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لم ترد بالأصل

HOSSAM MAGHRABY

OXIDANT STRESS IN CHILDREN WITH THALASSEMIA

THESIS

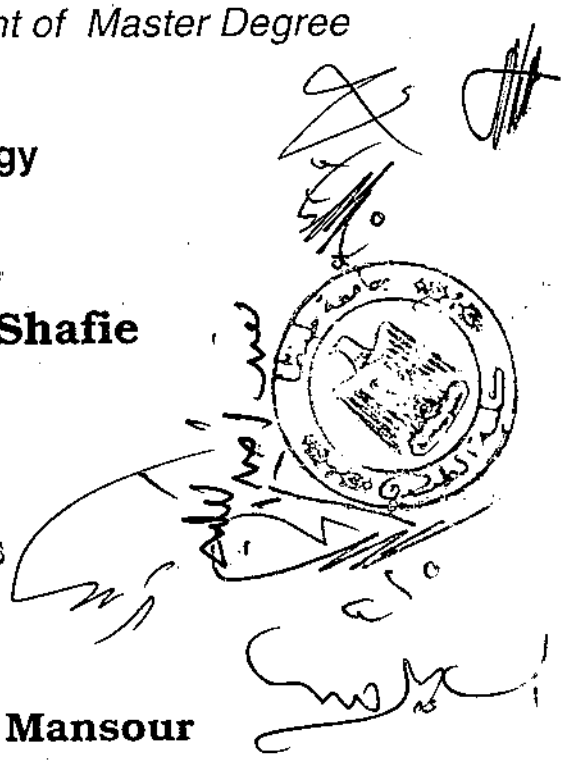
Submitted for the Partial Fulfillment of Master Degree

In

Clinical Pathology

By

Ghada Hamed El-Shafie
(M.B.,B.Ch.,)



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قَالُوا سُبحانَكَ لا عِلم لَنَا إلاّ ما علَمتنا إنّكَ أنتَ العَليمُ الحكيم

صدق الله العظيم

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To

my

Parents

LIST OF ABBREVIATIONS

ADP	:	Adenosine diphosphate.
ALT	:	Alanin transaminase.
AST	:	Aspartate transaminase.
αTH	:	α-tocopherol.
ATP	:	Adenosine triphosphate.
CCL3	:	Tricholoromethyl radical.
Cu	:	Copper.
(CUZN) SOD:		Cuprozinc superoxide dismutase.
DFO	:	Desferrioxamine.
DNA	:	Deoxyribonucleic acid.
$\Delta O^{\cdot -}_2$	:	Delta form of singlet oxygen.
e'	:	Electron.
EDTA	:	Ethylene diamine tetra-acetic acid.
ELT	:	Euglobin lysis time.
Fe^{2+}	:	Ferrous.
Fe^{3+}	:	Ferric.
fl	:	Femtoliter.
G-PX	:	Glutathione peroxidase.
GSH	:	Reduced Glutathione.
GSSG	:	Oxidized Glutathione.
H+	:	Hydrogen ion.
H_2O_2	:	Hydrogen peroxide.
Hb	:	Haemoglobin
HbA	:	Adult haemogloin
HbF	:	Fetal haemogloin
HOCL	:	Hypochlorus acid.
HPLC	:	High performance liquid chromatography.
LCR	:	Locus control region.
LO^{-}	:	Alkoxy radicals.
$LOO\cdot$	:	Peroxyl radical.
LOOH	:	Lipid hydroperoxides.
MCH	:	Mean corpuscular haemogloin.
MCHC	:	Mean corpuscular haemogloin concentration.
MCV	:	Mean corpuscular volume.
MDA	:	Malonyldialdehyde.
MEOS	:	Microsomal ethanol oxidizing system.

Mn	:	Manganese.
MnSOD:		Manganese superoxide dismutase.
NAD+	:	Nicotinamide adenine dinucleotide.
NADH	:	Reduced nicotinamide adenine dinucleotide.
NADP	:	Nicotinamide adenine dinucleotide phosphate.
$O^{\cdot}_2$	:	Superoxide radical.
O_2	:	Singlet oxygen.
O_2	:	Molecular oxygen.
OFRs	:	Oxygem free radicals.
OH	:	Hydroxyl Radical.
pg	:	Picogram.
PT	:	Prothrombin time
PTT	:	Partial thromboplastin time
R-S	:	Thiyl radical
RBCs	:	Red blood cells.
RNA	:	Ribonucleic acid.
ROO	:	Lipid hydroperoxides.
ROS	:	Reactive oxygen species.
Se	:	Selenium.
$\Sigma O^{\cdot}_2$	:	Sigma form of singlet oxygen.
SOD	:	Superoxide dismutase.
TBA	:	Thiobarbituric acid.
TBARS	:	Thiobarbituric acid reactive substances.
UV	:	Ultraviolet.
WBC	:	White blood cells.

CONTENTS

INTRODUCTION

INTRODUCTION

Oxygen free radicals are extreemly reactive species that although crucial to a wide range of normal biological processes are potentially damaging if produced in excessive amounts. They can disrupt cell membrane (through lipid peroxidation), destroy cell enzyme functions, alter DNA and lead to cell death (Sullivan and Newton 1988).

Physiologic oxidant challenge almost certainly begins with generation of superoxide ($O_2^{\cdot -}$). The enzymatic or spontaneous dismutation of ($O_2^{\cdot -}$) yields peroxide (H_2O_2). The reaction of H_2O_2 with ferrous iron is the actual $OH^{\cdot}$ generating step. The importance of ($O_2^{\cdot -}$) lies in its ability to reduce Fe +++ back to Fe++. There by reading the iron for another cycle of $OH^{\cdot}$ formation. Consequently, availability of reducing agents is critical because their absence would make $OH^{\cdot}$ generation self limited as determined by amounts of ferrous iron. A variety of reducing agents present within the RBCs e.g. GsH, flavinoids and ascorbate potentially can substitute for O_2 (Hebbel, 1986 and Halliwell 1992).

The erythrocyte antioxidant mechanism may convently be divided into two parts. Some systems function to detoxify free radicals before they cause damage and other exist to repair damage only after it has occurred. The dismutation of O_2 is accelerated enormously by superoxide dismutase (SOD), the lipid peroxy radicals ($LOO^{\cdot}$) are converted to lipid hydroperoxides

(LOOH) by vitamen E (αTH) (Whitting, 1980), The only detoxification mechanism specifically located in the RBC membrane. Other non enzymatic antioxidants such as Vit C and β caroten may have a role in this defence system (Mohamed et al., 1996).

Thalassemic erythrocytes contain more lipid per cell which is susceptible to pro-oxidation, the distribution of fatty acids in these cells suggests that auto-oxidation of that lipid may have occured. Auto-oxidation may be initiated by free radicals which are constantly formed in the normal red cell and may be specially prevalent when unstable haemoglobin is present. The low MCHC or some other intracellular defect of the thalassemic cells may allow such potent oxidants to find their way to the cell membrane (Rachimilewitz et al., 1976). An additional factor which might play a significant role in lipid membrane peroxidation and decreased levels of vitamen E is related to the excess amounts of iron. Ferrous iron is known to be catalyst in the non enzymatic auto-oxidation of unsaturated fatty acids, a reaction which is specifically inhibited by alpha tocopherol (Smith et al., 1986 and Mohamed et al., 1996).

Thiobarbituric acid reactive substances (TBARS) such as malonyldialdehyds (MDA) which is a secondary break down product of lipid peroxidation can be used as an indicator of RBCs sensitivity to pro-oxidant stimuli and for RBCs oxidative damage (Corrons et al., 1995).