Product sheet

HT-29 MTX E12 Cells | 305801

Informações gerais

Description	HT-29-MTX-E12 is a goblet cell-like subclone derived from the human colorectal adenocarcinoma cell line HT29 through selection with methotrexate (MTX), a process that induces differentiation toward mucus-secreting phenotypes. Among several subclones developed from HT29-MTX, the E12 subclone stands out due to its robust formation of confluent monolayers with tight junctions and a significantly thick, continuous mucus layer on the apical surface. This subclone features a higher proportion of mature goblet cells, as demonstrated by Alcian Blue staining, transmission electron microscopy (TEM), and expression of mucin genes MUC1 and MUC2. In fact, MUC1 and MUC2 mRNA levels were substantially higher in HT-29-MTX-E12 compared to other subclones and parent HT29 cells, correlating with a mucus thickness of approximately 142 ± 51 µm-comparable to the in vivo intestinal environment. Functionally, HT-29-MTX-E12 has been shown to model the barrier properties of the human intestinal mucus layer, particularly in evaluating the absorption of lipophilic drugs. The presence of a thick mucus barrier significantly reduces the apparent permeability coefficients (Papp) of lipophilic compounds such as testosterone and various barbiturates when compared to mucus-free Caco-2 cells. For example, testosterone showed a 43% reduction in Papp in HT-29-MTX-E12, highlighting the impact of mucus on drug diffusion. Despite having a leakier epithelial barrier than Caco-2 cells, HT-29-MTX-E12 maintains physiological relevance through its mucus-producing capacity, making it a valuable in vitro model for investigating intestinal drug absorption and the influence of mucus on permeability.
Organism	Human
Tissue	Colon
Disease	Colon adenocarcinoma
Synonyms	HT29-MTX-E12, MTX-E12

Características

Age	44 years
Gender	Female
Ethnicity	Caucasian
Cell type	Epithelial
Growth properties	Adherent

Dados regulatórios

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Citation	HT-29-MTX-E12 (Cytion catalog number 305801)
Biosafety level	1
NCBI_TaxID	9606
CellosaurusAccession	CVCL_G356

Dados biomoleculares

Mutational profile	Mutation: APC, Simple, p.Glu853Ter (c.2557G>T), Heterozygous (from parent cell line).Mutation, APC, Simple, p.Thr1556Asnfs*3 (c.4666dupA) (c.4666_4667insA), Heterozygous (from parent cell line).Mutation, BRAF, Simple, p.Val600Glu (c.1799T>A), Heterozygous (from parent cell line).Mutation, PIK3CA, Simple, p.Pro449Thr (c.1345C>A), Heterozygous (from parent cell line).Mutation, SMAD4, Simple, p.Gln311Ter (c.931C>T), Homozygous (from parent cell line).Mutation, TP53, Simple, p.Arg273His (c.818G>A), Homozygous (from parent cell line).

Manuseio

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
Dissociation Reagent	Accutase
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.

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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.

Controle de Qualidade e Análise Molecular

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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.