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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

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Additional Info:

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).

Culture Medium:

Dulbecco's Modified Eagle's Medium (DMEM) modified to contain 2 mM L-glutamine, 4500 mg/L glucose and 10% of fetal bovine serum.

Subculturing:

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.

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: 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 $125 \times g$ 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).

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References:

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