MDCK [NBL-2] Cell Line

Catalog No.: AMS.EP-CL-0154

Origin and General Characteristics

Cell Name	MDCK [NBL-2] Cell Line
Organism	Canis familiaris
Age	Adult
Tissue	Kidney
Morphology	Epithelial
Growth Properties	Adherent
Descriptions	This line is hyperdiploid and there is a bi-modal chromosome number distribution. There are no consistent identifiable marker chromosomes. One normal X chromosome is present in most spreads.The cells are positive for keratin by immunoperoxidase staining. MDCK cells have been used to study processing of beta amyloid precursor protein and sorting of its proteolytic products.
Biosafety Level	1

Culture Conditions and Handling

Complete Growth Medium	MEM +10% FBS +1% P/S
Subculturing	Remove and discard culture medium. Briefly rinse the cell layer with DPBS solution to remove all traces of serum that contains trypsin inhibitor. Add 1.0 to 2.0 mL of Trypsin-EDTA solution to flask and observe cells under an inverted microscope until cell layer is dispersed (usually within 2 to 3 minutes). Cells that are difficult to detach may be placed at 37°C to facilitate dispersal. Add 4.0 to 6.0 mL of complete growth medium and aspirate cells by gently pipetting. Add appropriate aliquots of the cell suspension to new culture vessels.
Subcultivation Ratio	1:2-1:4
Medium Renewal	Every 2 to 3 days
Cryopreservation	Freeze medium: 60% Basal medium+30% FBS+10% DMSO Storage temperature: Liquid nitrogen vapor phase
Culture Conditions	Atmosphere: Air, 95%; CO2, 5% Temperature: 37°C

Special Features of the Cell Line

Gene Expression	The Cells Are Positive For Keratin By Immunoperoxidase Staining.
Applications	This cell line is a suitable transfection host and is useful for influenza research.

Recommendations for handling of cryopreserved cells

1. The cell is packaged by dry ice. When receiving the cell, please make sure that the vial is still frozen. If there is cell thawing in the tube, please take photo before experiment or storage.
2. If immediate culturing is not intended, the cryovial(s) must be stored in liquid nitrogen (-196°C) or at least at

-80°C after arrival.

If immediate culturing is intended, please follow these instructions:

3. Quickly thaw by rapid agitation in a 37°C water bath within 45-90 seconds. The water bath should have clean water containing an antimicrobial agent. As soon as the sample has thawed, remove the cryovial from the water bath.
   From now on, all operations should be carried out under aseptic conditions.
4. Transfer the cryovial to a sterile flow cabinet and wipe with 70% alcohol. Carefully open the vial and transfer the cell suspension into a 15 ml centrifuge tube containing 9 ml of cell complete medium (room temperature or 37°C).
5. In order to reduce cell damage, add 1ml of cell complete medium into cryovial, slightly pipette, then use a pipettor to add 1 ml of suspension into the centrifuge tube. Resuspend the cells carefully. Centrifuge at 300×g for 3 min and discard the supernatant. The centrifugation step may be omitted, but in this case the remains of the freeze medium have to be removed 24 hours later.
6. Resuspend the cells carefully in 10ml fresh cell culture medium and transfer them into one or two T25 cell culture flasks.

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