

NON INVASIVE AND LABEL FREE ANALYSIS OF OSTEOGENIC EVENTS DURING MINERALIZATION OF HUMAN BONE MARROW DERIVED STEM CELLS

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ABSTRACT:

In the field of tissue engineering, there is a high demand for non-invasive and label free measurements towards long term monitoring of cellular activities. To achieve this goal, a confocal hyper spectral Raman microspectroscopy [1], based on home-built spectral detection technology is designed which makes it possible to observe *in vitro* development of tissues from cultured cells. Vibrational Raman microspectroscopy is an optical technique with high chemical specificity, allowing *in situ* chemical analysis of complex systems by non invasive and label free methods. Human bone marrow derived stem cells are cultured under the influence of osteogenic mineralization medium that provide physical and chemical cues to guide differentiation and assembly into three dimensional mineralized tissue.

Results showed successful culture of human bone marrow derived stem cells over a period of more than 8 weeks under carefully maintained conditions. Over the culture period, the stem cells attached, spread and grew until they reached the stage of confluence. The presence and variation in the events of mineralization over ECM could be detected with confocal Raman microspectroscopy, which is not feasible with conventional techniques and staining procedures [2]. By week 4 of the culture period, mineralized extracellular matrix (ECM) showing *de novo* bone formation was detected. Further culture from week 4 till week 8 showed the transformation of *de novo* bone nodules towards crystalline bone nodules similar to *in vivo* bone in mineral composition. Confocal Raman microspectroscopy gives detailed information on events and transformations in mineralization and is capable to correlate those events with the organic phase.

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