A comparative study between different types of umbilical cord stem cells in treatment of experimental diabetic rats

A thesis submitted for the partial fulfillment of M.Sc. Degree in

Pharmaceutical Sciences (Biochemistry)

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رسالة توطئة للحصول على درجة الماجيستير في العلوم الصيدلية

(تخصص كيمياء حيوية)

مقدمة من

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تحت إشراف

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بسم الله الرحمن الرحيم

صدق الله العظيم

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Publications related to the Thesis

I. Poster presented in 20th ISCT Annual Meeting
April 23-26, 2014, Paris, France

Cytotherapy
Volume 16, Issue 4, Supplement, April 2014, Pages S66
20th Annual ISCT Meeting

Comparing umbilical cord blood stem cells and wharton's jelly mesenchymal stem cells regarding their differentiation potential to insulin producing cells

M.M. Kamal[1], H.O. El-Mesallamy[1], L.N. Hammad[2], R.F. El-Demerdash[2]

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Introduction

The number of patients suffering from Diabetes Mellitus (DM) is growing in an alarming rate which makes DM the most prevalent and serious metabolic disease. Now, cell therapy treatment options for diabetic patients are under extensive study. Interestingly, umbilical cord (UC) has been proved to be a good source of mesenchymal stem cells (MSCs), namely from umbilical cord blood (UCB-MSCs) and Wharton's jelly (WJ-MSCs).

Objectives

We thought to investigate the difference between these 2 important banking sources of stem cells and to compare their differentiation potentials towards insulin producing cells (IPCs) in vitro and their potential use for treatment of streptozotocin (STZ) induced diabetic rats invivo.

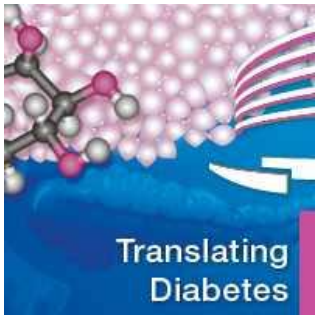
Materials and methods

Both UCB-MSCs and WJ-MSCs were isolated from UC and expanded for several passages. Expression of typical MSCs surface antigens and adipogenic differentiation potential as an example of mesenchymal lineage was used to verify MSCs identity. Afterwards, both UCB-MSCs and WJ-MSCs were induced to differentiate into IPCs, then the differentiated cells were assessed both genetically by determining the expression of Nestin, as stem cell marker and key markers of mature β-cells such as Pdx-1, Mafa and Nkx2.2 using qRT-PCR, and functionally by measuring insulin secretion after glucose challenge (Glucose stimulated insulin secretion; GSIS); a hall mark of functional β-cells.

Results and conclusions

WJ appeared to be a much more homogenous and potential source for MSCs as compared to UCB. Interestingly, both UCB-stem cells and WJ-MSCs were successfully differentiated to IPCs. Yet, the resulting IPCs from WJ-MSCs were to a limited extent functioning better than those obtained from UCB-MSCs. Both cell types were able to decrease fasting blood glucose level transiently in STZ induced diabetic mice. Taken together, we can conclude that WJ could represent a potential source of cells in the field of DM cell therapy rather than UCB.

II. Poster presented in EMBO/EMBL Symposium-Translating Diabetes April 30-May 3, 2014, Heidelberg, Germany



The generation of insulin producing cells from Wharton's jelly mesenchymal stem cells in comparison to umbilical cord blood mesenchymal stem cells

Introduction: The number of patients suffering from Diabetes Mellitus (DM) is growing in an alarming rate which makes DM the most prevalent metabolic disease. Now, cell therapy treatment options for diabetic patients are under extensive study. Interestingly, umbilical cord (UC) has been proved to be a good source of mesenchymal stem cells (MSCs), namely; umbilical cord blood (UCB-MSCs) and Wharton's jelly (WJ-MSCs).

Objectives: we thought to investigate the difference between these 2 important banking sources of stem cells and to compare their differentiation potentials towards insulin producing cells (IPCs) invitro and their potential use for treatment of streptozotocin-induced diabetic rats invivo.

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LIST OF ABBREVIATIONS

ANOVA	Analysis of variance
ASCs	Adult stem cells
BM	Bone marrow
BW	Body weight
CB	Cord blood
CD	Clusters of differentiation
cDNA	Complementary deoxy nucleic acid
Ct	Cycle threshold
DM	Diabetes mellitus
DMEM	Dulbecco's modified Eagle's medium
dNTP	Deoxy nucleotide tri-phosphate
ELISA	Enzyme linked immuno-sorbent assay
ESCs	Embryonic stem cells
FACS	Fluorescence-activated cell sorting
FBG	Fasting blood glucose
FBS	Fetal bovine serum
FITC	Fluorescein isothiocyanate
GDM	Gestational diabetes mellitus
GSIS	Glucose stimulated insulin secretion
GVHD	Graft-versus-host disease
H&E	Hematoxylin and eosin stain
HG-DMEM	High glucose- Dulbecco's modified Eagle's medium
HG-KRB	High glucose-Kreb's ringer bicarbonate
HLA	Human leukocyte antigen
HSCs	Hematopoietic stem cells
IDDM	Insulin dependent diabetes mellitus
IDF	International diabetes federation
Ig	Immunoglobulin
IPCs	Insulin producing cells
IR	Insulin resistance
ISCT	International society for cellular therapy
Isl-1	Insulin gene enhancer protein
KRB	Kreb's ringer bicarbonate

LG- KRB	Low glucose-Kreb's ringer bicarbonate
LG-DMEM	Low glucose-Dulbecco's modified Eagle's medium
MafA	V-maf musculoaponeurotic fibrosarcoma oncogene homolog
MHC	Major histocompatibility complex
MNCs	Mononuclear cells
MSCs	Mesenchymal stem cells
NA	Nicotinamide
Ngn-3	Neurogenin-3
NIDDM	Non-insulin dependent diabetes mellitus
P	Passage
PBS	Phosphate-buffered saline
PCR	Polymerase chain reaction
Pdx-1	Pancreatic and deodenal homebox1
PE	Phycoerythrin
Pen/Strep/ Ampho	Penicillin /streptomycin /amphotercin B
qRT-PCR	Quantitative reverse transcriptase polymerase chain reaction
RNase	Ribonuclease enzyme
RT	Reverse transcriptase enzyme
STZ	Streptozotocin
Taq	*Thermusaquaticus*
UC	Umbilical cord
UCB	Umbilical cord blood
UCWJ	Umbilical cord Wharton's jelly
WHO	World health organization
WJ	Wharton's jelly
α-MEM	Alpha minimum essential medium

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