Human dental pulp stem cells cultured in xenogeneic-free supplemented media

Summary

Introduction: The topic of the study is the cultivation of dental pulp stem cells (hDPSCs) in a xenogeneic-free culture medium. It is not permissible to use cells upon growing under the influence of xenogeneic (extraneous) substances in human clinical practice. The most frequently used in cultivation of hDPSCs is fetal calf serum (FCS/FBS). Unfortunately, these supplements are widespread in hMSCs cultivation, and all gold standard hMSCs properties were postulated in cells cultivated using these supplements. This raises the basic question if and how xenogeneic blood derivatives affect the properties of cells and their growth characteristics. There are two options for replacing these xenogeneic substances in the culture medium: the so-called serum-free media, or human blood supplements, ideally autologous ones. The conducted research was aimed at identifying the effects of xenogeneic and human blood supplements on basic hDPSCs characteristics that are fundamental to introduce the cell therapy into regular medical practice.

Method: By culturing 12 hDPSC lines obtained from adult, deciduous, and natal teeth in 12 different culture media, we investigated the effect of FCS, human blood derivatives, i.e., blood plasma (HP), and platelet-rich blood plasma (PRP) of different concentrations (2%, 10%, 20%) on cultured cells using growth characteristics and phenotypic analysis. First, all lines were cultured up to the 15th passage under standard culture conditions and differentiated into osteogenic, chondrogenic, and adipogenic cell lines to demonstrate stemness.

Results: According to the results of this study, the blood derivative with the highest growth support for dental pulp stem cells of permanent teeth (hADPSCs) and natal teeth (hNDPSCs) is PRP at the concentration of 10%. hDDPSCs grew best in cultivation medium with 10% HP. Phenotypic analysis showed remarkable differences in the expression of cluster of differentiation markers. Cells cultured in media with human blood derivatives have the higher neurogenic potential and a phenotype resembling the embryonic stem cells (hESCs), while cells cultured with FCS tended to express features of the hematopoietic lineage resembling the progenitor cells.

Conclusion: In our study, we demonstrated the effect of the various types of blood substitutes on the hDPSCs proliferation rate and phenotype. The human blood derivatives at the concentration of 10% are an ideal substitute for FCS due to their positive effect on hDPSCs proliferative activity, and support hDPSCs the undifferentiated state and neurogenic potential.

Key words: Mesenchymal stem cells, dental pulp, human blood derivatives, cultivation.