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DATA SHEET

Human Normal Kupffer Cell Culture - Frozen Vial

Culture – Frozen Vial

Catalog number: 36167-17N

Description: Frozen Ampule (1.2×10^6 cells) of 1×10^6 viable cells upon thawing, shipped with dry-ice. Also available in T25, T75, T150, and T225 tissue culture flask with plated cells, shipped at room temperature. The Clonal selected Human Normal Kupffer Cell Culture was derived from Human Normal liver tissue. They were maintained in Celprogen's Human Normal Kupffer Cell Culture Complete Growth Media and sub-cultured every 24 to 48 hours on Human Normal Kupffer Cell Culture Extracellular Matrix.

Source: Human Normal Liver Tissue

Donors: All donors from which the Cells were derived were pre-screened; donors tested negative for the usual blood donation infectious disease panel (ABO/RH, Hepatitis B Surface Antigen, HIV 1 and 2, Syphilis, hepatitis B Core, Human T Lymphocyte Virus 1 and 2, Hepatitis C Virus, Antibody Screen, Nucleic Amplification Test for HIV 1 HCV, West Nile Virus and Antibodies to Trypanosoma cruzi (the agent of Chagas disease).

Mycoplasma test: Negative-PCR and mycoplasma agar methods

Sterility: Negative for bacteria, yeast, and mold

Storage Conditions: Liquid nitrogen vapor phase for frozen Ampule of Human Normal Kupffer Cell Culture. For plated cells in tissue culture flask, upon receipt of the cells wipe the flask with 70% ethanol and transfer to sterile tissue culture hood. In the tissue culture hood remove the media from the cells and wash the cells with 1X PBS sterile solution, for 2-3 minutes, remove the 1X PBS solution and then Trypsinize. After Trypsinization of the Cells neutralize the Trypsin with equal volume of Human Normal Kupffer Cell Culture Complete Growth Media with Serum and collect the Cell suspension in sterile conical centrifuge tube in the tissue culture hood. Centrifuge the cell suspension at 100g for 7 minutes in centrifuge. Plate cells 5×10^5 cells per pre-coated flasks with Human Normal Kupffer Cell Culture Extracellular Matrix for Expansion in Human Normal Kupffer Cell Culture Complete Growth Medium.

Positive Markers: CD68, TNF-a, IL-6, CD34, F4/80 Antibody



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**Morphology
& Proliferation:**

Mixed population of cells with approximately 95% attached cells and the other 5.0% in suspension, need to change cell culture media every day after 48 hours of initial cell culture or when the media starts changing color to slight yellow from pink. Fast growing cell culture, change media with Celprogen's Human Normal Kupffer Cell Culture Complete Growth Media with the appropriate Human Normal Kupffer Cell Culture Extracellular Matrix. Temperature 37⁰C in 5% CO₂ humidified incubator.

Sub-culturing:

1. Thaw the vial with gentle agitation in a 37⁰C water bath or a dry 37⁰C shaking incubator. For water bath thawing keep the O-ring out of the water.
2. Remove the thawed vial and wipe with 70% ethanol. Then transfer to the tissue culture hood.
3. Transfer the vial contents to a sterile centrifuge tube, and gently add pre-warmed Human Normal Kupffer Cell Culture Complete Growth Media to the centrifuge tube. Use additional Human Normal Kupffer Cell Culture Complete Growth Media to rinse the vial and transfer the liquid to the centrifuge tube, repeat this once more to ensure you have all the cells transferred to the 15ml centrifuge tube. Centrifuge the cells at 100g for 7 minutes. Remove the supernatant and re-suspend the cell pellet in 500ul of Human Normal Kupffer Cell Culture Complete Growth Medium.
4. Add the 500ul of cells to T75 flask pre-coated with Human Normal Kupffer Cell Culture Extracellular Matrix with 10ml of Human Normal Gall Kupffer Cell Culture Complete Growth Medium.
5. Incubate the cells in the T75 flask at 37⁰C in a 5% CO₂ humidified incubator. Perform 100% Media Change every 24 to 48 hours.
6. Medium renewal every other day or 2-3 days, sub-culturing ratio: 1:2 or 1:3 depending on cell density.
7. Refer to protocols, flow diagrams and videos for more detail. <http://celprogen.com/tech.htm>

Freezing Medium: Available for purchase Cat# M36167-17NFM

Trypsin: Available for purchase Cat# T1509-014

IX PBS: Available for Purchase Cat# P1408-013

Storage temperature: Liquid nitrogen vapor phase

Product Orders: Before submitting an order you will be asked to read and accept the terms and conditions of Celprogen's Material Transfer Agreement (MTA).

Permits/Forms: In addition to the MTA mentioned above, other CELPROGEN and/or regulatory permits may be required for the transfer of this CELPROGEN material. Anyone purchasing CELPROGEN material is ultimately responsible for obtaining the permits.



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**Notices
& Disclaimers:**

CELPROGEN products are intended for laboratory research purposes only. They are not intended for use in Humans. The product, Human Normal Kupffer Cell Culture – Frozen Vial is established and manufactured by CELPROGEN Inc., and is for Research Use Only. This product is not for resale or may not be transferred to a third party prior to written request and approval by CELPROGEN Inc.