

بسم الله الرحمن الرحيم



HOSSAM MAGHRABY



شبكة المعلومات الجامعية التوثيق الالكتروني والميكروفيلم



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جامعة عين شمس

التوثيق الإلكتروني والميكروفيلم

قسم

نقسم بالله العظيم أن المادة التي تم توثيقها وتسجيلها
على هذه الأقراص المدمجة قد أعدت دون أية تغيرات



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تحفظ هذه الأقراص المدمجة بعيدا عن الغبار



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**Histological Effect of Platelet Rich Plasma
Versus Adipose Tissue Derived Mesenchymal
Stem Cells on Carbon Tetrachloride Induced
Liver fibrosis in Albino Rat**

Thesis

Submitted for Partial fulfillment of
M.D. Degree in Histology

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بِسْمِ اللَّهِ الرَّحْمَنِ الرَّحِيمِ

قالوا

سبحانك لا علم لنا
إلا ما علمتنا إنك أنت
العليم العظيم

صدق الله العظيم

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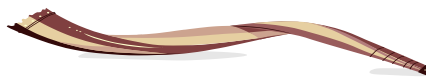
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DEDICATION

To those who touched my life, provided the support and guidance whenever needed.

To my dearly beloved mother and father.

To my husband.

To my daughter, to my sons, to my sisters and brothers

To my professors



Thank you all

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List of Abbreviations

Abb.	Full term
µl.....	Microliter
ADP	Adenosine diphosphate
ADSCs	Adipose tissue derived mesenchymal stem cells
AFLD.....	Alcoholic fatty liver disease
ALD	Alcoholic liver disease
ALT\AST	Alanine transaminase\Aspartate transaminase
AT	Adipose tissue
ATP	Adenosine tri-phosphate
bFGF	Basic fibroblast growth factor
BM	Bone marrow
BMSCs.....	Bone marrow mesenchymal stem cells
CCL3.....	Trichloromethyl free radical
CCL3O2-	Trichloroperoxyl radical
CCL4.....	Carbon tetrachloride
CD.....	Cluster of differentiation
c-Met.....	Mesenchymal to epithelial transition receptor
CYP.....	Cytochrome P
DMEM.....	Dulbecco's modified Eagle's medium
ECM.....	Extracellular matrix
EDTA.....	Ethylene diamine tetra-acetate
EGF	Epidermal growth factor
EGFR	Epidermal growth factor receptor
EV	Extracellular vesicles
FBS	Fetal bovine serum
GFs.....	Growth factors
gm	Gram
hASCs	Human Adipose-derived Stem Cells

List of Abbreviations Cont...

Abb.	Full term
HBV	Hepatitis B virus
HCV	Hepatitis C virus
HDL	High-density lipoproteins
HGF	Hepatocyte growth factor
HLCs	Hepatocyte-like cells
HSC.....	Hepatic Stellate Cell
IFN- γ	Interferon- γ
IGF	Insulin like growth factor
IL.....	Interleukin
IRF	Interferon regulatory factor
JAK	Janus kinase
KCs	Kupffer cells
kg	Kilogram
LDL.....	Low-density lipoproteins
lncRNA H19	Human long-chain non-coding RNA
LP-PRP	Leukocyte-poor PRP
LPS	Lipopolysaccharide
LR-PRP.....	Leukocyte-rich PRP
Ly6C	Lymphocyte Ag 6C
MFBs	Myofibroblasts
ml	Milliliter
NAFLD	Non-alcoholic fatty liver disease
NASH	Non-alcoholic steatohepatitis
PAP	Platelet average plasma
PAS	Periodic acid-schiff reagent.
PBS	Phosphate buffered Saline
PCNA.....	Proliferating Cell Nuclear Antigen

List of Abbreviations Cont...

Abb.	Full term
PCR.....	Polymerase chain reaction
PDGF	Platelet derived growth factor
PHx	Partial hepatectomy
PPP.....	Platelet poor plasma
PRP	Platelet rich plasma
RER.....	Rough endoplasmic reticulum
ROS.....	Reactive oxygen species
rpm.....	Revolution per minute
SER.....	Smooth endoplasmic reticulum
SPSS	Statistical Package for the Social Sciences
SRY.....	Sex determining region of Y chromosome
STAT1	Signal transducers and activators of transcription 1
SVF	Stromal vascular fraction
TAA	Thioactamide
TGF.....	Transforming growth factor
TGFβ.....	Transforming growth factor beta
TIRAP.....	Toll-interleukin 1 receptor domain containing adaptor protein
TNF.....	Tumor necrosis factor
uPA	Urokinase-type plasminogen activator
VGEF.....	Vascular endothelial growth factor
VLDLs	Very low-density lipoproteins
WBCs.....	White blood cells
Wnts.....	Wingless-related integration site
α- SMA	Alpha smooth muscle actin

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Abstract:

Background: Liver cirrhosis is a significant lethal health problem all over the world. Liver transplantation is the only efficient treatment. Unfortunately, it is limited due to many obstacles. Therefore, alternative treatments are required for cirrhosis. Platelet rich plasma (PRP) and adipose tissue derived mesenchymal stem cells (ADSCs) are a challenging measure for treatment of cirrhosis.

Aim of the work is to assess the effect of PRP versus ADSCs on experimentally induced liver fibrosis in albino rats.

Material and Methods: forty-two adult female rats were included in this study. In addition, twenty young-weaned albino rats (five males and fifteen female) were used for preparation of ADSCs and PRP. The adult females were divided into three main groups: **Group I: control group.** **Group II:** that received carbon tetrachloride (CCL₄) in dose of 0.5 mg/kg twice weekly for six weeks. They were subdivided into 2 subgroups; **subgroup IIA** where rats were sacrificed after the last injection and **subgroup IIB** where rats were left for spontaneous recovery for four weeks after the last CCL₄ injection. **Group III:** which were further subdivided into 2 subgroups; **subgroup IIIA** where rats were subjected to fibrosis then received PRP in a dose of 1ml/kg twice weekly for four weeks and then were sacrificed. Whereas, **subgroup IIIB** that were subjected to fibrosis and then received ADSCs in a dose of 3×10^6 cells/rat once. Liver specimens were prepared for histological and immune-histochemical techniques. Morphometrical and statistical studies were done.

Results: CCL₄ injection resulted in distortion of the liver architecture and multiple injuries in hepatocytes including vacuolations in the cytoplasm and pyknotic nuclei. In addition, it resulted in hepatic fibrosis especially surrounding the central vein and portal areas. Treatment by PRP resulted in an obvious histological improvement in hepatic structure and most

hepatocytes showed acidophilic cytoplasm with basophilic granules and vesicular nuclei and significant decrease in hepatic fibrosis ($p < 0.05$). Unfortunately, few areas in the field of hepatic tissue were not improved. Moreover, there was significant increase in the mean number of Proliferating Cell Nuclear Antigen (PCNA). In addition, there was a significant decrease in the area percentage of α -SMA positive cells, the mean number of caspase-3 positive cells and area percentage of anti-vimentin ($p < 0.05$). ADSCs treatment also resulted in improvement of hepatic structure in the form of significant decrease in hepatic fibrosis and significant increase in the mean number of PCNA. There was also a significant decrease in the area percentage of α -SMA positive cells, the mean number of caspase-3 positive cells and area percentage of anti-vimentin as compared to untreated groups ($P < 0.05$).

Conclusion: There was non-significant difference between PRP and ADSCs in treatment of hepatic fibrosis. Accordingly, PRP could be an adjuvant treatment for hepatic fibrosis which needs more investigations.

Key words: liver fibrosis, CCL4, PRP, ADSCs, rat.