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BCRJ Code: 0400

Cell Line: CCD-18Co

Species: Homo sapiens

Vulgar Name: Human

Tissue: Colon

Morphology: **Fibroblast**

Disease: Normal

Growth Properties: Adherent

Sex: Female

Age/Ethinicity: 2 Month / Black

Biosafety: 1

Addtional Info: The line begins to senesce at about PDL = 42.

Culture Medium: DMEM Low glucose + fetal bovine serum to a final concentration of 10%.

> Remove medium, and rinse with 0.25% trypsin, 0.053 mM EDTA solution. Remove the solution and add an additional 1 to 2 mL of trypsin-EDTA solution. Allow the flask to sit at room temperature (or at 37°C) until the cells detach. Add fresh culture medium, aspirate and dispense into new culture flasks. Note: Growth of the cells is enhanced by addition of tumor necrosis factor

alpha (TNF alpha) to the medium.

Subculturing Medium

Renewal:

Subculturing:

Every 2 to 3 days

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Subculturing

Subcultivation Ratio:

1:2 to 1:3

Culture Conditions:

Atmosphere: air, 95%; carbon dioxide (CO2), 5% Temperature: 37°C

SAFETY PRECAUTION: Is highly recommend that protective gloves and

clothing always be used and a full face mask always be worn when handling frozen vials. It is important to note that some vials leak when submersed in

Cryopreservation:

Thawing Frozen Cells:

95% FBS + 5% DMSO (Dimethyl sulfoxide)

liquid nitrogen and will slowly fill with liquid nitrogen. Upon thawing, the conversion of the liquid nitrogen back to its gas phase may result in the vessel exploding or blowing off its cap with dangerous force creating flying debris. 1. Thaw the vial by gentle agitation in a 37°C water bath. To reduce the possibility of contamination, keep the Oring and cap out of the water. Thawing should be rapid (approximately 2 minutes). 2. Remove the vial from the water bath as soon as the contents are thawed, and decontaminate by

the water bath as soon as the contents are thawed, and decontaminate by dipping in or spraying with 70% ethanol. All of the operations from this point on should be carried out under strict aseptic conditions. 3. For cells that are

sensitive to DMSO is recommended that the cryoprotective agent be removed immediately. Transfer the vial contents to a centrifuge tube containing 9.0 mL complete culture medium and spin at approximately 125 x g for 5 to 7 minutes. 4.Discard the supernatant and Resuspend cell pellet with the

recommended complete medium (see the specific batch information for the culture recommended dilution ratio). 5. Incubate the culture in a appropriate atmosphere and temperature (see "Culture Conditions" for this cell line). NOTE: It is important to avoid excessive alkalinity of the medium during recovery of the cells. It is suggested that, prior to the addition of the vial

contents, the culture vessel containing the growth medium be placed into the

incubator for at least 15 minutes to allow the medium to reach its normal pH

(7.0 to 7.6).

Sugarman BJ, et al. Recombinant human tumor necrosis factor-alpha: effects on proliferation of normal and transformed cells in vitro. Science 230:

943-945, 1985. PubMed: 3933111 Hinterleitner TA, et al. II-1 stimulates intestinal myofibroblast COX gene expression and augments activation of CI-secretion in T84 cells. Am. J. Physiol. 271: C1262-C1268, 1996. PubMed:

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ATCC: CRL-1459

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References:







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