PROBETEX
Experimental Pathology Resources™

Data Sheet

1 X 10^6 cells

Lot # W501

Rat Metanephric Mesenchymal Cells (CRMM-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

Probetex rat metanephric mesenchymal (CRMM-1) cells are derived from metanephric mesenchyme at gestational day 13 (Arar et al, 2000). The cells were identified and their purity was assessed by phase-contrast microscopy showing a stellate cell shape (Figure 1) in low density and expression phenotypic markers vimentin (Figure 2) by immunofluorescence microscopy. The cells are negative for the epithelial cell markers pancytokeratin, Endo A cytokeratin and UB marker *Dolichos biflorus*. When combined with mouse ureteric cells (CMUB-1), CMMM-1 cells organize into capillary-like structures, cysts and tubuloid arrangements in three-dimensional growth in Matrigel® implanted subcutaneously into SCID mice (Barnes et al, 2008). Each stock re-animates with a doubling time of approximately 12 hours.

Directions for use

Please read carefully before starting re-animation.

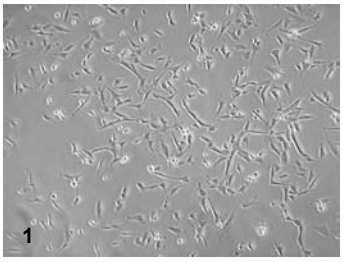
Re-animation: Thaw the cells quickly in a 37°C water bath. Wipe down the outside with 70% ethanol before bringing the vial into your cell culture hood. Transfer the cells to 10 ml of warm growth medium. Centrifuge the cells at ~400 x g for 5 min. Resuspend the cells in a total of 20 ml growth medium (DMEM high glucose, 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 0.05% 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. 10-20-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.



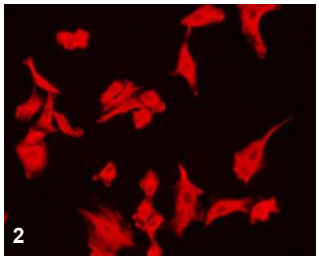


Figure 1. Phase contrast micrograph of CMMM-1 cells showing cells in a stellate shape. Figure 2. Fluorescence microscopy of CMMM-1 displaying a mesenchymal marker vimentin.

Arar M, Xu Y, Elshihabi I, Barnes JL, Ghosh Choudhury, G, Abboud HE: Platelet-derived growth factor receptor-β regulates migration and DNA synthesis in metanephric mesenchymal cells. J Biol Chem 275:9527-9533, 2000.

Barnes JL, Velagapudi C, Nilsson R-P, Barnes VL, Arar M, Abboud HE: Induction of tubulogenesis by co-culture of mouse kidney metanephric mesenchymal and ureteric bud stem cell lines. J Am Soc Nephrol. 19:104A, 2008.

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