Product sheet

Caco-2 Cells | 300137

General information

Description	Caco-2 cells serve as an advanced in vitro model for the human intestinal barrier, primarily due to their differentiation into a cell monolayer that closely resembles the enterocytes lining the small intestine. When culturing the Caco2 cell line on tissue culture filter inserts with polycarbonate filters, Caco-2 cells undergo spontaneous differentiation. The differentiation of Caco2 cells results in the expression of specialized cell types, complete with microvilli, enzymes, and transporters, paralleling the complex features and mechanisms found in an in vivo situation. In the context of intestinal absorption studies models, Caco-2 cells, which were derived from a human colorectal adenocarcinoma patient, are instrumental due to their ability to develop high TEER values, signifying intact tight junctions and epithelial barrier function. These properties are crucial for assays like the cholesterol efflux assay and investigations into cellular transport, including the movement of lipid nanoparticles and the detection of protein interactions. Caco-2 cells are pivotal for intestinal absorption studies, providing a reliable in vitro approximation of the intestinal epithelium. Mimicking intestinal enterocytes, these cells facilitate analyses of oral drug absorption by simulating the intestinal barrier. Researchers utilize Caco-2 cells to predict how substances traverse the intestinal mucosa, which is essential for the pharmacokinetic profiling of oral medications. Furthermore, they are a key tool in investigating intestinal cholesterol uptake, homeostasis and transport, which are vital processes for understanding lipid metabolism and associated diseases. Caco-2 cells remain a cornerstone in colon carcinoma and toxicology research, not only for their relevance to human gastrointestinal studies but also for their role in providing detailed insights into the biliary pathway, the metabolism of xenobiotics within the colon, cancer and toxicology research.
Organism	Human
Tissue	Colon
Disease	Adenocarcinoma
Applications	Model of the GI (gastrointestinal) tract, measurement of the Trans-Epithelial/Endothelial Eletrical Resistance (TEER). Caco-2 cells develop high TEER values of up to 2000 cm2 (as measured by CLS using the CellZscope, nanoAnalytics, Münster, Germany).
Synonyms	CaCo-2, CACO-2, Caco 2, CACO 2, CACO2, CaCo2, CaCO2, Caco2, Caco-II

Characteristics

Age	72 years
Gender	Male
Ethnicity	Caucasian

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Morphology	Epithelial-like
Growth properties	Adherent

Regulatory Data

Citation	CaCo-2 (Cytion catalog number 300137)
Biosafety level	1
NCBI_TaxID	9606
CellosaurusAccession	CVCL_0025

Biomolecular Data

Receptors expressed	Heat stable enterotoxin (Sta, E. coli), epidermal growth factor (EGF), retinoic acid binding protein I and retinol binding protein II, keratin positive.
Antigen expression	Blood Type O, Rh+, HLA class II negative
Isoenzymes	Me-2, 1, PGM3, 1, PGM1, 1, ES-D, 1, AK-1, 1, GLO-1, 1, G6PD, B.
Tumorigenic	Yes, in nude mice. Form moderately well differentiated adenocarcinomas consistent with colonic primary (grade II)
Virus resistance	Human immunodeficiency virus (HIV, LAV)
Ploidy status	(P14), hypertetraploid
MSI-status	Stable (MSS)

Handling

Culture Medium	EMEM (MEM Eagle), w: 2 mM L-Glutamine, w: 2.2 g/L NaHCO3, w: EBSS (Cytion article number 820100a)
Supplements	Supplement the medium with 10% FBS and 1% NEAA

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Dissociation Reagent	Accutase
Doubling time	60 to 70 hours
Subculturing	Remove the old medium from the adherent cells and wash them with PBS that lacks calcium and magnesium. For T25 flasks, use 3-5 ml of PBS, and for T75 flasks, use 5-10 ml. Then, cover the cells completely with Accutase, using 1-2 ml for T25 flasks and 2.5 ml for T75 flasks. Let the cells incubate at room temperature for 8-10 minutes to detach them. After incubation, gently mix the cells with 10 ml of medium to resuspend them, then centrifuge at 300xg for 3 minutes. Discard the supernatant, resuspend the cells in fresh medium, and transfer them into new flasks that already contain fresh medium.
Seeding density	1 x 10^4 cells/cm^2 will result in a 90% confluent monolayer in about 4 days
Post-Thaw Recovery	After thawing, plate the cells at 5 x 10^4 cells/cm^2 and allow the cells to recover from the freezing process and to adhere for at least 24 hours.
Freeze medium	As a cryopreservation medium, we use complete growth medium (including FBS) + 10% DMSO for adequate post-thaw viability, or CM-1 (Cytion catalog number 800100), which includes optimized osmoprotectants and metabolic stabilizers to enhance recovery and reduce cryo-induced stress.

Thawing and Culturing Cells	1. Confirm that the vial remains deeply frozen upon delivery, as cells are shipped on dry ice to maintain optimal temperatures during transit. 2. Upon receipt, either store the cryovial immediately at temperatures below -150°C to ensure the preservation of cellular integrity, or proceed to step 3 if immediate culturing is required. 3. For immediate culturing, swiftly thaw the vial by immersing it in a 37°C water bath with clean water and an antimicrobial agent, agitating gently for 40-60 seconds until a small ice clump remains. 4. Perform all subsequent steps under sterile conditions in a flow hood, disinfecting the cryovial with 70% ethanol before opening. 5. Carefully open the disinfected vial and transfer the cell suspension into a 15 ml centrifuge tube containing 8 ml of room-temperature culture medium, mixing gently. 6. Centrifuge the mixture at 300 x g for 3 minutes to separate the cells and carefully discard the supernatant containing residual freezing medium. 7. Gently resuspend the cell pellet in 10 ml of fresh culture medium. For adherent cells, divide the suspension between two T25 culture flasks; for suspension cultures, transfer all the medium into one T25 flask to promote effective cell interaction and growth. 8. Adhere to established subculture protocols for continued growth and maintenance of the cell line, ensuring reliable experimental outcomes.
Incubation Atmosphere	37°C, 5% CO_2, humidified atmosphere.
Shipping Conditions	Cryopreserved cell lines are shipped on dry ice in validated, insulated packaging with sufficient refrigerant to maintain approximately −78 °C throughout transit. On receipt, inspect the container immediately and transfer vials without delay to appropriate storage.
Storage Conditions	For long-term preservation, place vials in vapor-phase liquid nitrogen at about −150 to −196 °C. Storage at −80 °C is acceptable only as a short interim step before transfer to liquid nitrogen.

Quality Control & Molecular Analysis

Sterility	Mycoplasma contamination is excluded using both PCR-based assays and luminescence-based mycoplasma detection methods. To ensure there is no bacterial, fungal, or yeast contamination, cell cultures are subjected to daily visual inspections.