PROBETEX
Experimental Pathology Resources™

Data Sheet

1 X 10^6 cells

Lot # W452

Rat Glomerular Mesangial Cells (CRGMes-1)

Materials supplied:

This package contains 1.0 X 10^6 cells in 1 ml of cryoprotective medium (Basal Eagle's medium with Hank's BSS, 5% FBS, 7.5% DMSO) plus antibiotic/antimycotic.

Description

Rat glomerular mesangial (CRGMes-1) cells were isolated by differential sieving from the cortex of Sprague Dawley kidneys. The cells were identified and their purity was assessed by phase-contrast microscopy showing a stellate, kite-like shape (Figure 1A) in subconfluent culture. Immunofluorescence microscopic evaluation of phenotypic markers show positive staining for Thy1.1 antigen, alpha-smooth muscle actin (Figure 1B), desmin, vimentin, and fibronectin. The cells are negative for endothelial and epithelial markers factor VIII and cytokeratin, respectively. Each stock re-animates with a doubling time of approximately 12 hours.

Directions for use

Please read carefully before starting re-animation.

Re-animation: Thaw the cells quickly in a 37°C water bath. 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% CO_2. Replace medium 24-48 hours after re-animation.

Subculturing: Remove the medium from confluent, viable cells. Rinse with Mg^{2+}/Ca^{2+}-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 suggested.

Cryopreservation: Treat the cells the same way as for subculturing. Determine the number of viable cells and resuspend them at 2.0 X 10^6 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.

Adventitious Agents:

This product is certified free of bacteria, fungi, and mycoplasma.

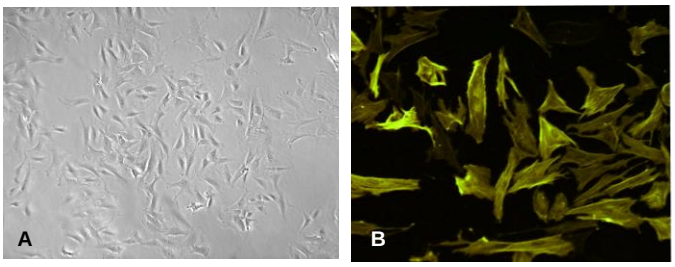

Figure 1. (A) Phase contrast micrograph of CRGMes-1 cells near confluence showing stellate, kite-like cell shape. (B) Fluorescence microscopy shows positive α-smooth muscle actin.

1. Barnes JL, Hevey KA. Glomerular mesangial cell migration in response to platelet-derived growth factor. Laboratory Investigation. 62:379-382, 1990.
2. Barnes JL, Hevey KA. Glomerular mesangial cell migration. Response to platelet secretory products. American Journal of Pathology. 138:859-866, 1991.

Probetex, Inc.
7418 John Smith, Suite A
San Antonio, TX 78229

(210) 616-9515 FAX: (210) 616-9914
e-mail: info@probetex.com