

ABSTRACT

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Title of diploma thesis: Characterization of different differentiation protocols of H9c2 cardiomyoblasts

Cell differentiation is a common process in the development of multicellular organisms leading to an alteration of cell's appearance and function. The process of differentiation plays a significant role in the reparation of damaged tissues too. Some chemicals may have negative effects on these processes and therefore affect human health.

This thesis focuses on characterization of differentiation processes of H9c2 cell line using various published differentiation protocols, and to describe changes over time in the morphology (occurrence of multinucleated and elongated cells) and mRNA expression of selected markers typical for the cardiac (cTnT, Hand2, GATA4) or skeletal (myogenin) muscle phenotype. We monitored the process of the differentiation for 15 days and the individual protocols differed by the addition of fetal bovine serum (FBS, 1 % or 10 %) and all-trans-retinoic acid (ATRA, 10 nmol/L) to the growth medium Dulbecco's Modified Eagle's Medium.

From what we have observed, the results show that all of the utilized protocols led to the cell differentiation that changed quantitatively and qualitatively as well as with time. The age of the culture had an important effect as well. Among others, relatively homogenous change was observed in the cardiac troponin T and for myogenin gene expression. The effect of serum deprivation was manifested mainly at the beginning of the differentiation process (up to the 9th day), later on more changes were observed in the medium with the higher FBS concentration. Regarding the homogeneity of the acquired results, we have decided to focus on the protocol which uses the 10% FBS medium with the addition of ATRA in the future experiments and confirm the changes in detail.