

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

Cancer Cell Line

Catalogue number SCC209

Pack Size $\geq 1 \times 10^6$

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 surrogates¹ 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 $\geq 1 \times 10^6$ 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

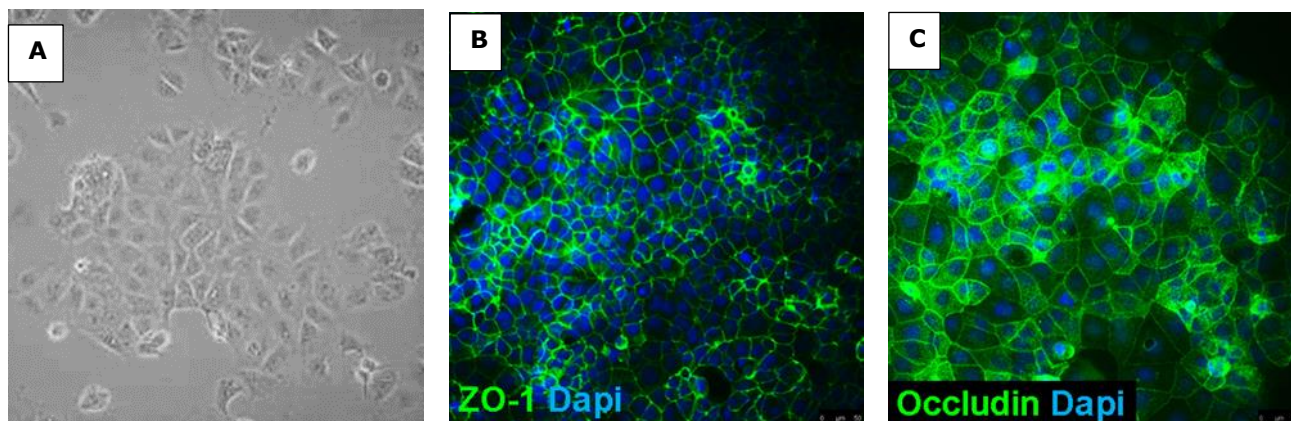


Figure 1. Bright-field image of Caco-2/TC-7 cells one day after thaw (A). Caco-2/TC-7 cells express ZO-1 (B, Cat. No. MABT339) and Occludin (C, Cat. No. ABT146).

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 $300 \times g$ 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% CO₂.

Subculturing Cells

1. Do not allow the cells to grow to confluency. Caco-2/TC-7 cells should be passaged at ~80-85% confluency.
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 $300 \times g$ 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.

Academic Use Agreement

Subject to local law

THIS PRODUCT MAY ONLY BE USED BY INDIVIDUALS EMPLOYED BY AN ACADEMIC INSTITUTION AND IS INTENDED SOLELY TO BE USED FOR ACADEMIC RESEARCH, WHICH IS FURTHER DEFINED BELOW. BY OPENING THIS PRODUCT, YOU ("PURCHASER") HEREBY REPRESENT THAT YOU HAVE THE RIGHT AND AUTHORITY TO LEGALLY BIND YOURSELF AND/OR YOUR EMPLOYER INSTITUTION, AS APPLICABLE, AND CONSENT TO BE LEGALLY BOUND BY THE TERMS OF THIS ACADEMIC USE AGREEMENT. IF YOU DO NOT AGREE TO COMPLY WITH THESE TERMS, YOU MAY NOT OPEN OR USE THE PRODUCT AND YOU MUST CALL MILLIPORESIGMA ("SELLER") CUSTOMER SERVICE (1-800-645-5476) TO ARRANGE TO RETURN THE PRODUCT FOR A REFUND.

"Product" means Caco-2/TC-7 Human Colon Cell Line (SCC209).

"Academic Research" means any internal in vitro research use by individuals employed by an academic institution. Academic Research specifically excludes the following uses of whatever kind or nature:

- Re-engineering or copying the Product
- Making derivatives, modifications, or functional equivalents of the Product
- Obtaining patents or other intellectual property rights claiming use of the Product
- Using the Product in the development, testing, or manufacture of a Commercial Product
- Using the Product as a component of a Commercial Product
- Reselling or licensing the Product
- Using the Product in clinical or therapeutic applications including producing materials for clinical trials
- Administering the Product to humans
- Using the Product in collaboration with a commercial or non-academic entity

"Commercial Product" means any product intended for: (i) current or future sale; (ii) use in a fee-for-service; or (iii) any diagnostic, clinical, or therapeutic use.

Access to the Product is limited solely to those officers, employees, and students of PURCHASER's academic institution who need access to the Product to perform Academic Research. PURCHASER shall comply with all applicable laws in its use and handling of the Product and shall keep it under reasonably safe and secure conditions to prevent unauthorized use or access.

These use restrictions will remain in effect for as long as PURCHASER possesses the Product.

COMMERCIAL OR NON-ACADEMIC ENTITIES INTERESTED IN PURCHASING OR USING THE PRODUCT MUST CONTACT licensing@emdmillipore.com AND AGREE TO SEPARATE TERMS OF USE PRIOR TO USE OR PURCHASE.

GMO

This product contains genetically modified organisms.
Este producto contiene organismos genéticamente modificados.
Questo prodotto contiene degli organismi geneticamente modificati.
Dieses Produkt enthält genetisch modifizierte Organismen.
Ce produit contient des organismes génétiquement modifiés.
Dit product bevat genetisch gewijzigde organismen.
Tämä tuote sisältää geneettisesti muutettuja organismeja.
Denna produkt innehåller genetiskt ändrade organismer.

Notice

We provide information and advice to our customers on application technologies and regulatory matters to the best of our knowledge and ability, but without obligation or liability. Existing laws and regulations are to be observed in all cases by our customers. This also applies in respect to any rights of third parties. Our information and advice do not relieve our customers of their own responsibility for checking the suitability of our products for the envisaged purpose.

The information in this document is subject to change without notice and should not be construed as a commitment by the manufacturing or selling entity, or an affiliate. We assume no responsibility for any errors that may appear in this document.

Technical Assistance

Visit the tech service page at SigmaAldrich.com/techservice.

Terms and Conditions of Sale

Warranty, use restrictions, and other conditions of sale may be found at SigmaAldrich.com/terms.

Contact Information

For the location of the office nearest you, go to SigmaAldrich.com/offices.

The life science business of Merck operates
as MilliporeSigma in the U.S. and Canada.

Merck and Sigma-Aldrich are trademarks of Merck KGaA, Darmstadt, Germany or its affiliates. All other trademarks are the property of their respective owners. Detailed information on trademarks is available via publicly accessible resources.

© 2023 Merck KGaA, Darmstadt, Germany and/or its affiliates. All Rights Reserved.

Document Template 20306518 Version 5.0

20551742 Rev 01/23

