

**Klemen Zupančič** finished undergraduate study of Biotechnology at University of Ljubljana in June 2009. He acquired first experiences with the laboratory work at Biotechnical Faculty under the supervision of Hrvoje Petković, Ph.D. when he was working with Propionibacterium sp. He also worked at Department of Animal Science at Biotechnical Faculty under the supervison of Tanja Kunej, Ph.D. where he cooperated on bioinformatics project. At the end of last year of undergraduate study he has been given the opportunity to do a research work for his graduation thesis under the supervision of Miomir Knezević, Ph.D., Asist. Prof. and Darja Marolt, Ph.D. at prof. Gordana Vunjak-Novakovic's Laboratory for Stem Cells and Tissue Engineering at Columbia University, New York. Currently he is continuing to study Biomedicine at University of Ljubljana and does PhD research at National Institute of Biology. He has also helped kick-start several start-up companies among which GenePlanet d.o.o. and BPK d.o.o. are the most successful.

## Graduation thesis: The effect of culture in three dimensional environment on growth and differentiation of human stem cells

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The main obstacle, when manipulating stem cells in the laboratory is that the path of their differentiation is difficult to control. More and more studies are investigating the effects of three-dimensional (3D) environment on differentiation of stem cells, which is different and unique for every tissue type. Cell scaffolds can be used as 3D environment, among which hydrogels are notable (collagen, hyaluronic acid, alginate). In experiments for this thesis we investigated the effects of 3D environment on growth and differentiation of human stem cells in comparison to classic forms of 3D environment (pellets of cells and embryoid bodies). Our experiments were done on human embryonic stem cells (hESC) and adipose stem cells (hASC). We estimated viability of cells using Live/Dead assay, metabolic activity using XTT assay, number of cells by measuring quantity of DNA, and for determining gene expression and thus differentiation path real-time PCR was used. hASC grew better in collagen gel than in hyaluronic acid, gene expression of cells in collagen was similar to control groups. When cultures of hESC were observed, the cells, grown in hyaluronic acid were significantly more undifferentiated compared to the cells, grown in collagen, that showed similar differentiation pathway to the cells in embryoid bodies. Based on our results we can conclude, that different cell environments can significantly affect cell differentiation. In combination with growth factors, hydrogels offer different possibilities for culture of stem cells.