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ENDOTHELIAL DIFFERENTIATION PROMOTED BY HYPOXIA IN HUMAN AMNIOTIC FLUID-DERIVED STEM CELLS

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Methods We isolated HAFSCs from amniotic fluid of the second trimester of gestation. We observed the growth features

by MTT. We detected surface antigens by flow cytometry. Moreover, we detected mRNA levels of OCT-4, Nanog by RT-PCR, and Observed adipogenic and osteogenic differentiation. We focused on endothelial differentiation by detecting v WF, CD31 and CD133 by flow cytometry, and compared mRNA levels of VEGF, v WF and CD31 under hypoxia by real-time RT-PCR. We recorded capillary-like structures of the cells plated on Matrigel under hypoxia.

Results The cells liked fibroblast and expanded rapidly. The growth curves showed S shape. The mRNA levels of OCT-4, Nanog were observed. Flow cytometry analysis showed the cells were positive for CD29 CD44 CD73 CD90 HLA-ABC and negative for CD34 CD45 HLA-DR. After adipogenic and osteogenic differentiation, lipid droplets and calcium mineralisations were observed by Oil Red O staining and von Kossa staining. After endothelial differentiation, flow cytometry analysis showed 20% vWF 38% CD31 31% CD133. Real-time RT-PCR showed VEGF, v WF and CD31 mRNA levels increased significantly at 6h under hypoxia ($p<0.05$). The capillary-like structures were observed at 6 hours of hypoxia when plated the cells on Matrigel.

Conclusion HAFSCs can differentiate into endothelial cells with VEGF and bFGF. Under hypoxia conditions, within a short time the mRNA levels up-regulated along with time. The angiogenic capability were more significant compared with the control groups.