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Oral Presentations

O-43
Evaluating senescence of amniotic fluid mesenchymal stem cell in different passages by Q-PCR analysis of *FoxM1* gene

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Background: Stem cells are undifferentiated cells that have the ability to transform and differentiate into different cell types. These cells have two important characteristics that differentiate them from others. They have the ability to reproduce unlimited and remain in the undifferentiated state. The most important mature types of these cells are mesenchymal cells, which become more susceptible to accumulation of cell damage with passing time and increase longevity. These damages can help to improving recovery, senescence and finally death of cells. Various factors cause the intrinsic and harmful process of senescence such as internal factors as genetics, the expression of some genes as *P53*, Nuclear factor NF-kappa-B and Forkhead Box M1 (*FoxM1).* free radicals and external factors such as environmental changes that affect the function of the cell. FoxM1 is a member of the Forkhead transcription family, which has been actively involved in regulating organism growth, differentiation and cell proliferation and is important for the expression of cell cycle-dependent genes in the G2 phase.

Objective: The main goal of this study was to evaluate the expression level of FoxM1 gene as a marker of senescence in mesenchymal stem cells isolated from human amniotic fluid at different passages.

Materials and Methods: Totally 37 amniotic fluid samples were obtained from pregnant mothers referred to the PND Department of Yazd Reproductive Sciences Institute. After culturing successive passages and examining the cells morphologically and characterizing them by flow cytometry, their aging status was evaluated in several passages by using beta-galactosidase (X-gal Cinna Gene) staining. After RNA extraction by Tripure kit and cDNA synthesis by Thermo Fisher kit, 10 samples in passages 4 and 7 were evaluated for FoxM1 gene expression change as a marker of aging and GAPDH gene as internal reference using (quantitative PCR) technique. The data were analyzed using GraphPad Prism and SPSS version16 software.

Results: Microscopic examination and staining of beta-galactosidase showed that mesenchymal stem cells isolated from amniotic fluid enter the aging stage in different passages. Comparing the results of *FoxM1* gene expression in different passages (2, 4, and 7), showed a statistical meaningful increase of expression in old cells compared to young cells ($F = 10.43$; $P < 0.001$). Despite the increased expression of *FoxM1* gene in the passage 4 compared to the young cells in passage 2, indicated that there was no significant difference between two groups ($t = 1.134$, $p<0.05$). Comparison of *FoxM1* gene expression in aging cells of passage 7 compared with young cells of passage 2, showed that the increase was statistically significant ($t = 3.758$; $p < 0.003$).

Conclusion: *FoxM1* gene expression in cellular aging has an effective role in preventing cellular aging, and control of aging-related traits includes reducing cell doubling time, regenerative power, and differentiation potential.

Key words: *Mesenchymal stem cells, Amniotic fluid, Cellular senescence, FoxM1 gene expression, Q-PCR.*