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IMMORTALIZED CELL LINES

P10890-IM

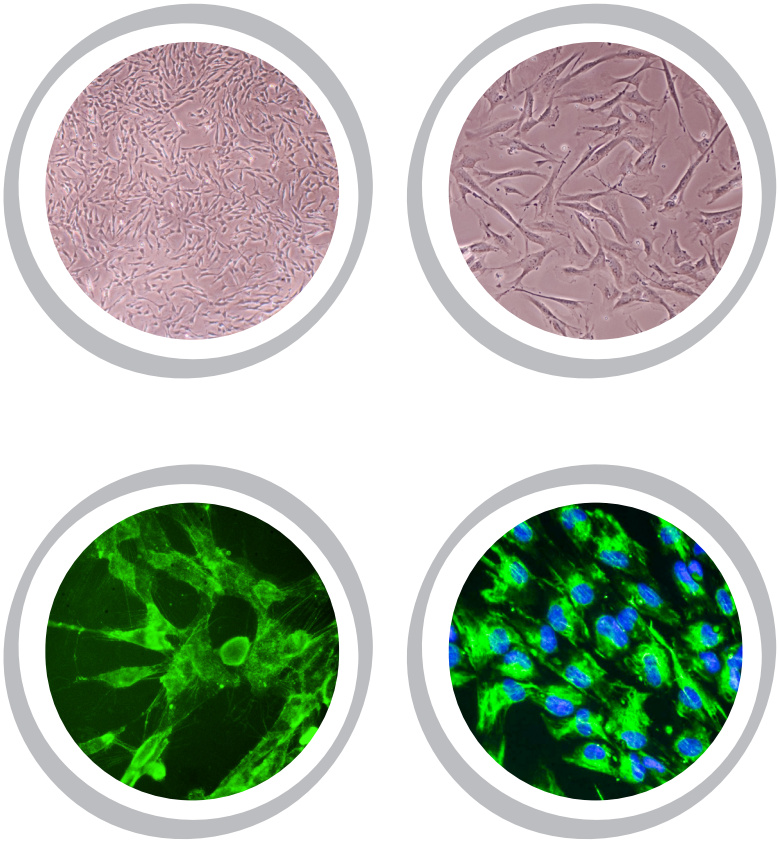
Human Dental Pulp Mesenchymal Stem Cells (hDPSCs) are multipotent cells residing within the dental pulp tissue. They possess the ability to differentiate into various mesenchymal lineages, including osteogenic, chondrogenic, and adipogenic fates, and exhibit immunomodulatory properties. These cells are of increasing interest for regenerative medicine and tissue engineering applications due to their accessibility, plasticity, and paracrine signaling potential. Immortalized hDPSCs represent a valuable tool for studying stem cell biology, dental tissue regeneration, and for the development of advanced cell-based therapeutic strategies.

Human Dental Pulp Mesenchymal Stem Cells were immortalized using two lentiviral vectors: one expressing SV40 large T antigen and another co-expressing BMI1 and HPV E7. Unlike primary cells, which undergo replicative senescence, the transduced cells showed stable proliferation beyond 30 passages, demonstrating effective immortalization through combined targeting of cell cycle and senescence pathways.

IMMORTALIZED HUMAN STEM CELLS

DENTAL PULP MESENCHYMAL

Product Type: Immortalized Human Dental Pulp Mesenchymal Cells
Catalog Number: P10890-IM
Immortalization: SV40 Large T Antigen, BMI1 and HPV E7. G418 resistant.
Number of cells: >1x10^6 cells (cryopreserved vials)
Storage: Liquid Nitrogen
Recommended Medium: Mesenchymal Stem Cell Medium Kit (Ref: P60115)
Product Characterization: Positive for CD105 and Vimentin.



CD105 Vimentin

IMMORTALIZED CELL LINES

About Immortalized Human Dental Pulp Mesenchymal Stem Cells

Immortalized Human Dental Pulp Mesenchymal Stem Cells were generated by transducing primary cells with two types of lentiviral vectors: one expressing the SV40 large T antigen (Lenti-SV40) and another co-expressing BMI1 and HPV E7 (Lenti-BMI1-E7). Immortalized cells were cultured in parallel with primary cells to assess their proliferation capacity. While primary cells underwent replicative senescence after a few passages, the dual-transduced cells exhibited stable and long-term proliferation, having been expanded beyond 30 passages without signs of growth arrest. The SV40 large T antigen promotes immortalization by inactivating tumor suppressor proteins such as p53 and Rb, while BMI1 and E7 further enhance cell cycle progression and prevent senescence, thereby ensuring continuous cell proliferation. This combination of viral oncoproteins efficiently bypasses critical cell cycle checkpoints, making it particularly useful for generating immortalized cell lines. The resulting immortalized cells maintain consistent morphology, a stable proliferative phenotype, and are suitable for long-term in vitro applications, providing a valuable resource for further studies in regenerative medicine and tissue engineering.

Culturing conditions

1 IMMEDIATELY UPON DELIVERY

1.1 Remove the vial from the shipping container to check for freezing.

1.2 Transfer the frozen vial to liquid nitrogen until ready to thaw.

2 THAWING CELLS:

2.1 Prepare a fibronectin coated flask (2 µg/cm2, T-75 flask is recommended). Add 10 ml of sterile Dulbecco's phosphate buffered saline (DPBS) to a T-75 flask and then add 150 µl of fibronectin stock solution (1 mg/ml, Innoprot cat. no. P8248). Leave the flask in incubator overnight.

2.2 Prepare "Thawing medium" by combining 500 ml of basal medium, 25 ml of fetal bovine serum, 5 ml of Growth supplement and 5 ml of penicillin/streptomycin solution.

2.3 Thaw cells rapidly in a 37ºC water bath; avoid allowing the sample to warm to 37ºC. Cryovials should be cool to the touch when removed.

2.4 Remove the vial, wipe it dry, and transfer it to a sterile field.

2.5 Rinse the vial with 70% ethanol, then wipe to remove excess. Open the vial and resuspend its contents using a 1 ml Eppendorf pipette.

2.6 Dispense the contents into a 25 cm^2 culture flask with warm complete media (FBS percentage can be increased up to 10% for better culture establishment).

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Culturing conditions

2.7 Place the flask in the incubator.

2.8 For optimal results, avoid disturbing the culture for 16 hours after initiation. Change the growth medium the next day to remove unattached cells, then every other day thereafter.

3 MAINTENANCE OF THE CULTURE:

3.1 Change medium 48 hours after establishing a subculture.

3.2 Subculture when cells are over 90% confluent.

4 SUBCULTURING:

Remove medium, rinse with 0.05% trypsin-EDTA solution. Add 1 to 2 mL of trypsin-EDTA solution and allow the flask to sit until cells detach. Add fresh culture medium, aspirate, and dispense into new culture flasks.

Recommended subcultivation ratio of 1:2 to 1:6.

Medium Renewal: 2 to 3 times per week.

Reagents for cryopreservation: Cryostor S10.

Quality Control / Biosafety

The cells test negative for HIV-1, HBV, HCV, mycoplasma, bacteria, yeast and fungi.

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