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BCRJ Code: 0111

Cell Line: HT-29

Species: Homo sapiens

Vulgar Name: Human

Colon Tissue:

Morphology: **Epithelial**

Disease: Colorectal Adenocarcinoma

Growth Properties: Adherent

Sex: **Female**

44 Year / Caucasian Age/Ethinicity:

Applications: This cell line is a suitable transfection host.

Amelogenin: X CSF1PO: 11,12 D13S317: 11,12 D16S539: 11,12 D5S818: 11,12 D7S820: 10 THO1: 6,9 **DNA Profile:**

TPOX: 8,9 vWA: 17,19

Yes, in nude mice; forms well differentiated adenocarcinoma consistent with colonic primary (grade **Tumor Formation::**

I); tumors also form in steroid treated hamsters

secretory component of IgA; carcinoembryonic antigen (CEA); transforming growth factor beta **Products:**

binding protein; mucin

Biosafety: 1

> Ultrastructural features reported for HT-29 cells include microvilli, microfilaments, large vacuolated mitochondria with dark granules, smooth and rough endoplasmic reticulum with free ribosomes,

lipid droplets, few primary and many secondary lysosomes. The cells express urokinase receptors, but do not have detectable plasminogen activator activity [PubMed ID: 8381394]. HT-29 cells are negative for CD4, but there is cell surface expression of galactose ceramide (a possible alternative receptor for HIV). The line is positive for expression of c-myc, K-ras, H-ras, N-ras, Myb, sis and fos oncogenes. The p53 antigen is overproduced, and there is a G -> A mutation in codon 273 of the p53 gene resulting in an Arg -> His substitution. N-myc oncogene expression was not detected. There is a

G -> A mutation in codon 273 of the p53 gene resulting in an Arg -> His substitution.

Culture Medium: McCoy's 5A Medium is modified with fetal bovine serum to a final concentration of 10%.



Addtional Info:







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Subculturing:

Remove and discard culture medium. Briefly rinse the cell layer with PBS without calcium and magnesium to remove all traces of serum that contains trypsin inhibitor. Add 2.0 to 3.0 mL of Trypsin-EDTA solution to flask and observe cells under an inverted microscope until cell layer is dispersed (usually within 5 to 15 minutes). Note: To avoid clumping do not agitate the cells by hitting or shaking the flask while waiting for the cells to detach. Cells that are difficult to detach may be placed at 37°C to facilitate dispersal. Add 6.0 to 8.0 mL of complete growth medium and aspirate cells by gently pipetting. Add appropriate aliquots of the cell suspension to new culture vessels. Incubate cultures at 37°C. 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:

2 to 3 times per week

Subculturing Subcultivation Ratio:

1:3 to 1:8

Culture Conditions:

Atmosphere: air, 95%; carbon dioxide (CO2), 5% Temperature: 37°C

Cryopreservation:

95% FBS + 5% DMSO (Dimethyl sulfoxide)

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.

Thawing Frozen Cells:

- 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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