

Lec1 Cells | 305010

General information

Description

The Lec1 cell line is a mutant clone selected for its resistance to wheat germ agglutinin, derived from the parental CHO clone Pro-5. This selection process resulted in a cell line with a specific glycosylation defect, characterized by the presence of N-linked carbohydrates with a blocked Man5-GlcNAc2-Asn intermediate. This blockage is due to the absence of N-acetylglucosaminyltransferase I (GlcNAc-TI), an enzyme critical for the progression of glycan synthesis to more complex forms. As a result, Lec1 cells accumulate glycoproteins with truncated, high-mannose type oligosaccharides.

Lec1 cells are invaluable for the study of glycoprotein biosynthesis, particularly in understanding how altered N-linked glycosylation affects cell function. Researchers utilize Lec1 cells to investigate the impact of glycosylation on protein folding, stability, receptor function, and intracellular trafficking. Additionally, these cells provide a unique platform for studying the compartmentalization of viral infection-induced or foreign DNA transfection-induced endogenous glycoproteins. The simplified glycan structures in Lec1 cells also make them ideal for producing glycoproteins that are easier to analyze in various experimental contexts.

They are primarily used in vitro for mechanistic studies and biotechnological applications involving glycoprotein production and analysis.

Organism Chinese hamster

Tissue Ovary

Synonyms CHO-Lec1, CHO Lec1, Pro-Lec1.3C, Pro-5 Lec1.3c, Pro-5WgaRI3C

Characteristics

Age Adult

Morphology Epithelial

Growth properties Adherent

Regulatory Data

Citation Lec1 (Cytion catalog number 305010)

Biosafety level 1

NCBI_TaxID 10029

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CellosaurusAccession CVCL_3440

Biomolecular Data**Handling**

Culture Medium Alpha MEM, w: 2.0 mM stable Glutamine, w/o: Ribonucleosides, w/o: Deoxyribonucleosides, w: 1.0 mM Sodium pyruvate, w: 2.2g/L NaHCO₃

Supplements Supplement the medium with 10% FBS

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.

Seeding density 2 to 4 x 10⁴ cells/cm²

Fluid renewal 2 to 3 times per week

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 $300 \times g$ 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°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

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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.