

**NCM460 Cells | 305430**

**General information**

**Description**

The NCM460 cell line is derived from normal human colon mucosal epithelial cells, providing a critical in vitro model for studying human intestinal physiology and pathology. This cell line was established from histologically normal tissue isolated during surgery from a gastric cancer patient, specifically from the transverse colon margin considered free of malignant changes. NCM460 cells exhibit characteristics typical of gastrointestinal epithelial cells, including the expression of markers such as villin and human secretory component, confirming their epithelial origin. Importantly, these cells maintain a non-tumorigenic phenotype, as demonstrated by their inability to grow in soft agar and lack of tumor formation in nude mice.

The culture of NCM460 cells requires specialized conditions to support their growth as a mixed suspension-monolayer system, reflecting varying stages of epithelial differentiation. The presence of mucin-positive cells and neuroendocrine marker expression in some subpopulations suggests a retained multilineage capability, indicative of a stem-like component within the cell population. This property makes NCM460 particularly useful for studies on cell differentiation, drug transport, and epithelial barrier functions.

NCM460 has been widely applied in research focusing on colon cancer progression, allowing comparisons between normal and diseased epithelial cells. It also serves as a platform for investigating the effects of dietary components, pharmaceuticals, and other external factors on colon epithelial health and disease. This cell line offers a robust tool for advancing our understanding of gastrointestinal biology at the cellular and molecular levels.

**Organism** Human

**Tissue** Colon, mucosa

**Disease** Normal

**Synonyms** NCM-460

**Characteristics**

**Age** 68 years

**Gender** Male

**Ethnicity** Hispanic

**Morphology** Epithelial-like

**Cell type** Epithelial cell

## NCM460 Cells | 305430

**Growth properties** Adherent

## Regulatory Data

**Citation** NCM460 (Cytion catalog number 305430)

**Biosafety level** 1

**NCBI\_TaxID** 9606

**CellosaurusAccession** CVCL\_0460

## Biomolecular Data

**Tumorigenic** No, tested in nude mice and athymic mice

## Handling

**Culture Medium** DMEM, w: 4.5 g/L Glucose, w: 4 mM L-Glutamine, w: 3.7 g/L NaHCO<sub>3</sub>, w: 1.0 mM Sodium pyruvate (Cytion article number 820300a)

**Supplements** Supplement the medium with 10% FBS and 1% NEAA

**Dissociation Reagent** Accutase

**Doubling time** 32-38 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.

**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.

## NCM460 Cells | 305430

### 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  $-150^{\circ}\text{C}$  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  $37^{\circ}\text{C}$  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  $300 \times g$  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

$37^{\circ}\text{C}$ , 5%  $\text{CO}_2$ , humidified atmosphere.

### Shipping Conditions

Cryopreserved cell lines are shipped on dry ice in validated, insulated packaging with sufficient refrigerant to maintain approximately  $-78^{\circ}\text{C}$  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  $-196^{\circ}\text{C}$ . Storage at  $-80^{\circ}\text{C}$  is acceptable only as a short interim step before transfer to liquid nitrogen.

## Quality Control & Molecular Analysis

**NCM460 Cells | 305430**

**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.