et al. Activation of four homeobox gene clusters in human embryonal carcinoma cells induced to differentiate by retinoic acid. *Differentiation* 37: 73-79, 1988. PubMed: 2898410 Fenderson BA, et al. Glycolipid core structure switching from globo- to lacto- and ganglio- series during retinoic acid-induced differentiation of TERA-2-derived human embryonal carcinoma cells. *Dev. Biol.* 122: 21-34, 1987. PubMed: 3297853 Andrews PW, et al. Pluripotent embryonal carcinoma clones derived from the human teratocarcinoma cell line Tera-2. Differentiation in vivo and in vitro. *Lab. Invest.* 50: 147-162, 1984. PubMed: 6694356 Andrews PW, et al. Different patterns of glycolipid antigens are expressed following differentiation of TERA-2 human embryonal carcinoma cells induced by retinoic acid, hexamethylene bisacetamide (HMBA) or bromodeoxyuridine (BUdR). *Differentiation* 43: 131-138, 1990. PubMed: 2373286 Gonczol E, et al. Cytomegalovirus replicates in differentiated but not in undifferentiated human embryonal carcinoma cells. *Science* 224: 159-161, 1984. PubMed: 6322309 Andrews PW. Retinoic acid induces neuronal differentiation of a cloned human embryonal carcinoma cell line in vitro. *Dev. Biol.* 103: 285-293, 1984. PubMed: 6144603 Hirka G, et al. Differentiation of human embryonal carcinoma cells induces human immunodeficiency virus permissiveness which is stimulated by human cytomegalovirus coinfection. *J. Virol.* 65: 2732-2735, 1991. PubMed: 1850047 Dewji NN, Singer SJ. Cell surface expression of the Alzheimer disease-related presenilin proteins. *Proc. Natl. Acad. Sci. USA* 94: 9926-9931, 1997. PubMed: 9275228 Baldassarre G, et al. Transfection with a CRIPTO anti-sense plasmid suppresses endogenous CRIPTO expression and inhibits transformation in a human embryonal carcinoma cell line. *Int. J. Cancer* 66: 538-543, 1996. PubMed: 8635871

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