

<u>1 X 10⁶ cells</u> **Lot #** <u>W433</u>

Rat Glomerular Epithelial Cells (CRGEp-1)

Materials supplied

This package contains 1.0 X 10⁶ cells in 1 ml of cryoprotective medium (Basal Eagle's medium with Hank's BSS, 5% FBS, 7.5% DMSO) plus antibiotic/antimycotic.

Description

Probetex renal glomerular epithelial (CRGEp-1) cells are derived from male Sprague-Dawley rats. The cells were grown from isolated decapsulated adherent glomeruli in culture. The cells were characterized to be glomerular epithelial based on phenotypic criteria of a polyhedral shape and growth in islands (Figure 1), expression of tight junction proteins such as (ZO-1), and the ability to form "domes" when grown on plastic. The cells stain positive by immunofluorescence for cytokeratin, desmin, vimentin, ezrin, and the specific glomerular epithelial marker synaptopodin (Figure 2). The cells are also susceptible to the cytotoxic effects of aminonucleoside. The cells are negative for mesangial and endothelial markers. Each stock can quickly re-animate with a doubling time of 12 hours.

Directions for use

Please read carefully before starting re-animation.

<u>Re-animation:</u> Thaw the cells quickly in a 37° C waterbath. Wipe down the outside with 70% ethanol before bringing the vial into your cell culture hood. Transfer the cells to 10 ml of warm growth medium. Centrifuge the cells at ~400 x g for 5 min. Resuspend the cells in a total of 20 ml growth medium (RPMI-1640, 10% Fetal Bovine Serum, with or without antibiotics) and transfer to a T-75 flask. Incubate at 37° C and 5% CO₂. Replace medium 24-48 hours after re-animation.

<u>Subculturing</u>: Remove the medium from confluent, viable cells. Rinse with Mg²⁺/Ca²⁺-free Phosphate-Buffered Saline (PBS). Detach the cells by addition of 0.25% trypsin/EDTA

solution and incubate the cells for up to 10 min at 37°C. Add growth medium and collect the cells. Centrifuge the cells at 400 x g for 5 min. Resuspend the cells in fresh growth medium. 8-fold to 10-fold split ratios are recommended.

<u>Cryopreservation</u>: Treat the cells the same way as for subculturing. Determine the number of viable cells and resuspend them at 2.0 X 10⁶ cells/ml in growth medium. Add an equal volume of cryoprotective medium for a recommended final DMSO concentration of 7.5%. The recommended storage condition is liquid nitrogen vapor phase.

<u>Adventitious Agents</u>: This product is certified free of bacteria, fungi and mycoplasma.

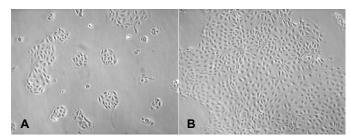
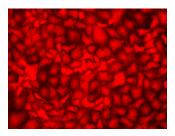


Figure 1. Phase contrast micrograph showing growth in a polyhedral shape and growth pattern of small "islands" (A) and near confluence (B).

Figure 2. Localization of the glomerular epithelial marker synaptopodin by immuno-fluorescence.



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