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Product Information

Human Adipose Derived Stem Cells (hADSC)

Catalog Number	10HU-001 (Normal) 10HU-222 (Type 1 Diabetes) 10HU-007 (Type 2 Diabetes) 10HU-232 (Obesity)	Cell Number	0.5 million cells/vial; 1.0 million cells/vial (available for Normal)
Species	*Homo sapiens*	Storage Temperature	Liquid Nitrogen

Description

Human Adipose-Derived Stem cells (ADSCs) are isolated from human lipoaspirate tissue collected during elective surgical liposuction procedures. ADSCs are available for normal donors, patients with Type 1 Diabetes, Type 2 Diabetes, or Obesity (BMI>30). It has been shown that the ADSCs demonstrate very similar phenotypic and functional characteristics to that of bone marrow-derived mesenchymal stem cells. Thousands of articles have been published on ADSCs using a variety of terminology, including adipose-derived mesenchymal stem cells (AD-MSCs), adipose MSCs (AMSCs), adipose-derived adult stem (ADAS) cells, and adipose stromal/stem cells (ASCs). Normal human ADSCs have been reported to differentiate into many different lineages including chondrogenic, osteogenic, adipogenic and neural. And have been applied in studies include stem cell differentiation, regenerative medicine [1], and cell therapy [2].

iXCells Biotechnologies offers normal human adipose-derived stem cells (hADSC) from adipose tissues from single donor and cryopreserved at passage 1. hADSC are positive for CD29, CD44, CD73, CD90, CD105, and negative for CD14, CD31, CD45 (Figure 1). hADSC can be in vitro differentiated into adipocytes and osteoblasts (Figure 2 and 3) using Adipocyte Differentiation Medium (Cat # MD-0005) and Osteogenic Differentiation Medium (Cat # MD-0006). hADSC can be further expanded using Adipose-derived Stem Cell Growth Medium (Cat # MD-0003). These cells are negative for HIV-1, HBV, HCV, mycoplasma, bacteria, yeast, and fungi.

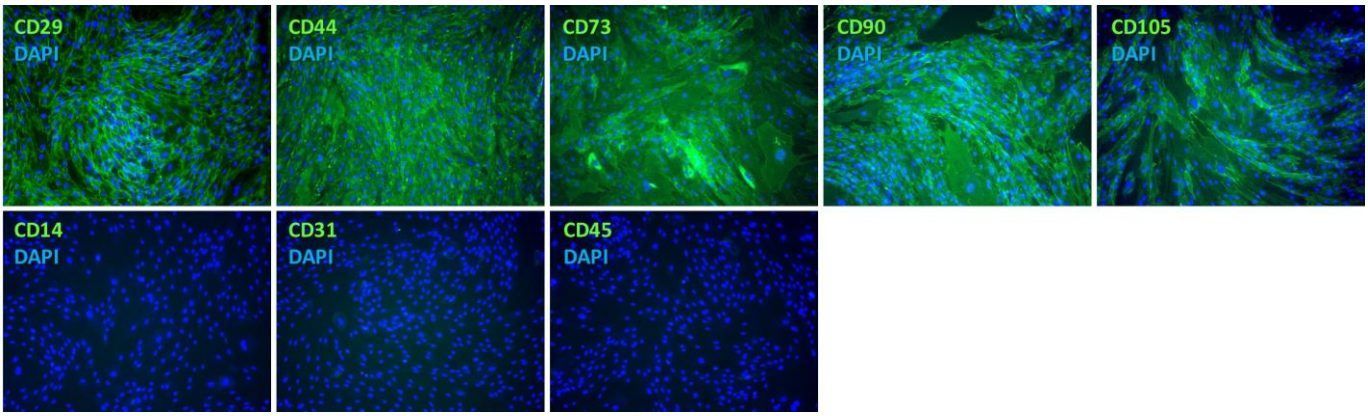

Figure 1. Immunostaining of cell surface markers of human ADSC.

Product Details

Tissue	Human adipose tissue from liposuction procedures
Package Size	0.5 million cells/vial; 1.0 million cells/vial
Passage Number	P1
Shipped	Cryopreserved
Storage	Liquid nitrogen
Growth Properties	Adherent
Media	Adipose-Derived Stem Cell Growth Medium (Cat # MD-0003) Adipocyte Differentiation Medium (Cat # MD-0005) Osteogenic Differentiation Medium (Cat # MD-0006)

Protocols

Standard Culture Procedure

1. Upon receipt of the frozen hADSC, it is recommended to thaw the cells and initiate the culture immediately in order to retain the highest cell viability.
2. To thaw the cells, put the vial in 37°C water bath with gentle agitation for ~1 minute. Keep the cap out of water to minimize the risk of contamination.
3. Pipette the cells into a 15ml conical tube with 5ml fresh Adipose-derived Stem Cells Growth Medium (Cat # MD-0003).
4. Centrifuge at 1,000rpm (~220g) for 5 minutes at room temperature.
5. Remove the supernatant and re-suspend the cells in fresh Adipose-derived Stem Cells Growth Medium.
6. Culture the cells in one 100 mm dish or one T75 flask. Change medium every 3~4 days.
7. When cells reach >85% confluence, freeze them or subculture cells as following
8. Aspirate the culture medium, and wash once with sterile PBS (5ml/T75 flask).
9. Add ~2 ml of 0.25% Trypsin-EDTA to the flask and incubate for ~3 minutes at 37°C. Neutralize the enzyme by adding 2-3 volumes of cell culture medium.
10. Centrifuge 1,000rpm (~220g) for 5min and re-suspend the cells in desired volume of medium.
11. Seed new culture vessels at 5×10^3 cells/cm^2.

Safety Precaution: *it is highly recommended that protective gloves and clothing should be used when handling frozen vials.*

Adipocyte Differentiation Protocol (12 well plate format)

1. Grow hADSCs in Adipose-Derived Stem Cell Growth Medium (Cat# MD-0003) to >95% confluency.
2. Aspirate the growth medium and replace with 1.5 ml fresh growth medium/well, let the cells grow for 2~3 more days.
3. Aspirate the growth medium, apply 1.5 ml Adipocyte Differentiation Medium (Cat# MD-0005) per well to the cells.

   Note: Cells at this stage may detach from dish easily, so do not use pump to aspirate off the medium at this step. Use pipet and slowly remove the medium instead. Add Adipocyte Differentiation Medium very gently to avoid cell detachment.
4. Change fresh Adipocytes Differentiation Medium every 3 days (slowly remove and add the medium as described above).
5. Culture hADSCs in Adipocytes Differentiation Medium for 10-14 days, and analyze the percentage of cells with oil-droplet formation by Oil Red O Staining (Figure 2).

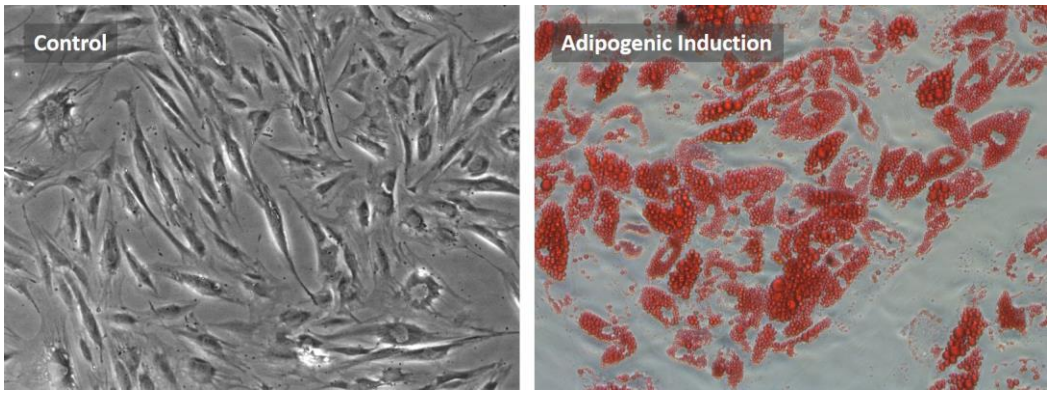

Figure 2. Human ADSC adipocyte differentiation (Day 14 post adipogenic induction).

Osteogenic Differentiation Protocol (12 well plate format)

1. Grow hADSCs in Adipose-Derived Stem Cell Growth Medium (Cat# MD-0003) to ~80% confluency.
2. Carefully aspirate the growth medium, then apply 1.5 ml Osteogenic Differentiation Medium per well (Cat# MD-0006) to the cells.
3. Change fresh Osteogenic Differentiation Medium every 3 days. Be careful not to disturb the cell monolayer.
4. Culture the hADSCs for 3-4 weeks, and osteoblasts can be detected by Alizarin Red S staining (stain the extracellular calcium deposit) (Figure 3).

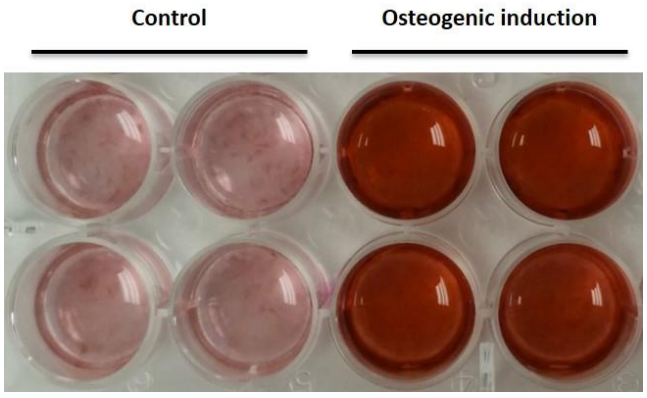

Figure 3. Human ADSC osteogenic induction (Day 24 post osteogenic induction). Alizarin Red S staining of osteoblasts. The extracellular calcium deposit was stained in bright orange-red color.

References

[1] Harasymiak-Krzyzanowska I et al. Adipose tissue-derived stem cells show considerable promise for regenerative medicine applications. Cell Mol Biol Lett. 2013; 18(4): 479-493.

[2] Bertheuil N, et al and Tarte K. Adipose-derived stromal cells: history, isolation, immunomodulatory properties and clinical perspective. Ann Chir Plast Esthet. 2015;60(2): 94-102.

Disclaimers