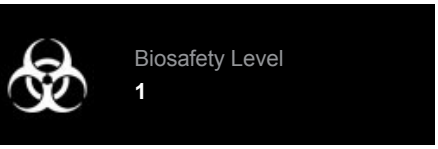
ATCC

Product Sheet

NTERA-2 cl.D1 [NT2/D1] (ATCC® CRL-1973™)

Please read this FIRST



Intended Use

This product is intended for research use only. It is not intended for any animal or human therapeutic or diagnostic use.

Complete Growth Medium

The base medium for this cell line is ATCC-formulated Dulbecco's Modified Eagle's Medium, Catalog No. 30-2002. To make the complete growth medium, add the following components to the base medium: fetal bovine serum to a final concentration of 10%.

Citation of Strain

If use of this culture results in a scientific publication, it should be cited in that manuscript in the following manner: NTERA-2 cl.D1 [NT2/D1] (ATCC® CRL-1973™)

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PO Box 1549
Manassas, VA 20108 USA
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Description

Organism: *Homo sapiens*, human
Tissue: testis; derived from metastatic site: lung
Disease: malignant pluripotent embryonal carcinoma
Age: 22 years
Gender: male
Morphology: epithelial-like,differentiation changes phenotype
Growth Properties: adherent
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
Cytogenetic Analysis: This is a hypotriploid human cell line with the modal chromosome number of 63 in 48% of cells examined. However, cells with 62 chromosome counts also occurred at a rather high frequency (24%). The rate of polyploidy was 1.6%., About 12 marker chromosomes are constantly found in most cells. They include: der(9)t(1;9)(q25;q34.3); del(1)(q25); der(13)t(11;13)(q13;q34); t(Xq1q); and eight others., At least two markers are found only in some cells. The normal Y chromosome was found in all cells. Only single copies of normal chromosomes 1, 10, 11 and 13 were present. Others were mostly in two or three copies per cell.

Batch-Specific Information

Refer to the Certificate of Analysis for batch-specific test results.

SAFETY PRECAUTION

ATCC highly recommends 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.

Unpacking & Storage Instructions

1. Check all containers for leakage or breakage.
2. Remove the frozen cells from the dry ice packaging and immediately place the cells at a temperature below -130°C, preferably in liquid nitrogen vapor, until ready for use.

Handling Procedure for Frozen Cells

To insure the highest level of viability, thaw the vial and initiate the culture as soon as possible upon receipt. If upon arrival, continued storage of the frozen culture is necessary, it should be stored in liquid nitrogen vapor phase and not at -70°C. Storage at -70°C will result in loss of viability.

1. Thaw the vial by gentle agitation in a 37°C water bath. To reduce the possibility of 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 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. 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. Resuspend cell pellet with the recommended complete medium (see the specific batch information for the culture recommended dilution ratio). 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 complete 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). pH (7.0 to 7.6).
5. Incubate the culture at 37°C in a suitable incubator. A 5% CO_2 in air atmosphere is recommended if using the medium described on this product sheet.

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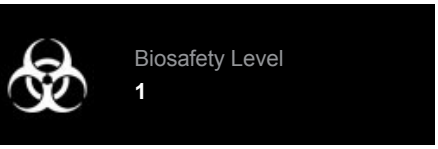

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Handling Procedure for Flask Cultures

The flask was seeded with cells (see specific batch information) grown and completely filled with medium at ATCC to prevent loss of cells during shipping.

1. Upon receipt visually examine the culture for macroscopic evidence of any microbial contamination. Using an inverted microscope (preferably equipped with phase-contrast optics), carefully check for any evidence of microbial contamination. Also check to determine if the majority of cells are still attached to the bottom of the flask; during shipping the cultures are sometimes handled roughly and many of the cells often detach and become suspended in the culture medium (but are still viable).
2. **If the cells are still attached,** aseptically remove all but 5 to 10 mL of the shipping medium. The shipping medium can be saved for reuse. Incubate the cells at 37°C in a 5% CO_2 in air atmosphere until they are ready to be subcultured.
3. **If the cells are not attached,** aseptically remove the entire contents of the flask and centrifuge at 125 x g for 5 to 10 minutes. Remove shipping medium and save. Resuspend the pelleted cells in 10 mL of this medium and add to 25 cm^2 flask. Incubate at 37°C in a 5% CO_2 in air atmosphere until cells are ready to be subcultured.

Subculturing Procedure

Subcultures are prepared by scraping. Cells from confluent cultures (approximately 20 million cells per 75 cm^2) 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 X 10^6 viable cells per 75 cm^2 flask.
Medium Renewal: Every 2 to 3 days

Cryopreservation Medium

Complete growth medium described above supplemented with 5% (v/v) DMSO. Cell culture tested DMSO is available as ATCC Catalog No. 4-X.

Comments

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 X 10 exp6 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).

References

References and other information relating to this product are available online at www.atcc.org.

Biosafety Level: 1

Appropriate safety procedures should always be used with this material. Laboratory safety is discussed in the current publication of the *Biosafety in Microbiological and Biomedical Laboratories* from the U.S. Department of Health and Human Services Centers for Disease Control and Prevention and National Institutes for Health.

ATCC Warranty

ATCC® products are warranted for 30 days from the date of shipment, and this warranty is valid only if the product is stored and handled according to the information included on this product information sheet. If the ATCC® product is a living cell or microorganism, ATCC lists the media formulation that has been found to be effective for this product. While other, unspecified media may also produce satisfactory results, a change in media or the absence of an additive from the ATCC recommended media may affect recovery, growth and/or function of this product. If an alternative medium formulation is used, the ATCC warranty for viability is no longer valid.

Disclaimers

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