In the Name of Allah the Most Gracious and the Most Merciful

Isolation of Dental Pulp Stem Cells and their Ex Vivo Differentiation into Odontoblasts

THESIS

Submitted to the Faculty of Oral and Dental Medicine

Cairo University

In partial fulfillment of the requirements for the

Master's degree in Endodontics

By

Nermeen Elmotaz Bellah Ahmed, B.D.S. (2002)

Assistant Researcher (Oro-dental Genetics Department)

National Research Center

Supervisors

Prof. Dr. Siza Yacoub Zakhary

Professor of Endodontics

Department of Endodontics

Faculty of Oral and Dental Medicine

Cairo University

Prof. Dr. Eman Hassan Anwar Aboul-Ezz

Professor and head of Oro-Dental Research Division

National Research Center

To my beloved mom and dad,

For you being the light of my life,

My heart and soul,

From you I get my strength and belief,

Thank you for everything,

You are my all

ACKNOWLEDGMENTS

To Dr. Misako Nakashima, thank you for accepting me in your lab in the first place. Thank you for teaching me, helping me to know better and pushing me to do my best. You are a great scientist and indeed a very kind person. Being to your lab was a dream that came true. With your knowledge, guidance and support this work became possible. I am forever grateful to you.

To my professor Dr. Eman Hassan Anwar Aboul-Ezz, this work would not have been possible without your support and encouragement. Under your supervision, I chose this topic and began the thesis. Thank you for believing in me and having faith in my abilities, even when I felt down. Thank you for always being there when I needed you. You are a real example of how a mentor should be. I am forever in your debt.

To my professor Dr. Siza Yacoub Zakhary, thank you so much for your support and encouragement. Thank you for having patience with me, supporting me in this topic and seeing me through to the very end. You are a great professor and a wonderful person. Words cannot explain my deep gratitude and respect to you.

To Dr. Tarek Hamed El Badry, Thank you for helping me to make this thesis look better with your wonderful to the point remarks and thank you for your help in writing this thesis.

I send enormous thanks to the National Society of Human Genetics which I have the honor to be a member of, and to its head, the great professor Dr. Samia A. Temtamy, for their support and help. Without their financial and spiritual support, this work would have never been done. For my great professor Dr. Samia A. Temtamy, nothing can give her what she really deserves. She is and always will be my idol.

A special thanks to the entire team at the oral regeneration laboratory at the National Institute of Longevity Sciences (NILS), Obu, Japan especially Iohara Koichiro and Li Zheng for their help in the scientific execution of the experiments. Although it was difficult for them to communicate in English, they did their best to teach me, guide me and help me through my experiments. I really owe them a lot.

To all my friends, thank you for everything. Thank you for your support, trust and always being there for me, especially when I was depressed the most. Being your friend is a gift, which I should be thankful for, my entire life.

I cannot end without thanking my family, on whose constant encouragement and love I have relied throughout my time in Japan. Their patience, love, and continuous prayers were the guidance for me in my hardest times. Although it was very hard for them to have me away, they always helped me through the tough time and made it easy for me. To them I bow in respect.

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List of Abbreviations

BM: Basement membrane

BMP: Bone morphogenic protein

BMSSC: Bone marrow stromal stem cells

β actin: Beta actin

CD: Clusters of differentiation

cDNA: Complementary Deoxyribonucleic acid

DEJ: Dentino-enamel junction

DEPC: Diethyl procarbonate

DMEM: Dulbecco modified eagle's media

DNA Deoxyribonucleic acid

DP: Dental papilla

DPSC: Dental pulp stem cell

DSPP: Dentin sialophosphoprotein

EBM-2: Endothelial basal cell medium-2

ECM: Extracellular matrix

EDTA: Ethylenediamine tetraacetic acid

ESC: Embryonic stem cell

FACS: Florescence-activated cell sorting

FBS: Fetal bovine serum

GAG: Glycosaminoglycans

Gdf: Growth differentiation factor

GFAP: Glial fibrillary acid protein

HEPS: 1-(2-Hydroxyethyl)-1 piperazineethanesulfonic acid buffer solution

HSC: Hematopoietic stem cell

IDE: Inner dental epithelium

MACS: Magnetic-activated cell sorting

MC: Mesenchymal cell

$MgCl_2$: Magnesium chloride

mRNA: Messenger ribonucleic acid

MSC: Mesenchymal stem cell

PBS: Phosphate buffered solution

PCR: Polymerase chain reaction

PDL: Periodontal ligament

PDLSC: Periodontal ligament stem cell

PI: Propidium iodide

PPARϒ2: Peroxisome proliferating activated receptor gamma 2

rh-BMP2: Recombinant human bone morphogenic protein 2

rh-EGF: Recombinant human epidermal growth factor

rh-IGF-1: Recombinant human insulin growth factor-1

RNA: Ribonucleic acid

Rt-PCR: Reverse transcriptase polymerase chain reaction

SCAP: Stem cells from root apical papilla

SHED: Stem cells from human exfoliated deciduous teeth

SP: Side population

STAT3: Signal transducer and activator of transcription 3

TGF: Transforming growth factor

UV: Ultraviolet

VCAM-1: Vascular adhesion molecule-1

VEGF: Vascular endothelial growth factor

Introduction

Despite our extensive knowledge regarding the pathology of dental disease, restoration of diseased dental tissue to date remains fairly empirical. However, our increasing understanding of the exquisite regenerative potential of the dentine- pulp complex highlights the importance of characterizing fully the cellular and molecular processes under-pining dentin regeneration.

Pulpal exposure, due to caries shows very limited potential for pulp recovery due to bacterial infection of the pulp for substantial period of time, which compromises the defense reaction.

Vital pulp therapy aims to treat reversible pulpal injury and maintain pulp vitality and function. It includes two therapeutic approaches: indirect pulp capping in cases of deep dentinal cavities and direct pulp capping in cases of pulp exposures. Successful outcome for vital pulp therapy is very dependent on the type and location of injury, age of the tooth, treatment modality (capping material) and integrity of the cavity restoration.

Whilst the biological processes directed by the treatment strategy have received much attention, controversy still exists regarding the biological basis of the mechanism by which the capping material regulates healing and repair of the pulp in vital pulp therapy. Even if reparative dentin is formed, its orientation