

PLC/PRF/5 Cells | 300315**General information**

Description	The cells produce HBsAg. At present, there is no evidence that this cell line produces infectious Hepatitis B virus.
Organism	Human
Disease	Hepatocellular carcinoma
Synonyms	PLC-PRF-5, PLC PRF 5, PLC/PRF5, PLCPRF5, PLC-8024, PLC8024, PLC, Alexander cells, Alexander, Primary Liver Carcinoma/Poliomyelitis Research Foundation/5

Characteristics

Age	24 years
Gender	Male
Ethnicity	African
Cell type	Alexander cells
Growth properties	Adherent

Regulatory Data

Citation	PLC/PRF/5 (Cytion catalog number 300315)
Biosafety level	2
NCBI_TaxID	9606
CellosaurusAccession	CVCL_0485

Biomolecular Data

Products	Hepatitis virus B surface antigen (HBsAg)
Karyotype	Mean number: 56, heteroploid, with marker chromosomes

PLC/PRF/5 Cells | 300315**Handling**

Culture Medium	DMEM, w: 4.5 g/L Glucose, w: 4 mM L-Glutamine, w: 3.7 g/L NaHCO ₃ , w: 1.0 mM Sodium pyruvate (Cytion article number 820300a)
Supplements	Supplement the medium with 10% FBS
Dissociation Reagent	Accutase
Doubling time	35 to 40 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	1×10^4 cells/cm ²
Fluid renewal	2 times per week
Post-Thaw Recovery	After thawing, plate the cells at 5×10^4 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.

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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% CO₂, 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

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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.