

ISOLATION AND PRELIMINARY CHARACTERISATION OF STEM CELLS FROM HUMAN DENTAL PULP

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INTRODUCTION: Therapies based upon cell replacement and tissue engineering, underpinned by stem cell biology, are emerging as potentially powerful strategies in modern regenerative medicine. Recently, existing concepts of cell lineage, commitment and differentiation have been challenged by the use of adult stem cells as a source of pluripotent cells. The aim of this study was to investigate the potential of using human dental pulp tissue as a source of pluripotent stem cells.

METHODS: Fifty dental pulps were extirpated from healthy permanent teeth (third molars and premolars) extracted at Leeds Dental Institute. After splitting the teeth, the pulps were removed and divided into six transverse segments in an apical-coronal direction to investigate any site-specific differences in stem cell potential. Primary cells were isolated using standard organ culture methods, establishing a total of 100 cultures. Cells were cultured in basal media or under osteogenic, chondrogenic and adipogenic conditions and assessed by enumeration of colony forming units fibroblastic (CFU-F) formation, histological staining (alkaline phosphatase - ALP, alcian blue/sirius red, von Kossa, Oil Red, Toluidine Blue), biochemical assays (ALP, DNA content, sGAG), flow cytometry and light microscopical analysis.

RESULTS: The majority of human dental pulp derived stem cells (hDPSCs) demonstrated classic spindle shaped fibroblast-like morphology and growth characteristics. The proliferative potential of both the dental pulp derived CFU-F and primary cell cultures varied from tooth-to-tooth and from site-to-site within individual pulps. Cells derived from the

middle segments of the pulp demonstrated stronger ALP positive activity compared with those derived from the more apical and coronal regions. Microscopical analyses of the hDPSCs following culture in specialised media indicated osteogenic, chondrogenesis neural and epithelial morphologies whereas adipogenesis was low. Mineralized deposits were observed when the cells were cultured under osteogenic conditions. hDPSCs had surface phenotype similar to human mesenchymal stem cells (CD45⁻CD34⁻CD133⁻CD105⁺CD73⁺CD166⁺).

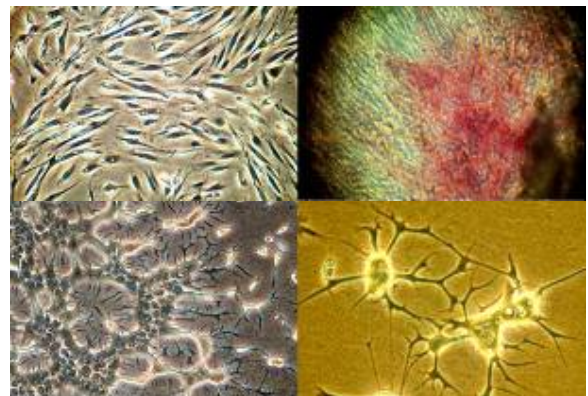


Fig. 1: Plasticity of human dental pulp stem cell.

DISCUSSION & CONCLUSIONS: We conclude that pluripotent stem cells are present within human dental pulp. The reasons for the observed site-specific differences in proliferative and osteogenic potential are unknown but they may reflect differences in the relative contributions of the different tissue compartments at these locations. This study, together with the work of others, indicates the potential for using dental pulp as a source of stem cells for future tissue replacement therapies and tissue engineering strategies.