# CRFK

305534

## GENERAL INFORMATION

### Description

The CRFK (Crandell-Rees Feline Kidney) cell line is a well-established model derived from the renal cortex of a normal feline embryo. It is widely used in virology research, particularly for the study of feline viruses such as feline infectious peritonitis virus (FIPV) and feline herpesvirus (FHV-1). CRFK cells are highly permissive to various feline viruses, which has made them invaluable for vaccine development and virological assays. For instance, these cells are used in the production and testing of live-attenuated vaccines, like the FVRCP vaccines, which protect against feline rhinotracheitis, calicivirus, and panleukopenia. The cell line's role in viral replication studies has revealed key insights into host-virus interactions and virus-induced cytopathic effects, such as syncytia formation and changes in cellular morphology.
Recent research has highlighted challenges associated with using CRFK cells for vaccine development due to contamination risks posed by endogenous retroviruses (ERVs). Specifically, CRFK cells can produce infectious RD-114 virus, a feline endogenous retrovirus, through recombination events. To address these safety concerns, genome-editing technologies like TALENs have been employed to knock out the env genes of the RD-114-related sequences, reducing the risk of ERV contamination in vaccine production. These modifications do not compromise the viability of CRFK cells, and the engineered RDKO_CRFK cells have shown improved safety profiles for use in manufacturing biologics.
Moreover, transcriptomic analyses of CRFK cells have demonstrated their differential gene expression profiles in response to viral infections. Upon infection with FIPV, CRFK cells upregulate various immune-related pathways, including Toll-like receptor signaling, MAPK, and JAK-STAT pathways, while also displaying significant downregulation of pathways involved in neuronal and cellular organization. These expression changes reflect the innate immune responses activated in epithelial cells and highlight differences compared to primary immune cells, such as macrophages, which exhibit distinct host responses under similar conditions. Such findings underscore the utility of CRFK cells in modeling early-stage host-pathogen interactions and evaluating antiviral strategies.

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| **Tissue** | Kidney |

### Applications

Widely used to manufacture vaccines for parvoviruses such as feline panleukopenia virus (FPLV) and canine parvoviruses (CPVs)

### Synonyms

- CrFK
- Crfk
- NBL-CrFK
- Crandell Reese Feline Kidney
- CFK
- Crandell Feline Kidney
- CCC
- Crandell's Cat Cell

## CLASSIFICATION

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| **Organism** | Cat |
| **Sex** | Female |
| **Growth properties** | Adherent |
| **Biosafety level** | 1 |

## CHARACTERISTICS

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| **Morphology** | Epithelial-like |

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## IDENTIFIERS

### Citation

CRFK (Cytion catalog number 305534)

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|---|---|
| **NCBI TaxID** | 9685 |
| **Cellosaurus** | CVCL_2426 |

## EXPRESSION AND SUSCEPTIBILITY

### Viruses

Persistently infected by feline endogenous virus RD-114

### Virus susceptibility

Feline panleukopenia virus, Feline calicivirus, Reovirus (Feline), Felid herpesvirus 1, Canine parvovirus, Rabies virus, Bovine viral diarrhea virus 1

## HANDLING AND CULTURE

### Culture medium

EMEM (MEM Eagle), w: 2 mM L-Glutamine, w: 2.2 g/L NaHCO3, w: EBSS (Cytion article number 820100a)

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| **Incubation atmosphere** | 37°C, 5% $CO_2$, humidified atmosphere. |

### Supplements

Supplement the medium with 10% FBS and 1% NEAA

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| **Dissociation reagent** | Accutase |

### 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.

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| **Fluid renewal** | 2 to 3 times per week |

### Thawing and culturing

Confirm that the vial remains deeply frozen upon delivery, as cells are shipped on dry ice to maintain optimal temperatures during transit.
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.
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.
Perform all subsequent steps under sterile conditions in a flow hood, disinfecting the cryovial with 70% ethanol before opening.
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.
Centrifuge the mixture at 300 x g for 3 minutes to separate the cells and carefully discard the supernatant containing residual freezing medium.
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.
Adhere to established subculture protocols for continued growth and maintenance of the cell line, ensuring reliable experimental outcomes.

## QUALITY CONTROL AND CRYOPRESERVATION

### 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.

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| **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. |

**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.

**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.

## DONOR

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| **Donor age** | 3 months |

## LEGAL AND SAFETY

**Safety precautions**

When planning to store a cryovial in liquid nitrogen for future thawing, it is mandatory to adhere to stringent safety measures. Appropriate protective gloves and clothing are essential, and the use of a face mask or safety goggles is required during the transfer of frozen samples to or from the liquid nitrogen tank. This is to mitigate the risk of injury from potential cryovial explosions upon removal, which can result in the projection of sharp fragments.

**Warranty**

At Cytion, we stand behind the quality and viability of our products.
We guarantee that our cells will be viable at the start of culture for
60 days after shipment from our facilities.
We assure that all materials conform to the specifications described in the
Certificate of Analysis and/or Product Sheet.
For full details, please refer to our
Warranty Statement.

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| **Material Transfer Agreement required** | Yes |

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