**Product sheet**

# HROC32 Cells | 300818

## General information

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| **Description** | This is one cell line of a series of tumor cell lines which have been established by PD Dr. Michael Linnebacher from Primary CRC resection specimens since 2006. |
| **Organism** | Human |
| **Tissue** | Colon ascendens, UICC IV |
| **Disease** | Primary adenocarcinoma, TNM stage T4N2M1R0L0V1 grading G2, Lk(n) + 9, Σ Lk(n) 14 |
| **Synonyms** | HROC32P |

## Characteristics

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| **Age** | 82 years |
| **Gender** | Female |
| **Ethnicity** | Caucasian |
| **Morphology** | Epithelial-like |
| **Growth properties** | Adherent |

## Regulatory Data

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| **Citation** | HROC32 (Cytion catalog number 300818) |
| **Biosafety level** | 1 |
| **NCBI_TaxID** | 9606 |
| **CellosaurusAccession** | CVCL_1D06 |

## Biomolecular Data

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| **Protein expression** | PTEN |

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| **Antigen expression** | CD15 +, CD24 +, CD44 +, CD55 +, CD58 +, CD50 +, CD 54 +, CD66acde +, CD71 +, CD102 +, CD326 +, CD80 -, CD86 -, EpCAM +, HLA-A2 +, MHC I +, MHC II + (IFN-g pretreated), Her2/neu + |
| **Tumorigenic** | Yes, in immune-suppressed nude mice |
| **Viruses** | Free of human pathogenic viruses SV40, JC/BK, HBV, HCV, HIV. |
| **MSI-status** | MSS |
| **Mutational profile** | APCwt, p53R282W, K-RasG12A, N-Raswt, H-Raswt SNP rs12628 at codon 27, PIK3CAst, BRafwt |

## Handling

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| **Culture Medium** | DMEM:Ham's F12 (1:1), w: 3.1 g/L Glucose, w: 2.5 mM L-Glutamine, w: 15 mM HEPES, w: 0.5 mM Sodium pyruvate, w: 1.2 g/L NaHCO3 (Cytion article number 820400a) |
| **Supplements** | Supplement the medium with 10% FBS |
| **Dissociation Reagent** | Accutase |
| **Doubling time** | 30 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** | 2 x $10^4$ cells/$cm^2$ |
| **Fluid renewal** | Every 3 to 5 days |
| **Post-Thaw Recovery** | 1 to 2 weeks |
| **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.<br>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.<br>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.<br>4. Perform all subsequent steps under sterile conditions in a flow hood, disinfecting the cryovial with 70% ethanol before opening.<br>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.<br>6. Centrifuge the mixture at 300 x g for 3 minutes to separate the cells and carefully discard the supernatant containing residual freezing medium.<br>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.<br>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.