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DATA SHEET

Product name:	Mouse Osteoblast -Plated T75

Catalog number: 11039-11-T75

Description:
Osteoblast T75 Plated Cells. Also available in T25, T150, and T225 tissue culture flasks with plated Cells, shipped at room temperature or Frozen vial shipped in Dry-ice. They were maintained in Celprogen's Osteoblast Complete Growth Medium and sub-cultured every 24 to 48 hours on Osteoblast Extra-cellular Matrix.

Source: Mouse Bone Marrow

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 Osteoblast . 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 of the cells and wash the cells with 1X PBS sterile solution, for 2-3 minutes, remove the PBS solution and then trypsinize. After trypsinization of the Cells neutralize the trypsin with equal volume of Osteoblast 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 $5x10^5$ cells per pre-coated flasks with Osteoblast Extra-cellular Matrix for Expansion in Osteoblast Complete Growth Medium.

Tests Performed:
Microbial: Negative
Incorporation of acetylated Low Density Lipoprotein (LDL): Positive

Positive Markers:
Alkaline Phosphatase, Fibronectin, IGFBP-3, Osteocalcin, Collagen 1, RUNX2, CD38

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 for pink. Fast growing cell culture. Change media with Celprogen's Osteoblast Complete Growth Medium with the appropriate Osteoblast Extra-cellular Matrix. Temperature 37^0C in 5% CO_2 humidified incubator.

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Sub-culturing:

1. Thaw the vial with gentle agitation in a 37^0C water bath or a dry 37^0C 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 Osteoblast Growth Media to the centrifuge tube. Use additional Osteoblast Complete 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 1500 RMP for 5 minutes. Remove the supernatant and re-suspend the cell pellet in 500ul of Osteoblast Complete Growth Medium.
4. Add the 500ul of cells to appropriately sized flask pre-coated with Osteoblast Extra-cellular Matrix with 15ml of Osteoblast Complete Growth Medium.
5. Incubate the cells in the T75 flask in a 37^0C in 5% CO_2 humidified incubator. Perform 100% Media Change every 24 to 48 hours.
6. Medium renewal every other or 2-3 days, sub-culturing ratio: 1:3

Freezing Medium: Available for purchase Cat# M11039-11FM

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.

Biosaftely Level 1

Notices & Disclaimers:

***CELPROGEN products are intended for laboratory research purposes only*. They are not intended for use in humans.**
This Product Osteoblast established and manufactured by CELPROGEN Inc., and is for Research Use Only. This product is not for re-sale or may not be transferred to third party prior to written request and approval by CELPROGEN Inc.