

P-24 Characterization of Human Multipotent Stem Cells Isolated from Second-trimester Amniotic Fluid

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Background & Objectives: The purpose of this study was to isolate adult stem cells from amniotic fluid obtained by second-trimester amniocentesis.

Method: Mesenchymal stem cell (MSC)-like cells were obtained from amniotic fluid obtained by second-trimester amniocentesis. The phenotypic characteristics of these amniotic fluid-derived MSCs (AFMSCs) were examined using RT-PCR, immunocytochemistry and telomerase activity assay. To examine their differentiation potential, AFMSCs were cultured in specific induction medium for the osteogenesis, chondrogenesis, adipogenesis and neurogenesis. After culture, cell differentiation was assessed by Von Kossa, Oil red O, Alcian Blue and Neu N stainings.

Results: Human AFMSCs were successfully isolated and maintained through 27 passages. RT-PCR analyses of the AFMSC at 9th passage showed the prominent expression of Oct-4, SCF, vimentin, CK18 and BMP-4 genes. Immunocytochemical study after 17 passages demonstrated the distinct expression of collagen I, II, III, IV and XII, fibronectin, HCAM, ICAM-1, PECAM-1, VCAM-1, α -smooth muscle actin, desmin, vWF, TRA-1-60, SSEA-3 and -4. Results of the telomerase activity assay indicated that AFMSC at 5th passage possess the activity. AFMSC cultured in the specific differentiation induction medium exhibited positive staining with each stain, implying that they could differentiate into osteocyte, adipocytes, chondrocyte and neuronal cells under appropriate conditions.

Conclusions: Profiles of gene expression, protein localization and telomerase activity assay of human AFMSC showed typical features of known adult stem cells. Considering their multi-differentiation potential, human AFMSC could be an excellent alternative source for the human cell therapy, replacing MSC and other fetal stem cells.

P-25 Human Amnion-derived Cells Have Mesenchymal Stem/Progenitor Cell Potential

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Background & Objectives: The aim of this study was to isolate adult stem cells from the human

amniotic membrane for the cell therapy.

Method: Mesenchymal stem cell (MSC)-like cells were obtained from the human amnion after a Caesarean section. The phenotypic characteristics of these amnion-derived cells (AMC) were examined using RT-PCR, immunocytochemistry and telomerase activity assay. To examine their differentiation potential, AMC were cultured in specific induction medium for the osteogenesis, chondrogenesis, adipogenesis and neurogenesis. After culture, cell differentiation was assessed by Von Kossa, Oil red O, Alcian Blue and Neu N stainings.

Results: Human AMC were successfully isolated and maintained through 9 passages. RT-PCR analyses of the AMC at 3rd passage showed the prominent expression of Oct-4, SCF, nestin, PAX-6, vimentin, N-CAM, CK18, BMP-4, GATA-4, AFP and HNF-4a genes. Immunocytochemical study after 6 passages demonstrated the distinct expression of collagen I, II, III and XII, fibronectin, HCAM, α -smooth muscle actin, desmin, SSEA-3 and -4. Results of the telomerase activity assay indicated that AMC at 3rd passage possess the activity. AMC cultured in the specific differentiation induction medium exhibited positive staining with each stain, implying that they could differentiate into osteocyte, adipocytes, chondrocyte and neuronal cells under appropriate conditions.

Conclusions: Profiles of gene expression, protein localization and telomerase activity assay of human AMC showed typical features of known adult stem cells. Considering their multi-differentiation potential, human AMC could be an excellent alternative source for the human cell therapy, replacing MSC and other fetal stem cells.

P-26 Comparison of Freezing Methods (Slow-freezing vs Vitrification) in the Aspect of Early Follicular Development (Morphology and Development Associated Genes)

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Background & Objectives: Cryopreservation of ovarian tissues has been performed using slow-freezing method. Recently, a rapid, simple and economical cryopreservation method, vitrification, has been newly applied to ovarian tissues. But, there are still controversies on the efficacy of these two methods. In this study, in order to compare the efficacy of two freezing methods, we evaluated the morphological development and also analyzed the development associated genes of ovarian primordial follicle after slow-freezing or vitrification.

Method: Slow cryoconserved or vitrified ovaries and fresh control from 1-day-old female mice were in-vitro cultured for 5 days. During culture, the ovaries were examined histologically to evaluate morphologically abnormalities and development of primordial follicles at 1 and 5 day after culture, respectively. The