Data Sheet
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Caco-2/TC-7 Human Colon Adenocarcinoma Cell Line

Cancer Cell Line

Catalogue number SCC209

Pack Size ≥ 1x10^6

Store at Liquid Nitrogen

FOR RESEARCH USE ONLY

Not for use in diagnostic procedures. Not for human or animal consumption.

Background

The Caco-2 cell line and clones are human colon carcinoma cells that spontaneously differentiate into mature enterocyte-like phenotypes. Cultured beyond confluency, these cells develop tight junctions, microvilli, and express enzymes and transporters characteristic of intestinal enterocytes.

The use of Caco-2 cells and clones as enterocyte surrogates[1] has enabled the in vitro study of the structure and polarization of the human intestinal barrier, the intestinal biotransformation of drugs, toxicology of the intestine, [2] the pathogenesis of enterovirulent bacteria, [3] and more over the past three decades. The prediction of oral bioavailability from in vitro assays is an invaluable tool in drug discovery and development. The Caco-2 assay1 remains the gold standard for in vitro prediction of intestinal drug permeability and absorption to date.[4]

The parental Caco-2 cell line have been shown to be heterogeneous with highly variable transport and permeability properties that are dependent upon culture conditions. Caco-2/TC-7 clone was isolated from a late passage of the parental Caco-2 cell line and comprises a more homogeneous population of cells that exhibits the characteristic functions of small intestinal enterocytes including organization of the apical brush border, distribution of the associated hydrolase sucrase-isomaltase (SI),[5] more developed intercellular tight junctions and a shorter differentiation time.

Source

The Caco-2/TC-7 Human Colon Adenocarcinoma Cell Line is a late passage clone of the non-clonal parental Caco-2 line.[5]

Short Tandem Repeat

D3S1358: 17	Penta E: 7	D16S539: 12, 13	D8S1179: 12
TH01: 6	D5S818: 12, 13	CSF1PO: 11	TPOX: 9, 11
D21S11: 30	D13S317: 11, 13, 14	Penta D: 9	FGA: 19
D18S51: 12	D7S820: 11, 12	vWA: 16, 18	Amelogenin: X

Cancer cell lines are inherently genetically unstable. Genetic instability may arise in the form of loss of heterozygosity of alleles at one or more genetic sites with increased passages.

Quality Control Testing

- Each vial contains ≥ 1x10^6 viable cells.

- Cells are tested negative for infectious diseases by a Human Essential CLEAR panel by Charles River Animal Diagnostic Services.
- Cells are verified to be of human origin and negative for inter-species contamination from mouse, rat, Chinese hamster, Golden Syrian hamster, and Non-human Primate (NHP) as assessed by a Contamination Clear panel by Charles River Animal Diagnostic Services.
- Cells are negative for mycoplasma contamination.

Storage and Handling

Caco-2/TC-7 cells should be stored in liquid nitrogen until use. The cells can be cultured for at least 10 passages after initial thawing without significantly affecting the cell marker expression and functionality.

Representative Data

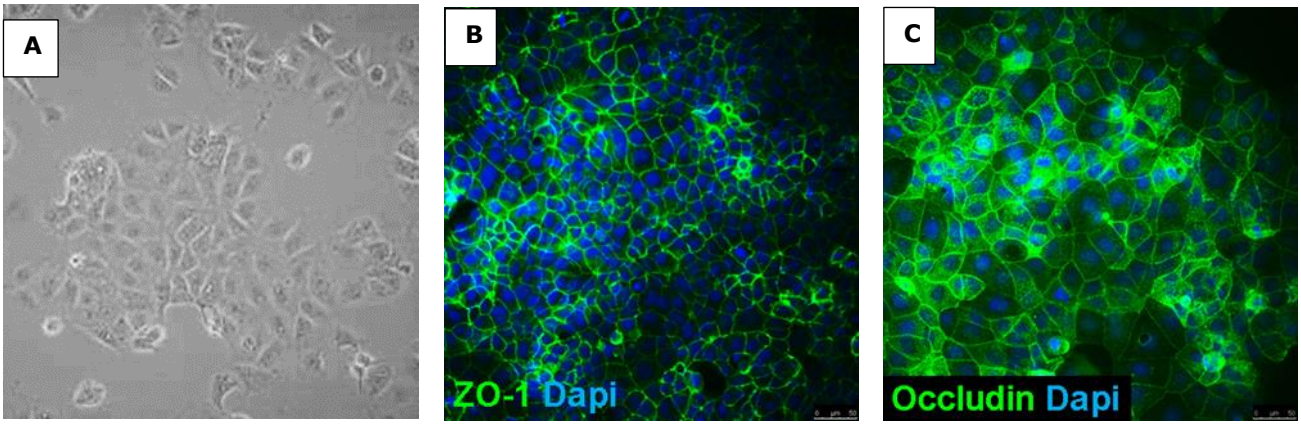

Figure 1. Bright-field image of Caco-2/TC-7 cells one day after thaw (A). Caco-2/TC-7 cells express ZO-1 (B, Cat. No. MABT339) and Occludin (C, Cat. No. ABT146).

Protocols

Thawing Cells

1. Do not thaw the cells until the recommended medium is on hand. Cells can grow on normal tissue cultureware surfaces without any additional coating.

   Caco-2/TC-7 Expansion Medium: Cells are thawed and expanded in Caco-2/TC-7 Expansion Medium comprised of High Glucose DMEM with L-Glutamine and sodium pyruvate (Cat. No. D6429) supplemented with 10% Fetal Bovine Serum (Cat. No. ES-009-B), 1x L-alanyl-L-glutamine (Cat. No. A0550-1G) and 1x Non-Essential Amino Acids (NEAA; Cat. No. TMS-001).
2. Remove the vial of frozen Caco-2/TC-7 cells from liquid nitrogen and incubate in a 37 °C water bath. Closely monitor until the cells are completely thawed. Maximum cell viability is dependent on the rapid and complete thawing of frozen cells.

   IMPORTANT: Do not vortex the cells.
3. As soon as the cells are completely thawed, disinfect the outside of the vial with 70% ethanol. Proceed immediately to the next step.
4. In a laminar flow hood, use a 1- or 2-mL pipette to transfer the cells to a sterile 15 mL conical tube. Be careful not to introduce any bubbles during the transfer process.

6. Using a 10 mL pipette, slowly add dropwise 9 mL of Caco-2/TC-7 Expansion Medium (Step 1 above) to the 15 mL conical tube.

   IMPORTANT: Do not add the entire volume of media all at once to the cells. This may result in decreased cell viability due to osmotic shock.
7. Gently mix the cell suspension by slowly pipetting up and down twice. Be careful not to introduce any bubbles.

   IMPORTANT: Do not vortex the cells.
8. Centrifuge the tube at 300 *x g* for 2-3 minutes to pellet the cells.
9. Decant as much of the supernatant as possible. Steps 5-8 are necessary to remove residual cryopreservative (DMSO).
10. Resuspend the cells in 15 mL of Caco-2/TC-7 Expansion Medium.
11. Transfer the cell mixture to a T75 tissue culture flask.
12. Incubate the cells at 37 °C in a humidified incubator with 5% CO_2.

Subculturing Cells

1. Do not allow the cells to grow to confluency. Caco-2/TC-7 cells should be passaged at ~80-85% confluence.
2. Carefully remove the medium from the T75 tissue culture flask containing the 80% confluent layer of Caco-2/TC-7 cells.
3. Rinse the flask with 10 mL 1X PBS. Aspirate after the rinse.
4. Apply 5-7 mL of Accutase and incubate in a 37 °C incubator for 3-5 minutes.
5. Inspect the flask and ensure the complete detachment of cells by gently tapping the side of the flask with the palm of your hand.
6. Add 5-7 mL of Caco-2/TC-7 Expansion Medium to the plate.
7. Gently rotate the flask to mix the cell suspension. Transfer the dissociated cells to a 15 mL conical tube.
8. Centrifuge the tube at 300 *x g* for 3-5 minutes to pellet the cells.
9. Discard the supernatant, then loosen the cell pellet by tapping the tip of the tube with a finger.
10. Apply 2-5 mL of Caco-2/TC-7 Expansion Medium to the conical tube and resuspend the cells thoroughly.

    IMPORTANT: Do not vortex the cells.
11. Count the number of cells using a hemocytometer.
12. Plate the cells to the desired density. Do not exceed 85-90% confluency. Recommended split ratio is 1:5.

Cryopreservation of Cells

Caco-2/TC-7 Human Colon Adenocarcinoma Cell Line may be frozen in Caco-2/TC-7 Expansion Medium and 10% DMSO using a Nalgene slow freeze Mr. Frosty container.

References

1. Gastroenterology 1989; 96(3): 736-749.
2. Cell Biol. Toxicol. 2005; 21(1): 1-26.
3. Microbiol. Mol. Biol. Rev. 2013; 77(3): 380-439.
4. Current Topics in Medicinal Chemistry 2018; 18(26): 2209-2229.
5. J. Cell Sci. 1994; 107(Pt 1): 213-225.

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